combine nicely, consume wisely

"The role of single food properties in oral processing behavior, intake and sensory perception of composite foods"

Arianne van Eck

Propositions

1. *1.* Food shape changes eating behavior more advantageously than food hardness. *(this thesis)*

2. *2.* Solid foods drive food oral processing behavior of composite foods, whereas liquid and semi-solid foods are stronger drivers of flavor perception.
   (this thesis)

3. Scientists should write news items in addition to scientific papers, as consumers still believe sweet can be tasted on the tip of the tongue only.

4. Providing city art such as the "plastic skyscraper whale" increases environmental awareness more effectively than education campaigns (Hedefalk *et al.* (2015) Environmental Education Research, 21:7, 975-990).

5. Making people pay by phone is more effective than changing the type of banknote-paper to reduce transmission of bacteria (Gedik *et al.* (2013) Antimicrob Resist Infect Control 2, 22).

6. Providing a proper coffee machine improves collaboration more than flex desks.

7. *Train pride* should become the new *flight shame.*

Propositions belonging to the PhD thesis, entitled

"Combine nicely, consume wisely: The role of single food properties in oral processing behavior, intake and sensory perception of composite foods."

Arianne van Eck

Wageningen, 19 June 2020

Combine nicely, consume wisely.

The role of single food properties in oral processing behavior, intake and sensory perception of composite foods.

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This research was conducted under the auspices of the Graduate School VLAG (Advanced studies in Food Technology, Agrobiotechnology, Nutrition and Health Sciences).

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Thesis
submitted in fulfilment of the requirements for the degree of doctor
at Wageningen University
by the authority of the Rector Magnificus,
Prof. Dr A.P.J. Mol,
in the presence of the
Thesis Committee appointed by the Academic Board
to be defended in public
on Friday 19 June 2020
at 1:30 p.m. in the Aula.

Arianne van Eck
Combine nicely, consume wisely. The role of single food properties in oral processing behavior, intake and sensory perception of composite foods.
242 pages.

PhD thesis, Wageningen University, Wageningen, The Netherlands (2020)
With references, with summary in English

ISBN: 978-94-6395-346-7
DOI: 10.18174/517695

Cover by: Hannah Romijn | www.hannahvormgeving.nl
Layout inside by: ProefschriftMaken | www.proefschriftmaken.nl

Never lose your sense of wonder

ABSTRACT

Combine nicely, consume wisely.

Consumers frequently combine two or more single foods within one bite; for example bread with spread or vegetables with dressing. Such food pairings are called composite foods. This research investigates the effects of food properties on oral processing behavior, intake and sensory perception of composite foods.

Results show that eating behavior of composite foods can be modified by relatively small changes in single food properties. In addition, sensory perception of composite foods is complex, as interactions between foods in mouth imply significant changes in sensory perception. Consequently, consumer sensitivity to discriminate between foods is reduced when a food is assessed together with an accompanying food.

This thesis provides new insights into how structural transitions of foods contribute to intake and perception of composite foods. This is of particular interest in the design of healthy or environmentally friendly foods, in which assuring excellent sensory quality still poses a challenge.

CONTENTS

GENERAL INTRODUCTION

1

1. GENERAL INTRODUCTION

Good food, good mood - Food consumption impacts health outcomes and consumers' well-being. Foods are consumed to obtain energy and nutrients for the human body; foods are also consumed to experience pleasure and reward. Although consumers pursue values such as health, perceived sensory characteristics are still a very important driver of food appreciation and pleasure during the course of eating. In daily life, this often presents a paradox: the healthier, nutritious food options are frequently less liked by consumers whereas highly palatable (often energy-dense) foods are easily overconsumed, contributing to diet-related non-communicable diseases. The large number of micronutrient deficiencies (2 billion people worldwide) (Initiative 2009) as well as the high prevalence of overweight and obesity (1.9 billion people worldwide) (World Health Organization 2018a) indicate that malnutrition is still a leading public health problem throughout the world. Moreover, the elderly population is growing, and prevalence of malnutrition is frequently observed among the elderly (Hickson 2006; Fávaro-Moreira *et al.* 2016). Ageing is known to cause declines in sensory functions, changes in salivary flow and composition, tooth loss and/or reduced jaw muscle strength, which in turn alter eating capability. Such a decrease in eating capability leads to reduced food intake, and consequently an increased risk of malnutrition (Fontijn-Tekamp *et al.* 2004; Ikebe *et al.* 2012; Vandenberghe-Descamps *et al.* 2016). Practical approaches to increase food and nutrient intake are therefore required to ensure healthy ageing.

Understanding which factors contribute to food intake (*i.e.* when, what, how much do people eat) and consumer perception (*i.e.* why do people eat/like what they eat) are of utmost importance for public health. Controlling food intake while maintaining sensory appreciation is still a challenge. Strategies to either increase healthy food intake or decrease total energy intake while retaining positive sensory characteristics are desired.

Food structure is known to affect both food intake (Robinson *et al.* 2014) and sensory perception (Devezeaux de Lavergne *et al.* 2017) via food oral processing behavior (Figure 1.1). Firstly, the field of food oral processing behavior is introduced (section 1.1), after which the influence on intake and perception is explained (section 1.3, section 1.4).

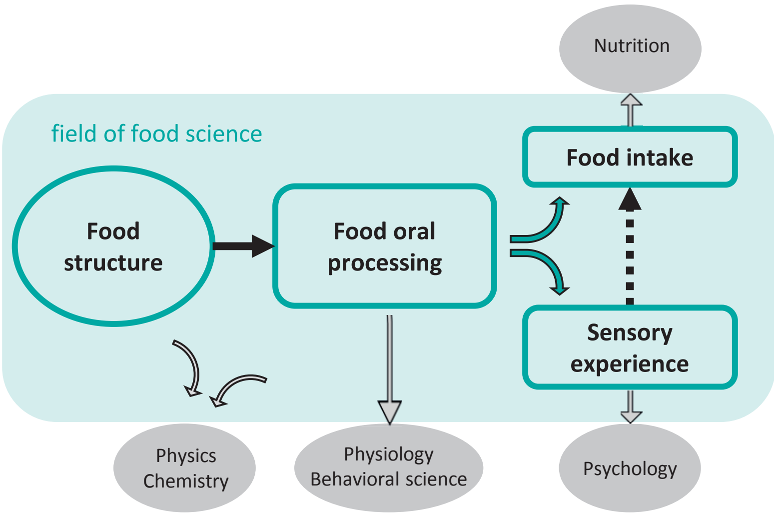

Figure 1.1: A multi-disciplinary approach highlighting the role of food oral processing in relation to food structure, food intake and sensory perception. The different aspects of food science (in green) relate to other disciplines including physics, chemistry, physiology, behavioral science, psychology and nutrition (in gray).

1.1 Food oral processing behavior

The field of food oral processing covers two main aspects: the way people eat foods (*i.e.* eating behavior) and the way the food structure changes in the mouth upon consumption (*i.e.* bolus properties). From the start of consumption, food structure generally decreases and food lubrication increases with mastication time, the end product being the food bolus. Food breakdown can be obtained by chewing, tongue movements, dilution with saliva or enzymatic breakdown by salivary enzymes. Chewing is the main process in the breakdown of solid foods, whereas tongue movements are most relevant for liquid foods, semi-solid foods and bolus transport. Increased food lubrication can be reached by saliva incorporation or moisture release from the food matrix (Witt & Stokes 2015; Mosca & Chen 2017). This process is schematically illustrated in Figure 1.2.

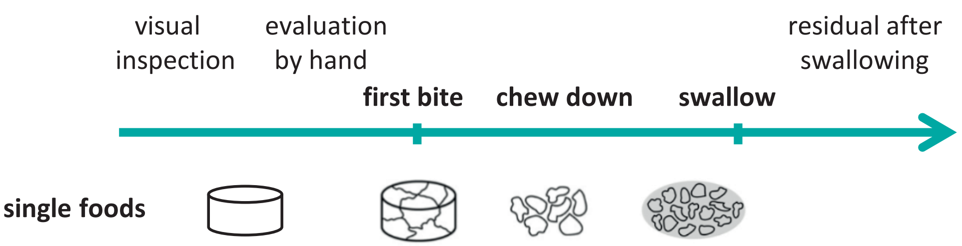

Figure 1.2: Schematic representation of different stages of food oral processing behavior of foods, showing structural breakdown and lubrication by saliva (in gray). Figure is adopted from Stieger and Van de Velde (2013).

The importance of food oral processing was already introduced in 1988 by Hutchings and Lillford (Hutchings & Lillford 1988). Their model visualizes the breakdown paths of a range of foods, indicating the effect of "time in mouth" on "the degree of structure" and "the degree of lubrication" of the foods. Hutchings and Lillford postulated that safe swallowing requires the food structure to be reduced below a certain level and food lubrication to be increased above a certain level.

1.2 Impact of food structure on food oral processing behavior

Food structure is known to influence food oral processing behavior. Recently, oral processing behavior of liquid (drinkable), semi-solid (spoonable) and solid (chewable) foods was regulated by their respective rheological and mechanical properties (Aguayo-Mendoza *et al.* 2019). For liquid and semi-solid foods, consumption time increased with increasing consistency. For solid foods, consumption time increased with increasing Young's modulus. Likewise, harder solid foods are known to be chewed for a longer time than softer foods (Mioche *et al.* 2003; Yven *et al.* 2005; Çakir *et al.* 2012b; Koc *et al.* 2013; Panouillé *et al.* 2014; Devezeaux de Lavergne *et al.* 2015a; Devezeaux de Lavergne *et al.* 2015b; Devezeaux de Lavergne *et al.* 2016a; Jourdren *et al.* 2016; Young *et al.* 2016a; Gao *et al.* 2017; Krop *et al.* 2018). In addition, increasing elasticity or dryness of foods has been observed to prolong consumption time (Wee *et al.* 2018).

1.3 Impact of food structure and food oral processing behavior on food intake

Food intake has recently been linked to food structure and food oral processing behavior (de Graaf & Kok 2010; de Graaf 2012; Robinson *et al.* 2014; McCrickerd & Forde 2017). Foods that require longer time in the mouth before swallowing (*i.e.* slow eating rate in g/min) have been associated with higher expected satiation (Forde *et al.* 2013b), higher fullness perception (Zhu *et al.* 2013; Ferriday *et al.* 2016) and reduced *ad libitum* food intake (de Wijk *et al.* 2008; Weijzen *et al.* 2009; Zijlstra *et al.* 2009; Bolhuis *et al.* 2011; Forde *et al.* 2013a; Bolhuis *et al.* 2014a; Fogel *et al.* 2017; Forde *et al.* 2017; Lasschuijt *et al.* 2017; McCrickerd *et al.* 2017). As an example, systematically increasing viscosity of liquids/semi-solids (de Wijk *et al.* 2008; Zijlstra *et al.* 2008; McCrickerd *et al.* 2017) and increasing hardness of solids (Bolhuis *et al.* 2014a; Lasschuijt *et al.* 2017) increased *ad libitum* food intake. Thus, food structure determines eating rate (*i.e.* how long the food has to be masticated before swallowing) which influences food intake.

1.4 Impact of food structure and food oral processing behavior on sensory perception

Sensory perception is also influenced by food structure and food oral processing behavior (Devezeaux de Lavergne *et al.* 2017). For example, fracture properties of foods influence sensory perception of a bite (Çakir *et al.* 2012b; Koc *et al.* 2013; Devezeaux de Lavergne *et al.* 2015b; Campbell *et al.* 2016; Devezeaux de Lavergne *et al.* 2016a; Krop *et al.* 2018). More specifically, fracture stress and fracture strain correlated with firmness and brittleness perception, respectively (Devezeaux de Lavergne *et al.* 2017). In addition, bolus fragmentation has been related to creaminess or graininess perception. Dynamic changes in bolus properties have also been observed to influence flavor release and perception, but the underlying mechanisms are not completely understood yet (Salles *et al.* 2011; Feron & Salles 2018). Thus, food structure determines structural transformations of food throughout mastication which in turn influence sensory perception.

Changes in oral processing behavior are also known to impact sensory perception. For example, in the case of ice creams, chewing resulted in prolonged fruity and coldness sensations, whereas letting them melt in mouth resulted in prolonged sweetness and firmness sensations (Doyennette *et al.* 2019).

1.5 Multidisciplinary approach: Methodologies to assess food oral processing behavior, food intake and sensory perception

As explained above, food oral processing behavior bridges food structure, food intake and sensory perception (Figure 1.1). Linking food structure to eating behavior is necessary to better understand which food properties influence eating rate and thereby trigger intake (Robinson *et al.* 2014; McCrickerd & Forde 2017). Linking food structure to bolus properties is necessary to better understand the structural transformations of food during mastication that trigger sensory sensations (Chen 2014; Devezeaux de Lavergne *et al.* 2017; Foegeding *et al.* 2017). Methodological aspects need to be considered when designing and interpreting multi-disciplinary studies, as outcomes depend on the methodology used. Commonly applied methodologies will be further discussed.

Methodologies to assess food and bolus properties

Many different methodologies have been developed to characterize food properties before and during consumption, since food structure varies considerably between foods and changes strongly with mastication time. The majority of methodologies are the same for food before consumption and the respective food bolus. In case of food properties before consumption, composition, rheological or mechanical properties, lubrication properties and microstructure are frequently studied (van der Linden 2012). With regard to bolus properties, composition and bolus hydration, rheological or mechanical properties, lubrication properties and particle size are frequently studied, as these have been suggested to play a key role in swallowing and sensory perception (Panouillé *et al.* 2016; Devezeaux de Lavergne *et al.* 2017). The specific method used to characterize mechanical and lubrication properties depends on the consistency and heterogeneity of the food. Particle size analysis is a measure of food comminution, which is determined for solid foods only.

Boli are generally collected by instructing panelists to expectorate food (*ex situ* bolus collection; “chew, expectorate and analyze” approach) just before swallowing. To study the breakdown kinetics in mouth, boli can also be collected at certain time points throughout mastication (Panouillé *et al.* 2016).

Methodologies to assess oral processing behavior

Video recordings, electromyography (EMG), electromyography combined with jaw-tracking (EMG-JT), videofluorography and ultrasonic echo-sonography have been used to assess eating behavior (Boyar & Kilcast 1986; Mioche *et al.* 2002; Casas *et al.* 2003; Hennequin *et al.* 2005; Vinyard & Fiszman 2016). Among these methods, videofluorography and ultrasonic echo-sonography have been used sporadically to record tongue and soft-tissue movements, whereas video recordings, EMG and EMG-JT are commonly used to study the relationship between food properties and eating behavior.

The recording of videos is an easy, non-invasive method to assess how different foods affect the consumption process (*e.g.* the number of chews, chewing time, chewing frequency, number of swallows, eating rate). No specialized equipment and little training for data collection and analysis are required, which are advantages of using video recordings (Hennequin *et al.* 2005; Wilson *et al.* 2013). However, the disadvantage of video recording is that no information can be collected with respect to muscle activities, chewing movements and chewing velocities. EMG and EMG-JT provide opportunities to measure these parameters. However, these techniques require specialized equipment and some training for the researcher. Despite the need for specialized equipment and expertise, EMG-JT methodology is extremely useful for understanding details of the chewing process. In particular, it provides information on physiological events elicited during oral processing (Hennequin *et al.* 2005; Vinyard & Fiszman 2016).

Methodologies to assess food intake

When assessing food intake behavior, the distinction between satiation and satiety needs to be considered (Blundell 1979; Forde 2018b). Satiation (sometimes also called intra-meal satiety) develops throughout the course of eating, and refers to the process that ends an eating episode such as a meal or snack (*e.g.* increased fullness, decreased desire to eat). Satiation is usually assessed by the amount of food consumed *ad libitum* (total weight, volume, energy) and appetite ratings upon meal termination. On the other hand, satiety (sometimes also called inter-meal or post-ingestive satiety) refers to the inhibition of further eating and relates to between consumption moments. Satiety is usually assessed by the duration of hunger suppression or the amount of food consumed during a subsequent meal. In such pre-load studies, participants consume a fixed amount of food, after which they score their appetite over a period of time and/or *ad libitum* intake of the next meal is determined. In addition, consumer expectations of fullness (expected satiation) and hunger suppression (expected satiety) delivered by foods have been quantified to better understand differences in food choice and intake behavior (Fiszman & Varela 2013; Forde *et al.* 2015).

Methodologies to assess sensory perception

Among sensory methods, one can distinguish between discrimination tests, descriptive analyses, rapid sensory methods and temporal methods. Discrimination tests such as same-different, paired comparison, duo-trio, triangle, tetrad or 3-AFC tests investigate whether any perceptible differences exist between two food products (Lawless & Heymann 2010). Descriptive sensory analyses such as Quantitative Descriptive Analysis (QDA) (Stone *et al.* 2008) are used to investigate how food products differ in specific sensory characteristics (Lawless & Heymann 2010; Stone *et al.* 2012). Although QDA has been used as the "golden standard" method for years, two major drawbacks of this method are that intensive panel training is required as well as that dynamic sensory changes are not necessarily captured (Murray *et al.* 2001). Therefore, the use of rapid sensory methods such as napping, Check-All-That-Apply (CATA) and Rate-All-That-Apply (RATA) have increased over the past years (Ares & Varela 2014; Ares & Varela 2018). Rapid sensory methods use either semi-trained panelists or naïve consumers. Research has shown that similar outcomes are obtained when comparing results of conventional descriptive analysis with trained panelists and rapid methods with untrained panelists (Dehlholm *et al.* 2012; Fiszman & Varela 2013; Oppermann *et al.* 2017; Liu *et al.* 2018; Aguiar *et al.* 2019). These studies show that both methods can provide very similar information, and that trained panels are not always necessary. As the above-mentioned sensory methods are static methods (overall or averaged perception), these methods do not provide any information on how perception changes over time. To obtain such information, temporal methods such as Temporal Dominance of Sensations (TDS), Temporal Check-All-That-Apply (TCATA), Time-Intensity and Progressive Profiling are often used (Pineau *et al.* 2009; Cadena *et al.* 2014; Pineau & Schlich 2015; Castura *et al.* 2018; Nguyen *et al.* 2018; Berget *et al.* 2020). Temporal methodologies are frequently used as complementary assessment to obtain a complete sensory profile. Sensory perception should be captured dynamically, as bolus properties change considerably during consumption by which texture and flavor sensations are triggered (see section 1.4). In this context, sensory trajectories obtained with TDS have been linked to changes in bolus properties over time (Saint-Eve *et al.* 2015; Devezeaux de Lavergne *et al.* 2016b), which confirms the added value of dynamic sensory evaluations.

1.6 Influence of environmental and individual variables on consumption behavior

Not only food structure, but also environmental and individual variables affect food oral processing behavior, intake and sensory perception (Figure 1.3). In this thesis, food properties were systematically varied (presented in green), whereas environmental and individual factors were kept constant as much as possible and are considered to be outside the scope of this thesis.

Environmental factors include location (laboratory setting vs. real-life, institution vs. restaurant) (Edwards *et al.* 2003; De Wijk *et al.* 2019); ambiance (cutlery, plate size and shape, music, ambient temperature, lighting) (McElrea & Standing 1992; Stroebele & de Castro 2006; Oberfeld *et al.* 2009; Sester *et al.* 2013; Reinoso Carvalho *et al.* 2015; Bolhuis & Keast 2016;

Motoki *et al.* 2018), social environment (eating alone vs. eating with others) (Hetherington *et al.* 2006; Hermans *et al.* 2012); distraction (eating while watching television) (Bellisle *et al.* 2004; Blass *et al.* 2006; Hetherington *et al.* 2006); portion size (Diliberti *et al.* 2004; Kral & Rolls 2004; Rolls *et al.* 2004; Rolls *et al.* 2006); packaging, labels and claims (Grunert & Aachmann 2016; Skaczkowski *et al.* 2016); and price. Throughout this thesis, all tests were performed in a sensory laboratory setting to standardize the environmental influence on the outcome.

Large differences in food oral processing behavior, intake and sensory perception have been observed between individuals (Næs *et al.* 2018). Such inter-individual variations have been attributed to gender (Nagasawa *et al.* 1997; Woda *et al.* 2006; Ketel *et al.* 2019); ageing (Kohyama *et al.* 2002; Mioche *et al.* 2004; Ketel *et al.* 2019); cross-cultural differences (Rozin 1988; Prescott & Bell 1995; Kim *et al.* 2018; Ketel *et al.* 2019; Pedrotti *et al.* 2019; Ketel *et al.* 2020); physiological differences such as masticatory ability, saliva characteristics and velum opening (Buettner *et al.* 2002; Engelen & Van Der Bilt 2008; Repoux *et al.* 2012; Mosca & Chen 2017; Mosca *et al.* 2019a; Ketel *et al.* 2020); different eating strategies (slow vs. fast chewers) (Yven *et al.* 2012; Robinson *et al.* 2014; Devezeaux de Lavergne *et al.* 2015a; McCrickerd & Forde 2017); familiarity and expectations (Santagiuliana 2019); or combinations of these factors. Throughout this thesis, we have worked with young, European, Caucasian adults that frequently consumed the test foods .

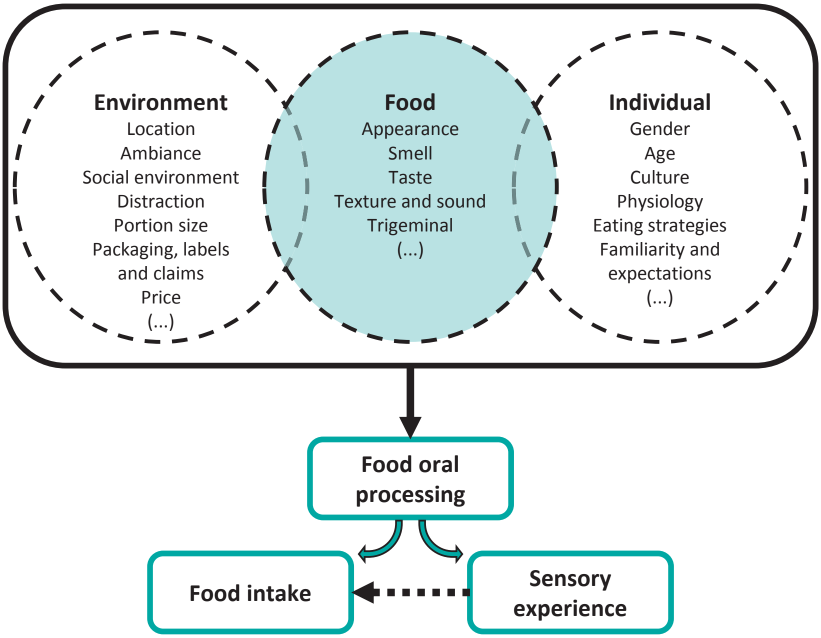

Figure 1.3: Representation of different variables (environment, food, individual) that are known to affect oral processing behavior, intake and sensory perception of foods (adapted from Dacremont and Sester (2019) and Devezeaux de Lavergne *et al.* (2017). In this thesis, food properties are systematically varied (presented in green), while the environmental and individual factors are kept constant.

1.7 What are composite foods?

In everyday life, consumers frequently combine foods with different composition and properties within a meal or within one bite. For example, bread is often combined with spread and/or cheese, yogurt with fruits and/or granola or various vegetables combined with dressing into a salad. Throughout this thesis, the term composite foods refers to foods that are composed of two single foods, *i.e.* one solid carrier food (*e.g.* bread, a vegetable) combined with one condiment (*e.g.* mayonnaise, cheese spread or dip). Yet, composite foods have received surprisingly little attention in the field of food science.

To better understand how single foods contribute to oral processing behavior of composite foods, food oral processing behavior of bi-layer model gels with contrasting mechanical properties was studied (Devezeaux de Lavergne *et al.* 2016b). Both layers contributed to oral processing behavior of the composite food gels, but the hard gel layers were found to influence oral processing behavior slightly more than soft gel layers. Likewise, several studies have been performed on commercially available foods. Addition of butter was observed to reduce the number of chewing cycles and total consumption time of dry carriers such as toast and cake (Gavião *et al.* 2004; Engelen *et al.* 2005). Addition of solid food particles (nuts, fruit pieces) to semi-solid yogurt increased the number of chews and total consumption time (Aguayo-Mendoza *et al.* ; Prinz & Lucas 1995; Tarrega *et al.* 2016; Morell *et al.* 2018). Thus, oral processing behavior of foods is affected by the presence of other foods, but little is known about the underlying mechanisms.

Also in terms of energy intake of composite foods, there is little knowledge on how specific single food properties determine food intake. Although food properties impact intake of single foods (see section 1.3), whether such changes in single food properties also influence intake of composite foods is less well-understood. Only few intake studies have been performed with composite foods with systematically varied food properties. For example, Bolhuis *et al.* (2014) assessed intake of hamburgers (bread, meat, tomato, ketchup) and rice salads (rice, vegetables) varying in hardness (hard/soft bread, raw/cooked vegetables). Harder composite foods led to slower eating rate and a lower energy intake than the soft versions (Bolhuis *et al.* 2014a). Mosca *et al.* (2019) assessed intake of yogurt with granola while varying granola particle size (large/small). Smaller granola particles led to slower eating rate and a lower intake than the larger particles (Mosca *et al.* 2019b). Apparently, single food properties can be used to modify eating behavior and *ad libitum* intake of composite foods. Sensory perception of composite foods is known to be different from that of single foods. From the start of consumption, consumers are exposed to the different structures and flavors present in composite foods, which increases sensory complexity perception (Paulsen *et al.* 2012; Tang *et al.* 2017; Santagiuliana *et al.* 2018a; Santagiuliana *et al.* 2018b). Upon mastication, the different foods are mixed in the mouth, leading to continuous transitions of the food structures present. Composite foods are generally highly liked by consumers, which is presumably related to intra-oral sensory variety perceived throughout consumption (Szczesniak & Kahn 1984; Hyde & Witherly 1993; Miele *et al.* 2010).

1.8 Rationale and thesis outline

Little is known about the mechanisms underlying oral processing behavior, food intake and sensory perception of composite foods, although this is the most common consumption context. In particular, a systematic understanding of how different single food properties contribute to oral processing behavior and consequently to intake and sensory perception of composite foods is still lacking. The research described in this thesis aims to investigate the effects of carrier and condiment properties on food oral processing behavior, food intake and sensory perception of composite foods (Figure 1.4). **Chapters 2 – 5** aim to determine the role of single food properties in food oral processing and intake of composite foods. **Chapters 2, 3, 6 and 7** aim to determine the role of single food properties in sensory perception of composite foods. By systematically varying single food properties, this thesis provides new insights into the mechanisms underlying food oral processing of composite foods, which enables to gain a better understanding of the structural transitions of foods that contribute to intake and/or perception of composite foods.

The aim of the first study was to investigate the effect of condiment addition on eating behavior, bolus formation and dynamic sensory perception of carrier foods (**Chapter 2**). Bread and crackers were combined with firm cheese, cheese spread and mayonnaise into composite foods (carrier food with condiment). Eating behavior, bolus properties at 33, 66 and 100% of total mastication time and dynamic sensory perception were examined for carriers foods without and with different condiments.

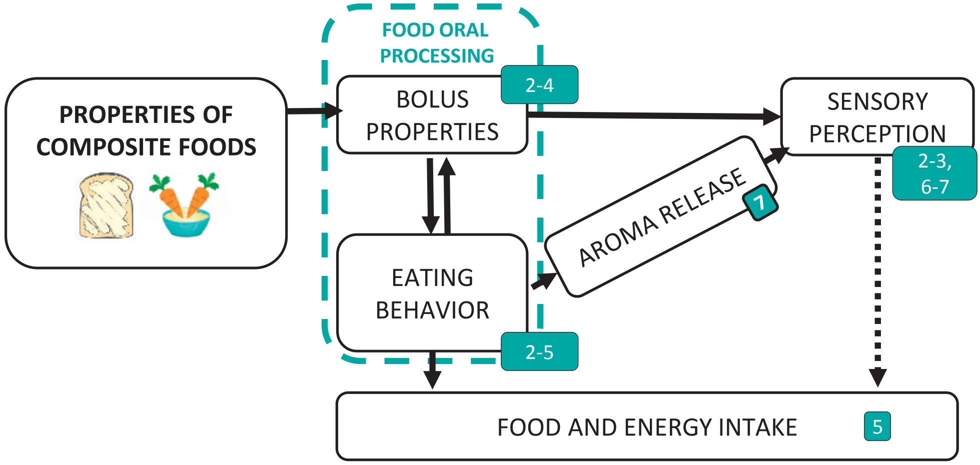

Figure 1.4: Schematic overview of the framework of this thesis. The numbers indicate the chapters in which the results are described. Sensory perception has an influence on intake, but this direct relation was not studied in this thesis and is therefore represented by a dotted arrow.

Chapters 3 and 4 aimed to investigate the influence of single food properties on eating behavior and bolus formation of composite foods. Mayonnaises systematically varying in fat content (full fat, low fat) or viscosity (thick, thin) were prepared to better understand the mechanisms by which condiments facilitate bolus formation of carrier foods. The different mayonnaises were combined with a range of carrier foods including fresh bread (**Chapter 3**), cooked potato (**Chapter 3**) and raw carrots (**Chapter 4**) to obtain knowledge across food categories and the effect of carrier water absorption capacity. Eating behavior and bolus properties were examined for carrier foods without and with different condiments.

As foods are available in a broad range of shapes and sizes, **Chapter 4** assessed eating behavior of raw carrots varying in shape (cube, julienne) and number/size of particles with similar total weight. A selection of these carrots varying in shape were combined with mayonnaise to validate whether the effect of food shape on eating behavior persists when assessed as a composite food. Eating behavior and bolus properties were examined for carrots without and with mayonnaise. **Chapter 5** aimed to investigate the role of cracker shape on eating behavior and *ad libitum* intake of crackers with cheese dip. In this study, crackers varying in shape (flat squares, finger-shape sticks) were combined with cheese dip varying in viscosity (thick, thin).

Single food properties were assumed to play a large role in sensory perception of composite foods, and this was investigated in **Chapters 3, 6 and 7**. In **Chapter 3**, fresh bread and cooked potato were combined with mayonnaises varying in fat content (full fat, low fat) and viscosity (thick, thin), and sensory perception was assessed by descriptive sensory analysis with a trained panel. In **Chapter 6**, carrier foods (bread, carrot) varying in hardness (soft, medium, hard) were combined with mayonnaises varying in fat content (full fat, low fat) or viscosity (thick, medium, thin). Dynamic sensory perception was assessed by naïve consumers using Temporal Dominance of Sensations (TDS) to investigate how consumers shift their attention from one to the other food throughout a bite. Secondly, static sensory perception was assessed by naïve consumers using the Rate-All-That-Apply (RATA) methodology to investigate the influence of one food on the sensory profile of the other food. The experiment in **Chapter 7** was designed to unravel the mechanisms by which single foods affect the sensory properties of composite foods. In-nose aroma release and dynamic aroma intensity perception were assessed simultaneously for mayonnaises without and with different carrier foods (bread, cooked potato) varying in hardness (soft, hard).

Finally, **Chapter 8** provides a general, integrated discussion of all studies and reflects on practical implications related to composite foods. Methodological considerations, suggestions for future research and main conclusions are also provided.

1

MAYONNAISE
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2

Oral processing behavior and dynamic sensory perception of composite foods: Toppings assist saliva in bolus formation

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Food Quality and Preference, 2019, 71: 497-509

ABSTRACT

Background and objective: Composite foods consist of combinations of single foods, such as bread with toppings. Single foods can differ considerably in their mechanical and sensory properties. This study aimed to investigate the effect of toppings on oral processing behavior and dynamic sensory perception of carrier foods when consumed as composite foods.

Methods: Two carriers (bread, crackers) and three toppings (firm cheese, cheese spread, mayonnaise) were selected and six carrier-topping combinations were prepared. Mastication behavior, bolus properties (33, 66 and 100% of total mastication time) and dynamic sensory perception were determined for single carriers and all carrier-topping combinations.

Results: Both carriers with cheese spread and mayonnaise were chewed shorter and with fewer chews than single bread and crackers, although twice the mass of food was consumed. These toppings contributed to a faster bolus formation by providing moisture, so that less saliva was incorporated into the bolus during mastication. As a result of the moisture incorporation, carrier boli with toppings were softened and perceived less firm and less dry than carrier boli alone. The largest effects of toppings on oral processing behavior and perception were found for liquid-like mayonnaise, and these effects were more pronounced in dry crackers than in moist bread.

Conclusions: We conclude that toppings assist saliva in bolus formation of carriers. Carriers drive oral processing behavior and texture perception whereas toppings drive overall flavor perception. This knowledge contributes to food design tailored for specific consumer segments and future personalized nutrition.

Key words: composite foods, carrier foods, toppings, oral processing, bolus properties, sensory perception

2.1 INTRODUCTION

Many foods that are frequently consumed are composed of combinations of multiple single foods such as bread with cheese, crackers with spreads or yogurt with cereals. Throughout this paper, the term composite foods will refer to foods that are composed of two single foods. The single foods can differ considerably in composition, mechanical properties and sensory characteristics (Szczesniak & Kahn 1984; Scholten 2017).

Oral processing behavior of composite foods is different from that of single foods. From the start of consumption, two single foods are mixed in the mouth, which results in complex oral processing behavior depending on the properties of both food components. Few studies investigated the impact of composite foods on oral processing behavior. One of those studies was performed by Hutchings *et al.* (2011), who studied oral processing behavior of gelatin and chocolate matrices with embedded peanuts. They found that the mastication behavior (chewing duration, number of chews, chewing frequency) was influenced by the type of matrix, and this was not altered by the type of peanuts (dry or moist) embedded in the matrix. The matrix was shown to influence breakdown of the embedded peanuts, as obvious from a variation in peanut particles size distributions. Larsen *et al.* (2015) found that the release of embedded inclusions from a gel matrix ensured differences in breakdown pathways. While these two studies focused on matrices with embedded components, Devezeaux de Lavergne *et al.* (2016b) investigated bi-layer model gels displaying mechanical contrast by combining two gel layers with different mechanical properties. Oral processing behavior of such gels showed oral processing characteristics between that of the two separate layers indicating the importance of the properties of both gel layers present. However, hard gel layers were found to influence oral processing behavior slightly more than soft gel layers. Only one research group investigated mastication behavior of commercially available composite foods (Gavião *et al.* 2004; Engelen *et al.* 2005). They investigated the effect of spreading butter on different carrier foods, such as bread, cake and toast. They found a significant difference in mastication behavior between carrier foods alone and carriers with butter. Addition of butter decreased the number of chewing cycles of the drier carriers, such as toast and cake, but not for the more moist bread (Engelen *et al.* 2005). The researchers hypothesized that butter facilitated the lubrication and bolus formation of carriers and thereby reducing the number of chewing cycles. However, no information is currently available on the mechanisms underlying bolus formation in such foods and its influence on mastication behavior.

Few studies have attempted to investigate the sensory perception of composite foods. Adding sauces to carrier foods (vegetables with gravy, salmon with culinary sauce) decreased the perceived intensity scores of such carrier foods (Meinert *et al.* 2011; Paulsen *et al.* 2012). In another study, the impact of different carriers (chicken broth, white rice and grilled chicken) on soy sauce perception was investigated (Cherdchu & Chambers 2014). They found that the carriers did not strongly affect the classification pattern of soy sauces, and differences between soy sauces were still observed. However, solid white rice and grilled chicken tended to modify the sensory properties of the soy sauce more than liquid chicken broth. In another study, dynamic texture perception was shown to increase in complexity (*i.e.* the number of dominant attributes increased) when model gel matrices contained two or more embedded inclusions (Tang *et al.* 2017). In the case of model bi-layer gels, dynamic sensory perception corresponded to an average of the two single layers they were composed of (Devezeaux de Lavergne *et al.* 2016b), while for breads with a crust, the harder crust dominated the dynamic texture perception (Gao *et al.* 2017).

Oral processing behavior is relevant because it imparts structural changes to the food, thereby impacting dynamic sensory perception, oro-sensory exposure time, satiation and food intake (Chen 2009; Chen 2015; Campbell *et al.* 2017b; Devezeaux de Lavergne *et al.* 2017; Wang & Chen 2017; Forde 2018a). Oral processing behavior and dynamic sensory perception have been extensively studied for model foods and various single foods such as bread (Koc *et al.* 2013; Panouillé *et al.* 2014; Witt & Stokes 2015; Jourdren *et al.* 2016; Le Bleis *et al.* 2016; Devezeaux de Lavergne *et al.* 2017; Gao *et al.* 2017), but little is known about oral processing behavior and sensory perception of composite foods, although these are often consumed. A multidisciplinary approach integrating food structure, oral processing behavior and dynamic sensory perception for composite foods may help to gain further understanding of how composite foods are perceived and which factors determine their perception. An understanding of the role of each single food may be used to control mastication behavior, bolus formation, sensory perception or liking of foods. Such insights are of relevance for food design tailored to specific consumer segments such as the increasing elderly population or people with decreased eating capabilities (Laguna & Chen 2016; Mosca & Chen 2016).

In this study, carriers (bread and crackers) and toppings (firm cheese, cheese spread and mayonnaise) were used as single foods, and they were combined into composite foods (carrier with topping). The aim of this study was to investigate the effect of toppings on oral processing behavior and dynamic sensory perception of these composite foods. Both carriers and toppings are expected to influence the oral processing behavior and dynamic sensory perception, however we hypothesize that carriers are the main determinant of oral processing behavior and texture perception since they require oral breakdown before being swallowed safely.

2.2 MATERIALS AND METHODS

2.2.1 Samples

Carrier foods and toppings were combined to form composite foods. Two commercial carriers were used, namely bread crumb (toast bread, Jacquet®, France) and crackers (mini-toast naturel, Haust®, The Netherlands), and three commercial toppings, namely firm cheese (AH Goudse jong belegen, Albert Heijn, The Netherlands), cheese spread (Kiri® mit Sahne, BEL Group, France) and low-fat mayonnaise (Licht en Romig, Calvé®, Unilever, The Netherlands). These foods were selected based on their similar fat content and difference in mechanical properties. The carriers (2) were combined with the toppings (3) to create 6 combinations. The single carriers and toppings (2 carriers and 3 toppings) were included as a reference, which gave a total of 11 samples.

Table 2.1 presents an overview of the composition and product properties of the single foods. The fat content was taken from the nutritional information on the product label. The moisture content of the single foods was determined gravimetrically (16 – 18 h at 105°C) in five replicates. This method was also used for the expectorated boli, and is explained in more detail in section 2.2.5. The water activity of the single foods was determined in three replicates using a LabMaster aw (Novasina®). The mechanical properties first peak force (associated to firmness), adhesiveness and cohesiveness were determined in 9 replicates using two cycle puncture tests with a Texture Analyzer (TA.XT Plus). This method was also used for the expectorated boli, and is explained in more detail in section 2.2.5.

The carriers were served at constant weight of 2.1 g, which was based on the weight of the single cracker. In addition, the dimensions of bread (35 x 35 x 8 mm) and crackers (35 x 35 x 8 mm) were comparable. A feasibility test was performed with 6 consumers to quantify a natural carrier-topping serving ratio. It was observed that consumers showed a natural eating behavior when carriers and toppings were combined in a 1:1 weight ratio. All three toppings were therefore also served at a constant weight of 2.1 g. This gave a total weight of 4.2 g for carrier-topping combinations (2.1 g carrier and 2.1 g topping). The toppings covered the carriers completely. Carrier-topping combinations were prepared just before serving in order to prevent moisture transfer from the toppings into the carriers.

2.2.2 Subjects

48 healthy subjects were initially recruited, out of which 18 subjects (7 male and 11 female) were selected to participate (25.6 ± 2.93 years, mean ± SD). Inclusion criteria were good dental health, no missing teeth and/or molars except wisdom teeth, no dental braces, no piercings in the mouth, no swallowing or mastication disorders and non-smoking habits. Only Caucasian adults were included in our study. Selection criteria assessed during the screening session were mechanically stimulated saliva flow rate, mastication time and liking of the carriers, toppings and composite foods. Subjects with low/high saliva flow rates (top

Table 2.1: Overview of composition (fat and moisture content), physical-chemical properties (water activity, first peak force, adhesiveness, cohesiveness, texture category) and serving size of single foods (2 carriers, 3 toppings). Mean values ± standard error of the mean are given.

Single foods	Fat content* (wt%)	Moisture content (wt%)	Water activity	First peak force** (N)	Adhesiveness** (g·s)	Cohesiveness**	Texture category	Serving size (g)
Carriers								
Bread	4	34.4 ± 1.0	0.91 ± 0.002	0.92 ± 0.08	-1.9 ± 4.9	0.77 ± 0.06	Soft solid	2.1
Cracker	4	2.7 ± 0.2	0.21 ± 0.015	16.92 ± 2.89	-7.7 ± 9.4	0.06 ± 0.03	Hard solid	2.1
Toppings								
Firm cheese	31	41.1 ± 0.5	0.96 ± 0.004	4.00 ± 0.05	-103.1 ± 21.9	0.75 ± 0.01	Soft solid	2.1
Cheese spread	30	57.0 ± 0.1	0.98 ± 0.001	1.11 ± 0.02	-221.9 ± 47.4	0.25 ± 0.02	Semi-solid	2.1
Mayonnaise	27	62.8 ± 0.2	0.97 ± 0.001	-	-	-	Plastic liquid	2.1

* The fat content was taken from the packages.
** The mechanical properties of mayonnaise were not analyzed since mayonnaise is a plastic liquid

and bottom 10%), fast/slow mastication times (top and bottom 10%) and who disliked the samples strongly (scores below 3 on 9 point hedonic scale) were excluded. Subjects gave written informed consent and received financial compensation for participation. The study protocol was submitted to the Medical Ethical Committee of Wageningen University (METC-WU) and exempted from ethical approval. All 18 subjects completed the study.

2.2.3 Experimental approach

Each subject attended 11 sessions of 45 minutes over a time period of three months. In the first two sessions, mastication behavior was characterized using video recordings. Subjects were instructed to chew each food as they would do naturally. These video recordings were used to determine the total mastication time for each sample by averaging the mastication times of all subjects and all replicates. Averaged total mastication times of bread without topping, with firm cheese, with cheese spread and with mayonnaise corresponded to 13.8±0.6 14.2±0.6 13.5±0.6 and 11.7±0.5 s (mean±SE), respectively. Averaged total mastication times of cracker without topping, with firm cheese, with cheese spread and with mayonnaise corresponded to 20.9±0.6 21.1±0.6 19.6±0.5 and 16.5±0.5 s (mean±SE), respectively. The third session was a training session, during which the sensory attributes and the procedures of the following sessions were introduced. Reference foods were used to acquaint the subjects with the different sensory attributes. The remaining eight sessions were used for bolus collection and sensory assessment. During these sessions, subjects were asked to expectorate the bolus for each sample after time points corresponding to 33, 66 and 100% of the averaged total mastication time using a stopwatch. In addition, subjects were asked to evaluate intensity of sensory attributes after one chew and at 33, 66 and 100% of total mastication time using progressive profiling method. Each method is explained in further detail in section 2.2.4 - 2.2.6.

For all sessions, samples were presented with three digit codes. Carriers and carrier-topping combinations were served on a plate, whereas the single toppings were served on a spoon. Subjects cleaned their palate after consumption of each sample with water and cucumber. Cucumber was used as palate cleanser instead of commonly used crackers, since using one of the samples as palate cleanser was not preferred. Cucumber was chosen because it is a relatively bland tasting solid food with high moisture content by which it could aid in the removal of cracker/bread/topping residues.

2.2.4 Characterization of mastication behavior using video recordings

Oral processing behavior of all samples by n=18 subjects was characterized in triplicate using video recordings. Four stickers were placed on the subjects face, which were later used as reference points during the video analyses. Two stickers with a defined distance of 5 cm were placed on the forehead, one sticker was placed on the nose and one sticker was placed on the chin. Subjects were seated in a chair with a camera in front of them. They were asked to chew each food as they would do naturally, while being video recorded. The subjects were instructed to maintain their head straight to the camera, and not to block their mouth or face with their hand while eating. Furthermore, they were instructed to indicate the moment of swallowing by raising their hand.

The videos were analyzed using Kinovea software (version 0.8.15), which is a free software that can be used to analyze mastication behavior. Each video was calibrated by setting the distance between the two stickers on the forehead as 5 cm. The path of the nose and chin sticker was tracked by the software in order to determine the chewing movements. All videos were analyzed separately by two of the authors, after which they compared their findings until agreement on the start of consumption and the moment of swallowing was reached for each video.

The parameters collected from the videos included total mastication time (s), number of chews, chewing frequency (chews/s), maximum vertical jaw movement (cm) and maximum lateral jaw movement (cm). Total mastication time was defined as the time period between the moment when the lips were closed after placing the food in the mouth and the moment of swallowing. Subjects generally take multiple swallows during mastication, but only the moment of the main swallow was used for analysis. Chewing frequency was calculated by dividing the number of chews by the mastication time. Maximum vertical and lateral jaw movements were defined as the maximum distance between the nose and chin in vertical and lateral directions, respectively. In addition, parameters describing oral processing behavior were also analyzed for tertiles of mastication time (*i.e.* 0 – 33%, 33 – 66% and 66 – 100% of mastication time) in order to analyze changes throughout mastication.

2.2.5 Characterization of bolus properties throughout mastication

Different bolus properties were analyzed at 33, 66 and 100% of total mastication time corresponding to early chew down, late chew down and the moment of swallowing, respectively. Boli were collected from n=18 subjects in quadruplicate; one replicate was photographed for illustration purposes, one replicate was used to determine the moisture and saliva content of the boli and two replicates were used to determine the mechanical properties of the boli. Within each replicate, the sample serving order was randomized with respect to sample type and expectoration time point. One replicate was presented in two different sessions, so that half of the samples was analyzed in the first session and the other half in the second session. All boli were analyzed for the different properties immediately after expectoration.

Images of expectorated boli

Pictures of expectorated boli were taken for all time points and all samples. The boli were photographed on a gray background using a digital camera (Canon IXUS 180). The camera was placed in a tripod at a distance of 40 cm above the bolus. One image was obtained for each bolus. These pictures were taken in order to illustrate differences between samples at different time points. They were not used for further image analysis.

Moisture and saliva content of expectorated boli

Boli were collected in petri dishes covered with lids and analyzed within an hour after expectoration in order to prevent moisture evaporation from the samples. 2 – 3 g of the expectorated boli were placed on aluminum dishes, weighed and dried for 16 – 18 h at 105°C in an atmospheric oven (Venti-line, VWR®). After drying, samples were cooled in a desiccator for 30 min and subsequently weighed. Bolus moisture content (MC) on a wet weight basis was calculated using $MC=(m_0-m_1)/m_0 \cdot 100\%$, where m0 is the weight of the sample before drying and m1 is the weight after drying. Bolus moisture content on a dry weight basis was calculated using $(MC_{db}= (m_0-m_1)/m_1$, which was subsequently used to calculate the saliva content (SC) per gram dry food by subtracting the moisture content on a dry weight basis of the product from the moisture content on a dry weight basis of the bolus (MC_{db}). The rate of saliva incorporation (g/min) was calculated by dividing the saliva content (SC) by mastication time. These calculations were based on the assumption that the bolus was fully expectorated. Few samples (8 out of 432 samples) resulted in slightly negative values for the saliva content, most likely due to small measurement errors. These samples were excluded from the statistical analyses.

Mechanical properties of expectorated boli

The mechanical properties of the expectorated boli were analyzed in duplicate using two cycle puncture tests with a Texture Analyzer (TA.XT Plus) fitted with a 500 g load cell. A cylindrical probe with a flat surface and a diameter of 4 mm was used. Punctures were performed up to a strain of 50% of the initial bolus height with a constant speed of 5 mm/s. The probe was then retrieved at the same speed of 5 mm/s and a resting time of 5 s was applied before a second puncture was performed. Three measurements, each at a different location of one expectorated bolus, were performed to obtain an averaged value. The mechanical properties of expectorated mayonnaise samples were not analyzed since these samples could not been detected by the Texture Analyzer due to their liquid behavior.

First peak force (associated to firmness), adhesiveness and cohesiveness were determined from the force-time curves as described before by Devezeaux de Lavergne *et al.* (2015b). First peak force was defined as the maximum peak force during the first puncture cycle. Adhesiveness was defined as the area under the negative force-time curve during the first cycle. Cohesiveness was defined as the ratio between the area under the (positive) force-time curve obtained during the second and first puncture cycle.

2.2.6 Characterization of dynamic sensory perception using progressive profiling

All samples were evaluated in duplicate for four sensory attributes (firmness, stickiness, dryness and flavor intensity) at four different moments of mastication (first chew, and 33, 66 and 100% of total mastication time) by n=18 subjects using progressive profiling. The attributes firmness (*i.e.* force required to push the tongue through the product to the upper palate), stickiness (*i.e.* degree to which the product adheres to any mouth surface while chewing) and dryness (*i.e.* dry and rough feeling on the tongue and oral cavity) were selected because they best describe the differences between the carriers and toppings and the expected changes thereof during oral processing. The attribute overall flavor intensity (*i.e.* the total amount of flavor) was included because the samples assessed differed strongly in flavor quality and to avoid dumping effects.

Subjects were asked to evaluate the sensory attributes immediately after expectoration. An unstructured 100 mm line scale anchored from "not at all" to "extremely" was used (Eye Question software, version 4.5.6). Sensory attributes were assessed according to a balanced design, so that 9 subjects started the sessions with the assessment of the attributes firmness and stickiness and the other 9 subjects started with the attributes dryness and flavor intensity.

2.2.7 Statistical data analyses

Results were reported as mean values with standard error (n=18 subjects). Outliers (Z-score>3.29) were removed from the data (typically less than 1.6% of all values). Data were checked for normality, and a log transformation was applied for the parameters peak force and adhesiveness in order to obtain normally distributed data. Linear mixed models were performed for all mastication, bolus and sensory parameters for single carriers and all carrier-topping combinations (lmerTest package)(RStudio, version 1.0.143). Significance level of $p<0.05$ was chosen. For the mastication parameters, carrier and topping were set as fixed effects, and subject and replicate were set as random effects. For the bolus and sensory parameters, carrier, topping and time were set as fixed effects, and subject and replicate were set as random effects. Then, a model including only the bolus and sensory data at 100% of mastication was carried out to study the differences in bolus and sensory properties between carriers with and without toppings at the moment of swallowing. For this analysis, carrier and topping were set as fixed effect, and subject and replicate were set as random effects. Interaction effects were included in all models, and random effects that were not significant were excluded from all models. The relationships between oral processing parameters throughout mastication of the different carriers with and without toppings were summarized using Principal Component Analysis (PCA) on subject averaged data (The Unscrambler X software, version 10.4.1).

2.3 RESULTS

2.3.1 Characterization of mastication behavior

For both carriers, mastication time and number of chews are summarized in Figure 2.1. Significant carrier effects (F=638.4, p<0.001; F=584.9, p<0.001), topping effects (F=44.0, p<0.001; F=26.9, p<0.001) and carrier-topping interactions (F=4.0, p=0.008; F=3.6, p=0.013) were found for both mastication time and number of chews (Table 2.2), indicating that the effects of toppings on carrier foods were not the same for bread and crackers. On average, crackers were processed longer in the mouth and with a higher number of chews compared to bread. For example, single crackers were masticated for 20.9±0.6 s with 27.7±0.8 chews, whereas single bread was masticated for 13.8±0.5 s with 17.8±0.8 chews. Addition of mayonnaise to bread significantly decreased total mastication time from 13.8±0.5 to 11.7±0.5 s (p<0.05), whereas no significant effects were found for addition of firm cheese and cheese spread. In the case of crackers, both mayonnaise and cheese spread had a significant effect on mastication time (p<0.05). The mastication time of crackers decreased from 20.9±0.6 to 19.6±0.5 s for cheese spread and to 16.5±0.5 s for mayonnaise. Addition of firm cheese to crackers did not significantly affect the total mastication time (21.1±0.6 s). Similar results were found for the number of chews required until swallowing. Addition of mayonnaise significantly decreased the number of chews required to swallow both bread and crackers, and cheese spread significantly decreased the number of chews for crackers only. Firm cheese did not lead to changes in number of chews for neither carriers. On average, similar effects of toppings on oral processing behavior of carriers were found for bread and crackers, but the effects were larger in crackers than in bread.

Chewing frequencies of the carriers with and without toppings varied between 1.25±0.02 and 1.36±0.03 chews/s (data not shown). Even though these differences in chewing frequencies were small, significant carrier effects (F=10.9, p=0.001) and topping effects (F=2.9, p=0.034) were found.

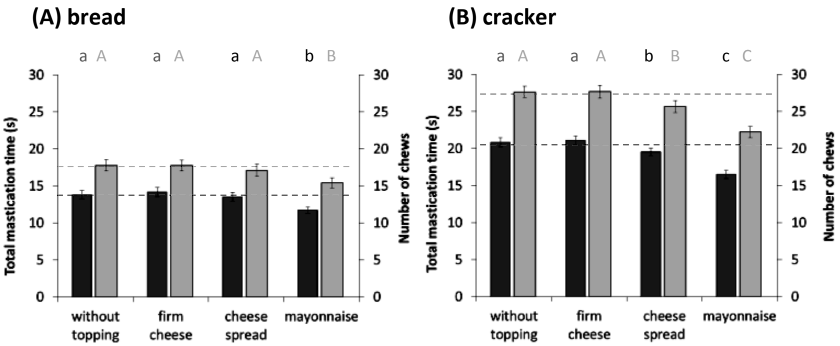

Figure 2.1: Total mastication time (dark gray) and number of chews (light gray) until swallowing for bread (A) and crackers (B) without topping and with different toppings (firm cheese, cheese spread and mayonnaise). Error bars represent standard error of the mean. Dashed lines represent averaged value of single carriers (n=18 subjects, in triplicate). Different letters indicate significant differences between means (p<0.05).

Jaw movements were recorded during mastication, and maximum vertical and lateral jaw movements were determined (data not shown). A significant carrier effect was found for both maximum vertical jaw movements ($F=16.4$, $p<0.001$) and maximum lateral jaw movements ($F=12.7$, $p<0.001$). The results show that jaw movements in both directions were larger for crackers than for bread. For example, the maximum vertical and lateral jaw movements of single bread were 19.7±0.6 and 19.1± 0.9 mm, whereas this was 22.4±0.8 and 20.3± 0.7 mm for single crackers. In case toppings were added, larger maximum vertical jaw movements were observed ($F=7.8$, $p<0.001$), but this effect was only significant after addition of firm cheese and cheese spread and not for mayonnaise. A significant topping effect was found for maximum lateral jaw movements ($F=3.8$, $p=0.011$), but none of the toppings affected the lateral jaw movements of single carriers significantly.

Besides characterization of total mastication behavior, the mastication parameters were also analyzed for tertiles of mastication time, *i.e.* early chew down (0 – 33% of total mastication time), middle chew down (33 – 66% of total mastication time), and late chew down (66 – 100% of total mastication time). Chewing frequency decreased towards the end of mastication. Averaged chewing frequency for all samples was 1.10±0.01 chews/s during late chew down, which was lower than during the earlier stages of mastication (1.28±0.01 chews/s for early chew down, 1.32±0.01 for middle chew down). In addition, maximum vertical jaw movements were largest during early chew down (19.4±0.2 mm), and decreased during middle and late chew down (16.4±0.2 and 16.4±0.2 mm). No differences in maximum lateral jaw movements throughout mastication were observed.

Table 2.2: Fixed effects of linear mixed models carried out for all mastication parameters, bolus properties and sensory properties of single carriers and carrier-topping combinations.

Measurement	Carrier			Topping			Time			Carrier:Topping			Carrier:Time			Topping:Time		
(A) Mastication behavior[a]	**F**	**p**		**F**	**p**					**F**	**p**							
Total mastication time (s)	638.4	<0.001	***	44.0	<0.001	***				4.0	0.008	**						
Number of chews until swallowing	584.9	<0.001	***	26.9	<0.001	***				3.6	0.013	*						
Chewing frequency (chews/s)	10.9	0.001	**	2.9	0.034	*				0.1	0.981	NS						
Maximum vertical jaw movement (mm)	16.3	<0.001	***	7.8	<0.001	***				1.7	0.158	NS						
Maximum lateral jaw movement (mm)	12.7	<0.001	***	3.8	0.011	*				0.2	0.906	NS						
	Carrier			**Topping**			**Time**			**Carrier:Topping**			**Carrier:Time**			**Topping:Time**		
(B) Bolus properties[b]	**F**	**p**		**F**	**p**		**F**	**p**		**F**	**p**		**F**	**p**		**F**	**p**	
Moisture content (wt%)	237.0	<0.001	***	41.3	<0.001	***	190.2	<0.001	***	0.2	0.9	NS	17.1	<0.001	***	2.3	0.033	*
Saliva content (g/g dry weight)	173.9	<0.001	***	54.6	<0.001	***	163.2	<0.001	***	3.1	0.026	*	7.4	<0.001	***	2.0	0.072	NS
Rate of saliva incorporation (g/min)	92.3	<0.001	***	4.3	0.006	**	15.8	<0.001	***	5.2	0.002	**	1.5	0.228	NS	2.5	0.024	*
Peak force (N)	55.7	<0.001	***	124.3	<0.001	***	123.6	<0.001	***	4.6	0.004	**	2.9	0.058	NS	1.1	0.336	NS
Adhesiveness (g·s)	99.0	<0.001	***	51.0	<0.001	***	108.6	<0.001	***	21.7	<0.001	***	54.8	<0.001	***	6.9	<0.001	***
Cohesiveness	298.5	<0.001	***	8.9	<0.001	***	3.5	0.03	*	6.4	<0.001	***	0.3	0.749	NS	2.2	0.037	*
	Carrier			**Topping**			**Time**			**Carrier:Topping**			**Carrier:Time**			**Topping:Time**		
(C) Sensory characteristics[b]	**F**	**p**		**F**	**p**		**F**	**p**		**F**	**p**		**F**	**p**		**F**	**p**	
Dryness	651.5	<0.001	***	115.3	<0.001	***	154.9	<0.001	***	0.2	0.925	NS	15.0	<0.001	***	1.9	0.054	NS
Firmness	245.7	<0.001	***	11.1	<0.001	***	160.6	<0.001	***	0.1	0.978	NS	30.9	<0.001	***	1.4	0.174	NS
Stickiness	21.4	<0.001	***	3.7	0.011	*	54.4	<0.001	***	0.8	0.472	NS	3.4	0.017	*	3.5	<0.001	***
Flavor intensity	30.2	<0.001	***	328.5	<0.001	***	75.0	<0.001	***	3.6	0.013	*	2.0	0.107	NS	6.1	<0.001	***

[a] F-values and p-values are derived from linear mixed models with carrier, topping and the interaction as fixed effect, and subjects and replicates as random effects.
[b] F-values and p-values are derived from linear mixed models with carrier, topping, time and the interactions as fixed effect, and subjects and replicates as random effects.
Significance is presented as NS (non-significant); * ($p<0.05$), ** ($p<0.01$), and *** ($p<0.001$).

Table 2.3: Fixed effects and descriptives (mean±SE) of bolus properties and sensory characteristics at moment of swallowing (t=100%) for bread and crackers with and without toppings, derived by linear mixed models.

	Carrier			Topping			Carrier:Topping		
(A) Bolus properties	**F**	**p**		**F**	**p**		**F**	**p**	
Moisture content (wt%)	55.2	<0.001	***	21.1	<0.001	***	0.2	0.865	NS
Saliva content (g/g dry weight)	96.4	<0.001	***	29.0	<0.001	***	1.4	0.258	NS
Rate of saliva incorporation (g/min)	45.7	<0.001	***	5.0	0.003	**	2.6	0.057	NS
Peak force (N)	32.6	<0.001	***	76.0	<0.001	***	0.6	0.634	NS
Adhesiveness (g·s)	0.0	0.835	NS	27.7	<0.001	***	3.8	0.010	*
Cohesiveness	134.8	<0.001	***	12.5	<0.001	***	1.9	0.125	NS
	Carrier			**Topping**			**Carrier:Topping**		
(B) Sensory characteristics	**F**	**p**		**F**	**p**		**F**	**p**	
Dryness	52.1	<0.001	***	25.3	<0.001	***	1.4	0.242	NS
Firmness	1.9	0.166	NS	10.2	<0.001	***	0.9	0.419	NS
Stickiness	0.4	0.553	NS	4.6	0.004	**	0.8	0.508	NS
Flavor intensity	14.9	<0.001	***	87.3	<0.001	***	1.7	0.174	NS

Table 2.3: Continued

	Bread (mean±SE)				Crackers (mean±SE)			
(A) Bolus properties	**without topping**	**firm cheese**	**cheese spread**	**mayonnaise**	**without topping**	**firm cheese**	**cheese spread**	**mayonnaise**
Moisture content (wt%)	55.0 ± 1.5 *b*	52.3 ± 0.9 *c*	55.1 ± 1.2 *b*	57.9 ± 1.1 *a*	50.4 ± 1.9 *b*	48.1 ± 1.2 *c*	51.2 ± 1.4 *b*	54.3 ± 1.5 *a*
Saliva content (g/g dry weight)	0.68 ± 0.06 *a*	0.50 ± 0.04 *b*	0.42 ± 0.07 *b*	0.46 ± 0.08 *b*	0.97 ± 0.06 *a*	0.66 ± 0.05 *b*	0.66 ± 0.07 *b*	0.74 ± 0.08 *b*
Rate of saliva incorporation (g/min)	4.5 ± 0.5 *b*	5.6 ± 0.4 *ab*	4.2 ± 0.7 *b*	5.1 ± 1.6 *a*	6.1 ± 0.6 *b*	6.2 ± 0.4 *ab*	5.9 ± 0.6 *b*	7.6 ± 0.8 *a*
Peak force (N)	0.49 ± 0.05 *b*	0.53 ± 0.03 *a*	0.49 ± 0.04 *ab*	0.19 ± 0.01 *c*	0.67 ± 0.08 *b*	0.69 ± 0.05 *a*	0.67 ± 0.06 *ab*	0.29 ± 0.03 *c*
Adhesiveness (g·s)	-22 ± 2 *b*	-26 ± 3 *ab*	-30 ± 3 *a*	-8 ± 1 *c*	-19 ± 2 *b*	-25 ± 3 *a*	-32 ± 4 *a*	-15 ± 3 *c*
Cohesiveness	0.46 ± 0.01 *c*	0.52 ± 0.01 *b*	0.53 ± 0.01 *ab*	0.52 ± 0.01 *a*	0.38 ± 0.01 *c*	0.39 ± 0.02 *b*	0.41 ± 0.02 *ab*	0.45 ± 0.02 *a*

	Bread (mean±SE)				Crackers (mean±SE)			
(B) Sensory characteristics	**without topping**	**firm cheese**	**cheese spread**	**mayonnaise**	**without topping**	**firm cheese**	**cheese spread**	**mayonnaise**
Dryness	45.3 ± 3.2 *a*	36.6 ± 3.0 *b*	28.4 ± 2.8 *c*	29.0 ± 3.3 *c*	62.3 ± 4.2 *a*	48.5 ± 3.8 *b*	44.6 ± 3.5 *c*	36.7 ± 3.3 *c*
Firmness	46.2 ± 2.6 *ab*	51.3 ± 3.4 *a*	45.5 ± 2.9 *b*	36.7 ± 3.0 *c*	51.5 ± 2.7 *ab*	50.3 ± 2.7 *a*	45.6 ± 3.2 *b*	41.2 ± 3.5 *c*
Stickiness	58.8 ± 2.7 *a*	62.2 ± 3.1 *a*	56.9 ± 3.6 *a*	48.7 ± 3.8 *b*	59.9 ± 3.5 *a*	59.3 ± 3.5 *a*	57.8 ± 3.6 *a*	54.1 ± 3.7 *b*
Flavor intensity	26.7 ± 2.9 *c*	56.8 ± 3.3 *b*	55.6 ± 2.8 *b*	68.9 ± 2.8 *a*	26.0 ± 3.1 *c*	50.4 ± 2.7 *b*	44.2 ± 3.4 *b*	60.9 ± 2.7 *a*

F-values and p-values are derived from linear mixed models with carrier, topping and the interaction as fixed effect, and subjects and replicates as random effects. Significance is presented as NS (non-significant); * ($p<0.05$), ** ($p<0.01$), and *** ($p<0.001$).
Different letters indicate significant differences between bread samples or cracker samples ($p<0.05$)

2

2.3.2 Characterization of bolus properties

Moisture content of boli and saliva incorporation

Figure 2.2 displays the moisture content as a function of mastication time for bread (Figure 2.2A) and crackers (Figure 2.2B). Bolus moisture content increased with mastication time, and more moisture was taken up at the beginning of mastication. More moisture uptake was observed for crackers compared to bread (carrier:time interaction, F=17.1, p<0.001). The moisture content of single crackers increased from 2.7±0.1 to 50.4±1.9% at the moment of swallowing, whereas for single bread it increased from 34.4±0.5 to 55.0±1.5%. When toppings were added to the carriers, the initial moisture content was larger than for the carriers alone, and moisture content increased to a lesser extent during mastication (carrier:topping interaction, F=2.3, p=0.033). Moisture contents at the moment of swallowing varied between 48.1±1.2 and 57.9±1.1 wt%, and a significant carrier effect (F=55.2, p<0.001) and topping effect (F=22.1, p<0.001) were observed (Table 2.3). On average, bread samples were swallowed at slightly higher moisture content than crackers. Carriers with firm cheese exhibited the lowest moisture content at the moment of swallowing, whereas this was the highest for carriers with mayonnaise.

Saliva content increased with increasing mastication time, and more saliva was incorporated at the beginning of mastication. Figure 2.3 shows the averaged saliva content at the moment of swallowing in bread (Figure 2.3A) and crackers (Figure 2.3B). A higher saliva content was found for crackers compared to bread (F=96.4, p<0.001), *e.g.* 0.97±0.06 g/g dry weight for single crackers and 0.68±0.06 g/g dry weight for single bread. In addition, a significant topping effect was found (F=29.0, p<0.001). All three toppings significantly decreased the amount of saliva incorporated in both bread and crackers. In bread, firm cheese, cheese spread and mayonnaise decreased the saliva content from 0.68±0.06 to 0.50±0.04, 0.42±0.07 and 0.46±0.07 g/g dry weight, respectively. In the case of crackers, the toppings decreased the saliva content from 0.97±0.06 to 0.66±0.05 g/g dry weight for firm cheese, to 0.66±0.07 g/g dry weight for cheese spread and to 0.74±0.08 g/g dry weight for mayonnaise.

Significantly higher rates of saliva incorporation were found for crackers than bread samples (F=45.7, p<0.001)(Table 2.3). As an example, the rate of saliva incorporation was 6.1±0.6 g/min for single crackers and 4.5±0.5 g/min for single bread. Furthermore, a significant topping effect was found (F=5.0, p=0.003), and only mayonnaise increased the rate of saliva incorporation of carriers significantly.

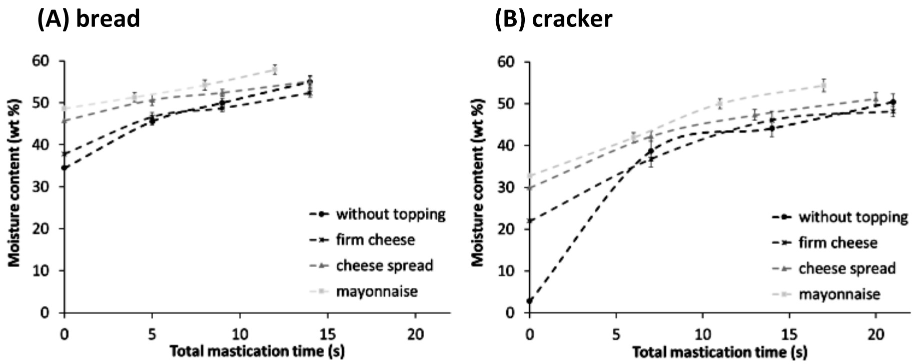

Figure 2.2: Bolus moisture content (wt%) for bread (A) and crackers (B) with and without toppings (n=18 subjects). Time points correspond to 33, 66 and 100% of total mastication time. The initial moisture content of samples is presented at t=0s. Error bars represent standard error of the mean. Dotted lines are added to guide the eye.

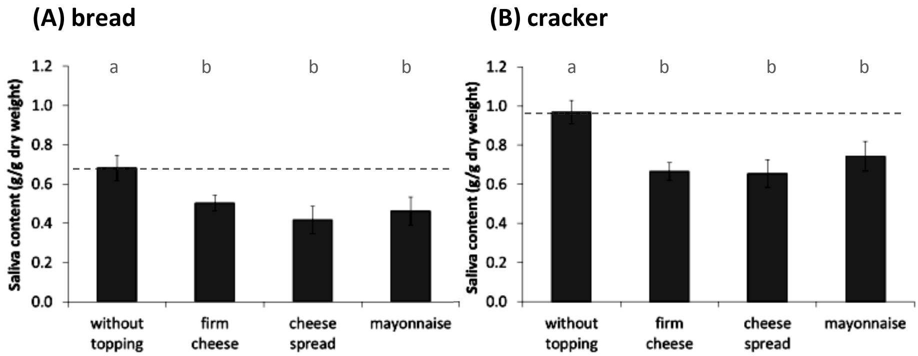

Figure 2.3: Bolus saliva content at moment of swallowing in bread (A) and crackers (B) with and without toppings. Error bars represent standard error of the mean. Dashed lines represent averaged value of single carriers (n=18 subjects). Different letters indicate significant differences (p<0.05).

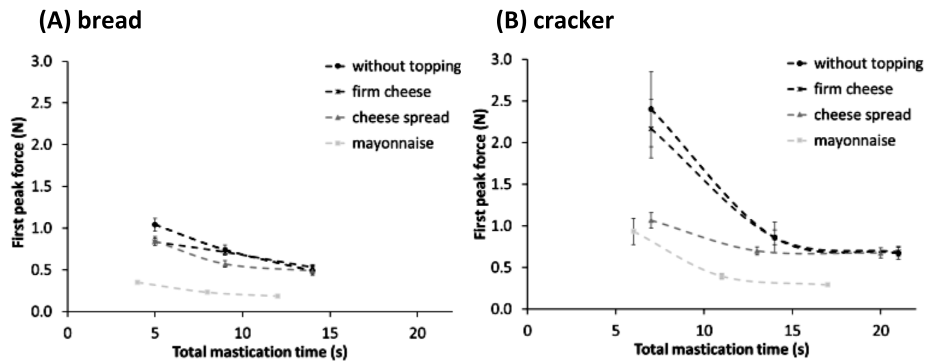

Figure 2.4: Bolus peak force during first puncture cycle (associated to firmness) for bread (A) and crackers (B) with and without toppings. Time points correspond to 33, 66 and 100% of total mastication time. Error bars represent standard error of the mean (n=18 subjects, in duplicate). Dashed lines are added to guide the eye.

Mechanical properties of boli

Figure 2.4 shows the average first peak force (associated to firmness) of the expectorated boli as a function of mastication time. The peak force of all samples decreased over mastication time, in particular during the first 33% of total mastication time, to reach a plateau at around 66% of total mastication time (time effect, F=123.6, p<0.001). A significant carrier:topping effect was found (F=4.6, p=0.004). Both the initial first peak force and its decrease during mastication was higher for crackers than for bread. For example, the peak force of single bread decreased from 0.92±0.08 to 0.49±0.05 N, while for the crackers a much larger decrease from 16.92±2.89 to 0.67±0.06 N was found. Addition of toppings to carriers decreased the first peak force of both carriers, especially at the early stage of mastication. After 33% of total mastication time, the peak force of single bread was 1.04±0.08 N. This decreased to 0.84±0.04, 0.85±0.05, and 0.35±0.02 N after addition of firm cheese, cheese spread and mayonnaise, respectively. In the case of crackers, the toppings decreased peak force at 33% of total mastication from 2.40±0.45 to 2.17±0.35 N for firm cheese, to 1.07±0.09 N for cheese spread, and to 0.93±0.16 N for mayonnaise. On average, the addition of mayonnaise (gray square) showed the largest decrease in peak force for both bread and crackers and resulted in the lowest peak forces during all stages of mastication.

Adhesiveness of boli from bread and cracker increased with increasing mastication time (data not shown). Significant carrier:topping, carrier:time and topping:time interactions were observed (Table 2.2). At the moment of swallowing, bread and crackers did not differ in adhesiveness, but a significant topping effect (F=27.7, p<0.001) and carrier:topping interaction (F=3.8, p=0.01) were observed (Table 2.3). In the case of topping addition, the adhesiveness of bread and crackers either increased or decreased. Addition of toppings showed an increase for firm cheese and cheese spread and a decrease for mayonnaise.

Small, but statistically significant differences between samples and time points were found for cohesiveness (Table 2.2). On average, bread boli were slightly more cohesive than cracker boli at all three time points (data not shown). Addition of firm cheese, cheese spread and mayonnaise increased the cohesiveness of single carriers in ascending order. At the moment of swallowing, a significant carrier effect (F=134.8, p<0.001) and topping effect (F=12.5, p<0.001) were observed (Table 2.3).

2.3.3 Characterization of sensory properties

Figure 2.5A and Figure 2.5B show the sensory scores for dryness perception as a function of the mastication time for bread and crackers. Dryness scores decreased with increasing mastication time for all samples. After one chew, single crackers (90.4±2.1 mm, Figure 2.5B) were perceived drier than single bread (65.4±3.4 mm, Figure 2.5A). A larger absolute decrease in dryness perception was observed for crackers than bread (carrier:time interaction, F=15.0, p<0.001). However, a difference in dryness between the carriers was maintained through mastication, and crackers (62.3±4.2 mm) were still perceived drier than bread (45.3±3.2 mm) at the end of mastication. Addition of toppings decreased dryness perception at all four mastication time points for both carriers ($F=115.3$, $p<0.001$) (Figure 2.5A, 2.5B). Although all three toppings decreased dryness perception, the type of topping added to the carriers determined the degree of the decrease. At the end of mastication, a significant carrier effect ($F=52.1$, $p<0.001$) and topping effect ($F=25.3$, $p<0.001$) were observed. Dryness scores of bread decreased significantly from 45.3±3.2 to 36.6±3.0 mm after addition of firm cheese, to 28.4±2.8 mm after addition of cheese spread, and to 29.0±3.3 mm after addition of mayonnaise ($p<0.05$). Similar results were found for the dryness perception of crackers, and dryness scores of crackers significantly decreased from 62.3±4.2 to 48.5±3.8, 44.6±3.5 and 36.7±3.3 mm after addition of firm cheese, cheese spread, and mayonnaise, respectively ($p<0.05$). Overall, the absolute decrease in dryness was largest after the addition of mayonnaise, followed by cheese spread and then firm cheese for both carriers at all mastication time points.

Figure 2.5C and Figure 2.5D display the sensory scores for firmness perception as a function of the mastication time for bread and crackers. Firmness scores decreased with increasing mastication time for all samples. After one chew, single crackers (86.6±2.6 mm, Figure 2.5D) were perceived firmer than single bread (58.3±3.5 mm, Figure 2.5C).

The absolute decrease in firmness through mastication was greater for crackers than for bread, resulting in similar firmness intensities at the moment of swallowing ($F=1.9$, $p=0.166$). Addition of toppings did not significantly change the firmness scores after one chew. Addition of toppings to carriers decreased firmness scores throughout mastication (Figure 2.5C, 2.5D), but the effect depended on the type of topping. The absolute decrease in firmness perception was greatest after the addition of mayonnaise, followed by cheese spread, and then firm cheese. At the moment of swallowing, a significant topping effect was found ($F=10.2$, $p<0.001$), and addition of mayonnaise significantly decreased the firmness intensity perception of both carriers (Table 2.3).

Stickiness increased with increasing mastication time, especially at the early stage of mastication (data not shown). At the moment of swallowing, no significant difference in stickiness was observed between bread and crackers ($F=0.4$, $p=0.553$), but a significant topping effect was found ($F=4.6$, $p=0.004$). Addition of mayonnaise to the carriers significantly decreased stickiness at the moment of swallowing ($p<0.05$)(Table 2.3); stickiness of single bread decreased from 58.8±2.7 to 48.7±3.8 mm after addition of mayonnaise, whereas the stickiness of cracker decreased from 59.9±3.5 to 54.1±3.7 mm.

Flavor intensity slightly increased with increasing mastication times (Figure 2.5E, 2.5F). Overall, flavor intensity was driven by the presence of toppings rather than carriers.

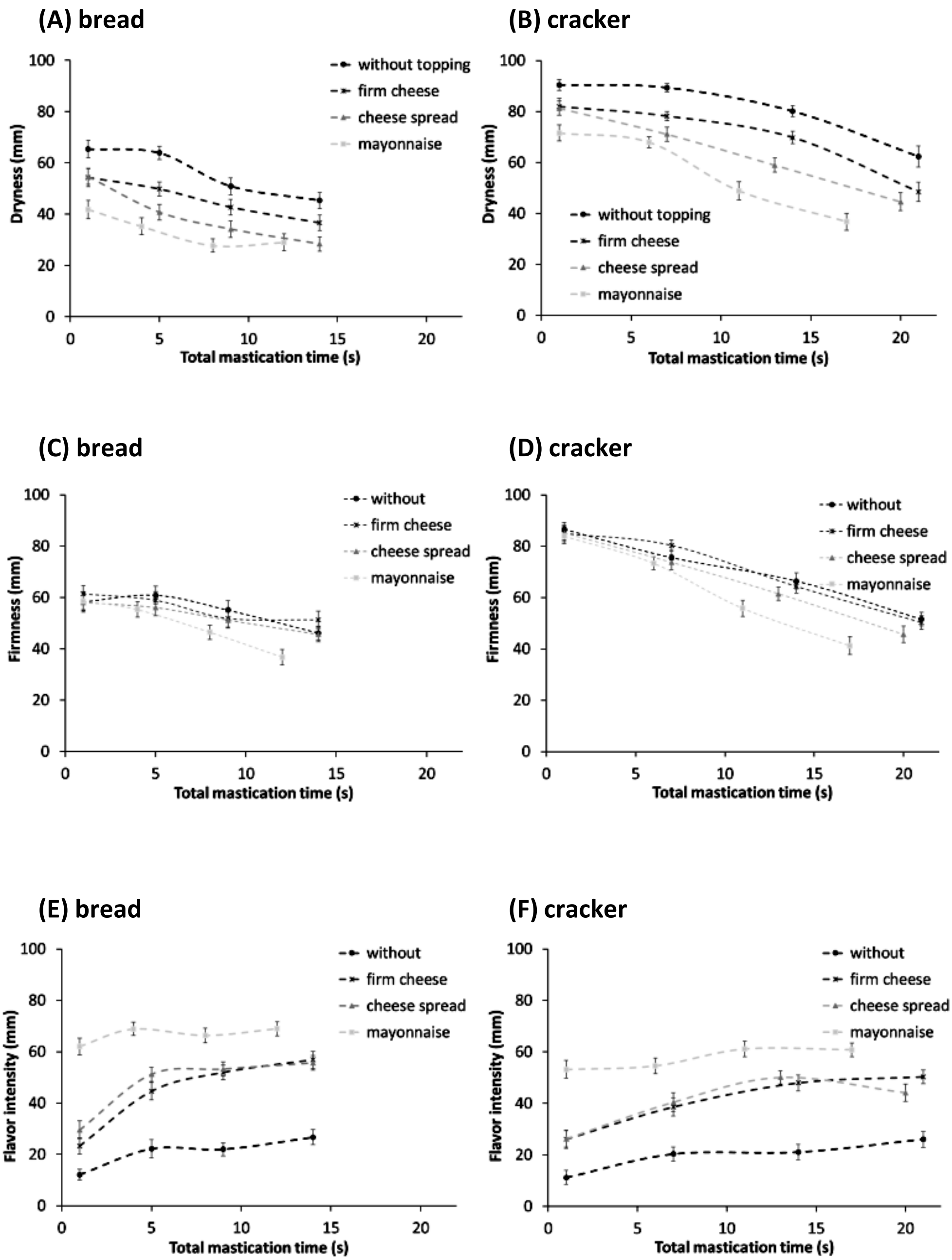

Figure 2.5: Dryness, firmness and flavor intensity for bread and crackers with and without toppings determined by progressive profiling (n=18 subjects, in duplicate): (A) dryness of bread; (B) dryness of crackers; (C) firmness of bread; (D) firmness of crackers; (E) flavor intensity of bread; and (F) flavor intensity of crackers. Time points correspond to 1 chew and 33, 66 and 100% of total mastication time. Error bars represent standard error of the mean. Dashed lines are added to guide the eye.

Addition of toppings to carriers increased flavor intensity scores at all four mastication time points. The degree of the increase was determined by the type of topping that was added to the carriers. The absolute increase in flavor intensity of both carriers was largest after the addition of mayonnaise (flavor intensity scores increased by a factor of 3), followed by cheese spread and firm cheese (flavor intensity scores increased by a factor of 2). Furthermore, the absolute increase in flavor intensity was larger when toppings were added to bread than crackers. At the moment of swallowing (Table 2.3), the flavor intensity of bread without topping, with firm cheese, cheese spread and mayonnaises were 26.7±2.9, 56.8±3.3, 55.6±2.8 and 68.9±2.8 mm, whereas this was 26.0±3.1, 50.4±2.7, 44.2±3.4 and 60.9±2.7 mm for crackers, respectively.

2.4 DISCUSSION

We investigated the contribution of toppings to carriers on the oral processing behavior and dynamic sensory perception of composite foods. Carriers (bread and crackers) and toppings (firm cheese, cheese spread and mayonnaise) were used as single foods, and they were combined into composite foods (carrier with topping) with contrasting composition, mechanical properties and sensory characteristics.

As was seen in Figure 2.1, spreading the toppings cheese spread and mayonnaise on carrier foods reduced total mastication time and number of chews until swallowing, and no effect was found for firm cheese. It is interesting to note that the weight of carriers alone was 2.1 g, while the weight of carrier-topping combinations doubled to 4.2 g (*i.e.* 2.1 g carrier and 2.1 g topping). Thus, even though addition of toppings to carriers doubled the sample weight, the carriers shortened the total mastication time in the case of cheese spread and mayonnaise while it remained similar for firm cheese. This implies that toppings influenced the mastication behavior of carriers. These data are consistent with those of Gavião *et al.* (2004) and Engelen *et al.* (2005) who showed that spreading butter on toast decreased the time and number of chewing cycles until swallowing. In those works, it was hypothesized that toppings facilitated saliva in the bolus formation of carriers leading to boli that are broken down and lubricated enough to be safely swallowed after shorter mastication times and less chews. This is indeed confirmed by the results of our current study. Addition of toppings to carriers led to less saliva incorporation (Figure 2.3) and a faster decrease in first peak force (Figure 2.4) of carrier boli. Addition of toppings to carriers led to decreased dryness and firmness perception (Figure 2.5). Due to their dry character, both bread and crackers require an increase in lubrication behavior and reduction of structure before they can be swallowed safely. These results show that toppings moistened and softened the bolus, and consequently less time had to be spent on reducing structure and increasing lubrication to safely swallow earlier. This confirms the hypothesis that toppings assist saliva in bolus formation.

Comparing the two different carriers (bread versus cracker), crackers have a lower moisture content and are harder than bread (Table 2.1). It is known that dry and/or hard foods are processed in the mouth for longer time because they require more saliva and/or need to be softened more (Hutchings & Lillford 1988; Chen *et al.* 2013). This is confirmed by the present study, which found longer mastication times and higher number of chews until swallowing for crackers than for bread (Figure 2.1). Hence, dry and/or hard foods were expected to benefit more from the facilitating effects of toppings than moist and/or soft foods. We indeed found that for crackers, the addition of toppings resulted in a larger absolute decrease in total mastication time and number of chews until swallowing (Figure 2.1), and this effect was less pronounced in bread. These findings are in line with previous studies. Engelen *et al.* (2005) reported that adding butter to toast and cake reduced the number of chewing cycles until swallowing, whereas no effect was found for bread (higher initial moisture content, softer). In addition, adding fluids to toast (low moisture content, hard), cake (low moisture content), and peanuts (low moisture content, hard) decreased the number of chews until swallowing, whereas this effect was not found for cheese (soft) and carrots (high moisture content, hard)(Pereira *et al.* 2006; Pereira *et al.* 2007; Van Der Bilt *et al.* 2007). These studies clearly indicate that mainly dry foods benefit from the facilitating effects of fluids or toppings leading to shortened mastication times, but no data was found on the bolus properties underlying this observation. Our present study highlights that indeed the bolus formation of crackers rather than bread was affected by the presence of toppings. Absolute decrease in saliva incorporation and first peak force after addition of toppings was larger for crackers compared to bread (Figure 2.3 and 2.4). Thus, dry and hard crackers with a low moisture content absorbed more moisture from the toppings than soft bread with a higher moisture content.

Different toppings (firm cheese, cheese spread and mayonnaise) impacted oral processing behavior and sensory perception of carriers differently. On average, mayonnaise had the largest impact on oral processing behavior of carriers, followed by cheese spread and only a small influence by firm cheese was observed. Similar findings were found for dryness and firmness perception. It appears that the higher the contrast in terms of mechanical properties and/or moisture content between toppings and carriers, the larger the influence of toppings on oral processing behavior and texture perception of carrier foods. These different effects of the three toppings are likely to be caused by variations in their initial composition and product properties. It seems that incorporation of toppings into bread and cracker boli depends on its consistency. Mayonnaise has the highest moisture content and can be characterized as a plastic liquid-like topping (Table 2.1). Due to its high moisture content and liquid behavior it was easily mixed with the carriers to form a cohesive bolus. Mayonnaise thereby softened the carrier boli most effectively (Figure 2.4) and consequently decreased dryness and firmness scores largely (Figure 2.5). Cheese spread also decreased mastication times and number of chews of carriers, but to a lesser extent than mayonnaise.

This could be attributed to less moisture incorporation into the carrier boli due to its semi-solid texture and slightly lower moisture content. Another possible explanation might be found in differences in adhesiveness between cheese spread and mayonnaise. Increased adhesiveness of foods requires longer cycle duration and increased muscle activities to remove it from oral surfaces and thereby increasing total mastication time. This has been shown for caramels (Çakir *et al.* 2012a). Cheese spread is more adhesive than mayonnaise and was perceived more sticky, which might have contributed to longer mastication time and a higher number of chews. Firm cheese had the lowest impact on both oral processing behavior and sensory perception of carriers, which can be explained by its solid-like consistency and limited ability to incorporate moisture in the boli. The facilitating effects of toppings on bolus formation of bread and crackers depends strongly on the consistency of toppings. Fat content is not likely to have caused the differences between toppings found in the present study, since all three toppings contained similar fat content (Table 2.1).

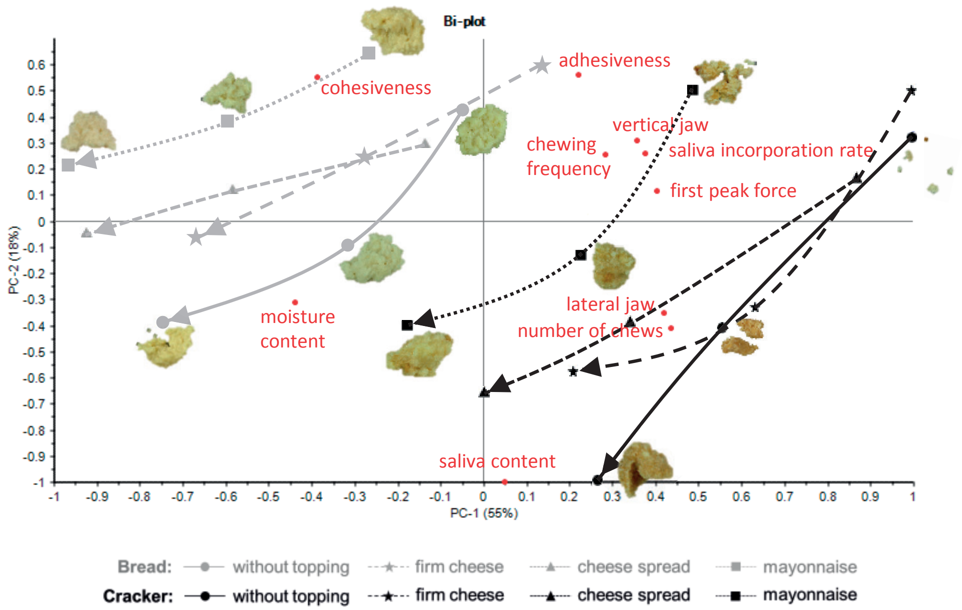

Figure 2.6: Principal component analysis (PCA) representing oral processing pathways of bread and crackers with and without toppings. Mastication behavior parameters (number of chews, chewing frequency, maximum vertical and lateral jaw movements) were taken at three stages during oral processing (*i.e.* 0-33, 33-66 and 66-100% of total mastication time). Bolus properties (moisture content, saliva content, saliva incorporation rate, first peak force, adhesiveness and cohesiveness) were taken at three time points throughout oral processing (33, 66 and 100% of total mastication time). Bread samples are presented in gray, and cracker samples in black. Lines were drawn to guide the eye. Pictures of single carriers and carriers with mayonnaise were presented close to the corresponding time points for illustration purposes.

Figure 2.6 shows a Principal Component Analysis (PCA) bi-plot of carriers with and without toppings over oral processing parameters throughout mastication to summarize the oral processing pathways of such foods. Mastication behavior parameters were taken at three stages during oral processing (*i.e.* 0-33, 33-66 and 66-100% of total mastication time) and bolus properties were taken at three time points throughout oral processing (33, 66 and 100% of total mastication time). As can be seen, bread samples are located on the left side of the first the principal component (PC1) and the cracker samples are located on the right side of PC1 (X axis, 55%). Oral processing of bread starts near bolus adhesiveness and cohesiveness, whereas oral processing of crackers starts near vertical jaw movements, saliva incorporation rate and bolus first peak force. Interestingly, all foods move towards the left and towards the bottom part of the plot with increasing mastication time, moving towards the parameters moisture content and saliva content. Carriers with toppings are positioned relatively close to the corresponding single carrier, showing that the oral processing pathways of composite foods are relatively close to that of the single carrier. This indicates that the oral processing pathways and oral processing behavior are driven by carriers rather than toppings, which might be due to the dominating texture of the carriers. This is consistent with previous studies that investigated model foods, in which a hard layer was found to dominate the oral processing behavior of bi-layer model gels containing hard and soft layers (Devezeaux de Lavergne *et al.* 2016b). In addition, the hard crust was found to dominate the dynamic texture perception of bread crumb with crust sample throughout consumption (Gao *et al.* 2017). Although the pathways of carriers with and without toppings were similar, the presence of a topping induced some changes to the oral processing pathways of single carriers (Figure 2.6). Carriers with firm cheese and cheese spread are positioned closest to its single carrier, whereas carriers with mayonnaise are positioned more towards the left top part of the plot. This illustrates that the addition of mayonnaise had the highest impact on the oral processing pathways of carriers.

When carrier foods were combined with toppings, the oral processing pathways and texture perception were influenced by both carriers and toppings, but in different ways. Carriers drove the oral processing behavior and texture perception because these need to be broken down into smaller particles and need to be lubricated before swallowing. Toppings moistened the carrier foods by which they adapted their oral processing pathways and texture perception.

Flavor perception is another important aspect of the dynamic sensory perception of composite foods. Flavor intensity increased with increasing mastication time. The addition of toppings to carriers increased flavor intensity scores at all four time points, indicating that flavor perception throughout consumption was driven by the toppings rather than the carriers. This increase in flavor intensity by adding toppings might be one of the factors that explains why bread and crackers are frequently consumed together with toppings. Besides, flavor intensities were lower for cracker-topping combinations than for bread-topping combinations (Figure 2.5, Table 2.3). This might be explained by the dry character of crackers, dry crackers might take up the topping to a larger extent than moist bread, leading to a lower flavor intensity perception. Another explanation might be the hard character of the crackers, hard cracker require more oral breakdown than soft bread, which might distract consumers attention from flavor perception. This result is in line with previous studies on single foods, showing that an increase in hardness of gels and candies decreased the perceived flavor intensities (Boland *et al.* 2006; Saint-Eve *et al.* 2011).

The present study has important implications for food design, and these insights allow the tailoring of oral processing behavior and subsequent consumer perception of composite foods. For example, adding toppings facilitated mastication and bolus formation of carrier foods, which could be applied in foods for elderly or people with decreased eating capabilities. Although this study focuses on the oral processing behavior and sensory perception of composite foods, the findings may well have implications for food intake and satiation. Addition of toppings to carrier foods decreased oro-sensory exposure time while it increased the total energy content of the food consumed. This might cause faster consumption of higher energy dense foods and subsequently this may result in increased consumers food intake.

2.5 CONCLUSION

This study shows that toppings (firm cheese, cheese spread and mayonnaise) impacted the mastication behavior, bolus formation and dynamic sensory perception of carrier foods (bread, crackers). Carriers to which semi-solid cheese spread and liquid-like mayonnaise were added were chewed for a shorter time and with fewer chews than single carriers bread and crackers, although twice the mass of food was orally processed. No effect was found for solid firm cheese. Less saliva incorporation and a faster decrease in bolus peak forces were observed after addition of toppings to carriers. Addition of toppings also decreased dryness and firmness perception, whereas flavor perception was increased at all stages of mastication. We conclude that toppings assist saliva in bolus formation of bread and crackers. In particular, liquid-like mayonnaise and semi-solid cheese spread moistened and softened the carriers, leading to shorter mastication times and a lower number of chewing cycles to break down and lubricate boli enough to be safely swallowed. As the effects were most pronounced after the addition of mayonnaise, followed by cheese spread and firm cheese, the consistency of the toppings seems to play a major role. More liquid-like toppings are more easily mixed with the carriers, leading to faster moisture incorporation into the bolus and a faster softening of the bolus. The effects of toppings were more pronounced in crackers than in bread, indicating that also the dryness of the carriers affects oral processing. Thus, oral processing depends on the mechanical properties of both the carrier and the topping. To develop a full understanding on the role of single foods on the oral processing behavior and sensory perception of composite foods, additional studies investigating carriers and toppings with systematically varied product properties will be required.

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3

Sauce it up: Capturing the influence of condiment properties on eating behavior, bolus formation and sensory perception of solid foods

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ABSTRACT

Background and objective: Condiments are foods that are rarely consumed on its own. Although addition of condiments to carrier foods is known to affect eating behavior and sensory perception, a systematic understanding of how different condiment properties contribute to oral processing behavior and sensory perception of solid carrier foods is lacking. This study aimed to understand the mechanisms by which condiments varying in composition or rheological properties facilitate bolus formation and influence chewing behavior and sensory perception of solid carrier foods.

Methods: Two carriers (bread, cooked potato) were combined with mayonnaises differing in fat content and viscosity. Electromyography with jaw tracking, bolus properties throughout mastication and descriptive sensory perception were determined.

Results: Addition of mayonnaises changed bolus properties of solid carrier foods considerably (decreased bread firmness, increased potato cohesiveness, increased lubrication of both bread and potato bolus) and consequently facilitated faster bolus formation. While addition of mayonnaises to bread and potatoes decreased the number of chewing cycles before swallowing, consumers did not change muscle activities nor jaw movements per chew. No effect of mayonnaise fat content on eating behavior of composite foods was observed. Low viscosity mayonnaise resulted in faster bolus formation of carriers leading to faster swallowing than high viscosity mayonnaise. Low viscosity mayonnaise penetrated faster into bread, where it provides faster bread softening. Low viscosity mayonnaise mixed easier with the potato pieces by which it enhanced adhesion between potato pieces. Both mayonnaise fat content and viscosity influenced sensory perception of composite foods considerably, especially in terms of fattiness and creaminess.

Conclusions: We conclude that eating behavior, bolus formation and sensory perception of solid carrier foods can be modified considerably by addition of condiments. While composition and rheological properties of condiments have a large effect on bolus formation and sensory perception of solid carrier foods, they have only a limited effect on eating behavior which is dominated by the properties of the solid carrier food. This could be an effective strategy to increase healthy eating, alter food intake for example for the elderly population, and increase food appreciation.

Key words: condiments, composite foods, eating behavior, bolus properties, sensory perception

3.1 INTRODUCTION

Condiments are popular foods, and can be obtained in large variation; toppings, seasonings, dressings, sauces, etc. As condiments are consumed multiple times per day, they contribute largely to our daily food intake (Spence 2018). In the Netherlands, condiments are consumed during the three main meals (breakfast, lunch, dinner) as well as during snacking moments between meals (Van Rossum *et al.* 2016). Furthermore, condiment consumption among the Dutch population increased by 20% in the period from 2007-2010 to 2012-2016) (Van Rossum *et al.* 2016). Yet, condiments received surprisingly little scientific attention in the field of sensory science and eating behavior.

Condiments are frequently added to solid carrier foods such as bread, vegetables, potatoes, fish and meat. We refer to the combination of a solid carrier food with a condiment as composite food, which will be used throughout this paper. Addition of condiments has been suggested to complement or enhance the flavor, taste and texture perception of carrier foods and to increase sensory pleasure (Spence 2018). Sensory complexity increases when two foods differing in mechanical properties and composition are combined into a composite food (Paulsen *et al.* 2012; Tang *et al.* 2017; Santagiuliana *et al.* 2018a; Santagiuliana *et al.* 2018b). Such inhomogeneous composite foods are generally highly liked by consumers, presumably related to intra-oral sensory variety perceived throughout consumption (Szczesniak & Kahn 1984; Hyde & Witherly 1993; Miele *et al.* 2010).

Addition of condiments affects eating behavior of solid carrier foods. When condiments were added to bread or crackers, fewer chews, shorter mastication times until swallowing and consequently faster eating rate were observed (Gavião *et al.* 2004; Engelen *et al.* 2005; van Eck *et al.* 2019b). Condiments moistened and softened bread boli, which led to faster formation of safe-to-swallow boli (van Eck *et al.* 2019b). Condiments also facilitated mastication of raw vegetables. Addition of mayonnaise to raw carrots resulted in fewer chews, shorter mastication times and faster eating rate (van Eck *et al.* 2019c). In this case, the faster eating was not explained by moisture uptake of boli, as carrots are assumed to not absorb moisture in contrast to bread and cracker boli. The mechanisms for changes in eating behavior caused by addition of condiments to solid carrier foods may therefore be different for different composite foods. Little is known about how rheological and physical-chemical properties and composition of condiments influences eating behavior of composite foods.
As condiments are commercially available in a wide range of compositions (*e.g.* fat content, moisture content) and/or textural properties (*e.g.* viscosity, friction), we previously studied the effect of type of condiment (solid cheese, cheese spread, mayonnaise) on oral processing behavior of bread or crackers (van Eck *et al.* 2019b). Mayonnaise was shown to have the largest impact on oral processing behavior of composite foods (*i.e.* least number of chews, shortest mastication time, fastest eating rate), followed by cheese spread and solid cheese which had only limited impact on oral processing behavior. These different effects of the

three types of condiments are likely to be caused by their initial food properties, suggesting that condiment consistency affects bolus formation of carriers (van Eck *et al.* 2019b). In addition, differences in bolus formation were also found for bread and crackers as food structure breakdown and bolus formation was affected by the textural properties of such carriers. Addition of condiments to solid carrier foods seems to facilitate bolus formation of composite foods in different ways. We hypothesize that adherence of separate solid carrier bolus pieces is enhanced by condiments which provide lubrication to composite food boli. However, a systematic understanding of how condiment properties and composition contribute to oral processing behavior, bolus formation and sensory perception of solid carrier foods is still lacking.

Using a multidisciplinary approach to investigate the link between food structure, oral processing behavior and bolus properties is therefore necessary to better understand the transformation of food properties during mastication that trigger sensory sensations (Chen 2014; Devezeaux de Lavergne *et al.* 2017; Foegeding *et al.* 2017). Such approach has been used previously within a broad range of single foods including model gels (Çakir *et al.* 2012b; Koc *et al.* 2013; Devezeaux de Lavergne *et al.* 2015b; Devezeaux de Lavergne *et al.* 2016a; Krop *et al.* 2018), meat (Mioche *et al.* 2003; Yven *et al.* 2005; Devezeaux de Lavergne *et al.* 2015a), bread (Panouillé *et al.* 2014; Jourdren *et al.* 2016; Gao *et al.* 2017) and biscuits (Young *et al.* 2016a). As composite foods involve textural changes of two separate foods simultaneously, linking composite food structure to oral processing behavior and sensory perception becomes more challenging.

The aim of this study was to understand the mechanisms by which condiments varying in composition or rheological properties facilitate bolus formation and influence chewing behavior and sensory perception of solid carrier foods. Condiments (mayonnaises) varying in fat content and viscosity were combined with different carrier foods (bread, cooked potato). Two different carrier foods were chosen based on their difference in water absorption capability. We hypothesize that bolus formation of composite foods is affected by condiment viscosity with moisture being absorbed faster by carrier foods when viscosity of condiments is low. We hypothesize that high fat or moisture content of condiments facilitates adherence of composite food boli. We hypothesize that eating behavior links to bolus properties and sensory perception of composite foods. By systematically varying the properties of the condiments, this study provides new insights into the mechanisms underlying food oral processing of composite foods, which enables us to gain a better understanding of the structural transitions of foods that contribute to perception. Such knowledge may be useful to increase healthy food intake with high consumer appreciation.

3.2 MATERIALS AND METHODS

3.2.1 Samples

Mayonnaises varying in fat content and viscosity were used, and all mayonnaises had a similar flavor profile. A commercial mayonnaise was used as full fat/high viscosity version (FF-HV; Hellmann's Real, Unilever, The Netherlands). The low fat / high viscosity (LF-HV) and low fat / low viscosity (LF-LV) mayonnaises were prepared for this study at a pilot plant and are not commercially available. Two types of LF-HV mayonnaise were prepared, in which either starch or xanthan was used as thickening agent (LF-HV-starch, LF-HV-xanthan). As starch can be degraded by salivary enzymes, the viscosity of mayonnaises with starch was assumed to decrease faster during oral processing than that of mayonnaise thickened with xanthan. Table 3.1 presents an overview of the composition and product properties of the mayonnaises.

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Table 3.1: Product properties of mayonnaises varying in fat content, viscosity and thickening agent.

	FF-HV	LF-HV-starch	LF-HV-xanthan	LF-LV
Fat content	Full fat (FF)	Low fat (LF)	Low fat (LF)	Low fat (LF)
(% w/w)	73	20	20	20
Viscosity	High viscosity (HV)	High viscosity (HV)	High viscosity (HV)	Low viscosity (LV)
at 1 s^{-1} (Pa·s)	60±12	92±22	149±16	2±0.2
at 10 s^{-1} (Pa·s)	9±2	13±3	16±1	0.4±0.04
at 100 s^{-1} (Pa·s)	1.3±0.3	1.9±0.4	1.6±0.1	0.1±0.01
Thickening agent	-	Starch	Xanthan	-
(% w/w)	-	5	3	-

Mayonnaises were combined with solid carrier foods to form composite foods. Two commercial carrier foods were used, namely bread (whole grain casino bread, Albert Heijn, The Netherlands) and purple potatoes (Solanum tuberosum, Albert Heijn, The Netherlands). Bread and potatoes were selected based on their difference in water absorption capability (*e.g.* bread is assumed to absorb moisture, whereas boiled potatoes are assumed to absorb less moisture). Dark bread and purple potatoes were chosen to increase color contrast between condiment and carrier foods in expectorated boli to facilitate qualitative visualization of mixing behavior of condiments with carriers in boli. Fresh bread without crust was cut in squares of 35x35x8 mm of approximately 3.5 g (moisture content: 44±3 wt%). Potatoes were peeled, cut in small beams of ~70x12.5x.12.5 mm of approximately 6.5 g, vacuum packed into heat-resistant plastic bags, and cooked sous-vide at 90ºC for 15 min (moisture content: 88±1 wt%). After cooking, all bags were cooled in ice water for 15 min and stored in the refrigerator (4 ºC) for up to six days.

For carrier-mayonnaise combinations, mayonnaise was spread on top of bread (simplified model for bread with spread), and potatoes were completely covered by mayonnaise (simplified model for potato salad with mayonnaise dressing). Approximately 3.5 g of mayonnaise was added to the carriers leading to a 1:1 weight ratio for bread-mayonnaise combinations and a 2:1 weight ratio for potato-mayonnaise combinations. This was based on the weight ratios of bread with spreads and vegetables with condiments of previous studies (van Eck *et al.* 2019b; van Eck *et al.* 2019c). Carrier-mayonnaise combinations were prepared just before serving to minimize moisture transfer of the mayonnaises into the carriers before consumption. In addition, carriers were assessed alone as a reference.

3.2.2 Oral breakdown of composite foods

Subjects

A group of 16 Caucasian, European females (22.9±2.5 years, mean±SD) participated in the study. All subjects were consumers of bread, potato and mayonnaise on a regular basis, had good dental health (self-reported), and were non-smokers (self-reported). A homogeneous group of subjects (n=16) was selected based on age, gender, mechanically stimulated saliva flow rate (3.2±0.5 g/min, mean±SD) and natural eating time from a selection of samples (13±3 s for both mayonnaise-bread combinations and mayonnaise-potato combinations, mean±SD), which were assessed during one hour long screening session (n= 33 recruited subjects). Subjects with low/high saliva flow rates (top and bottom 10%) and fast/slow mastication times (top and bottom 10%) were excluded from participation. All subjects gave written informed consent, completed the study and received financial compensation for participation.

Experimental approach

Subjects participated in 9 sessions of maximum 45 minutes over three months. In the first two sessions, natural eating behavior was characterized using video recordings to calculate bolus expectoration time points. The subsequent five sessions were used for bolus collection from which various bolus properties were characterized. In the last two sessions, mastication behavior using electromyography (EMG) and jaw tracking (JT) was determined.

For all sessions, samples were presented with three-digit codes in a random order following a completely randomized design. All samples were served on a spoon. Between each sample, subjects cleansed their palate with cold water and tea (Jasmine green tea, Twinings, UK) for at least 1 minute. They used tongue scrapers to aid the removal of oil from their tongue.

Characterization of eating behavior using electromyography and jaw tracking

Muscle activity and three-dimensional jaw movements were recorded simultaneously. Electromyographic (EMG) activity was recorded bilaterally from the superficial masseter, anterior temporalis, and anterior digastric muscles at 1000 Hz using surface electrodes

(BioFlex, BioResearch Assoc. Inc., Milwaukee, WI). Muscle position was located by palpation while participants clenched their teeth and electrodes were adhered to the skin overlying the muscles. Participants held a ground electrode to minimize electrical background noise. Lead wires were connected to the BioEMG III amplifier, which passed amplified EMG signals to a recording computer. Mandibular movements were recorded using a jaw tracking device (JT-3D, BioResearch Assoc., Inc.), which records incisor-point movements in three-dimensions. A small magnet was attached to the lower central incisors with a dental-grade adhesive (Stomahesive, ConvaTec, Princeton, NJ). Movement of the magnet was tracked by an array of sensors securely fit as a unit on the subject's head to record vertical, anteroposterior, and lateral components of the jaw relative to the cranium during mastication. During data collection, jaw kinematics (magnet position) and EMG signals were electronically recorded synchronously using BioPAK software (v6.0, BioResearch Assoc., Inc.).

Raw EMG data were band-pass filtered at 100-3000 Hz. To provide a single waveform for analyses, raw EMG data were transformed by calculating the root mean square (rms) of each digitized raw EMG signal at 2 ms intervals over a 42-ms time constant using LabView Graphical Programming System (National Instruments Corporation, Austin, TX) (Hylander & Johnson 1993; Vinyard *et al.* 2008). A chewing sequence was produced for each sample from the simultaneous recordings of jaw movements and rms-EMG activity. As with the video recordings, total mastication time (s), number of chews and chewing frequency (chews/s) were determined. More detailed parameters including chewing cycle duration (opening, closing, power stroke), chewing velocities (during opening, closing), chewing movements (vertical, anterior posterior and medial lateral direction) and muscle activities (temporalis, masseter and digastric; per chew and during the sequence) were also collected to comprehensively analyze jaw-muscle activity and jaw-kinematic patterns.

Characterization of bolus properties throughout consumption

Several bolus properties (moisture content, saliva content, fat content, mechanical properties, friction coefficient) were characterized at 33, 66 and/or 100% of the chewing sequence. These time points were defined for each sample by averaging the total mastication time of all subjects and all replicates extracted from the video recordings, following the method described by van Eck *et al.* (2019) (van Eck *et al.* 2019b). Subjects attended five bolus collection sessions over a time period of one month. These sessions were divided in three parts: (1) boli were collected at 33, 66 and 100% of total mastication for image acquisition, followed by dry matter analysis and subsequent fat content analysis), (2) boli were collected at 33, 66 and 100% of total mastication for the analysis of the mechanical properties (30 samples were served over two sessions), and (3) boli were collected at 100% of total mastication time for tribological analyses (10 samples were served over 1 session). Serving order was randomized for sample and time point. To collect the boli, subjects were instructed to chew a sample for a given number of seconds using a stopwatch, to expectorate

the bolus into a petri dish, and to cover the petri dish with a lid. All boli were analyzed immediately after expectoration to prevent moisture evaporation from the samples.
Bolus images: Pictures of expectorated boli were taken for all samples (n=10), all time points (n=3) and all subjects (n=16) using an image acquisition system (IRIS VA 400, AlphaMOS, France). Pictures were used to qualitatively illustrate differences between food boli, time points and subjects. Pictures were not analyzed further to obtain quantitative information.

Moisture and saliva content: Moisture and saliva content of boli were determined by dry matter content analysis for all samples and all time points. Boli were placed on aluminum dishes, weighed, dried for 16 – 18 h at 105°C in an atmospheric oven (Venti-line, VWR®), and weighed again after drying. Bolus moisture content (MC) on a wet weight basis was calculated using $MC=(m_0-m_1)/m_0 \cdot 100\%$, where m0 is the weight before drying and m1 is the weight after drying. Bolus moisture content on a dry weight basis was calculated using $MC_{db}=(m_0-m_1)/m_1$, which was subsequently used to calculate the saliva content (SC) by subtracting the moisture content of the product from the moisture content of the bolus (MC_{db}).

Fat content: Total fat content of all carrier-mayonnaise boli expectorated at 100% of mastication time were determined using Soxhlet extraction with petroleum ether. Total fat content determination was applied immediately after the dry matter analysis. Total fat content on a dry weight basis (FC_{db}) was calculated using $FC_{db}=f_1/db_0$ where f_1 is the weight of fat after evaporation and db_0 is the weight of the dried bolus before extraction. Plain carrier boli were not analyzed since fat is absent in these products.

Mechanical properties: Mechanical properties of boli presented in petri dishes were determined by a two cycle puncture test for all samples and all time points, following the method described by van Eck *et al.* (2019) van Eck *et al.* (2019b). A Texture Analyzer (TA. XT Plus) equipped with a 500 g load cell and a cylindrical probe with a diameter of 4 mm was used. Puncture tests were performed up to a strain of 50% of the initial bolus height (typically 10 mm) at three different locations of the expectorated bolus. A constant speed of 5 mm/s was used. After the first puncture, the probe was retracted from the samples and a resting time of 5 s was used before the second puncture was applied. Bolus firmness, adhesiveness and cohesiveness were determined from the force-time curves as described by Devezeaux de Lavergne *et al.* (2015b). Firmness was defined as the maximum peak force during the first puncture cycle. Adhesiveness (*i.e.* food sticking to probe) was defined as the area under the negative force-time curve measured during the retraction of the probe from the samples in the first cycle. Cohesiveness (*i.e.* food sticking together) was defined as the ratio between the area under the positive force-time curve obtained during the second and first puncture cycle.

Friction coefficients of boli: Coefficients of friction of all boli expectorated at 100% of mastication time were determined using a tribometer (TriboLab, Bruker, Karlsruhe, Germany), following the method described by Fuhrmann *et al.* (2019) (Fuhrmann *et al.* 2020). Boli expectorated at 33 and 66% of mastication time were not analyzed since these samples were too inhomogeneous. Each bolus was split in two samples, after which each half of the bolus was placed on the lower surface and gently leveled with a spoon. The lower surface was a rectangular container attached to a reciprocating drive, containing a roughened PDMS mat (60x40 mm) to mimic the in-mouth surface. The container was moved in an oscillating fashion over a distance of 6 mm with an increasing speed from 0 to 12 mm/s. The upper surface was a cylindrical probe with a diameter of 30 mm made from roughened PDMS (706 mm^2). The normal force applied between the upper surface and the bolus was 0.5 N. The two surfaces were cleaned with ethanol and water prior to each measurement. Coefficient of friction (COF) was calculated with the use of the advanced oscillating COF algorithm provided with the UMT viewer software.

3.2.3 Sensory descriptive analysis

Quantitative descriptive sensory analysis was performed using Unilever Foods' absolute scaling method (UFASM) with a trained panel consisting of Dutch women (n=12) with an average age of 58±8 years (mean±SD). All panelists were previously familiar with the sensory attribute lexicon and with a large variety of mayonnaises.

Two familiarization sessions of 3 hours took place to acquaint the panel with the different mayonnaises and with the carrier-mayonnaise combinations. The first session was used to discuss the attributes with definitions (Table 3.2). Attributes were based upon past lexicons developed for mayonnaise evaluation, and attributes applicable to carrier-mayonnaise combinations were determined during a panel discussion. The second training session was used to set the attribute order, after which the panel practiced with the FF-HV, LF-HV-xanthan and LF-LV mayonnaises without and with carriers. The panelists used a 15-step categorical scale ranging from 0 to 15, where 0 represented not at all and 15 represented extremely high intensity.

The panel attended two evaluation sessions of 2.5 hours each (1 hour – 30 min break – 1 hour) over two weeks. The sessions were organized by sample type: bread without/with mayonnaise was evaluated in the first session and potato without/with mayonnaise was evaluated in the second session. At the start of each session, the panel evaluated one warm-up sample (FF-HV) to avoid first-order-effects. All samples were coded with 3-digit random codes, evaluated in duplicate and presented in a random order following a balanced design. Between each sample, subjects cleansed their palate with cold water, tea (jasmine green tea, Twinings, UK) and crackers (Barber cream crackers, Burton's Biscuit Co., UK) for at least 2- 3 minutes.

Table 3.2: Sensory attributes and definitions used during the QDA evaluation.

Modality	Attribute	Definition
Odor	Overall odor intensity	The intensity of the odor totality
Taste	Overall taste intensity	The intensity of the taste totality
Mouthfeel	Dry	Dry and rough feeling on the tongue or in the mouth
	Firm	Degree of firmness (the force needed to press the sample between the tong and the palate)
	Sticky	Degree of stickiness
	Gummy	Degree of small soft gel particles or lumps
	Creamy	Degree of creaminess like whipped cream
	Fatty	Degree of fatty feeling
	Velvet	Degree of creamy feeling such as Calve full fat mayo (soft and velvet)
	Smooth	Degree of slippery feeling
	Salivating	Degree of salivation or mouthwatering due to secretion of saliva
	Absorbing	Degree of mayonnaise absorbance in the bread / the potato
	Chewing effort	Degree of effort to chew the sample / form a bolus
	Homogeneous	Degree of mixing of mayonnaise with the bread / the potato (in the mouth)
	Bread fibers *	Degree of a fiber feeling, due to the presence of bread in the mouth
	Potato particles *	Degree of a particles feeling, due to the presence of potato pieces in the mouth
Afterfeel	Residue	A substance remains in the mouth (in the molars)
	Fatty film layer	A fatty film, coating remains in the mouth
	Dry, rough	Dry and rough feeling remains on the tongue or in the mouth
	Cleaning effort	Degree of effort to clean the mouth after eating the sample

* The sensory attribute bread fibers was assessed for bread samples only. The sensory attribute potato particles was assessed for potato samples only.

3.2.4 Statistical data analyses

Data were collected for carriers (bread, potato) with mayonnaises varying in fat content (high/low), viscosity (high/low) and thickening agent (starch/xanthan). As a reference, carriers without mayonnaises (single bread, single potato) were assessed to show the impact of the different mayonnaises on eating behavior, bolus formation and sensory perception of carriers.

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Results are reported as mean values with standard error. To investigate the effect of mayonnaise properties on eating behavior, bolus formation and sensory perception of carrier-mayonnaise combinations, linear mixed models were performed using Lmer package (Kuznetsova *et al.* 2016). Mayonnaise, carrier and mayonnaise:carrier interaction were set as fixed effects and subject, serving order, session (if applicable) and replicate (if applicable) were set as random effects. Data on single carriers (*i.e.* without mayonnaise) were used illustratively, and were not included in the linear mixed models. Multiple factor analysis (MFA) was performed to compare the different data sets (video recordings, EMG and jaw tracking, bolus properties at moment of swallowing and static sensory characteristics) simultaneously, using FactoMineR package (Lê *et al.* 2008). For this analysis, only those parameters with a significant mayonnaise effect during mixed models were considered. Furthermore, Pearson's product-moment correlations (r) were used to determine relationships between averaged coefficient of friction of boli and sensory perception (smoothness, dry and rough afterfeel). R language (RStudio, version 1.0.143) was used to perform all statistical tests. Significance level of $p<0.05$ was chosen.

3.3 RESULTS AND DISCUSSION

The aim of the study was to understand the mechanisms by which condiments varying in composition or rheological properties facilitate bolus formation and influence chewing behavior and sensory perception of solid carrier foods. . The results of all measurements (bolus formation, eating behavior, sensory perception) are summarized in Table 3.3 together with the results of the statistical data analysis. In the following, to be concise results of different measurements are linked and discussed together, and are not always discussed one by one

To gain more insights into the mechanisms responsible for bolus formation during oral processing and the accompanying sensory perception, a full fat mayonnaise with high viscosity (FF-HV), a low fat mayonnaise with low viscosity (LF-LV), and two low fat/ high viscosity mayonnaises (LF-HV) were assessed in the present study. For the LF-HV mayonnaises, a thickening agent was added to compensate for the decrease in viscosity upon a reduction in fat content. Two different thickeners were investigated, starch (which is frequently used in the preparation of low fat mayonnaises; LF-HV-starch) and xanthan (LF-HV-xanthan). These thickeners were chosen based on their sensitivity towards amylase present in saliva. Starch is broken down by α-amylase, which was expected to decrease the viscosity throughout mastication. As a comparison, we also used xanthan, which is not broken down by amylase, and therefore the viscosity was expected to remain the same through consumption. Although the rheological properties of the two mayonnaises were similar (Table 3.1), perception of the two mayonnaises was quite different. The structure of LF-HV-xanthan mayonnaise was perceived as gummy (gummy mouthfeel was 7.5±0.7 for LF-HV-xanthan compared to 0.2±0.1, 0.5±0.3 and 0.0±0.0 for FF-HV, LF-HV-starch and LF-LV, respectively). Gummy perception dominated the eating experience and this was strongly disliked by the subjects, as free comments of the panel indicated. In contrast to xanthan, starch is used commonly as thickening agent in commercially available low fat mayonnaises. As a consequence, the following section focuses on comparisons between HF-HV, LF-LV and LF-HV-starch rather than with LF-HV-xanthan.

3.3.1 Eating behavior of carrier foods with added condiments

Eating behavior of carrier foods with and without condiments

Table 3.3 provides a general overview of the results. Clear differences in eating behavior were observed for bread and cooked potato when consumed without mayonnaises (Table 3.3A). Plain wholegrain bread was eaten with more chews per sequence (23±2 vs. 20±1), higher muscle activities per chew (3.0±1.0 vs. 2.4±0.1) and larger jaw movements per chew (17.1±0.5 vs. 15.8±0.3 mm in vertical direction) than cooked potato, although bite size was smaller for bread (3.5 g) than potatoes (6.5 g). This was to be expected, as number of chewing cycles, chewing forces and jaw movements are closely linked to the mechanical properties of food (Ishihara *et al.* 2011; Çakir *et al.* 2012b; Kohyama *et al.* 2017). Bread is chewier than potatoes and therefore requires more chewing effort.

The addition of mayonnaises decreased the number of chewing cycles and increased eating rate significantly for both solid carrier foods (Figure 3.1). On average, the number of chews decreased from 23±2 to 16±2 for one bite of bread (-30%) (Figure 3.1A) and from 20±1 to 17±1 for potato (-15%) (Figure 3.1B). However, addition of mayonnaise did not affect muscle activities and jaw movements per chew for bread and potato. This was unexpected, as addition of mayonnaise softened the bread and potato boli (Figure 3.4A, 3.4B). As firmness perception decreased (from 5.6±0.5 to 4.4±0.3 for bread, and from 6.9±0.4 to 6.2±0.4 for potato), it was expected that chewing would take less effort when mayonnaises are added to solid carrier foods, but this was not reflected in muscle activities and jaw movements per chew. Hence, subjects adopted their oral processing behavior when mayonnaises were added to bread and potatoes simply by decreasing number of chews and total chewing time until swallowing while maintaining muscle activities and jaw movements per chew. Apparently, chewing force and jaw movements are dominated by the solid carrier food rather than the condiment, and therefore changes in condiment properties do not change chewing effort.

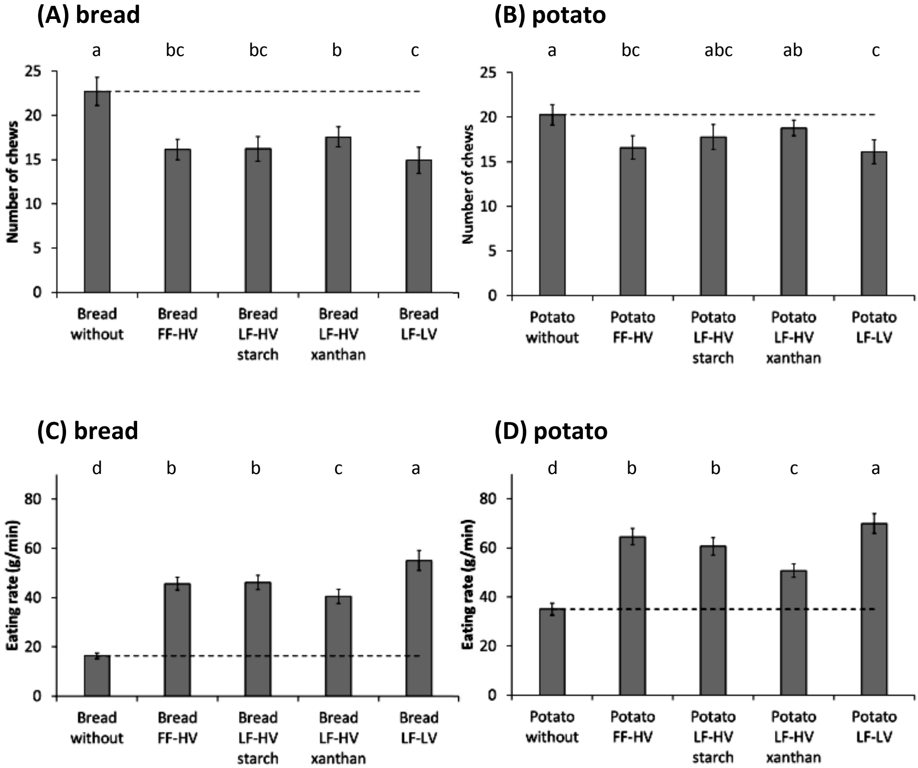


Figure 3.1: Number of chews required until swallowing and eating rate (g/min) for bread (A, C) and cooked potato (B, D) without and with different mayonnaises, determined using electromyography. Dashed lines represent the averaged value of single carriers. Error bars represent standard error of the mean. Different lower case letters indicate significant differences between means ($p<0.05$). The abbreviations are explained in Table 3.1.

Bread versus potato: The effect of condiment addition on eating behavior

A comparison of bread and potato carriers reveals that the effect of condiment addition on eating behavior depended on the type of carrier food. On average, the addition of mayonnaises decreased the number of chews from 23±2 to 16±2 for bread (-30%) and from 20±1 to 17±1 for potato (-15%), showing that condiments facilitated mastication of bread to a larger extent than that of cooked potato (both in absolute numbers and percentages). Hence, condiments seem to aid bolus formation of dry foods more than carriers with a high moisture content. In the case of low water content carriers, water might be absorbed faster and thus mixed easier with the carrier, whereas in high moisture content carriers water might be absorbed slower and mixing might occur mostly due to mechanical forces. In view of these findings, it should be noted that mayonnaise was mixed with bread in a 1:1 weight ratio, whereas potato was mixed with mayonnaise in a 2:1 weight ratio. Thus, a relatively higher amount of mayonnaise was present in bread-mayonnaise combinations than in the potato-mayonnaise combinations. This can contribute to the larger effect for the bread. We can therefore not

Table 3.3: Significance values (mayonnaise effect, carrier effect, mayonnaise:carrier interaction) and mean intensity scores (± standard error of the mean) describing eating behavior (A), bolus properties (B) and sensory evaluation (C) of carrier-mayonnaise combinations. Bread data is presented on page 62-65 and potato data is presented on page 66-69.

	Mayonnaise		**Carrier**		**MxC**	
	F	**p**	**F**	**p**	**F**	**p**
(A) EMG / Jaw Tracking						
Eating behavior						
Number of chews	18.6	**<0.001**	8.2	**0.005**	0.2	0.922
Chewing time (s)	16.0	**<0.001**	2.9	0.090	0.5	0.694
Chewing cycle duration (s)	3.6	**0.015**	22.5	**<0.001**	0.2	0.867
Opening duration (s)	6.5	**<0.001**	23.5	**<0.001**	2.6	0.052
Closing duration (s)	0.4	0.764	12.3	**<0.001**	0.2	0.918
Power stroke duration (s)	1.1	0.345	67.0	**<0.001**	0.6	0.599
Chewing frequency (chew/s)	3.1	**0.026**	20.1	**<0.001**	0.3	0.824
Eating rate (g/min)	18.0	**<0.001**	102.7	**<0.001**	0.9	0.420
Jaw movements						
Opening veloctiy (mm/s)	1.0	0.388	0.7	0.412	0.9	0.443
Closing velocity (mm/s)	2.0	0.108	13.8	**<0.001**	0.6	0.628
Vertical movement (mm)	5.1	**0.002**	20.8	**<0.001**	0.6	0.642
Anterior posterior movement (mm)	1.2	0.323	6.7	**0.010**	0.1	0.969
Medial lateral movement (mm)	1.5	0.222	1.5	0.230	1.0	0.392
Muscle activities						
Total activity per sequence	14.2	**<0.001**	17.4	**<0.001**	1.0	0.392
Total activity per chew	1.0	0.374	157.8	**<0.001**	1.6	0.193
Temporalis activity per sequence	10.5	**<0.001**	27.3	**<0.001**	1.1	0.343
Temporalis activity per chew	0.2	0.924	146.4	**<0.001**	1.2	0.318
Temporalis activity per chew (working-side)	0.2	0.918	124.4	**<0.001**	1.4	0.234
Temporalis activity per chew (balancing-side)	0.2	0.912	135.4	**<0.001**	0.8	0.505
Masseter activity per sequence	13.8	**<0.001**	31.4	**<0.001**	1.0	0.376
Masseter activity per chew	1.4	0.252	165.6	**<0.001**	2.0	0.116
Masseter activity per chew (working-side)	1.0	0.379	132.3	**<0.001**	1.6	0.202
Masseter activity per chew (balancing side)	1.2	0.301	121.7	**<0.001**	1.6	0.197
Digastric activity per sequence	13.0	**<0.001**	0.7	0.420	0.2	0.911
Digastric activity per chew	3.4	**0.019**	37.0	**<0.001**	0.5	0.685
Digastric activity per chew (working-side)	3.2	**0.025**	31.2	**<0.001**	0.8	0.489
Digastric activity per chew (balancing-side)	2.6	0.056	22.7	**<0.001**	0.4	0.752

Data on single bread (i.e. without mayonnaise) are presented for illustration purposes (light-gray column), and were not included in the linear mixed models. Significant p values (<0.05) are highlighted in bold. Different letters indicate significant differences between carrier-mayonnaise combinations. The abbreviations are explained in Table 3.1 and the sensory attributes are explained in Table 3.2.

Bread	Bread FF-HV		Bread LF-HV-starch		Bread LF-HV-xanthan		Bread LF-LV	
Mean ± SE	Mean ± SE		Mean ± SE		Mean ± SE		Mean ± SE	
23 ± 2	16 ± 1	*b*	16 ± 1	*b*	18 ± 1	*a*	15 ± 1	*b*
14.7 ± 0.9	10.4 ± 0.7	*bc*	10.4 ± 0.8	*b*	11.1 ± 0.7	*a*	9.2 ± 0.8	*c*
0.67 ± 0.02	0.66 ± 0.02	*ab*	0.66 ± 0.02	*ab*	0.66 ± 0.02	*a*	0.64 ± 0.02	*b*
0.23 ± 0.01	0.23 ± 0.01	*ab*	0.24 ± 0.01	*a*	0.23 ± 0.01	*a*	0.22 ± 0.01	*b*
0.27 ± 0.01	0.26 ± 0.01		0.26 ± 0.01		0.26 ± 0.01		0.26 ± 0.01	
0.16 ± 0.01	0.15 ± 0.01		0.15 ± 0.00		0.16 ± 0.01		0.15 ± 0.01	
1.5 ± 0.0	1.5 ± 0.0	*ab*	1.5 ± 0.0	*ab*	1.5 ± 0.0	*b*	1.6 ± 0.0	*a*
16 ± 1	46 ± 3	*b*	46 ± 3	*b*	41 ± 3	*c*	55 ± 4	*a*
40 ± 7	38 ± 7		38 ± 7		40 ± 7		36 ± 7	
36 ± 6	36 ± 6		37 ± 7		37 ± 7		34 ± 6	
17.1 ± 0.5	17.5 ± 0.5	*a*	17.9 ± 0.5	*a*	17.2 ± 0.5	*ab*	16.3 ± 0.6	*b*
7.1 ± 0.8	7.4 ± 0.7		7.6 ± 0.9		7.2 ± 0.7		6.9 ± 0.7	
7.9 ± 0.5	7.7 ± 0.4		8.2 ± 0.4		7.3 ± 0.4		7.4 ± 0.4	
67 ± 5	47 ± 3	*b*	46 ± 3	*b*	51 ± 4	*a*	39 ± 3	*b*
3.0 ± 0.1	3.0 ± 0.1		3.0 ± 0.1		2.9 ± 0.1		2.8 ± 0.1	
23 ± 2	16 ± 1	*b*	16 ± 1	*b*	18 ± 1	*a*	14 ± 1	*b*
1.04 ± 0.04	1.03 ± 0.04		1.05 ± 0.03		1.03 ± 0.04		1.01 ± 0.04	
0.52 ± 0.02	0.52 ± 0.02		0.53 ± 0.02		0.52 ± 0.02		0.51 ± 0.02	
0.52 ± 0.02	0.51 ± 0.02		0.52 ± 0.02		0.51 ± 0.02		0.50 ± 0.02	
21 ± 1	16 ± 1	*b*	16 ± 1	*b*	18 ± 1	*a*	13 ± 1	*b*
1.00 ± 0.04	1.00 ± 0.04		1.02 ± 0.03		1.00 ± 0.04		0.94 ± 0.04	
0.49 ± 0.02	0.50 ± 0.02		0.50 ± 0.02		0.50 ± 0.02		0.46 ± 0.02	
0.50 ± 0.02	0.50 ± 0.02		0.52 ± 0.02		0.50 ± 0.02		0.47 ± 0.02	
21 ± 1	14 ± 1	*bc*	15 ± 1	*b*	16 ± 1	*a*	13 ± 1	*c*
0.96 ± 0.04	0.94 ± 0.04	*ab*	0.97 ± 0.04	*a*	0.92 ± 0.04	*ab*	0.90 ± 0.03	*b*
0.49 ± 0.02	0.47 ± 0.02	*ab*	0.49 ± 0.02	*a*	0.45 ± 0.02	*ab*	0.45 ± 0.02	*b*
0.47 ± 0.02	0.47 ± 0.02		0.48 ± 0.02		0.45 ± 0.02		0.45 ± 0.02	

Table 3.3: Continued

	Mayonnaise		Carrier		MxC	
	F	p	F	p	F	p
(B) Bolus properties						
Compositional bolus properties						
Moisture content at swallowing (%wt)	613.1	**<0.001**	1819.7	**<0.001**	10.6	**<0.001**
Saliva content at swallowing (g/g dry product)	7.6	**<0.001**	80.0	**<0.001**	3.3	**0.024**
Fat content at swallowing (g/g dry product)	1268.9	**<0.001**	48.5	**<0.001**	4.1	**0.009**
Mechanical bolus properties						
Peak force at swallowing (N)	10.0	**<0.001**	14.7	**<0.001**	2.3	0.079
Adhesiveness at swallowing (N·s)	65.2	**<0.001**	117.7	**<0.001**	1.2	0.312
Cohesiveness at swallowing	19.8	**<0.001**	148.0	**<0.001**	11.9	**<0.001**
Tribological bolus properties						
Friction coefficient at swalllowing (8 mm/s)	10.9	**<0.001**	469.3	**<0.001**	12.2	**<0.001**
(C) Sensory characteristics						
Odour attribute						
Overall intensity	0.3	0.842	2.5	0.116	0.6	0.634
Overall intensity	11.1	**<0.001**	0.7	1.000	2.6	0.051
Mouthfeel attributes						
Dry	2.6	0.051	2.4	1.000	0.1	0.931
Firm	4.1	**0.008**	0.7	0.398	2.7	0.051
Sticky	28.4	**<0.001**	0.5	0.461	0.7	0.525
Gummy	35.8	**<0.001**	2.0	0.155	1.0	0.414
Creamy	20.9	**<0.001**	1.4	0.245	0.0	0.986
Fatty	46.0	**<0.001**	0.6	1.000	0.2	0.923
Velvet	6.0	**<0.001**	2.2	1.000	0.7	0.537
Smooth	0.4	0.776	1.2	0.281	0.3	0.833
Salivating	5.4	**0.001**	2.1	1.000	0.5	0.651
Absorbing	26.3	**<0.001**	3.2	1.000	8.9	**<0.001**
Chewing effort	0.7	0.540	0.4	0.514	2.8	0.044
Homogeneous	7.6	**<0.001**	0.0	0.886	3.8	**0.012**
Bread fibers*	1.8	0.151	-	-	-	-
Potato particles*	1.7	0.169	-	-	-	-
Afterfeel attributes						
Residue	3.6	**0.015**	0.3	1.000	0.7	0.564
Fatty film layer	24.6	**<0.001**	0.1	1.000	0.7	0.556
Dry Rough	0.3	0.797	0.3	1.000	0.4	0.741
Cleaning	5.1	**0.002**	0.0	1.000	0.5	0.668

* The sensory attribute bread fibers was assessed for bread samples only. The sensory attribute potato particles was assessed for potato samples only.

Data on single bread (i.e. without mayonnaise) are presented for illustration purposes (light-gray column), and were not included in the linear mixed models. Significant p values (<0.05) are highlighted in bold. Different letters indicate significant differences between carrier-mayonnaise combinations. The abbreviations are explained in Table 3.1 and the sensory attributes are explained in Table 3.2.

Bread	**Bread FF-HV**		**Bread LF-HV-starch**		**Bread LF-HV-xanthan**		**Bread LF-LV**	
Mean ± SE	Mean ± SE		Mean ± SE		Mean ± SE		Mean ± SE	
59.8 ± 2.0	48.4 ± 1.1	*b*	67.1 ± 0.8	*a*	66.5 ± 0.6	*a*	67.3 ± 0.5	*a*
0.86 ± 0.10	0.57 ± 0.04	*a*	0.72 ± 0.08	*a*	0.51 ± 0.05	*a*	0.58 ± 0.05	*a*
- - -	54 ± 1	*a*	25 ± 1	*b*	19 ± 1	*c*	28 ± 1	*b*
0.27 ± 0.02	0.10 ± 0.01	*b*	0.08 ± 0.01	*b*	0.15 ± 0.01	*a*	0.09 ± 0.01	*ab*
0.029 ± 0.002	0.016 ± 0.001	*b*	0.013 ± 0.001	*c*	0.024 ± 0.002	*a*	0.013 ± 0.001	*bc*
0.52 ± 0.01	0.55 ± 0.01	*a*	0.54 ± 0.01	*a*	0.56 ± 0.02	*a*	0.51 ± 0.01	*a*
1.21 ± 0.06	1.04 ± 0.05	*a*	0.92 ± 0.05	*b*	0.90 ± 0.02	*b*	0.86 ± 0.04	*b*
8.6 ± 0.4	8.2 ± 0.4		8.6 ± 0.3		8.2 ± 0.4		8.4 ± 0.4	
6.8 ± 0.4	10.6 ± 0.4	*a*	10.1 ± 0.4	*a*	8.5 ± 0.4	*b*	9.7 ± 0.4	*a*
6.8 ± 0.6	3.5 ± 0.2		3.7 ± 0.3		3.9 ± 0.3		3.0 ± 0.2	
5.6 ± 0.5	4.6 ± 0.3	*ab*	4.8 ± 0.2	*a*	4.5 ± 0.3	*a*	3.5 ± 0.3	*b*
1.6 ± 0.4	3.7 ± 0.3	*b*	4.3 ± 0.3	*b*	5.0 ± 0.3	*a*	2.4 ± 0.3	*c*
0.0 ± 0.0	0.6 ± 0.3	*b*	0.4 ± 0.3	*b*	4.2 ± 0.6	*a*	0.0 ± 0.0	*b*
- - -	4.5 ± 0.4	*a*	3.9 ± 0.4	*ab*	3.5 ± 0.4	*b*	2.3 ± 0.2	*c*
- - -	5.6 ± 0.3	*a*	4.9 ± 0.3	*b*	4.3 ± 0.3	*c*	3.3 ± 0.3	*d*
- - -	4.3 ± 0.5	*a*	4.0 ± 0.4	*a*	4.1 ± 0.4	*ab*	3.2 ± 0.3	*b*
0.7 ± 0.3	2.2 ± 0.5		2.4 ± 0.5		2.1 ± 0.5		2.1 ± 0.4	
1.5 ± 0.3	5.5 ± 0.3	*a*	5.6 ± 0.3	*a*	4.7 ± 0.3	*b*	5.4 ± 0.2	*a*
- - -	6.1 ± 0.4	*ab*	5.0 ± 0.4	*b*	2.9 ± 0.4	*c*	7.3 ± 0.5	*a*
4.0 ± 0.4	4.0 ± 0.3	*a*	4.0 ± 0.2	*a*	4.3 ± 0.4	*a*	3.4 ± 0.3	*a*
1.6 ± 0.6	5.5 ± 0.5	*a*	4.0 ± 0.5	*ab*	2.9 ± 0.5	*b*	5.3 ± 0.4	*a*
10.6 ± 0.7	8.9 ± 0.4		8.2 ± 0.4		8.5 ± 0.3		8.2 ± 0.4	
- - -	- - -		- - -		- - -		- - -	
5.7 ± 0.5	5.3 ± 0.4	*a*	5.0 ± 0.5	*ab*	5.5 ± 0.5	*a*	4.7 ± 0.5	*b*
0.4 ± 0.2	5.5 ± 0.3	*a*	4.6 ± 0.3	*b*	6.2 ± 0.4	*a*	4.1 ± 0.3	*b*
6.2 ± 0.5	5.5 ± 0.3		5.5 ± 0.3		5.5 ± 0.4		5.5 ± 0.4	
4.9 ± 0.4	4.6 ± 0.4	*b*	5.0 ± 0.4	*ab*	5.5 ± 0.5	*a*	4.5 ± 0.4	*b*

Table 3.3: Continued; results for potato without and with mayonnaises are shown.

	Mayonnaise		Carrier		MxC	
	F	p	F	p	F	p
(A) EMG / Jaw Tracking						
Eating behavior						
Number of chews	18.6	**<0.001**	8.2	**0.005**	0.2	0.922
Chewing time (s)	16.0	**<0.001**	2.9	0.090	0.5	0.694
Chewing cycle duration (s)	3.6	**0.015**	22.5	**<0.001**	0.2	0.867
Opening duration (s)	6.5	**<0.001**	23.5	**<0.001**	2.6	0.052
Closing duration (s)	0.4	0.764	12.3	**<0.001**	0.2	0.918
Power stroke duration (s)	1.1	0.345	67.0	**<0.001**	0.6	0.599
Chewing frequency (chew/s)	3.1	**0.026**	20.1	**<0.001**	0.3	0.824
Eating rate (g/min)	18.0	**<0.001**	102.7	**<0.001**	0.9	0.420
Jaw movements						
Opening veloctiy (mm/s)	1.0	0.388	0.7	0.412	0.9	0.443
Closing velocity (mm/s)	2.0	0.108	13.8	**<0.001**	0.6	0.628
Vertical movement (mm)	5.1	**0.002**	20.8	**<0.001**	0.6	0.642
Anterior posterior movement (mm)	1.2	0.323	6.7	**0.010**	0.1	0.969
Medial lateral movement (mm)	1.5	0.222	1.5	0.230	1.0	0.392
Muscle activities						
Total activity per sequence	14.2	**<0.001**	17.4	**<0.001**	1.0	0.392
Total activity per chew	1.0	0.374	157.8	**<0.001**	1.6	0.193
Temporalis activity per sequence	10.5	**<0.001**	27.3	**<0.001**	1.1	0.343
Temporalis activity per chew	0.2	0.924	146.4	**<0.001**	1.2	0.318
Temporalis activity per chew (working-side)	0.2	0.918	124.4	**<0.001**	1.4	0.234
Temporalis activity per chew (balancing-side)	0.2	0.912	135.4	**<0.001**	0.8	0.505
Masseter activity per sequence	13.8	**<0.001**	31.4	**<0.001**	1.0	0.376
Masseter activity per chew	1.4	0.252	165.6	**<0.001**	2.0	0.116
Masseter activity per chew (working-side)	1.0	0.379	132.3	**<0.001**	1.6	0.202
Masseter activity per chew (balancing side)	1.2	0.301	121.7	**<0.001**	1.6	0.197
Digastric activity per sequence	13.0	**<0.001**	0.7	0.420	0.2	0.911
Digastric activity per chew	3.4	**0.019**	37.0	**<0.001**	0.5	0.685
Digastric activity per chew (working-side)	3.2	**0.025**	31.2	**<0.001**	0.8	0.489
Digastric activity per chew (balancing-side)	2.6	0.056	22.7	**<0.001**	0.4	0.752

Data on single potato (i.e. without mayonnaise) are presented for illustration purposes (light-gray column), and were not included in the linear mixed models. Significant p values (<0.05) are highlighted in bold. Different letters indicate significant differences between carrier-mayonnaise combinations. The abbreviations are explained in Table 3.1 and the sensory attributes are explained in Table 3.2.

Potato	Potato FF-HV		Potato LF-HV-starch		Potato LF-HV-xanthan		Potato LF-LV	
Mean ± SE	Mean ± SE		Mean ± SE		Mean ± SE		Mean ± SE	
20 ± 1	17 ± 1	*b*	18 ± 1	*b*	19 ± 1	*a*	16 ± 1	*b*
12.5 ± 0.7	10.1 ± 0.7	*bc*	11.1 ± 0.8	*b*	11.9 ± 0.5	*a*	9.7 ± 0.7	*c*
0.61 ± 0.01	0.62 ± 0.01	*ab*	0.64 ± 0.02	*ab*	0.64 ± 0.02	*a*	0.61 ± 0.01	*b*
0.21 ± 0.01	0.22 ± 0.01	*ab*	0.23 ± 0.01	*a*	0.23 ± 0.01	*a*	0.21 ± 0.01	*b*
0.26 ± 0.01	0.27 ± 0.01		0.27 ± 0.01		0.27 ± 0.01		0.27 ± 0.01	
0.14 ± 0.01	0.14 ± 0.01		0.14 ± 0.01		0.14 ± 0.01		0.14 ± 0.01	
1.6 ± 0.0	1.6 ± 0.0	*ab*	1.6 ± 0.0	*ab*	1.6 ± 0.0	*b*	1.6 ± 0.0	*a*
35 ± 2	64 ± 3	*b*	61 ± 3	*b*	51 ± 3	*c*	70 ± 4	*a*
40 ± 7	39 ± 7		37 ± 7		37 ± 7		36 ± 7	
36 ± 6	34 ± 6		32 ± 6		34 ± 6		30 ± 6	
15.8 ± 0.3	16.4 ± 0.4	*a*	16.5 ± 0.4	*a*	16.3 ± 0.4	*ab*	15.6 ± 0.4	*b*
6.5 ± 0.8	6.5 ± 0.6		7.0 ± 0.8		6.9 ± 0.7		5.9 ± 0.5	
7.1 ± 0.3	7.3 ± 0.4		7.5 ± 0.4		7.5 ± 0.4		7.2 ± 0.4	
48 ± 3	39 ± 2	*b*	41 ± 3	*b*	48 ± 3	*a*	38 ± 3	*b*
2.4 ± 0.1	2.5 ± 0.1		2.4 ± 0.1		2.4 ± 0.1		2.5 ± 0.1	
16 ± 1	13 ± 1	*b*	13 ± 1	*b*	16 ± 1	*a*	13 ± 1	*b*
0.80 ± 0.04	0.80 ± 0.05		0.76 ± 0.04		0.80 ± 0.04		0.84 ± 0.05	
0.41 ± 0.02	0.41 ± 0.03		0.38 ± 0.02		0.40 ± 0.02		0.43 ± 0.02	
0.39 ± 0.02	0.39 ± 0.03		0.38 ± 0.02		0.40 ± 0.02		0.41 ± 0.02	
15 ± 1	12 ± 1	*b*	13 ± 1	*b*	16 ± 1	*a*	12 ± 1	*b*
0.77 ± 0.03	0.78 ± 0.04		0.76 ± 0.04		0.78 ± 0.03		0.79 ± 0.04	
0.38 ± 0.02	0.39 ± 0.02		0.38 ± 0.02		0.38 ± 0.02		0.39 ± 0.02	
0.39 ± 0.02	0.39 ± 0.02		0.38 ± 0.02		0.40 ± 0.02		0.40 ± 0.02	
17 ± 1	14 ± 1	*bc*	15 ± 1	*b*	16 ± 1	*a*	13 ± 1	*c*
0.85 ± 0.03	0.87 ± 0.03	*ab*	0.84 ± 0.04	*a*	0.84 ± 0.03	*ab*	0.85 ± 0.03	*b*
0.43 ± 0.02	0.43 ± 0.02	*ab*	0.42 ± 0.02	*a*	0.42 ± 0.02	*ab*	0.42 ± 0.02	*b*
0.42 ± 0.02	0.44 ± 0.02		0.42 ± 0.02		0.42 ± 0.02		0.43 ± 0.02	

Table 3.3: Continued

	Mayonnaise		Carrier		MxC	
	F	p	F	p	F	p
(B) Bolus properties						
Compositional bolus properties						
Moisture content at swallowing (%wt)	613.1	**<0.001**	1819.7	**<0.001**	10.6	**<0.001**
Saliva content at swallowing (g/g dry product)	7.6	**<0.001**	80.0	**<0.001**	3.3	**0.024**
Fat content at swallowing (g/g dry product)	1268.9	**<0.001**	48.5	**<0.001**	4.1	**0.009**
Mechanical bolus properties						
Peak force at swallowing (N)	10.0	**<0.001**	14.7	**<0.001**	2.3	0.079
Adhesiveness at swallowing (N·s)	65.2	**<0.001**	117.7	**<0.001**	1.2	0.312
Cohesiveness at swallowing	19.8	**<0.001**	148.0	**<0.001**	11.9	**<0.001**
Tribological bolus properties						
Friction coefficient at swalllowing (8 mm/s)	10.9	**<0.001**	469.3	**<0.001**	12.2	**<0.001**
(C) Sensory characteristics						
Odour attribute						
Overall intensity	0.3	0.842	2.5	0.116	0.6	0.634
Taste attribute						
Overall intensity	11.1	**<0.001**	0.7	1.000	2.6	0.051
Mouthfeel attributes						
Dry	2.6	0.051	2.4	1.000	0.1	0.931
Firm	4.1	**0.008**	0.7	0.398	2.7	0.051
Sticky	28.4	**<0.001**	0.5	0.461	0.7	0.525
Gummy	35.8	**<0.001**	2.0	0.155	1.0	0.414
Creamy	20.9	**<0.001**	1.4	0.245	0.0	0.986
Fatty	46.0	**<0.001**	0.6	1.000	0.2	0.923
Velvet	6.0	**<0.001**	2.2	1.000	0.7	0.537
Smooth	0.4	0.776	1.2	0.281	0.3	0.833
Salivating	5.4	**0.001**	2.1	1.000	0.5	0.651
Absorbing	26.3	**<0.001**	3.2	1.000	8.9	**<0.001**
Chewing effort	0.7	0.540	0.4	0.514	2.8	0.044
Homogeneous	7.6	**<0.001**	0.0	0.886	3.8	**0.012**
Bread fibers*	1.8	0.151	-	-	-	-
Potato particles*	1.7	0.169	-	-	-	-
Afterfeel attributes						
Residue	3.6	**0.015**	0.3	1.000	0.7	0.564
Fatty film layer	24.6	**<0.001**	0.1	1.000	0.7	0.556
Dry Rough	0.3	0.797	0.3	1.000	0.4	0.741
Cleaning	5.1	**0.002**	0.0	1.000	0.5	0.668

* The sensory attribute bread fibers was assessed for bread samples only. The sensory attribute potato particles was assessed for potato samples only.

Data on single potato (i.e. without mayonnaise) are presented for illustration purposes (light-gray column), and were not included in the linear mixed models. Significant p values (<0.05) are highlighted in bold. Different letters indicate significant differences between carrier-mayonnaise combinations. The abbreviations are explained in Table 3.1 and the sensory attributes are explained in Table 3.2.

Potato	Potato FF-HV		Potato LF-HV-starch		Potato LF-HV-xanthan		Potato LF-LV	
Mean ± SE	Mean ± SE		Mean ± SE		Mean ± SE		Mean ± SE	
83.0 ± 0.6	65.7 ± 0.4	*b*	79.8 ± 0.4	*a*	80.1 ± 0.5	*a*	80.2 ± 0.4	*a*
1.51 ± 0.18	0.73 ± 0.04	*b*	1.17 ± 0.11	*a*	1.02 ± 0.12	*ab*	1.03 ± 0.10	*a*
	60 ± 1	*a*	27 ± 1	*b*	23 ± 1	*c*	29 ± 1	*b*
0.13 ± 0.01	0.06 ± 0.01	*b*	0.06 ± 0.01	*b*	0.10 ± 0.01	*a*	0.09 ± 0.02	*ab*
0.009 ± 0.001	0.008 ± 0.001	*b*	0.003 ± 0.000	*c*	0.017 ± 0.001	*a*	0.006 ± 0.001	*bc*
0.29 ± 0.01	0.41 ± 0.02	*b*	0.28 ± 0.02	*c*	0.52 ± 0.02	*a*	0.38 ± 0.02	*b*
0.70 ± 0.03	0.52 ± 0.02	*b*	0.45 ± 0.01	*b*	0.67 ± 0.02	*a*	0.48 ± 0.01	*b*
10.1 ± 0.3	8.5 ± 0.2		8.5 ± 0.3		8.8 ± 0.3		8.5 ± 0.3	
6.9 ± 0.4	8.3 ± 0.3	*a*	8.5 ± 0.3	*a*	7.6 ± 0.3	*b*	8.1 ± 0.4	*a*
7.7 ± 0.3	5.1 ± 0.4		5.0 ± 0.4		5.4 ± 0.3		4.7 ± 0.4	
6.9 ± 0.4	5.9 ± 0.4	*ab*	6.4 ± 0.4	*a*	6.5 ± 0.3	*a*	6.1 ± 0.4	*b*
1.9 ± 0.4	3.5 ± 0.4	*b*	3.6 ± 0.4	*b*	5.0 ± 0.3	*a*	2.4 ± 0.2	*c*
1.4 ± 0.7	1.8 ± 0.6	*b*	1.9 ± 0.6	*b*	5.0 ± 0.5	*a*	2.1 ± 0.7	*b*
	3.9 ± 0.5	*a*	3.3 ± 0.4	*ab*	2.7 ± 0.4	*b*	1.6 ± 0.3	*c*
	5.7 ± 0.3	*a*	5.1 ± 0.4	*b*	4.7 ± 0.4	*c*	3.5 ± 0.4	*d*
	4.3 ± 0.5	*a*	3.5 ± 0.4	*a*	3.2 ± 0.4	*ab*	2.8 ± 0.3	*b*
0.1 ± 0.1	1.3 ± 0.5		1.0 ± 0.4		0.8 ± 0.3		1.1 ± 0.3	
3.1 ± 0.4	4.6 ± 0.3	*a*	4.4 ± 0.3	*a*	4.0 ± 0.3	*b*	4.5 ± 0.4	*a*
	4.5 ± 0.5	*a*	4.1 ± 0.4	*ab*	2.8 ± 0.5	*b*	3.9 ± 0.5	*ab*
5.6 ± 0.4	5.1 ± 0.4	*a*	5.4 ± 0.4	*a*	5.1 ± 0.4	*a*	5.4 ± 0.4	*a*
0.9 ± 0.3	4.8 ± 0.5	*a*	4.4 ± 0.4	*a*	4.0 ± 0.6	*a*	4.1 ± 0.4	*a*
- - -	- - -		- - -		- - -		- - -	
11.4 ± 0.4	9.2 ± 0.4		8.8 ± 0.4		8.8 ± 0.4		9.3 ± 0.4	
6.4 ± 0.6	5.8 ± 0.5	*a*	5.4 ± 0.5	*ab*	5.4 ± 0.5	*a*	4.8 ± 0.4	*b*
0.4 ± 0.2	5.4 ± 0.3	*a*	4.6 ± 0.4	*b*	5.6 ± 0.3	*a*	3.8 ± 0.4	*b*
6.5 ± 0.3	5.5 ± 0.4		5.8 ± 0.5		6.0 ± 0.3		5.5 ± 0.4	
5.4 ± 0.5	4.9 ± 0.4	*b*	5.0 ± 0.4	*ab*	5.5 ± 0.4	*a*	5.0 ± 0.4	*b*

draw firm conclusions on the effect of absorption or speed of mixing on eating behavior. The reason why different ratios were chosen was that these weight ratios are naturally applied by consumers when preparing these composite foods and hence represent realistic consumption context (van Eck *et al.* 2019b; van Eck *et al.* 2019c). Thus, when considering naturally applied condiment:carrier weight ratios, condiments assist bolus formation of relatively dry foods such as bread more than carrier foods with a high moisture content such as cooked potatoes.

Effect of condiment properties on eating behavior of composite foods

When comparing mayonnaises differing in fat content (FF-HV versus LF-HV-starch), no significant differences in eating behavior were observed (Table 3.3A). Hence, as eating behavior is not affected by fat content, replacing a full-fat condiment by a low fat alternative (~2743 vs. 1125 kJ/100 mL) can lower energy intake without affecting eating behavior

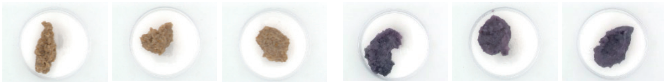
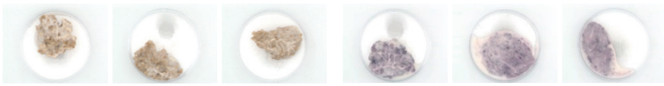
When comparing mayonnaises differing in viscosity (LF-HV-starch versus LF-LV), carriers with low viscosity mayonnaise were swallowed after slightly shorter chewing time than those with high viscosity mayonnaise (from 10±1 to 9±1 s for bread and from 11±1 to 10±1 s for potato, $p=0.033$). This is likely due to the faster diffusion of low viscosity mayonnaise than the high viscosity version into and throughout the bread bolus, leading to faster moistening. Although these differences seem relatively small, we have to consider that carrier-condiment combinations are not eaten as a single bite but as part of a meal. Consequently, over the consumption of multiple bites, adding condiments with a lower viscosity result in faster eating rate (from 46±3 to 55±4 g/min for bread and from 61±3 to 70±4 g/min for potato; Figure 3.1) which might lead to higher food intake than when condiments with a higher viscosity are chosen.

3.3.2 Effect of condiments on bolus properties and sensory perception of carrier foods

Bread versus potato: Effect of condiment addition on bolus formation and sensory perception

Addition of mayonnaises changes the bolus properties and sensory perception of bread and cooked potato (Table 3.3B, 3.3C). Throughout consumption, mayonnaises were mixed with carrier foods leading to structural changes. Typical pictures of expectorated boli are shown in Table 3.4 for illustration purposes. Part of the mayonnaise was present on the outer surface of the bolus throughout the entire process of mastication, by which it could contribute to the lubriaction of saliva. Indeed, with the presence of mayonnaise, the coefficient of friction decreased (Figure 3.2) and less saliva incorporation was required before swallowing (Figure 3.3). The addition of mayonnaise also led to a decrease in peak force for the bread bolus (Figure 3.4A) and an increase in cohesiveness for potato bolus (Figure 3.4D). These results indicate that the mechanism by which condiments assist saliva in bolus formation differ between bread and potato. This is most likely due to the water absorption capability of these products. In the case of bread, the saliva/mayonnaise diffuses into the bread, where

Table 3.4: Representative pictures of expectorated boli at three time points throughout mastication (33, 66 and 100% of total mastication time) for bread (left) and cooked potato (right) without and with different mayonnaises. Chewing times before expectoration (s), moisture content (wt%), saliva content (g/g dry weight) and fat content (wt%) are reported for each sample and time point (mean ± standard error of the mean).

	Bread boli			**Potato boli**		
Sample	**33%**	**66%**	**100%**	**33%**	**66%**	**100%**
Without						
Chewing time (s)	7	14	21	6	12	17
Moisture content (%)	54±1	57±1	60±2	82±0	83±0	83±0
Saliva content (g/g dry weight)	0.4±0.0	0.6±0.1	0.9±0.1	1.3±0.1	1.7±0.1	1.5±0.2
Fat content (%)	-	-	-	-	-	-
						
FF-HV						
Chewing time (s)	6	11	17	5	11	16
Moisture content (%)	42±1	45±1	48±1	65±1	66±1	66±1
Saliva content (g/g dry weight)	0.4±0.0	0.4±0.0	0.4±0.0	0.6±0.0	0.7±0.0	0.7±0.0
Fat content (%)	-	-	54±1	-	-	60±1
						
LF-HV-starch						
Chewing time (s)	6	11	17	5	10	15
Moisture content (%)	63±1	64±1	67±1	77±1	79±0	80±0
Saliva content (g/g dry weight)	0.3±0.1	0.5±0.1	0.7±0.1	0.8±0.1	1.0±0.1	1.2±0.1
Fat content (%)	-	-	25±1	-	-	27±1
						
LF-HV-xanthan						
Chewing time (s)	6	12	17	6	11	17
Moisture content (%)	62±0	65±1	66±1	79±0	79±1	80±0
Saliva content (g/g dry weight)	0.2±0.0	0.4±0.1	0.5±0.0	0.6±0.1	0.9±0.1	1.0±0.1
Fat content (%)	-	-	19±1	-	-	23±1
						
LF-LV						
Chewing time (s)	5	9	14	5	10	15
Moisture content (%)	62±1	65±1	67±0	79±1	80±0	80±0
Saliva content (g/g dry weight)	0.2±0.0	0.4±0.1	0.6±0.0	0.7±0.1	0.9±0.1	1.0±0.1
Fat content (%)	-	-	28±1	-	-	29±1
						

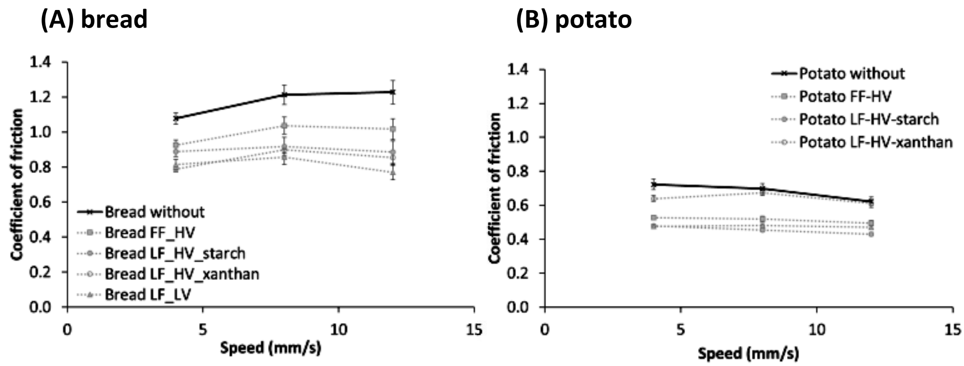


Figure 3.2: Bolus friction coefficient at the moment of swallowing as a function of speed for bread (A) and cooked potato (B) without and with different mayonnaises. Error bars represent standard error of the mean. The abbreviations are explained in Table 3.1.

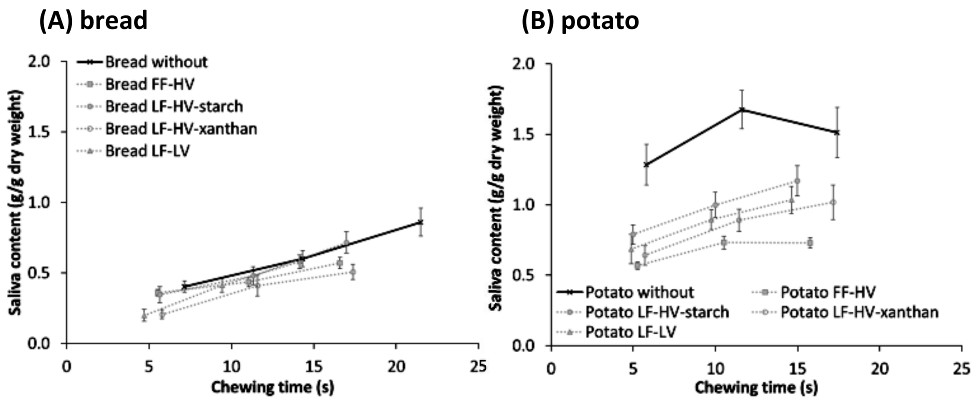


Figure 3.3: Bolus saliva content at the moment of swallowing for bread (A) and cooked potato (B) without and with different mayonnaises. Error bars represent standard error of the mean. The abbreviations are explained in Table 3.1.

it provides the moistening and softening of the bread to form a compact moistened bolus without really falling apart into pieces. In the case of potato, no direct saliva incorporation is obtained as potato already contains a high water content. Instead, the potato is first broken down in multiple pieces. Saliva and additional moisture is then used to adhere bolus pieces together to form a safe-to-swallow bolus. For bread and potato, saliva facilitates bolus formation, but by different mechanisms. Regardless of the mechanism, bolus properties for safe swallowing were reached after shorter chewing times with the addition of mayonnaise for bread and potato. Apparently, also when moisture is used for adherence, low viscosity condiments are beneficial, as they can easily spread between potato boli fragments. Consequently, due to moisture incorporation consumers perceived carriers with mayonnaises as smoother (from 0.7±0.3 to 2.2±0.2 for bread, from 0.1±0.1 to 1.1±0.2 for potato), less dry (from 6.8±0.6 to 3.5±0.1 for bread, from 7.7±0.3 to 5.1± 0.2 for potato), less firm (from 5.6±0.5

to 4.4±0.1), and the fibrous structure of bread was perceived to a lesser extent (from 10.6±0.7 to 8.4±0.2). In addition, the presence of individual potato pieces was perceived to a lesser extent (from 11.4±0.4 to 9.0±0.2), indicating increased cohesiveness perception. Hence, the measured bolus properties were in line with the perceived sensory characteristics.

Effect of condiment properties on bolus properties and sensory perception of composite foods

When comparing mayonnaises differing in fat content (*i.e.* FF-HV versus LF-HV-starch), bolus composition distinctly differed in moisture and fat content. As expected, bolus with FF-HV mayonnaise was higher in fat, whereas the bolus with LF-HV-starch mayonnaise was higher in moisture content (Table 3.3B, Table 3.4). As no differences in eating behavior were observed between full-fat and low-fat mayonnaise for both bread and potato (Figure 3.1), this indicates that fat and moisture facilitate bolus formation of these carriers to a similar extent. In the case of bread, this is probably logical, as the diffusion of the mayonnaise

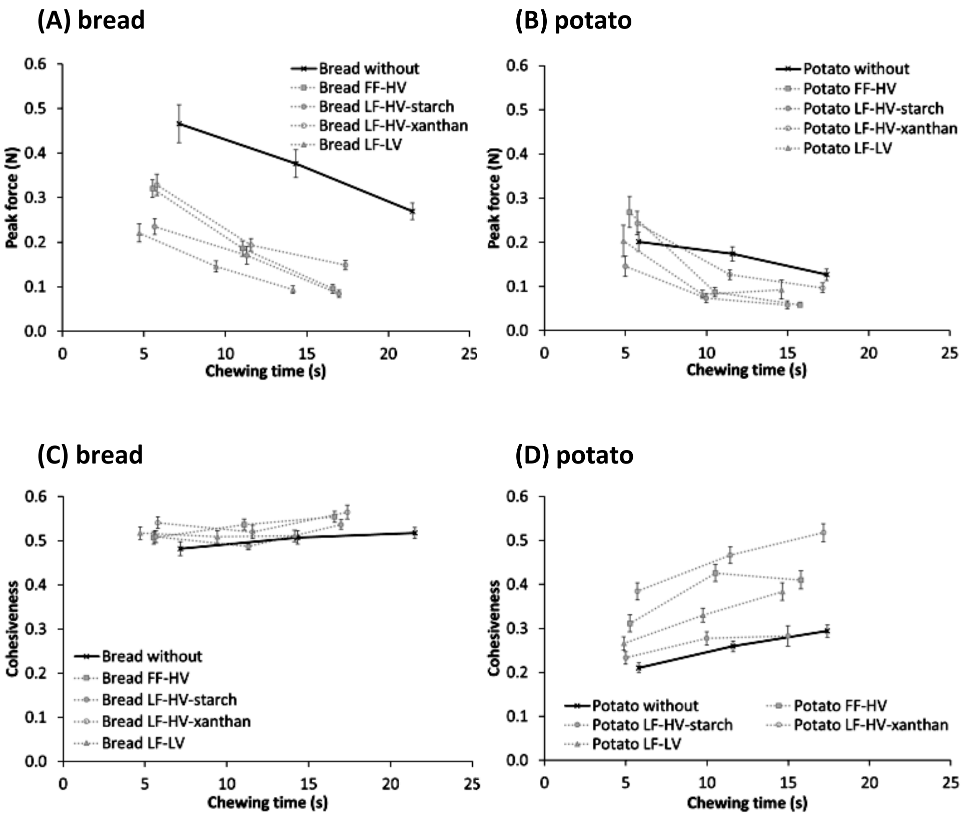


Figure 3.4: Bolus mechanical properties (peak force, cohesiveness) throughout mastication for bread (A, C) and cooked potato (B, D) without and with different mayonnaises. Peak force refers to bolus firmness, cohesiveness refers to the degree to which food sticks together. Error bars represent standard error of the mean. The abbreviations are explained in Table 3.1.

is driven by viscosity, which is the same for both mayonnaises. For the potato, larger differences could have been expected. As the mayonnaise in this case is used as a "glue" to adhere the particles together, the fat content could have had a beneficial effect. Apparently in this case, the composition of the mayonnaise does not change the affinity towards the potato to change adherence. Also with respect to sensory perception, little differences were observed, as 18 out of 20 attributes were not significantly influenced by mayonnaise fat content (Table 3.3C). Only the attribute fatty (during consumption and after swallowing) was significantly different.

The previous results on eating behavior (Figure 3.1, section 3.3.1) indicate that viscosity seems to be more important in bolus formation than fat content. When comparing mayonnaises differing in viscosity (*i.e.* LF-HV-starch versus LF-LV), some changes in bolus properties throughout mastication were indeed found. In case of bread, bolus peak force was lower in the presence of low viscosity mayonnaise than in the presence of high viscosity mayonnaise (Figure 3.4A), already at an early stage of consumption. Low viscosity mayonnaise is able to penetrate into the bread bolus faster than high viscosity mayonnaise, as low viscosity liquids have a higher diffusion rate and are more prone to capillary action by the small pores in the bread structure (Hicsasmaz & J.T. 1995). This leads to faster penetration, by which coefficient of friction is more effectively reduced (Figure 3.2A) and a safe-to-swallow bolus is formed after shorter time. This was evidently perceivable, as bread carriers with low viscosity mayonnaises were rated significantly less firm (3.5±0.3 compared to 4.8±0.2) and more absorbing (7.3±0.5 compared to 5.0±0.4) than with high viscosity mayonnaise. In the case of potatoes, bolus cohesiveness increased to a larger extent at an earlier stage of consumption with the addition of low viscosity mayonnaise compared to high viscosity mayonnaise (Figure 3.2D). This indicates that low viscosity mayonnaise mixed easier with the potato pieces to form a cohesive and safe-to-swallow bolus after shorter times. Apparently, also in this case, the viscosity is the driving force for fast adherence of particles and bolus formation. As no differences between firmness were observed (Figure 3.2B), this indicates that swallowing of potato-condiment combinations may be more related to cohesiveness and not to bolus firmness.

Regarding sensory perception, it is important to mention that the sensory evaluations were largely impacted by mayonnaises viscosity (Table 3.3C). Sensory perception therefore seems to be more dominated by viscosity than by fat content. In the case of bread, 7 out of 20 attributes were significantly different for LF-HV-starch and LF-LV mayonnaise, and 5 out of 20 attributes for potatoes. In particular, carrier-mayonnaise combinations were perceived as significantly less creamy, fatty and velvety when low viscosity mayonnaise was added. These attributes are desired mouthfeel attributes contributing to food appreciation (Richardson-Harman *et al.* 2000; Ares *et al.* 2010). Thus, decreasing condiment viscosity probably leads to a lower food appreciation.

Role of lubrication in swallowing and sensory perception of composite foods
Lubrication behavior of foods has been suggested by many others to influence both the ease of swallowing and smoothness perception (Stokes *et al.* 2013; Campbell *et al.* 2017a; Sarkar & Krop 2019). Figure 3.2 shows the friction coefficients for the carrier boli mixed with different mayonnaises. These results show that lower friction coefficients are obtained for composite foods with added mayonnaise compared to the carriers without mayonnaise. So in addition to faster diffusion and mixing, mayonnaises can also aid bolus formation and swallowing by increasing lubrication (lower friction). We also examined whether lubrication behavior was linked to specific sensory attributes. The coefficient of friction correlated to perceived smoothness (r = -0.82 for bread, r = -0.85 for potato), perceived creaminess (r = -0.62 for bread, r = -0.62 for potato) and dry, rough after-feel perception (r = 0.91 for bread, r = 0.92 for potato).

To better understand how condiments affect eating behavior and smoothness perception of composite foods, it is desired to gain information on the lubrication ability of the ingredients present in the food boli (*i.e.* the role of fat content and viscosity). Although condiment properties tend to impact tribological behavior of bread-mayonnaise combinations (Figure 3.2A), this effect is not clearly observed in potato-mayonnaise combinations (Figure 3.2B). It is not yet clear how fat and moisture are distributed throughout the bolus and how that influences lubrication. In addition, for similar moisture content, particle sizes of boli fragments can influence lubrication (Fuhrmann *et al.* 2020). We hypothesize that dry foods such as bread soften by moisture uptake by which lubrication is more effectively supported than by adding fat. In the case of potato, either water or fat is used to keep the particles together without such a softening effect. Therefore, less effect of condiment properties on lubrication may have been observed.

3.3.3 Practical implications
Our results showed that addition of any mayonnaise to carrier foods contributed to faster eating rates by providing bolus softening (bread), adherence of bolus pieces together (potato) and/or lubrication (bread, potato). Faster eating rate can lead to higher energy food intake (de Graaf & Kok 2010; Robinson *et al.* 2014; McCrickerd & Forde 2017). Historically, condiments are high in fat and energy. Nevertheless, providing condiments to a meal might help consumers to promote healthy eating. For instance, addition of small amounts of condiments to vegetables or whole grain staple foods (that are generally less liked due to its flavor or texture) might increase their intake so that recommended daily intakes are reached. In addition, when targeting the elderly population at risk of malnutrition due to impaired oral processing capacities (Hickson 2006; Pauly *et al.* 2007), addition of condiments to solid foods assists bolus formation and can consequently contribute to a desired increase in food intake.

Modest nuances in energy intake of composite foods could be achieved by changing specific condiment properties. As summarized in the MFA individuals map (Figure 3.5), the fat content of condiments did not have a large impact on eating behavior, bolus properties nor sensory characteristics of carrier-condiment combinations, as the FF-HV and LF-HV-starch are positioned close to each other. Fat content is inherently related to energy density of foods, and full fat condiments also bring an increase in both fat and calorie intake, which is usually not desirable. The present study highlights that changing the fat content of condiments did not affect eating behavior and only modestly affected the sensory perception of composite foods.

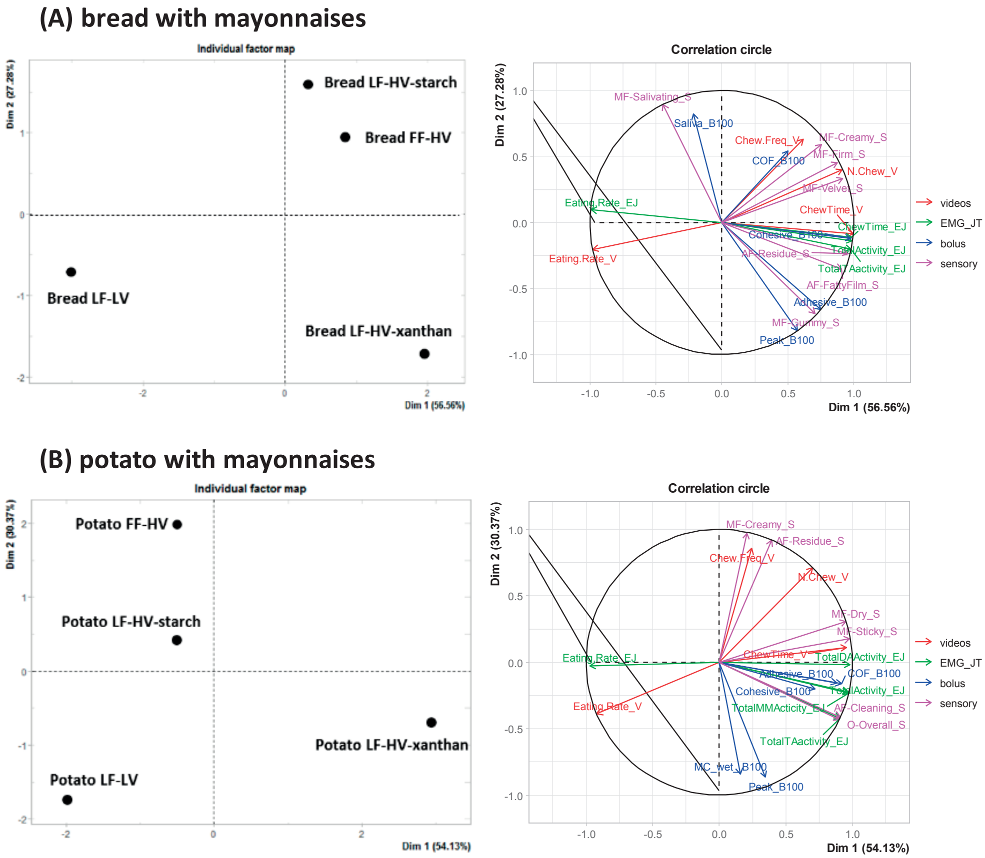


Figure 3.5: Multiple Factor Analysis (MFA) on the four different datasets (video recordings, EMG and jaw tracking, bolus properties at the moment of swallowing, and sensory characteristics) for bread with different mayonnaises (A) and cooked potato with different mayonnaises (B). Only those parameters with a significant mayonnaise effect during mixed models were considered (Table 3.3). The individuals map (samples) is shown on the left, and the variables map (parameters) is shown on the right. Different colors indicate different datasets, and only the 20 variables with the highest contribution are displayed in words. The abbreviations are explained in Table 3.1.

These data indicate that replacing full-fat with low fat condiments, when assessed with carrier foods, could be a promising strategy to reduce fat and thereby energy intake among the general population in a relatively unconscious way. Changing the viscosity of condiments, on the other hand, had a larger effect on both the chewing behavior and sensory perception of the carrier-condiment combinations (Figure 3.5), as LF-HV-starch and LF-LV mayonnaises are positioned further apart. Carriers with low viscosity mayonnaise were chewed with fewer chews for a shorter time, resulting in faster bread softening and faster potato adherence, and were perceived as less creamy, fatty and velvety than the high viscosity mayonnaise combinations. Consequently, changing the viscosity of condiments influences eating rate (this is desired to impact intake), but this is also likely to affect food appreciation. Such changes may be used to target different intake behavior of consumer groups. For example, increasing viscosity with less fat could be used to decrease food intake by increasing eating rate, adding low viscosity condiments to vegetables may increase uptake of vegetables of children, and adding low viscosity condiments may increase total food intake of elderly.

3.4 CONCLUSIONS

Consumers frequently combine foods that vary greatly in composition, structure and energy density. This study shows that addition of condiments facilitates bolus formation of solid carrier foods, regardless of specific condiment properties. All condiments assisted saliva by increasing lubrication (lower coefficient of friction, less saliva incorporation) and impacting the degree of bolus structure (decreased firmness of bread bolus, increased cohesiveness of potato bolus) allowing the bolus to be safely swallowed after fewer chews and shorter chewing times. Although number of chews and chewing time were largely decreased with the addition of condiments, subjects did not adapt their muscle activities per chew nor jaw movements. When comparing the influence of different condiment properties (fat content, viscosity), only small effects on eating behavior were observed. Condiment fat content did not influence eating behavior. Condiment viscosity influenced eating behavior and less viscous condiments were chewed with a higher eating rate (g/min). Viscosity was shown to play an important role in the bolus formation due to two different events: (1) water absorption for dry foods to soften the bolus, and (2) using moisture to adhere separate bolus particles into a bolus.

We conclude that eating behavior of carrier-condiment combinations is mainly affected by the presence of condiments and to a much smaller extent by the specific properties of the condiment. These results suggest that addition of condiments might largely alter food intake, which could be an effective strategy to increase healthy eating or to decrease or increase food intake in different consumer groups.

MAYONNAISE

Shape up! How shape, size and addition of condiments influence eating behavior towards vegetables

4

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Food & Function, 2019, 10: 5739

ABSTRACT

Background and objective: Practical approaches to increase consumption of healthy foods such as vegetables are needed. This study determined the influence of shape, size and condiment properties on eating behavior of carrots.

Methods: Eating behavior (mastication time, number of chews, chewing frequency, eating rate) was determined for carrots with same total weight but different shapes (cube, julienne), and varying in size, number of pieces and aspect ratio. To investigate the effect of condiment addition on eating behavior of carrots, mayonnaises varying in fat content and viscosity were combined with carrots, and mastication behavior and bolus properties were determined.

Results: Carrots presented in one large cube required the lowest mastication effort (shortest mastication time, fewest chews) among all pre-cut carrots. Carrot cubes required less mastication effort leading to higher eating rates than carrots julienne. Mayonnaises, in particular those with high fat content or low viscosity, contributed to faster bolus formation of carrots. Carrots were swallowed with less particles of larger sizes when mayonnaises were added. These results indicate that a specific particle size is not a prerequisite to induce swallowing, and that other bolus properties such as lubrication or cohesiveness trigger the urge to swallow.

Conclusions: We conclude that eating behavior of carrots can be controlled by relatively small changes in both carrot and condiment properties. To increase carrot intake by increasing eating rate, we suggest to avoid cutting of carrots or to add condiments, which could be an effective strategy to increase vegetable consumption or to decrease mastication effort to target the elderly population.

Key words: shape, size, condiments, eating behavior, bolus properties

4.1 INTRODUCTION

Vegetables are considered healthy because of their low energy content and their high vitamin, mineral, phytochemical and fiber content (Slavin & Lloyd 2012). Therefore, vegetable consumption is beneficial for human health. People who eat vegetables as part of their daily diet have a reduced risk of many chronic diseases such as obesity, cardio-vascular diseases and certain forms of cancer (Wang *et al.* 2014). Although the beneficial health effects of vegetables are well-known, the actual recommended daily intake of vegetables is still not met by the majority of the population (Diethelm *et al.* 2012; Vereecken *et al.* 2015). In the Netherlands, the mean vegetable intake was 127 g/day in 2012-2014, and only 15% of Dutch adults met the recommended amount of 200 g/day (Van Rossum *et al.* 2016; Van Rossum *et al.* 2017). Practical approaches to increase the consumption of vegetables are needed, and therefore a better understanding of the factors that influence vegetable intake is necessary.

One of the approaches to increase vegetable intake is by controlling eating rate, as faster eating rates (g/min) have been related to higher food intake (Robinson *et al.* 2014). Eating rates are known to depend on both consumer characteristics (Devezeaux de Lavergne *et al.* 2015a; Ketel *et al.* 2019) and food properties (Forde *et al.* 2013b; Forde *et al.* 2017; van den Boer *et al.* 2017). Unfortunately, consumer characteristics such as age, gender and ethnicity are fixed and cannot be influenced. Food properties such as food texture and shape, on the other hand, can be modified, which can be used as an effective strategy to influence eating rate and intake of vegetables.

Vegetables naturally differ in shape and size, and consumers use multiple preparation techniques such as cutting, cooking, steaming and baking before vegetable consumption. These preparation techniques often soften the vegetables. It is well-known that softer foods require lower mastication efforts (mastication time, number of chews, muscle activity), are consumed at higher eating rates leading to higher intake, than harder foods (Bolhuis *et al.* 2014a; Campbell *et al.* 2017b; Lasschuijt *et al.* 2017; Aguayo-Mendoza *et al.* 2019). However, less is known about the effect of shape and size of vegetables on eating behavior and *ad libitum* intake. Kohyama *et al.* (2007) investigated eating behavior of carrot cubes (20x20x20 mm, ~7 g) and finely cut carrots (fine strips of 1x1.5x30 mm, ~7 g). Finely cut carrots required longer mastication time, more chews with a higher muscle activity, and were consumed at lower eating rates than carrot cubes (Kohyama *et al.* 2007). However, it is not well understood why there is a difference in eating behavior between carrot cubes and carrots julienne. The effect of carrot shape on bolus formation was not determined in this study. Investigating the bolus properties of carrots varying in shape might help to explain the observed differences in eating behavior. Recently, Goh *et al.* (2017) and Liem *et al.* (2019) investigated the influence of vegetable size on vegetable intake. Intake of whole carrots (160x30x20 mm, ~70 g) and carrot cubes (20x20x20 mm, ~10 g) by children was determined in a controlled school setting and a real-life movie setting. Intake of whole carrots was higher than intake of carrot cubes in both settings (consumption increased with 75% in the movie setting), demonstrating that carrot shape influences carrot intake (Goh *et al.* 2017; Liem & Russell 2019). However, it is not clearly understood how the size of vegetables influences eating behavior and bolus formation. Taking the mastication behavior and bolus properties of vegetables into consideration allows to better understand the mechanisms underlying vegetable intake. Based on this knowledge, practical approaches to increase the consumption of vegetables might be devised.

Vegetables are frequently consumed together with condiments, for example with dips (vegetables cut in beams), as Russian salad (vegetables cooked and cut in small cubes) or coleslaw (vegetables cut julienne). Little is known about the effect of condiment addition and its properties on eating behavior of vegetables. In the case of carrier foods with relatively low water content such as toast or crackers, it is known that condiments facilitate bolus formation by moistening and softening food boli, which leads to shorter mastication times to break down and lubricate boli enough to be safely swallowed (Gavião *et al.* 2004; Engelen *et al.* 2005; van Eck *et al.* 2019b). The shorter mastication times are due to the ability of toast and crackers to absorb moisture from saliva or condiments, which softens these foods by increasing moisture content (van Eck *et al.* 2019b). Vegetables are quite different from toast and crackers, as they have a high moisture content and are speculated to not absorb moisture from saliva or condiments during mastication. Therefore, vegetables require different oral processing strategies than toast and crackers (Jalabert-Malbos *et al.* 2007; Chen *et al.* 2013). The impact of condiments on bolus formation is expected to be different for vegetables than for toast or crackers. Instead of acting as a food softener, the condiments will just be mixed with the vegetables. Further understanding of the effect of condiment properties on mastication behavior of vegetables such as carrots may be used to control food intake of different vegetable-condiment combinations.

The objective of this study was to determine the effect of shape, size and addition of condiments differing in physicochemical properties on eating behavior of carrots. We chose mayonnaise as a representative for the condiment, as it is often used with carrots to make salads or used as an ingredients for dips. Raw carrots were cut into cubes and julienne pieces of various sizes and aspect ratios, which were assessed with and without mayonnaise. Carrots cut into cubes or julienne represent simplified model foods for vegetables with dip and vegetable salads such as coleslaw. We hypothesize that carrots varying in shape are consumed with different mastication efforts and eating rates leading to different bolus properties at the moment of swallowing. The addition of mayonnaise to carrots is hypothesized to stick carrot pieces together by which it aids the bolus formation of carrots.

4.2 MATERIALS AND METHODS

4.2.1 Carrot preparation

An overview of all carrot samples is presented in Table 4.1. Fresh raw carrots (Daucus carota; winter carrot; purchased from local retailer in Wageningen, Netherlands) were peeled and cut into cubes (15x15x15 mm) and julienne pieces (90x3x2 mm), which were further cut to obtain cube and julienne samples varying in number and size of carrot pieces. All samples were served at similar total weight of approximately 4.2 g. For the carrot cubes, carrots were cut into one large cube, eight medium-sized cubes, 27 small-sized cubes and 64 very small-sized cubes using a knife. The sample codes used for the cubes are C_1, C_8, C_27 and C_64, in which the first letter refers to the shape (Cube C), and the number refers to the number of pieces. For the julienne carrots, carrots were cut using a spiral slicer (Gefu Spiral Slicer Spirelli 2.0) and a knife into eight long julienne pieces, 24 medium-long julienne pieces and 64 short julienne pieces. The sample codes used for the carrot julienne are J_8, J_24 and J_64, in which the first letter refers to the shape (Julienne J), and the number refers to the number of pieces. All carrot samples were prepared freshly the morning of the session and vacuum packed until serving to minimize water loss.

4.2.2 Mayonnaise preparation

The mayonnaise formulations and properties are summarized in Table 4.2. Mayonnaises were prepared using sunflower oil, egg yolk, lemon juice and mustard. All ingredients were purchased from a local retailer in Wageningen, the Netherlands. Three mayonnaises (full fat/high viscosity, low fat/high viscosity, low fat/low viscosity) varying in fat content (77 and 28% w/w) and viscosity (high viscosity: approximately 190 Pa·s at 1s-1 ; low viscosity: approximately 21 Pa·s at 1s-1) were prepared. The sample codes used for the mayonnaises are FF_HV, LF_HV, and LF_LV, in which the first two letters refer to full fat (FF) or low fat (LF), and the last two letters to high viscosity (HV) or low viscosity (LV). Mayonnaises were prepared at room temperature (20 ± 1 ºC) using a mixer (Thermomix, Vorwerk, Germany, speed 3). For the FF_HV mayonnaise, 16 g egg yolk, 3 g lemon juice and 5 g mustard were mixed and stirred for 10 s, after which 80 g sunflower oil was gradually added in 3 min. For the low-fat mayonnaises (LF_HV and LF_LV), first xanthan (5 g for LF_HV and 2 g for LF_LV) (E415, Pit&Pit bvba, Belgium) was gradually added to 50 g water under continuous stirring, and the mixture was heated at 80 ºC for 5 minutes until all xanthan was dissolved. The solution was cooled down to room temperature, and then, 16 g egg yolk, 3 g lemon juice and 5 g mustard were added and mixed for 10 s. 30 g sunflower oil was then added slowly in 3 min. All mayonnaises were mixed for 2 min after addition of the oil.

Table 4.1: Overview of carrot samples varying in shape, number and size.

Sample code	C_1	C_8|	C_27	C_64	J_8	J_24	J_64
Carrot images							
Shape	cube	cubes	cubes	cubes	julienne	julienne	julienne
Number of pieces	1	8	27	64	8	24	64
Size of pieces	large	medium	small	very small	long	medium	short
Length of one piece (mm)	15	7.5	5	3.75	90	30	11
Width of one piece (mm)	15	7.5	5	3.75	3	3	3
Height of one piece (mm)	15	7.5	5	3.75	2	2	2
Aspect ratio of one piece[a]	1	1	1	1	30	10	4
Surface area of one piece (mm^2)	1350	338	150	84	912	312	125
Total surface area of all pieces (mm^2)	1350	2700	4050	5400	7296	7488	7968
Total weight of all pieces (g)	4.2	4.2	4.2	4.2	4.2	4.2	4.2

[a] Aspect ratio is here defined as the length of one piece divided by the width of one piece.

Table 4.2: A) Formulations of mayonnaises and relative concentrations of all ingredients (% w/w) and B) properties of mayonnaises varying in fat content and viscosity (mean±SD).

	Full Fat / High Viscosity (FF_HV)		Low Fat / High Viscosity (LF_HV)		Low Fat / Low Viscosity (LF_LV)	
(A) Formulation	*Mass (g)*	*Concentration (% w/w)*	*Mass (g)*	*Concentration (% w/w)*	*Mass (g)*	*Concentration (% w/w)*
Sunflower oil	80	*77*	30	*28*	30	*28*
Egg yolk	16	*15*	16	*15*	16	*15*
Lemon juice	3	*3*	3	*3*	3	*3*
Mustard	5	*5*	5	*5*	5	*5*
Xanthan	-	-	5	*5*	2	*2*
Water	-	-	50	*46*	50	*47*
(B) Mayonnaise properties						
Fat content (% w/w)	77		28		28	
Viscosity at 1 s^{-1} (Pa·s)	186±79		193±37		21±5	
Viscosity at 10 s^{-1} (Pa·s)	29±8		15±3		4±1	
Viscosity at 100 s^{-1} (Pa·s)	5±1		2±1		1±0	

To characterize the flow properties of the three mayonnaises, flow curves were determined using a rheometer (Anton Paar Rheometer 301) equipped with an Inset I-PP50/SS plate and a CP50-1 cone. The viscosity of the mayonnaises was measured by putting 0.5 g mayonnaise on the plate, applying a resting period of 5 min, and then shearing at shear rates ranging from 1 s-1 to 1000 s-1 in 5 min (30 measurement points of 10 s each). Measurements were performed for each new batch (n=5) in duplicate at room temperature (20 ± 1 ºC).

4.2.3 Carrots varying in shape

The mastication behavior of carrots varying in shape, number of pieces and size of pieces (Table 4.1) was determined. 17 subjects (7 male and 10 female, between 20 and 34 years old) participated. All subjects were European, Caucasian healthy adults with good dental health (self-reported), non-smoking habits (self-reported) and frequent carrot consumers (self-reported). Each subject attended one session of 45 min during which mastication behavior was determined using video recordings (section 4.2.5). The mastication behavior of four carrot cubes (C_1, C_8, C_27 and C_64) and three julienne carrots (J_8, J_24 and J_64) was quantified. This yielded a total of 7 samples, which were assessed in duplicate. All samples were served at a weight of 4.2 g. Samples were served on a spoon in randomized order following a completely randomized design. Samples were coded with 3 digit codes. Subjects cleaned their palate after consumption of each sample with water and crackers. Subjects gave written informed consent and received financial compensation for participation.

4.2.4 Carrots without and with mayonnaises varying in fat content and viscosity

The mastication behavior and bolus properties of carrots differing in shape without and with mayonnaises varying in fat content and viscosity (FF_HV, LF_HV and LF_LV) were determined. Carrot cubes (C_1) and carrots julienne (J_8) were used, since these carrots were the most different with respect to mastication behavior (see section 4.3.1). 20 subjects (9 male and 11 female, between 18 and 25 years old) participated. All subjects were European, Caucasian healthy adults with good dental health (self-reported), non-smoking habits (self-reported), frequent carrot consumers (self-reported), and no allergies or intolerances for mayonnaises. Seven of the subjects also participated in the previous test (carrots varying in shape), whereas 13 participants performed only this one. Each subject attended three sessions of 60 min over a time period of four weeks. Mastication behavior was determined in duplicate in the first session (section 4.2.5). Bolus properties (particle size distribution, retention of carrot in the bolus) were determined in the following two sessions (section 4.2.6). Carrots (C_1, J_8) without and with mayonnaises (FF_HV, LF_HV, LF_LV) were assessed, which gave a total of 8 samples. Carrots were served at approximately 4.2 g and carrots with added mayonnaises at approximately 6.2 g (4.2 g carrot with 2.0 g mayonnaise). This carrot/mayonnaise weight ratio corresponds to a common vegetable/mayonnaise serving ratio used to prepare coleslaw. For carrot cubes, mayonnaise was added on top of the cubes. For carrots julienne, carrots were premixed with the mayonnaises in a plastic container before serving. Samples were served on a spoon in randomized order following a completely randomized design. Samples were coded with 3 digit codes. Subjects cleaned their palate after consumption of each sample with water and crackers. Subjects gave written informed consent and received financial compensation for participation.

4.2.5 Characterization of mastication behavior using video recordings

Mastication behavior (total mastication time, number of chews, chewing frequency and eating rate) was characterized in duplicate using video recordings, as described by Aguayo-Mendoza *et al.* (2019) and van Eck *et al.* (2019) (Aguayo-Mendoza *et al.* 2019; van Eck *et al.* 2019b). Subjects were seated in a chair with a video camera placed in front of them at a distance of approximately 50 cm. At the start of the session, the researcher placed two stickers with a distance of 5 cm on the subjects' forehead, one sticker on the subjects' nose tip, one sticker on the subjects' chin, and one sticker on the subjects' Adam's apple. These stickers were used as reference points during the video analyses. Subjects were instructed to chew each sample as they would do naturally, to maintain their head straight to the camera, not to block their face with their hand, and to raise their hand to indicate the moment of swallowing.

Video analyses were performed using Kinovea software (version 0.8.15) to determine total mastication time (s), number of chews and chewing frequency (chews/s). Each video was calibrated by setting the distance between the two stickers on the forehead as 5 cm. The

Y coordinates of the nose and chin stickers were extracted over time. All analyses were performed at 15 frames per second.

Total mastication time (s) was defined as the time period from placing the sample in the mouth (*i.e.* the moment the lips were closed after placing the food in the mouth) until the moment of swallowing (*i.e.* the moment that the subjects raised their hand to indicate swallowing, verified by a movement of the Adam's apple sticker). Subjects generally took multiple swallows during mastication, and only the main swallow (*i.e.* the moment they swallowed the bolus) was used to determine total mastication time. The number of chews was determined by tracking the difference in Y coordinates between the nose and the chin sticker (*i.e.* vertical displacement) during total mastication time. Chewing frequency was calculated by dividing the number of chews by the mastication time. Eating rate (g/min) was calculated by dividing the sample weight by the total mastication time in minutes.

4

4.2.6 Characterization of bolus properties

Subjects were instructed to chew the samples and to expectorate the bolus at the moment of swallowing (*i.e.* 100% of total mastication time). This time point was determined for each sample by averaging the mastication times of all subjects and all replicates obtained during the video recordings. Boli were collected in pre-weighed containers covered with lids to prevent moisture evaporation from the samples. Immediately after bolus expectoration, subjects were instructed to take a sip of water, to thoroughly rinse their mouth and to expectorate the debris in a separate container.

Carrot retention and loss

The containers with expectorated bolus were weighed to determine the weight of the total bolus. After weighing, the bolus was sieved with a 1 mm sieve with water for 1 min, washed with acetone, and left in the sieve for 1 min. This process caused the mayonnaise and/or saliva to pass through the sieve and the majority of carrot particles to be retained. Hence, carrot particles less than 1 mm were discarded (typically less than 1% of all carrot particles). The retained carrot particles were weighed in order to determine the weight of carrot retained in the bolus (mb). This approach (*i.e.* sieving, washing, drying and weighing) was also applied to the debris samples to determine the weight of carrot retained in the debris (md). Mass of lost carrots (mlost) due to swallowing was calculated using $m_{lost}=m_0-(m_b+m_d)$, where m_0 is the weight of carrot before consumption (*i.e.* 4.2 g). This calculation was based on the assumption that the bolus was fully consumed (typically no carrot pieces were left on the spoon).

Particle size distribution

Following carrot retention and loss, particle size distribution of carrots in the bolus was determined using image analysis. For each bolus, the researcher placed two times 0.5 g of carrot particles obtained from the bolus in a petri dish (120x120x17 mm). Hence, 1 g of the total bolus (0.5 g in duplicate) was used for particle size characterization. Individual particles were separated from each other using a spatula. The petri dishes were placed on a flatbed scanner (Canon CanoScan 9000F MarkII) and a color picture with a black background was taken. Pictures were imported into ImageJ (version 1.51f, National Institute of Health, USA) to conduct particle size analysis. Pictures were converted to an 8-bit image, after which a black and white threshold was applied to obtain a binary image. For each image, the number of particles, average d50 particle area (mm^2) and average particle roundness were obtained. Since carrot particles were not spherical, the area rather than diameter was taken as a measure to represent particle size.

4.2.7 Data analysis

Results were reported as mean values with standard error. Outliers (Z-score>3.29) were removed from the data (typically less than 1.2% of all values). To investigate the mastication behavior of carrots varying in shape, size and number, linear mixed models were performed with shape as fixed effect and subject, replicate and serving order as random effects using Lmer package (Kuznetsova *et al.* 2016). In addition, Pearson's product-moment correlations (r) were used to determine the relationships between carrot shape properties (number of pieces, surface area, aspect ratio) and mastication behavior parameters (mastication time, number of chews, eating rate). To investigate the addition of mayonnaises on mastication behavior and bolus properties of carrots, linear mixed models were performed with mayonnaise, shape and mayonnaise:shape as fixed effects and subject as random effect using Lmer package (Kuznetsova *et al.* 2016). In addition, Principal Component Analysis (PCA) was performed on the bolus properties data, in which the mastication behavior parameters (total mastication time, number of chews) were plotted as supplementary variables using SensorMineR package (Husson *et al.* 2014). R language (RStudio, version 1.0.143) was used to perform all statistical tests. Significance level of $p<0.05$ was chosen.

4.3 RESULTS AND DISCUSSION

4.3.1 Influence of shape, number and size of carrot pieces on mastication behavior

Figure 4.1 shows the total mastication time (A), the number of chews until swallowing (B) and the eating rate (C) for the different carrot samples. All samples had similar weight (approximately 4.2 g), so the differences can be attributed to a separate effect of the shape, size or number of pieces. Total mastication time ($F=25.5$, $p<0.001$), number of chews required to swallow carrots ($F=15.3$, $p<0.001$) and eating rate ($F=21.1$, $p<0.001$) were significantly influenced by the way carrots were cut before consumption. Even though subjects adapted the time and number of chews to a large extent, their chewing frequencies did not differ between samples varying in the number, size and shape of carrot pieces ($F=0.6$, $p=0.695$). Chewing frequency of all carrot samples was about 1.4 chews/s. This is in line with previous studies on eating behavior of raw carrots (Kohyama *et al.* 2007; Forde *et al.* 2013b; Aguayo-Mendoza *et al.* 2019).

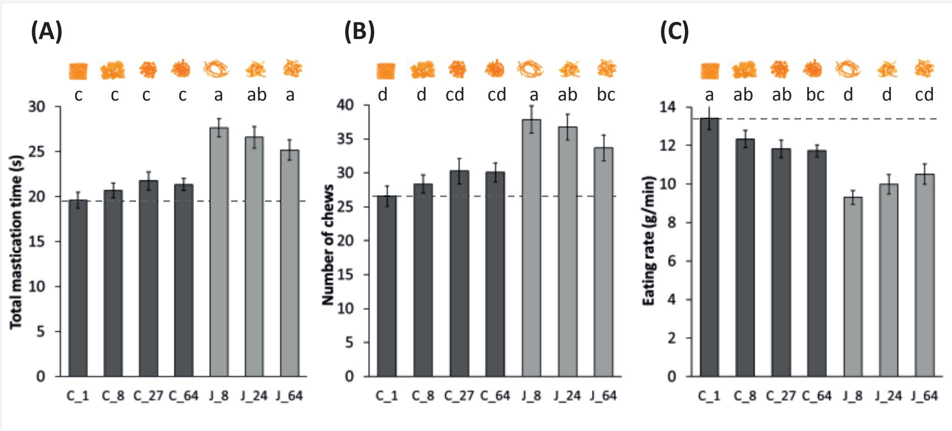

Figure 4.1: Total mastication time (A), number of chews until swallowing (B) and eating rate (C) for carrots varying in shape, number and size of carrot pieces. The sample codes refer to the shape (C for cube, J for julienne) and the number of carrot pieces (Table 4.1). Carrots cut in cubes are presented in dark gray, carrots cut julienne in light gray. Mean values (n=17 subjects, in duplicate) together with standard error of the mean are shown. The dashed line represents the mean values of one carrot cube (C_1). Different letters indicate significant differences between means (p<0.05). Pictures of the carrot samples are presented for illustration purposes.

Carrots cut into one cube (C_1) were consumed with the shortest chewing time (20±1 s), fewest chews (27±2) and highest eating rate (13.4±0.6 g/min) among all samples tested. Cutting carrots into multiple smaller cubes (C_8, C_27 and C_64) did not significantly affect mastication times (21±1s, 22±1s and 21±1s), numbers of chews (28±1, 30±2, 30±1) and eating rates (12.3±0.4 g/min, 11.8±0.5 g/min, 11.7±0.3 g/min) compared to C_1. Much larger effects were observed when carrots were cut into julienne pieces (J_8, J_24 and J_64), leading to increased total mastication times, increased number of chews and decreased eating rates. Consumption of J_8, J_24 and J_64 required 28±1 s and 38±2 chews (p<0.001, p<0.001), 27±2 s and 37±2 chews (p<0.001, p<0.001) and 25±1 s and 34±2 chews (p<0.001, p<0.001), respectively. Eating rates of J_8, J_24 and J_64 were 9.3±0.4, 10.0±0.5 and 10.5±0.5 g/min, respectively. This result indicates that cutting carrots, in particular into julienne shape, increases rather than reduces mastication effort required to form a bolus that is safe to swallow and consequently reduces eating rate. This observation might be explained by the selection function, which describes that larger particles are more likely to be selected by the molars to be further broken-down during chewing than smaller particles (Lucas & Luke 1983; Van der Glas *et al.* 1987). Pre-cut carrots have a higher number of smaller particles already at the beginning of mastication, which leads to a lower particle selection chance and consequently the consumption of smaller carrot pieces requires a higher mastication effort to break down all particles to a certain size to form a bolus that is safe to swallow. Similar findings were reported by Kohyama *et al.* (2007), who found that finely cutting carrots increased consumers mastication effort (Kohyama *et al.* 2007). However, for other foods such as gelatin gels, surimi gels or roast pork, no effect of pre-cutting on the mastication effort has been observed (Kohyama *et al.* 2005; Kohyama *et al.* 2007; Kim *et al.* 2015). This discrepancy could be attributed to the different mechanical properties of these foods. In the case of soft and tough foods such as gels or roast pork, swallowing might be triggered more by factors such as cohesiveness or lubrication than by particle size, and consequently initial particle size and number have little effect on mastication effort. However, in the case of harder foods such as carrots, particle size is a main factor in swallowing determination (Peyron *et al.* 2004; Jalabert-Malbos *et al.* 2007). Harder foods require smaller bolus particles before the bolus can be swallowed. Consumers therefore use more time and effort to break down all initial particles to a smaller size and to bind these particles together into a cohesive bolus that is safe to swallow (Nishinari *et al.* 2019).

A comparison of carrot cubes and julienne carrots with similar number of pieces reveals that shape alters the mastication behavior of carrot pieces to a large extent. Carrot cubes were chewed for shorter time, with fewer chews and with higher eating rate than julienne carrots (Figure 4.1). Samples consisting of 8 (C_8), 27 (C_27) and 64 (C_64) carrot cubes were chewed for 21±1, 22±1 and 21±1 s, whereas this was 28±1, 27±1 and 25±1 s for samples consisting of 8 (J_8), 24 (J_24) and 64 (J_64) julienne pieces, respectively (Figure 4.1A). This effect was significant for all sample comparisons; containing 8 pieces ($p<0.001$), 27 versus 24 pieces ($p<0.001$) and the 64 smallest pieces ($p=0.002$). The difference in mastication time is also reflected in differences in number of chews and eating rates. C_8, C_27 and C_64 required 28±1, 30± and 30±1 number of chews and were consumed at eating rates of 12.3±0.4, 11.8±0.5 and 11.7±0.3 g/min, whereas this was 38±2, 37±2 and 34±2 chews and 9.3±0.4, 10.0±0.5 and 10.5±0.5 g/min for J_8, J_24 and J_64 (Figure 4.1B, 4.1C). The number of pieces was not significantly correlated with total mastication time ($r=0.05$, $p=0.446$), number of chews ($r=0.04$, $p=0.501$) and eating rate ($r=-0.09$, $p=0.154$)

These differences in oral processing parameters are related to the shape of the pieces, and might be explained by differences in aspect ratio or surface area (Table 4.1). To evaluate how these factors are correlated, Figure 4.2 shows mastication time, number of chews and eating rate versus aspect ratio and total surface area of all pieces. Aspect ratio and total surface area of all pieces are both positively correlated with mastication time (aspect ratio: $r=0.37$, $p<0.001$; surface area: $r=0.42$, $p<0.001$), number of chews (aspect ratio: $r=0.30$, $p<0.001$; surface area: $r=0.33$, $p<0.001$) and eating rate (aspect ratio: $r=-0.34$, $p<0.001$; surface area: $r=-0.41$, $p<0.001$). Surface area seems to be stronger correlated with the eating behavior measures than aspect ratio. Aspect ratio and total surface area of all pieces are higher for julienne than for cubes. A higher aspect ratio and surface area would increase the possible contact between the pieces and the teeth, and thereby increase the biting chance. However, this does not appear to decrease mastication effort (total mastication time, number of chews). In the case of multiple smaller julienne pieces, they are distributed throughout the oral cavity and more effort is therefore required to place them between the teeth before

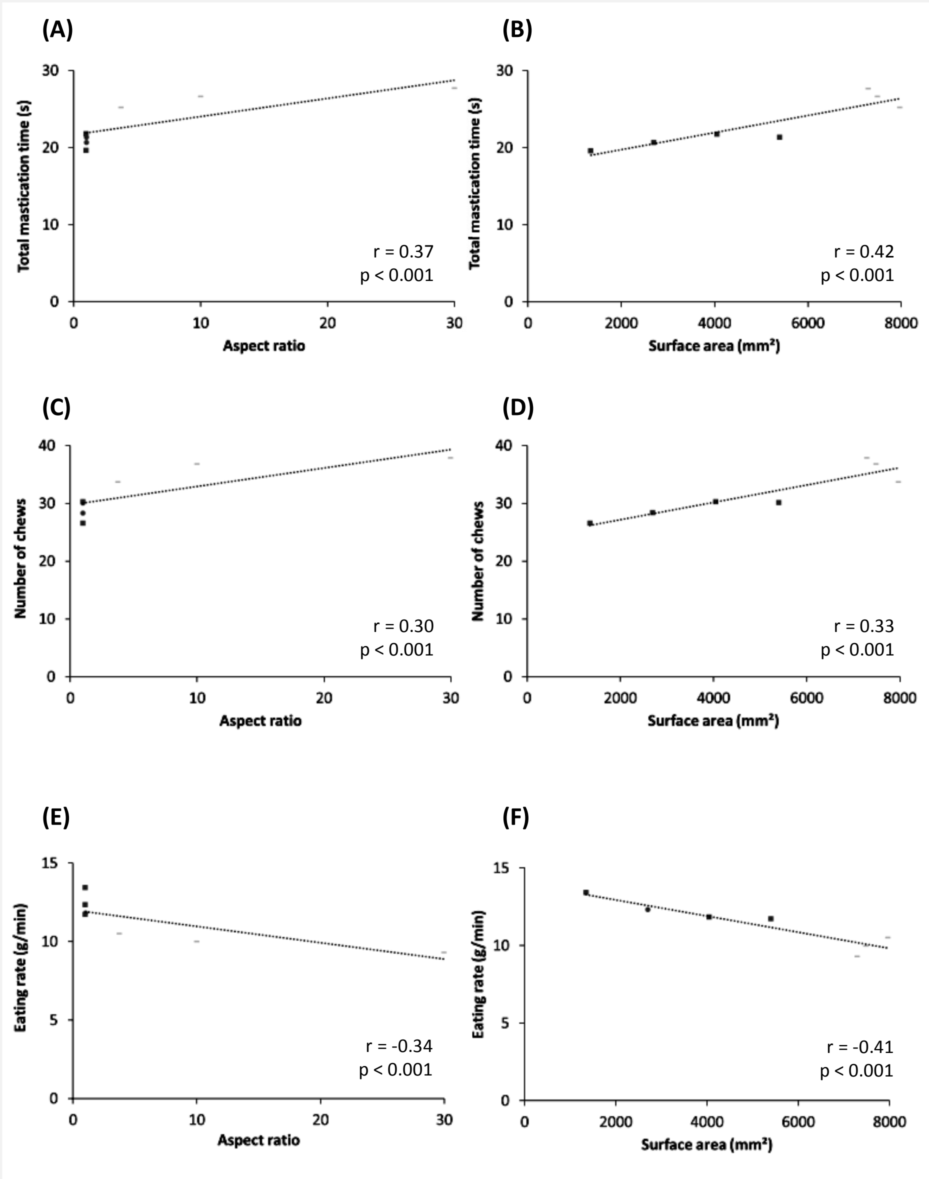

Figure 4.2: Correlations among the carrot shape properties aspect ratio (on the left) and total surface area of all pieces (on the right) and the mastication behavior parameters total mastication time (A, B), number of chews (C, D) and eating rate (E, F). The dark-gray squares (■) represent the cube samples and the light-gray bars (▬) represent the julienne samples. Pearson's product-moment correlations (r) and p-values are shown in the graph.

one can bite. In addition, in the case of a higher surface area, also more saliva (and thus more mastication time) might be needed to paste the julienne particles together to form a bolus that is lubricated and cohesive enough to be safely swallowed. Thus, food particles with a high aspect ratio and/or a high surface area are more difficult to form a safe-to-swallow bolus than those with a low aspect ratio and/or a low surface area. Together, these data highlight that shape has a large impact on mastication effort required to safely swallow raw carrots and consequently also on eating rate. A controlled eating rate could potentially influence vegetable intake. Specific implications of these findings will be discussed later in more detail (section 4.3.3).

4.3.2 Effect of addition of mayonnaises varying in fat content and viscosity on oral processing behavior of carrots

To investigate the effect of condiment addition on oral processing behavior, mayonnaises that differed in viscosity and fat content were added to carrots (one large cube, C_1 and 8 julienne pieces, J_8). Although mastication effort (total mastication time, number of chews until swallowing, eating rate) of carrots (C_1, J_8) with mayonnaises was mainly driven by the carrot properties, an effect of mayonnaise properties was also observed for mastication time, number of chews and eating rate (mayonnaise effect; $F=10.1$, $p<0.001$ for total mastication time; $F=5.1$, $p=0.002$ for number of chews; $F=115.8$, $p<0.001$ for eating rate). Addition of mayonnaise to carrots did not affect chewing frequencies (around 1.3 to 1.4 chews/s) of carrot cubes and carrots julienne ($F=1.4$, $p=0.257$). As shown in Figure 4.3, addition of mayonnaise reduced total mastication time, number of chews until swallowing and increased eating rate of carrots. Although the effects were larger for carrots julienne than for carrot cubes (shape effect; $F=262.3$, $p<0.001$ for total mastication time; $F=133.3$, $p<0.001$ for number of chews; $F=203.5$, $p<0.001$ for eating rate), mayonnaise properties (fat content, viscosity) influenced mastication behavior of C_1 and J_8 carrots in a similar way (no mayonnaise:shape interactions; $F=1.4$, $p=0.257$ for total mastication time; $F=0.9$, $p=0.439$ for number of chews; $F=0.7$, $p=0.576$ for eating rate). When comparing mayonnaises varying in fat content and viscosity, a significant decrease in total mastication time of carrots was observed for full fat (FF_HV) and low viscosity (LF_LV) mayonnaise ($p<0.001$, $p<0.001$), but not for low fat high viscosity (LF_HV) mayonnaise ($p=0.489$). For example, total mastication time of J_8 decreased from 25±1 to 21±1, 24±1 and 23±1 s after addition of FF_HV, LF_HV and LF_LV mayonnaise, respectively. This difference in mastication time is also reflected in differences in number of chews and eating rates (Figure 4.3B, 4.3C). Our results show that mayonnaises can aid mastication of carrots and increase eating rate, and that this effect is not driven by mayonnaise fat content alone nor viscosity alone.

When comparing the high viscosity mayonnaises varying in fat content (FF_HV and LF_HV), the results indicate that full fat mayonnaise aids mastication of carrots more than low fat mayonnaise. It could be argued that oil increased lubrication or that oil adhered the

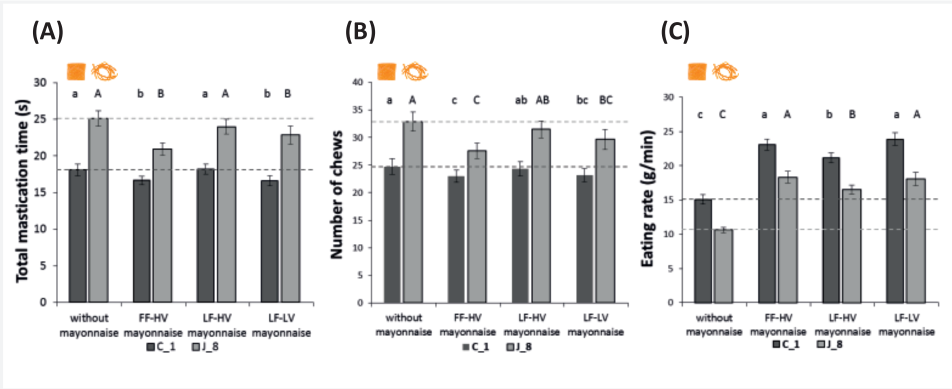

Figure 4.3: Total mastication time (A), number of chews until swallowing (B) and eating rate (C) for carrot cube C_1 (dark gray) and carrots julienne C_8 (light gray) without mayonnaise and with different mayonnaises (**F**ull **F**at/**H**igh **V**iscosity, **L**ow **F**at/**H**igh **V**iscosity, **L**ow **F**at/**L**ow **V**iscosity). Carrots alone were served at approximately 4.2 g, and carrots with mayonnaise were served at approximately 6.2 g (*i.e.* 4.2 g carrot, and 2 g mayonnaise). Error bars represent standard error of the mean. Dashed lines represent mean values of carrots without mayonnaise (n=20 subjects, in duplicate). Different lower case letters indicate significant differences between cube (C_1) without or with different mayonnaises (p<0.05). Different capital letters indicate significant differences between julienne (J_8) without or with different mayonnaises (p<0.05).

carrot fragments more efficiently together than water, and subsequently triggered earlier swallowing. Another possible explanation might be that the presence of a relative high amount of xanthan in the LF_HV mayonnaise slowed down bolus formation. Although the mayonnaise had a similar viscosity profile as the FF-HV mayonnaise (Table 4.2), the LF_HV mayonnaise had a gummy structure, possibly causing slower adherence of the mayonnaise to the carrot particles. As a consequence, a higher effort was required to adhere carrot particles together compared to that of the full fat version, by which more time was needed to form a bolus that is safe to swallow. When comparing the low fat mayonnaises varying in viscosity (LF_HV and LF_LV), a low viscous mayonnaise shortened mastication of carrots more than the high viscous mayonnaise. This could be attributed to faster carrot particle adherence due to the lower viscosity, and consequently triggered swallowing after shorter mastication times. Hence, mayonnaises with a high fat content or a low viscosity adhered to and lubricated carrot particles better, leading to shorter mastication times to form a bolus that is safe enough to be swallowed. Specific implications of these findings will be discussed later in more detail (section 4.3.3).

The condiments seem to influence the eating behavior by affecting the bolus formation. To gain more insights into this effect, the carrot bolus properties (particle size, particle retention) at the moment of swallowing were determined (Figure 4.4, Table 4.3). In case of carrots without mayonnaise, 0.5 g bolus of a carrot cube (C_1) consisted of 74±4 particles with a d50 of 4.8±0.3, whereas carrot julienne (J_8) consisted of 155±6 particles with a d50 of 3.9±0.2 mm^2. The presence of mayonnaises clearly influenced carrot particle size distributions of both carrot cubes and carrots julienne (Figure 4.4, Table 4.3). Carrots were broken down into a bolus consisting of fewer particles of larger sizes when any of the mayonnaises were added. On average, the influence of mayonnaise on the bolus properties of carrots was larger for carrots julienne (J_8) than carrot cubes (C_1) (mayonnaise:shape interaction effects; $F=14.2$, $p<0.001$ for particle count; $F=7.0$, $p<0.001$ for particle size; $F=3.3$, $p=0.024$ for carrot retention in bolus). In the case of carrots julienne (J_8) with FF_HV, LF_HV and LF_LV mayonnaise, 0.5 g of bolus consisted of 117±6, 97±4 and 114±4 particles with a d50 of 5.9±0.3, 7.5±0.4 and 5.8±0.3 mm^2, respectively, which are fewer particles of much larger sizes than in the bolus without mayonnaise (155±6 particles with a d50 of 3.9±0.2 mm^2). In addition, the presence of mayonnaise increased the mass of carrot retained in the bolus for carrots julienne (J_8)(Table 4.3). Consequently, addition of mayonnaise decreased the mass of carrot lost (mlost) during chewing carrots julienne. These results show that mayonnaises assist saliva in bolus formation of carrots. This is consistent with our previous hypothesis that mayonnaises increase bolus cohesiveness by adhering carrot pieces together and at the same time provide lubrication. Due to the larger surface area of the julienne, more particles were adhered by the mayonnaise and consequently included in the bolus. Due to the incorporation of the mayonnaise into the bolus, the carrot boli can be safely swallowed at larger particle sizes after shorter eating times. Although condiments are known to facilitate bolus formation of dry, low moisture content foods such as bread or crackers (Gavião *et al.* 2004; Engelen *et al.* 2005; van Eck *et al.* 2019b), this is the first time

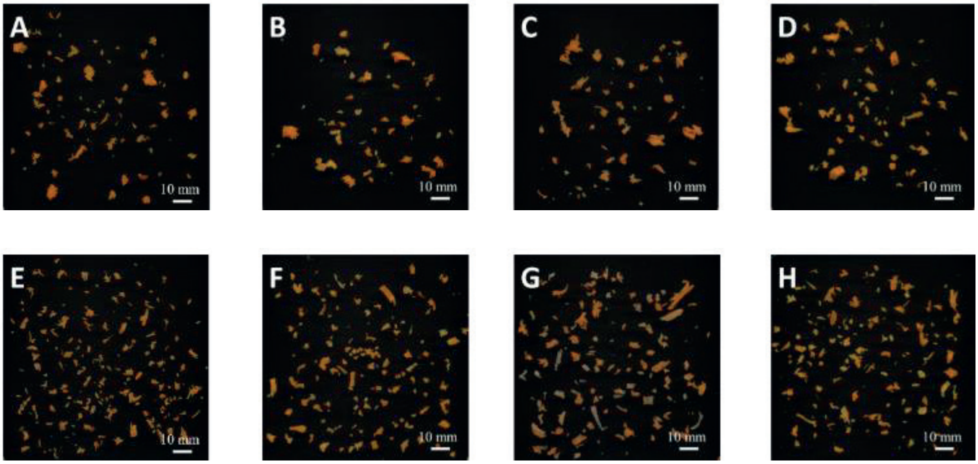

Figure 4.4: Pictures of expectorated carrot fragments (0.5 g) at the moment of swallowing of one subject (A-D: carrot cube C_1 without mayonnaise, and with **F**ull **F**at/**H**igh **V**iscosity, **L**ow **F**at/**H**igh **V**iscosity and **L**ow **F**at/**L**ow **V**iscosity; E-H: carrots julienne J_8 without mayonnaise, and with **F**ull **F**at/**H**igh **V**iscosity, **L**ow **F**at/**H**igh **V**iscosity and **L**ow **F**at/**L**ow **V**iscosity).

Table 4.3: Fixed effects and descriptives (mean±SE) of bolus properties at the moment of swallowing (100% mastication time) for carrot cubes and carrots julienne without and with different mayonnaises (**F**ull **F**at/**H**igh **V**iscosity, **L**ow **F**at/**H**igh **V**iscosity, **L**ow **F**at/**L**ow **V**iscosity), derived by linear mixed models.

	Particle count (-)		Particle size d_{50} (mm²)		Particle roundness (-)		Mass carrot in bolus (g)		Mass carrot in debris (g)		Mass carrot lost (g)	
(A) Statistics	***F***	***p***	***F***	***p***	***F***	***p***	***F***	***p***	***F***	***p***	***F***	***p***
Shape effect	497.8	***	0.9	0.335	121.3	***	18.7	***	14.1	**	16.5	***
Mayonnaise effect	39.7	***	25.7	***	0.5	0.707	11.9	***	12.4	**	3.6	*
Mayonnaise:Shape effect	14.2	***	7.0	***	2.1	0.098	3.3	*	1.9	0.127	2.6	0.052

(B) Descriptives	Particle count (-)		Particle size d_{50} (mm²)		Particle roundness (-)	Mass carrot in bolus (g)		Mass carrot in debris (g)		Mass carrot lost (g)	
Cube C_1	74 ± 4	*a*	4.8 ± 0.3	*b*	0.56 ± 0.004	2.6 ± 0.1	*b*	0.3 ± 0.04	*a*	1.3 ± 0.1	*a*
Cube C_1 with FF_HV	69 ± 4	*ab*	5.5 ± 0.2	*ab*	0.56 ± 0.004	2.4 ± 0.2	*b*	0.2 ± 0.05	*b*	1.5 ± 0.1	*a*
Cube C_1 with LF_HV	59 ± 4	*c*	5.9 ± 0.4	*a*	0.57 ± 0.005	2.9 ± 0.1	*a*	0.1 ± 0.03	*c*	1.2 ± 0.1	*b*
Cube C_1 with LF_LV	62 ± 3	*bc*	6.1 ± 0.4	*a*	0.57 ± 0.005	2.5 ± 0.1	*b*	0.2 ± 0.04	*b*	1.5 ± 0.1	*a*
Julienne J_8	155 ± 6	*A*	3.9 ± 0.2	*C*	0.54 ± 0.004	1.9 ± 0.2	*C*	0.5 ± 0.06	*A*	1.9 ± 0.1	*A*
Julienne J_8 with FF_HV	117 ± 6	*B*	5.9 ± 0.3	*B*	0.54 ± 0.005	2.2 ± 0.2	*B*	0.3 ± 0.04	*B*	1.7 ± 0.2	*A*
Julienne J_8 with LF_HV	97 ± 4	*C*	7.5 ± 0.4	*A*	0.52 ± 0.004	2.6 ± 0.2	*A*	0.2 ± 0.04	*C*	1.5 ± 0.2	*B*
Julienne J_8 with LF_LV	114 ± 4	*B*	5.8 ± 0.3	*B*	0.53 ± 0.004	2.4 ± 0.2	*AB*	0.3 ± 0.04	*B*	1.6 ± 0.1	*A*

F-values and p-values are derived from linear mixed models with shape, mayonnaise and the interaction as fixed effect, and subject as random effect.
Significance is presented as NS (non-significant); * (p<0.05), ** (p<0.01), and *** (p<0.001).
Different small letters indicate significant differences between cube samples without/with mayonnaise (p<0.05).
Different capital letters indicate significant differences between julienne samples without/with mayonnaise (p<0.05).

that condiments are shown to assist bolus formation of foods with a high moisture content without moisture absorbing capacity. Hence, the mechanism by which condiments facilitate bolus formation of carrier foods depends on the properties of the carrier foods. In the case of dry bread and crackers, the condiments provide moistening and softening. In the case of carrots, condiments are used to provide increased bolus cohesiveness. Providing lubrication seems to play a role in both dry and moist carrier foods and is therefore likely to be an important parameter in safe swallowing.

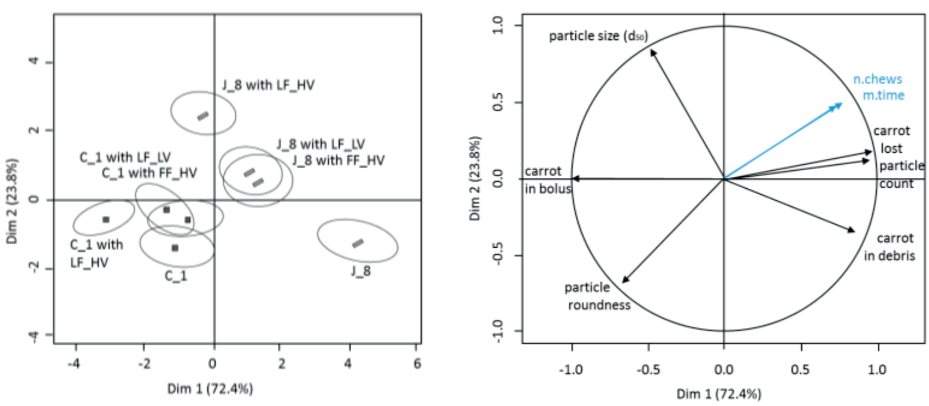

Figure 4.5: Principal Component Analysis (PCA) of the bolus parameters of carrot cubes (C_1, ■) and carrots julienne (J_8, ▬) without mayonnaise and with different mayonnaises (**F**ull **F**at/**H**igh **V**iscosity, **L**ow **F**at/**H**igh **V**iscosity, **L**ow **F**at/**L**ow **V**iscosity) at 100% of total mastication time. Mastication behavior parameters are plotted as supplementary variables (blue lines). The individuals map (samples) is shown on the left, and the variables map (attributes) is shown on the right. The ellipse level of confidence was 0.95.

Mayonnaises varying in fat content and viscosity impacted the mastication behavior of carrots differently. FF_HV and LF_LV mayonnaises significantly decreased the mastication effort (total mastication time, number of chews) required for the consumption of carrot cubes and carrots julienne (Figure 4.3), whereas little effects were observed for LF_HV mayonnaise. Although the LF_HV mayonnaise had the lowest impact on mastication behavior, it largely influenced carrot bolus properties including carrot particle size and carrot retention in the bolus (Table 4.3). Addition of the LF_HV mayonnaise decreased the number of particles, increased the size of the particles and increased carrot retention in the bolus to a larger extent than the FF_HV and LF_LV mayonnaises. This shows that when carrots are consumed in combination with mayonnaise, it is not the particle size that determines the swallowing threshold (Hutchings & Lillford 1988). Instead other bolus properties such as lubrication or cohesiveness seem to dominate the swallowing threshold and trigger the urge to swallow.
As summarized in the Principal Component Analysis (PCA) bi-plot, mastication behavior and bolus properties of carrots are mainly affected by carrot shape (PC1, 72.4%). Carrots julienne

were chewed for a longer time with more chews to decrease the degree of structure (*i.e.* higher number of particles). As a consequence of this longer chewing time, less carrot was retained in the bolus likely due to intermediate swallows. However, when mayonnaise was added, the amount of carrots in the bolus was increased more for the julienne than for the cubes. The type of mayonnaise induced some smaller changes to the mastication behavior and bolus properties of carrots (PC2, 23.8%). 95%-confidence ellipses indicate that julienne without mayonnaise (J_8) differed significantly from all julienne-mayonnaise combinations, whereas cube without mayonnaise (C_1) differed significantly from cube with LF_HV mayonnaise only, and not from the FF_HV and LF_LV mayonnaise. These results indicate that mayonnaise had a larger impact on bolus formation of julienne carrots than for carrot cubes.

4.3.3 Practical implications

Practical approaches to increase vegetable consumption are desired, as many people do not meet the recommended daily intake of vegetables. Results showed that changing the shape of carrots affected mastication time (Figure 4.1A) and eating rate (Figure 4.1C), which are known to influence food intake within a meal (Forde *et al.* 2013b; Robinson *et al.* 2014; Forde *et al.* 2017; van den Boer *et al.* 2017). We observed that carrot cubes were consumed with less mastication effort (shorter mastication time, fewer chews) and with higher eating rates than julienne carrots. Furthermore, carrots cut into one large cube were consumed with lower mastication effort and higher eating rate than for small carrots already cut into smaller pieces. Hence, cutting vegetables in particular into elongated, julienne pieces (high aspect ratio, high surface area) prolongs mastication time, and slows down eating rate. This would potentially reduce vegetable intake. On the other hand, consuming vegetables in larger pieces may increase vegetable intake. Thus, vegetable shape influences eating rate and potentially influences vegetable intake. It has to be noted that changing vegetable properties within one bite (standardized eating) can be different from changing vegetable properties in a snack or a complete meal consisting of multiple bites (free eating). Recently, Goh *et al.* (2017) and Liem *et al.* (2019) investigated *ad libitum* intake of carrots served as whole carrots (unit size: ~70 g) and carrot cubes (unit size: ~10 g). They found that children consumed more of the whole carrots than of the diced carrots, which is in agreement with our hypothesis (Goh *et al.* 2017; Liem & Russell 2019). However, the properties of one bite are expected to be relatively similar for whole carrots and carrot cubes (*i.e.* one large piece that is easy to place between the teeth or molars), and the difference in intake between whole carrots and carrot cubes is therefore not likely to be due to a difference in mastication effort as was seen for the carrot cubes and julienne carrots. Other mechanisms such as an increased consumption effort (a consumer has to collect smaller carrots more often than whole carrots) or an effect of unit size (the larger the meal, the more consumers tend to eat) are likely to be involved in the total intake between whole carrots and carrot cubes. Even though different mechanisms underlie consumption of carrots varying in shape and size, we recommend to consume carrots as a whole or pre-cut into large pieces to increase carrot intake. Although we investigated carrots only, we believe that this concept holds for other vegetables and fruits. Changing the shape of the vegetable can be easily implemented by vegetables producers and/or adopted by consumers, and could therefore be a promising strategy to increase vegetable intake among the population. In addition, many vegetables or fruits are naturally present at single bite sizes varying in size (*e.g.* cherry tomatoes, strawberries, Brussels sprouts, etc.) and we speculate that eating rate and potential intake is higher when such foods are served at a larger single bite size.

Another relatively simple approach to increase vegetable consumption could be to provide a condiment together with the vegetable, since we observed that addition of mayonnaises to raw carrots shortened mastication time (Figure 4.3A) and increased eating rate (Figure 4.3C). This approach is therefore also expected to increase vegetable intake. Addition of mayonnaises contributes to faster bolus formation by providing lubrication and adhering the carrot pieces together. Thus, to increase vegetable and energy intake, we recommend to consume vegetables accompanied by condiments. This strategy might unfortunately bring also an increase of the calorie intake, which is usually not desirable. However, such an increase in both vegetable and energy intake is desired among the elderly population since they are at risk of undernutrition (Hickson 2006; Pauly *et al.* 2007). Ageing induces changes in oral physiology and mastication efficiency, and elderly chew solid foods longer and with more chews than the younger population (Mioche *et al.* 2004; Ketel *et al.* 2019). When designing foods targeting the elderly population, it is important to be aware that pre-cutting foods does not necessarily decrease mastication effort; it can even increase mastication effort. As discussed above, adding condiments to solid foods, in particular those condiments with a low viscosity or a high fat content, aids chewing and is expected to facilitate increased food intake of older consumers with decreased eating capabilities.

Next to food intake, nutrient absorption from vegetables into the human body is important to assure the beneficial impact of nutrients on human health. As discussed before, initial carrot shape, and the presence and type of mayonnaise affect carrot particle sizes at the moment of swallowing (Figure 4.4, Table 4.3). We speculate that this might have important implications for the bioaccessibility of micronutrients present in carrots, such as α-carotene, β-carotene, vitamin K and vitamin B6. For raw carrots, it has been shown that carotene bioaccessibility increases when carrot particle sizes decrease at the moment of swallowing (Hedrén *et al.* 2002; Lemmens *et al.* 2010). We observed that carrot cubes (C_1) were swallowed with larger particle sizes than carrots cut into long julienne pieces (J_8). To facilitate micronutrient absorption, we would therefore recommend to cut vegetables before consumption, leading to longer mastication times and consequently further particle breakdown before swallowing. This recommendation is in contrast to our recommendation on food intake. From an increased bioaccesibility perspective we recommend to pre-cut vegetables, whereas from a decreased *ad libitum* intake perspective we recommend to eat whole/large vegetables. To develop a full picture of vegetable intake and subsequent nutrient absorption, additional multi-disciplinary studies are needed that combine eating behavior, bolus properties and digestion of vegetables. Further, the present study showed that addition of mayonnaises increased carrot particle sizes at the moment of swallowing, and thus a decrease in carotene bioaccessibility can be expected. On the other hand, the presence of lipids is known to facilitate carotenoid accessibility (Xavier & Mercadante 2019). Brown *et al.* (2004) found a greater absorption of carotenoids when raw carrots were consumed in combination with salad dressings containing fat (Brown *et al.* 2004). Thus, although larger carrot pieces were swallowed with the presence of mayonnaises, this lower accessibility might be compensated by a higher uptake due to the presence fat. This holds in particular for the full fat mayonnaise (FF_HV). The combination of these findings provides some support that the addition of condiments to carrots, regardless of the particle size, can increase carotenoid accessibility.

4.4 CONCLUSIONS

This study highlights that carrot shape has a large impact on mastication effort required to safely swallow carrots. Carrots cut in one large cube required the shortest chewing time and the least chews among all pre-cut carrots (cubes and julienne). Carrot cubes were chewed for a shorter time, with fewer chews and with higher eating rate than carrots julienne with similar number of pieces and an equal total weight. Aspect ratio and surface area increase with pre-cutting and are higher for julienne than for cube carrots. Consequently, more chewing and more saliva is needed to break down and pack the carrot particles together to form a lubricated and cohesive bolus that is safe to swallow. Furthermore, addition of mayonnaise reduced total mastication time until swallowing and increased eating rate of both carrot cubes and carrots julienne. Carrots were swallowed with less particles of larger sizes when mayonnaises were added. A specific particle size is therefore not a prerequisite to induce swallowing. This indicates that other bolus properties such as lubrication or cohesiveness rather than carrot particle size triggered the urge to swallow. In particular, full fat mayonnaise and low viscous mayonnaise lubricated and adhered carrot particles more than low fat high viscosity mayonnaise, leading to shorter mastication times and a lower number of chewing cycles to form boli that can be safely swallowed. We conclude that oral processing behavior of carrot-mayonnaise combinations is mainly affected by carrot shape and to a smaller extent by the presence and type of mayonnaise. These results suggest that relatively small changes in the vegetable or condiment properties can largely alter consumption time and eating rate, which could be an effective strategy to increase vegetable consumption or to decrease mastication effort to target the elderly population.

5

Shape matters: How cracker shape modifies *ad libitum* snack intake of crackers with cheese dip

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ABSTRACT

Background and objective: Food properties influence eating rate and food intake within a meal. Nowadays, snack consumption increases and snacks contribute substantially to our daily energy intake. Studying the extent to which food properties contribute to snack intake is of interest. This study investigated the influence of both cracker shape and cheese viscosity on *ad libitum* snack intake of cracker-cheese combinations.

Methods: Forty-four unrestrained participants (13 males, 23±3 years, BMI 21±2 kg/m2) participated in four late afternoon snack sessions [2x2 randomized crossover design]. Iso-caloric crackers were baked into flat squares and finger-shape cylindrical sticks (40x40x3 and 60x10x10 mm; 2.5 g per cracker) and combined with a cheese dip varying in viscosity (thick or thin). Crackers and cheese dip were served in separate large bowls containing approximately 80 crackers and 500 g cheese. Participants were instructed to dip the crackers into the cheese bowl. Participants consumed crackers with cheese *ad libitum* while watching a movie of 30 minutes. Dipping behavior and oral processing behavior were measured simultaneously by hidden balances under the cheese bowls and video recordings.

Results: Cracker intake (28±1 crackers per session) was not influenced by cracker shape nor cheese viscosity. Cheese intake was higher for flat squared than finger-shape crackers (13.2 g, 131 kJ, 15% difference, p=0.016), as a larger amount of cheese was scooped with flat squared crackers than with finger-shape crackers (2.9±0.2 vs. 2.3±0.1 g cheese per dip, p<0.001). The larger energy intake when using flat crackers seems to be unconscious and did not trigger earlier satiation. Cheese viscosity did not affect cheese intake. Eating rate over snacking time decreased by reducing bite frequency (p<0.001) while dip size remained fairly constant (p=0.12).

Conclusions: We conclude that changing food shape is an easy, subtle and yet underexplored approach that modifies eating behavior and consequently food intake.

Key words: shape, snacks, composite foods, oral processing behavior, bite size, eating rate, *ad libitum* food intake

5.1 INTRODUCTION

With the increase of overweight and obesity, developing approaches to lower food intake is an ongoing public health challenge. In 2016, more than 1.9 billion adults (~39% of the world's adult population) were overweight, of which 650 million were obese (World Health Organization 2018b). Nowadays, snack consumption increases (Bellisle *et al.* 2003; Piernas & Popkin 2010; Dunford & Popkin 2018) and contributes substantially to our daily energy intake by their high energy density (de Graaf 2006). For instance, snack consumption is responsible for 459 and 579 kcal per day for US children (2-18 years, 2011-2014) (Dunford & Popkin 2018) and US adults (≥19 years, 2003-2006) (Piernas & Popkin 2010). Therefore, gaining insights into factors contributing to snack intake is of interest.

Nowadays, more attention is given to eating behavior and how to change this to decrease food intake. There is much evidence that food and energy intake is effectively lowered by decreasing eating rate (amount of food consumed per unit of time, in g/min) (de Graaf & Kok 2010; de Graaf 2012; Robinson *et al.* 2014; McCrickerd & Forde 2017). In this context, oral exposure duration thus appears to be a main determinant of food intake (Zijlstra *et al.* 2009; Wijlens *et al.* 2012; Bolhuis *et al.* 2014b; Lasschuijt *et al.* 2017). Consequently, energy intake rate (energy consumed per unit of time, in kcal/min), *ad libitum* intake and thereby overconsumption can be reduced when foods are consumed with slower eating rates.

Eating rate depends on both consumer characteristics (Devezeaux de Lavergne *et al.* 2015a; McCrickerd & Forde 2017; Ketel *et al.* 2019) and food properties (Forde *et al.* 2013b; Forde *et al.* 2017; van den Boer *et al.* 2017). Individuals have consistent habitual eating strategies for any type of food: those who eat faster were observed to take larger bites from different types of foods and consume typically more energy (Robinson *et al.* 2014; McCrickerd & Forde 2017). Changing food properties, on the other hand, has been reported to change eating rate and thereby intake. Well-known examples are changing viscosity in liquids/semi-solids or changing hardness in solids (de Wijk *et al.* 2008; Zijlstra *et al.* 2008; Lasschuijt *et al.* 2017; McCrickerd *et al.* 2017). For example, *ad libitum* intake of chocolate flavored foods was 14 or 30% lower for a semi-liquid or semi-solid version than for a liquid version, respectively (Zijlstra *et al.* 2008). Similar trends were found in rice porridges, as thick porridge was consumed slower than the thinner version, by which intake was decreased with approximately 12% (McCrickerd *et al.* 2017). Lasschuijt *et al.* (2017) showed that increasing gel hardness increased oral exposure duration and subsequently decreased food intake by 21.5% (Lasschuijt *et al.* 2017). These food textural characteristics are well-known to influence food oral processing behavior including oral exposure times, chewing and eating rate and consequently regulate *ad libitum* food intake.

Variation in food shape can also influence oral processing behavior and food intake, but this aspect is less studied. Recently, we investigated the effect of carrot shape on oral processing behavior within one bite (van Eck *et al.* 2019c). Carrot cubes were chewed for a shorter time, with fewer chews and with higher eating rate than carrots julienne served at equal weight (van Eck *et al.* 2019c). Goh *et al.* (2017) and Liem & Russell (2019) investigated whether differences in shape and serving size of vegetables (whole vs. diced carrots) affected *ad libitum* intake among children. They found that intake of whole carrots was significantly higher than intake of diced carrots (Goh *et al.* 2017; Liem & Russell 2019). However, the mechanisms by which food shape influences eating behavior and/or intake are not well understood and require further investigation.

Although some strategies are known to change eating behavior of single foods, little is known about changes in eating behavior when foods are consumed in combination with other foods. For example, bread or crackers are often combined with cheese or spreads, salad with dressing or yogurt with cereals. Such foods that are composed of two or more single foods are termed composite foods. From the start of consumption, the combined foods are mixed in the mouth resulting in versatile textural properties and thereby less predictable oral processing behavior. Previously, we showed that different condiments affected the eating rate of bread and cracker to a different extent. Eating rate was especially influenced by the consistency of the condiment. Fastest eating rate of crackers and breads were obtained when combined with mayonnaise, followed by cheese spread and then firm cheese (van Eck *et al.* 2019b). The influence of condiment consistency was confirmed in another study, which demonstrated that low viscous mayonnaise accelerated eating rate of composite foods more than high viscous mayonnaise (manuscript in preparation). Bolhuis *et al.* (2014) assessed the effects of varying hardness of bread (soft vs. hard) and vegetables (raw vs. cooked) on total meal energy intake, and found that energy intake was 13% lower in the harder version of each food (Bolhuis *et al.* 2014a). Recently, Mosca *et al.* (2019) assessed eating rate and *ad libitum* intake of yogurts with added granola pieces varying in particle size (6 and 12mm). Modification of granola particle size changed eating rate by 7% and *ad libitum* intake by 5% (Mosca *et al.* 2019b). These studies showed that variations in single food properties can affect eating rate sufficiently to affect total energy intake of composite foods.

In this work we aim at investigating the influence of food properties on eating rate and *ad libitum* intake of snacks. We use crackers and processed cheese for this purpose, and evaluated the snacking behavior of these products during video watching, resembling a real life situation. In this study, the primary objective was to investigate the influence of cracker shape and cheese viscosity on *ad libitum* food intake of cracker-cheese combinations. Secondly, relationships between oral processing characteristics, dynamic dipping behavior and *ad libitum* intake were investigated. We hypothesize that both cracker shape and cheese viscosity change bite size (and the ratio between cracker and cheese), as cheese might be easier scooped when crackers have a larger surface area or when cheeses have a higher viscosity. We hypothesize that single products can be optimized for their shape and consistency, which influences bite size and/or eating rate of composite foods, and thereby regulates food intake. Such an approach offers the possibility to develop snacks that assist in energy intake regulation.

5.2 MATERIALS AND METHODS

5.2.1 Samples

A 2x2 factorial design was used with two cracker shapes (flat squares / finger-shape sticks) and two cheese viscosities (high / low) yielding four cracker with cheese dip combinations.

5

Crackers

Two cracker shapes were prepared: flat squares (40x40x3 mm; surface area per cracker 3680 mm^2) and finger-shape sticks (60x10x10 mm; surface area per cracker 2600 mm^2). Pictures of both crackers are shown in Figure 5.2. Crackers were prepared from the same dough, baked into the two shapes with an equal weight per piece to assure equal flavor and calorie content between the two shapes (~1620 kJ/100 g). The dough was based on a commercial cracker recipe, and was prepared from the following ingredients: wheat flour (Edelweiss), tap water, dried yeast, salt, barley syrup, icing sugar, margarine and bakery enzyme (Biobake BPN). Equal cracker weight before and after baking (weight after baking: 2.4±0.1 g for the squares, 2.5±0.1 g for the sticks, n=12) and similar moisture content (5±2 wt% for the squares, 7±0 wt% for the sticks, n=10) was assured by adjusting baking time and temperature. To characterize the hardness of the two shapes, three-point bend tests were performed with a Texture Analyzer (TA.XT Plus, Stable Micro Systems, United Kingdom) fitted with a 5 kg load cell and a constant test speed of 2 mm/s. The mean force needed to break the crackers was 5.2±1.5 and 7.6±3.0 N (mean±SD, n=26) for the flat squares and finger-shape crackers, respectively.

Cheese dip

Cheese dips were based on commercially available processed cheese dip (Cheez Dippers, Fromageries Bel Production, Lons le Saunier, France). The creaming (stirring at 80°C) and cooling process during production was slightly adapted to obtain cheese dips with different viscosities but with similar flavor and identical calorie content (~996 kJ/100 g). Flow curves of the two cheese dips were determined using a rheometer (MCR 301 Rheometer, Anton Paar Benelux BVBA, Belgium) equipped with an Inset I-PP50/SS plate and a CP50-1 cone. A resting period of 5 min was applied, after which the viscosity was measured between shear rates ranging from 1 to 1000 s-1 within a time period of 7.5 minutes at 20°C. The cheese dip with a viscosity of 124 Pa·s at 1 s-1, of 16 at 10s-1 and 2 at 100 s-1 is referred to as thick, whereas the cheese dip with viscosities of 55 Pa·s at 1 s-1, 9 at 10 s-1 and 1 at 100 s-1 as thin. At the end of the last snack session, participants (n=44) received both cheeses on a spoon in a randomized serving order, and were asked to choose the thickest cheese (2-AFC) out of a pair. Thirty-two participants (74%) chose the high viscous cheese as the thickest sample, indicating that the difference in viscosity between cheeses led to a difference in thickness perception. Cheeses were removed from the refrigerator the afternoon before evaluation and allowed to equilibrate to a temperature of 17 °C (17±3°C for the high viscosity cheese, 17±4°C for the low viscosity cheese).

5.2.2 Participants

Participants were recruited from Wageningen and surroundings using social media and printed posters. Participants were selected to be cracker and cheese consumers (at least once a month, self-reported) and they had to fulfil the following criteria: age between 18-35 years old, European nationality, Caucasian ethnicity, BMI between 18.5-30 kg/m2, good general and oral health (self-reported). Participants were excluded from the study if they smoked or had a food allergy or intolerance to any of the ingredients present in both foods assessed in the study. Participants were also excluded if they had difficulties with chewing, swallowing and/or eating in general, had dental braces (not including a dental wire), had smell or taste disorders, used medication that may affect the function of taste/smell/mastication/salivation, had a history of eating disorders, had followed an energy restricted diet during the last 2 months, gained or lost 5 kg of body weight over the last half year or if they were pregnant or lactating.

Participants were invited to a screening session to check whether they were eligible to participate in the study. Participants completed the screening questionnaire including general information, medical information and information about allergies, product use and eating patterns), the Dutch Eating Behavior questionnaire (DEBQ) (Van Strien *et al.* 1986), and the three-factor eating questionnaire (TFEQ) (Stunkard & Messick 1985).

Eighty-five participants participated in the screening session. After the screening, 51 participants were found eligible and available. These 51 participants (13 males, 23±3 years old, BMI of 21±2 kg/m2) completed the study. Participants were kept naïve to the primary outcome of the study. Participants were told that the study aimed to determine liking of crackers with cheese while watching different nature documentaries. Participants were debriefed regarding the actual study objective after the study.

Participants received a monetary incentive for their participation, and gave written informed consent before the start of the study. The study was registered at the Dutch Trial register (NL7741; http://www.trialregister.nl). The experimental protocol of the study was submitted to and exempted from ethical approval by the medical ethics committee of Wageningen University (NL70240.081.19, ABR70240). The medical ethical committee judged that the study does not fall within the remit of the 'Medical Research Involving Human Subjects Act'. The ethical committee evaluated the experimental protocol and decided that the study is lawfully not obliged to obtain ethical approval from a recognized medical research ethics committee.

5.2.3 Experimental design

The study had a 2x2 randomized crossover design [two crackers varying in shape and two added cheese dips varying in viscosity]. Participants attended four test days over a time period of four weeks, one session per week, so that they assessed each cracker-cheese combination once . The four cracker-cheese combinations were served in random order, following a Latins square Williams design, and labelled with three-digit codes.

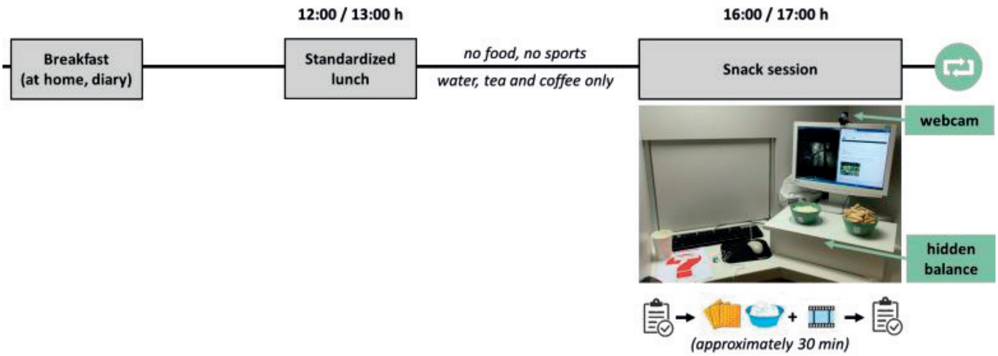

Figure 5.1: Experimental design displaying the different methodologies throughout the day. The snack session started with an appetite and liking evaluation, followed by *ad libitum* cracker with cheese dip consumption while watching a 30 minute movie (during which oral processing behavior and dipping behavior were monitored by a webcam and hidden balance under the cheese bowl), and concluded by another appetite and product liking evaluation.

Crackers with cheese dip were assessed during the late afternoon snack session (Figure 5.1), since late afternoons are a common consumption moment for crackers with cheese according to cheese producers. Participants received a large bowl of pre-weighed crackers (~200 g, approximately 80 pieces) and a large bowl of pre-weighed cheese dip (~500 g), which is about twenty times more than the commercially available snack product Cheez Dippers (Fromagerie Bel, Lons le Saunier, France). They also received a pre-weighed glass of tap water (~300 g) that they could consume freely during the session. Then, they were asked to watch a nature documentary of about 30 min, and to consume crackers with cheese dip until they were comfortably full. While consuming the cracker-cheese samples, cheese dipping behavior was recorded by hidden balances under the cheese bowls. In addition, participants were video recorded to extract oral processing behavior parameters.

Standardization of hunger

Firstly, participants were instructed to eat the same breakfast around the same time for all test session days, and to record this in an online diary. Prospective morning snacks were also recorded in this diary. Secondly, participants were provided with a standardized lunch at either 12:00 hours or 13:00 hours, depending on the time of the test session (16:00 or 17:00 hours, respectively). The lunch consisted of tomato soup, whole-grain bread slices, hummus, Nutella and a fruit yoghurt. For each participant, the amount of food was calculated based on 22% of their daily energy needs (Schofield, WHO, 1985). Participants self-filled the sandwiches and they were instructed to finish the lunch plate within 30 minutes. Thirdly, participants were requested not to consume anything except water, tea or coffee without milk and sugar nor to perform sports in between lunch and the afternoon test session.

Nature documentaries

Four different episodes (Galapagos Islands, Scottish Highlands, Great Barrier Reef and Namibia Desert) of a nature documentary (Nature's Microworlds, BBC Earth) were shown during the test sessions. The presentation order of the documentaries was randomized among participants following a Latins square Williams design. The four episodes were equally liked by consumers ($p=0.20$), and the type of episode did not affect eating behavior nor intake ($p>0.05$).

Nature documentaries were added to the study design for three reasons. Firstly, crackers with cheeses are generally consumed in a distractive state, *e.g.* during a social event or when watching television or a movie. Using a documentary as a distractive state is easily to control, and was therefore chosen for this study. Secondly, participants automatically turned their head straight to the screen when watching a documentary, which allowed to determine oral processing behavior easily without participants being too much aware of the camera recording it. Thirdly, in order to avoid participants from gobbling the samples to finish the session earlier, participants were told that they had to watch the entire documentary before evaluating final appetite and liking.

Appetite ratings, hedonic ratings and meal termination

At the beginning of the test session, participants rated appetite including hunger, fullness and thirst. After the first bite, they rated how much they liked the crackers, cheese dip and the combination. Immediately after the documentary ended, participants were asked to indicate how much they liked the documentary. Then, they rated their appetite and product liking again. Finally, participants indicated why they terminated consumption: I was full, the flavor of crackers with cheese was not pleasant anymore, I was bored, on a five-point scale anchored from totally disagree to completely agree. Ratings of appetite, product liking and documentary liking were assessed using a 100 pt VAS anchored with not at all and very much. Data were acquired by Qualtrics survey software (version October 2019, Qualtrics, USA).

Ad libitum food intake

Table 5.1 provides an overview of the *ad libitum* intake, bite size and oral processing behavior parameters extracted throughout the experiment. Participants were instructed to consume crackers with cheese dip until they were comfortably full. About 200 g of crackers and 500 g of cheese dip were served in separate bowlsParticipants could ask for more crackers and cheese if required. In total, an additional portion of crackers was requested three times (two different participants). No additional portion of cheese was requested.

Ad libitum cracker and cheese intake was calculated by subtracting the weight of the bowls before and after consumption. Parameters extracted were total cracker intake (in grams, in kJ), total cheese intake (in grams, in kJ) and total food intake (in grams, in kJ). In addition, averaged cheese:cracker weight ratio was calculated by dividing the cheese intake in grams by the cracker intake in grams.

Cheese dipping behavior

Cheese dipping behavior throughout snacking was monitored by precision balances (Kern, type PCD 10K0.1, KERN & Sohn GmbH, Germany). Cheese bowls were placed on the balances hidden in a table setup, so that participants did not see the balance. The balances were connected to a computer by an USB cable, which allowed to continuously record the weight of the cheese bowls (Software BalanceConnection, Kern, KERN & Sohn GmbH, Germany). The weight (g) of the cheese bowl was recorded by the hidden balance for every second of the complete snacking event of 30 minutes.

The weight of the cheese bowl reduced with each bite, which allowed to determine the amount of cheese consumed per dip and dynamic evolutions in dip size during consumption. From these data, the number of dips and the averaged dip size (g) were calculated.

Oral processing behavior

Oral processing behavior throughout snacking was determined using video recordings. A webcam (Microsoft LifeCam studio) was placed on top of the computer screen, just above the nature documentary that was presented on the screen to ensure that participants looked into the camera (face-on).

Videos were decoded using Simple Video Coder Software (Barto *et al.* 2017). A coding scheme was developed to record the frequency and the duration of three key events (*i.e.* new cracker with cheese dip, re-dipped cracker, sip of water) and two additional behavioral events (*i.e.* cracker without cheese, two crackers consumed at the same time) during a complete snacking event of about 30 minutes. Coding of all video recordings was performed by two trained video coders, who watched several videos together until they agreed on the coding scheme. Approximately 10% of the videos were randomly selected and codification was validated by both.

Parameters directly extracted from the video recordings include the number of crackers eaten, number of bites taken, number of dips taken, number of sips of water, total snacking time (min), total oral exposure time (s) and averaged oral exposure time per bite (s/bite). Total snacking time was defined as the time between the first bite and the last swallow. Total oral exposure time was defined as the cumulated period that food remains in the mouth during the snacking event. Averaged oral exposure time was defined as the averaged period that food remains in the mouth per bite. Subjects can take multiple swallows, but only the moment of the main swallow was used for analysis.

Additional oral processing behavior parameters including averaged cracker bite size (g/bite), eating rate (g/min) and energy intake rate (kJ/min) were calculated by combining the video and intake data. Averaged cracker bite size was calculated by dividing the amount of crackers consumed (g) by the total number of bites. Eating rate (g/min) and energy intake rate were calculated by dividing the amount of crackers and cheese dip eaten (g and kJ) by the total snacking time (min).

5.2.4 Statistical data analysis

Results were reported as mean values with standard error. During the snack sessions, participants were instructed to eat the crackers with cheese dip *ad libitum* until they felt comfortably full. When less than 10 g of crackers (n=6) or less than 10 g of cheese dip (n=1) were consumed, data were excluded from the statistical analyses. In total, statistical data analyses were performed on results of 44 participants (13 male, 23±3 years, BMI 21±2 kg/m2). One-way analyses of variance (ANOVA) on appetite and liking data were conducted to assure equal appetite at the start of the snack sessions and to compare products effects on appetite and liking. The effect of cracker shape and cheese viscosity on intake, bite size and oral processing behavior parameters (Table 5.1) were analyzed with linear mixed models using Lmer package (Kuznetsova *et al.* 2016). Shape, viscosity and shape:viscosity interaction were set as fixed effects and participant and session were set as random effects. The effect of consumption time on dynamic dipping behavior was analyzed with linear mixed models with time, sample and time:sample as fixed effects and participant and session as random effects. Multiple factor analysis (MFA) was performed on different data sets (intake, oral processing behavior, dipping behavior) to compare consumers differing in dipping behavior using FactoMineR package (Lê *et al.* 2008). R language (RStudio, version 1.0.143) was used to perform all statistical tests. Significance level of $\alpha<0.05$ was chosen.

Table 5.1: Overview of the *ad libitum* intake, bite size and oral processing behavior parameters extracted throughout the experiment.

Parameter	Definition	Obtained from
(A) Ad libitum food intake		
Cracker intake (g, kJ)	Total amount of crackers (intake in grams or energy intake in kJ) consumed during the snacking event	Intake
Cheese intake (g, kJ)	Total amount of cheese dip (intake in grams or energy intake in kJ) consumed during the snacking event	Intake
Total food intake (g, kJ)	Total amount of the crackers and cheese dip (intake in grams or energy intake in kJ) consumed during the snacking event	Intake
Water intake (g)	Total amount of water (in grams) consumed during the snacking event	Intake
(B) Oral processing behavior		
Number of crackers	Total number of crackers consumed during the snacking event	Videos
Number of bites	Total number of bites during the snacking event; this also includes bites of crackers without cheese and re-bites	Videos
Number of sips of water	Total number of sips of water consumed during the snacking event	Videos
Total snacking time (min)	Time between the first bite and the last swallow	Videos
Total oral exposure time (min)	Cumulated period that food remains in the mouth during the snacking event	Videos
Oral exposure time per bite (s/bite)	Averaged period that food remains in the mouth per bite	Videos
Eating rate (g/min)	The amount of food consumed per unit of time (i.e. cracker intake in grams divided by total oral exposure time)	Intake + videos
Energy intake rate (kJ/min)	The amount of energy consumed per unit of time (i.e. cracker intake in kJ divided by total oral exposure time)	Intake + videos
(C) Dipping behavior		
Number of dips	Total number of dips during the snacking event; this also includes crackers that are dipped multiple times.	Balances
Cheese dip size (g) [a]	Averaged amount of cheese dip consumed per bite	Balances
Cheese:cracker weight ratio	Averaged amount of cheese in grams relative to the averaged amount of crackers in grams	Intake

5.3 RESULTS

5.3.1 Ad libitum food intake

Figure 5.2 shows food intake of cheese and crackers (Figure 5.2A) and cheese energy intake (Figure 5.2B) for the four cracker/cheese combinations. *Ad libitum* cracker intake was not influenced by cracker shape (p=0.94) nor cheese viscosity (p=0.77). *Ad libitum* cheese intake was significantly influenced by cracker shape (p=0.016); finger-shape crackers led to a 15% (13.2 g, 131 kJ) lower intake of cheese dips compared to that of flat squares (Figure 5.2). Consequently, averaged total energy intake was 1971±138 kJ when cheese dips were served with finger-shape crackers and 2106±145 kJ when served with flat squares (total energy intake reduction of 7%). Cheese viscosity did not affect cheese intake (p=0.83) nor total energy intake (p=0.97). Total water consumption throughout the snacking session (239±115 g, mean±SD) was not influenced by cracker shape (p=0.96) nor cheese viscosity (p=0.62).

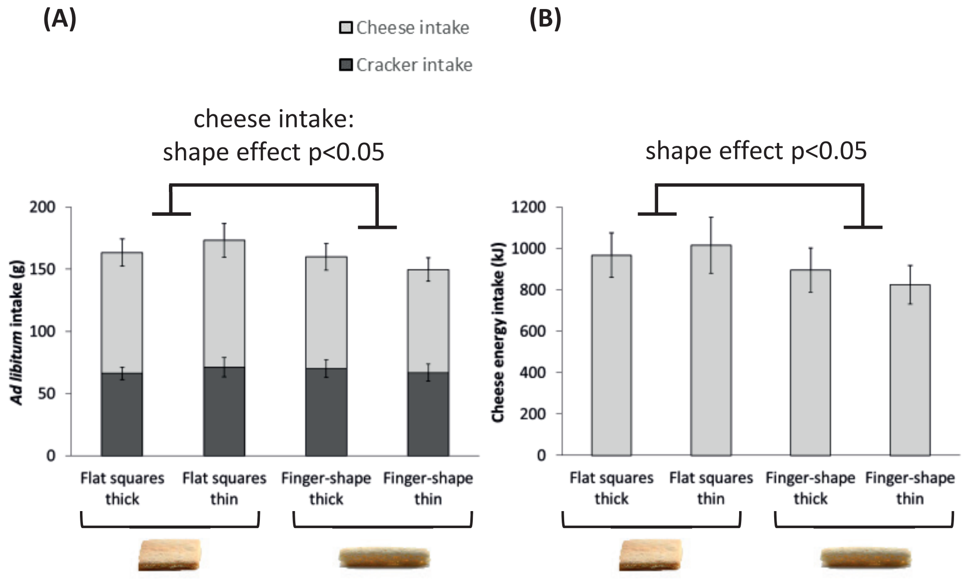

Figure 5.2: Intake of crackers varying in shape (dark gray bars) consumed together with cheese dips varying in viscosity (light gray bars), expressed in grams (A). Energy intake by cheese dip consumption, expressed in kilojoules (B). Error bars represent standard error of the mean (n=44). Pictures of crackers varying in shape are presented for illustration purposes.

5.3.2 Oral processing behavior

Number of crackers, number of bites, number of sips of water, total snacking time and oral exposure time per bite did not differ significantly between samples (Table 5.2B). On average, participants consumed 28±1 crackers with 46±2 bites, regardless of cracker shape and cheese viscosity. Although oral exposure time per bite did not differ significantly between samples, flat squares tended to be chewed slightly shorter (14.7±0.5 s) than finger-shape crackers (15.2±0.5 s). Consequently, over the total snacking event, total oral exposure time was found to be significantly shorter for flat squares (10±1 min) than for finger-shape crackers (11±1 min) (p=0.025).

Eating rate and energy intake rate of crackers with cheese dip were significantly influenced by cracker shape (p<0.001; p<0.001), but not by cheese viscosity nor by a shape:viscosity interaction (Figure 5.3). Flat squares resulted in a higher eating rate and higher energy intake rate than finger-shape crackers.

Table 5.2: Mean intensity scores (± standard error of the mean, n=44) and significance values (cracker shape, cheese viscosity, shape:viscosity interaction) describing *ad libitum* intake (A), oral processing behavior (B) and dipping behavior (C) of crackers with cheese dip. Significant p values (<0.05) are highlighted in bold.

	flat squares thick			flat squares thin			finger-shape thick			finger-shape thin			Cracker shape	Cheese viscosity	Shape: viscosity
	Mean	±	*SE*	*Mean*	±	*SE*	*Mean*	±	*SE*	*Mean*	±	*SE*	p	p	p
(A) Ad libitum intake															
Crackers															
Intake (g)	66	±	5	71	±	8	70	±	7	67	±	7	0.94	0.77	0.20
Energy intake (kJ)	1073	±	83	1155	±	124	1136	±	115	1084	±	109	0.94	0.77	0.20
Cheese dip															
Intake (g)	97	±	11	102	±	14	90	±	11	83	±	9	**0.016**	0.83	0.27
Energy intake (kJ)	968	±	108	1016	±	136	896	±	106	825	±	94	**0.016**	0.83	0.27
Crackers with cheese dip															
Intake (g)	163	±	15	173	±	19	160	±	17	150	±	15	0.09	0.98	0.20
Energy intake (kJ)	2041	±	173	2170	±	234	2032	±	205	1909	±	187	0.16	0.97	0.19
Water															
Intake (g)	242	±	19	236	±	18	241	±	18	236	±	14	0.96	0.61	0.94
(B) Oral processing behavior															
Number of crackers	29	±	2	28	±	3	29	±	3	28	±	3	0.98	0.17	0.90
Number of bites	46	±	4	44	±	3	49	±	4	46	±	3	0.14	0.18	0.77
Number of sips of water	6	±	1	6	±	0	6	±	1	6	±	1	0.60	0.88	0.96
Total snacking time (min)	24	±	1	25	±	1	23	±	1	25	±	1	0.57	0.17	0.31
Total oral exposure time (min)	10	±	1	10	±	1	12	±	1	11	±	1	**0.025**	0.14	0.38
Oral exposure time per bite (s)	15	±	1	15	±	1	15	±	1	15	±	1	0.39	0.73	0.54
Eating rate (g/min)	17	±	1	17	±	1	14	±	1	15	±	1	**<0.001**	0.50	0.99
Energy intake rate (kJ/min)	211	±	14	217	±	17	183	±	12	189	±	12	**<0.001**	0.37	0.98
(C) Dipping behavior															
Number of dips	31	±	2	34	±	3	38	±	3	36	±	2	**0.020**	0.80	0.24
Cheese dip size (g)	2.9	±	0.2	2.8	±	0.2	2.3	±	0.2	2.4	±	0.2	**<0.001**	0.92	0.23
Cheese:cracker weight ratio	1.6	±	0.1	1.5	±	0.1	1.3	±	0.1	1.3	±	0.1	**<0.001**	0.53	0.69

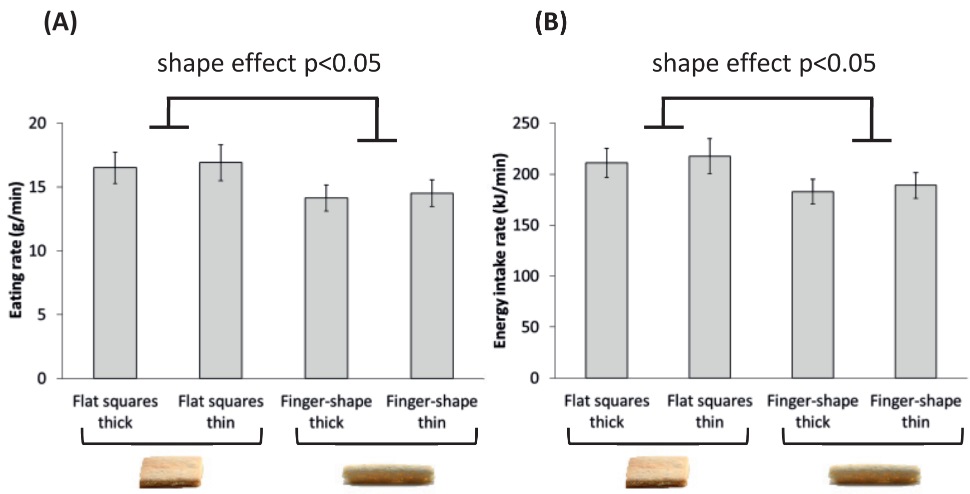

Figure 5.3: Eating rate (A) and energy intake rate (B) of crackers varying in shape consumed together with cheese dips varying in viscosity. Error bars represent standard error of the mean (n=44). Pictures of crackers varying in shape are presented for illustration purposes.

5.3.3 Cheese dipping behavior

Participants dipped crackers in a large bowl containing cheese dip, and the shape of the cracker had a large impact on cheese dipping behavior (Table 5.2C). The number of dips was lower for flat squares (33±2) than finger-shape crackers (37±2) (p=0.020). Cheese dip size was larger for flat squares (2.9±0.2 g) than finger-shape crackers (2.3±0.1 g) (p<0.001). Consequently, cheese:cracker weight ratio was larger for flat squares (1.5±0.1) than finger-shape crackers (1.3±0.1) (p<0.001). Cheese viscosity and shape:viscosity interactions did not significantly influence cheese dipping behavior (Table 5.2C).

Dynamic dipping behavior shows that number of dips (and thereby also other factors such as the eating rate, energy intake rate) decreased with increasing snacking time (p<0.001), regardless of cracker shape and cheese viscosity (Figure 5.4B). On average, participants dipped 9 – 11 times in the first five minutes, whereas this was only 2 – 3 times in the last five minutes of the snacking session. Dip size remained relatively constant throughout the snack session of 30 minutes (p=0.12) (Figure 5.4C), resulting in a steadily higher consumption of cheese with the presence of flat squares compared to that with finger-shape crackers. No time:sample interaction was found for the number of bites (p=0.79) and dip size (p=0.96), which indicates that the effect of time was comparable for all cracker-cheese combinations.

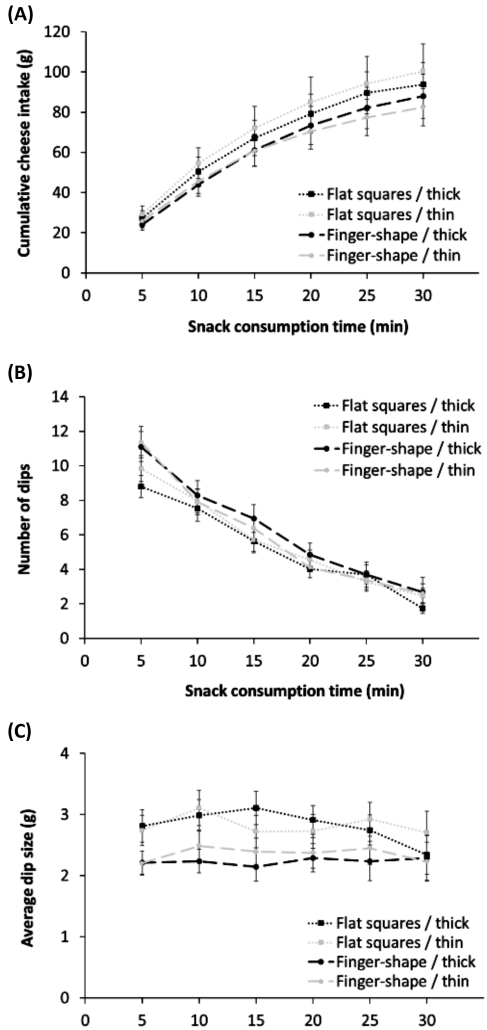

Figure 5.4: Cumulative cheese intake (A), number of dips (B) and averaged cheese dip sizes (C) during the consumption of crackers with cheese dip within a 30-minute afternoon snack session. Time points are averaged for every 5 minutes. Error bars represent standard error of the mean.

Individual differences in cheese dipping behavior

Participants differed in their dipping behavior. Most participants (n=23) dipped crackers more than once per cracker (*i.e.* they dipped the cracker into the cheese, bit part of the cracker, and dipped the remaining part of cracker into the cheese once more). Some participants (n=9) dipped crackers only once per cracker. In addition, some participants adapted their behavior according to the shape of the cracker, as some participants (n=8) dipped the finger-shape crackers twice or multiple times and the flat squares once per cracker, whereas others (n=3) dipped the flat squares twice or multiple times and the finger-shape crackers once.

Multiple Factor Analysis (MFA) was performed to represent the different dipping behavior groups in relation to the different data matrices (intake, food oral processing behavior and dipping behavior). Visual inspection of Figure 5.5 shows that the first dimension mostly describes the difference in dipping behavior (Dim 1: 58.72%). When comparing only-once-dippers and more-than-once-dippers, they applied a similar number of dips (34±3 vs. 37±2) but the only-once-dippers consumed far more cheese per bite (3.3±0.2 g) than the more-than-once dippers (2.3±0.1 g). Consequently, the only-once-dippers snacked faster (20±1 vs. 13±1 g/min) leading to a higher amount of cracker (87±9 vs. 55±3 g), cheese (111±11 vs. 86±7 g) and total energy (2510±225 vs. 1747±105 kJ) than more-than-once-dippers.

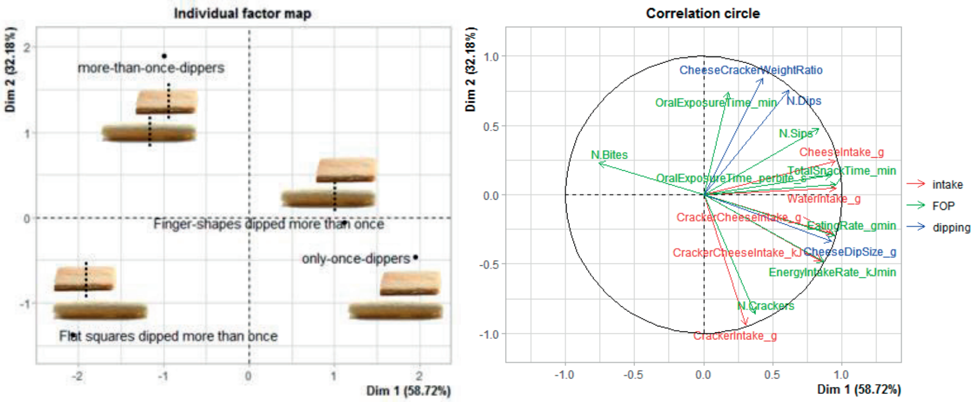

Figure 5.5: Comparison of four groups of participants varying in dipping behavior (crackers were dipped more than once per cracker, n=23; crackers were dipped once per cracker, n=9; only finger-shape crackers were dipped more than once per cracker, n=8; and only flat squares were dipped more than once per cracker, n=3) that consumed crackers with cheese dip *ad libitum*. Multiple Factor Analysis (MFA) was performed on four different datasets (intake, oral processing behavior, dipping behavior) that are presented in different colors. The individuals map (groups differing in dipping behavior) is shown on the left, and the variables map (parameters) is shown on the right. The different parameters are explained in Table 5.1. Pictures of crackers varying in shape are presented for illustration purposes, and the dotted lines represent two or multiple dips per cracker.

5.3.4 Appetite and hedonic ratings

Hunger ratings decreased from 60±1 to 24±1 pt ($p<0.001$), fullness ratings increased from 36±2 to 70±2 pt ($p<0.001$) and thirst ratings decreased from 50±2 to 35±2 pt ($p<0.001$) during the snacking session. Ratings of hunger, fullness and thirst both before and after the snack session did not differ between the different cracker-cheese combinations (Table 5.3).

Overall, the crackers with cheese dip combinations were rated as positively pleasant (*i.e.* 64±3 to 75±2 pt on a 100 pt VAS), and the hedonic ratings after the snack session did not differ among the four combinations ($p=0.38$) (Table 5.3).

Table 5.3: Appetite (A) and hedonic (B) ratings before and after the snack session (mean±SE, n=44). Significant p values (<0.05) are highlighted in bold.

	flat squares thick				**flat squares thin**				**finger-shaped thick**				**finger-shaped thin**				**p-value**
Parameters	*Mean*	*±*	*SE*		*Mean*	*±*	*SE*		*Mean*	*±*	*SE*		*Mean*	*±*	*SE*		
(A) Appetite ratings																	
Hunger																	
before	60	±	3		61	±	3		62	±	3		57	±	3		0.618
after	23	±	3		24	±	3		26	±	3		24	±	3		0.856
Fullness																	
before	34	±	3		36	±	3		36	±	3		37	±	3		0.937
after	71	±	3		74	±	3		69	±	3		67	±	3		0.471
Thirst																	
before	49	±	3		52	±	3		49	±	3		51	±	3		0.878
after	35	±	4		37	±	3		37	±	3		32	±	3		0.694
(B) Hedonic ratings																	
Cracker liking																	
before	64	±	3	*a*	65	±	3	*a*	56	±	3	*b*	55	±	3	*b*	**0.027**
after	64	±	4	*a*	63	±	3	*a*	51	±	3	*a*	53	±	3	*b*	**0.004**
Cheese liking																	
before	68	±	3		70	±	3		68	±	3		69	±	3		0.967
after	60	±	3		66	±	3		61	±	3		65	±	3		0.490
Cracker with cheese liking																	
before	74	±	2	*ab*	75	±	2	*a*	68	±	3	*bc*	67	±	2	*c*	**0.033**
after	68	±	3		70	±	3		64	±	3		66	±	3		0.381

Different letters indicate significant differences between cracker-cheese combinations (LSD posthoc, p<0.05).

5.4 DISCUSSION

Cheese dip intake was substantially influenced by the shape of cracker it was combined with. Flat squared crackers led to a 15% higher cheese intake (13.2 g, 131 kJ) than finger-shape crackers. This effect of cracker shape on *ad libitum* cheese intake can be explained by a difference in dip size, as the flat squares were consumed with 26% more cheese dip. The flat squares have a 41% larger surface area than the finger-shape crackers, which makes it easier to scoop more cheese on the cracker leading to a larger bite size. Participants unconsciously consumed more cheese with flat square crackers than with finger-shape crackers and, most importantly, they do this without differences in self-reported fullness. This is in line with another study, which compared spoon and fork users, showing that spoon users ate faster leading to higher food intake than fork users, probably because more food fitted on a spoon than on a fork (Bolhuis & Keast 2016). In general, larger bite sizes are associated with an underestimated, increased food intake by consumers (Weijzen *et al.* 2009; Zijlstra *et al.* 2009; Bolhuis *et al.* 2013). The present results show that by solely changing the shape of a single food, one can regulate food intake of food composites without changing liking of the food. This has important implications for future snack design since modifications of food shape are easy to implement by the food industry, and foods do not require ingredient reformulation while product liking is maintained.

Cracker intake was not influenced by cracker shape nor cheese viscosity. Participants steadily consumed 28±1 crackers throughout a snacking event of 30 minutes. Also the number of bites and thereby cracker bite size remained constant (on average: 46±2 bites). It may be that consumers apply a certain habitual consumption effort (*i.e.* reaching for a cracker, taking a bite) that results in certain familiar intake. Likewise, others found that consumers applied an almost equal number of bites in different yogurt-granola conditions (Mosca *et al.* 2019b). Several studies suggest that intake can be influenced by a small difference in consumption effort. For example, intake was lower when snack foods were served as smaller nibbles compared to that of large bars (Weijzen *et al.* 2008). Similarly, children consumed a lower amount of diced carrots than whole carrots (Goh *et al.* 2017; Liem & Russell 2019). In both cases, intake was lower when consumers had to consume the foods into more smaller bites (increased effort). Apparently, consumers use a constant, habitual consumption effort before they terminate snacking (see also Figure 5.4). In case of the present study, cheese intake was influenced by modification of cracker shape. In contrast to previous studies (Weijzen *et al.* 2008; Goh *et al.* 2017; Liem & Russell 2019), the crackers differed in shape but had the same weight per piece. Therefore, our study demonstrates that snack intake can be modified without changing convenience or consumption effort of the consumer. Such knowledge can be applied in future snack development.

Cheese viscosity was also expected to influence bite size, as viscosity can change the ease at which it can be scooped onto the crackers and/or the ease of safe-to-swallow bolus formation. However, cheese viscosity did not affect intake, food oral processing behavior nor dipping behavior of crackers with cheese dip. To be able to dip crackers into the cheese, the cheese dips could not be too solid nor too liquid. Consequently, the viscosity range was relatively small. We suggest that the difference in viscosity might not have been large enough to affect eating behavior and intake (viscosity differed with a factor 2). Even though differences in viscosity were instrumentally present and perceived by 74% of the participants, the difference in viscosity was apparently too small to affect oral exposure time, eating rate and subsequent intake of the cracker-cheese combinations. In previous studies where a viscosity effect on oral exposure time, eating rate and/or intake was observed, products were used with much larger viscosity differences (*e.g.* liquid, semi-liquid vs. semi-solid milk-based products; carrier foods combined with cheese spread vs. mayonnaise) (de Wijk *et al.* 2008; Zijlstra *et al.* 2008; van Eck *et al.* 2019b). Apparently, the difference in viscosity should be large enough to be able to affect oral processing behavior, eating rate and intake.

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In our experimental settings, participants were free to choose their own way of consumption (Figure 5.5), which resulted in large differences in eating/dipping behavior and intake between participants. Most participants (52%) dipped the cracker into the cheese, bit part of the cracker, and dipped the remaining part of cracker into the cheese once more (more-than-once-biters), whereas others (20%) dipped each cracker once and consumed it at once (only-once-biters). Interestingly, such differences in biting and dipping behavior impacted total food intake considerably. Multiple dipping led to less cheese per cracker compared to one dip per cracker (3.3±0.2 vs. 2.3±0.1 g). Those who consumed one dip per cracker showed higher intakes for crackers (87±9 vs. 55±3 g) and cheese (111±11 vs. 86±7 g). This implies that taking smaller bite sizes slows down eating rate and reduces energy intake, in agreement with other studies (Weijzen *et al.* 2009; Zijlstra *et al.* 2009; Mishra *et al.* 2012; Bolhuis *et al.* 2013; Bolhuis & Keast 2016; James *et al.* 2018).

Although large differences were observed between different participants, dipping behavior per participant remained relatively constant throughout a snacking session of 30 minutes. The amount of cheese dipped on a cracker did not differ between the beginning and end of consumption. Although one could expect that consumers add less cheese to the cracker with increasing snacking time as a result of satiation or sensory specific satiety, this was not the case. One can also argue the other way around, that consumers add more cheese with increasing snacking as a result of product liking. However, this was also not the case. Bite sizes remain constant throughout the snacking episode and consumers use their own standardized dip size and dipping strategy until they terminate snacking.

Although crackers are frequently consumed with cheese, they are also consumed with other spreads or toppings (*e.g.* butter, cream, hummus, jam, Nutella). We speculate that the effect of cracker shape on food intake can be generalized towards other spreads and toppings. This is of particular relevance, as dips, spreads and toppings are generally high in energy, fat and/or sugar (de Graaf 2006). Thus, changing the shape of crackers and likely also other carrier foods (*e.g.* raw vegetables, potato chips, pretzels) seems to be a promising approach to reduce intake of such less desirable nutrients. The advantage of simply changing the shape of a carrier food is that producers do not have to change the ingredients nor the recipe so that sensory perception and liking is probably maintained. Therefore, modifying food shapes seems to be a promising strategy to unconsciously affect food and energy intake. This can be a particularly effective strategy to target unrestrained consumers which are not actively looking at moderating their calorie intake.

The results of this study shows that food intake of snacks can easily be reduced by 15%. Snack consumption is responsible for 579 kcal per day for US adults (Piernas & Popkin 2010). Although snacks are not the major part of the total energy intake of consumers throughout the day, such a reduction can still contribute to a healthier diet. The contributions of snack foods to dietary intake is increasing (Bellisle *et al.* 2003; Piernas & Popkin 2010; Dunford & Popkin 2018), and therefore a reduction of 15% becomes of relevance. For an even larger impact on total food intake, such strategy of changing food shape could also be transferred to main meals such as French fries with sauce.

6

Adding condiments to foods: How does static and dynamic sensory perception change when bread and carrots are consumed with mayonnaise?

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Food Quality and Preference, 2019, 73: 154-170

ABSTRACT

Background and objective: Foods with condiments such as bread with spreads or vegetables with dips are frequently consumed. The aim of this study was to understand how dynamic and static sensory perception changes when foods are consumed together with condiments.

Methods: Two carriers (bread, carrot) varying in hardness were combined with condiments (mayonnaises) varying in fat content and viscosity to obtain model composite foods. Dynamic sensory perception was assessed using Temporal Dominance of Sensations (TDS) with attribute lists describing both carrier- and condiment-related attributes. Static sensory perception was evaluated using Rate-All-That-Apply (RATA) with attribute lists descriptive for either bread, carrot or mayonnaise.

Results: Carrier foods (bread, carrot) had a larger influence on dynamic and static sensory perception of carrier-condiment combinations than condiments (mayonnaises). Sensations related to mayonnaises (sour, creamy) were dominant at later stages of consumption when these were combined with harder bread or carrots. Hard bread or carrots reduced intensities of several mayonnaise-related attributes (sour, dairy when combined with bread; creamy, after taste when combined with carrots) to a larger extent than soft bread or carrots. Consumer sensitivity to discriminate between foods was not affected by the presence of other food items when differences in bread, carrots or mayonnaise properties were large. In case of smaller differences between food properties, consumer sensitivity to discriminate between foods declined and depended on the food type it was combined with.

Conclusions: We conclude that the product properties of both solid carrier foods and condiments and their interaction during consumption impact dynamic and static sensory perception of carrier-condiment combinations.

Key words: bread, carrot, mayonnaise, condiments, Temporal Dominance of Sensations (TDS), Rate-All-That-Apply (RATA)

6.1 INTRODUCTION

Many foods are combined with condiments and are eaten within one bite, such as bread with spreads or vegetables with dips. Properties of such carrier foods can differ considerably from the properties of the condiment in composition, mechanical properties and sensory characteristics. During consumption, foods are mixed in the mouth inducing continuous changes in sensory characteristics depending on the properties of the carriers and condiments.

Few studies have investigated the sensory perception of foods to which condiments or sauces have been added. It has been shown that flavor complexity increased when sauces were added to salmon (Paulsen *et al.* 2012), and texture complexity increased when inclusions were embedded in gel matrices (Tang *et al.* 2017). Furthermore, addition of gravy to vegetables (broccoli, cauliflower and potato) reduced the flavor intensity of the vegetables and increased gravy-related attributes (Meinert *et al.* 2011). Similar results were found by Paulsen *et al.* (2012) showing that addition of culinary sauces to salmon reduced salmon flavor intensities and increased basic taste intensities corresponding to the culinary sauces. Cherdchu and Chambers (2014) investigated the effect of different carriers (chicken broth, white rice and grilled chicken) on soy sauce perception. Differences between soy sauces were still perceived regardless of the presence of different carriers. However, white rice and grilled chicken tended to modify the sensory properties of soy sauces to a larger extent than liquid chicken broth. Cherdchu and Chambers (2014) suggested that these differences in perception might be related to the solid versus liquid consistency of the foods. Recently, we showed that condiments (cheese, cream cheese and mayonnaise) affected the sensory perception of carriers (bread, cracker) by influencing the structural transitions during oral processing (van Eck *et al.* 2019b). Carriers were shown to absorb moisture from the liquid-like condiments throughout mastication, and consequently they were perceived as less firm and less dry. All these studies discussed above indicate that the sensory profile of foods was altered by the addition of condiments, and vice versa. However, limited information is currently available on the influence of certain product properties of foods, *e.g.* mechanical properties, absorption capability, moisture content, fat content, viscosity, etc., on the sensory perception of carrier-condiment combinations. Further understanding on the relations between such food properties and sensory perception of carrier-condiment combinations may be used to control sensory perception or liking of different carrier-condiment combinations.

Next to studies on sensory perception, also sensory assessments are generally performed with carrier foods or condiments alone rather than with combinations thereof. However, condiments such as mayonnaise, ketchup or mustard are nearly always consumed in combination with other foods. This raises the question how perception of condiments and carriers changes when one is added to the other. Combining carriers with condiments is thought to increase stimulus complexity and to decrease sensitivity to detect differences between samples (Kroll & Pilgrim 1961; Lawless & Heymann 2010; Carpenter *et al.* 2012; Stone *et al.* 2012). For example, while condiments differing in composition might be perceived different when consumed on their own, perceptual differences between them might disappear when the condiments are added to certain foods. Hence, investigating the sensory perception of condiments that are added to different carriers provides essential information that is more representative of the natural consumption context. Similarly, carriers to which a condiment is added may also be perceived differently.

The aim of this study was to investigate how dynamic and static sensory perceptions of foods change when they are combined with condiments. In this study, carrier foods (bread, carrot) varying in hardness (soft, medium, hard) were combined with condiments (mayonnaise) varying in fat content and viscosity. These carrier-condiment combinations represent simplified model food for sandwiches and salads. We decided for this study to combine one carrier with one condiment only to be able to vary mechanical properties and composition in a controlled manner. Bread and carrot are expected to influence the sensory profiles of mayonnaises differently, since bread absorbs (part of) the mayonnaise resulting in a compact bolus, whereas carrots do not absorb mayonnaise leading to a bolus of dispersed carrot particles in a fluid mayonnaise/saliva matrix. We hypothesize that both carriers and condiments influence the dynamic sensory perception of carrier-condiment combinations, but that the texture sensations of the solid carriers dominate the perception. The combination of carriers with a condiment is expected to decrease the sensory intensity scores of both the carriers and the condiments. We hypothesize that discrimination sensitivity between carriers or condiments is decreased when assessed as carrier-condiment combinations.

6.2 MATERIALS AND METHODS

6.2.1 Samples

Carrier foods and condiments were combined to form carrier-condiment combinations with varying properties. Two commercial carriers were used, namely bread and carrots. These foods were selected based on their difference in bolus formation (*i.e.* bread can absorb (part of) a condiment, whereas carrots do not absorb condiments). One type of commercial condiment was selected, namely mayonnaise. 24 carrier-mayonnaise combinations were prepared following a 2x3x4 design: two carriers (bread, carrot) with three hardness levels (soft, medium, hard) were combined with four mayonnaises varying in fat content and viscosity (full fat/high viscosity, low fat/high viscosity, low fat/medium viscosity, low fat/low viscosity). Detailed sample preparation is explained in the following paragraphs.

Bread samples without crust were prepared by using different preparation methods for fresh white bread (Plaisir de mie toastbrood, Jacquet®, France) to obtain bread samples with three hardness levels (soft, medium, hard). Fresh bread was used as the soft bread, it was toasted for 3 min (Philips health grill HD4409, setting 3) to obtain medium bread, and it was oven-dried for 40 min at 100ºC (Venti-line, VWR®) to obtain hard bread. The soft and medium bread (SB and MB) were prepared just before sensory evaluation in order to maintain its texture, whereas hard bread (HB) was prepared one day before the sensory evaluation and stored in airtight containers. Bread was served in squares of 35x35x8 mm of approximately 2 g for soft and medium bread and of approximately 1.5 g for hard bread. To characterize the difference in hardness between the three bread samples, uniaxial compression tests were performed with a Texture Analyzer (TA.XT Plus, Stable Micro Systems, United Kingdom) fitted with a 50 kg load cell. A plate with a diameter of 100 mm was used. Measurements were performed at room temperature (20 ± 1 °C) with a constant speed of 1 mm/s in five replicates. The mean force needed to compress the bread samples (surface area = 1225 mm^2) to 20% strain was 4.8±0.4, 16.4±1.2 and 127.5±8.5 N (mean±SE) for SB, MB and HB, respectively (Table 6.1).

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Fresh carrots (winter carrot; purchased from local retailer in Wageningen, Netherlands) were cooked sous-vide to obtain carrots with three hardness levels (soft, medium, hard). First, carrots were peeled, cut in beams (90x15x15 mm), and vacuum packed into heat-resistant plastic bags. Carrots were cooked at 90ºC for 60 min to obtain soft carrots (SC), at 85ºC for 45 min to obtain medium carrots (MC), and at 85ºC for 5 min to obtain hard carrots (HC) with a cooked carrot flavor. After cooking, all bags were cooled in ice water and stored in the refrigerator (4 ºC) for up to three days. Carrots were cut into cubes of 15x15x15 mm of approximately 4 g just before sensory evaluation. To characterize the difference in hardness between the three carrot samples, uniaxial compression tests were performed with a Texture Analyzer (TA.XT Plus) fitted with a 50 kg load cell. A plate with diameter of 100 mm was used. Measurements on carrot cubes were performed at room temperature (20 ± 1 °C) with a constant speed of 1 mm/s up to a strain of 80%. The orientation of the carrot cube relative to the uniaxial compression direction was not controlled for. The mean fracture stress of carrots, prepared on different days, was measured in triplicate. The mean fracture stresses were 125±5, 305±11 and 1490±26 kPa (mean±SE) for SC, MC and HC, respectively (Table 6.1).

Three commercial mayonnaises varying in fat content and viscosity were used, namely full fat/high viscosity (FF-H; Mayonaise, Calvé®, Unilever, The Netherlands), low fat/high viscosity (LF-H; Licht & Roming, Calvé®, Unilever, The Netherlands), and low fat/low viscosity (LF-L; Slasaus Naturel, Calvé®, Unilever, The Netherlands) mayonnaises. A fourth mayonnaise with low fat/medium viscosity (LF-M) was obtained by mixing the commercially available LF-H and LF-L mayonnaises in a 1:1 weight ratio. The fat content was taken from the nutritional information on the product label, and corresponded to 69, 27, and 26 wt% for FF-H, LF-H, and LF-L mayonnaise, respectively. Accordingly, the fat content of the prepared LF-M was 26.5%.

Table 6.1: Overview and codes of the carriers (bread, carrot), mayonnaises and carrier-mayonnaise combinations included in the present study. Hardness values of the bread and carrots and fat content and viscosity of the mayonnaises are given.

		Mayonnaises (n=4)			
		Full fat (FF) High viscosity (H)	Low fat (LF) High viscosity (H)	Low fat (LF) Medium viscosity (M)	Low fat (LF) Low viscosity (L)
	Fat content, viscosity[*] / *Hardness*[**]	*69 wt%* *12.9, 3.7 Pa·s*	*27 wt%* *13.1, 4.3 Pa·s*	*26.5 wt%* *6.6, 2.3 Pa·s*	*26 wt%* *3.7, 1.4 Pa·s*
Bread (n=3)					
Hard (HB)	*127.5±8.5 N*	HB FF-H	HB LF-H	HB LF-M	HB LF-L
Medium (MB)	*16.4±1.2 N*	MB FF-H	MB LF-H	MB LF-M	MB LF-L
Soft (SB)	*4.8±0.4 N*	SB FF-H	SB LF-H	SB LF-M	SB LF-L
Carrot (n=3)					
Hard (HC)	*1490±26 kPa*	HC FF-H	HC LF-H	HC LF-M	HC LF-L
Medium (MC)	*305±11 kPa*	MC FF-H	MC LF-H	MC LF-M	MC LF-L
Soft (SC)	*125±5 kPa*	SC FF-H	SC LF-H	SC LF-M	SC LF-L

* Mayonnaises varying in fat content and viscosity were used. The fat content (wt%) was taken from the nutritional information on the product label. Flow curves were determined to characterize the difference in viscosity (Pa·s, at shear rates of 10 and 50 s-1) between the four mayonnaises.

** Hardness of the bread and carrots was varied. Uniaxial compression tests were performed to characterize the difference in hardness between the three bread samples (force in N to compress to 20% strain, mean±SD) and the three carrot samples (fracture force in kPa, mean±SD).

To characterize the difference in viscosity between the four mayonnaises, flow curves were determined using a rheometer (MCR 301 Rheometer, Anton Paar Benelux BVBA, Belgium) equipped with an Inset I-PP50/SS plate and a CP50-1 cone. The viscosity of the mayonnaises was measured by putting 0.5 g mayonnaise on the plate and shearing at shear rates ranging from 1 s-1 to 1000 s-1 after a resting period of 5 minutes. Viscosities of 12.9, 13.1, 6.6 and 3.7 Pa·s at a shear rate of 10 s-1 and viscosities of 3.7, 4.3, 2.3 and 1.4 Pa·s at a shear rate of 50 s-1 were obtained for FF-H, LF-H, LF-M and LF-L mayonnaise, respectively (Table 6.1). Mayonnaise was served at a weight of approximately 2 g.

Carrier-mayonnaise combinations were prepared just before serving in order to minimize moisture transfer of the mayonnaises into the carriers before consumption. The mayonnaise was served on top of the carriers and covered them completely. An overview of all carrier-mayonnaise combinations with its codes is given in Table 6.1.

6.2.2 Consumer panel

Subjects were recruited from Wageningen University & Research campus using flyers, posters and social media. All subjects were Caucasian adults and were consumers of bread, carrot and mayonnaise on a regular basis. Other inclusion criteria were no allergies or intolerances for gluten or eggs, good dental health (self-reported), and non-smoking habits (self-reported). None of the subjects was familiar with the sensory methodologies used in the present study nor had any previous training in the sensory evaluation of bread, carrot or mayonnaise. 64 consumers (16 male and 48 female, 18 – 62 years) were recruited for the Temporal Dominance of Sensations (TDS) evaluations, and 66 consumers (17 male and 49 female, 18 – 58 years) for the Rate-All-That-Apply (RATA) evaluations. 33 subjects participated in both tests (TDS and RATA), all other subjects performed either TDS or RATA. RATA evaluation was performed at least two months after TDS evaluation, and no effect of previous TDS evaluation on the RATA evaluation was observed. Subjects gave written informed consent before the start of the study and they received financial compensation for their participation.

6.2.3 Sensory evaluation procedures

Environmental conditions

The sensory evaluations were carried out at the sensory facilities at Wageningen University & Research. Samples were presented with 3 digit codes, and served at room temperature in randomized order according to a completely randomized design. Bread and bread-mayonnaise combinations were served on a plate, whereas carrot, mayonnaise and carrot-mayonnaise combinations were served on a spoon. Subjects were seated in individual sensory booths, and evaluated the samples under normal light conditions. Subjects were instructed to not eat, drink or brush their teeth one hour prior to the test, and to not wear strong perfume or lotion during the test. Subjects cleaned their palate after each sample with water, tea (Jasmine green tea, Twinings, UK) and green apple for at least 1 min. Green apple was used as palate cleanser instead of crackers as commonly done, since cracker is comparable to the hard bread used in the present study, and therefore not preferred as a palate cleanser.

Temporal Dominance of Sensations (TDS) and liking

Bread and carrots with and without mayonnaise were evaluated using Temporal Dominance of Sensations (TDS) (n=64 subjects). Mayonnaises alone were not assessed by TDS due to their considerably shorter consumption times compared to the other samples.

The sensory attribute lists were prepared based on a literature search followed by a pilot test with 8 consumers (not participating in the real experiment, 4 male and 4 female, age between 22 and 29 years) using a Check-All-That-Apply (CATA) methodology. They could add any missing attribute to the provided list. The attributes that were most frequently selected were included in the final attribute list. Ten sensory attributes were included for the evaluation of bread with and without mayonnaise (Table 6.2A) and ten sensory attributes were included for the evaluation of carrot with and without mayonnaise (Table 6.2B).

Each subject attended 2 sessions of 60 min over a time period of three weeks. Subjects evaluated bread with and without mayonnaise in one session and carrot with and without mayonnaise in the other session. Bread and carrot evaluations were counterbalanced, meaning that half of the subjects started with the bread evaluations and half of the subjects started with the carrot evaluations. Subjects received the attributes and definitions by email before each session, and they were instructed to familiarize themselves with these attributes. An introduction to the sensory attributes and a live demonstration of the procedures were given at the start of each session to familiarize the subjects with the attributes and procedures. Each session started with a warm-up sample (SB with FF-H, MC with LF-M) so that subjects could familiarize themselves with the procedure and the attributes. Subjects evaluated first 7 samples, followed by a break, and then continued with 8 samples.

For each sample, subjects were instructed to put the entire sample in the mouth and simultaneously click the start button. Then, they had to select the attributes that were perceived dominant (*i.e.* the attribute that catches most of the attention at that moment in time) during consumption by a single click on the attribute button. The selected attribute remained dominant until a new attribute was selected. Subjects were free to select as many attributes as they perceived and they could select the same attribute multiple times. Subjects were instructed to click the stop button when perception ended after swallowing the sample. Attributes order was randomized for each subject, but not for each sample. Data were acquired by TimeSens software (version 1.1.601.0, ChemoSens, FR).

Table 6.2: Sensory attributes and definitions used during TDS evaluation. Different attribute lists were used for the evaluation of bread with and without mayonnaise (A) and for the evaluation of carrot with and without mayonnaise (B).

	Sensory attribute	Definition
(A) Bread session		
Basic taste	Salty	Salty taste, associated with salt
	Sour	Sour taste, associated with sour ingredients such as lemon
	Sweet	Sweet taste, associated with sugar
Texture/Mouthfeel	Adhesive	Sensation of adhesiveness, due to adherence of bread to the teeth/molars/palate
	Creamy	Sensation of creaminess, described as a full, fatty or smooth mouthfeel
	Crispy	Sensation of crispiness, described as the force required to bite/chew the bread while causing a high sound
	Dry	Sensation of dryness, due to the absence of water or a lack of saliva
	Soft	Sensation of softness, described as no or little force required to bite/chew the product
	Thick	Sensation of thickness, associated with products with a high viscosity
	Thin	Sensation of thinness, associated with products with a low viscosity (liquid-like)
(B) Carrot session		
Basic taste	Salty	Salty taste, associated with salt
	Sour	Sour taste, associated with sour ingredients such as lemon
	Sweet	Sweet taste, associated with sugar
Texture/Mouthfeel	Creamy	Sensation of creaminess, described as a full, fatty or smooth mouthfeel
	Crunchy	Sensation of crunchiness, described as the force required to bite/chew the carrot while causing a low sound
	Soft	Sensation of softness, described as no or little force required to bite/chew the product
	Particles	Sensation of particles, due to the presence of carrot pieces in the mouth
	Thick	Sensation of thickness, associated with products with a high viscosity
	Thin	Sensation of thinness, associated with products with a low viscosity (liquid-like)
	Watery	Sensation of watery, due to the presence of water/moisture

Rate-All-That-Apply (RATA)

Bread, carrot, mayonnaise, bread-mayonnaise combinations and carrot-mayonnaise combinations were evaluated using the Rate-All-That-Apply (RATA) methodology (n=66 consumers). Currently, there is no standard method for the evaluation of carrier-condiment combinations. Therefore, the RATA methodology was adapted to allow the assessment of such combinations. Sensory evaluation was divided into two parts: (1) evaluation of carrier characteristics for carriers and carrier-mayonnaise combinations, (2) evaluation of mayonnaise characteristics for mayonnaises and carrier-mayonnaise combinations. Three attribute lists descriptive for bread, carrot or mayonnaise were used. Subjects were instructed

Table 6.3: Sensory attributes and definitions used during the RATA evaluation. Different attribute lists were used for the evaluation of bread characteristics, carrot characteristics and mayonnaise characteristics.

	Bread	**Carrot**	**Mayonnaise**	**Definition**
Basic taste	Bitter	Bitter	Bitter	Bitter taste, associated with coffee or caffeine
	Salty	Salty	Salty	Salty taste, associated with salt
	Sour	Sour	Sour	Sour taste, associated with sour ingredients such as lemon
	Sweet	Sweet	Sweet	Sweet taste, associated with sugar
Flavor	Baked			Baked flavor, associated with freshly baked bread
		Cooked		Cooked flavor, associated with cooked/processed carrots
			Dairy	Dairy flavor, associated with creamy dairy products like yogurt or ice cream
		Green		Green flavor, associated with green plants or vegetables
	Yeast			Yeast flavor, characterized as a fermented or yeast-like, associated with beer or bread
Texture/ Mouthfeel	Adhesive	Adhesive	Adhesive	Sensation of adhesiveness, due to adherence of product to the teeth/molars/palate
	Chewy	Chewy	-	Sensation of chewiness, described as resistance to chewing/breakdown
	Cohesive	Cohesive	Cohesive	Sensation of cohesiveness, described as the degree to which the product stays together or "remains as a whole"
	-	-	Creamy	Sensation of creaminess, described as a full, fatty or smooth mouthfeel
	Crispy	-	-	Sensation of crispiness, described as the force required to bite/chew the bread while causing a high sound
	-	Crunchy	-	Sensation of crunchiness, described as the force required to bite/chew the carrot while causing a low sound
	Dense	-	-	Sensation of denseness, due to the absence of air cavities in the bread, associated with a compact bread
	Dry	-	-	Sensation of dryness, due to the absence of water or a lack of saliva
	-	Fibrous	-	Sensation of fibrousness, associated with woodiness or the presence of fibers
	Grainy	-	-	Sensation of graininess, due to the presence of granular bread pieces/particles in the mouth
	-	Particles	-	Sensation of particles, due to the presence of carrot pieces in the mouth
	-	-	Salivating	Sensation of salivating or mouthwatering, due to secretion of saliva during food consumption
	Soft	Soft	Soft	For bread/carrot: Sensation of softness, described as no or little force required to bite/chew the product For mayonnaise: Sensation of softness or velvety, described as a low force required to deform the mayonnaise
	-	-	Thick	Sensation of thickness, described as the resistance to flow, associated with products with a high viscosity
	-	Watery	-	Sensation of watery, due to the presence of water/ moisture
After taste	After taste	After taste	After taste	After taste, described as the bread/carrot/mayonnaise taste/flavor intensity that remains in the mouth
After feel	Residue	Residue	-	Sensation of residue, due to product residue/pieces that remain in the mouth
	-	-	Residual film	Sensation of residual film, sensation of mayonnaise residue or fatty film that remains in the mouth

to focus on sensory perception of one of the food items (bread, carrot or mayonnaise) when carrier-mayonnaise combinations were assessed. Hence, subjects assessed bread attributes for bread and bread-mayonnaise combinations, carrot attributes for carrot and carrot-mayonnaise combinations, and mayonnaise attributes for mayonnaises, mayonnaise-bread and mayonnaise-carrot combinations during different sessions.

The method was piloted by a feasibility test with 6 consumers (not participating in the real experiment, 3 male and 3 female, age between 22 and 35 years). The pilot test confirmed that consumers could evaluate attributes related to either the carrier or mayonnaise when a carrier-mayonnaise combination was served. Furthermore, all attributes that were selected or mentioned during this feasibility test were included in the final attribute lists. Sixteen, sixteen and thirteen sensory attributes were included for the evaluation of bread, carrot and mayonnaise characteristics, respectively (Table 6.3).

Each subject attended 5 sessions of 60 min over a time period of five weeks. Subjects attended one familiarization session, after which they participated in four sensory evaluation sessions. The familiarization session was used to acquaint the subjects with the samples, the sensory attributes and definitions, and the RATA evaluation procedure (including the 9-box scale and its anchors). When subjects reported that the procedures were clear, they continued with a practice exercise. They practiced with the bread attribute list for two bread-mayonnaise combinations, with the carrot attribute list for two carrot-mayonnaise combinations, and with the mayonnaise attribute list for two bread-mayonnaise and two carrot-mayonnaise combinations. Samples used in this session were soft carriers (bread and carrot) with LF-L mayonnaise and hard carriers with FF-H mayonnaise, which represented the sample scope of the present study. During this exercise, it was highlighted that subjects had to evaluate only one of the food items present (bread, carrot or mayonnaise) depending on the attribute list they were evaluating. After the exercise, the origin of the selected attributes (bread, carrot or mayonnaise) was discussed with subjects to make sure that they indeed evaluated either the bread, carrot or mayonnaise characteristics.

The four sensory evaluation sessions were organized by attribute list and food type: bread characteristics of bread and bread-mayonnaise combinations were evaluated in one session, carrot characteristics of carrots and carrot-mayonnaise combinations were evaluated in one session, mayonnaise characteristics of mayonnaises and bread-mayonnaise combinations were evaluated in one session, and mayonnaise characteristics of mayonnaises and carrot-mayonnaise combinations were evaluated in one session. The order of the sessions was randomized between subjects. Subjects received the attributes and definitions by email before each session, and were instructed to familiarize themselves with these attributes. Each session started with a warm-up sample (SB with FF-H, MC with LF-M) so that subjects could familiarize themselves with the procedure. For one breads session and one carrot session, subjects evaluated first 7 samples, followed by a break, and then continued with 8 samples (in total 15 bread samples with/without mayonnaise and 15 carrot samples with/without mayonnaise). For the two mayonnaise sessions, subjects evaluated first 7 samples, followed by a break, and then continued with 7 samples (in total 28 mayonnaise samples with/without carriers).

For each sample, subjects were instructed to check all attributes that applied and subsequently score the perceived intensity of these attributes on a 9-box scale anchored from low to high. Subjects were instructed to consume the sample in two bites. They were instructed to evaluate the taste/flavor attributes after the first bite, to evaluate the texture/mouthfeel attributes after the second bite, and to thereafter evaluate the after taste and after feel attributes. Attributes order was randomized within modality (taste, flavor, texture/mouthfeel) for each subject, but not for each sample. Data were acquired by Qualtrics survey software (version November 2017, Qualtrics, USA).

6.2.4 Data analysis

TDS data analysis

Bread and carrot data were analyzed separately. Panel performance was checked, and all data were included in the data analysis. TDS data was time-standardized between 0 (*i.e.* time at which the first attribute was selected) and 1 (*i.e.* time that the stop button was selected). TDS curves and band plots were generated to visualize the data. Chance line of 0.10 and significance level of 0.05 were used. Band plots were displayed above the TDS curves to allow following the evolution of dominant attributes along consumption time easily (Galmarini, Visalli, & Schlich, 2017). While TDS curves represent the dominance rate of all attributes (significant and not significant attributes) over standardized time, band plots report only the significant dominant attributes over time without detailing dominance rates. Consequently, TDS curves provide more detailed information about dynamic sensory perception whereas band plots allow for a more convenient, visual comparison of multiple samples. Principal Component Analysis (PCA) with an ellipse level of confidence of 0.95 was performed on the dominance durations of sensations to evaluate the differences between samples. PCA sensory trajectories were obtained according to the method described by Lenfant *et al.* (2009), to evaluate differences between samples during the mastication period. Samples were plotted over three time points corresponding to early (33%), middle (66%) and late chew down (100%), which is about one time point per 5 s of consumption. Similar trajectories were obtained when shorter time intervals were chosen (data not shown). TimeSens software (version 1.1.601.0, ChemoSens, FR) was used to plot all figures.

RATA data analysis

Results were reported as mean values with standard error (n=66 subjects). Non-checked attributes were treated as intensity=0, and RATA intensity scores (0-9) were treated as continuous data (Meyners *et al.* 2016; Oppermann *et al.* 2017). Data were checked for serving order, and serving order did not significantly affect any of the sensory attributes ($p>0.05$). Linear mixed models were performed for all attributes of all three attribute lists (Kuznetsova *et al.* 2016). For these analyses, carrier, mayonnaise and carrier:mayonnaise were set as fixed effects, and subject was set as random effect. Significance level of $p<0.05$ was chosen. PCA with an ellipse level of confidence of 0.95 was performed to summarize the relationships between sensory attributes (Husson *et al.* 2014). R language (RStudio, version 1.0.143) was used to perform all statistical tests.

6.3 RESULTS

6.3.1 Temporal Dominance of Sensations (TDS) of bread or carrots with and without mayonnaise

Band plots and TDS curves were obtained for carriers that were evaluated with and without mayonnaise. Figure 6.1 shows a selection of the band plots and TDS curves representing the range of products investigated, including bread and carrot with their two most different hardness levels (hard, soft) without and with one mayonnaise (LF-L). The band plots and TDS curves of the remaining samples are shown in Appendix 1 and 2. Visual inspection of TDS curves and band plots showed clear differences between the three hardness levels for both bread and carrots. For hard bread, crispy was the dominant sensation at the beginning, dry at the middle and adhesive at the end of consumption (Figure 6.1A). For soft bread, soft and dry were dominant sensations at the beginning of consumption and sweet and adhesive from the middle towards the end of consumption (Figure 6.1C). A higher number of attributes were dominant for medium bread, and these dominant attributes (crispy, soft, dry, sweet and adhesive) coincided with the dominant attributes of both the hard and soft bread. In the case of carrots, hard carrots were perceived crunchy at the beginning and particles and sweet towards the end of consumption (Figure 6.1E), whereas soft carrots were perceived soft at the beginning, and sweet and watery from the middle towards the end of consumption (Figure 6.1G). For medium carrots, the dominant sensations changed more frequently during consumption, and these sensations (crunchy, soft, sweet, particles, watery) go along with the ones selected in hard and soft carrots.

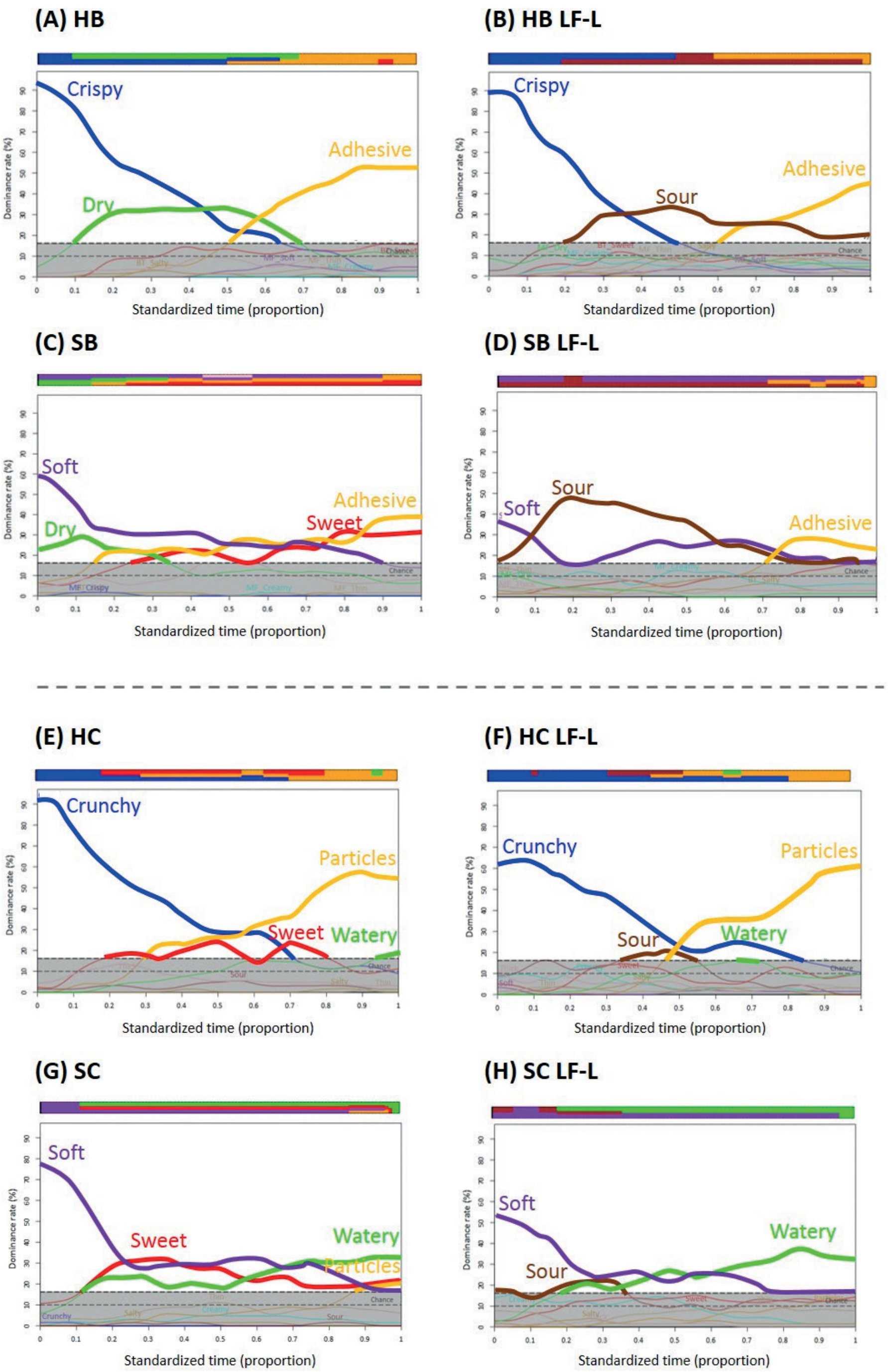


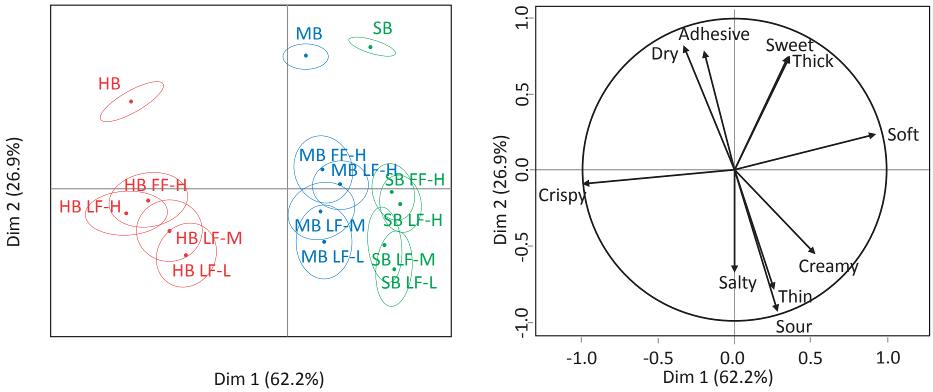
Figure 6.1: Band plots and TDS curves of bread alone (A, C), bread-mayonnaise combinations (B, D), carrots alone (E, G), and carrot-mayonnaise combinations (F, H) over standardized eating time. Carriers without mayonnaise are presented in the left column and carrier-mayonnaise combinations are presented in the right column. The abbreviations are explained in Table 6.1.

Visual inspection of the band plots and TDS curves shows some differences for carriers without and with mayonnaises (Figure 6.1). In the case of bread, dry was not a significantly dominant sensation anymore when mayonnaise was present, while in the case of carrots, sweet was not a significantly dominant sensation anymore. These attributes were replaced by attributes related to mayonnaises, *e.g.* creamy, sour and sweet, especially at the middle of consumption. The dominant sensations depended on the type of mayonnaise, *i.e.* creamy, sour and sweet for FF-H, creamy and sour for LF-H and LF-M, and sour for LF-L (Figure 6.1B, 6.1D, 6.1F, 6.1H). Such sensations, for example sourness, tended to appear in an earlier stage of consumption when mayonnaises were combined with soft carriers than when combined with hard carriers.

Figure 6.2 presents the PCA plots on dominance durations for carriers with and without mayonnaises. In the case of bread (Figure 6.2A), 89.1% of total variance was explained by the first two components. PC1 (62.2%) distinguished between the three hardness levels and PC2 (26.9%) distinguished between bread and bread-mayonnaise combinations. In the case of carrots (Figure 6.2B), 94.9% of total variance was explained by the first two components. PC1 (84.3%) distinguished between the three hardness levels and PC2 (10.6%) distinguished mainly between carrots and carrot-mayonnaise combinations.

Sensory trajectories of the dominance rates at different time points are shown in Figure 6.3. The first two principal components account for 80.1% of the total variance among bread samples with and without mayonnaise and for 81.9% of the total variance among carrot samples with and without mayonnaises. An effect of hardness on the sensory trajectories was observed for both bread and carrots. In case of bread (Figure 6.3A), hard, medium and soft bread started the trajectories at very different positions (red, blue and green), but the difference in position decreased towards the end of the trajectories with all samples converging towards adhesive, sweet and thick. A different pattern was observed for carrots (Figure 6.3B). Hard, medium and soft carrots started the trajectories at very different positions, and the differences in position remained towards the end of the trajectories. For both bread and carrots, a relatively small effect of mayonnaise addition on the trajectories was observed. The type of mayonnaise did not strongly affect the sensory trajectories of carrier-mayonnaise combinations.

(A) bread



(B) carrot

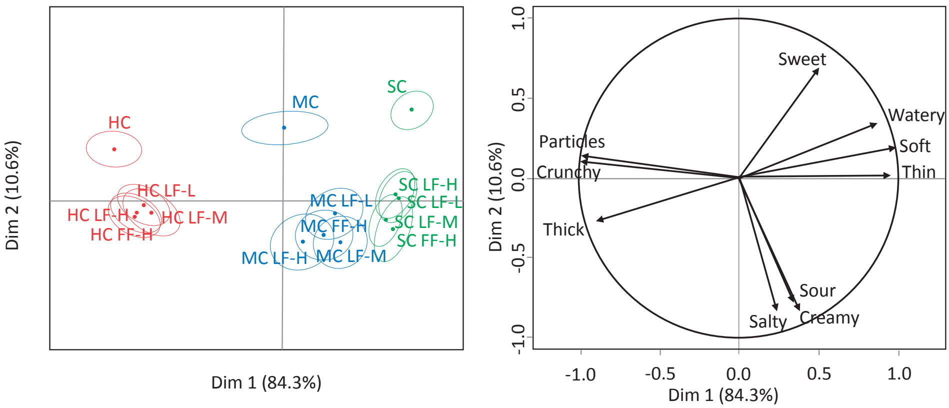


Figure 6.2: Principal Component Analysis (PCA) on the TDS dominance durations for bread with and without mayonnaises (A) and carrot with and without mayonnaises (B). The individuals map (samples) is shown on the left, and the variables map (attributes) is shown on the right. The ellipse level of confidence was 0.95. Different colors indicate carriers varying in hardness (red: hard carriers, blue: medium carriers, green: soft carriers). The abbreviations are explained in Table 6.1 and the attributes are explained in Table 6.2.

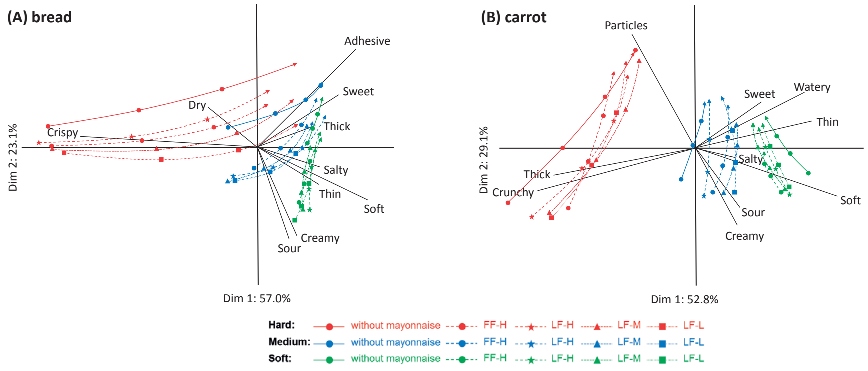


Figure 6.3: Principal Component Analysis (PCA) bi-plot representing the sensory trajectories of bread with and without mayonnaises (A) and carrot with and without mayonnaises (B) over standardized perception time. The dots represent 0, 33, 66 and 100% of perception time. The lines are added to guide the eye. The abbreviations are explained in Table 6.1 and the attributes are explained in Table 6.2.

6.3.2 Rate-All-That-Apply (RATA)

Rate-All-That-Apply (RATA) of bread and carrots with and without mayonnaise

The intensity scores of bread samples with and without mayonnaises are summarized in Table 6.4. A hardness effect was observed for 14 out of 16 attributes, indicating clear differences between the three bread samples with varying hardness level. A mayonnaise effect was observed for 7 out of 16 attributes, and no hardness:mayonnaise interactions were found. On average, adding mayonnaises to bread increased salty and sour intensity of bread, whereas it decreased sweet, yeasty, crispy, dry and after taste intensity of bread. Low fat mayonnaises tended to increase bread sourness and to decrease bread sweetness and after taste to a larger extent than full fat mayonnaises.

Table 6.5 summarizes the mean intensity scores of carrots with and without mayonnaises. A hardness effect was found for 12 out of 16 attributes, a mayonnaise effect was observed for 6 out of 16 attributes, and no hardness:mayonnaise interactions were found. On average, addition of mayonnaises to carrots increased sour and decreased bitter, sweet, green and after taste intensities of carrots. Low fat mayonnaises tended to increase carrot sourness and wateriness and to decrease carrot sweetness to a larger extent than full fat mayonnaises. These results are comparable with those found for the bread samples.

Figure 6.4 presents PCA plots on sensory intensity scores of carriers that were assessed with and without mayonnaise. The first two components explained 80.1% and 87.4% of the total variances for the bread (Figure 6.4A) and carrots (Figure 6.4B), respectively. Similar results were obtained for both carriers. PC1 (65.9% for bread, 73.5% for carrots) distinguished between the three hardness levels, and PC2 (14.2% for bread, 13.9% for carrot) accounted for the presence of mayonnaises. 95%-confidence ellipses indicate that soft, medium and hard carriers without mayonnaise differed significantly from each other. It can also be observed that soft, medium and hard carriers were still perceived different from each other upon the addition of mayonnaise regardless of its fat content or viscosity. One exception was observed for bread with LF-H mayonnaise, as the ellipses of SB LF-H and MB LF-H overlapped.

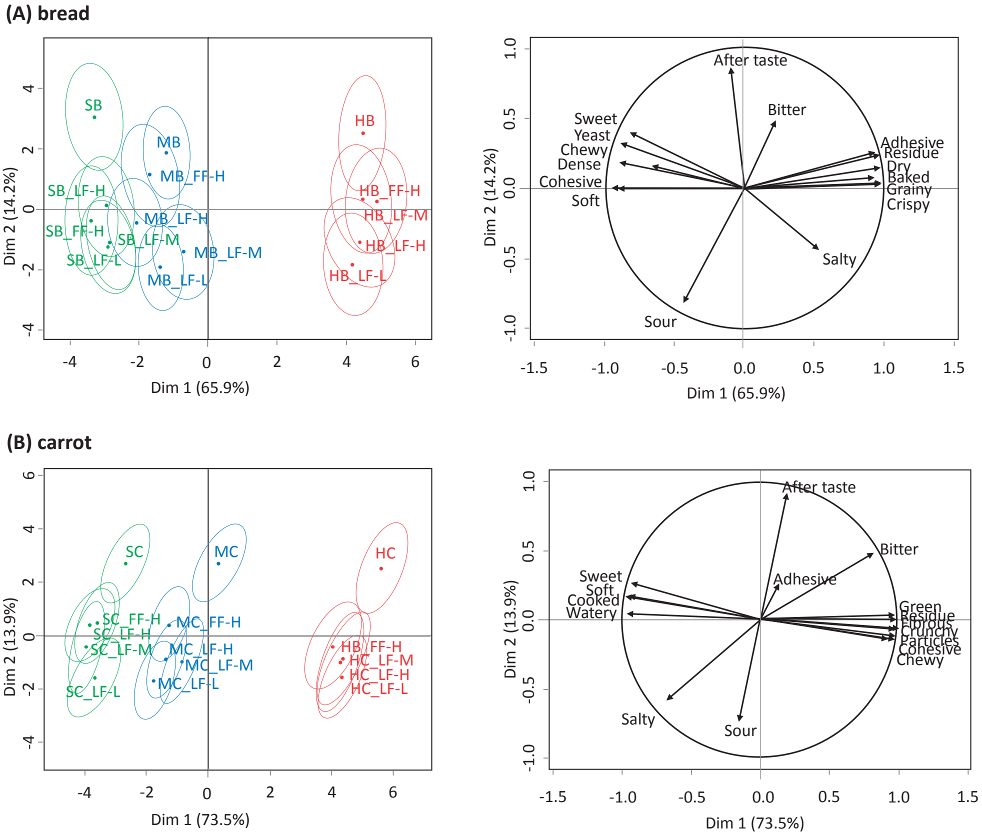


Figure 6.4: Principal Component Analysis (PCA) on the RATA intensity scores for bread with and without mayonnaises (A) and carrot with and without mayonnaises (B). The individuals map (samples) is shown on the left, and the variables map (attributes) is shown on the right. The ellipse level of confidence was 0.95. Different colors indicate carriers varying in hardness (red: hard carriers, blue: medium carriers, green: soft carriers). The abbreviations are explained in Table 6.1 and the attributes are explained in Table 6.3.

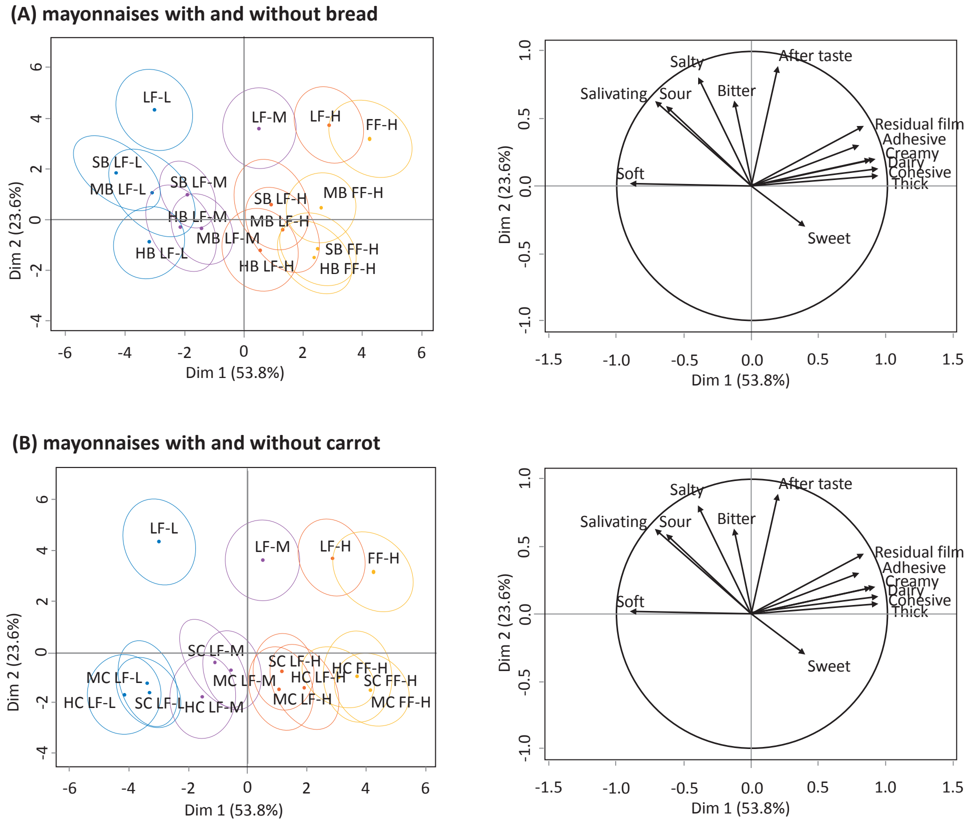


Figure 6.5: Principal Component Analysis (PCA) on the RATA intensity scores for mayonnaises with and without bread (A) and mayonnaises with and without carrot (B). The individuals map (samples) is shown on the left, and the variables map (attributes) is shown on the right. The ellipse level of confidence was 0.95. Different colors indicate mayonnaises varying in fat content and viscosity (yellow: FF-H, orange: LF-H, purple: LF-M, blue: LF-L). The abbreviations are explained in Table 6.1 and the attributes are explained in Table 6.3.

Rate-All-That-Apply (RATA) of mayonnaises with and without bread or carrots

The mean intensity scores of mayonnaises without and with bread or carrots are summarized in Table 6.6. A mayonnaise effect was observed for 12 out of 13 attributes showing that the four mayonnaises with varying fat content and viscosity were perceived clearly different. A carrier effect was observed for all attributes. On average, the presence of carrot or bread decreased the intensity scores of the attributes salty, sour, dairy, adhesive, cohesive, creamy, salivating, thick, after taste and residual film of the mayonnaises. Sour intensity scores decreased to a larger extent when mayonnaises were combined with carrots than with bread, whereas creamy intensity scores decreased to a larger extent when mayonnaises were combined with bread instead of with carrots. Sour and dairy intensity scores decreased to a larger extent with increasing bread hardness, and creamy and after taste intensity scores decreased to a larger extent with increasing carrot hardness. A mayonnaise:carrier interaction was observed for thick ($F=3.8$, $p<0.001$), indicating that the effects of carriers on mayonnaise perception were not the same for each mayonnaise. Bread lowered the thickness perception of all four mayonnaises, whereas carrots only lowered the thickness perception of mayonnaise with medium and low viscosity, *e.g.* LF-M and LF-L, but not with high viscosity, *e.g.* FF-H and LF-H (data not shown).

Figure 6.5 presents PCA plots on mayonnaise intensity scores of mayonnaises that were consumed with and without bread (Figure 6.5A) or carrots (Figure 6.5B). The first two components explain 77.4% of the total variance. PC1 (53.8%) distinguishes between the four mayonnaises, and PC2 (23.6%) accounts for the respective presence of bread or carrots. 95%-confidence ellipses indicate that LF-L, LF-M and FF-H mayonnaises differed from each other (*i.e.* non-overlapping ellipses), and that LF-H did not differ from LF-M and FF-H. It can also be observed that this discrimination between mayonnaises was influenced by the presence of bread (Figure 6.5A) or carrots (Figure 6.5B). With the presence of bread, regardless of its hardness, the ellipses of LF-L and LF-M overlapped but they differed from LF-H and FF-H mayonnaise. With the presence of hard or medium carrots, LF-L, LF-M and FF-H mayonnaises differed from each other, but LF-H overlapped with FF-H or LF-M. In the case of soft carrots, all four mayonnaises differed from each other.

6.4 DISCUSSION

6.4.1 Dynamic sensory perception of carrier-mayonnaise combinations

When carriers (bread, carrot) and mayonnaise were combined in one bite, TDS results showed that sensations related to both food items were perceived dominant during consumption. The dynamic sensory profiles of carriers with mayonnaises were driven by the texture of the solid carriers, *e.g.* bread or carrot (Figure 6.1). In agreement with literature on single foods (Albert *et al.* 2012; Jager *et al.* 2014; Rodrigues *et al.* 2018), we observed that texture sensations related to the mechanical properties of foods (*i.e.* crispy, crunchy and soft) dominated the early stages of the consumption of carrier-mayonnaise combinations. In addition, we observed that texture sensations related to the bolus properties of carrier-mayonnaise combinations (*i.e.* adhesive, particles, watery) dominated the last stage of consumption. This is in accordance with previous studies on bread and wheat flakes, showing that the attribute stickiness became dominant towards the moment of swallowing (Peyron *et al.* 2011; Panouillé *et al.* 2014). This high impact of texture might be explained by the structure of the solid foods studied. As both bread and carrots are solids, they require a large amount of food breakdown and an increase in lubrication by the addition of saliva during oral processing in order to form a bolus that is safe to swallow. These changes during oral processing have a large effect on the sensory perception. This is in agreement with our earlier observations, in which we found that bread and crackers determined oral processing behavior of bread-condiment and cracker-condiment combinations (van Eck *et al.* 2019b).

Hence, the texture of solid carrier-condiment combinations is the main determinant for the dynamic sensory profiles rather than its taste.

The degree of hardness of bread and carrots clearly impacted the dynamic perception of mayonnaise, as mayonnaise-related attributes (*i.e.* creamy, sweet, sour) tended to appear in earlier stages of consumption when they were combined with soft carriers (*i.e.* at 0 – 15% of normalized consumption time) compared to when they were combined with hard carriers (*i.e.* at 10 – 40% of normalized consumption time). Changing food hardness changes oral processing behavior (*i.e.* longer chewing time, more salivation) and consequently taste and flavor release from the mayonnaise matrix. In addition, foods with a hard texture might distract more attention from the mayonnaises, thereby delaying dominant mayonnaise perception. To develop a full understanding on the role of carriers and condiments on dynamic perception of carrier-condiment combinations, additional studies including *in vivo* taste/flavor release with simultaneous sensory assessments will be required.

6

6.4.2 Effect of mayonnaise addition on the sensory properties of bread and carrots

Taste and flavor intensities of bread and carrots were clearly influenced by the presence of mayonnaises (Tables 6.4, 6.5). Mayonnaises reduced the characteristic taste and flavor intensities of the bread (*i.e.* sweet, yeast) and carrots (*i.e.* bitter, sweet, green). This is likely due to a dilution effect (Kroll & Pilgrim 1961), since the concentration of taste and flavor compounds of a single food is reduced by the addition of another food and thereby decreasing the intensity of its characteristic tastes and flavors. On the other hand, mayonnaises enhanced the salty intensity of bread and the sour intensity of both bread and carrots, indicating not only dilution of taste and flavor occurs but also enhancement. This enhanced intensity might be an effect of the taste profile of the mayonnaise directly, but another explanation is that consumers were not able to differentiate between the taste and flavor coming from mayonnaise and bread or carrots. The present results indicate that the stronger taste and flavor intensity of mayonnaises overruled the characteristic taste and flavor of the relatively bland bread and carrots. Although mayonnaises affected the taste and flavor intensities of bread and carrots in a similar way, additional effects on texture and mouthfeel intensities were observed depending on the textural properties of either bread or carrots. As the bread is able to take up moisture, presence of mayonnaise decreased bread crispiness and dryness probably due to moisture transfer from the mayonnaises into the bread (van Eck *et al.* 2019b). As the carrots do not have the ability to absorb moisture, very little effects of mayonnaise on carrot texture and mouthfeel were found. One exception was observed for carrots with LF-L mayonnaise, since the watery mouthfeel of carrots increased with the presence of the low viscosity mayonnaise. A possible explanation is that consumers were not able to differentiate between the water coming from the carrot and the mayonnaise, or that the higher sourness of the LF-L mayonnaise increased the saliva secretion leading to an increased watery perception.

Table 6.4: Significance values and mean intensity scores (± standard error of the mean) of bread attributes, obtained for bread samples without and with mayonnaises by using RATA methodology and the bread attribute list. The abbreviations are explained in Table 6.1 and the attributes are explained in Table 6.3.

	Bread Hardness		Mayonnaise		Bread varying in hardness (mean±SE)					
Taste/Flavor	F		F		HB		MB		SB	
Bitter	0.9	NS	0.2	NS	0.6 ± 0.1		0.5 ± 0.1		0.5 ± 0.1	
Salty	7.3	***	3.5	**	1.8 ± 0.1	*A*	1.7 ± 0.1	*A*	1.5 ± 0.1	*B*
Sour	8.2	***	16.0	***	0.8 ± 0.1	*B*	0.9 ± 0.1	*B*	1.1 ± 0.1	*A*
Sweet	13.5	***	3.3	*	2.4 ± 0.1	*B*	2.8 ± 0.1	*A*	3.0 ± 0.1	*A*
Baked	192.1	***	1.7	NS	5.1 ± 0.2	*A*	3.4 ± 0.1	*B*	1.8 ± 0.1	*C*
Yeast	73.3	***	5.6	***	2.4 ± 0.1	*C*	3.2 ± 0.1	*B*	4.0 ± 0.1	*A*
Mouthfeel/Texture										
Adhesive	77.1	***	1.8	NS	5.0 ± 0.1	*A*	3.7 ± 0.1	*B*	3.2 ± 0.1	*C*
Chewy	49.1	***	2.0	NS	2.4 ± 0.1	*C*	4.1 ± 0.1	*A*	3.1 ± 0.1	*B*
Cohesive	97.1	***	2.2	NS	1.4 ± 0.1	*C*	3.3 ± 0.1	*A*	3.0 ± 0.1	*B*
Crispy	3092.3	***	3.1	*	7.8 ± 0.1	*A*	1.6 ± 0.1	*B*	0.2 ± 0.0	*C*
Dense	57.0	***	1.9	NS	1.4 ± 0.1	*B*	2.9 ± 0.1	*A*	2.6 ± 0.1	*A*
Dry	403.4	***	14.4	***	5.8 ± 0.2	*A*	2.5 ± 0.1	*B*	1.6 ± 0.1	*C*
Grainy	226.7	***	0.1	NS	3.4 ± 0.2	*A*	1.3 ± 0.1	*B*	0.7 ± 0.1	*C*
Soft	1167.4	***	1.5	NS	0.2 ± 0.0	*C*	3.4 ± 0.1	*B*	6.3 ± 0.1	*A*
After taste/After feel										
After taste	0.2	NS	7.9	***	3.3 ± 0.1		3.3 ± 0.1		3.3 ± 0.1	
Residue	210.4	***	1.8	NS	5.3 ± 0.1	*A*	3.6 ± 0.1	*B*	3.0 ± 0.1	*C*

	Bread Hardness		Mayonnaise		Bread without or with mayonnaises (mean±SE)				
Taste/Flavor	F		F		without	with FF-H	with LF-H	with LF-M	with LF-L
Bitter	0.9	NS	0.2	NS	0.6 ± 0.1	0.5 ± 0.1	0.6 ± 0.1	0.5 ± 0.1	0.5 ± 0.1
Salty	7.3	***	3.5	**	1.4 ± 0.1 *b*	1.8 ± 0.1 *a*	1.8 ± 0.1 *a*	1.8 ± 0.1 *a*	1.6 ± 0.1 *ab*
Sour	8.2	***	16.0	***	0.4 ± 0.1 *d*	0.9 ± 0.1 *c*	1.2 ± 0.1 *ab*	0.9 ± 0.1 *bc*	1.2 ± 0.1 *a*
Sweet	13.5	***	3.3	*	2.9 ± 0.2 *ab*	3.0 ± 0.2 *ab*	2.6 ± 0.2 *c*	2.5 ± 0.2 *c*	2.7 ± 0.2 *bc*
Baked	192.1	***	1.7	NS	3.8 ± 0.2	3.4 ± 0.2	3.2 ± 0.2	3.4 ± 0.2	3.3 ± 0.2
Yeast	73.3	***	5.6	***	3.6 ± 0.2 *a*	3.3 ± 0.2 *ab*	3.3 ± 0.2 *a*	2.9 ± 0.2 *c*	3.0 ± 0.2 *bc*
Mouthfeel/Texture									
Adhesive	77.1	***	1.8	NS	4.2 ± 0.2	4.0 ± 0.2	3.9 ± 0.2	3.9 ± 0.2	3.8 ± 0.2
Chewy	49.1	***	2.0	NS	3.6 ± 0.2	3.1 ± 0.2	3.3 ± 0.2	3.0 ± 0.2	3.1 ± 0.2
Cohesive	97.1	***	2.2	NS	2.6 ± 0.2	2.7 ± 0.2	2.8 ± 0.2	2.3 ± 0.2	2.4 ± 0.2
Crispy	3092.3	***	3.1	*	3.5 ± 0.3 *a*	3.1 ± 0.3 *b*	3.0 ± 0.3 *b*	3.1 ± 0.3 *b*	3.2 ± 0.3 *ab*
Dense	57.0	***	1.9	NS	2.5 ± 0.2	2.3 ± 0.2	2.5 ± 0.2	2.1 ± 0.2	2.1 ± 0.2
Dry	403.4	***	14.4	***	4.3 ± 0.2 *a*	3.1 ± 0.2 *b*	3.1 ± 0.2 *b*	3.0 ± 0.2 *b*	3.1 ± 0.2 *b*
Grainy	226.7	***	0.1	NS	1.9 ± 0.2	1.8 ± 0.2	1.8 ± 0.2	1.8 ± 0.2	1.8 ± 0.2
Soft	1167.4	***	1.5	NS	3.1 ± 0.2	3.4 ± 0.2	3.4 ± 0.2	3.3 ± 0.2	3.2 ± 0.2
After taste/After feel									
After taste	0.2	NS	7.9	***	3.6 ± 0.1 *a*	3.5 ± 0.1 *a*	3.3 ± 0.1 *ab*	3.2 ± 0.1 *b*	2.8 ± 0.1 *c*
Residue	210.4	***	1.8	NS	4.1 ± 0.1	4.0 ± 0.2	4.0 ± 0.2	3.9 ± 0.2	3.8 ± 0.1

F-values and p-values are derived from linear mixed models with hardness, mayonnaise and hardness:mayonnaise interaction as fixed effects, and subjects as random effects. No significant interaction effects were found and interaction effects are therefore not included in the table. Probabilities are presented as * ($p<0.05$), ** ($p<0.01$), and *** ($p<0.001$). Different capital letters indicate significant differences between bread hardness ($p<0.05$), *i.e.* HB, MB and SB. Different lower case letters indicate significant differences between bread without or with different mayonnaises ($p<0.05$), *i.e.* bread without mayonnaise and with FF-H, LF-H, LF-M and LF-L.

Table 6.5: Significance values and mean intensity scores (± standard error of the mean) of carrot attributes, obtained for carrot samples without and with mayonnaises by using RATA methodology and the carrot attribute list. The abbreviations are explained in Table 6.1 and the attributes are explained in Table 6.3.

	Carrot Hardness		Mayonnaise		Carrots varying in hardness (mean±SE)		
Taste/Flavor	**F**		**F**		**HC**	**MC**	**SC**
Bitter	37.9	***	14.5	***	1.8 ± 0.1 *A*	1.2 ± 0.1 *B*	0.9 ± 0.1 *C*
Salty	2.0	NS	1.6	NS	0.9 ± 0.1	1.0 ± 0.1	1.0 ± 0.1
Sour	0.1	NS	6.5	***	0.9 ± 0.1	1.0 ± 0.1	1.0 ± 0.1
Sweet	120.4	***	2.9	*	2.5 ± 0.1 *C*	3.8 ± 0.1 *B*	4.6 ± 0.1 *A*
Cooked	1387.0	***	1.6	NS	0.5 ± 0.1 *C*	4.8 ± 0.1 *B*	7.2 ± 0.1 *C*
Green	540.6	***	12.7	***	6.3 ± 0.1 *A*	2.6 ± 0.1 *B*	1.5 ± 0.1 *C*
Mouthfeel/Texture							
Adhesive	0.3	NS	0.2	NS	1.3 ± 0.1	1.2 ± 0.1	1.3 ± 0.1
Chewy	390.1	***	1.7	NS	5.1 ± 0.2 *A*	2.4 ± 0.1 *B*	1.2 ± 0.1 *C*
Cohesive	105.7	***	0.3	NS	3.5 ± 0.2 *A*	1.8 ± 0.1 *B*	1.4 ± 0.1 *C*
Crunchy	1868.2	***	1.3	NS	7.2 ± 0.1 *A*	2.4 ± 0.1 *B*	0.5 ± 0.1 *C*
Fibrous	340.4	***	2.1	NS	4.6 ± 0.1 *A*	2.3 ± 0.1 *B*	1.2 ± 0.1 *C*
Particles	303.9	***	1.8	NS	5.1 ± 0.1 *A*	2.8 ± 0.1 *B*	1.9 ± 0.1 *C*
Soft	1477.7	***	0.6	NS	0.3 ± 0.0 *C*	3.8 ± 0.1 *B*	7.0 ± 0.1 *A*
Watery	229.2	***	2.5	*	2.4 ± 0.1 *C*	4.0 ± 0.1 *B*	5.4 ± 0.1 *A*
After taste/After feel							
After taste	0.1	NS	23.7	***	3.3 ± 0.1	3.2 ± 0.1	3.2 ± 0.1
Residue	143.2	***	2.1	NS	4.0 ± 0.1 *A*	2.8 ± 0.1 *B*	2.4 ± 0.1 *C*

Table 6.5: Continued

	Carrot Hardness		Mayonnaise		Carrots without or with mayonnaises (mean±SE)									
Taste/Flavor	F		F		without		with FF-H		with LF-H		with LF-M		with LF-L	
Bitter	37.9	***	14.5	***	1.9 ± 0.2	*a*	1.1 ± 0.1	*b*	1.2 ± 0.1	*b*	1.2 ± 0.1	*b*	1.0 ± 0.1	*b*
Salty	2.0	NS	1.6	NS	0.8 ± 0.1		0.9 ± 0.1		1.1 ± 0.1		1.0 ± 0.1		1.1 ± 0.1	
Sour	0.1	NS	6.5	***	0.8 ± 0.1	*c*	0.8 ± 0.1	*c*	0.9 ± 0.1	*bc*	1.1 ± 0.1	*ab*	1.2 ± 0.1	*a*
Sweet	120.4	***	2.9	*	3.7 ± 0.2	*ab*	3.8 ± 0.2	*a*	3.7 ± 0.2	*ab*	3.3 ± 0.2	*c*	3.4 ± 0.2	*bc*
Cooked	1387.0	***	1.6	NS	4.2 ± 0.2		4.4 ± 0.2		4.1 ± 0.2		4.1 ± 0.2		4.0 ± 0.2	
Green	540.6	***	12.7	***	4.3 ± 0.2	*a*	3.0 ± 0.2	*c*	3.5 ± 0.2	*b*	3.3 ± 0.2	*bc*	3.3 ± 0.2	*bc*
Mouthfeel/Texture														
Adhesive	0.3	NS	0.2	NS	1.3 ± 0.1		1.2 ± 0.1		1.2 ± 0.1		1.2 ± 0.1		1.3 ± 0.1	
Chewy	390.1	***	1.7	NS	3.0 ± 0.2		2.7 ± 0.2		2.9 ± 0.2		2.8 ± 0.2		3.1 ± 0.2	
Cohesive	105.7	***	0.3	NS	2.3 ± 0.2		2.3 ± 0.2		2.1 ± 0.2		2.3 ± 0.2		2.3 ± 0.2	
Crunchy	1868.2	***	1.3	NS	3.5 ± 0.2		3.2 ± 0.2		3.5 ± 0.2		3.2 ± 0.2		3.3 ± 0.2	
Fibrous	340.4	***	2.1	NS	3.0 ± 0.2		2.5 ± 0.2		2.7 ± 0.2		2.7 ± 0.2		2.6 ± 0.2	
Particles	303.9	***	1.8	NS	3.5 ± 0.2		3.2 ± 0.2		3.1 ± 0.2		3.3 ± 0.2		3.2 ± 0.2	
Soft	1477.7	***	0.6	NS	3.7 ± 0.2		3.8 ± 0.2		3.6 ± 0.2		3.6 ± 0.2		3.7 ± 0.2	
Watery	229.2	***	2.5	*	3.7 ± 0.2	*b*	3.8 ± 0.2	*b*	3.9 ± 0.2	*ab*	4.0 ± 0.2	*ab*	4.2 ± 0.2	*a*
After taste/After feel														
After taste	0.1	NS	23.7	***	4.1 ± 0.2	*a*	3.0 ± 0.1	*b*	3.0 ± 0.1	*b*	3.1 ± 0.1	*b*	2.9 ± 0.1	*b*
Residue	143.2	***	2.1	NS	3.3 ± 0.2		3.0 ± 0.1		3.0 ± 0.1		3.0 ± 0.1		3.1 ± 0.1	

F-values and p-values are derived from linear mixed models with hardness, mayonnaise and hardness:mayonnaise interaction as fixed effects, and subjects as random effects. No significant interaction effects were found and interaction effects are therefore not included in the table.

Probabilities are presented as * ($p<0.05$), ** ($p<0.01$), and *** ($p<0.001$). Different capital letters indicate significant differences between carrot hardness ($p<0.05$), *i.e.* HC, MC and SC. Different lower case letters indicate significant differences between carrot without or with different mayonnaises ($p<0.05$), *i.e.* carrot without mayonnaise and with FF-H, LF-H, LF-M and LF-L.

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Table 6.6: Significance values and mean intensity scores (± standard error of the mean) of mayonnaise attributes, obtained for mayonnaise samples without and with carriers (bread, carrot) by using RATA methodology and the mayonnaise attribute list. The abbreviations are explained in Table 6.1 and the attributes are explained in Table 6.3.

	Mayonnaise		**Carrier**		**Mayonnaises (mean±SE)**							
Taste/Flavor	**F**		**F**		**FF-H**		**LF-H**		**LF-M**		**LF-L**	
Bitter	1.0	NS	5.5	***	0.8±0.1		1.0±0.1		0.9±0.1		0.9±0.1	
Salty	7.7	***	6.5	***	2.5±0.1	*B*	2.6±0.1	*B*	2.9±0.1	*A*	2.9±0.1	*A*
Sour	71.2	***	19.0	***	3.5±0.1	*D*	4.0±0.1	*C*	4.6±0.1	*B*	5.2±0.1	*A*
Sweet	4.5	**	2.7	*	2.7±0.1	*A*	2.4±0.1	*B*	2.4±0.1	*B*	2.3±0.1	*B*
Dairy	73.4	***	6.0	***	4.0±0.1	*A*	3.3±0.1	*B*	2.9±0.1	*C*	2.3±0.1	*D*
Mouthfeel/Texture												
Adhesive	19.6	***	3.5	**	2.3±0.1	*A*	2.0±0.1	*B*	1.7±0.1	*C*	1.5±0.1	*C*
Cohesive	68.4	***	7.2	***	2.4±0.1	*A*	2.2±0.1	*B*	1.5±0.1	*C*	1.0±0.1	*D*
Creamy	153.7	***	16.3	***	5.4±0.1	*A*	5.0±0.1	*B*	4.0±0.1	*C*	3.0±0.1	*D*
Salivating	10.6	***	2.6	*	2.7±0.1	*B*	2.8±0.1	*B*	3.1±0.1	*A*	3.3±0.1	*A*
Soft	49.2	***	3.6	**	3.6±0.1	*C*	3.7±0.1	*C*	4.4±0.1	*B*	5.1±0.1	*A*
Thick*	264.8	***	28.5	***	4.4±0.1	*A*	3.8±0.1	*B*	2.3±0.1	*C*	1.2±0.1	*D*
After taste/After feel												
After taste	2.7	*	39.0	***	3.8±0.1	*A*	3.5±0.1	*B*	3.6±0.1	*AB*	3.6±0.1	*AB*
Residual film	47.1	***	15.3	***	3.0±0.1	*A*	2.6±0.1	*B*	2.3±0.1	*C*	1.9±0.1	*D*

	Mayonnaise		Carrier		Mayonnaises without or with carriers (mean±SE)													
Taste/Flavor	F		F		without carrier		with HB		with MB		with SB		with HC		with MC		with SC	
Bitter	1.0	NS	5.5	***	1.1±0.1	*a*	0.9±0.1	*ab*	1.0±0.1	*ab*	1.2±0.1	*a*	0.8±0.1	*bc*	0.7±0.1	*c*	0.7±0.1	*c*
Salty	7.7	***	6.5	***	3.2±0.2	*a*	2.5±0.1	*c*	2.8±0.1	*b*	2.7±0.1	*bc*	2.6±0.1	*bc*	2.5±0.1	*b*	2.7±0.2	*bc*
Sour	71.2	***	19.0	***	5.2±0.1	*a*	4.2±0.2	*cd*	4.3±0.2	*c*	4.8±0.1	*b*	4.0±0.1	*de*	3.8±0.1	*e*	3.9±0.1	*de*
Sweet	4.5	**	2.7	*	2.2±0.1	*bc*	2.6±0.2	*a*	2.6±0.1	*a*	2.5±0.1	*ab*	2.1±0.1	*c*	2.5±0.1	*ab*	2.5±0.2	*ab*
Dairy	73.4	***	6.0	***	3.6±0.2	*a*	2.9±0.1	*c*	3.2±0.1	*b*	3.2±0.2	*b*	2.8±0.1	*c*	3.2±0.1	*b*	2.9±0.2	*bc*
Mouthfeel/Texture																		
Adhesive	19.6	***	3.5	**	2.3±0.2	*a*	1.9±0.1	*b*	2.0±0.1	*b*	1.9±0.1	*b*	1.7±0.1	*b*	1.8±0.1	*b*	1.8±0.1	*b*
Cohesive	68.4	***	7.2	***	2.2±0.2	*a*	1.4±0.1	*e*	1.7±0.1	*cd*	1.6±0.1	*de*	1.8±0.1	*bd*	1.9±0.1	*abc*	1.8±0.1	*bd*
Creamy	153.7	***	16.3	***	5.2±0.2	*a*	3.8±0.2	*d*	4.2±0.2	*c*	4.1±0.1	*cd*	4.1±0.2	*c*	4.3±0.2	*bc*	4.6±0.2	*b*
Salivating	10.6	***	2.6	*	3.3±0.2	*a*	2.9±0.2	*b*	2.9±0.2	*b*	3.0±0.1	*ab*	2.8±0.1	*b*	3.0±0.2	*b*	2.8±0.2	*b*
Soft	49.2	***	3.6	**	4.0±0.2	*bc*	4.3±0.2	*ab*	4.4±0.2	*a*	4.4±0.2	*a*	3.8±0.2	*c*	4.2±0.2	*ab*	4.4±0.2	*a*
Thick*	264.8	***	28.5	***	3.9±0.2	*a*	2.1±0.1	*d*	2.5±0.2	*c*	2.4±0.2	*cd*	3.3±0.2	*b*	3.2±0.2	*b*	3.1±0.2	*b*
After taste/After feel																		
After taste	2.7	*	39.0	***	4.9±0.1	*a*	3.4±0.1	*b*	3.6±0.1	*b*	3.6±0.1	*b*	2.9±0.1	*c*	3.4±0.1	*b*	3.6±0.1	*b*
Residual film	47.1	***	15.3	***	3.2±0.1	*a*	2.3±0.1	*bc*	2.5±0.1	*b*	2.3±0.1	*bc*	2.1±0.1	*c*	2.3±0.1	*bc*	2.3±0.1	*bc*

F-values and p-values are derived from linear mixed models with carrier, mayonnaise and carrier:mayonnaise interaction as fixed effects, and subjects as random effects. Interaction effects are not included in the table, since only one significant interaction effect was found (thick).

Probabilities are presented as * (p<0.05), ** (p<0.01), and *** (p<0.001). Different capital letters indicate significant differences between mayonnaises varying in fat content and viscosity (p<0.05), *i.e.* FF-H, LF-H, LF-M and LF-L. Different lower case letters indicate significant differences between mayonnaises without or with carrier (p<0.05), *i.e.* mayonnaises without or with bread (HB, MB and SB) or carrots (HC, MC and SC).
* Thick: A significant interaction effect was observed (F=3.8, ***).

Not only the presence of mayonnaise, but also specific mayonnaise properties had an effect on the perception of the bread and carrots. Low fat mayonnaises increased sourness in both bread and carrots and wateriness in carrots to a larger extent than full fat mayonnaises did, as well as a larger decrease in the sweetness of bread and carrots and after taste of bread. This may be an effect of the lower fat content directly, as a subsequent higher amount of aqueous phase is available to dilute sensory effects. More water addition leads to a decrease in tastant concentration such as sweetness and after taste, and at the same time to an increased watery perception. Although the effect of fat content on the dilution effect may be plausible, we could not confirm this in our study, as slight differences in the flavor profiles of the different mayonnaises were present. According to the sensory profiles of the mayonnaises (Table 6.6), the low fat mayonnaise was perceived as slightly more sour than the full fat mayonnaise. This higher sourness in low fat mayonnaises might have overpowered the taste and flavor perception of the relatively plain bread and carrots to a larger extent than full fat mayonnaise did.

In the case of bread, we expected that both the fat content and the viscosity of mayonnaises would have an influence on the texture properties of bread. We expected a larger decrease in crispiness and dryness of bread after the addition of both low fat mayonnaises due to the larger fraction of the hydrophilic continuous phase and low viscosity mayonnaises due to the higher ability to be absorbed by the bread. However, no effect of the fat content nor the viscosity profile on bread crispiness and dryness was observed. This can be explained by the fact that RATA is a static sensory method, and consumers scored their perceived intensity of an attribute after one bite. This procedure does not take into account specific changes in intensity over the chewing sequence. Thus, even though low fat and low viscosity mayonnaises might be absorbed faster by the bread, similar crispness and dryness intensity scores are obtained when using RATA methodology. Therefore, more knowledge on the effect of fat content and viscosity of mayonnaise on crispness and dryness perception of bread throughout consumption is necessary to gain insight in the exact role of these properties of mayonnaise on bread perception.

6.4.3 Effect of bread and carrot addition on sensory properties of mayonnaises

All sensory intensity scores of mayonnaises were clearly impacted by the presence of bread or carrots (Table 6.6). The addition of bread or carrots decreased several taste and flavor intensities of mayonnaises including salty taste, sour taste and dairy flavor. This finding is consistent with the work of other studies (Meinert *et al.* 2011; Paulsen *et al.* 2012; Cherdchu & Chambers 2014) and can be explained by a dilution effect (Kroll & Pilgrim 1961). In addition, the intensity scores of multiple texture and mouthfeel attributes of mayonnaises were decreased when mayonnaises were combined with bread or carrot including adhesive, cohesive, creamy, salivating, thick and residual film. This indicates that the texture of solid foods prevails over the sensory perception of the mayonnaises. This finding corroborates the results of Cherdchu and Chambers (2014), who observed that solid carriers had a larger impact on the flavor profile of soy sauces than liquid carriers. These results imply that addition of solid foods has a larger effect on the sensory profile of liquid foods than then the addition of liquid foods to solid foods.

An effect of the type of carrier matrix can be seen from the fact that bread and carrots affected the intensity scores of certain mayonnaise attributes in different ways (Table 6.6). The presence of carrots but not bread decreased the bitterness intensity scores of mayonnaise, whereas the presence of bread but not carrots increased the sweetness intensity scores of mayonnaise. Furthermore, carrots decreased the sourness intensity scores of mayonnaises to a larger extent than bread, whereas bread decreased the thickness intensity scores of mayonnaises to a larger extent than carrots. Although this might be explained by a difference in mass between bread and carrots (2 g of bread vs. 4 g of carrot), we hypothesize that the perception of certain mayonnaise characteristics depends on other properties of food matrix it is combined with (*e.g.* mayonnaise absorption or sensory interactions). Bread and carrots are assumed to differ in the way the bolus is formed (*i.e.* bread takes up (part of) the mayonnaise and carrots do not), which can explain the differences in mayonnaise perception after addition of these two carriers. Different taste, flavor and texture interactions might have occurred between mayonnaise and bread and between mayonnaise and carrots leading to the differences in mayonnaise perception identified. Such aroma, taste and texture interactions have been observed in liquids (Tournier *et al.* 2007), sequential tastings of foods and beverages (Bastian *et al.* 2010; Donadini *et al.* 2013; Donadini & Fumi 2014; Donadini *et al.* 2015; Galmarini *et al.* 2015; Galmarini *et al.* 2017) or sequential tastings of different foods (Aaslyng & Frost 2008; Aaslyng & Frost 2010), and such sensory interactions become even more complex when two foods are combined and eaten within one bite.

The effect of food hardness is evident from the fact that a larger decrease in the intensity scores of certain mayonnaise characteristics was found for harder carriers than for softer carriers (Table 6.6). Harder bread decreased the sourness and dairy flavor intensity scores of mayonnaises to a larger extent than softer bread, and harder carrots decreased the creaminess and after taste intensity scores of mayonnaises to a larger extent than softer carrots did. Previous studies have shown that in the case of single foods, the degree of hardness also has an effect on sensory perception. For example, for gels and candies, an increase in hardness decreased perception of the flavor intensities (Boland *et al.* 2006; Saint-Eve *et al.* 2011). This research shows the hardness of one food can also influence the perceived sensory intensity of a second food when consumed within one bite. This effect can be ascribed to three different mechanisms. Firstly, harder solid foods might distract more attention from the more liquid-like mayonnaises and thereby decreasing its intensity scores to a larger extent (*i.e.* cognitive mechanism). Secondly, harder solid foods influence the oral processing behavior during breakdown including mastication time, number of chews and salivation. This could have led to changes in tastant or flavor release (*i.e.* physiological effect). Thirdly, the harder bread was drier with more absorption capacity, and this consequently might have led to more mayonnaise absorption and decreased intensity scores (*i.e.* physical effect).

6.4.4 Sensitivity to discriminate between different breads, carrots or mayonnaises when consumed as combinations

The sensory profiles of bread or carrots varying in hardness were perceived different from each other when assessed alone and when combined with any of the mayonnaises, regardless of the fat content or viscosity of the mayonnaises (Figure 6.4). Thus, even when breads and carrots were combined with various mayonnaises, the sensory profiles of breads and carrots varying in hardness did not overlap and subjects were able to differentiate between bread and carrots. The reason why breads and carrots differing in hardness were still perceived as different when they were combined with a condiment is most likely due to the large differences in the hardness of the breads (compression forces varied between breads by a factor of at least 3x (Table 6.1)) and the carrots (fracture stresses varied between carrots by a factor of 2x (Table 6.1)). It might therefore be interesting to reduce the variation in hardness to see if and/or when differences in perception between solid foods disappear when combined with a condiment. We hypothesize that a minimal difference in hardness needs to be present to be perceived as a difference in hardness (Santagiuliana *et al.* 2018b), and that this minimal difference will become larger when a carrier is combined with a condiment.

Although differences in the bread and carrot properties were perceived when combined with mayonnaise, differences in mayonnaise properties were not always perceived when combined with bread or carrots. Sensory profiles of mayonnaises with varying properties (fat content, viscosity) were perceived different from each other when consumed by itself, as the differences in mayonnaise properties were considerably large (*i.e.* FF-H, LF-M, LF-L) (Figure 6.5). When mayonnaises were combined with carriers, bread and carrots affected the sensitivity to discriminate between these mayonnaises differently. In the case of bread (Figure 6.5A), regardless of the hardness of the bread, the sensory profiles of LF-M and LF-L mayonnaise started overlapping. Thus, LF-M and LF-L mayonnaises were different when assessed alone, but less when assessed together with bread. In the case of carrots (Figure 6.5B), the sensitivity to discriminate between FF-H, LF-M and LF-L mayonnaises was not affected by the presence of carrots regardless of the hardness of the carrots. Discrimination between mayonnaise characteristics was therefore not affected by carrots. In addition, although the sensory profiles of FF-H and LF-H mayonnaises overlapped when assessed alone, they did not overlap when assessed with soft and medium carrots. These results show that the perception of consumers towards certain products depends on the foods they are combined with and affects sensory sensitivity. Differences between mayonnaises with different formulations can be perceived when assessed on its own, but these differences may disappear or enhance when assessed together with another food. These findings have important implications for product development of foods. When designing products for specific carrier-condiment combinations, sensory assessment of such combinations must be considered since it could provide additional information about the sensory interactions between the foods present.

6.4.5 Sensory assessment of carrier foods with condiments: TDS and RATA methodologies

Naive consumers could discriminate between carrier-mayonnaise combinations with varying properties by using both TDS and RATA. TDS was an insightful method to reveal the time periods of consumption and the sensory attributes during which the carrier or the condiment dominated sensory perception of the carrier-mayonnaise combinations. This dynamic information could not be obtained using RATA. RATA showed that naive consumers were able to evaluate the sensory attributes of the carrier or the mayonnaise separately even when these two food items were consumed together. Consequently, RATA provided insights on the effect of carriers on the sensory profile of condiments, and vice versa. RATA was therefore a powerful methodology to reveal and quantitatively describe changes in sensitivity to discriminate between different carriers and mayonnaises when they were consumed in combination. This information could not be obtained using TDS. Certainly, this was easier for evaluation of texture than for taste or flavor attributes for the untrained subjects. Consumers were not always able to differentiate between tastes and flavors coming from the carrier or the condiment. Training of subjects might allow to achieve better discrimination between tastes and flavors coming from the carrier or condiment. This should be taken into consideration when one wants to use this method to investigate other carrier-condiment pairs. Furthermore, we assessed attributes that were specific to the carrier or the condiment. The choice of having three different attribute lists (bread, carrot and mayonnaise) might have prevented finding carrier:mayonnaise interaction effects. In addition, we did not include attributes related to sensations that occur only when carriers and condiments are combined and that are absent in single carriers or single condiments. Adding additional, more holistic, attributes might be considered in future research on sensory evaluation of composite foods.

6.5 CONCLUSION

This study demonstrates that perception of carrier foods and condiments changes when they are combined with each other. Both TDS and RATA can be used to assess carriers with condiments, and combining both methodologies provides complementary information on the sensory perception of such composite foods with complex sensory characteristics. Carriers (bread, carrot) to which a condiment (mayonnaise) was added resulted in increased dynamic sensory complexity throughout consumption since sensations related to both carrier and mayonnaise characteristics were perceived. In addition, the dynamic sensory profiles of the carrier-mayonnaise combinations were dominated by the texture of the solid carriers, especially in the beginning and end of consumption, whereas additional sensations related to the mayonnaises appeared in the middle stage of consumption. The dominant role of the solid carriers can be attributed to the food breakdown and an increase in lubrication before swallowing, leading to continuous changes in food structure and texture perception, which thereby drives dynamic sensory perception of carriers with condiments. Furthermore, sensations related to mayonnaises tended to appear in an earlier stage of consumption when they were combined with softer carriers than with harder carriers. Also the static sensory profiles of carriers and condiments change when they are consumed together in one bite. The addition of solid (bread, carrot) to liquid-like (mayonnaise) foods was shown to have a larger effect on the sensory profiles than the addition of liquid-like to solid foods. Harder bread or carrots affected some sensory intensity scores of mayonnaises to a larger extent than softer ones, and little effects of fat content and viscosity of mayonnaises on the sensory profiles of bread and carrots were observed. The sensitivity to discriminate between bread or carrots varying in hardness was not influenced by the presence of mayonnaise, which can be attributed to the large differences in hardness between samples. Similarly, when the differences between the mayonnaises were large (full fat/high viscosity vs. low fat/low viscosity), the sensitivity to discriminate between mayonnaises was not influenced by the presence of bread nor carrots. However, when the differences between the mayonnaises were smaller (low fat/medium viscosity vs. low fat/low viscosity), the addition of bread decreased the sensitivity to discriminate between mayonnaises and the sensory profiles started to overlap.

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Cognitive factors drive flavor perception of condiments more than in-nose flavor release when consumed as composite foods

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ABSTRACT

Background and objective: When condiments such as mayonnaise are combined with carrier foods such as bread and potatoes, aroma release and perception is complex and changes dynamically throughout consumption. This study investigated the effect of condiment and carrier properties on *in vivo* aroma release and perception.

Methods: Mayonnaises varying in fat content (high/low) and viscosity (high/low) were spiked with two lemon aroma compounds (limonene, citral). Carriers differing in moisture absorption capacity (bread, potato) and hardness (hard/soft) were combined with mayonnaises. In-nose aroma release and perception of lemon intensity were assessed dynamically and simultaneously.

Results: Mayonnaise properties affected aroma release and perception congruently; higher viscosity decreased both aroma release and perception, and higher fat content increased aroma release and perception. When mayonnaises were combined with carriers, aroma release and perception were no longer congruent. Addition of carriers to mayonnaises increased aroma release and decreased perception of aroma intensity.

Conclusions: We conclude that cognitive effects are likely to modulate aroma perception of composite foods.

Key words: nose-space aroma release, sensory perception, mayonnaise, carrier foods

7.1 INTRODUCTION

Condiments such as spreads, dressings or sauces are a key ingredients in many different culinary traditions, and are often used to enhance the flavor of a carrier food or dish (Sikora *et al.* 2008). Condiment addition such as mayonnaises to carrier foods such as bread, crackers and carrots has recently been shown to increase eating rate (van Eck *et al.* 2019b; van Eck *et al.* 2019c), potentially leading to an increase in energy intake. In the last decades, food industry put considerable efforts into development of fat reduced condiments to provide consumers with less energy dense foods. These modifications imply a significant change in flavor release and perception. Food flavor profiles are partly due to perception of aroma compounds, which are released from the food matrix and reach the olfactory receptors located in the human nasal cavity (retronasal pathway) (Robert-Hazotte *et al.* 2019). In addition, some aroma compounds remain in the breath air when food is swallowed, and contribute to aroma perception after swallowing (Mayr *et al.* 2003). Aroma release is a rather complex process, which is influenced by food composition, food structure and dynamic changes thereof during oral processing (Tournier *et al.* 2007; Buettner & Beauchamp 2010; Poinot *et al.* 2013).

Fat reduction impacts aroma release and perception in a direct and/or indirect way. Fat or other hydrophobic phases present in the food matrix can bind hydrophobic aroma compounds and reduce their vapor pressure (physicochemical effect). Thus, increasing fat content in the food matrix reduces the release of hydrophobic aroma compounds both in the vapor phase and during consumption (Wendin *et al.* 1999; Carrapiso 2007; González-Tomás *et al.* 2007; Linforth *et al.* 2010; Frank *et al.* 2011). Vice versa, in the case of fat reduction, less hydrophobic aroma compounds are retained in the food matrix leading to higher aroma release and intensity perception (Miettinen *et al.* 2002; Miettinen *et al.* 2003; Arancibia *et al.* 2011). Changing fat content also largely impacts rheological properties which can affect aroma release and perception in an indirect way. Higher volume fraction of dispersed fat increases the viscosity, which reduces aroma release by slowing down diffusion of aroma compounds (Cook *et al.* 2003a).

Hydrocolloids are commonly added to fat-reduced foods to maintain desired rheological properties. Several studies demonstrated that an increase in viscosity through addition of hydrocolloids reduces aroma release and perception (Baines & Morris 1987; Roberts *et al.* 1996; Baek *et al.* 1999; Cook *et al.* 2003b; Malone *et al.* 2003; Bylaite *et al.* 2005; Boland *et al.* 2006; Xu *et al.* 2017). This effect has been shown for various liquid and semi-solid foods including milk-like foods and yogurts (Kora *et al.* 2003; Saint-Eve *et al.* 2006; Bult *et al.* 2007). Reduction of aroma release and perception caused by an increase in food viscosity due to hydrocolloid addition likely has a physicochemical origin. Hydrocolloids may change release kinetics, reduce aroma release and consequently perception by slowing down aroma diffusion or by binding aroma compounds through different types of attractive interactions.

Several studies reported an effect of food properties (composition, rheological properties) on aroma release explaining subsequent sensory perception (Baek *et al.* 1999; Boland *et al.* 2006; Saint-Eve *et al.* 2006), while other studies did not find congruencies between *in vivo* aroma release and subsequent sensory perception (Hollowood *et al.* 2002; Weel *et al.* 2002; Lethuaut *et al.* 2004; Cook *et al.* 2005). For example, in case of flavored gels, increasing gel hardness did not affect aroma release but did decrease perceived aroma intensity (Weel *et al.* 2002). In this context, it has been suggested that cognitive interactions due to cross-modal associations between food texture and aroma perception play a role (Tournier *et al.* 2007; Tournier *et al.* 2009). It is also plausible that changes in oral processing behavior (e.g. dilution with saliva, structure breakdown by chewing or influence of mouth temperature) induce changes in aroma release and potentially perception (Buettner & Beauchamp 2010).

Flavored condiments such as mayonnaises are usually consumed together with rather plain carrier foods such as potatoes and bread, and dynamic interactions between condiments and carriers can influence aroma release and perception. Addition of carrier foods to condiments is generally known to decrease overall perceived flavor intensity of condiments (Meinert *et al.* 2011; Paulsen *et al.* 2012; Cherdchu & Chambers 2014; van Eck *et al.* 2019a). Addition of bread or carrots to mayonnaises has been shown to reduce perceived intensities of several mayonnaise-related flavor attributes (*e.g.* sour, dairy, creamy) (van Eck *et al.* 2019a). However, the mechanisms underlying the reduction in flavor perception upon carrier addition are not known. To the best of our knowledge, it has not been investigated whether the decreased flavor perception is due to physiochemical interactions leading to a lower delivery of aroma compounds into the nasal cavity or whether other mechanisms such as different eating behaviors or cognitive interactions due to cross-modal associations come into play. To summarize, it is not well understood how carrier addition changes flavor release and perception of condiments. Providing insights into in-nose aroma release of condiment-carrier pairings can lead to a better understanding of factors contributing to sensory perception and intake of complex food combinations.

As shown by our earlier work, not only the presence of a carrier food, but also the type of carrier effects sensory properties of condiments. Crackers decreased overall perceived intensity of a variety of condiments (firm cheese, cheese spread, mayonnaise) to a larger extent than bread (van Eck *et al.* 2019b). Harder carrier foods decreased the perception of mayonnaise to a larger extent than softer carriers (van Eck *et al.* 2019a). Besides sensory attribute intensity, also dynamic perception of sensory properties of condiments were affected by addition of carriers. Mayonnaise-related attributes (*i.e.* creamy, sweet, sour) were perceived as dominant sensations earlier during consumption when combined with softer carriers than when combined with harder carriers. These results show that the type and mechanical properties of carrier foods affect static and dynamic sensory properties of food-condiment combinations. The mechanisms underlying the reduction in flavor perception and the changes in dynamics of flavor perception upon carrier addition are not known. We hypothesize that aroma perception of condiment-carrier food combinations is driven by physicochemical effects on aroma release (condiment aroma release is decreased as condiment aroma compounds bind to the carrier). Cognitive effects are believed to play a role in sensory perception of condiment-carrier combinations (cognitive distraction from flavor perception of condiment by carrier addition and cross-modal interactions). Studying in-nose aroma release coupled to dynamic sensory analysis might therefore help to unravel the mechanisms underlying this phenomenon.

This study aimed to get a better insight into the relation between in-nose aroma release and dynamic aroma perception of condiments (mayonnaise) when consumed with and without carriers (bread, potatoes). Mayonnaise properties were varied by varying fat content (high, low) and viscosity (high, low) to understand the effect of different physicochemical properties on in-nose aroma release and perception. Carrier foods with different moisture absorption capacity (bread, potato) varying in texture (soft, hard) were tested to investigate the role of carrier type on aroma release and perception of condiments (Figure 7.1).

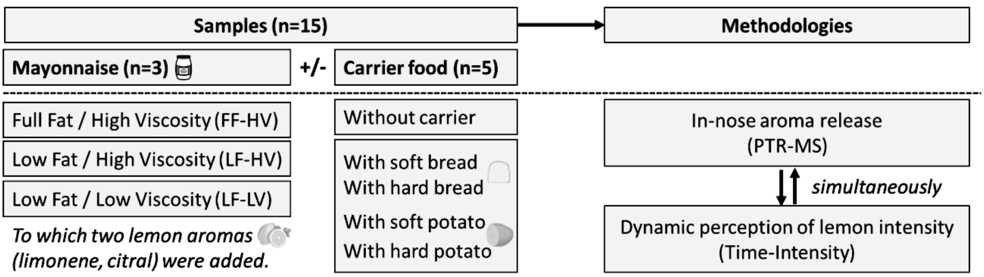

Figure 7.1: Experimental design outlining the approach. Mayonnaises varying in fat content (high/low) and viscosity (high/low) were tested without carrier food (n=3) and together with carrier foods differing in moisture absorption capacity (bread, potato) and hardness (hard/soft). Mayonnaises were spiked with two lemon aroma compounds (limonene, citral), which allowed to characterize in-nose aroma release and dynamic lemon intensity perception.

7.2 MATERIALS AND METHODS

7.2.1 Samples

Three different mayonnaises varying in fat content and viscosity were prepared, namely full fat/high viscosity (FF-HV; 69% w/w oil; Calvé De echte, Unilever, The Netherlands), low fat/high viscosity (LF-HV), and low fat/low viscosity (LF-LV) mayonnaises. For the low fat mayonnaises, a 2.5 or 1.0% xanthan in water solution (E415, Pit&Pit bvba, Belgium) was gradually spooned into the FF-HV mayonnaise following a 1.6:1.0 weight ratio to create the LF-HV or LF-LV mayonnaises (26.5% w/w oil) with similar oil droplet sizes, respectively. Two lemon aroma compounds varying in hydrophobicity, citral (*Mw* = 152 g/mol , *log P* = 2.76, 1 mg/g mayonnaise) and limonene (*Mw* = 136 g/mol , *log P* = 4.2, 1 mg/g mayonnaise), were gently mixed into the mayonnaises using a spatula. The addition of these compounds made the mayonnaise easier to track during the Proton Transfer Reaction Mass Spectrometry (PTR-MS) analysis and easier to be perceived by the panelists. The two compounds were chosen based on their aroma, their different physical/chemical properties and their masses after some preliminary measurements on volatile organic compounds (VOCs) emissions on both mayonnaises and carriers to verify interferences. Mayonnaises were served at a weight of 2 g.

Mayonnaises were assessed alone and in combination with different carrier foods. Two commercial carrier foods were used, namely bread (Plaisir de mie toastbrood, Jacquet®, France) and potatoes (Waxy potatoes, Albert Heijn, The Netherlands). Mayonnaise on bread represents a simplified model food for sandwiches and mayonnaise on potato represents a simplified model food for salads. Bread cubes without crust (35x35x8 mm) were served fresh and oven-dried for 40 min at 100ºC (Venti-line, VWR®) to obtain two bread samples with varying properties. Peeled potato cubes (30x12x12 mm) were cooked sous-vide at 90ºC for 15 and 45 min to obtain two potato samples with varying properties. Carrier-mayonnaise combinations were prepared just before serving in order to minimize moisture transfer of the mayonnaises into the carriers before consumption.

Table 7.1 presents an overview of the composition and product properties of the mayonnaises (fat content, viscosity, oil droplet size) and the carrier foods (firmness, water activity). Mayonnaises properties were measured each morning before data collection (n=10 days of data collection) and carrier properties were measured for each new preparation batch (n=4 batches) to ensure that samples were stable over the data collection period. To determine the viscosity of the mayonnaises, mayonnaises were sheared at shear rates ranging from 1 s-1 to 1000 s-1 after a resting period of 5 minutes using a rheometer (MCR 301 Rheometer, Anton Paar Benelux BVBA, Belgium) equipped with an Inset I-PP50/SS plate and a CP50-1 cone. The oil droplet size of the mayonnaises ($D_{3,2}$) was measured by light scattering (Mastersizer 2000, Malvern Instruments) in triplicate using the refractive index of sunflower oil (1.469). To determine the firmness of the carrier foods, uniaxial compression tests were performed with a Texture Analyzer (TA.XT Plus, Stable Micro Systems, United Kingdom) fitted with a 50 kg load cell, a cylindrical plate with a diameter of 100 mm and a constant speed of 1 mm/s. Bread samples were compressed until 20% strain, and the mean force needed to compress the bread samples was calculated. Potato samples were compressed until 50% stain and the mean fracture stress of the potatoes was calculated. The water activity of the carrier foods was measured using a LabMaster aw (Novasina®).

Table 7.1: Product properties of mayonnaises varying in fat content and viscosity (A) and the carrier foods bread (B) and potato (C) varying in preparation methods (mean±SD).

(A) Mayonnaise properties	Full fat / High viscosity (FF-HV)	Low fat / High viscosity (LF-HV)	Low fat / Low viscosity (LF-LV)
Fat content (w/w %)	70	27	27
Viscosity at 1 s^{-1} (Pa·s)	84±19	73±12	11±3
Viscosity at 10 s^{-1} (Pa·s)	13±3	10±1	2±1
Viscosity at 100 s^{-1} (Pa·s)	2.1±0.4	1.3±0.2	0.3±0.1
$D_{3,2}$ (µm)	10.9±2	7.9±1	7.1±1

(B) Bread properties	Fresh bread	Dried bread
Compression force (N)	5±2	100±38
A_w	0.92±0.02	0.20±0.16
(C) Potato properties	**Soft potato**	**Semi-hard potato**
Fracture stress	40±17	198±132
A_w	0.98±0.01	0.99±0.00

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7.2.2 Subjects

A group of 14 Caucasian, European females (23±3 years,) participated in the study. An homogeneous group of subjects was selected based on their mechanically stimulated saliva flow rate (1.4±0.6 g/min, mean±SD)(Engelen *et al.* 2005), size of the oral cavity (73.5±10.4 g water, mean±SD)(Alsanei & Chen 2014) and natural eating time of the samples (16±5 s, mean±SD)(van Eck *et al.* 2019b), which were assessed during one selection session of one hour based on procedures described elsewhere. In addition, they had non-smoking habits (self-reported), good dental health (self-reported), and were consumers of mayonnaise, bread and potato on a regular basis. All subjects gave written informed consent, completed the study and received financial compensation for participation.

7.2.3 Chewing protocol

Subjects were instructed to follow a chewing protocol to minimize the influence of individual differences in mastication behavior on aroma release and perception throughout consumption. Subjects were instructed to consume each sample within one bite, and to swallow after 20 seconds of consumption (timer was shown on the screen). In the case of mayonnaises alone, they were instructed to swirl samples in their mouth. In the case of mayonnaise-carrier combinations, they were instructed to chew the sample with a frequency of 1 chew/s (*i.e.* approximately 20 chews) using a metronome and the timer on the screen. Furthermore, subjects were asked to raise their hand each time they swallowed, which was recorded by the researcher. In addition, they were asked to keep their mouth closed during all the evaluations.

7.2.4 Nose-space analysis, data extraction and peak selection

In vivo aroma release was measured using a commercial PTR-MS instrument (Ionicon Analytik GmbH, Innsbruck, Austria) equipped with a time of flight and a quadrupole ion guide (PTR-QiTOF). H3O+ was used as precursor ion, and the ionization conditions were the following: 1000 V drift voltage, 60.0 °C drift temperature, 3.8 mbar drift pressure, resulting in an E/N ratio of 133 Td. Acquisition was set to 1 spectrum per second. Sampling was carried out via a heated (95 °C) inlet tube with an inlet flow of 45.02 sccm. The mass resolution (m/Δm) was at least 5000.

The nose-space experimental set up was adapted from previous PTR-MS in-nose studies (Heenan *et al.* 2012; Charles *et al.* 2015; Pedrotti *et al.* 2019). For each measurement, laboratory air was sampled for 20 s. After that, subjects were asked to insert two teflon tubes (diameter: 6.8 mm, length: 6.4 cm, connected to the heated inlet tubes) in the nose. They were asked to breath normally through their nose, and subjects' breath was sampled for 60 s. Then they consumed the samples for 20 s. After swallowing the sample, subjects kept on breathing for 90 s. This led to a total sampling time of 190 s (Figure 7.2). Samples were assessed in triplicate by each subject.

PTR-MS data were treated with TOFO office software (Department of Food Quality and Nutrition, Edmund Mach Foundation) as described in Cappellin *et al.* (2011). A total of 247 mass peaks were extracted from 20 m/z to 250 m/z and in-nose concentration was calculated. From that, 73 peaks were selected for the further analysis based on pilot experiment report, literatures and the high concentration of the release curve for the relevant aroma compounds of citral, limonene, mayonnaise, food carriers and the exhaled gases from participants. In the work only mass peaks corresponding to the two lemon aroma compound are considered: m/z 138.139 and 153.131 tentatively identified as the limonene isotope ($^{13}CC_9H_{16}H^+$) and citral ($C_{10}H_{16}OH^+$) were chosen as representative examples, respectively. The m/z 81.070 ($C_6H_9^+$) and 135.119 ($C_{10}H_{15}^+$) were chosen as main fragments of limonene and citral.

For each mass peaks a release curve was obtained by plotting between peak concentration (ppbV) and time (seconds). Each release curve, was divided in 4-time separate windows: lab air session (1-20 s), breathing (21-80 s), mastication session (81 s to first swallowing point) and post-swallowing session (first swallowing point until 195 s). Each part of the curve was averaged for all the panel and superposed to create an average release curve for each sample. For comparing the different mayonnaises aroma release and the food carrier interactions, the baseline (signal before the sample was ingested) was then subtracted and three main parameters were extracted from each individual release curve: the area under the curve (AUC_R), the maximum concentration (I_{max}_R) and the time to reach the maximum concentration (T_{max}_R).

7.2.5 Time-intensity (TI) sensory methodology

Dynamic lemon aroma intensity of mayonnaises was determined using the time-intensity (TI) methodology (Cliff & Heymann 1993). Subjects were instructed to place the sample in the mouth and simultaneously click the start button. Then, they continuously scored the lemon intensity over time by moving the cursor horizontally on a 100 mm unstructured line scale anchored from not at all to very (Eye Question software, version 4.11.19). The total duration of the evaluation was set at 110 s, meaning that subjects evaluated lemon intensity during chewing (approximately 20 s) and after the sample had been swallowed (approximately 90 s). Intensity scores were recorded with an interval time of 500 ms. From the time-intensity profiling, the total area under the curve (AUC_S), the maximum perceived intensity (I_{max}_S) and the time to maximum intensity (T_{max}_S) were obtained (Figure 7.2). In the present study, Liu&MacFie standardization was applied to correct for individual signature curves (Liu & MacFie 1990; Lawless & Heymann 2010).

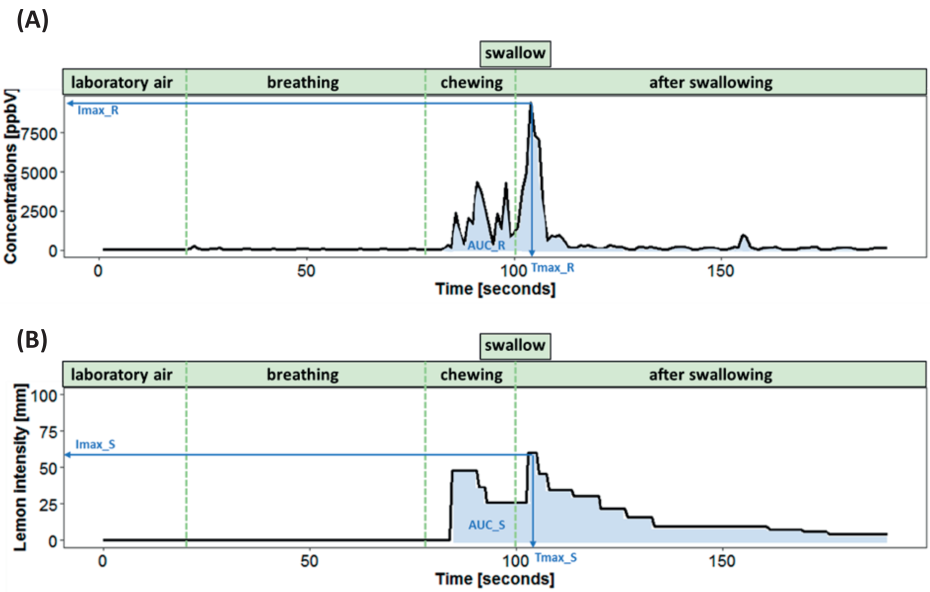

Figure 7.2: Example of an aroma release curve (A) and a time intensity curve (B), in which the different sampling procedures and panel instructions are indicated in green. Parameters obtained from the curves are shown in blue.

7.2.6 Experimental approach

Subjects participated in 10 sessions over a time period of one month. Subjects were firstly trained over four sessions of 1 hour, after which dynamic aroma perception and *in vivo* aroma release were determined simultaneously by using TI and PTR-MS during the subsequent six sessions of maximum 1.5 hours.

During the four training sessions, subjects were acquainted with the chewing protocol (training 1 and 2) and the TI methodology (training 3 and 4). The first training started with an introduction to the chewing protocol (section 7.2.3), after which the subjects practiced the protocol. During the second training session, subjects were familiarized with the nose tubes used to connect the subjects' nasal cavity with the PTR-MS. During this session, subjects continued practicing the chewing protocol while having the nose tubes in their nose. The third session was used to introduce TI methodology to the subjects, after which they practiced with the tasting protocol, nose tubes and TI methodology using mayonnaises spiked with lemon aroma. The fourth session was a pilot experiment, during which they practiced with the tasting protocol, nose tube and TI methodology for all samples included in the present study.

During the six data collection sessions, subjects were requested to not eat, drink, or brush their teeth two hours before the experiment and to not wear perfume or lotion. All samples were assessed following a 3x5 design: three mayonnaises with five carrier conditions (without carrier, with fresh bread, with oven-dried bread, with shortly cooked potato, with long cooked potato). Samples were assessed in triplicate leading to a total of 45 nose-space measurements and sensory analyses for each subject. Each replicate was assessed over two sessions. Within each replicate, samples were presented in a random order following a completely randomized design. Samples were presented with three digit codes, and served on a spoon to facilitate easy intake. Between each sample, subjects cleansed their palate for at least 6 minutes using cold water, hot water and tongue scrapers to aid the removal of oil from their tongue. No other palate cleansers were used, since they might affect the volatile release of follow-up samples.

7.2.7 Statistical data analyses

Results were reported as mean values with standard error (n=14 subjects, in triplicate). Outliers (Z-score>3.29 or Z-score<-3.29) were removed from the data. To investigate the effect of mayonnaise properties, linear mixed models were performed on a subset of the data including the data of the single mayonnaises (*i.e.* without carriers) only. For this analysis, mayonnaise was set as fixed effect and subject, replicate, serving order and session were set as random effects using Lmer package (Kuznetsova *et al.* 2016) To investigate the interplay between mayonnaise properties and carrier properties, linear mixed models were performed with mayonnaise, carrier and mayonnaise:carrier interaction as fixed effects and subject, replicate, serving order and session as random effects. This analysis was performed for bread and potato carrier separately. In addition, Multiple Factor Analysis (MFA) was performed on the selected mass peaks from PTR-MS analysis and on the Time-Intensity data by using FactoMineR package (Lê *et al.* 2008). Only the AUC was used in this case and data were scaled to unit variance before performing the analysis. (Lê *et al.* 2008)R language (RStudio, version 1.0.143) was used to perform all statistical tests. Significance level of $p<0.05$ was chosen.

7.3 RESULTS

7.3.1 In-nose aroma release and dynamic lemon perception of mayonnaises without carriers: effect of viscosity and fat content

Dynamic aroma release and dynamic lemon intensity profiles of single mayonnaises (without carrier food) are shown in Figure 7.3. Table 7.2 provides a summary of all aroma release (AUC_R, I_{max}_R and T_{max}_R) and perception (AUC_S, I_{max}_S and T_{max}_S) parameters. As can be seen from Figure 7.3A – 7.3D, limonene and citral display different release profiles. While limonene was released fast, resulting in a sharp peak (Figure 7.3A, 7.3B), citral was released slowly throughout consumption (later T_{max}_R, see Table 7.2) resulting in a broader peak (Figure 7.3C, 7.3D). Although both compounds are hydrophobic, citral has a higher boiling temperature and lower vapor pressure due to its higher molecular weight and its molecular structure, resulting in a lower volatility than limonene.

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Mayonnaise viscosity (LF-HV vs. LF-LV) clearly affected in-nose aroma concentrations (Figure 7.3A, 7.3C) and dynamic lemon intensity perception (Figure 7.3E). *In vivo* limonene release, *in vivo* citral release and lemon intensity perception decreased with increasing mayonnaise viscosity. For example, in case of limonene (m/z 138.139), AUC_R decreased by 69% and I_{max}_R decreased by 74% with increasing viscosity (Table 7.2). Congruently, with respect to sensory perception, AUC_S decreased by 31% and I_{max}_S decreased by 23%. The times to reach the maximum concentration and intensity (T_{max}_R, T_{max}_S) were not significantly affected by mayonnaise viscosity.

Mayonnaise fat content (FF-HV vs. LF-HV) also affected in-nose aroma concentrations (Figure 7.3B, 7.3D) and dynamic lemon intensity perception (Figure 7.3F). *In vivo* limonene release, *in vivo* citral release and lemon intensity perception decreased by fat reduction from 70 to 27 wt%. For example, AUC_R of limonene (m/z 138.139) decreased by 72% and AUC_S decreased by 45% with decreasing fat content. Similar trends were found for the Imax values (Table 7.2). A reduction of fat content slowed down the release of limonene ($p<0.05$, T_{max}_R), but no significant effect was observed for citral release.

7.3.2 In-nose aroma release and dynamic lemon perception of mayonnaise with carrier foods

Figure 7.4 shows averaged in-nose limonene release, in-nose citral release and perceived lemon intensity curves for FF-HV mayonnaise with/without carrier foods. The release and perceived intensity curves of the other two mayonnaises (LF-HV and LF-LV) are provided as supplementary data in the paper, as addition of carriers affected release and perception of the different mayonnaises in a relatively similar way. In-nose limonene and citral release parameters (AUC_R, I_{max}_R and T_{max}_R) and perceived lemon parameters (AUC_S, I_{max}_S and T_{max}_S) of mayonnaises without and with carriers are presented in Table 7.3. Overall, in-nose limonene and citral release increased with the addition of food carriers, whereas simultaneous lemon intensity perception of mayonnaises decreased. Bread and potato affected aroma release and perception of mayonnaises in a slightly different way. The results of bread and potato addition are therefore reported separately in the following sub sections.

Effect of bread addition on in-nose aroma release and aroma perception of mayonnaises

Addition of bread increased in-nose limonene and citral release of mayonnaises, regardless of bread texture (Figure 7.4A, 7.4C and Table 7.3). For example, in case of limonene (m/z 138.139), AUC_R increased by 136% and 144% after addition of soft and hard bread, respectively ($p<0.001$; $p<0.001$). For citral (m/z 135.199 and 153.131), AUC_R increased with addition of bread, but this effect was only significant for LF-HV mayonnaise. Similar trends were observed for I_{max}_R values. Bread texture (soft vs. hard) did not affect limonene and citral release concentrations significantly.

The time to reach maximum aroma concentration (T_{max}_R) was affected by addition of bread, regardless of bread texture (Table 7.3). Overall, T_{max}_R was reached earlier for mayonnaise-bread combinations than for mayonnaises consumed without bread. These differences in T_{max}_R were significant for LF-HV but not for FF-HV nor LF-LV.

Addition of bread decreased lemon intensity perception of mayonnaises (Figure 7.4E, Table 7.3). However, the effect of bread on lemon intensity perception was not the same for each mayonnaise (significant mayonnaise:bread interaction). For FF-HV mayonnaise, AUC_S and I_{max}_S were lowered by 11 and 8% with soft bread ($p>0.05$; $p>0.05$) and by 21 and 22% with hard bread ($p=0.003$; $p=0.001$). For LF-LV mayonnaise, AUC_S and I_{max}_S decreased by 10 and 10% with soft bread ($p>0.05$; $p>0.05$) and by 22 and 15% with hard bread ($p=0.030$; $p>0.05$). No significant effect was observed for LF-HV. Hence, bread hardness partly affected lemon intensity perception of mayonnaises. T_{max}_S was not significantly affected by the addition of bread.

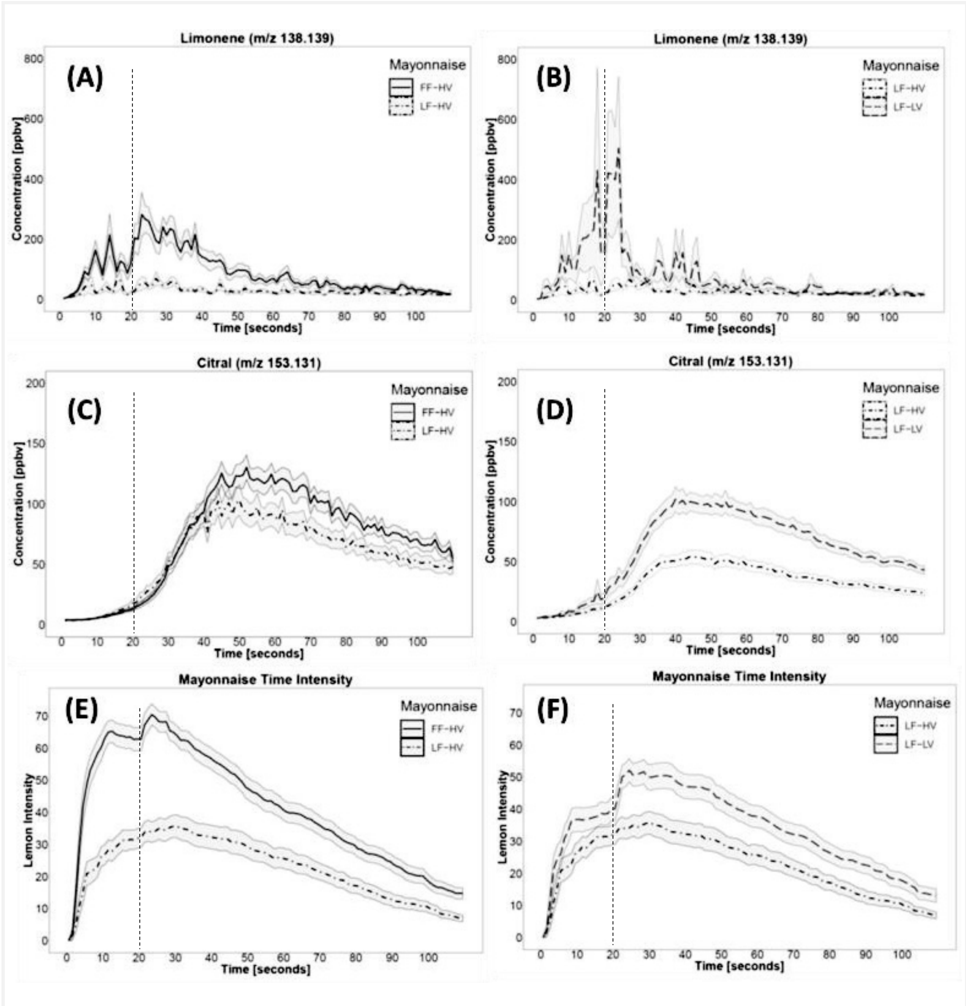

Figure 7.3: Averaged in-nose limonene release (m/z = 138.139) (A, B), in-nose citral release (m/z = 153.131) (C, D) and lemon intensity perception (E, F) during mastication and after swallowing for mayonnaise varying in viscosity and fat content (n=14 subjects, in triplicate). Mayonnaise differing in viscosity (LF-HV and LF-LV) are presented on the left (A, C, E), and the mayonnaise varying in fat content (FF-HV and LF-HV) are presented on the right (B, D, F). The shaded bars represent the standard error of the mean. The moment of swallowing is indicated as dashed line at 20 s.

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Table 7.2: Summary of parameters (mean±SE) describing *in vivo* limonene release, *in vivo* citral release and dynamic lemon intensity perception for mayonnaises varying in fat content (FF = full fat, LF = low fat) and viscosity (HV = high viscosity, LV = low viscosity). The release parameters AUC_R, I_{max}_R, T_{max}_R correspond to the area under the curve, the maximum concentration and time to reach the maximum concentration. The sensory parameters AUC_S, I_{max}_S and T_{max}_S correspond to the total area under the curve, the maximum perceived intensity and the time to reach the maximum perceived intensity.

		Mayonnaise		FF-HV				LF-HV				LF-LV			
AUC		*F*	*p*	*mean*	±	*SE*		*mean*	±	*SE*		*mean*	±	*SE*	
AUC_R (ppbV·s)	*m/z 81.070*	17.0	<0.001	94331	±	6988	*a*	25840	±	3907	*b*	72744	±	14208	*a*
limonene	*m/z 138.139*	10.3	<0.001	9617	±	701	*a*	2695	±	407	*b*	8571	±	2003	*a*
AUC_R (ppbV·s)	*m/z 135.119*	25.8	<0.001	4210	±	277	*b*	3228	±	321	*b*	6226	±	424	*a*
citral	*m/z 153.131*	23.9	<0.001	7550	±	538	*b*	6007	±	608	*b*	11767	±	850	*a*
AUC_S (mm·s)		29.0	<0.001	9198	±	506	*a*	5096	±	489	*c*	7362	±	566	*B*
I_{max}		*F*	*p*	*mean*	±	*SE*		*mean*	±	*SE*		*mean*	±	*SE*	
I_{max}_R (ppbV)	*m/z 81.070*	9.4	<0.001	8521	±	785	*a*	2586	±	468	*b*	8085	±	1616	*a*
limonene	*m/z 138.139*	5.9	<0.01	857	±	80	*ab*	269	±	50	*b*	1030	±	271	*a*
I_{max}_R (ppbV)	*m/z 135.119*	26.3	<0.001	88	±	5	*b*	72	±	7	*b*	134	±	8	*a*
citral	*m/z 153.131*	24.8	<0.001	173	±	12	*b*	145	±	14	*b*	267	±	16	*a*
I_{max}_S (mm)		30.6	<0.001	77	±	3	*a*	48	±	4	*c*	62	±	4	*b*
T_{max}		*F*	*p*	*mean*	±	*SE*		*mean*	±	*SE*		*mean*	±	*SE*	
T_{max}_R (s)	*m/z 81.070*	4.7	<0.05	29	±	2	*b*	39	±	3	*a*	34	±	3	*ab*
limonene	*m/z 138.139*	3.8	<0.05	28	±	2	*b*	38	±	3	*a*	33	±	3	*ab*
T_{max}_R (s)	*m/z 135.119*	0.0	NS	51	±	2		49	±	2		50	±	3	
citral	*m/z 153.131*	0.3	NS	56	±	2		52	±	2		52	±	2	
T_{max}_S (s)		4.8	<0.05	33	±	3	*b*	39	±	3	*ab*	43	±	3	*a*

lower case letters: significant differences between mayonnaise varying in fat content and viscosity (p<0.05).

Effect of potato addition on in-nose aroma release and aroma perception of mayonnaises

Addition of potato to mayonnaises increased both limonene and citral release (Figure 7.4B, 7.4D, Table 7.3). For example, in case of limonene release (m/z 138.139), AUC_R increased by 45 and 43% after addition of soft and hard potato ($p<0.001$ and $p<0.001$). In case of citral release (m/z 135.119), AUC_R increased by 8% with soft potato ($p>0.05$) and by 21% with hard potato ($p<0.001$). Hence, potato texture (soft vs. hard) affected citral release concentrations. Similar trends were observed for I_{max}_R.

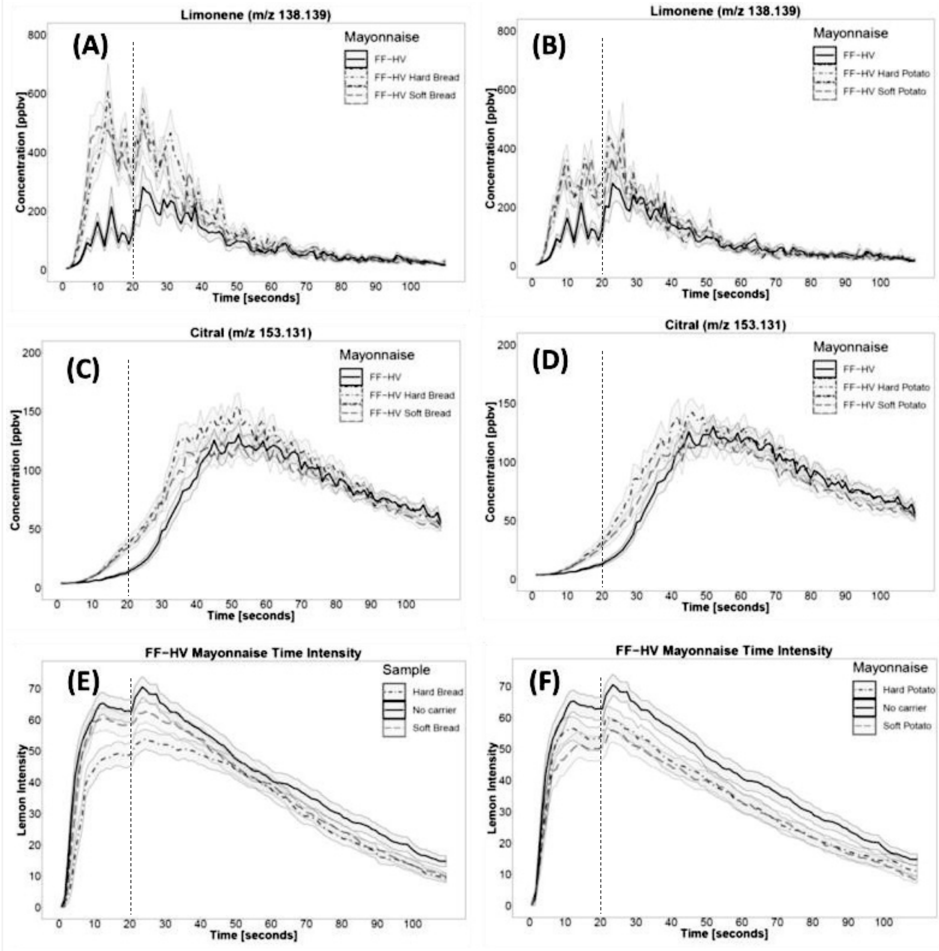

Figure 7.4: Averaged in-nose limonene release (m/z = 138.139) (A, B), in-nose citral release (m/z = 153.131) (C, D) and lemon intensity perception (E, F) during mastication and after swallowing for mayonnaise without and with different food carriers (n=14 subjects, in triplicate). Mayonnaise (*i.e.* FF-HV mayonnaise) with bread carriers (soft, hard) is presented in yellow on the left (A, C, E), and the mayonnaise with potato carriers (soft, hard) is presented in pink on the right (B, D, F). The shaded bars represent the standard error of the mean. The moment of swallowing is shown as gray dashed line at 20 s.

Table 7.3: Summary of parameters (mean±SE) describing *in vivo* limonene release, *in vivo* citral release and dynamic lemon intensity perception for mayonnaises without and with carrier foods. The effect of bread and potato are presented separately. The release parameters AUC_R, I_{max}_R, T_{max}_R correspond to the area under the curve, the maximum concentration and time to reach the maximum concentration. The sensory parameters AUC_S, I_{max}_S and T_{max}_S correspond to the total area under the curve, the maximum perceived intensity and the time to reach the maximum perceived intensity.

		Bread		Mayo:Bread		
		F	*p*	*F*	*p*	
AUC						
AUC_R (ppbV·s)	*ms 81.070*	63.1	p<0.001	1.1	NS	
limonene	*ms 138.139*	42.4	p<0.001	1.3	NS	
AUC_R (ppbV·s)	*ms 135.119*	13.1	p<0.001	4.5	p<0.01	*FF-H*
citral						*LF-H*
						LF-L
	ms 153.131	6.2	p<0.01	6.0	p<0.001	*FF-H*
						LF-H
						LF-L
AUC_S (mm·s)		7.0	p<0.01	4.0	p<0.01	*FF-H*
						LF-H
						LF-L
Imax						
Imax_R (ppbV)	*ms 81.070*	34.4	p<0.001	0.5	NS	
limonene	*ms 138.139*	17.9	p<0.001	1.4	NS	
Imax_R (ppbV)	*ms 135.119*	11.2	p<0.001	2.3	NS	
citral	*ms 153.131*	4.5	p<0.05	4.8	p<0.001	*FF-H*
						LF-H
						LF-L
Imax_S (mm)		9.1	p<0.001	3.5	p<0.01	*FF-H*
						LF-H
						LF-L
Tmax						
Tmax_R (s)	*ms 81.070*	32.8	p<0.001	4.2	p<0.01	*FF-H*
limonene						*LF-H*
						LF-L
	ms 138.139	30.4	p<0.001	4.0	p<0.01	*FF-H*
						LF-H
						LF-L
Tmax_R (s)	*ms 135.119*	6.3	p<0.01	1.3	NS	
citral	*ms 153.131*	1.4	NS	0.7	NS	
Tmax_S (s)		0.3	NS	1.4	NS	

capital letters: significant differences between mayonnaise without/with bread carriers (p<0.05)

No carrier				With soft bread				With hard bread			
mean	*±*	*SE*		*mean*	*±*	*SE*		*mean*	*±*	*SE*	
64613	±	6001	*B*	149536	±	9825	*A*	158206	±	9186	*A*
6995	±	770	*B*	16553	±	1278	*A*	17085	±	1173	*A*
4210	±	277	*A*	4532	±	248	*A*	5119	±	307	*A*
3228	±	321	*B*	5970	±	400	*A*	5562	±	450	*A*
6226	±	424	*A*	6766	±	423	*A*	6590	±	460	*A*
7550	±	538	*A*	7833	±	477	*A*	8909	±	595	*A*
6007	±	608	*B*	10828	±	775	*A*	9953	±	859	*A*
11767	±	850	*A*	11571	±	736	*A*	10902	±	821	*A*
9198	±	506	*A*	8214	±	437	*AB*	7307	±	488	*B*
5096	±	489	*A*	5844	±	490	*A*	5515	±	481	*A*
7362	±	566	*A*	6608	±	522	*AB*	5767	±	484	*B*
6414	±	658	*B*	12880	±	926	*A*	13414	±	937	*A*
720	±	99	*B*	1613	±	175	*A*	1533	±	147	*A*
97	±	5	*B*	121	±	5	*A*	128	±	7	*A*
173	±	12	*A*	176	±	12	*A*	203	±	14	*A*
145	±	14	*B*	248	±	19	*A*	223	±	20	*A*
267	±	16	*A*	268	±	19	*A*	250	±	22	*A*
77	±	3	*A*	71		3	*AB*	61		3	*B*
48	±	4	*A*	48		3	*A*	48		3	*A*
62	±	4	*A*	56		3	*A*	52		4	*A*
29	±	2	*A*	24	±	2	*A*	26	±	2	*A*
39	±	3	*A*	24	±	2	*B*	22	±	2	*B*
34	±	3	*A*	21	±	1	*B*	23	±	2	*B*
28	±	2	*A*	24	±	2	*A*	25	±	1	*A*
38	±	3	*A*	23	±	2	*B*	22	±	2	*B*
33	±	3	*A*	19	±	1	*B*	24	±	2	*B*
50	±	1	*A*	43	±	1	*B*	46	±	1	*AB*
54	±	1		52	±	1		55	±	1	
38	±	2		38	±	2		40	±	2	

7

Table 7.3: Continued

		Potato		Mayo:Potato		
		F	*p*	*F*	*p*	
AUC						
AUC_R (ppbV·s)	*ms 81.070*	32.5	p<0.001	2.2	NS	
limonene	*ms 138.139*	21.8	p<0.001	1.9	NS	
AUC_R (ppbV·s)	*ms 135.119*	9.0	p<0.001	2.2	NS	
citral	*ms 153.131*	4.5	p<0.05	3.1	p<0.05	*FF-H*
						LF-H
						LF-L
AUC_S (mm·s)		10.5	p<0.001	2.1	NS	
Imax						
Imax_R (ppbV)	*ms 81.070*	20.0	p<0.001	2.4	NS	
limonene	*ms 138.139*	10.4	p<0.001	1.7	NS	
Imax_R (ppbV)	*ms 135.119*	9.1	p<0.001	2.3	NS	
citral	*ms 153.131*	5.4	p<0.01	2.8	p<0.05	*FF-H*
						LF-H
						LF-L
Imax_S (mm)		9.8	p<0.001	1.4	NS	
Tmax						
Tmax_R (s)	*ms 81.070*	20.1	p<0.001	1.5	NS	
limonene	*ms 138.139*	16.1	p<0.001	1.5	NS	
Tmax_R (s)	*ms 135.119*	5.8	p<0.01	0.7	NS	
citral	*ms 153.131*	1.9	NS	0.2	NS	
Tmax_S (s)		0.2	NS	1.0	NS	

capital letters: significant differences between mayonnaise without/with bread carriers (p<0.05)

No carrier				With soft potato				With hard potato			
mean	*±*	*SE*		*mean*	*±*	*SE*		*mean*	*±*	*SE*	
64613	±	6001	*B*	118421	±	8157	*A*	118637	±	7441	*A*
6995	±	770	*B*	12697	±	1015	*A*	12350	±	803	*A*
4552	±	227	*B*	4957	±	195	*B*	5722	±	246	*A*
7550	±	538	*A*	7476	±	527	*A*	8091	±	605	*A*
6007	±	608	*B*	8365	±	577	*AB*	9536	±	772	*A*
11767	±	850	*A*	10706	±	634	*A*	12201	±	658	*A*
7236	±	335	*A*	6085	±	307	*B*	6200	±	300	*B*
6414	±	658	*B*	11860	±	927	*A*	11370	±	898	*A*
720	±	99	*B*	1373	±	151	*A*	1361	±	155	*A*
97	±	5	*B*	110	±	5	*B*	127	±	6	*A*
173	±	12	*A*	172	±	14	*A*	182	±	14	*A*
145	±	14	*B*	204	±	15	*AB*	233	±	20	*A*
267	±	16	*A*	260	±	17	*A*	295	±	19	*A*
62	±	2	*A*	54	±	2	*B*	57	±	2	*AB*
34	±	1	*A*	25	±	1	*B*	27	±	1	*B*
33	±	1	*A*	24	±	1	*B*	27	±	1	*B*
50	±	1	*A*	45	±	1	*B*	46	±	1	*B*
54	±	1		51	±	1		51	±	1	
38	±	2		38	±	2		37	±	2	

The time to reach maximum aroma concentration of mayonnaises (T_{max}_R) was affected by the addition of potatoes, regardless of potato texture (Table 7.3). On average, T_{max}_R was reached after shorter times after the addition of potato.

Regarding sensory perception, presence of potato carriers decreased perceived lemon intensity. AUC_S decreased by 19% with addition of soft potato ($p<0.001$) and by 17% with addition of hard potato ($p=0.002$). Similar effects were observed for I_{max}_S, but the effect was significant for soft potato (reduction by 15% $p<0.001$) but not for hard potato (reduction by 9%; $p>0.05$). T_{max}_S was not significantly affected by addition of potato carriers.

7.3.3 Results overview: in-nose aroma release and sensory perception of mayonnaise-carrier combinations

A Multiple Factor Analysis (MFA) analysis was conducted to summarize the effect of mayonnaise viscosity, mayonnaise fat content, carrier addition and carrier texture on aroma release and perception of mayonnaises (Figure 7.5). PC1 explained 44.9% of total variance and accounted mainly for differences in mayonnaise fat content (Figure 7.5B). In this case, the 95% confidence ellipses highlight two clusters: one with full fat mayonnaise (FF-HV) and the other with the two low fat mayonnaises (LF-HV and LF-LV). PC2 explained 34.3% of total variance and accounted for sample differences in viscosity (Figure 7.5A) and carrier addition (Figure 7.5C). In this case, the 95% confidence ellipses highlight two clusters: one with low viscosity mayonnaises (LF-LV) and one with high viscosity mayonnaises (LF-HV and FF-HV). Figure 7.5C highlights a difference between food carriers: bread samples are positioned further away from the single mayonnaises than potato samples, indicating that bread had a larger overall impact on lemon aroma release and intensity perception than potato.

To summarize, increasing mayonnaise viscosity or decreasing mayonnaise fat content reduced lemon aroma release and simultaneous lemon intensity perception. The two lemon aroma compounds (limonene, citral) had slightly different release patterns, with limonene being released faster and with higher concentration due to its higher volatility than citral. When mayonnaises were combined with carriers, aroma release and perception were no longer congruent. Addition of bread and potato to mayonnaises enhanced lemon aroma release and decreased simultaneous lemon intensity perception. When comparing the different carrier foods, addition of bread increased lemon aroma release concentrations more than potato. Bread hardness did not influence lemon aroma release, but harder bread tended to decrease lemon intensity perception to a larger extent than soft bread. Potato hardness did not influence aroma release, but softer potato tended to decrease lemon intensity perception slightly more than harder potato.

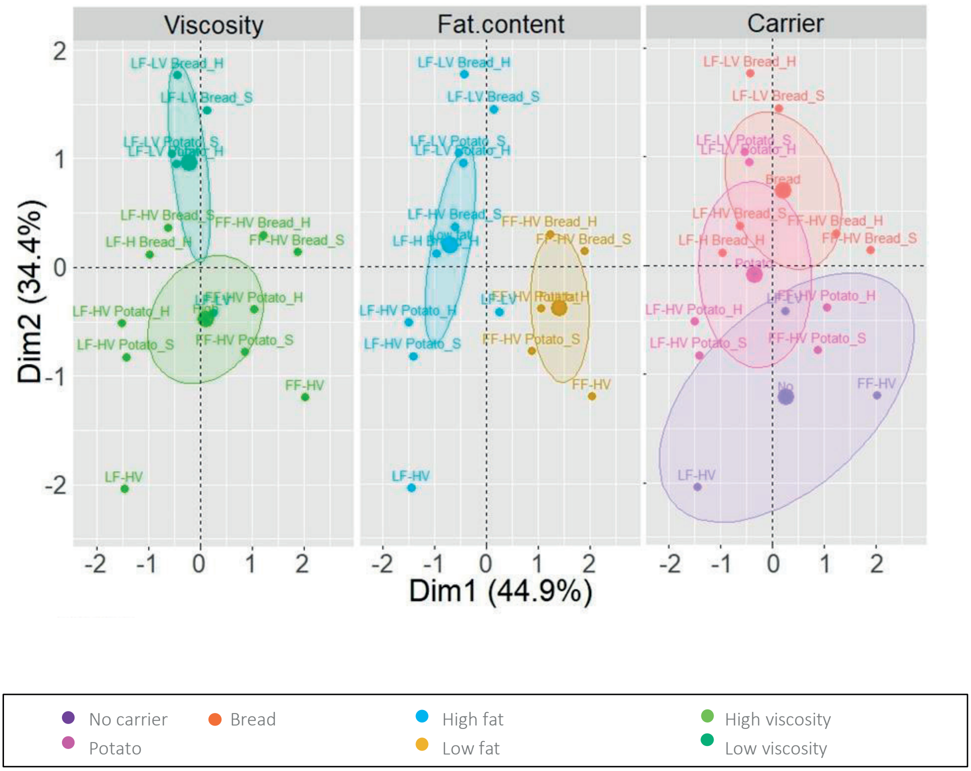

Figure 7.5: Scatter plot of multiple factor analysis (MFA) of in-nose aroma release and dynamic lemon perception data, in which the mayonnaise viscosity effect (A), the mayonnaise fat content effect (B) and the carrier effect (C) are highlighted using different colors.

7.4 DISCUSSION

Regarding the in-nose aroma release of condiment-carrier combinations, we see that aroma release from condiments (mayonnaises) is enhanced when consumed together with carriers (bread or potatoes) compared to consumption without carriers. We hypothesized that carriers would actually lower aroma release by binding aroma compounds (*i.e.* physicochemical effect). Although such interaction might have occurred through physical non-covalent bonds between carriers and condiments, a higher concentration of aroma compounds was released in the nose with the addition of carrier foods. This indicates that other mechanisms play a larger role in actual in-nose aroma release. We suggest that, during the 20 s of consumption, the difference in food oral processing between mayonnaise and mayonnaise in combination with carriers explain the increase in in-nose aroma release. Despite a standardized consumption protocol (mayonnaise without carrier: swirl in mouth for 20 s; mayonnaise with carrier: chew with 1 chew/s for 20 s; section 7.2.3), mayonnaise-carrier combinations required chewing to safely break down the food before swallowing, whereas the single mayonnaises did not require chewing and they were just swirled around in the mouth. The chewing might have induced more aroma release. Moreover, as a result of chewing and mixing, the surface area of mayonnaise-carrier combinations might have increased since the carrier might have been broken down into multiple smaller bolus pieces. As the mayonnaise is now distributed over a larger area, a higher transfer of aroma compounds from the mayonnaise to the vapor phase might have taken place. This could explain why total aroma released increased. This was also reflected in the time required to reach the maximum aroma concentration (T_{max}_R), which was faster in case of the carrier-mayonnaise combinations than for single mayonnaises (Table 7.3). In addition, the velum-tongue border has been observed to open more frequently during consumption of solid foods than liquid foods (Buettner *et al.* 2002), which could increase the ability of aroma compounds to pass to the nasal cavity ahead of swallowing. Such an effect of oral processing behavior on *in vivo* aroma release is consistent with previous research (Hansson *et al.* 2003; Pionnier *et al.* 2004; Aprea *et al.* 2006; Tarrega *et al.* 2008; Raithore & Peterson 2018; Doyennette *et al.* 2019). Addition of solid carrier foods to condiments thus increases oral movements, in-mouth food manipulations and food's surface area, and thereupon favors an increase in in-nose aroma release of condiments throughout consumption.

Higher in-nose aroma release with the addition of carriers foods was still maintained after participants swallowed the sample. Thus, also after the moment of swallowing, higher in-nose aroma concentrations were observed for mayonnaise-carrier combinations than for single mayonnaises. Such effect might be explained by differences in bolus properties and consequent oral retention. When mayonnaise is consumed on its own, it is mixed with saliva leading to a liquid-like bolus that is easily swallowed and we assume that little product remains in the mouth after swallowing. When mayonnaise is consumed with a carrier, it is mixed with both the carrier and saliva leading to a relatively cohesive solid bolus that easily sticks to different oral surfaces (teeth, tongue, palate) upon swallowing. In this case much more product remains in the mouth which likely leads to longer aroma release into the nasal cavity after swallowing.

The type of carrier food (bread versus potato) affected *in vivo* aroma release of mayonnaises, since bread increased nasal aroma concentrations to a larger extent than potatoes (Figure 7.4, Table 7.3). We suggest that this result could be partly explained by a difference in moisture absorption capacity between bread and potato. Bread is a dry, low water content product, so it absorbs moisture from the mayonnaises (van Eck *et al.* 2019b). This effectively increases the oil content in the mayonnaises, and decreases the volume of the continuous aqueous phase, through which the aroma compounds need to diffuse before reaching the air phase. The increase in oil content therefore leads to a larger surface area from which the aroma compounds can be released. Consequently, the aroma compounds are assumed to diffuse faster into the nasal cavity for breads. Another explanation for the lower release of aroma compounds from potato can be found in the properties of the starch in cooked potatoes. Potatoes contain starch granules, which are gelatinized upon cooking. The gelatinization leads to release of amylose from the granules into the continuous phase, whereas amylopectin resides mostly within the granules. Consequently, starch (mainly amylose) becomes available for interactions with hydrophobic aroma compounds after cooking through hydrophobic interactions. It is known that gelatinized starch retains hydrophobic aroma compounds including limonene to a larger extent than starch granules (Boutboul *et al.* 2002). Such interactions can limit aroma release, and could explain the lower release for potato. Together these results show that mayonnaise aroma release depended on the properties of the carriers it is combined with.

The texture of carrier foods did not significantly influence mayonnaise aroma release. It is important to note that a standardized consumption protocol was used (section 7.2.3), meaning that both soft and hard carrier foods were chewed 20 times at the same chewing frequency. This did not allow participants to adapt oral behavior based on texture, and presumably resulted in similar nasal air flows and release patterns, and this could be the reason why we see no effect of texture on release. In case of free eating, differences in aroma release of mayonnaises depending on the texture of the carrier food might occur, since softer foods generally require fewer chews than harder foods, likely to result in different nasal air flows which in turn can affect in-nose aroma release. For example, in case of cheese, firmer cheeses were chewed for a longer time and broken down into more bolus pieces by which both the release rate and the total amount of released aroma were increased (Tarrega *et al.* 2011; Repoux *et al.* 2012).

Inter-individual variation between subjects is known to affect oral behavior, aroma release and perception (Charles *et al.* 2000; Labouré *et al.* 2014; Pedrotti *et al.* 2019). To alleviate such subject variation, we selected a specific panel (young, female, Caucasian) and standardized their way of chewing by training with a chewing protocol (section 7.2.3). A next step would involve studies investigating aroma release of condiment-carrier combinations among participants with different eating behaviors (slow vs. fast eaters), as eating rate is known to affect bolus formation, sensory perception and food intake (Robinson *et al.* 2014; Devezeaux de Lavergne *et al.* 2017).

Also mayonnaise properties (viscosity, fat content) were observed to influence aroma release of mayonnaises considerably. As both viscosity and fat content have been shown to influence aroma release of single foods in previous studies, these results are discussed only shortly throughout this paper. Increasing mayonnaise viscosity by adding more xanthan resulted in lower aroma release and perception (LF-LV vs. LF-HV). Viscosity is known to play a relevant role in aroma release, as diffusion rate of aroma compounds is hindered by an increase in viscosity (Malone *et al.* 2003; Weterings *et al.* 2019). In addition, xanthan has been suggested to physically interact with hydrophobic aroma compounds by trapping them into a so-called “hydrophobic cavity” (Milas *et al.* 1990; Bylaite *et al.* 2005). Decreasing mayonnaise fat content while keeping the same viscosity resulted in decreased aroma release and perception (FF-HV and LF-HV). A similar observation was reported by Wendin *et al.* (1997), who found that decreased fat content tended to decrease the perceived lemon intensity in mayonnaise (Wendin *et al.* 1997). However, these results do not support the general theory that the partitioning of hydrophobic aromas into aqueous phases and air is greatly reduced with increasing fat/oil content (González-Tomás *et al.* 2007; Arancibia *et al.* 2011; Frank *et al.* 2011). This discrepancy may be due to different factors. Firstly, aroma compounds may interact with xanthan in low fat emulsions (LF-HV), which was added to compensate for the difference in viscosity due to the reduction of fat. So even though lowering oil content could provide the expected increase in aroma release, interactions with xanthan might have been more pronounced, eventually leading to a decrease in aroma release. Secondly, the FF-HV mayonnaise contains a higher number of fat droplets when compared to the LF-HV. This results in more interfacial area between oil and the continuous aqueous phase, and therefore interaction with the saliva may be increased and eventual transfer to the air phase. This may ultimately lead to a higher aroma release and an accompanying higher aroma concentration in the nose space (Tarrega *et al.* 2019).

To summarize, our study highlights that aroma release from mayonnaises is enhanced when they are consumed together with carrier foods such as bread or potatoes. Intuitively, one would expect that this increase in aroma release would be reflected in an increase in aroma perception. However, when looking at the sensory perception of the mayonnaise-carrier combinations, carrier addition actually decreased perceived aroma intensity of mayonnaises (Figure 7.4E-7.4F, Table 7.3). This decrease in perceived intensity is in line with previous studies, indicating that flavor intensity of soy sauce and mayonnaise decreased with addition of solid carrier foods (Cherdchu & Chambers 2014; van Eck *et al.* 2019a). The present study shows that the lower perceived intensity is not due to a lower delivery of aroma compounds into the nasal cavity, as aroma release was increased with addition of carriers (Figure 7.4A-7.4D, Table 7.3). This misalignment between perception and aroma release in case of mayonnaise-carrier combinations indicates that carriers modify condiment perception via other ways independent of actual in-nose aroma concentrations. We therefore suggest that cognitive effects play a pivotal role in the modulation of condiment-carrier perception, *i.e.* consumers pay more attention to texture and/or chewing with the presence of carriers, whereupon the aroma of condiments appears to be less intense. Recently, White *et al.* (2019) stressed that the influence of cognitive processes on sensory evaluation should be considered more by food scientists (White *et al.* 2019). They revealed that consumer perception is shaped by the way attention is distributed among sensory sensations. This phenomenon has been mainly discussed in the light of aroma-taste mixtures, showing that attention was directed to one or a few elements of multisensory mixtures. In the present study, condiments were evaluated in combination with solid carrier foods, which added another dimension (*i.e.*

texture / the process of chewing) to the aroma perception of the mayonnaises. We argue that cognitive attention was thereby drawn to the process of chewing. In case of condiment-carrier combinations, focusing on a specific task (*e.g.* chewing) might thus limit conscious perception of other senses (*e.g.* aroma) present, and this cognitive influence is important to keep in mind in sensory evaluation of complex foods.

Although such a cognitive effect seems plausible, it is important to acknowledge a possible sensory dumping effect (a well-known limitation of the Time-Intensity methodology) (Lawless & Heymann 2010). Carriers with different texture properties were added to mayonnaises with lemon aroma, and participants were asked to evaluate lemon intensity only. Subjects probably perceived differences in texture, and where asked to evaluate lemon aroma intensity only which might have led to the projection of perceived differences and changes in texture into lemon intensity. To minimize potential dumping effect, the perceived textural differences were carefully discussed during the multiple training sessions. Subsequently, the panel practiced with the evaluation of aroma intensity, while being aware of the possible differences in texture. In this context, it is known that transfer of aroma compounds into the nasal cavity follows swallow breath (Buettner *et al.* 2001). Thus, aroma perception is known to increase just after swallowing. When looking at our Time-Intensity data (Figure 7.3E-7.3F, 7.4E-7.4F), we observe a consistent increase in perceived lemon intensity just after 20 s. This demonstrates that our panel functioned very well since they clearly perceived this increase in aroma after swallowing, which strongly suggests that our panel was capable to perceive and evaluate aroma intensity. We therefore assume the sensory dumping effect to be small. Hence, the influence of cognitive effects on sensory perception of complex foods has to be considered.

The novelty of the present study is the fact that simultaneous aroma release and perception was assessed for condiment-carrier combinations and not only in model foods or single foods. Combining condiments and carrier foods increases the complexity of the food consumed, which is more representative of the common consumption context. In summary, in-nose aroma release matched perceived aroma intensity when mayonnaise was consumed alone (*i.e.* when higher aroma concentrations were released in the nose, also higher perceived aroma intensity values were reported). This was not the case for more complex foods such as condiment-carrier combinations. Addition of carriers increased in-nose aroma release but decreased the perceived aroma intensity of mayonnaises. Since this decreased aroma perception was not due to a lower delivery of aroma compounds into the nasal cavity, we conclude that aroma release alone does not explain sensory perception of composite foods. In case of composite foods, cognitive effects are likely to modulate perception of more complex food combinations, which supports the idea that not only physicochemical characteristics, but also consumers' cognitive mode should be considered in food design with excellent consumer appreciation.

MAYONNAISE
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GENERAL DISCUSSION

8

8. GENERAL DISCUSSION

Combine nicely, consume wisely - The research described in this thesis aimed to investigate the role of properties of single foods in oral processing behavior, sensory perception (*combine nicely*) and food intake (*consume wisely*) of composite foods. The composite foods used were composed of a condiment (*i.e.* mayonnaise, cheese) and a solid carrier food (*i.e.* bread, cracker, carrot, potato). Condiment and carrier properties were systematically varied (Figure 8.1) with respect to condiment type, condiment fat content, condiment viscosity, carrier type, carrier hardness and carrier shape. The main results and interpretations of this thesis are summarized and discussed in section 8.1. Then, methodological considerations (section 8.2), suggestions for future research (section 8.3) and main conclusions (section 8.4) are provided.

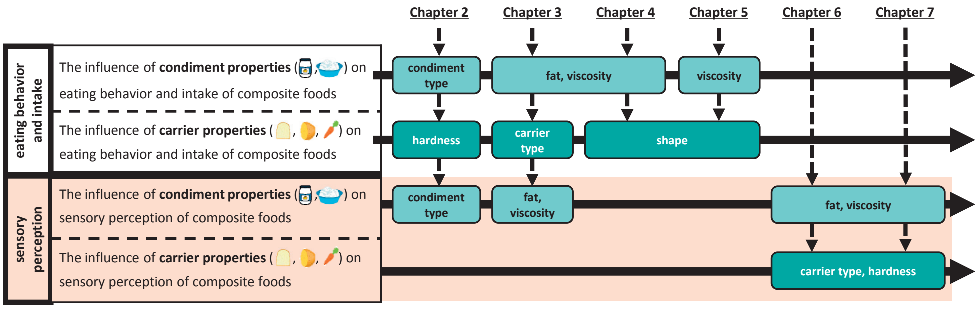


Figure 8.1: Schematic overview showing in which chapters condiment properties (light color) and/or carrier properties (dark color) were varied to determine the effect of single food properties on eating behavior, intake and/or sensory perception of composite foods.

8.1 Discussion and interpretations of main results

Summary of the main outcomes

The main findings of this thesis are summarized based on the effect of addition of condiments (Figure 8.2), condiment properties (Figure 8.3A) and carrier properties (Figure 8.3B).

As represented in Figure 8.2, addition of condiments assisted saliva in bolus formation of carrier foods (lower friction, softening bread, adhering vegetable pieces together into a cohesive bolus) leading to increased eating rate. Addition of condiments to carrier foods increased overall perceived flavor intensity, whereas carrier flavor and texture intensities decreased. With respect to dynamic sensory perception, perception was dominated by the texture of the solid carriers, especially in the beginning and end of consumption, whereas additional flavor sensations related to the condiments appeared in the middle stage of consumption.

8

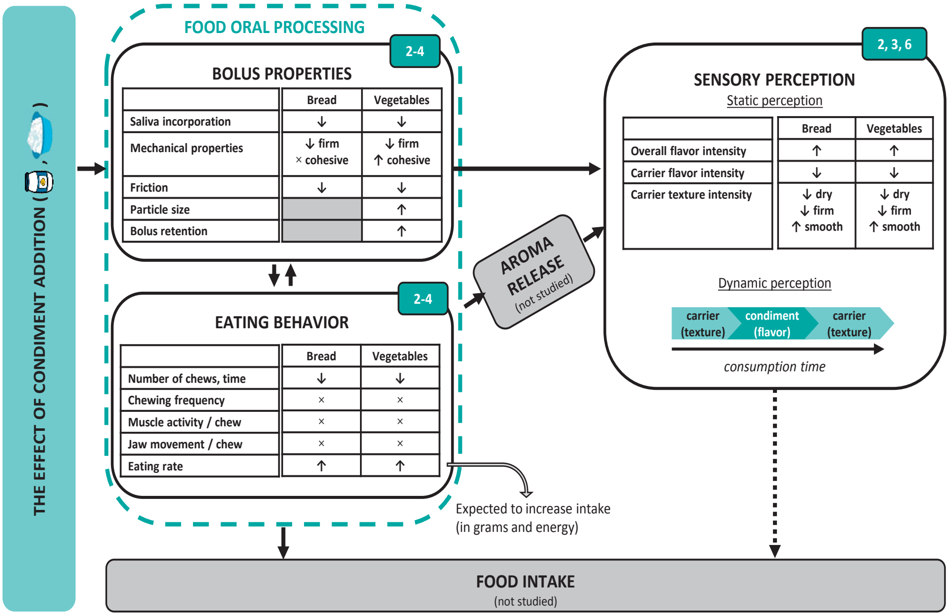
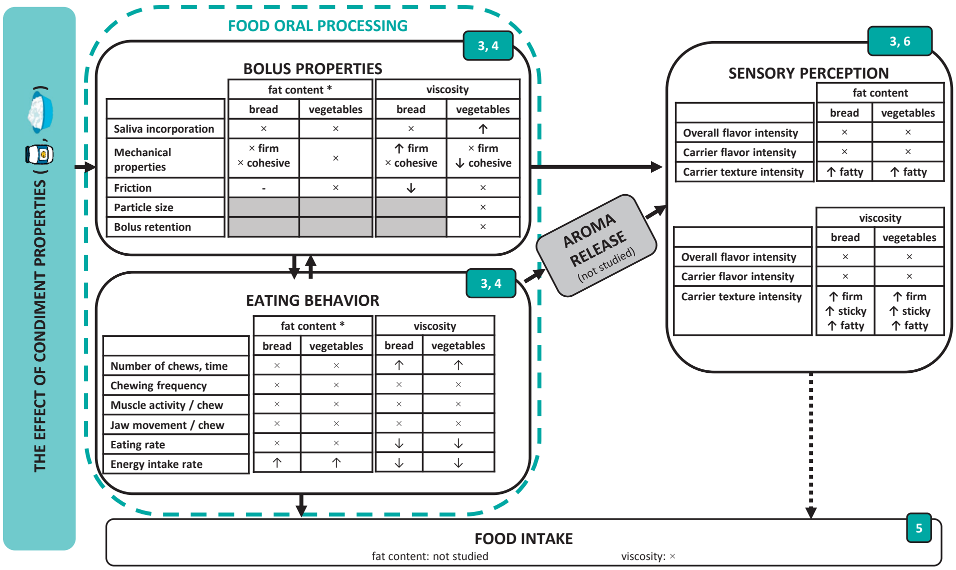


Figure 8.2: Schematic overview of the effect of condiment addition on food oral processing behavior, food intake and sensory perception of composite foods. Increased effects, decreased effects and no effects are illustrated with '↑', '↓' and '×', respectively. Gray boxes represent parameters that were not studied in this thesis. The numbers indicate the different chapters in which the results are described.

A



* The influence of fat content studied in Chapter 4 is not incorporated in this overview due to possible interactions with xanthan.

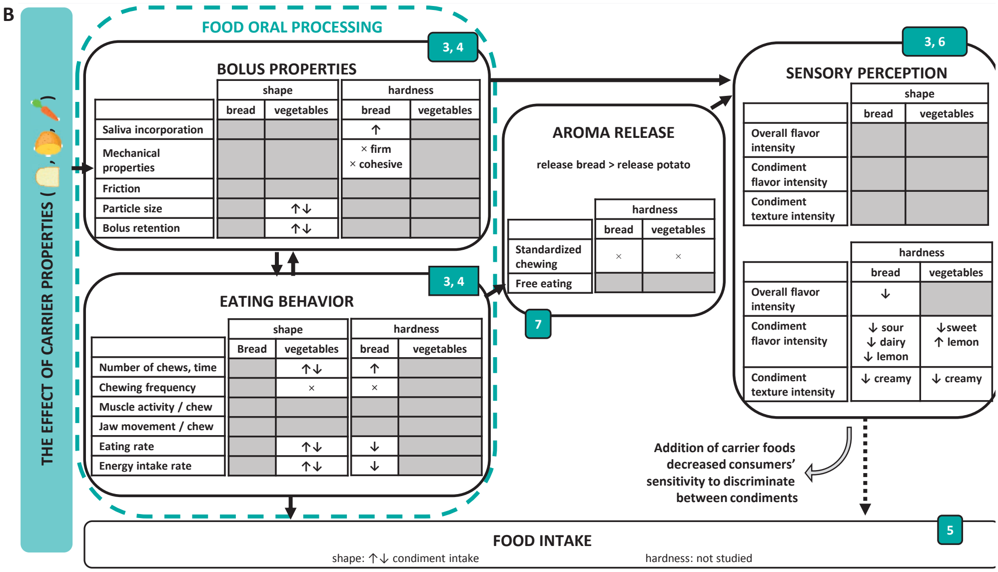


Figure 8.3: Schematic overview of the effect of condiment properties (A) and carrier properties (B) on food oral processing behavior, food intake and sensory perception of composite foods. Increased effects, decreased effects and no effects are illustrated with '↑', '↓' and '×', respectively. Gray boxes represent parameters that were not studied in this thesis. The numbers indicate the different chapters in which the results are described.

Single food properties impact eating behavior, bolus properties and sensory perception of composite foods in various ways. As represented in Figure 8.3A, increasing condiment fat content (from low fat to full fat) had a minor effect on eating behavior of composite foods (0-5% difference in eating rate), but increased perception of fattiness considerably. Low fat mayonnaises lubricated bread boli to a larger extent than full fat mayonnaise, whereas mayonnaise fat content did not affect mechanical properties nor lubrication of potato boli. Increasing condiment viscosity (from salad dressing to mayonnaise) reduced eating rate of composite foods by 13-16%, and enhanced firmness, stickiness and fattiness perception. Increasing mayonnaise viscosity enhanced firmness and friction of bread boli, whereas it reduced cohesiveness of potato boli.

The addition of a carrier food reduced consumer sensitivity to discriminate between condiments. With respect to specific carrier properties (Figure 8.3B), increasing carrier hardness (from bread to cracker) reduced eating rate of composite foods by 29-33%. Flavor perception of condiments decreased with increasing carrier hardness, whereas aroma release was not affected when a standardized chewing protocol was used. Carrot shape influenced eating behavior, with raw carrot cubes being consumed with 17% higher eating rate than carrots cut julienne. Varying cracker shape affected consumers' dipping behavior of cheese dips, with more cheese being consumed per dip (2.9±0.2 vs. 2.3±0.1 g cheese per dip). This led to higher *ad libitum* cheese intake (15%) when flat, square shaped crackers were used compared to finger shaped crackers.

The role of single food properties in eating behavior of composite foods

In everyday life consumers combine different foods, which influences their food intake. For example, condiments are added to provide lubrication to solid foods to facilitate safe-to-swallow bolus formation and subsequently increase food intake among the elderly (Cichero 2017). This thesis indeed proves that the addition of condiments to solid foods is an effective strategy to ease bolus formation (**Chapters 2-4)**.

Swallowing thresholds have been under scientific discussion since many years. There are a few studies that postulated the existence of a universal swallowing threshold, meaning that for a given food certain bolus properties should be reached to trigger swallowing, and that such a threshold is similar across consumers (Fontijn-Tekamp *et al.* 2004; Peyron *et al.* 2004). For example, no inter-individual variation (n=10) was observed in particle size distributions at the moment of swallowing for different nuts nor for different vegetables (Peyron *et al.* 2004). On the other hand, several studies suggested an individual rather than a universal swallowing threshold, as bolus properties at the moment of swallowing were found to differ considerably between consumers (Loret *et al.* 2011; Yven *et al.* 2012; Devezeaux de Lavergne *et al.* 2015a; Young *et al.* 2016a; Young *et al.* 2016b). Throughout this thesis, we selected participants considering their demographics and chewing rate (**Chapters 2, 3**), but we still observed large differences among participants in bolus properties at the moment of swallowing . This further supports the idea of an individual swallowing threshold.
In general, the scientific community agrees with Hutchings and Lillford's philosophy of the breakdown path, indicating that food structure has to be reduced to a certain level and food lubrication has to be above a certain level for the bolus to be swallowed safely (Hutchings & Lillford 1988). In line with this model, condiments were observed to soften bread and cracker structure (**Chapters 2, 3**) and to enhance lubrication by lowering friction (**Chapter 3**). However, we also showed that bolus cohesion is another extremely important factor in safe swallowing, as condiments facilitated bolus formation by adhering solid pieces together into a cohesive bolus **(Chapters 3, 4**). This holds in particular for food boli that consist of multiple, separate pieces, as such boli increase the risk of choking. It should be noted that the model of Hutchings and Lillford does not include bolus cohesion nor mentions intake or perception. Although the importance of bolus cohesion in safe swallowing has already been discussed in other studies focusing on single foods, including bread (Panouillé *et al.* 2014), biscuits (Young *et al.* 2016b), and breakfast cereals (Loret *et al.* 2011; Peyron *et al.* 2011), bolus cohesion still appears to be an underreported and undervalued aspect of safe swallowing.

The effect of food shape on eating rate is a yet underexplored topic. This thesis highlights that variation in food shape influences oral processing behavior and food intake considerably (**Chapters 4, 5**). It is noteworthy that cracker shape influenced *ad libitum* food intake unconsciously, as no differences in self-reported fullness were observed (**Chapter 5**).

Variation in food shape instead of food texture is an advantageous strategy, in particular because this strategy does not require food reformulations (thereby assuming unaffected consumer perception) and can be applied relatively easily by food manufacturers. Although the underlying mechanisms are not well understood, it can be expected that food shape can influence eating behavior via different routes, including (1) consumption effort (from package/plate/bowl to mouth) and (2) chewing effort (from bite to swallow). Firstly, in case of consumption effort, consumers are thought to apply a certain habitual consumption effort (*i.e*. reaching for a piece, taking a bite). Indeed, **Chapter 5** shows that consumers took a similar number of cracker bites among sessions. In other studies such an effect of shape was also observed, and indicated that changing consumption effort by shape might be used to either increase or decrease intake. For instance, total intake was lower when foods were served as multiple small pieces compared to one larger piece (Weijzen *et al.* 2008; Goh *et al.* 2017; Liem & Russell 2019), likely because consumers had to reach for their food more often. Secondly, in case of chewing effort, some shapes require higher chewing effort in mouth to form a safe-to-swallow bolus. For instance, as shown in **Chapter 4**, carrots cut julienne required more chews until swallowing than carrots cut into cubes. Similarly, yogurt with many small granola pieces required higher chewing effort leading to lower intake than yogurt with few large granola pieces (Mosca *et al.* 2019b). Intuitively, one expects that smaller pieces require less chewing effort than larger pieces. This is probably true if one would compare chewing one small piece with one large piece. However, as shape/size was changed but total weight was kept constant, this resulted in a larger number of smaller pieces. In both cases, more time and saliva were required to adhere the larger number of smaller pieces into a cohesive safe-to-swallow bolus. Further research is needed to compare different types of solid foods and different shapes to validate this general theory about the influence of food shape on eating behavior and food intake.

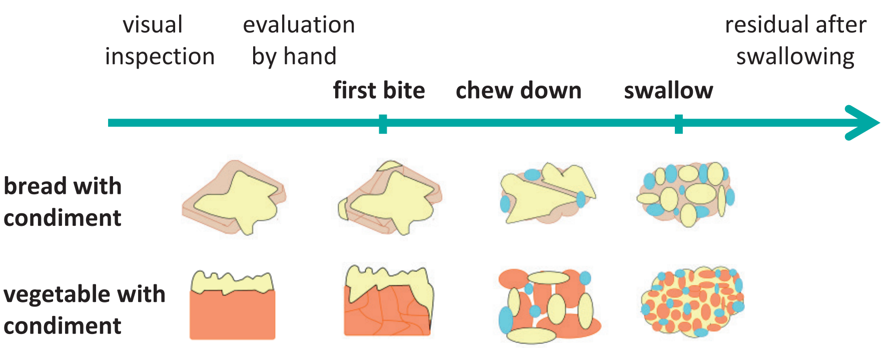


Figure 8.4: Schematic representation of different stages of food oral processing behavior of composite foods, showing structural breakdown of the carrier food (bread: in brown, vegetable: in orange) and lubrication by condiment (in yellow) and saliva (in blue).

This thesis focused on solid carrier foods combined with condiments (bread-condiment and vegetable-condiment combinations) only. This thesis demonstrated that condiments assisted saliva in bolus formation of solid carriers by decreasing bolus structure, increasing bolus lubrication, increasing bolus cohesion, or a combination of these three (Figure 8.4). To validate a general theory about eating behavior of carrier-condiment combinations, the following question can be raised: do condiments assist bolus formation of any type of solid food? As the assistance by condiments was observed for two distinctly different carriers (dry, moisture absorbing bread and vegetables with a high moisture content), it is believed that this concept holds for any solid carrier that is combined with a condiment (*e.g.* pasta with sauce or meat with gravy). It is important to bear in mind that, depending on the carrier and condiment properties, the magnitude of the effect as well as the underlying mechanisms might differ between carrier foods. For instance, condiment addition allowed carrier boli to be safely swallowed after fewer chews and shorter chewing times by different mechanisms, as condiments reduced the firmness of bread bolus, whereas they enhanced cohesiveness of potato bolus (**Chapter 3**; Figure 8.4). Largest facilitation effects are expected for dry foods, as these foods can absorb and incorporate moisture from condiments. Smallest facilitation effects are expected for very tough foods like meat, as these foods require intensive structure breakdown by the molars before swallowing regardless of a small increase in lubrication by condiments. In addition, the amount of condiment present (*i.e.* carrier:condiment weight ratio) might influence the magnitude of the effect. Decreasing carrier:condiment weight ratio by increasing the amount of condiment is expected to facilitate bolus formation leading to an increase in eating rate. When carrier:condiment weight ratio is large, eating behavior of the composite food is expected to be similar to eating behavior of the carrier. In other studies, fluids were added to crackers, and a higher volume of water (10 vs. 5 ml) reduced the number of chewing cycles until swallowing (Pereira *et al.* 2007). This suggests that bolus formation facilitation by condiments depends on the carrier:condiment ratio.

Composite foods do not only cover carrier-condiment combinations, but one can also think of other examples such as bread with chocolate sprinkles (solid with solid), yogurt with granola (semi-solid with solid), yogurt with syrup (semi-solid with liquid) or bouillon with croutons or vermicelli (liquid with solids). This raises the question whether findings from this thesis would be generalizable among a broad range of composite foods with very different characteristics. This thesis demonstrated that solid carrier foods drove eating behavior likely because these required more (extensive) structure breakdown, whereas the condiments tuned eating behavior to a smaller extent. For other composite foods, it is speculated that the food requiring the highest degree of structural breakdown drives eating behavior of composite foods. Indeed, recent findings presented that solid peach pieces regulated the mastication of yogurt with peach to a large extent (Aguayo-Mendoza *et al.*). In summary, although one food may drive eating behavior, other accompanying foods will adjust eating behavior depending on their specific properties.

This thesis highlights that, for composite foods, properties of single foods can be changed to achieve either a faster or slower eating rate. Approaches to increase eating rate and food intake are required to increase healthy food consumption and/or to contribute to healthy ageing. On the other hand approaches to decrease eating rate and intake are required to contribute to prevention of overweight and related chronic diseases among the general population. To increase eating rate and subsequent intake, this thesis on composite foods highlights that one can consider to soften the carrier food (**Chapters 2, 6**), to increase carrier dimensions (**Chapter 4**), to change carrier shape (**Chapter 5**) or to use a condiment with a low viscosity (**Chapters 2-4**). Such mechanistic knowledge is extremely valuable when targeting the elderly population. Vice versa, to reduce eating rate and subsequent intake, one can consider to increase carrier hardness (**Chapters 2, 6**), to decrease carrier serving size (**Chapter 4**), to change carrier shape (**Chapter 5**), to remove the condiment (**Chapters 2-4**) or to increase condiment viscosity (**Chapters 3, 4**). These practical guidelines to modify eating rate and intake of composite foods are summarized in Figure 8.5.

The role of single food properties in sensory perception of composite foods

Sensory characteristics of foods are important determinants for consumer acceptance: if food does not taste good, it is not accepted and not eaten. Foods are rarely consumed alone, and interactions with accompanying foods provoke a significant change in sensory perception.

For composite foods, consumer attention may shift from one food to another within one bite and back, and Temporal Dominance of Sensations was used to reveal which foods dominate sensory perception at which stages of consumption (**Chapter 6**). Dynamic texture perception of carrier-condiment combinations was dominated by solid carrier foods, in particular at the beginning and end of a bite. Solid foods require substantial oral breakdown before they can be swallowed safely, which leads to continuous changes in food structure thereby driving dynamic texture perception of carrier-condiment combinations. Similar trends were found for yogurt with granola, in which texture perception was mainly dominated by granola characteristics such as crunchy and sticky (van Bommel *et al.* 2019). Dynamic flavor perception was dominated by the condiment rather than the carrier (**Chapter 6**). This is in line with common consumer behavior, as condiments are used to enhance the flavor of a dish in many different culinary traditions (Sikora *et al.* 2008). Taken together, product development of solid carrier foods with a bland flavor should focus on the textural properties and efforts on flavor properties could be limited. Regarding product development of condiments, focus should be on the flavor properties and efforts on the texture properties could be limited. For other types of composite foods, we speculate that the food requiring the highest degree of structural breakdown will drive texture perception (when present above a certain ratio) (Aguayo-Mendoza *et al.*). Speculation about dynamic flavor perception in composite foods

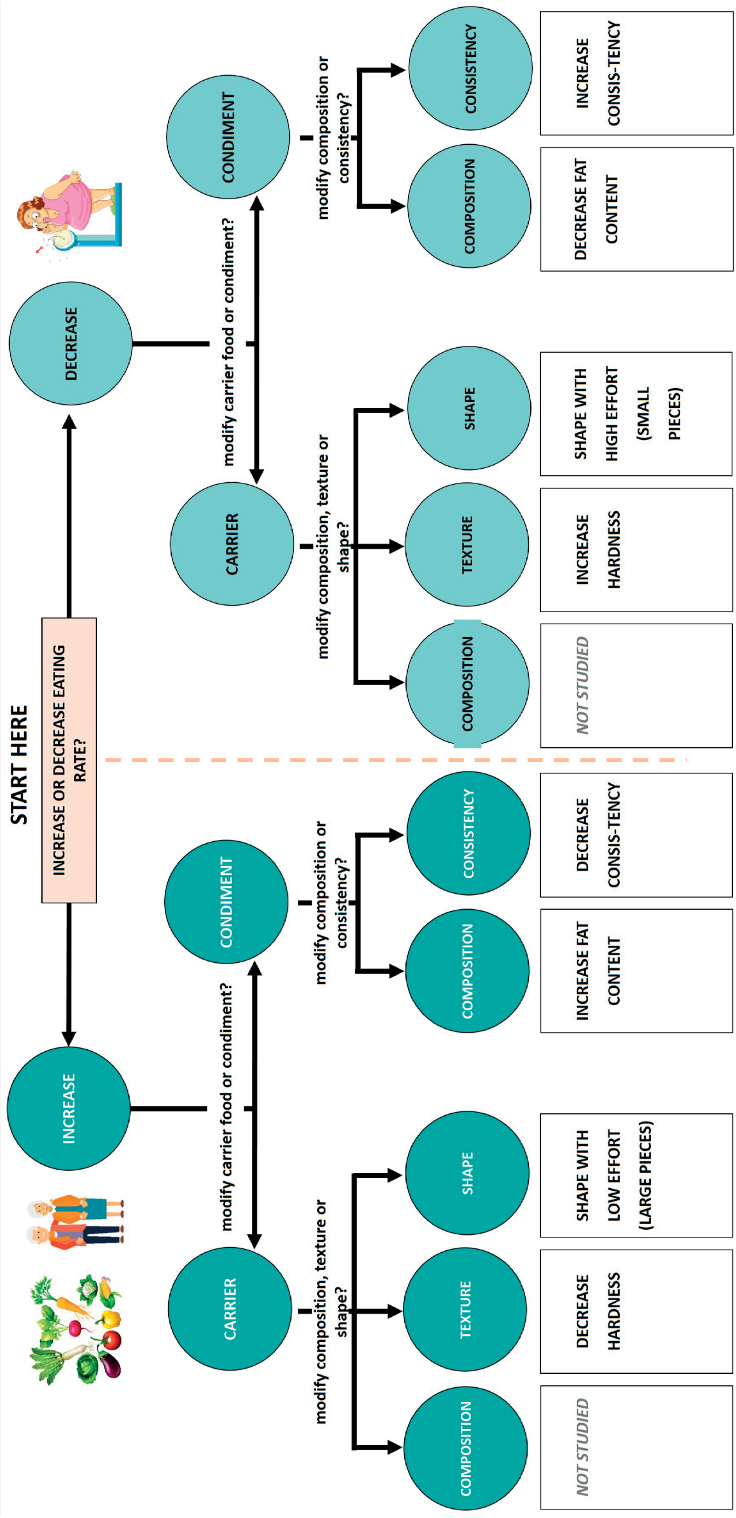


Figure 8.5: Decision chart showing how eating rate of carrier-condiment combinations can be increased (left side) or decreased (right side) by changing single food properties.

is more challenging, as flavor intensities differ substantially within food categories (*e.g.* vegetable bouillon vs. curry soup or watermelon vs. durian fruit).

In everyday life, consumers already combine certain foods to influence sensory perception. For example, condiments are added as a strategy to increase intake of vegetables or other unfamiliar foods among children (Pliner & Stallberg-White 2000; Cichero 2017). The results of this thesis show that both texture perception and flavor perception are changed when single foods are combined into composite foods. In general, perceived sensory intensity of a food decreases when an accompanying food is added (**Chapters 6, 7**) (Meinert *et al.* 2011; Paulsen *et al.* 2012; Cherdchu & Chambers 2014). Both physicochemical and cognitive mechanisms have been suggested to explain this observation. From a physicochemical view, foods might interact in mouth and thereby reduce flavor release, leading to suppressed perception. Also a dilution effect might occur, as the concentration of flavor compounds from a single food is reduced by the addition of another food, thereby decreasing the intensity of its characteristic flavors (Kroll & Pilgrim 1961). From a cognitive view the presence of accompanying foods might distract consumers' attention away from the product of interest, leading to suppressed perception. In this context, **Chapter 7** shows that cognitive effects play a key role in sensory perception of composite foods since the presence of an accompanying food increased the delivery of aroma compounds into the nasal cavity, but did result in lower perceived sensory intensity). Thus, single foods' flavor perception becomes less intense in the presence of accompanying foods, which can be desired in case of less-liked foods. However, the question whether the addition of accompanying foods and such a decrease in flavor perception impacts consumers' liking and consumers' actual eating behavior remains to be answered.

Assessing sensory perception of foods is important in product design. Sensory evaluations are generally performed with single foods, also when it concerns foods that are rarely consumed alone. For example, in case of mayonnaise, a spoon of mayonnaise rather than a mayonnaise-food combination is usually assessed during sensory evaluations. This questions the relevance of assessing sensory properties of single foods when the foods are commonly consumed with other foods. This raises the question how foods that are rarely consumed on its own such as dressings, sauces, condiments, spreads etc. should be assessed. This thesis demonstrates that sensory characteristics of mayonnaise are influenced by the presence of carrier foods and specific product properties such as carrier hardness (**Chapters 2-3, 6-7**). In addition, a range of mayonnaises was perceived to be different when consumed by themselves, but these differences were not always perceived when combined with a carrier food such as bread or vegetables (**Chapter 3**). Thus, sensory analyses of the single food itself tell only part of the story and might even be misleading. Although assessing composite foods rather than single foods would be more realistic, this may be unpractical considering the large range of possible food combinations to be assessed, especially when also taking into account that different food combinations are used around the globe. This is also an extremely time-consuming and expensive approach. Therefore, the recommendation with respect to sensory assessment of single foods that are rarely consumed on their own: perform discrimination tests within a range of commonly used accompanying foods to validate whether the new reformulated product is distinguished from the current or competitor product. The type of carrier food can differ between countries or consumer groups. As a consequence, this means that multiple food-food combinations might have to be assessed depending on the frequent consumption context of the targeted consumer group. If differences are clearly perceived, one can decide to perform rapid sensory methodologies to obtain additional information about the sensory interactions within composite foods.

Nowadays, sensory analysis is usually used to match new products to current market products and/or competitor products. However, whether the new product should exactly match the reference product could be challenged. As discussed above, the perceived sensory intensity of foods decreases when accompanying foods are added. Furthermore, this thesis demonstrates that consumer sensitivity to discriminate between condiments declined when they were assessed together with a carrier food (**Chapter 6**). Thus, although sensory profiles of condiments could be discriminated when consumed by themselves, such differences were not always perceived when combined with a carrier food. This indicates that the presence of accompanying foods might distract consumer attention away from the product of interest, which might allow more flexibility in product development. This can be of particular interest in the design of health-promoting foods (*e.g.* low in calories, reduced fat, reduced sugar, reduced salt, increased protein, gluten-free, etc.) and environmentally friendly foods (*e.g.* plant-based meat replacers, insects, etc.). The production of such foods is nowadays technically feasible, but assuring excellent sensory quality still poses a challenge. Considering frequently used accompanying foods of such healthy and/or sustainable foods might allow to develop healthier, more sustainable food combinations that are well-liked by consumers in a more affordable way.

8.2 Methodological considerations

Studying composite foods is more representative of a daily-life consumption context than studying single foods. In this thesis, we made a first attempt to understand eating behavior and sensory perception of composite foods by combining one solid carrier with one condiment within one bite. Throughout the thesis, distinctly different carriers have been tested to be able to generalize results across different composite food categories. Foods with a relatively low moisture content (bread, crackers) and those with a relatively high moisture content (potatoes, carrots) have been tested, as they were assumed to require different oral processing strategies before swallowing. In this thesis, mayonnaise has been used as a condiment in five out of six studies, as product properties such as fat content and viscosity were relatively easy to vary. For the *ad libitum* intake study (**Chapter 5**), cheese dips were used instead of mayonnaise, as Dutch consumers combine crackers more frequently with cheese than mayonnaise.

As perception is a dynamic process, perceptional changes upon consumption of multiple bites might occur so that single bite assessments may deviate from evaluations performed on complete meals. Recently, another study indicated that relatively similar dynamic sensory profiles were obtained by single bite vs. multiple bite assessments, but indeed multiple bite assessments allowed to capture a buildup of certain sensations such as sticky or fatty coating throughout the eating episode (van Bommel *et al.* 2019). As this thesis was the first attempt to study eating behavior and sensory perception in composite foods, single bite assessments were used for practical reasons.

Also other factors, such as inter-individual and environmental factors, have been shown to influence eating behavior and perception (**Chapter 1, section 1.6**). As this thesis focused on the role of food properties, individual and environmental aspects were kept constant to minimize their impact on the parameters of interest. Consequently, all studies were performed in sensory laboratories with healthy young, European, Caucasian adults to reduce subject variation. This raises the question whether results can be generalized among the world population. Addition of condiments is expected to facilitate bolus formation of foods regardless of consumer characteristics. However, the magnitude of the effect might be different, as elderly people might benefit more from condiment addition due to decreased eating capability (*e.g.* changes in salivary flow and composition, tooth loss and/or reduced jaw muscle strength) (Fontijn-Tekamp *et al.* 2004; Ikebe *et al.* 2012; Vandenberghe-Descamps *et al.* 2016). Similarly, as elderly have decreased taste capability, sensory differences perceived by our young panels might not be perceived by the elderly. As many different aspects such as culture, familiarity, expectations, and environment are known to affect consumer perception and appreciation, further speculations are challenging and require further investigation.

Throughout the thesis, we applied existing methodologies that were adjusted if necessary to determine eating behavior, bolus properties and sensory perception. Although electromyography (EMG) with or without jaw tracking (JT) has been used as the "golden standard" method for years, nowadays video recordings are more frequently used to determine eating behavior (see also **Chapters 2, 4 and 5**). The recording of videos is an easy, non-invasive method to assess how different foods affect the chewing process (*e.g*. the number of chews, chewing time, chewing frequency, eating rate). No specialized equipment and little researcher training for data collection and analysis are required, which is an advantage of using video recordings (Hennequin *et al.* 2005; Wilson *et al.* 2013). However, the disadvantage of video recording is that no information can be gathered with respect to muscle activities, chewing movements and chewing velocities. Recording of EMG or EMG-JT on the other hand, does provide opportunities to measure these parameters. However, these techniques require specialized equipment and some training. Despite the need for specialized equipment and expertise, EMG-JT methodology is extremely useful in understanding the details of the chewing process. In particular, it provides information on physiological events elicited during oral processing (Hennequin *et al.* 2005; Vinyard & Fiszman 2016). As EMG and EMG-JT are more sensitive and less subjective than video recordings, using these methods is recommended when assessing foods with subtle differences in composition or structure, like we did in **Chapter 3**.

With respect to bolus properties of composite foods, we showed that it is possible to determine the moisture content, saliva content, fat content and firmness of a broad range of composite foods (**Chapter 2-4**). Bolus cohesion is an important bolus property, but not studied very well yet. If measured, texture profile analysis (TPA) methodology is currently used, but this method comes with many limitations (Peleg 2019). This indicates the need for new methods that allow better quantification of bolus cohesion. Lubrication is another important factor in swallowing. Although methodologies are available to measure bolus lubrication properties for simple liquid foods and gel-like foods, approaches for solids and/or more heterogeneous foods are still lacking (Fuhrmann *et al.* 2020). In this thesis, it was possible to determine lubrication properties of composite foods (**Chapter 3**), but only at the moment of swallowing when boli were relatively soft and homogeneous. Finally, in the case of composite foods, other bolus properties become of relevance such as mixing behavior or fat distribution throughout the bolus. Currently, as far as we know, such methodologies are still missing and require further investigation.

Regarding sensory perception, several studies were performed with naive consumer panels (**Chapter 2, 6, 7**), showing results that were in line with data from an experienced, trained panel (**Chapter 3**). Hence, even though composite foods are sensorially more complex than single foods, consumers are able to identify differences in sensory profiles of such composite foods. Consequently, a trained panel is not necessarily required during sensory assessments of composite foods.

Finally, in **Chapter 7**, aroma release and sensory perception were determined simultaneously using PTR-MS combined with Time-Intensity methodology. The advantage of Time-Intensity is the opportunity to dynamically track the intensity perception of a flavour of interest. A well-known disadvantage of Time-Intensity is the potential occurrence of sensory dumping (Lawless & Heymann 2010), as participants can score the intensity of one specific sensory attribute only (and they are not able to report perceived changes in other sensory attributes. Therefore, combining in-nose aroma release with other dynamic sensory methodologies such as TDS or TCATA might be worth investigating, to allow participants to also report perceived changes in texture perception throughout consumption.

8.3 Future research

From single bite to meals and diets

This thesis shows that even small modifications of only one food modify eating rate, food intake and sensory perception of composite foods. A next step would involve studies investigating the role of single food properties on eating rate, intake and perception of a complete meal, *i.e.* composite foods that are composed of more than two single foods. Next, dietary interventions comparing a "fast diet" versus "slow diet" will be of interest to validate the current findings into daily life practices and public health applications. Recently, higher *ad libitum* intake (~500 kcal/day) was reported for ultra-processed diets with a high eating rate (37 g/min) compared to unprocessed diets with a low eating rate (30 g/min) (Hall *et al.* 2019). In this context, one can argue that a difference in eating rate rather than industrial food processing was responsible for the difference in intake (Forde *et al.* 2020). Therefore, clean dietary interventions investigating which food properties within the diet affect eating rate and food intake are required.

In everyday life, consumers have their own way of food consumption, which results in large differences in eating behavior and sensory perception between consumers. For instance, some people prefer to add a little bit of sauce to their dish whereas others like to add plenty of sauce. Consequently, differences in carrier:condiment weight ratios should be taken into consideration in future research on composite foods to account for differences in consumption behavior and perception between consumers. Another source of variation during consumption can be found in bite size. As observed in **Chapter 5**, differences in bite size (more-than-once-biters consuming small bites vs. only-once-biters consuming large bites) led to changes in *ad libitum* intake. Although it is known that bite size affects eating behavior and intake (Weijzen *et al.* 2009; Zijlstra *et al.* 2009; Mishra *et al.* 2012; Bolhuis *et al.* 2013; Bolhuis & Keast 2016; James *et al.* 2018), future studies are needed to investigate which single food properties impact bite sizes and subsequent intake and/or perception of composite foods.

Personalized food design?

With regard to personalized food design, more attention should be paid to inter-individual variation in eating rate (slow vs. fast eaters). Eating rate is influenced by demographics (gender, age, ethnicity); with males, young and Caucasian consumers chewing faster than females, elderly and Asian consumers (Ketel *et al.* 2019). Within a demographical group, eating rate largely depended on personal habits, leading to significant changes in bolus formation and sensory perception of single foods (Devezeaux de Lavergne *et al.* 2015a). In addition, such habitual eating strategies are consistent within individuals (*i.e.* those who eat one meal faster, also eat other meals faster) (Robinson *et al.* 2014; McCrickerd & Forde 2017) and constant over time (*i.e.* those who eat faster at young age, still eat faster several years later) (Fogel *et al.* 2018). Now, it would be relevant to understand how inter-individual variation in eating rate affects oral processing behavior, intake and sensory perception of composite foods. We hypothesize that slow eaters are more sensitive to small modifications in single food properties than fast eaters. Consequently, the suggested approach to change single food properties to modify composite food intake might be more effective in slow chewers than fast chewers, as slow eaters may be aware of changes in reformulated foods whereas fast eaters are not. Whether changing composite food properties is an efficient strategy to slow down fast eaters should still be validated.

Digestion and nutrient uptake

Food oral processing not only affects food intake and sensory perception, oral food breakdown is also the first step of digestion and of great importance for nutrient uptake (Hoebler *et al.* 1998; Hoebler 2000; Wang & Chen 2017). Physicochemical properties at the moment of swallowing, including particle size, are known to influence the digestive process. For composite foods, this thesis indicates that bolus properties at the moment of swallowing depend on single food properties. It is speculated that such differences in bolus formation have important implications for nutrient uptake. In addition, many desired micronutrients are fat-soluble, such as vitamin A, D, E and K. We speculate that the presence of fat-containing accompanying foods might therefore enhance bioaccessibility of such nutrients, as was observed for carrots combined with mayonnaise (Brown *et al.* 2004). To develop a full picture of nutrient uptake from composite foods, additional multi-disciplinary studies are needed that combine food structure, food oral processing behavior and digestion.

8.4 Main conclusions

The increasing demand for practical approaches to regulate food intake while maintaining excellent sensory characteristics represents an opportunity, yet also a challenge. The results in this thesis have shown that properties of either carrier foods or condiments can be used to alter eating behavior and sensory perception of composite foods. With respect to eating behavior, relatively small changes in properties (changing condiment viscosity, carrier hardness or carrier dimensions or shape) already have a significant effect on eating rate. Such physical changes in structure can be used to reduce overconsumption or to increase healthy food intake without having to change food formulations or nutrient composition to a large extent. We conclude that sensory perception of composite foods is complex, and sensory characteristics of one food are influenced by the specific properties of the other food present. Consumer sensitivity to discriminate between foods declined when these were assessed together with an accompanying food. This was not due to reduced delivery of aroma compounds into the nasal cavity, indicating that cognitive effects such as distraction play a role in sensory perception of composite foods. This supports the idea that both food design and cognitive factors should be used to modulate consumer perception. These findings can be of particular interest in the design of health-promoting foods.

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SUMMARY

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SUMMARY

Background and objective: Consumers frequently combine two or more foods within one bite; for example bread with spread and/or cheese, or vegetables with dressing combined into a salad. Such food pairings are called composite foods. Although consumers already combine certain foods to influence intake and/or sensory perception, composite foods have received surprisingly little scientific attention in the field of sensory science and eating behavior. The research described in this thesis aims to investigate the effects of food properties on food oral processing behavior, food intake and sensory perception of composite foods. This thesis provides new insights into how structural transitions of foods contribute to intake and/or perception of composite foods.

Methods: Composite foods were prepared by combining one condiment (*i.e.* mayonnaise or cheese) together with one solid carrier food (*i.e.* bread, cracker, carrot or potato). Single food properties including composition (condiment fat content), texture (condiment viscosity, carrier hardness) or serving dimensions (carrier shape) were systematically varied. The subjects that participated in the studies were healthy, young, European, Caucasian adults. A broad range of methodologies is used throughout the thesis. Eating behavior (chewing time, number of chews, eating rate, muscle activity, jaw movements) was examined using video recordings and/or electromyography combined with jaw tracking. Various bolus properties were characterized at certain time points throughout mastication (33, 66 and 100% of total mastication time), including saliva incorporation, mechanical properties, lubrication properties, particle size distribution and bolus retention. *Ad libitum* intake and appetite ratings were determined as measures of satiation. Sensory perception was assessed using dynamic (Temporal Dominance of Sensation, Progressive Profiling, Time-Intensity) and static methods (Descriptive Analysis, Rate-All-That-Apply). In-nose aroma release was determined using Proton Transfer Reaction-Mass Spectrometry (PTR-MS).

Results: Addition of condiments facilitated bolus formation of carrier foods leading to an increased eating rate (**Chapters 2-4**). Condiments assisted saliva in bolus formation of carriers by decreasing bolus firmness, increasing lubrication and/or increasing cohesiveness (**Chapter 3**). Changing condiment fat content had a minor effect on eating rate of composite foods, whereas substantial differences in eating rate were observed for condiment viscosity, carrier hardness and carrier shape. Increasing condiment viscosity (from salad dressing to mayonnaise) decreased the eating rate by 13-16% (**Chapter 3**). Increasing carrier hardness (from bread to crackers) decreased the eating rate by (same comment as above) 29-33% (**Chapter 2**). In case of carrier shape, cutting carrots differently (from cube to julienne) decreased the eating rate by 17% (**Chapter 4**). In addition, varying cracker shape affected dipping behavior and *ad libitum* intake of cheese dips. A larger amount of cheese was scooped with flat squared crackers than with finger-shape crackers (2.9±0.2 vs. 2.3±0.1 g) increasing the cheese intake by 15% (**Chapter 5**).

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Sensory perception of composite foods depends on the sensory characteristics of both single foods present. Dynamic sensory profiles were dominated by the texture of the carrier foods, especially in the beginning and the end of consumption, whereas condiments mainly contributed to flavor sensations perceived in the middle of consumption (**Chapter 6**). Combining single foods into composite foods decreased the perceived sensory intensity of single foods; and reduced consumer sensitivity to discriminate between single foods with small differences (**Chapter 6**). In addition, such decrease in sensory intensity perception was not due to a lower delivery of aroma compounds into the nasal cavity, as in-nose aroma release of condiments increased with the presence of a carrier food (**Chapter 7**). Consequently, cognitive effects are likely to play a key role in sensory perception of composite foods.

Conclusions: Oral processing behavior, intake and sensory perception of composite foods depend on the properties of both single foods present. Eating behavior of composite foods can be controlled by relatively small changes in either carrier or condiment properties. To increase eating rate, we suggest softening the carrier food, increasing carrier dimensions (one larger piece instead of many smaller pieces), changing carrier shape and/or adding a condiment with a low viscosity. Vice versa, to decrease eating rate, we suggest increasing carrier hardness, decreasing carrier dimensions size, changing carrier shape, or increasing condiment viscosity. Sensory perception of composite foods is complex, and interactions between single foods in mouth imply a significant change in sensory perception. Consumer sensitivity to discriminate between foods reduced when they were assessed together with an accompanying food. This can be of particular interest in the design of healthy and/or environmentally friendly foods, in which assuring excellent sensory quality still poses a challenge.

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ACKNOWLEDGEMENTS

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ACKNOWLEDGEMENTS

Good company in a journey makes the way seem shorter
Izaak Walton

I am grateful for this PhD journey, and all the ways it has shaped me. As I reflect on my personal PhD journey, I feel extremely supported by my colleagues, family members and friends who all played a significant role over the past years. Good company does not only make the way seem shorter, it also provides help, joy and security to the journey. It is therefore a great honor to acknowledge the good company that surrounded me during my PhD journey.

Vincenzo, Markus and Elke, I cannot imagine a better supervision team! Your research expertise and personal characteristics perfectly complemented each other; thank you so much for your guidance, trust and (always and ever) support. **Vincenzo**, I still remember one of our first conversations in which we speculated that our different working styles (*you: sauce chef, me: dessert chef*) would go well together. Now, four years later, I would like to accept our hypothesis, as working together has been a true pleasure. Thanks for your helicopter view, humor and our walk+coffee meetings. **Markus**, you have been a true mentor, always in for a chat, and full of priceless advise. Thanks for teaching me how to convince, persuade and which battles to choose. Whatever happened, you were there, always willing to help and you made sure that everything was fine in the end. Meanwhile, your WhatsApp stickers & GIFs always made me smile. Thank you, **Elke**, for always making time to discuss next research steps, to answer my questions and to check my manuscripts, presentations or other output. My manuscripts would not have been as they are without your thorough feedback. Thanks for providing your apartment during the FPH potluck dinners; it was a joy to explore so many different cuisines (and drinks) over the last four years.

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I am particularly grateful for the collaborations with **Chris & Erin, Michele and Dieuwerke**, since your contribution truly helped to broaden the scope of my PhD research. **Chris & Erin**, I am thankful that you came all the way from Ohio, USA to help me with the EMG-JT analyses and provided me with a crash course physiology plus data analysis. I loved seeing you participate in the Dutch culture, by taking the bike to the university and eating Dutch pancakes. **Michele**, from studying together to writing a paper together - who would ever have guessed that… I am glad that I got to know you better from both a professional and private perspective. Thanks for all your hard work, your cheering voice recordings and all the acro-yoga fun. **Dieuwerke**, waar ons contact begon met fijne gesprekken tijdens FQD-activiteiten, wandelingetjes of het halen van goede koffietjes, mocht dit (als kers op de taart) resulteren in het uitvoeren van een gezamenlijke studie. Bedankt voor je openheid, je vertrouwen en je zorgzaamheid – super waardevol. Also many thanks to the PhD students (**Eva, Marco, Monica, Philipp, Roelien**), other supervisors (**Betina, Gerry, Guido, Rene**) and

project partners (**Amelie, Benoit, Cecile, Marcel, Martijn, Michel, Stefanie, Veronica, Yves**) that were part of the *Smooth Bite For All* project. It was a pleasure to work together; thanks for the inspiring discussions, exchange of knowledge and fruitful collaborations.

I would like to gratefully acknowledge the MSc students I supervised: **Niels, Niki, Christien, Carla, Adelina, Rutger, Arpavee, Desiree, Robin, Sophie, Anouk** and **Dimitra**. We had lots of fun during too many sample preparations, video analyses, bolus samples or because of funny participants (Have you ever watched videos of participants dancing while tasting? *We did!* Have you ever seen participants stealing samples from their neighbor participant? *We did!*). I could not have performed all the work without your committed help; thanks to you all.

Many thanks to everyone within the Food Quality and Design, Food Physics and Human Nutrition groups. Food Physics, thanks for adopting me – I have always felt very welcome. **Corine, Kimberley** en **Lysanne** (FQD) en **Els** (FPH), jullie zijn de onzichtbare helden achter ieders PhD project! Geen enkele vraag was jullie te gek, een oplossing werd door jullie altijd snel gevonden of gecreëerd. **Charlotte, Erik, Frans, Geert, Harry, Mike, Miranda** en **Xandra**, bedankt voor al jullie technisch advies en hulp bij mijn onmogelijke praktische vragen (Is er plek voor 500 potten mayonaise? Is er daarnaast ook nog plek voor 300 kg kaas? Hoe krijgen we de uitgespuugde samples zo snel mogelijk geanalyseerd in het lab?). Thanks to my FPH office mates **Belinda, Claudine, Luka and Philipp**, we talked thousands of nothings while they meant millions of somethings to me. A special thanks to my FQD "flex" office mates **Annelies, Femke & Jonna** (also known as the FQD Dutchies). Hoewel menigeen (lees: Vincenzo) denkt dat onze dagen gevuld zijn met eten, kletsen, meer eten en meer kletsen, waren onze gezamenlijke office uurtjes verrassend productief. Ik wil jullie in het bijzonder bedanken voor jullie luisterende oortjes en onvoorwaardelijke hulp. **Marlou,** onze thuiswerk dagen in Utrechtse koffietentjes waren een welkome afleiding en bron van inspiratie. **Alex** en **Steven**, goede koffie gecombineerd met jullie openheid, humor en sarcasme is een perfecte combinatie, waarmee jullie altijd weer een brede lach op mijn gezicht toverden!

Met veel liefde wil ik mijn familie, huisgenootjes en vrienden bedanken. **Papa** en **mama**, wie had dat gedacht, dat jullie kleine meisje, die in Wageningen ging studeren om maar GEEN onderzoeker te hoeven worden, nu haar PhD afrondt. Bedankt voor jullie onvoorwaardelijke liefde - jullie zullen altijd in mij geloven, me steunen, voor me klaarstaan & trots op me zijn! **Opa & oma** en **oma**, van iedereen zijn jullie ongetwijfeld het meest blij en opgelucht als ik na een conferentie of reis weer veilig terug in Nederland ben. Het maakt voor jullie niet uit wat ik precies doe, jullie zullen altijd enorm trots zijn. **Jorinke, Manon** en **Sanne&Bas**, bedankt dat ik altijd bij jullie mocht binnenstormen voor een luisterend oor of een dikke knuffel. **Annelies, Bart, Esther, Eva, Femke, Frederike, Gerben, Gerline, Heidi, Hella, Jeroen, Jonna, Kelli, Kim, Kristel, Marit, Miriam, Roelien, Sarah, Silke, Simon, Thelma en Tom,** de mooiste

herinneringen maak je met de mensen dichtbij je! Dankjulliewel voor alle heerlijke PhD afleidingen (etentjes, koffietjes, taartjes, biertjes, wijntjes, oesters, whisky'tjes, feestjes, klimmen, acro-yoga, andere sporten, reisjes en veel meer) – zo veel gedeelde herinneringen die ik in mijn hart koester! **Gerben,** dankjewel voor alle koffietjes (veel te vroeg in de ochtend), gedeelde avonturen en de vrijheid die je me hebt gegeven om me te kunnen ontwikkelen tot wie ik nu ben. **Hannah**, heel erg bedankt voor je creativiteit in het ontwerpen van de cover – het is super mooi geworden. **Frederike**, fijn om jou als vriendin te hebben – bijzonder hoe onze vriendschap de afgelopen jaren heeft mogen groeien. Bedankt voor je vertrouwen, je vrolijke spraakberichtjes & je enorme gastvrijheid in het prachtige Zwitserland (als ik weer eens een ontspannings- of schrijfreisje zocht). **Kim**, dankjewel dat ik alles met je kan delen en dat je altijd voor me klaar zult staan (hoe ver we soms ook fysiek bij elkaar vandaan zijn). In het bijzonder dankjewel voor onze bijzondere tijd in Suriname. Hier bleek dat we naast goede vriendinnetjes, ook een zeer efficiënt schrijf duo zijn (al zou Tom ons waarschijnlijk eerder omschrijven als een koffieverslaafd duo haha). **Simon**, ik ben dankbaar dat onze paden mochten kruisen. Dankjewel voor alle "vooral voor je eigen plezier" support tijdens het afronden van mijn PhD!

Tot slot, **Roelien en Annelies**, ik ben super blij om jullie als mijn paranimfen (en collega's en goede vriendinnen) aan mijn zijde te hebben. Lieve **Roelien**, ruim vier jaar geleden begonnen we allebei aan onze PhD. Ik ben dankbaar dat ik jou hier in Wageningen heb mogen leren kennen. Het delen van de sensorische keuken verliep altijd soepel en de conferentie reisjes naar Edinburgh, Nottingham & Verona waren meer dan gezellig. In onze vrije tijd gingen we samen sporten, kerstdinertjes eten, zeilen of naar surf festivalletjes, op avontuur in een mistig Duitsland en zo veel meer. Dankjewel voor alles! Lieve **Annelies**, onze vriendschap ontstond tijdens onze MSc in het prachtige Kopenhagen. Tegenwoordig gaat er vrijwel geen dag voorbij waarin wij elkaar niet spreken (soms op meerdere kanalen door elkaar heen haha). Jouw steun tijdens het afronden van mijn PhD was super waardevol; dankjewel voor je kritische blik, het meedenken en meelezen. Daarnaast ben ik ongelooflijk dankbaar voor onze vriendschap, en in het bijzonder voor jouw grote hart waarmee je ALTIJD voor me klaar staat!

I cherish all of you and these shared, most special moments in my heart and memory!

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ABOUT THE AUTHOR

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Arianne van Eck was born on the 30th of March, 1992 in Veenendaal, the Netherlands. In 2010, she enrolled in the BSc program Human Nutrition and Health at Wageningen University. As part of her BSc, she took courses on communication and neurosciences at VU Amsterdam to broaden her knowledge. For her BSc thesis, she wrote a literature review on the effect of adherence to the Mediterranean diet on cognitive functioning in elderly. Arianne obtained her BSc degree in 2013, after which she enrolled in the MSc specialization Sensory Science at Wageningen University. As part of her MSc program, she followed an Erasmus semester at the university of Copenhagen, Denmark. For her MSc thesis, she investigated the effect of oil coatings on subsequent sweetness perception of liquids, which has resulted in a scientific publication. To finish her MSc degree, Arianne did a six months internship in the Snack flavor Creation Application department of Givaudan, the Netherlands. For her internship, she developed potato chips flavors, and performed sensory evaluation of these newly developed flavors by using a trained panel. She graduated in 2015. At the beginning of 2016, Arianne was appointed as a PhD candidate at TIFN and Food Quality and Design group at Wageningen University. Her PhD project was part of the public-private project called "Smooth bite for all". Her research focused on the role of single food properties in oral processing behavior, intake and sensory perception of composite foods. During her PhD project, Arianne attended various courses and she presented her work at several international conferences (nine oral presentations, three poster presentations). She was involved in teaching and supervising BSc and MSc students (twelve students). In 2019, she co-organized the 2nd Young EFFoST day in Rotterdam, the Netherlands.

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Arianne can be contacted by email: vaneckarianne@gmail.com.

OVERVIEW OF COMPLETED TRAINING ACTIVITIES

Discipline specific courses

2017	16th European school on rheology	Leuven, Belgium
	Healthy and sustainable diets: synergies an trade-offs (VLAG)	Wageningen, NL
	Advanced food analysis (VLAG)	Wageningen, NL
2016	Sensory perception & food preference: Affective drivers of food choice (VLAG)	Wageningen, NL

General courses

2019	Writing propositions (WGS)	Wageningen, NL
2017	Programming in VBA	Groningen, NL
	Introduction to R (VLAG)	Wageningen, NL
	Presenting with impact (WGS)	Wageningen, NL
	Scientific writing (WGS)	Wageningen, NL
	Scientific publishing (WGS)	Wageningen, NL
2016	Multivariate analysis for food/data sciences (VLAG)	Wageningen, NL

Optional courses and activities

2019	Organization Young EFFoST day 2019	Rotterdam, NL
2018	Spanish (beginners)	Utrecht, NL
	PhD study tour to Australia	Australia
2016	PhD study tour to Italy	Italy

LIST OF PUBLICATIONS

Publications in peer-reviewed journals

van Eck, Wijne, Fogliano, Stieger & Scholten (2019) *Shape up! How shape, size and addition of condiments influence eating behavior towards vegetables.*
Food & Function 10(9), 5739-5751.

van Eck, Fogliano, Galindo-Cuspinera, Scholten & Stieger (2019) *Adding condiments to foods: How does static and dynamic sensory perception change when bread and carrots are consumed with mayonnaise?*
Food Quality and Preference 73, 154-170.

van Eck, Hardeman, Karatza, Fogliano, Scholten & Stieger (2019) *Oral processing behavior and dynamic sensory perception of composite foods: Toppings assist saliva in bolus formation.*
Food Quality and Preference 71, 497-509.

Camacho, **van Eck**, van de Velde & Stieger (2015) *Formation dynamics of oral oil coatings and their effect on subsequent sweetness perception of liquid stimuli.*
J Agric Food Chem 63(36), 8025-8030.

Submitted for publication

van Eck, Franks, Vinyard, Galindo-Cuspinera, Fogliano, Stieger, Scholten: *Sauce it up: Capturing the influence of condiment properties on eating behavior, bolus formation and sensory perception of solid foods.*

van Eck, van Stratum, Achlada, Goldschmidt, Scholten, Fogliano, Stieger, Bolhuis: *Shape matters: How cracker shape modifies ad libitum snack intake of crackers with cheese dip.*

van Eck, Pedrotti, Brouwer, Supapong, Fogliano, Scholten, Biasioli, Stieger: *What drives in vivo aroma release and perception of condiments consumed with carrier foods?*

Conference presentations

2019 33rd EFFoST International Conference (Rotterdam, Netherlands)
oral presentation

13th Pangborn Sensory Science Symposium (Edinburgh, United Kingdom)
oral presentation, flash poster presentation

19th ICC conference (Vienna, Austria)
oral presentation (invited speaker)

2018 32nd EFFoST International Conference (Nantes, France)
two oral presentations

8th European Conference on Sensory and Consumer Research (EuroSense) (Verona, Italy)
oral presentation, poster presentation

5th International Conference on Food Oral Processing (Nottingham, United Kingdom)
two oral presentations

3rd Food Structure and Functionality Forum Symposium (Montreal, Canada)
oral presentation, poster presentation

2016 4th International Conference on Food Oral Processing (Lausanne, Switzerland)
poster presentation

COLOPHON

The research described in this thesis was performed within the framework of TiFN.
Financial support from Wageningen University and TiFN for this research and for printing this thesis is gratefully acknowledged.


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