Preference and satiety

Short- and long-term studies on food acceptance, appetite control and food intake

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Stellingen

1. Bij volwassenen is het energie-gehalte van voedingsmiddelen niet van invloed op de ontwikkeling van smaakvoorkeuren (*dit proefschrift*).

2. Volwassenen leren niet te compenseren voor verschillen in energie-inhoud van voedingsmiddelen (*dit proefschrift*).

3. Hedonische responsies verkregen in het laboratorium met de 'taste-and-spit' test overschatten de optimale concentraties van smaakcomponenten in voedingsmiddelen (*o.a. dit proefschrift*).

4. Liking is a dynamic process (*o.a. dit proefschrift*).

5. You eat what you like, but come to like what you eat (*dit proefschrift; PJ Rogers & DJ Mela in 'Your food: Whose choice?', 1992, Chapter 2, pp. 26-36*).

6. Waarom iets lekker is, is moeilijker aan te geven dan waarom iets niet lekker is.

7. Een bevinding in lijn van de verwachting is makkelijker te publiceren dan een bevinding die hier lijnrecht tegenover staat.

8. Het verstandigst is hij die weet wat hij niet weet (Socrates, 470-399 voor Christus).

9. Veel GSM-gebruikers zijn niet op de hoogte van het spreekwoord: '*Spreken is zilver, zwijgen is goud*'.

10. Competitie volleybal is apart voor mannen en vrouwen. Dat dit ook bij bridge het geval is, is opmerkelijk.

Stellingen behorende bij het proefschrift

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Liesbeth H. Zandstra. Wageningen, 26 april 2000.

Aan mijn ouders

Abstract

Preference and satiety. Short- and long-term studies on food acceptance, appetite control and food intake.

Ph.D.-thesis by Liesbeth H. Zandstra, Division of Human Nutrition & Epidemiology, Wageningen University, Wageningen, the Netherlands. April 26, 2000.

This thesis describes experiments studying the impact of nutritionally modified foods on food acceptance and appetite control. The major outcomes of the studies relate to (1) predictive validity of laboratory sensory tests on food consumption, (2) effects of macronutrient and energy content manipulations on long-term food acceptance and appetite control, and (3) effects of repeated food consumption, variety and changes in pleasantness on long-term food acceptance and food intake. On the basis of the studies in this thesis we conclude that laboratory hedonic ratings collected after the taste-and-spit test tend to overestimate the optimal preferred concentration of taste substances in foods. Also, adults do not readily learn or express compensation for a reduced-energy food after repeated experience with the food under realistic eating conditions. Finally, repeated exposure to foods can lead to changes in liking over time. Liking can either increase or decrease following repeated consumption. Whether liking increases or decreases with exposure depends on the sensory properties of foods, the type of food product, the availability of different varieties of foods, and the context in which the foods are consumed. This implies that reduced-energy foods with initial good acceptance may, over time, sustain or even increase in their acceptance. The development of palatable reduced-energy foods should therefore be encouraged in order to increase their initial acceptance and, as a consequence, their use and acceptance on the longer-term.

Contents

1

General introduction

Introduction

In the Netherlands as well as in other industrialised countries, there is still a discrepancy between dietary recommendations and actual food consumption. Most people have a higher intake of (saturated) fat and a lower fruit and vegetable consumption than recommended, which is of concern given their links with cardiovascular diseases (Lichtenstein *et al.*, 1998; Kushi *et al.*, 1985), obesity (Bolton & Woodward, 1994; Seidell, 1995) and cancer (Potter, 1996; WCRF, 1997). In recent years governmental and public health organizations have therefore actively promoted dietary recommendations, for example to increase consumption of fruit and vegetables and to reduce fat intake (Dutch Nutrition Council, 1986; Riedstra *et al.*, 1993; Ministerie van VWS, 1998). Although there have been changes in total fat intake since the publication of the official recommendations in 1986 (Dutch Nutrition Council, 1986), further changes are needed to meet recommendations.

Over the period 1988-1998 the mean percentage of energy derived from fat declined in parallel with total energy from 40% to 36%, which is only slightly higher than the recommended upper level of 35% of energy intake (Voedingscentrum, 1998). However, the mean percentage of energy derived from saturated fat remained relatively unchanged at 16% and 14% (range 12-15%) in 1988 and 1998 respectively, still well above the recommended 10% of energy intake (Voedingscentrum, 1998). Furthermore, the mean fruit and vegetable consumption declined by 10% over the period 1988-1998 to a mean consumption of 1 piece of fruit and 90-130 g of vegatables per day, which does not meet the recommended consumption of two pieces of fruit and 150-200 g of vegetables per day (Voedingscentrum, 1998).

In order to more effectively stimulate people to change their diet towards current dietary recommendations, a better understanding of why and in what way people choose their foods is needed. This understanding is of importance for both health professionals and product marketeers. Health professionals need insight into factors determining food choice and dietary patterns for the development of more effective nutrition education programmes. For product marketeers a better understanding in food choice processes is needed to develop appropriate strategies in marketing and advertising, and to develop modified or new food products which meet the preferences and dietary needs of consumers.

Food choice

Food choices, and hence nutrition behaviour, are determined by a large range of potential factors. Shepherd (1985) developed a model to illustrate the different factors *influencing consumers' food choice and food intake*. This framework includes three dimensions related to (1) the food (e.g. energy content), (2) the person making the food choice (e.g. experience, beliefs), and (3) the external economic and social environment in which the choice is made (e.g. price, availability). Two main factors related to food choice are the sensory perception of physical-chemical properties of foods and the physiological effects of foods (Shepherd, 1988). These two factors are the central focus of this thesis: preference, the individual's hedonic evaluation of sensory perception of foods, and satiety, a state and/or process related to the physiological effects of foods. The learned association between preference and satiety is of particular interest.

Mechanisms of acquisition of food preferences and aversions

Several researchers suggested that (dis)likes of foods can be formed through a learned association of the contexts and physiological consequences of ingesting foods with the sensory characteristics of those foods (Pelchat & Rozin, 1982; Kern *et al.*, 1993; Mela, 1995; Zellner *et al.*, 1983; Booth *et al.*, 1982). The clearest example of this type of learning is the conditioned taste aversion, which results when ingestion of a food is followed by a negative postingestive consequence, such as nausea or illness. This learned taste aversion usually requires only one exposure to the food, and, as a result, the food may be avoided for many years (Pelchat & Rozin, 1982). In a similar but probably more subtle way, preferences for foods may be acquired when associations are formed between the sensory characteristics and positive postingestive consequences of foods, such as those after consuming high energy foods (Kern *et al.*, 1993; Booth *et al.*, 1982). The latter association provides a mechanism by which people develop likes for energy-dense foods. Figure 1.1 shows a schematic representation of relevant factors influencing the acquisition of preferences.

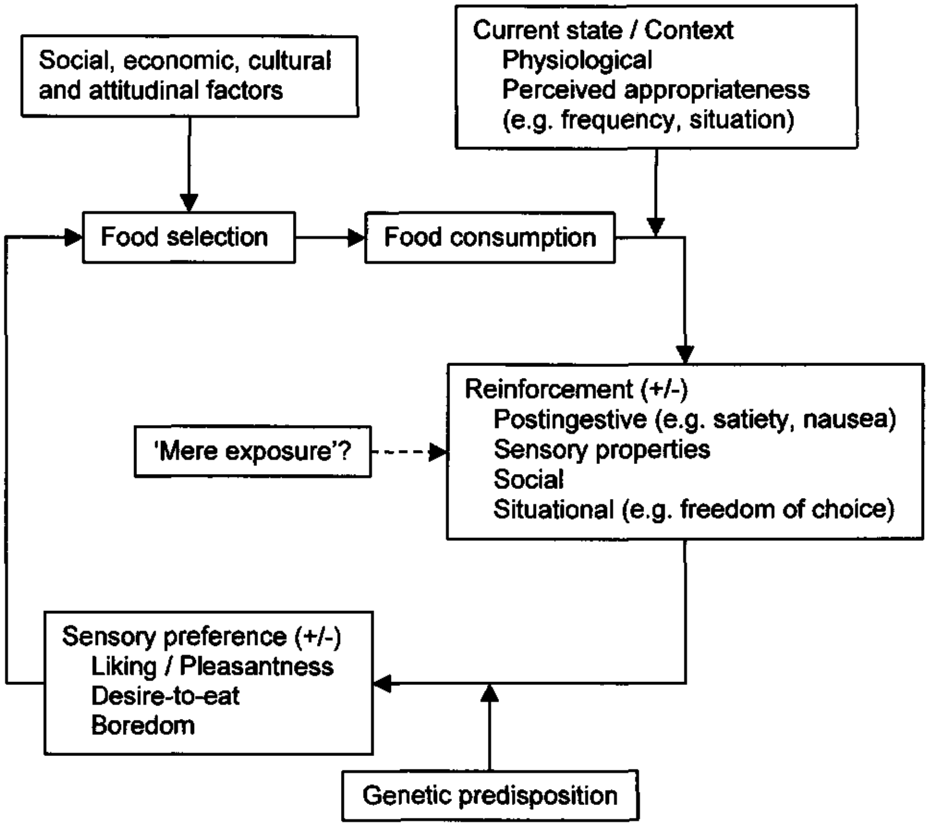

Figure 1.1 Schematic representation of factors influencing the acquisition of sensory preferences, based on Mela (1995).

Possible implications of acquisition of food preferences on food acceptance and food intake

The associative learning processes have potentially important implications for the long-term use and acceptance of reduced-energy foods. Reduced-energy foods should theoretically enable a lower consumption of energy and/or fat and sugar in the diet, in line with current dietary recommendations. The use of reduced-energy foods might therefore assist in managing weight control and the prevention of obesity and nutrition related diseases. The latter is of particular importance given the strong increases in the prevalence of obesity over the last 15 years (Seidell, 1995).

Reduced-energy foods usually have a reduced fat and/or sugar content and contain substances that mimic the sensory attributes of fat and carbohydrates. From this point of view, long-term acceptance of reduced-energy foods may decrease over time, since the rewarding properties (e.g. via satiety or metabolic effects) of these foods may be lower than their higher energy counterparts. As a consequence, despite initially high acceptance, consumers might be less likely to continue to like and use these reduced-energy foods over longer periods of time. The short-term regulation of food intake might also be affected by acquired associations between energy content of foods and their sensory characteristics (Shaffer & Tepper, 1994; Booth *et al.*, 1982; Booth *et al.*, 1976). The effectiveness of the use of reduced-energy foods over longer periods of time will depend on the precision of the food intake regulation. At present, the understanding of these issues is incomplete. It is therefore important to investigate the effects of energy manipulations of foods on long-term food acceptance, regulation of food intake and appetite control.

The associative learning processes may also underlie so-called 'mere exposure' effects. The mechanism of 'mere exposure' was originally proposed by Zajonc (1968), who suggested that mere exposure to a stimulus was a sufficient condition for enhancement of one's attitude toward that stimulus. This hypothesis has received support by a few food-related studies which found that mere exposure to new foods in particular leads to increased liking (Pliner, 1982; Stang, 1974; Crandall, 1984). High frequency or extended exposure to foods may, however, also lead to a decrease in liking and, as a result, a lower food acceptance (Porcherot & Issanchou, 1998; Ye & Van Raaij, 1997). To what extent repeated exposure affects food acceptance may depend on the duration and intensity of exposure, eating context, availability of different varieties of particular foods, the degree of freedom of choice, and the initial pleasantness of the foods consumed. Longer-term studies on the effects of repeated exposure, variety and changes in pleasantness on long-term food acceptability and food intake are lacking.

Aims of this thesis

This thesis was part of an EU-project, entitled 'Understanding and improving the selection and acceptance of foods for health promotion'. The overall objective of this EU-project was to enhance understanding and achievement of short- and long-term consumer acceptance of food products which can specifically contribute to meet current dietary goals. The focus was on consumer and food characteristics associated with the selection, continued acceptance, and dietary implications of a range of nutritionally modified alternatives to traditional food and ingredients. Four participants were involved in the project: Institute of Food Research, Reading, UK; University of Helsinki, Helsinki, Finland; Norwegian Food Research Institute MATFORSK, Ås, Norway; Wageningen University, Wageningen, the Netherlands.

The studies conducted in the Netherlands were a blend of short- and long-term laboratory and in-home trials, on food preferences and on short-term regulation of food intake. These studies are described in **Chapters 2-7** of this thesis (Table 1.1). In **Chapter 8** we discuss the main findings, conclusions and implications of the studies described in the preceding chapters.

This thesis has the following objectives:

1. To investigate the predictive validity of laboratory sensory tests relative to food consumption.
2. To investigate the effects of repeated food consumption, variety and changes in pleasantness on long-term food acceptance and food intake.
3. To investigate the effects of macronutrient and energy content manipulations on long-term food acceptance and appetite control.

Table 1.1 Summary of setting, period and focus of the studies described in this thesis.

	Setting	Period	Focus
Chapter 2	In-lab	Short-term	Food preference
Chapter 3	In-lab	Short-term	Short-term regulation of food intake
Chapter 4	In-lab	Long-term	Food preference
Chapter 5	In-home	Long-term	Food preference
Chapter 6	In-home	Long-term	Short-term regulation of food intake
Chapter 7	In-home	Long-term	Food preference (conditioned)

Studies on food preference

It is important to distinguish between different aspects of food acceptance and selection: *preference*, *liking* or *pleasantness*, *desire-to-eat* and *boredom*. *Preference* refers to a choice of one item rather than another. Preference judgements are often a mixture of many different factors (including price, availability, beliefs (e.g. concerns about health, nutrition, body weight)). *Liking* is an affective response to foods. The statement 'I like X better than Y' is not equivalent to the statement 'I prefer X to Y'. The latter describes a choice, whereas the 'like' statement describes an experience of pleasure. It is quite possible to 'prefer' one item to another, while not liking either. In many studies, liking for the taste/flavour of food is used as the main predictor of preference (Rozin, 1989). *Desire-to-eat* refers to wanting or intending to consume a food. Desire-to-eat is influenced by general liking for a food, but also by current physiological state and situational context (time of the day, perceived appropriateness). *Boredom* refers to a decrease in liking caused by satiation with specific attributes repeatedly delivered by the consumed food and/or a decrease in perceived appropriateness.

In sensory research the perception of sensory attributes of a food is distinguished from the hedonic evaluation of that food. Examples are the perceived taste (e.g. perceived sweetness intensity), aroma, texture and appearance. The relationship between concentration and perceived intensity increases monotonically (via a non-linear psychophysical function), whereas the relationship between the perceived perception and pleasantness is typically a parabolic inverted U shaped function (pyschohedonic function) (Moskowitz *et al.*, 1974). In other words, while intensity ratings increase with rising concentration, hedonic ratings show an optimum or breakpoint at a certain concentration level.

Most tests in sensory laboratories concern hedonic ratings of small samples of food to which subjects are briefly exposed. These tests are relatively inexpensive and easy to conduct, but they differ considerably from the daily life situation in which larger food samples are eaten. The predictive validity of these laboratory sensory tests as an indicator of consumption in free living situations has therefore been questioned (Lucas & Bellisle, 1987). In **Chapter 2** we investigated the predictive validity of four different laboratory sensory tests relative to consumption. Pleasantness and perceived intensity were determined in a taste-and-spit test, a taste-and-swallow test, a fixed quantity test of 300 g and an *ad libitum* consumption test.

Several studies showed that enhancing the pleasantness of foods increases food intake within one meal (Bobroff & Kissileff, 1986; Yeomans *et al.*, 1997). However, the longer-term relationships of pleasantness with food intake are unclear. Another issue is whether or not the relationship between pleasantness and food intake remains stable over time. The longer-term effects of manipulated pleasantness on *ad libitum* food intake, liking and appetite are reported in **Chapter 4**.

Some foods and/or drinks are consumed day after day for several years (e.g. bread, cheese, coffee), whereas other foods are only eaten occasionally, even though they may be highly liked. Apparently, foods differ in their appropriateness for consumption frequency, or differ in their capacity to produce boredom. The degree of available variety and the degree of freedom of choice may have an important role in the development of boredom. The effects of repeated exposure, the availability of different varieties of foods and the degree of freedom of choice on long-term food acceptance are reported in **Chapter 5**.

A preference for sweet and aversion for sour and bitter is present at birth (Steiner, 1977). These innate preferences are not fixed but can soon be modified by learning and experience with food and eating, especially via the impact of associative learning (Rozin *et al.*, 1998; Baeyens *et al.*, 1995). The latter includes the acquisition of (dis)likes through a learned association of a food's taste with its postingestive effects (Pelchat & Rozin, 1982; Kern *et al.*, 1993; Mela, 1995; Booth *et al.*, 1982) (Figure 1.1). A few studies have shown that children are able to learn to unconsciously associate the sensory properties of foods with their postingestive consequences through repeated consumption ('conditioned flavour preferences') (Kern *et al.*, 1993; Johnson *et al.*, 1991). In these studies, manipulation of energy from either carbohydrates (Birch *et al.*, 1990) or fat (Kern *et al.*, 1993; Johnson *et al.*, 1991) in novel-flavoured yoghurts led to relative increases in preference for the flavours associated with higher energy content. There is little information about the operation of this mechanism in adults under realistic conditions. In **Chapter 7** we investigated the role of postingestive factors and development of conditioned flavour preferences in adults in an in-home setting.

Studies on satiety

The control of appetite involves two different processes, satiation and satiety. These two processes determine the size and frequency of eating episodes (Blundell & Halford, 1994; Blundell *et al.*, 1994). Satiation is the process that terminates the eating within a meal, and influences the size of meals and snacks. Satiety is the state that occurs after eating of a food or meal has ended, and inhibits further eating, which influences the frequency of meals and snacks. These two processes, therefore, control events going on within-meals or between-meals (Blundell & Rogers, 1991). The present thesis focuses on both satiation (**Chapter 2, 4**) and satiety (**Chapter 3, 4 and 6**).

A variety of experimental paradigms have been used to investigate the effect of macronutrient and energy content of foods on subsequent energy intake. For example, a number of studies have investigated the effects of macronutrients or energy on satiety using a preload - test meal paradigm. This involves a design in which individuals consume fixed amounts of foods (preloads) varying in macronutrients or energy, and the measurement of the effect of that preload on subsequent food intake. A preload - test meal design was used in **Chapter 3 and 6**. Other studies have examined the effect of energy, fat and/or carbohydrates on satiation. Satiation is assessed by providing individuals with foods varying in composition and measuring the amount of foods consumed when the foods are freely available (**Chapter 2 and 4**).

Energy compensation is the term that refers to the extent to which subsequent energy intake is adjusted in response to the energy ingested through the previously consumed food. The results of a few studies suggest that the ability to show energy compensation might be less precise in older than in young subjects (Birch & Deysher, 1985; Roberts *et al.*, 1994; Rolls *et al.*, 1995). Young children (2-5 years old) compensated over a short-term period for a high energy intake at one meal by a lower energy intake at the next meal, and *vice versa* (Birch & Deysher, 1986; Birch & Deysher, 1985). Elderly subjects showed a less precise energy compensation than young adults (Roberts *et al.*, 1994; Rolls *et al.*, 1995). Up to now, however, no experiment has been conducted in which people in different stages of their life cycle have been compared. In **Chapter 3** we investigated the effects of consuming preloads on subsequent food intake in children, and younger and older adults.

The results of Chapter 3 were limited to a single exposure to the preload. It might be that the precision of energy compensation improves following repeated exposure to the preload due to the development of learned associations between satiety and the flavour characteristics of the food (Booth *et al.*, 1982; Booth *et al.*, 1976; Specter *et al.*, 1998). The effects of repeated exposure to the preloads on subsequent food intake in adults were therefore examined in **Chapter 6**.

Finally, **Chapter 8** discusses the main findings, conclusions and implications of the studies described in this thesis.

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2

Laboratory hedonic ratings as predictors of consumption*

Elizabeth H. Zandstra, Cees de Graaf, Hans C.M. van Trijp, Wija A. van Staveren

Abstract

The objective of this study was to investigate the predictive validity of laboratory sensory tests on consumption. Thirty-six subjects (30 women, 6 men; mean age 21 years) participated in a taste-and-spit test, a taste-and-swallow test, a fixed quantity test in which an amount of 300 grams was consumed, and an *ad libitum* consumption test. Subjects rated on a 10-point category scale pleasantness and perceived sweetness intensity of yoghurts varying in sucrose concentration (1%, 5.9%, 10%, 17.6% and 30% w/w). Consumption was measured in the *ad libitum* consumption test. Results showed that the optimal sucrose concentration as determined by the taste-and-spit test (10% w/w) was higher than that determined from the taste-and-swallow test, the fixed quantity test and the *ad libitum* consumption test (5.9% w/w). The mean of within-subject's correlation coefficients between pleasantness ratings and amount of yoghurt eaten in the *ad libitum* consumption test were 0.45, 0.62, 0.75 and 0.81 for the taste-and-spit test, the taste-and-swallow test, the fixed quantity test and the *ad libitum* consumption test, respectively. The perceived sweetness intensity did not change as function of the sensory test used. We conclude that pleasantness ratings collected after the taste-and-swallow test give a better prediction of consumption than the taste-and-spit test.

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Introduction

Most tests in sensory laboratories concern hedonic ratings of small samples of food to which subjects are briefly exposed. These tests are relatively inexpensive and easy to conduct, but they differ considerably from the daily life situation in which larger portions of foods are eaten in combination with other foods and beverages. In *ad libitum* consumption tests under laboratory conditions, subjects are exposed to foods for longer periods of time assuming that such a longer exposure reflects more actual use conditions. These *ad libitum* consumption tests are, however, more time consuming and more expensive compared to brief-exposure tests. Several studies have examined the extent to which these short laboratory hedonic ratings can predict how much and how often people will consume the tested products. These studies have shown equivocal results.

Lucas & Bellisle (1987) presented yoghurt in five sucrose concentrations (from 2% w/w to 29% w/w) to 18 subjects. They reported that a higher sucrose concentration was preferred in a taste-and-spit test than in an *ad libitum* consumption test under laboratory conditions. The intake of yoghurt correlated positively with the hedonic ratings of the taste-and-spit test but less strongly so than with the *ad libitum* consumption hedonic ratings (r=0.52 vs. r=0.70). Bellisle *et al.* (1988), Pérez *et al.* (1994) and Monneuse *et al.* (1991) replicated the study of Lucas & Bellisle (1987) with, respectively, salt in mashed potatoes and sucrose and aspartame in yoghurts. The results of these three studies revealed that the optimal preferred concentration derived from the taste-and-spit test was higher than that determined from the *ad libitum* consumption test. Other studies attempted to replicate this finding but failed to confirm the discrepancies in hedonic ratings between brief-exposure and *ad libitum* testing conditions (Daillant & Issanchou, 1991; Popper *et al.*, 1989; Shepherd *et al.*, 1991).

Two possible explanations for these diverging results are the different experimental tasks that subjects had to perform during the brief-exposure test (spitting vs. swallowing) and the types of response scales used to determine the optimal preferred concentration. In 4 studies subjects had to spit out the sample during the brief-exposure test (Bellisle *et al.*, 1988; Daillant & Issanchou, 1991; Pérez *et al.*, 1994; Lucas & Bellisle, 1987), but not in three other studies (Monneuse *et al.*, 1991; Popper *et al.*, 1989; Shepherd *et al.*, 1991). Furthermore, the results of Daillant & Issanchou (1991) and Shepherd *et al.* (1991) were based on a relative-to-ideal rating scale, while the results of the other studies were based on a 9-point hedonic category scale (Monneuse *et al.*, 1991; Bellisle *et al.*, 1988;

Pérez *et al.*, 1994; Lucas & Bellisle, 1987; Popper *et al.*, 1989). However, the differences in the two methodological aspects do not concur with the differences in results. Therefore, these two explanations can not completely explain the inconsistency in results.

The present study examined sensory perceptions and preferences of sweetened yoghurts in a laboratory *ad libitum* consumption test and various types of laboratory brief-exposure tests. As brief-exposure tests a taste-and-spit test, a taste-and-swallow test and a consumption of a fixed quantity of 300 grams were used. We used a 10-point category scale, because subjects in the Netherlands are familiar with this type of rating in the Dutch school system.

Materials and methods

Subjects

Thirty-six healthy subjects (30 women and 6 men; mean age 21 years, range 18 to 27 years; mean body mass index 22 kg/m^2) were recruited by advertisements in university buildings and word of mouth. Subjects were students or staff members of Wageningen Agricultural University. Subjects were not informed about the actual purpose of the study but were told beforehand that the sensory tests consisted of tasting and eating sweetened yoghurts. Individuals suffering from diabetes mellitus were excluded from participation. The study was approved by the Medical Ethical Committee of the Division of Human Nutrition & Epidemiology.

Stimuli

The stimuli were five plain full fat yoghurts (Coberco, Arnhem, the Netherlands) varying in sucrose concentrations: 1%, 5.9%, 10%, 17.6% and 30% w sucrose/w yoghurt. The yoghurts were prepared in the morning prior to sensory testing and were refrigerated at 3°C until the moment of serving. The 10% w/w is the standard concentration of sucrose in commercially available fruit flavoured yoghurts.

Sensory tests

The present study used four different sensory tests: a 'taste-and-spit' test, a 'taste-and-swallow' test, a 'fixed quantity (300 g)' test and an '*ad libitum* consumption' test. Instructions for each of the sensory tests were:

1. Taste-and spit test
a) to rinse with water before each stimulus
b) to sip from the 20 ml of yoghurt and to spit out the sample
c) to rate the yoghurt on pleasantness and sweetness intensity for the sample
d) to rate the yoghurt on pleasantness for the sample considered as a large quantity.

2. Taste-and-swallow test
a) to rinse with water before each stimulus
b) to sip from the 50 ml of yoghurt and to swallow as much as they felt appropriate
c) to rate the yoghurt on pleasantness and sweetness intensity for the sample
d) to rate the yoghurt on pleasantness for the sample considered as a large quantity.

3. Fixed quantity test
a) to rinse with water before the stimulus
b) to eat a fixed quantity of 300 grams of yoghurt
c) to rate the yoghurt at the point of finishing on pleasantness and sweetness intensity.

4. *Ad libitum* consumption test
a) to rinse with water before the stimulus
b) to eat as much as they wanted from the yoghurt, and to ask for more if they wished
c) to rate the yoghurt at the point of finishing on pleasantness and sweetness intensity.

Procedure

Subjects judged the stimuli in carrels used for sensory research. Subjects were instructed both orally and in writing to rate pleasantness and perceived sweetness intensity on a 10-point category scale in the four sensory tests. Pleasantness was measured by a hedonic scale where faces were used to indicate the different degrees of like or dislike (without anchor terms). The response scale for the determination of the perceived sweetness intensity ranged from 'not at all sweet' on the left hand side to 'extremely sweet' on the right hand side.

Each subject participated in 11 sessions at a fixed time on 11 successive days. For each subject the first session included first the taste-and-spit test and secondly the taste-and-swallow test, with a 15 minute rest period in between. In the next ten sessions subjects completed the *ad libitum* consumption test and the fixed quantity test. The order of the *ad libitum* consumption test and the fixed quantity test was randomly determined for each subject. Cards next to the yoghurt indicated if subjects had to eat as much as they wanted or to eat all that was served. The order of presentation of the yoghurts was randomly determined within each sensory test.

The yoghurts for the taste-and-spit test were served in transparent medicine cups of 25 ml, for the taste-and-swallow test in transparent glasses of 200 ml and for the fixed quantity test and the *ad libitum* consumption test in transparent glass bowls of 300 ml. The yoghurts in the taste-and-spit and the taste-and-swallow test were handed one by one to each subject. The amounts consumed in the *ad libitum* consumption test were recorded by weighing back the left overs. Subjects could ask for more if they wished.

Just before and just after finishing each sensory test, subjects rated subjective feelings of hunger and satiety on a visual analogue scale of 150 mm. There were six different appetite related items: 'appetite for a meal', 'appetite for something sweet', 'appetite for something savoury', 'oversatiety/overfullness', 'feeble weak with hunger' and 'appetite for a snack'.

Statistical analyses

A repeated measure analysis of variance (GLM-SAS) was used to analyse differences between the four sensory tests in the perceived sweetness intensity and pleasantness responses. Main effects used in the model were concentration, method, subject and their mutual interactions. Differences in the shapes of the psychophysical and the psychohedonic functions of the four sensory tests were analysed by examining the F-ratio for the (method)x(concentration) interaction. Differences in optimal sucrose concentration among the four sensory tests were judged by visual inspection, and by statistical analysis. We used a non-linear regression-based model to estimate the optimal sucrose concentration (model: pleasantness = $a + b(\log x) + c(\log x)^2$, with x as sucrose concentration). The estimated optimal sucrose concentration was calculated by putting the first derivative equal to zero.

Individual Spearman correlation coefficients between amount of yoghurt eaten in the *ad libitum* consumption test and pleasantness were computed for each sensory test. The Fisher's Z transformation was used to convert these correlation coefficients into z scores in order to perform a one-way analysis of variance. The mean of the inverse *z*-transformed correlation coefficients and their 95% confidence intervals were calculated.

Ratings on the 150-mm visual analogue scale for the feelings of hunger and satiety were read by an Optical Mark reader and converted to scores from 1 (weak) to 25 (strong). The change in appetite ratings was determined by subtracting the ratings taken just before sensory testing from the ratings taken right after sensory testing. A difference in the change in appetite ratings in the fixed quantity test was determined by using analysis of variance and Tukey-test. ANOVA and Tukey-test were also conducted on the amount of yoghurt consumed with concentration, hunger (= 'appetite for a meal' ratings in the *ad libitum* consumption test) and subject as independent variables.

Data analysis was carried out with the SAS statistical software package (SAS Institute Inc., 1990). We considered differences significant at $p < 0.05$.

Results

Concentration-intensity functions

The shapes of the concentration-intensity curves of the four sensory tests were parallel [$F(12,409)=0.53$; $p=0.896$] (Figure 2.1). Sweetness intensity ratings increased with sucrose concentration for each of the sensory tests [$F(4,409)=702.66$; $p<0.0001$]. The mean of perceived sweetness intensity ratings did not change as a function of method [$F(3,103)=0.74$; $p=0.531$].

Concentration-pleasantness functions

Visual inspection of Figure 2.2 suggests that the optimal sucrose concentration as determined by the taste-and-spit test was higher (10% w/w) than that determined from the other tests (5.9% w/w). The estimated optimal sucrose concentration confirmed the visual inspection (Table 2.1).

The shapes of the curves of the four sensory tests were not identical [(method)x(concentration) interaction: [$F(12,409)=3.03$; $p<0.05$]. This was due to the curve of the taste-and-spit test, since the curves of the other consumption tests were parallel [(method)x(concentration) interaction: $F(8,277)=1.57$; $p=0.13$]. Mean pleasantness responses of the taste-and-swallow test were lower than the fixed quantity test and the *ad libitum* consumption test [$F(2,70)=9.24$; $p<0.05$].

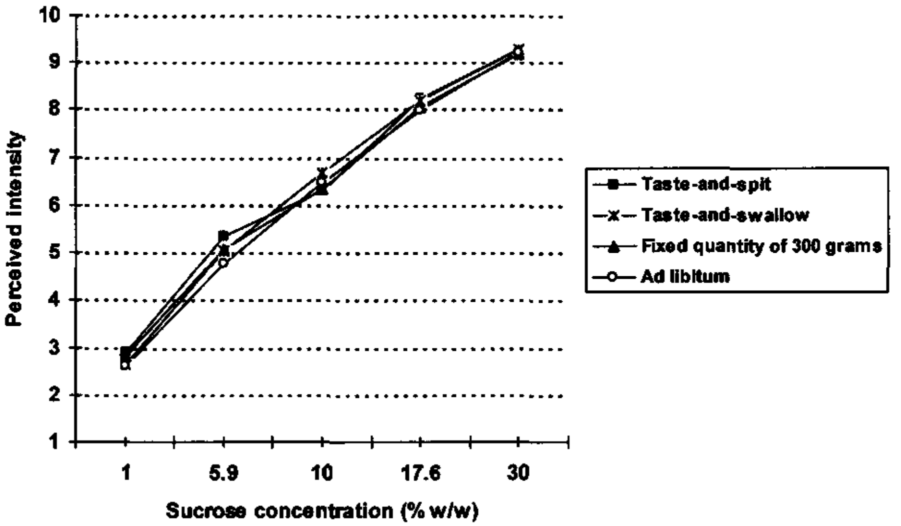

Figure 2.1 Mean responses on perceived intensity of the taste-and-spit test, the taste-and-swallow test, the fixed quantity (300 g) test and the ad libitum consumption test as function of sucrose concentration in yoghurt.

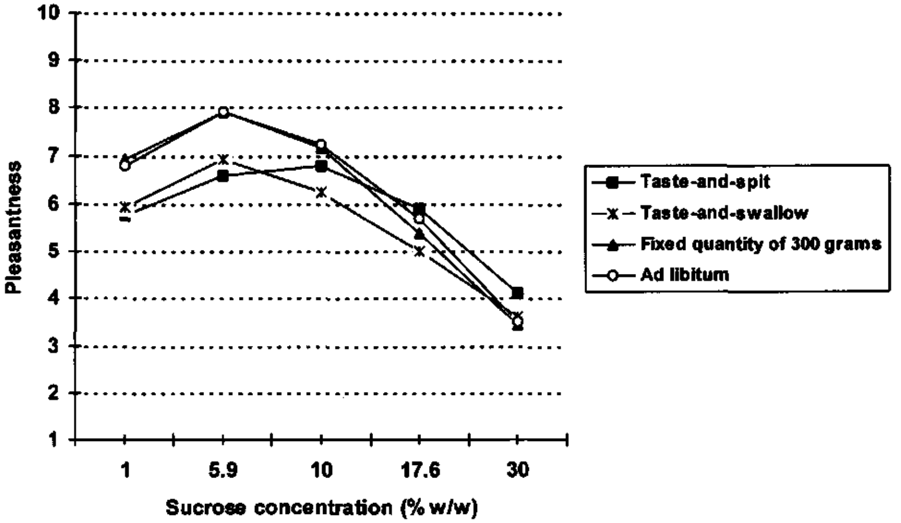

Figure 2.2 Mean pleasantness responses of the taste-and-spit test, the taste-and-swallow test, the fixed quantity (300 g) test and the ad libitum consumption test as function of sucrose concentration in yoghurt.

On average, the pleasantness responses of the taste-and-spit test and the taste-and-swallow test in which samples were considered as a large quantity were lower than the fixed quantity test and the *ad libitum* consumption test [$F(3,103)=18.11$, $p<0.0001$]. In the taste-and-spit test the optimal sucrose concentration shifted from 10% w/w in the laboratory sample to 5.9 % w/w in the same sample but considered as a large quantity.

Table 2.1 Estimated optimal sucrose concentration for each of the sensory tests.

	Estimated optimal concentration (% w/w)
Taste-and-spit	6.6
Taste-and-swallow	4.2
Fixed quantity (300 g)	2.8
Ad libitum consumption	2.9
Taste-and-spit considered as a large quantity	4.7
Taste-and-swallow considered as a large quantity	3.3

Amount of yoghurt eaten

The amount of yoghurt eaten varied with the concentration of sucrose [$F(4,114)=9.48$; $p<0.0001$] (Figure 2.3). Subjects ate significantly less at the 30% w/w sucrose yoghurt than at the other yoghurts, and they ate more of the 5.9% than the 17.6% w/w yoghurt. By comparing Figure 2.2 and Figure 2.3, it can be seen that the hedonic curves of the taste-and-swallow, the fixed quantity test and *ad libitum* consumption test were quite close to the curve of the amount of yoghurt eaten. The standard deviations of the amounts of yoghurt eaten were lower for the optimal yoghurt than for the other yoghurts (respectively 84, 55, 75, 111 and 116, ranging from 1 to 30 % w/w sucrose).

Individual pleasantness ratings in each of the sensory tests correlated positively with the amount of yoghurt eaten (Figure 2.4). The three outliers in each of the sensory test in Figure 2.4 were the same subjects ('large eaters'). We included the 3 subjects in our analyses because the results were the same when we analysed the data after excluding these subjects. The mean of within subject's correlation coefficients between consumption and pleasantness ratings ranged from 0.45 in the taste-and-spit test to 0.81 in the *ad libitum* consumption test (Table 2.2). The mean correlation coefficient of the taste-and-spit test was significant lower than the fixed quantity test and the *ad libitum* consumption test [F(3,138)=5.96; p<0.001]. No difference was found between the correlation coefficient in the taste-and-spit test and the taste-and-swallow test when subjects were asked to consider the sample as a large quantity [F(1,68)=1.75; p=0.19].

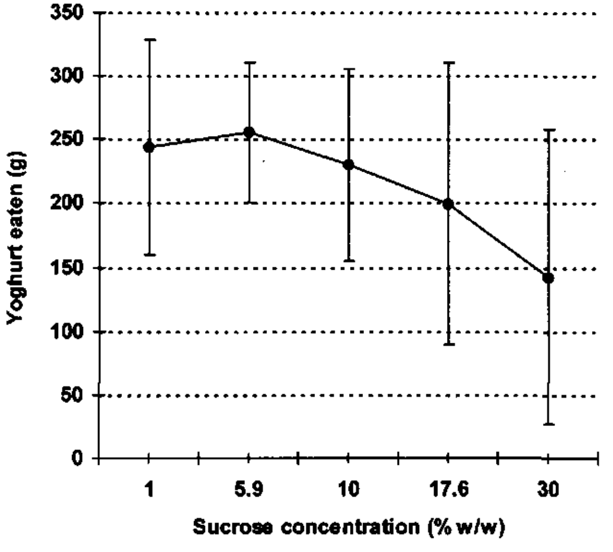

Figure 2.3 Amount of yoghurt eaten ± SD (gram) in the ad libitum consumption test as function of sucrose concentration in yoghurt.

Table 2.2 Mean of within subjects correlation coefficients (r) and 95% confidence intervals (95% CI) between the amount of yoghurt eaten in the ad libitum consumption test and pleasantness ratings for each of the sensory tests.

	r	95% CI
Taste-and-spit	0.45	(0.26 - 0.61)
Taste-and-swallow	0.62	(0.47 - 0.73)
Fixed quantity (300 g)	0.75	(0.59 - 0.85)
Ad libitum consumption	0.81	(0.73 - 0.88)
Taste-and-spit considered as a large quantity	0.53	(0.36 - 0.66)
Taste-and-swallow considered as a large quantity	0.66	(0.51 - 0.78)

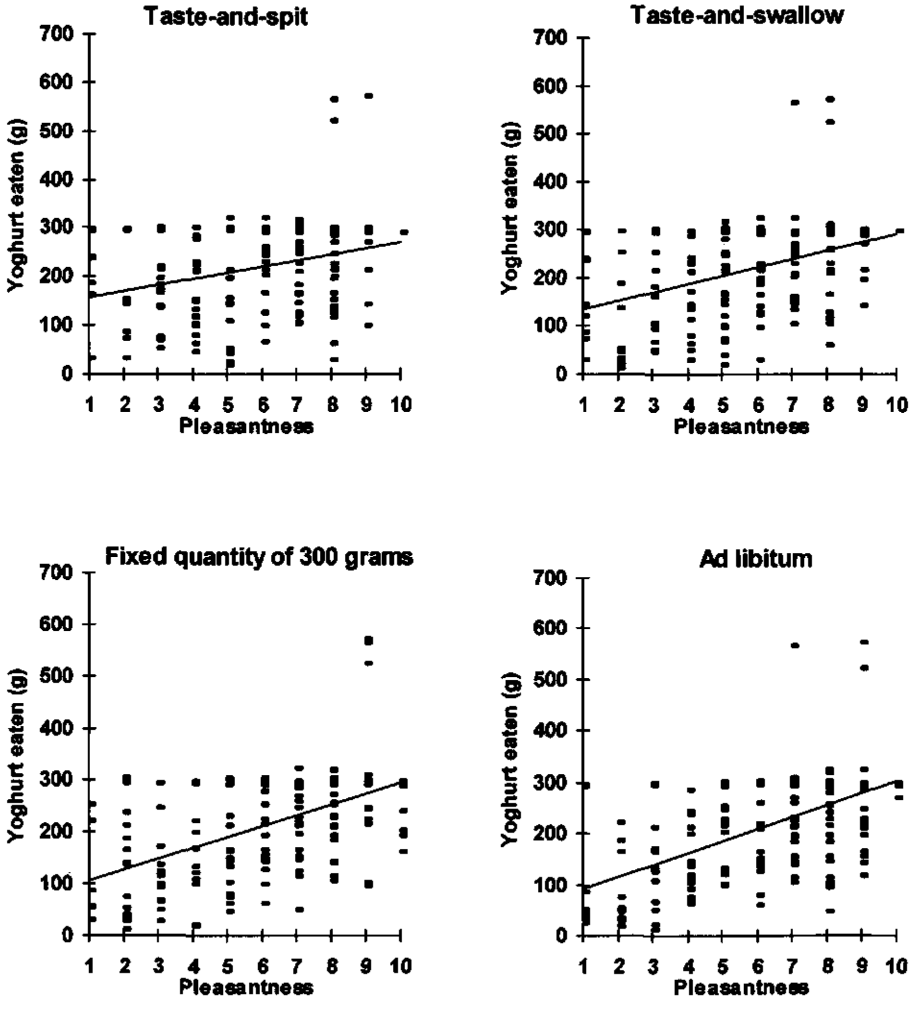

Figure 2.4 Individual pleasantness ratings --derived from the taste-and-spit test, the taste-and-swallow test, the fixed quantity (300 g) test and the ad libitum consumption test-- plotted against amount of yoghurt eaten (g) in the ad libitum consumption test.

Feelings of hunger and satiety in fixed quantity (300 g) test and *ad libitum* consumption test

Clear differences were found in change in 'appetite for something sweet' ratings between the five sucrose concentrations in the fixed quantity test and in the *ad libitum* consumption test [$F(4,138)=8.6$; $p<0.0001$ and $F(4,138)=3.3$; $p=0.01$, respectively] (Figure 2.5). The change at the 30% w/w sucrose concentration yoghurt was higher than all other yoghurts in the fixed quantity test, and higher than the 1% w/w sucrose concentration yoghurt in the *ad libitum* consumption test. In the fixed quantity test the change in the other appetite ratings did not significantly differ [$F(4,138)<1.2$; $p>0.3$]. In the *ad libitum* consumption test the change in 'appetite for a meal' concurred with the actual weight consumed, and was significantly higher after the 5.9% w/w sucrose concentration yoghurt than after the 30% w/w sucrose concentration yoghurt [$F(4,138)=4.3$; $p=0.003$]. In the *ad libitum* consumption test no significant change in the other appetite ratings was found [$F(4,138)<2.1$; $p>0.1$].

Discussion

The main results of this study show that pleasantness ratings collected after the taste-and-swallow test give a better prediction of food consumption than the taste-and-spit test. Furthermore, perceived sweetness intensity ratings do not differ among the sensory test used.

The optimal sucrose concentration in the taste-and-spit test (10% w/w) was higher than in the taste-and-swallow test, the fixed quantity test and the *ad libitum* consumption test (5.9% w/w). This finding is in agreement with previous studies (Monneuse *et al.*, 1991; Bellisle *et al.*, 1988; Pérez *et al.*, 1994; Lucas & Bellisle, 1987) which concluded that short brief-exposure tests give a biased estimation of the optimal concentration, but not with other studies which did not find a difference in optimal concentration between the sensory test used (Daillant & Issanchou, 1991; Popper *et al.*, 1989; Shepherd *et al.*, 1991).

Also, we found that the yoghurt consumption correlated best with pleasantness ratings of the *ad libitum* consumption test ($r=0.81$), and worst with pleasantness ratings of the taste-and-spit test ($r=0.45$). This observation confirms the results of a study by Hellemann & Tuorila (1991) who found that the hedonic responses after *ad libitum* consumption of bread correlated better with actual consumption than responses of the taste-and-spit test ($r=0.82$ vs. $r=0.63$). The mean of within subjects correlation coefficient in the taste-and-swallow test in the present study

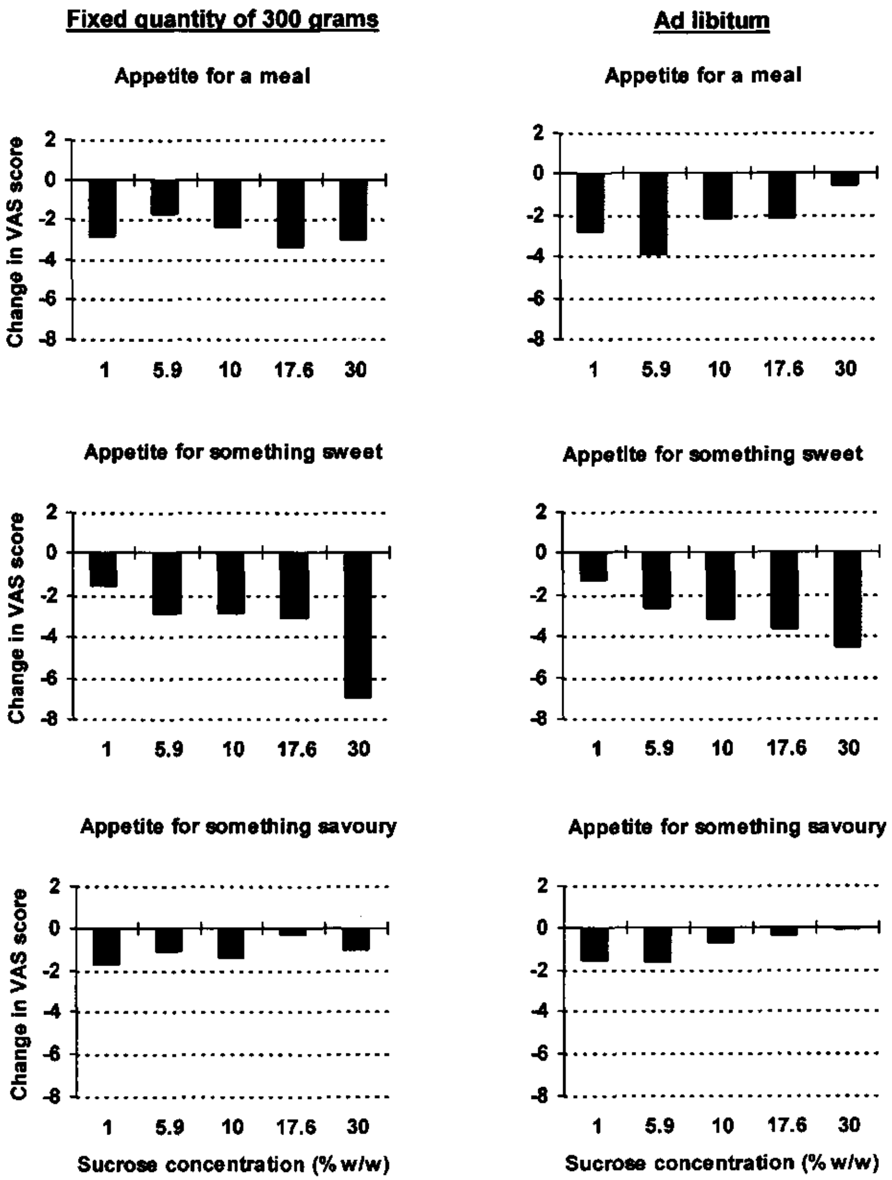

Figure 2.5 Change in 'appetite for a meal', 'appetite for something sweet' and 'appetite for something savoury' ratings in the fixed quantity (300 g) test and the ad libitum consumption test. The rating taken just before sensory testing is subtracted from the rating taken right after sensory testing.

(r=0.62) was lower than in the *ad libitum* consumption test (r=0.81), but still (non-significantly) higher than the taste-and-spit test (r=0.45). Differences in optimal concentration and within subjects correlation coefficients observed in the present study and previously imply that the taste-and-spit test is a poor predictor of consumption. In addition, the taste-and-swallow test gives a better prediction of consumption than the taste-and-spit test.

Mean pleasantness responses for the taste-and-spit test and the taste-and-swallow test were generally lower than the fixed quantity test and the *ad libitum* consumption test. The unpleasant element of spitting in a small bucket as well as inadequate stimulation of taste receptors in the taste-and-spit test could have influenced these ratings. In the taste-and-swallow test the size of the food sample eaten was probably not large enough to perform an overall judgement of the food. Another explanation for the generally lower pleasantness responses might be the day effect we introduced in the procedure by pairing the taste-and-spit test and taste-and-swallow test in session 1. When the taste-and-swallow test was conducted, all subjects had already been exposed to the test samples from the preceding taste-and-spit test. This exposure may have effected the judgements in the taste-and-swallow test, in the same way that re-tasting of samples can alter judgements in sensory procedures.

Furthermore, the ratings of the taste-and-spit test and the taste-and-swallow test (in which subjects ingested 0-50 ml) do not include the postingestive rewarding effects of ingesting substantial amounts of yoghurt with sucrose (Sclafani, 1997). However, the taste-and-spit test had the advantage that the measurements were conducted under standardised conditions. Subjective feelings of hunger and satiety, for example, were less controlled in the *ad libitum* consumption test in which subjects were allowed to consume as much yoghurt as they wished; this may have influenced the ratings for pleasantness. Moreover, social convention to finish eating what is in the whole bowl, is an important factor which likely influenced the data of the *ad libitum* consumption test (Mook & Votaw, 1992). Visual inspection of Figure 2.4 with the individual consumption data shows a limit at 300 g which is probably due to this social convention.

The optimal sucrose concentration in the taste-and-spit test shifted from 10% w/w in the laboratory sample to 5.9% w/w in the same sample but considered as a large quantity. Mattes & Mela (1986) also asked their subjects to consider each food presented as both an isolated laboratory sample and as a food prepared for normal consumption. The preferred sweetness of coffee and oatmeal was higher when the samples were spat out and perceived as a 'sample' as opposed to the swallowed samples. Adding the question --consider the food sample as a large quantity-- might therefore be an option to improve the predictive power of the taste-and-spit test for amounts consumed.

In the fixed quantity test and the *ad libitum* consumption test clear differences were found in change in 'appetite for something sweet' ratings between the five sucrose concentrations. The liking for sweet foods in general was reduced by consuming the sweetened yoghurt, which could be interpreted as sensory-specific satiety (Hetherington *et al.*, 1989; Rolls *et al.*, 1984). The ingestion of the highly sweetened yoghurts (> 10% w/w sucrose) in the fixed quantity test and the *ad libitum* consumption test resulted in a steeper slope of the hedonic curves compared to the taste-and-spit test and the taste-and-swallow test. This steeper slope confirms the phenomenon of sensory specific satiety.

For the food industry, catering and nutrition education programmes it is important to know if improvement of hedonic factors will lead to increased consumption. From this study it can be concluded that pleasantness ratings collected after the taste-and-swallow test give a better prediction of consumption than the taste-and-spit test. Rating a small sample while anticipating consuming a larger amount of this sample might be an option to improve the predictive power on consumption of the taste-and-spit test. The question of whether the same findings will be observed with non-sweet taste substances is still unclear. There is a greater sensitivity to sweet at the front of the tongue in contrast to, for example, to bitter. Consequently, differences in pleasantness ratings between the spitting and swallowing task might be more pronounced when testing with non-sweet taste substances (e.g. bitter, sour, salty). The generalisation of our findings to other age groups (e.g., children, elderly) should be investigated as well, since age has an effect on taste and smell perception (Zandstra & De Graaf, 1998). Especially for elderly with inadequate nutrition it is important to develop palatable 'senior' foods which may improve food enjoyment and may therefore result in an increment of food intake. Finally, one should realise that the hedonic factor is only one out of many factors determining the amount of food eaten, amongst other social and economic factors (Drewnowski, 1997; Logue, 1986). We have limited our study to laboratory methods. In these methods we assumed that the laboratory consumption reflects the consumption in the natural environment. Future work should concentrate on evaluating the ability of sensory and hedonic responses to predict consumption outside the laboratory, in homes and other real-life situations (Meiselman, 1992).

Acknowledgements

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3

Short-term regulation of food intake in children, young adults and the elderly*

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Wija A. van Staveren

Abstract

The present study investigated the effects of consuming preloads with different macronutrient and energy contents on subsequent intake and subjective feelings of hunger and satiety in children, young adults and the elderly. A 'preload-test meal' design was applied to 30 children (4-6 y), 33 young adults (18-26 y) and 24 elderly (61-86 y). Subjects were given four different strawberry yoghurt preloads that varied in energy and macronutrient content, or no yoghurt. Children, young and elderly adults consumed 200, 340 and 300 g of the preload, respectively. One yoghurt was low-fat, low-carbohydrate and low in energy (= the control; 0.7 MJ/500 g serving), one yoghurt was high-fat and medium in energy (71 en% of fat; 2 MJ/500 g serving), one yoghurt was high-carbohydrate and medium in energy (87 en% of carbohydrate; 2 MJ/500 g serving) and the fourth yoghurt was high-fat & high-carbohydrate and high in energy (42 en% of fat and 53 en% of carbohydrate; 3 MJ/500 g serving). 90 Minutes after preload consumption, subjects had an attractive *ad libitum* lunch-buffet. Energy intake at lunch and subjective feelings of hunger and satiety were analysed. The ability to compensate at lunch did not differ among the three age groups. Compared to the no-preload condition, all children, young and elderly adults ate significantly less after the high-fat & high-carbohydrate yoghurt. The energy compensation observed in the children ranged between -21% and 34%, in the young adults between 15% and 44% and in the elderly between 17% and 23%. Hunger responses were clearly different between the young and elderly adults. The young adults showed larger differences in their appetite ratings than the elderly, indicating that the elderly were less sensitive to the energy content of the preload than the young adults. We conclude that the ability to regulate the food intake within a preload -90 min- test meal paradigm did not differ among children, young adults and elderly.

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Introduction

Overweight and obesity are generally recognised to increase the risk of cardiovascular diseases and diabetes, and as a consequence there has been much debate about the underlying physiology of obesity. The prevalence of overweight in adults in European countries has been increasing over the last 15 years so that at least half of the adults have a body mass index higher than 25 kg/m^2 (Seidell, 1995a; Seidell, 1995b). Overweight in children is a rising public health concern given its assumed increasing prevalence and its association with excess morbidity and mortality in adulthood (Must, 1996; DiPietro *et al.*, 1994; Srinivasan *et al.*, 1996; Cook & Grothe, 1996; Gortmaker *et al.*, 1996).

On the other hand, a decrease in energy intake is shown in elderly Europeans (De Groot *et al.*, 1996). This may be a result of social influences (e.g., lack of help with shopping, bereavement, poverty), physical aspects (e.g., dentition problems, mobility disability), physiological factors (e.g., loss of appetite, decreased taste and odour perception) or a combination of some factors (Rolls *et al.*, 1995; Schiffman & Warwick, 1993; Roberts *et al.*, 1994; Morley, 1996; Morley & Silver, 1988). This so-called 'anorexia of aging' may have serious consequences, such as severe weight loss after the age of 70 years that is associated with nutrition-related diseases (Rajala *et al.*, 1990; Mowé *et al.*, 1994; Losonczy *et al.*, 1995).

The control of food intake could play an important role in the development of obesity in children and young adults and anorexia in the elderly. The results of a number of studies suggest that the precision of food intake regulation declines with increasing age (Birch & Deysher, 1985; Birch & Deysher, 1986; Roberts *et al.*, 1994; Rolls *et al.*, 1995). In Birch and Deysher's studies (1985 and 1986) young children two to five years old compensated over a short-term period for a high energy intake at one meal by a lower energy intake at the next meal, and *vice versa*. Elderly men in the study of Roberts *et al.* (1994) compensated less well for long-term (three weeks) imposed changes in energy balance than did young men. Rolls *et al.* (1995) reported that food intake compensation after different yoghurt preloads was less precise in elderly than in young men. The data on adult's abilities to show energy compensation are less clear, most likely because of variations in experimental designs (Rolls & Hammer, 1995; Shide *et al.*, 1995; De Graaf *et al.*, 1992; Blundell *et al.*, 1993; Rolls *et al.*, 1991; Foltin *et al.*, 1990; Caputo & Mattes, 1992). Up to now there has been no experiment in which all three age groups, i.e. children and young and elderly adults, have been compared.

The present experiment was designed to investigate the short-term regulation of food intake at three different stages in life. We used a preloading paradigm with a 'preload-test meal' design in children, young adults and elderly. Adjustments in energy intake were determined in a meal consumed *ad libitum* in response to covert changes of fat, carbohydrate and energy in a fixed yoghurt preload. We also measured subjective feelings of hunger and satiety in young adults and elderly to test whether elderly are less sensitive to the energy content of foods than young adults.

Materials and methods

Subjects

The study involved 88 subjects divided into three age-groups: 30 children (4-6 y), 33 young adults (18-26 y) and 24 elderly (61-86 y). The children attended a primary school in Bennekom, a village near Wageningen (The Netherlands). None of the children had a food allergy or a chronic health problem; a parental informed consent was obtained for each child. Young adults were healthy students of Wageningen University. Independently-living elderly were recruited throughout Wageningen and its surroundings by advertisements and by word of mouth. Elderly taking any medications that would affect food intake were excluded from participation.

Table 3.1 Characteristics of the subjects classified by age group (mean ± SD).

	Children (*n*=30)	Young adults (*n*=33)	Elderly (*n*=24)
Age (years)	4.5 (0.6)	22 (1.9)	75.5 (5)
Height (cm)	112.4 (4.8)	174.4 (8.1)	165.3 (0.1)
Weight (kg)	20.2 (2.4)	71.0 (9.6)	72.4 (8.9)
BMI (kg/m^2)	16.0 (1.2)	23.3 (2.3)	26.6 (3.5)
Restraint score[a]	-	2.2 (0.7)	2.9 (0.8)
Men/women (*n*)	18/12	5/28	6/18

[a] The restraint score was measured with Van Strien's Dutch Eating Behavior Questionnaire (1986). The minimum value is 1 (no restraint), the maximum value is 5 (high restraint).

The young adults and elderly signed an informed consent before participating in the study. They were told that the experiment was meant to assess sensory perception and feelings of hunger and satiety. Table 3.1 shows the main characteristics of these subjects in the study.

At the end of the study the young adults and elderly were paid and the children were given a small present. The experiment was approved by the Medical Ethical Committee of the Division of Human Nutrition & Epidemiology of Wageningen Agricultural University.

Design

The study had a 'preload-test meal' design. Five different preloads were offered in a random sequence that varied for each subject, resulting in a within-subjects repeated-measures design with each subject serving as his own control. The five experimental days took place in two weeks for each age group. The study with the elderly was conducted in the months July and August, the young adults in October and the children in November. A test day was run before the first experimental day to familiarize the subjects with the experimental procedure. The data obtained from this day were excluded from further analysis.

At the present time there is no agreement on how long the time interval between preload and test meal should be (Mattes, 1996; Rolls *et al.*, 1991; Houniet *et al.*, 1997; Rolls & Hammer, 1995). Several studies used a short time interval of about 20 - 30 min (Birch & Deysher, 1985; Birch & Deysher, 1986; Birch *et al.*, 1989), while other studies used an interval of more than 90 min (Hulshof *et al.*, 1995; Birch *et al.*, 1993; Johnson *et al.*, 1991; Drewnowski *et al.*, 1994; De Graaf *et al.*, 1993; Blundell *et al.*, 1993). In the present study we chose a time interval of 90 min. We believe that with a shorter time interval the volume and weight effects rather than the energy content and macronutrient effects of the preload will be measured on appetite and energy intake. Another argument in favour of a 90 min time interval is that the digestion of regular amounts of carbohydrates and fats results in the largest differences after 60 - 90 min in appetite related physiological responses, such as CCK (Maas *et al.*, 1997), insulin (Jenkins *et al.*, 1987), glucose (Jenkins *et al.*, 1987; Wolever *et al.*, 1991) and diet-induced thermogenesis (Visser *et al.*, 1995; Blundell *et al.*, 1993).

Table 3.2 Ingredients, macronutrient content and energy density of the control, high-fat, high-carbohydrate, and high-fat & high-carbohydrate yoghurts.

g/500 g serving	Control (non-blended)	High-fat (blended)	High-CHO[a] (blended)	High-fat & high-CHO[a] (blended)
Skimmed yoghurt	420	-	328	-
Full cream yoghurt	-	305	-	223
Aspartame	0.3	0.3	-	-
Strawberry, pureed	80	90	80	80
Fat-reduced whipped cream (25% fat)	-	105	-	105
Rice flour	-	-	25	25
Soft white sugar	-	-	60	60
Water	-	-	7	7
Total weight (g)	500	500	500	500
MJ (kcal)	0.7 (157)	2 (457)	2 (447)	3 (729)
Protein (g) (En%)	17 (45)	14 (12)	16 (14)	13 (7)
Fat (g) (En%)	0.4 (2)	36 (71)	0.3 (1)	33 (42)
Carbohydrate (g) (En%)	21 (54)	20 (17)	95 (87)	94 (53)
Age group	Control (MJ/served portion[b])	High-fat (MJ/served portion[b])	High-CHO[a] (MJ/served portion[b])	High-fat & high-CHO[a] (MJ/served portion[b])
Children	0.3	0.8	0.8	1.2
Young adults	0.5	1.3	1.3	2.1
Elderly	0.4	1.1	1.1	1.8

[a] High-CHO = High-carbohydrate.

[b] The children, young adults and elderly consumed 200, 340 and 300 grams of the preload, respectively.

Preloads

The preloads consisted of four strawberry yoghurts in which the energy, fat and carbohydrate content was covertly manipulated (Table 3.2). One yoghurt was low-fat, low-carbohydrate and low in energy (= the control; 0.7 MJ/500 g serving), one yoghurt was high-fat and medium in energy (71.4 en% of fat; 2 MJ/500 g serving), one yoghurt was high-carbohydrate and medium in energy (86.6 en% of carbohydrate; 2 MJ/500 g serving) and the fourth yoghurt was high-fat & high-carbohydrate and high in energy (41.5 en% of fat and 52.5 en% of carbohydrate; 3 MJ/500 g serving). There was one no-preload condition as well. The children, young adults and elderly consumed 200, 340 and 300 grams of the preload, respectively.

Test meal

The test meal consisted of a lunch-buffet consumed *ad libitum*. The lunch-buffet was composed of bread (brown or white), sweet and savoury sandwich fillings, fruits and drinks. Drinks were semi-skimmed milk, orange juice, coffee and tea. Fruits included apples and oranges. Sweet sandwich fillings comprised chocolate paste and jam, and savoury sandwich fillings were cheese, ham and peanut butter.

Subjects were allowed to choose whatever and as much as they wanted from the served foods. The sandwiches for the children were prepared in advance and were served on a tray in the middle of the table in front of the children. Coffee and tea was not offered to the children.

Measurements

Energy intake at lunch

Intake of food from the self-selected lunch-buffet was analysed by determining the wrappings and weighing all the amounts of food remaining on the plates. The amounts consumed were converted to energy and macronutrients with the help of the Dutch food composition table (NEVO, 1993).

Subjective feelings of hunger and satiety

Young adults and elderly rated their feelings of hunger and satiety of appetite by means of a double slash on six 150 mm visual analogue scales as described by Hulshof *et al.* (1995). The children did not complete these ratings. The six items were: 'appetite for a meal'; 'appetite for something sweet'; 'appetite for something savoury'; 'oversatiety/overfullness'; 'feeble, weak with hunger'; 'appetite for a snack'.

Sensory ratings preload

While consuming the preload, the young adults and elderly rated the pleasantness and perceived intensity of 3 sensory attributes on 150 mm visual analogue scales. For pleasantness the left- and right-hand side were anchored with the terms 'very unpleasant' and 'very pleasant'. For the perceived intensity of creaminess, sweetness and strawberry flavour, 'weak' and 'strong' were on the left- and right-hand sides of the scales.

Procedure

The young adults and elderly ate their breakfast at 8:00 h at home. They were asked to eat the same breakfast on each of the experimental days and to register the consumption of the breakfast in a diary. Every subject was asked not to eat or to drink anything between breakfast and preload, and between preload and the subsequent test meal. At 10:00 h the young adults and elderly were offered the preload at the departmental dining room. They scored their feelings of hunger and satiety on a visual analogue scale just before consumption of the preload and 15, 45, 75, 105 and 150 minutes after that rating. At 11:45 h, ninety minutes after the consumption of the preload, the subjects could eat whatever they wanted from the items of the test meal. During lunch, the young adults and elderly were sitting in groups, with 6 to 8 subjects per table.

The children got the preload at 9:45 h in primary school during their regular classroom snack period. The no-preload condition was planned on the same day for all children; the other preloads were served randomly. Prior to the study the children were trained by the teacher, using games about taste and food choice ('What is taste?' 'What would you choose from a lunch-buffet and why?'). The children did not eat or drink anything at school except the preload and the test meal. The lunch started 75 minutes after consuming the preload (at 11:15 h) in order to finish eating within the school hours (12:00 h). During lunch the children sat in the classroom at their own tables in seven groups of 4-5 children. Each group was supervised by a parent who was instructed before the study not to interfere with the children's food choice.

Statistical analyses

Statistical analyses were carried out with the statistical software package SAS (SAS Institute Inc., 1990). The data of the elderly were part of another study in which the effects of lunch environment (cosy vs. non-cosy setting) on food intake were determined. In the present study we used the data of the elderly in the cosy setting, since this environment was analogous to the cosy setting of the children and young adults.

A repeated-measures analysis of variance (α = 0.05) was performed to evaluate the effects of the preloads on energy intake at lunch using a model with preload, age group and subjects nested within age group as main effects. The (age group)x(preload) interaction reflected differences in ability to compensate at lunch between the age groups. Fourteen of the 30 children did not successfully complete the entire protocol because of failing to consume the amount of preload required in one or two conditions. Therefore, we used for safety two different methods to analyse the data because we did not want to miss a difference if one existed. First, we used the SAS Proc Mixed Procedure, which deals with unbalanced data and assumes that missing observations are missing at random. Second, we used the SAS Proc GLM Procedure in which we restricted the analysis to subjects who successfully completed the protocol (thereby excluding 14 children). Finally, we used a two-way analysis of variance (excluding the no-preload condition) to specify the effects of carbohydrate and fat in the preloads on energy intake at lunch. Differences in food choice data were assessed using the chi-square test.

Energy compensation was calculated as the difference of the mean lunch intake in the no-preload condition and the mean lunch intake for each of the yoghurt conditions divided by the energy of the preload, multiplied by 100. A positive value indicates that subjects (partial) compensate at lunch, while a negative value indicates that subjects over-eat at lunch.

Appetite ratings on scales for the 'feelings of hunger and satiety' dimensions were read automatically by an optical mark reader and were converted into scores from 1 (weak) to 25 (strong). Analysis of variance (α = 0.05) and Tukey-test were applied to investigate the effects of the preloads on absolute ratings, with age group and type of preload as the fixed within subject factor and subjects nested into age group as the random factor.

Results

Energy intake at lunch

The ability to compensate among the five conditions at lunch did not differ between the three age groups [(age group)x(preload) interaction $F(8,293)=1.33$; $p=0.23$]. The results did not change when the analysis were restricted to the subjects who successfully completed the protocol [(age group)x(preload) interaction $F(8,279)=1.30$; $p=0.25$; see Figure 3.1]. The effect of the preloads on the energy intake at lunch was significant within each age group [$F(4,60)=3.1$; $p=0.02$ for children; $F(4,128)=3.7$; $p=0.01$ for young adults; $F(4,92)=6.3$; $p=0.0002$ for elderly]. Compared to the no-preload condition, all children, young adults and elderly ate significantly less after the high-fat & high-carbohydrate yoghurt. Furthermore, the elderly ate significantly less, whereas the children ate significantly more after the high-carbohydrate yoghurt compared to the no-preload condition.

The two-way analysis with fat and carbohydrate as the main factors (excluding the no-preload condition) suggested that the different energy intake at lunch was mainly due to the carbohydrate content and not to the fat content of the preloads [carbohydrate: $F(1,60)=4.0$; $p=0.05$ for children; $F(1,128)=5.2$; $p=0.03$ for young adults; $F(1,92)=4.8$; $p=0.03$ for elderly] [fat: $F(1,60)=1.3$; $p=0.26$ for children; $F(1,128)=0.1$; $p=0.75$ for young adults; $F(1,92)=5.7$; $p=0.02$ for elderly].

There was an incomplete energy compensation for each of the three age groups (see Table 3.3). The energy compensation observed in the 30 children ranged between -21% and 34%, in the young adults between 15% and 44% and in the elderly between 17% and 23%.

Table 3.3 Mean energy compensation (%) after the different yoghurt conditions classified by age group.

Preload	Children (*n*=30) (% compensation)	Young adults (*n*=33) (% compensation)	Elderly (*n*=24) (% compensation)
Control	-21	44	23
High-fat	-7	15	19
High-CHO[a]	-7	29	19
High-fat & high-CHO[a]	34	20	17

[a] High-CHO = High-carbohydrate.

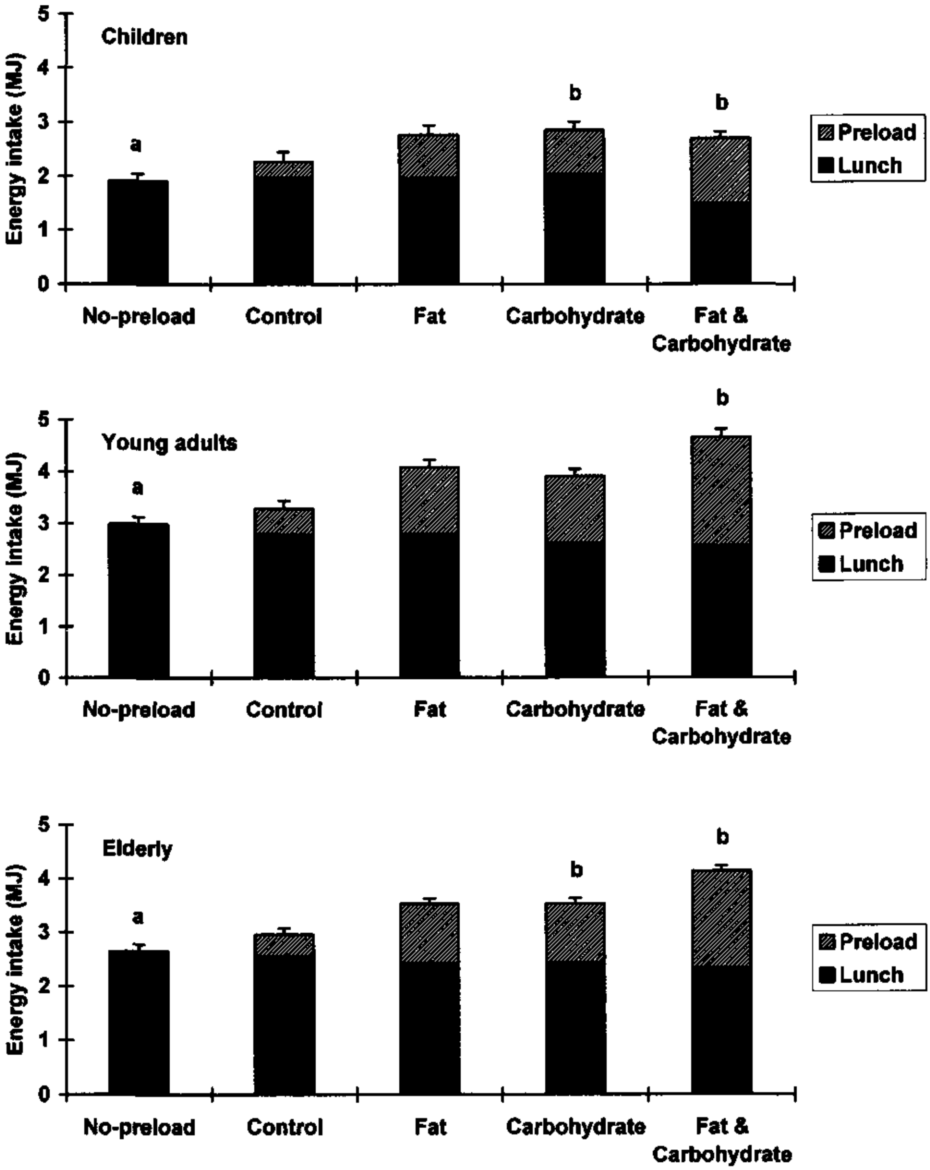

Figure 3.1 Mean energy intake at lunch + SEM (MJ) after the different preloads for 30 children, 33 young adults and 24 elderly. Dissimilar letters indicate a significant difference among the preloads.

The children's food choice pattern at lunch differed from that of the young adults and elderly (χ^2>8.9; p<0.01 for all food products). The children ate more white bread with sweet spread than the other age groups. They chose in general an apple as fruit and orange juice as a cold drink. The elderly mainly consumed brown bread with savoury spread and an orange as fruit. For drinks they choose milk and coffee. The young adults mainly drank tea, and apart from that, had a similar food choice pattern to the elderly.

Subjective feelings of hunger and satiety

The absolute ratings of the items 'appetite for a meal', 'appetite for something sweet' and 'appetite for something savoury' are presented in Figure 3.2. The mean responses for the items 'feeble, weak with hunger' and 'appetite for a snack' were similar to the responses of the item 'appetite for a meal', while the curve of responses of the item 'oversatiety/overfullness' was the reverse.

Analysis of the appetite responses revealed a significant time effect for the six items across the five conditions [all F(5,1090)>106.4; p<0.0001] (see Figure 3.2). A group effect (young adults vs. elderly) for the six appetite items was found [all F(1,1416)>4.7; p<0.03] such that the appetite ratings between the conditions differed more in the young adults than in the elderly. The young adults gave lower ratings after the high-fat yoghurt, the high-carbohydrate yoghurt and the high-fat & high-carbohydrate yoghurt compared to the no-preload condition for all appetite items [all F(4,950)>15.8; p<0.0001]. The elderly gave only lower ratings after the consumption of the high-fat & high-carbohydrate yoghurt compared to the no-preload condition for all appetite items, except for 'appetite for something sweet' where the appetite ratings were lower after the consumption of the high-fat yoghurt [all F(4,685)>3.3; p<0.01]. Neither the young adults nor the elderly had differences in the appetite ratings between the high-fat and high-carbohydrate yoghurts.

Compared to the no-preload condition, the young adults generally showed larger differences in their appetite ratings than the elderly [(age group)x(preload) interaction: all F(4, 219)> 3.2; p<0.02].

Sensory ratings preload

Analysis of variance of the sensory ratings of the yoghurt preloads showed no significant differences in rated pleasantness, perceived intensity of strawberry flavour or perceived intensity of sweetness (Table 3.4) [F(3,96)=0.4; p=0.74, F(3,96)=1.4;

p=0.26 and F=(3,96)=2.6; p=0.06, respectively, for young adults; and F(3,69)=0.01; p=0.99, F(3,69)=1.3; p=0.28; F(3,69)=1.7; p=0.18, respectively, for elderly]. The only statistically significant difference found was in the rating of the creaminess of the yoghurt: the young adults rated the creaminess of the high-carbohydrate and high-fat & high-carbohydrate yoghurt higher than the control and the high-fat yoghurt [F(3,96)=29.4; p<0.0001] and the elderly rated the creaminess of the high-fat & high-carbohydrate yoghurt higher than of the three other yoghurts [F(3,69)=3.6; p=0.02].

Table 3.4 Mean responses (SD) (millimeters from left-hand anchor) for pleasantness and sensory attributes of the preloads of 33 young adults and 24 elderly subjects. Dissimilar letters indicate a significant difference among the preloads.

	Control	High-fat	High-CHO[a]	High-fat & high-CHO[a]
Young adults (*n*=33)				
Pleasantness	88 (27)	93 (33)	89 (32)	85 (40)
Creaminess	76 (31)[A]	73 (36)[A]	109 (22)[B]	123 (25)[B]
Sweetness	85 (32)	95 (36)	100 (30)	102 (29)
Strawberry flavour	89 (31)	87 (35)	89 (39)	77 (37)
Elderly (*n*=24)				
Pleasantness	80 (29)	76 (28)	81 (30)	75 (34)
Creaminess	84 (27)[C]	74 (31)[C]	83 (30)[C]	103 (23)[D]
Sweetness	86 (33)	82 (35)	93 (29)	95 (32)
Strawberry flavour	79 (32)	68 (32)	81 (29)	76 (33)

[a] High-CHO = High-carbohydrate.

Discussion

The results of the present study show that the ability to regulate the food intake within a preload -90 min- test meal paradigm does not differ among children, young adults and elderly. There is evidence for an incomplete caloric compensation in each of the three age groups. Compared to the no-preload condition, the young adults show generally larger differences in their appetite ratings than the elderly, suggesting that elderly may be less sensitive to the preload manipulations than young adults.

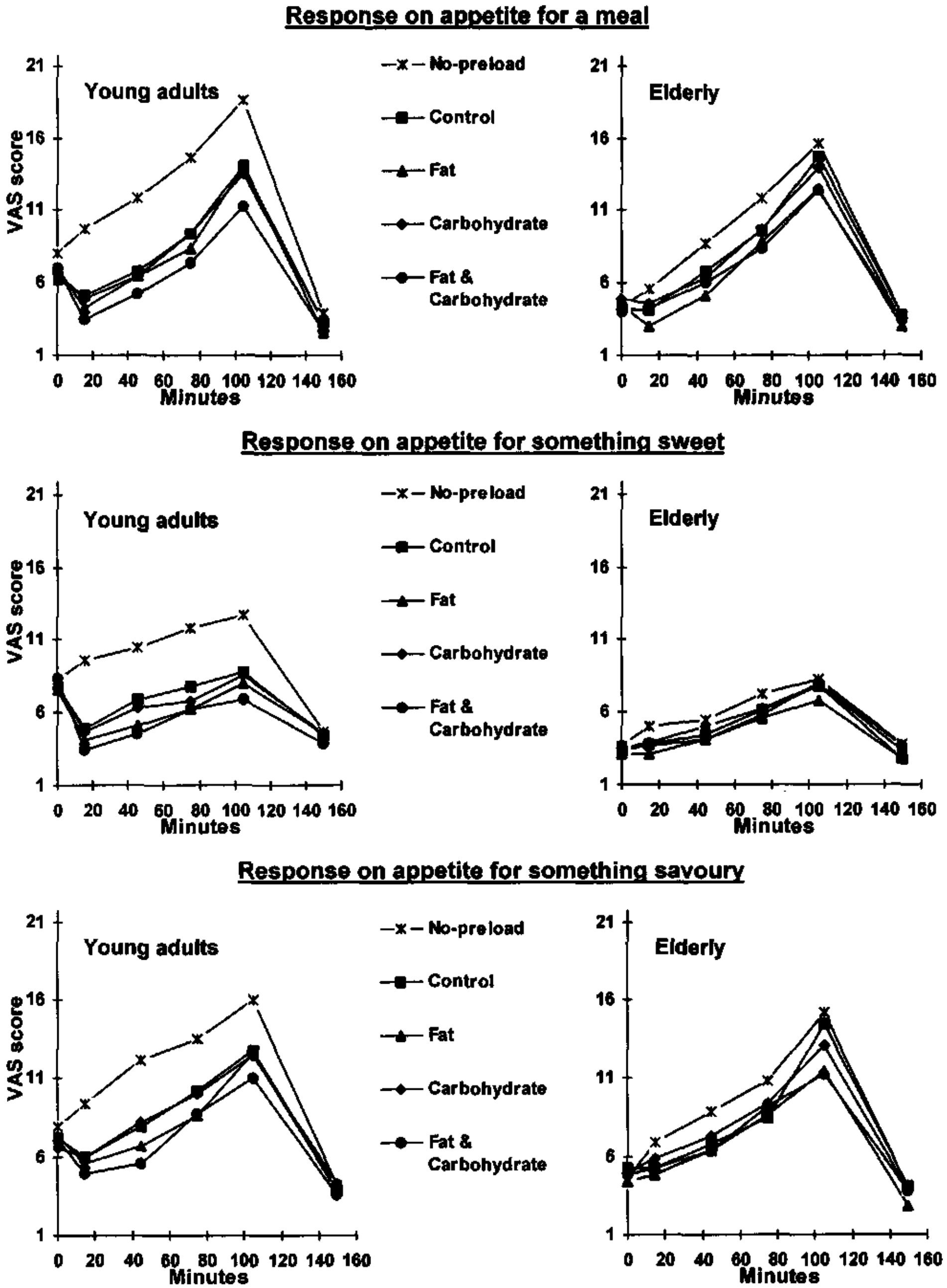

Figure 3.2 Absolute ratings 'appetite for a meal', 'appetite for something sweet' and 'appetite for something savoury' from 1 (weak) to 25 (strong) after different preloads for 33 young adults and 24 elderly.

In accordance with our findings, Birch *et al.* (1993) and Johnson *et al.* (1991) found an incomplete compensation in children (2-4 y) after a 90 min time interval. However, two studies by Birch and Deysher (1985 and 1986) showed complete compensation for, respectively, 18 and 21 children (2-5 y) using a 20 min time interval. Based on this research Birch and co-workers suggested that children are more sensitive to physiological, internal cues than adults or older children who may be more susceptible to eat in response to external cues (e.g. experience with food and eating, socio-cultural factors) (Birch & Deysher, 1985; Birch & Deysher, 1986; Birch *et al.*, 1989).

Both Rolls *et al.* (1995) and Roberts *et al.* (1994) showed an impairment in the food intake regulation in elderly individuals compared to young adults. The preload - test meal design of the present study is analogous to the design used in the study of Rolls *et al.* (1995). There are two major differences between their study and our study: the amount of yoghurt consumed --500 g in their study for both the young and elderly men, respectively, and 340 and 300 g in our study for the young and elderly adults, respectively--, and the length of the time interval --30 min in their study vs. 90 min in our study--. Rolls *et al.* (1995) found a larger difference in energy intake at lunch between the no-preload condition on the one hand and the control, the high-fat and the high-carbohydrate condition on the other hand than in our study. However, in the study of Rolls *et al.* (1995), the difference in energy intake at lunch between the control, the high-fat and the high-carbohydrate condition was small for both the adults and the elderly. This is in agreement with our data. We therefore think that the lunch intake data after the no-preload condition in the study of Rolls *et al.* (1995) are mainly due to weight effects of the preload. The basis for this explanation was recently confirmed by two studies of Rolls *et al.* (1998) and De Graaf & Hulshof (1996).

Rolls *et al.* (1991) reported that the time interval between preload and subsequent test meal may affect the degree of compensation; a nearly perfect complete compensation was shown in young adults after 30 minutes, and a compensation of 61% to 90% was found after 90 minutes. In the present study we chose a 90-minute time interval because we believe that a 20-minute interval does not measure the physiological effects (e.g. CCK, insulin), but only volume and weight effects of the preload, whereas a longer time interval includes the physiological postingestive and postabsorptive effects as well.

Hunger responses were clearly different between young adults and elderly.

Compared to the no-preload condition, the young adults generally showed larger differences in their appetite ratings than the elderly, suggesting that elderly might be less sensitive than young adults to the energy content of the preload. The results confirm the visual analogue data of Rolls *et al.* (1995) who showed that the older men were less hungry than the young participants in all conditions. Clarkston *et al.* (1997) concluded that aging is associated with reduced desire to eat and hunger after a meal. De Castro (1993) indicated as well that food intake did not affect hunger to the same extent in elderly as it did in younger subjects. He suggested accordingly that food intake in the elderly is primarily influenced by external factors. Therefore, elderly may require an increased conscious control over food intake than younger adults in order to prevent nutrition-related diseases.

No differences in effects of fat and carbohydrate on subjective ratings of the young and elderly adults were found, which is in line with most studies that have been conducted with young adults (Rolls & Hammer, 1995; Rolls *et al.*, 1991; De Graaf *et al.*, 1992; Foltin *et al.*, 1990; Warwick *et al.*, 1993).

The visual analogue ratings of the yoghurts indicate that the young adults and elderly subjects could hardly distinguish the yoghurts on the basis of the sensory properties. The preloads in the present study were perceived as equal in palatability, sweetness and flavour intensity, only not in creaminess. The results of a recent study suggest that palatability may affect satiation but not satiety (De Graaf *et al.*, 1999). Since the preloads were equally palatable, we assume that subjects were responding on the basis of physiological cues related to the energy and macronutrient contents of the yoghurts, with minimal influence of orosensory stimulation.

It should be noted that the elderly participants in our study represented a more heterogeneous group than the younger participants. The elderly subjects had on average a higher BMI and were more restrained than the young adults and children. Lean, unrestrained adults may differ from obese, restrained adults in their ability to regulate the food intake (Rolls *et al.*, 1994). In particular obese women and those concerned with regulation of body weight may be relatively insensitive to the satiety value of fat (Rolls *et al.*, 1994). The higher mean body weight of the elderly subjects followed the trend for increased body weight until late middle age (Cornoni-Huntley *et al.*, 1991). The few restrained elderly (n=6) in the present study did not differ in energy compensation at lunch compared to the non-restrained elderly ($<4\%$; $p>0.3$). We assume that the higher body weight and restrained eating score of the elderly did not highly affect the results of the energy intake at lunch.

The children's food choice differed from the young adults and elderly. The children tended to select the foods they were not allowed to eat or drink at home. They ate more white bread with sweet sandwich spreads and drank less milk compared to the 329 children (aged 4-7) of the Dutch National Food Consumption Survey -a survey carried out in 1992 in order to obtain information on eating behaviors of several population groups in the Netherlands- (Voorlichtingsbureau voor de Voeding, 1993). Davis (1928, 1939) reported that in the absence of adult control, infants were able to choose their own foods to maintain growth and health. She argued that her results provided evidence for an innate, automatic mechanism that regulates both the quantity and the variety of foods selected. However, she only offered nutritious and unsweetened foods that were considered to be good and healthy for children (Story & Brown, 1987). Klesges *et al.* (1991) showed that children selected sweet foods over non-sweet, more nutritious foods when they were allowed to eat in a free choice situation. Children are less sensitive for the sweetness of sucrose than young adults (Zandstra & De Graaf, 1998; James *et al.*, 1997), which could be the reason why the children preferred higher sucrose levels in the foods. Results of the same study of Klesges *et al.* (1991) showed that parental influences had a marked effect on the children's food selection: both the threat of parental monitoring and actual monitoring decreased the sugar content of foods selected and the total energy content of the meal (Klesges *et al.*, 1991). The presence of the five supervising parents at lunch could have influenced the children's food selection. We tried to minimise this influence by instructing the parents prior to the study not to interfere with the children's food choice.

Finally, the results of the present study are limited to a single exposure to the preload. It has been reported that food acceptance patterns are modified by postingestive consequences (positive/negative) of food intake. Likewise, children and young adults can learn to adjust their food intake after high energy preloads within a few exposures (Birch *et al.*, 1990; Kern *et al.*, 1993; Birch & Deysher, 1985). A study with repeated exposure to the preloads might result in a better regulation.

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4

Short- and long-term effects of changes in pleasantness on food intake*

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Abstract

This study concerns the effects of pleasantness on *ad libitum* food intake, liking and appetite over 5 successive days. Pleasantness was manipulated by varying the salt level in bread. Thirty-five students consumed *ad libitum* sandwiches for lunch, made with bread individually perceived as low, medium or high in pleasantness, in a balanced cross-over design. Pleasantness and desire-to-eat the sandwich were rated at first bite, after the consumption of each sandwich and at the end of the lunch. Fullness was rated just before and at several intervals after lunch. On the first day, the students ate less of the least pleasant bread than of the medium and most pleasant bread. On the fifth day, however, consumption of all the breads was similar. For the least pleasant bread, energy intake at lunch, desire-to-eat and fullness all increased over days, whereas these variables remained constant for the medium and most pleasant bread. Mean pleasantness ratings for all breads remained unaltered across the days. We conclude that, with repeated exposure, the desire-to-eat, fullness and intake of a less preferred food can increase over time. Thus, the relationship between pleasantness and food intake changes over this period.

* Appetite 2000, **34**, 1-8

Introduction

Sensory properties of a food have an effect on food intake (Drewnowski, 1997; Rolls *et al.*, 1982; Shepherd, 1988). Several studies showed that enhancing the pleasantness of a food increases its intake within one meal (Bellisle *et al.*, 1984; Bobroff & Kissileff, 1986; Rolls *et al.*, 1982; Hill *et al.*, 1984; Yeomans *et al.*, 1997; Yeomans, 1996; Spiegel *et al.*, 1993; Zandstra *et al.*, 1999).

Until now, however, no study has focused on longer-term relationships of pleasantness with food intake. Over a longer period a shift in pleasantness might occur by 'mere exposure' effects, in which repeated experience leads to increased liking (Pliner, 1982; Shepherd, 1988), or through reinforcing peri- or postingestive effects whereby a previously neutral or even unpleasant taste can become preferred (Rogers, 1990; Ramirez, 1990; Sclafani, 1997; Rozin *et al.*, 1998; Zellner *et al.*, 1983). Another issue is whether or not the relationship between pleasantness and food intake remains stable over time.

The purpose of the present study was to investigate the effects of manipulated pleasantness on appetite, liking and *ad libitum* food intake within single meals eaten over a period of 5 days. We used bread varying in sodium chloride concentration to generate comparable foods varying in pleasantness.

Methods

Subjects

The study population consisted of 25 women and 11 men, all healthy students of Wageningen University. Subjects with a body mass index higher then 25 kg/m^2 and subjects with restrained eating behaviour (score >2.37 for men and >3.24 for women) according to the Dutch Eating Behaviour Questionnaire (Van Strien *et al.*, 1986) were excluded from participation.

Table 4.1 Characteristics of the subjects (mean (SEM)).

	Total (*n*=35)	Women (*n*=25)	Men (*n*=10)
Age (years)	21.1 (0.4)	20.6 (0.3)	22.4 (0.4)
Height (cm)	173.1 (1.4)	169.0 (0.9)	183.3 (1.1)
Weight (kg)	67.8 (1.6)	64.2 (1.4)	76.7 (1.0)
BMI (kg/m^2)	22.5 (0.3)	22.4 (0.4)	22.8 (0.2)
Restraint score[a]	2.1 (0.1)	2.3 (0.1)	1.5 (0.1)

[a] Dutch Eating Behaviour Questionnaire (Van Strien *et al.*, 1986). The minimum value is 1 (no restraint), the maximum value is 5 (high restraint).

One subject dropped out in the second week for personal reasons: his data were excluded from statistical analysis. Table 4.1 shows the main characteristics of the remaining 35 subjects. The study was approved by the Medical Ethical Committee of the Division of Human Nutrition & Epidemiology of Wageningen University. Subjects were told that the study aimed to investigate taste perception and subjective feelings of hunger and satiety in young adults.

Food products

Breads varying in sodium chloride concentrations (low, medium and high levels of NaCl) were used as foods differing in levels of pleasantness. The levels of sodium chloride selected for the experiment were chosen on the basis of results of a pilot study in which 34 volunteers rated the pleasantness of brown bread samples, without spread, varying in sodium chloride concentration (0 to 5 g per 100 g of flour), from not at all pleasant (scored 1) to extremely pleasant (scored 10) at the extremes of ten dashes (Figure 4.1).

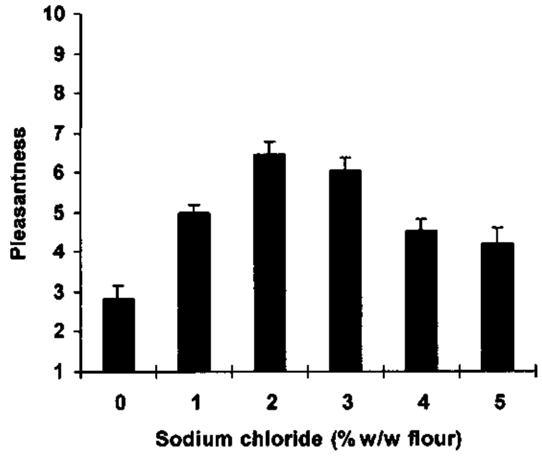

Figure 4.1 Results of the pilot study: mean (+ SEM) pleasantness ratings as a function of concentration of sodium chloride in bread from 1 (not at all pleasant) to 10 (extremely pleasant) (n=34).

The volunteers from the pilot study did not participate in the main study. The pilot study showed that on average the 0% NaCl bread was less pleasant than the 2% NaCl bread, with scores for the 4% NaCl bread in between these. The 5% NaCl bread was not chosen for the main experiment since repeated use of high NaCl concentrations (e.g. 5% solutions of NaCl) may produce oral irritation (Green & Gelhard, 1989). The commercially available bread is 2% NaCl. The breads were baked by a local bakery.

In the main experiment we determined for each subject the stimulus with the lowest, medium and highest pleasantness ratings (LP, MP, and HP breads) according to the individual's averaged pleasantness ratings on day 1 of each set of 5 days per salt level (see Table 4.2).

Table 4.2 Classification of subjects in low, medium and high levels of pleasantness according to the individual's averaged pleasantness ratings on day 1 of each set of 5 days per salt level.

Pleasantness:	Least (*n*=35)	Medium (*n*=35)	Most (*n*=35)
0% (w/w) NaCl bread	28	7	0
2% (w/w) NaCl bread	1	11	23
4% (w/w) NaCl bread	6	17	12

Procedure

Subjects consumed the bread as sandwiches (see below) between 12:15 h and 13:00 h each day for 3 weeks (excepting weekends) in a randomized, balanced cross-over study. The bread was served as part of a sandwich, because subjects in the Netherlands typically eat such lunches (Voedingscentrum, 1998). LP, MP, and HP sandwiches were consumed for 5 days in the departmental dining room. Subjects were seated at large tables in groups of 6-8 subjects and talking was allowed. Subjects ate their usual breakfast on the experimental days at 8:00 h at home and were allowed to drink coffee or tea and to eat a snack at 10:00 h, with the restriction that the breakfast and drink/snack at 10:00 h were the same during the whole study. All subjects were asked not to eat or to drink anything else other than breakfast, drink/snack and lunch until 14:30 h. Thus, subjects were not allowed to eat or drink for about 90 minutes after consumption of the bread. Appetite was measured just before and at several times after lunch.

Subjects wrote down at what time they started and stopped eating. At the end of the lunch they answered the question 'Why did you stop eating?'. Subjects could choose from a number of reasons: 'I felt full', 'The lunch was less tasty', 'Lunch time was over', 'Everyone else was finished', 'For some other reason ... (= open-

ended question)'. Subjects were asked to rank-order as many reasons as they thought important, from most (starting with 1) to least important.
Food intake was measured by weighing amounts served and leftovers. The amounts consumed were calculated to energy and macronutrients with the help of the Dutch nutrient database (NEVO, 1993).

Lunch menu

Brown sandwiches (a 35 g) were cut from loaves and were prepared in advance with standardised amounts of butter (5 g/sandwich) and spread on open slices. Subjects had to eat savoury and sweet spreads in turn within a lunch, in order to eliminate changes in food intake due to differences in spread instead of differences in pleasantness. Savoury spreads included per sandwich ham (13 g), cheese (16 g) and peanut butter (14 g) and sweet spreads comprised per sandwich chocolate paste (10 g), apple syrup (14 g) and jam (14 g). Fruits included apples and oranges, and drinks included coffee, tea, semi-skimmed milk, butter milk and orange juice.

Pleasantness and desire-to-eat

While consuming the lunch subjects rated pleasantness of each sandwich on a 10-point scale labelled at the ends 'not at all pleasant' and 'extremely pleasant' at the first bite, after the consumption of each sandwich and at the end of the lunch. Additionally, just before consumption of each sandwich, subjects rated their desire-to-eat that specific sandwich, with 'not at all strong' and 'extremely strong' on the left- and right-hand side, respectively of a 10-point scale. Each rating made was visible during subsequent ratings.

Appetite

Subjects were instructed to rate their appetite by a mark on a 150-mm line at 12:10, 13:00, 13:15, 13:30, 14:00 and 14:30 h. Appetite was assessed using four phrases: 'appetite for a meal', 'satiety (fullness)', 'faint with hunger' and 'appetite for a snack'. Subjects were instructed that 'appetite for a meal' referred to appetite for a whole meal, either a cooked or a sandwich meal; 'satiety (fullness)' referred to a feeling having eaten too much; 'faint with hunger' referred to a very strong urge to eat and 'appetite for a snack' referred to appetite for any snack. The left- and right-hand side of the rating line were anchored with the terms 'weak' and 'strong' respectively. The ratings were read automatically by an optical mark reader and converted into scores from 1 (weak) to 25 (strong).

Statistical analyses

Differences in energy intake at lunch and subjective feelings of hunger and satiety among lunches with LP, MP and HP bread were tested by repeated-measures analysis of variance, using the procedure GLM (General Linear Model) of the SAS software package (SAS Institute Inc., 1990).

Testing started with an extensive model with group (six subjects in six groups who received the same salt level sequence), pleasantness and day as main effects. To correct for possible carry-over effects, subject nested into group was added, and (group)x(pleasantness) was included as interaction term. In all cases, the model could be simplified to subject, pleasantness, day and (day)x(pleasantness) as main and interaction effects.

Differences in the slopes in change in pleasantness across days were analysed by examining the F-ratio for the (day)x(pleasantness) interaction. Day effects were analysed within each level of pleasantness. In the case of a statistically significant effect ($p < 0.05$) in the analysis of variance, day means among LP, MP, and HP bread were compared by Tukey's test. Chi-squared analyses were run on the frequencies of reasons for ending the lunch.

Changes in pleasantness and desire-to-eat ratings within a meal were calculated by subtracting the response to the last sandwich from the response to the first sandwich. Similar analyses of variance were performed on the changes in pleasantness and desire-to-eat ratings.

Results

Lunch intake

On the first day, intake at lunch was lower for LP than for MP and HP bread, with the means varying significantly [$F(2,67)=6.54$; $p=0.003$], whereas energy intake did not vary among the three levels of pleasantness on the fifth day [$F(2,64)=0.01$; $p=0.99$] (Figure 4.2). This increase across the days in intake of the LP bread was reflected in a borderline significant day effect [$F(4,129)=2.49$; $p=0.05$]. No significant variation across days was found in energy intake at lunch for MP and HP bread [$F(4,135)=1.83$ and 1.53, respectively, both $p>0.13$].

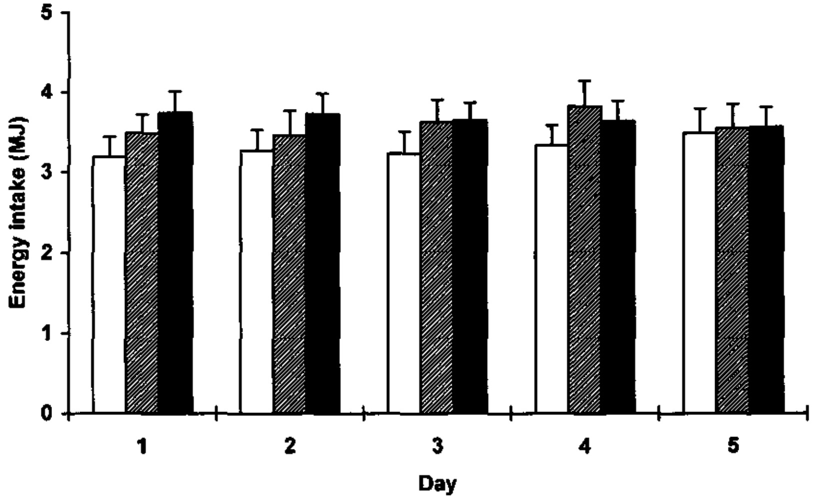

Figure 4.2 Mean (+ SEM) energy intake at lunch, with bread perceived as low (□), medium (▨) and high (■) in pleasantness from day 1 to day 5 (n=35).

Reasons for stopping eating

Mean meal duration (± SEM) for the LP, MP, and HP breads was 21.4 (± 0.7), 22.9 (± 0.8) and 23.3 (± 0.6) min, respectively. On all days, the most frequent response given as first reason was 'I felt full' (Table 4.3).

Table 4.3 Percentages of reasons for ending the lunch on day 1 and day 5 (n=35).

	Pleasantness					
	As first reason			As second reason		
	Least	Medium	Most	Least	Medium	Most
Day 1						
I felt full (%)	26	74	80	9	-	9
The lunch was less tasty (%)	46	14	6	57	43	18
Lunch time was over (%)	-	-	-	3	3	3
Everyone else was finished (%)	-	6	-	-	3	-
Day 5						
I felt full (%)	58	71	77	18	3	9
The lunch was less tasty (%)	18	12	15	18	14	18
Lunch time was over (%)	-	3	-	6	6	3
Everyone else was finished (%)	3	-	-	-	-	-

'I felt full' was least frequently given for the LP bread as first reason [χ^2=9.0; p=0.01]. 'Lunch was less tasty' was more frequently given for the LP bread than for the MP and HP bread situations both as first reason [χ^2=18.1; p=0.0001], and as second reason [χ^2=7.4; p=0.02]. No variations among the three levels of pleasantness were found in frequency or ranking of the other reasons [χ^2<6.3; p>0.05].

Across days, the frequency as first response for the LP bread of 'I felt full' increased from 26% to 58%, and of 'Lunch was less tasty' decreased from 46% to 18%. For the MP and HP bread the frequency and ranking of all reasons remained similar from day 1 to day 5.

Changes in pleasantness

The overall mean pleasantness ratings (± SEM) were 4.5 (± 0.2), 7.0 (± 0.1) and 8.0 (± 0.04) for the LP, MP, and HP bread. The ratings of the LP, MP, and HP breads were stable over time [F(4,467)=0.67; p=0.62].

Changes in pleasantness ratings from first to last sandwich within the meal across the days are shown in Figure 4.3. Within-meal changes in pleasantness ratings for all breads remained the same across the five days [F(4,467)=0.29; p=0.88].

Within each lunch, pleasantness ratings between the start and end of the lunch of the LP bread did not significantly decline on any day [T<1.8; p>0.09]. Pleasantness ratings decreased between the start and the end of the lunch at most days of the MP bread ([T>2.1; p<0.05] for day 2, 3 and 5, and [T<1.9; p>0.07] for day 1 and 4) and of the HP bread ([T>2.3; p<0.03] for day 1, 2, 3 and 5, and [T=1.1; p=0.3] for day 4).

Changes in desire-to-eat (within the lunch)

The overall mean desire-to-eat ratings (± SEM) were 7.3 (± 0.1), 7.5 (± 0.1) and 7.6 (± 0.1) for the LP, MP, and HP breads, respectively. On the first day, the desire-to-eat ratings over the lunch period decreased significantly more with the LP bread than with MP or HP breads (Figure 4.3) [F(2,68)=12.19; p=0.0001]. However, on the fifth day the decrease in desire-to-eat ratings was not significantly different amongst the three levels of pleasantness [F(2,68)=1.86; p=0.2]. This result was reflected in a significant main effect of day for LP bread [F(4,130)=5.56; p<0.001], and a borderline significant interaction effect of (day)x(pleasantness)

[F(8,456)=1.80; p=0.07]. The decrease in desire-to-eat ratings for MP and HP bread remained similar across the five days [F(4,135)=0.62 and 0.89, p=0.65 and 0.47, respectively].

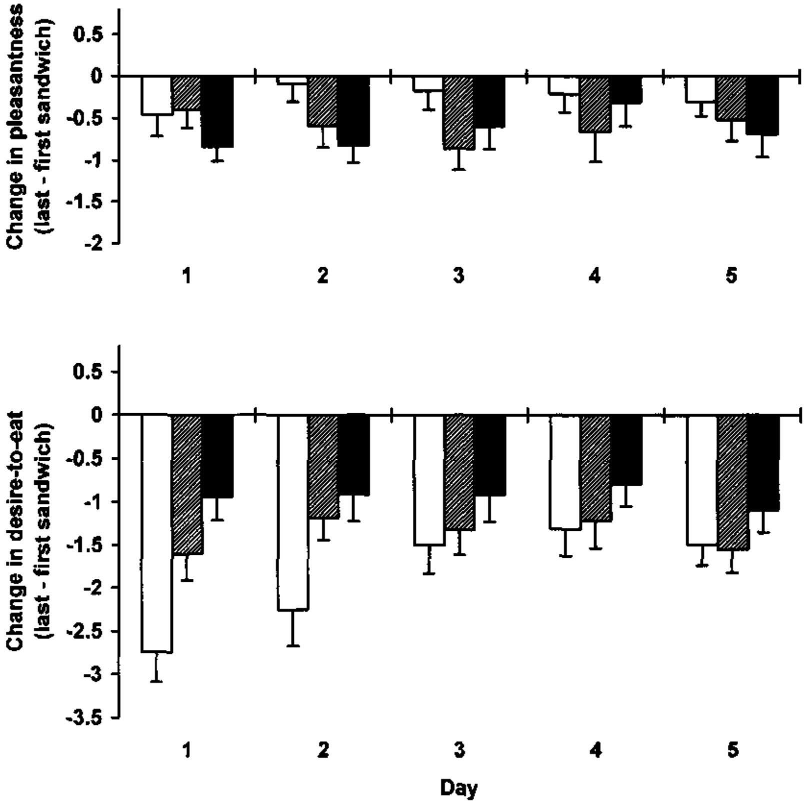

Figure 4.3 Mean (- SEM) change in pleasantness and desire-to-eat of bread perceived as low (□), medium (▨) and high (■) in pleasantness from day 1 to day 5 (n=35). The change is calculated by subtracting the response at the last sandwich from the response at the first sandwich.

Appetite

Mean responses across the days for 'faint with hunger', 'appetite for a snack' and 'appetite for a meal' were about identical, while the responses for 'satiety (fullness)' were the inverse of these. Analysis of variance revealed statistically different ratings for the four appetite items between the three levels of pleasantness averaged across the days [all F(2,3000)>8.36; p<0.001].

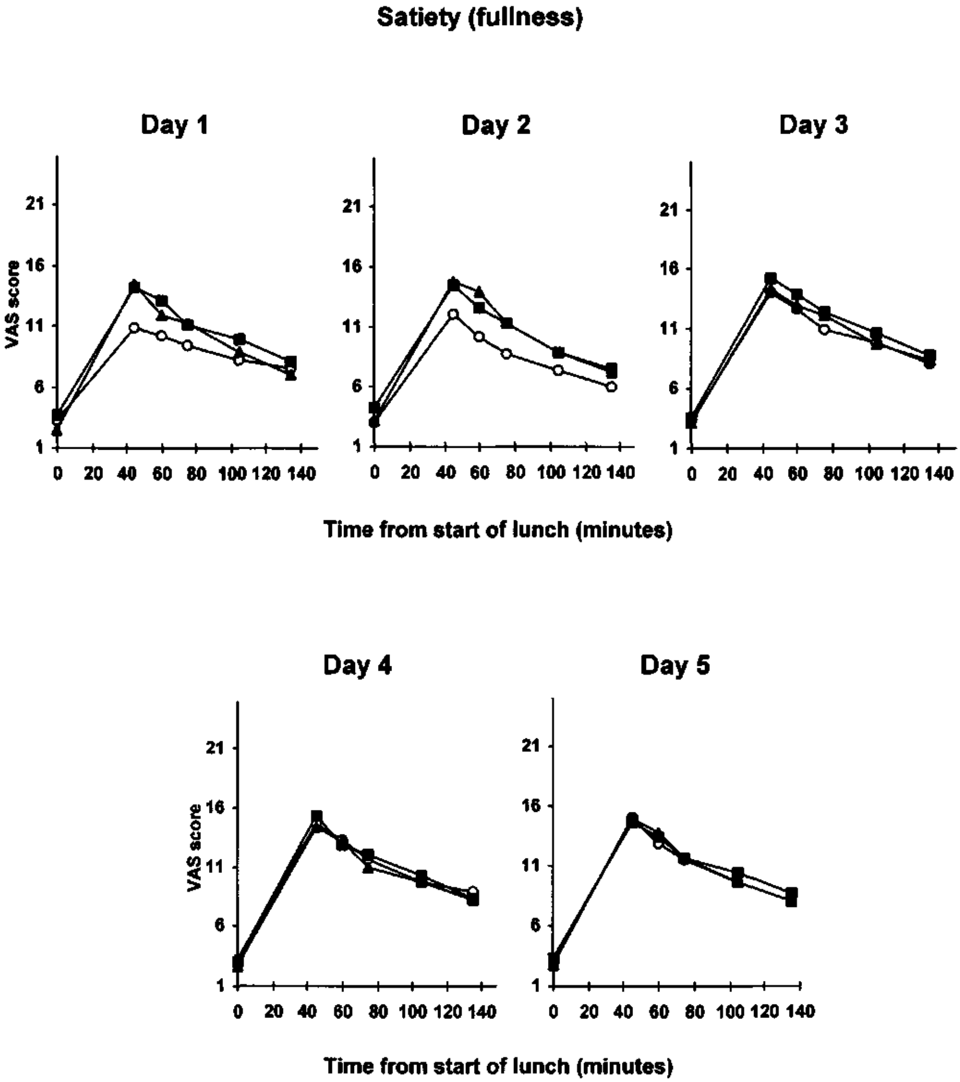

Figure 4.4 Mean ratings 'satiety (fullness)' from 1 (weak) to 25 (strong) after bread perceived as low (–○–), medium (–■–) and high (–▲–) in pleasantness from day 1 to day 5 (n=35). The ratings are shown from just before the lunch (0 minutes), right after the lunch (45 minutes) until 90 minutes after the lunch (135 minutes).

Mean ratings for 'faint with hunger', 'appetite for a snack' and 'appetite for a meal' for the LP bread were higher than for MP and HP breads, and ratings for 'satiety (fullness)' for the LP bread ratings were lower than for both of the other breads.

The rating 'satiety (fullness)' within the LP bread was lower on days 1 and 2 than on days 3, 4 and 5 [$F(4,972)=8.8$; $p=0.0001$] (Figure 4.4). Similar differences were found for 'appetite for a meal', 'faint with hunger' and 'appetite for a snack', but these differences did not reach statistical significance [$F(4,972)<2.25$; $p>0.07$]. The appetite ratings within the MP and HP bread were similar across the days [$F(4,986)<1.21$; $p>0.31$].

Discussion

The results of this study indicate that over a period of repeated exposure to a less preferred food, the pleasantness of that food remained unaltered, while the desire-to-eat, satiety and actual intake of that food increased. During the first day pleasantness had a clear effect on food intake, whereas at the fifth day the relationship between pleasantness and food intake was less clear.

At the first lunch there was a remarkable decrease of desire-to-eat ratings for the less preferred sandwiches compared to the medium and most preferred sandwiches. Over a few days, this decline in desire-to-eat ratings following the consumption of the least preferred sandwiches dissipated, and became similar to ratings made after consumption of the medium and most preferred sandwiches. In contrast, the decrease in pleasantness ratings from first to last sandwich remained unaltered across the days for all sandwich types. The difference in results between desire-to-eat and pleasantness ratings justifies the importance of clarifying the difference between questions about 'liking' for a food and 'desire to eat' a food (Rogers, 1990; Rogers & Blundell, 1990; Berridge, 1996; Gibson & Desmond, 1999). If subjects had only rated pleasantness, they might have used ratings of pleasantness to indicate an underlying change in desire-to-eat, rather than a real change in pleasantness (hedonic value).

Compared to the most pleasant bread, we found a lower intake of the least pleasant bread and not of the medium pleasantness bread on the first day. Yeomans (1996) reported that manipulation of the pleasantness of a pasta meal by the addition of three levels of oregano resulted in a higher intake in the most palatable condition. Bobroff & Kissileff (1986) also showed that, for a liked food, the meal intake, post-meal satiety rating and post-meal palatability ratings were higher than for a disliked food. In contrast with Yeomans (1996) and Bobroff & Kissileff

(1986), Guy-Grand *et al.* (1994) did not observe an increase in food intake during their highly palatable sandwich condition, and suggested that palatability affects meal size only beyond a minimum threshold. Another explanation for this difference could be that the palatability range of foods in the latter study was narrower than in the other two studies (Bellisle *et al.*, 1984). The palatability range between the least and most pleasant bread in the present study was clearly much wider than between the medium and most pleasant bread (overall mean 4.5, 7.0 and 8.0 for the least, medium and most pleasant bread) which could be the reason why the palatability affected intake and appetite in the low and not in the medium pleasantness condition.

The changes across days in energy intake at lunch for the least pleasant bread was in accordance with the changes in appetite ratings: In the first two days subjects consumed less of the least pleasant bread, which resulted in lower scores for 'satiety (fullness)'. Under short-term laboratory conditions, subjective feelings of hunger and satiety correlate significantly with food intake (De Graaf, 1993; Blundell & Rogers, 1991). In contrast to this hunger-food intake association, Mattes (1990) argued that hunger ratings cannot be used as a proxy measure for food intake outside the laboratory. That is, eating can occur when hunger is low (e.g., when highly preferred food is offered unexpectedly) and not at other times when hunger is high (e.g., when other acitivities or cognitive contraints have priority) (Hill *et al.*, 1995).

Overall, 'I felt full' was by far the most frequent reason given for ending the lunch. The 'I felt full' response was less frequently given for the least pleasant bread than for the medium and most pleasant breads. The second most common response given in the least pleasant condition was, as expected, 'The lunch was less tasty'. This is in line with the results of Mook & Votaw (1992) who found that feelings of fullness and reasons such as 'The food is all gone' were chosen much more often than reasons indicating a change in palatability such as 'The foods tastes less good'. In other words subjects do not generally perceive pleasantness to be the most important factor in terminating meals. However, this only holds in the case that the pleasantness remains within the acceptable margins (e.g. above the minimum threshold (Guy-Grand *et al.*, 1994)). If this is not the case, subjects will not even eat or choose the meal.

Pleasantness ratings between the start and the end of the lunch decreased for the MP and HP bread at most days, as has been reported in previous studies (Yeomans, 1996; Hetherington, 1996; Rolls *et al.*, 1984; Rolls *et al.*, 1981;

Yeomans & Symes, 1999), but not for LP bread. These previous studies suggested that the phenomenon of 'sensory specific satiety' might be the explanation for the decline in pleasantness within a meal (Yeomans, 1996). Sensory specific satiety has been investigated extensively by Rolls and collegues (Rolls *et al.*, 1981; Rolls *et al.*, 1984; Rolls, 1986). They showed that, within a meal, the pleasantness and desire-to-eat foods which have been eaten decline more than those foods which have not been eaten. It cannot be concluded on the basis of this study that sensory specific satiety caused the decline in pleasantness ratings, since subjects only rated pleasantness of eaten foods and not of other uneaten foods with (dis)similar sensory properties. However, the decrease in pleasantness between the start and the end of a meal is consistent with the idea of Cabanac (1971) that the pleasantness of taste changes with physiological usefulness or satiety (negative gustatory alliesthesia). The decision to end the lunch would then be hedonically driven, which was, however, not the case for the MP and HP bread in the present study. The relationship between pleasantness, desire-to-eat and satiety within meals also changed over days for the LP bread. This suggests that the underlying mechanism to end a meal is not a simple reduction in pleasantness, but a more complicated acquired response.

Adaptation to the different salt levels of bread might have affected the results of the present study (Bertino *et al.*, 1982; Bertino *et al.*, 1986; Beauchamp *et al.*, 1990). Some studies suggested that as a result of restriction of sodium intake, individuals came to prefer less salty foods and found the previously preferred level of salt in foods to be too salty. We assume that adaptation did not occur within the week of the present study since the shift in preference has been suggested to take a number of weeks (Bertino *et al.*, 1982; Bertino *et al.*, 1986; Beauchamp *et al.*, 1990). Moreover, the perceived intensities of saltiness of the breads of different salt levels remained constant over each set of 5 days.

In conclusion, we found that during a period of repeated consumption of a less preferred food, the desire-to-eat, satiety and the actual intake of that food can increase over time. To our knowledge, the present study is the first which has investigated longer term effects of pleasantness changes on food intake. However, the results of this study have been restricted to bread served as lunch only. More research is needed on long-term effects of pleasantness changes on the intake of other food products served at other meals as well. The understanding of hedonic responses to foods on the longer term, and the differential influences on liking and desire-to-eat, might then be of help in the improvement of the selection and acceptability of healthy food products.

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5

Effects of variety and repeated in-home consumption on product acceptance*

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Abstract

The present study was designed to investigate the effect of variety on long-term product acceptance and consumption in a home-use situation. Subjects (n=105) consumed a meat sauce once a week at dinner at home for a period of 10 weeks. Three variety groups were designed. The *monotony* group (n=45) consistently received the same flavour of meat sauce across all 10 weeks; the *imposed variation* group (n=30) received one from three different flavours of the meat sauce in random order, and the *free choice* (n=30) in variation group was allowed to choose among three flavours of the meat sauce. Results showed a substantial increase in boredom and decline in acceptance ratings after repeated consumption. As hypothesized, this effect was the largest for the *monotony* group and was least pronounced in the *free choice* group, with the *imposed variation* group in between. Consumption data were in line with these acceptance ratings; the *monotony* group consumed less of the food than the *free choice* group over that time. In conclusion, repeated consumption of a food product only once a week at home resulted in a remarkable increase of boredom over time. The boredom effect was the largest for subjects who consistently received the same food, and was least pronounced for subjects who were allowed to choose among three different flavours of the food.

* Submitted

Introduction

Laboratory sensory tests, in which subjects test small food samples in short exposure periods, were originally developed to predict long-term product acceptance and consumption. The predictive validity of these laboratory sensory tests on consumption and long-term acceptability in free living situations has been questioned for years (Zandstra *et al.*, 1999; Monneuse *et al.*, 1991; Daillant & Issanchou, 1991; Pérez *et al.*, 1994; Lucas & Bellisle, 1987; Popper *et al.*, 1989; Meiselman, 1992; Goldman, 1994; Griffin & Stauffer, 1990).

There is ample evidence that respondents change their opinions about a food product after repeated exposure to the same food product over longer periods of time (Porcherot & Issanchou, 1998; Vickers & Holton, 1998; Siegel & Pilgrim, 1958; Schutz & Pilgrim, 1958; Kamen & Peryam, 1961; Rolls & De Waal, 1985). These changes in pleasantness might occur by reinforcing postingestive effects (Sclafani, 1997; Rozin *et al.*, 1998; Birch *et al.*, 1990; Bolles *et al.*, 1981; Zellner *et al.*, 1983) or 'mere exposure' effects (Bornstein, 1989; Zajonc, 1968), whereby repeated exposure leads to an *in*creased liking (Porcherot & Issanchou, 1998; Stang, 1974; Crandall, 1984), or the other way around, by product irritation (e.g. boredom), whereby repeated exposure leads to a *de*creased liking (Siegel & Pilgrim, 1958; Van Trijp, 1995; Lévy & Köster, 1999; Hetherington *et al.*, 1998). A better understanding of both phenomena is of importance for product marketeers, particularly those involved in fast moving consumer goods (e.g. foods). They need to know not only whether consumers initially like the food product, but above all whether consumers continue to like and buy the food product after repeated consumption.

To what extent repeated consumption affects food acceptance may depend on the availability of different varieties of particular foods and the degree of freedom of choice (Kamen & Peryam, 1961; Pliner *et al.*, 1980; Deci & Ryan, 1987). The number of studies which have focused on the effect of repeated in-home consumption on food product acceptance is quite limited (Mela *et al.*, 1993; Porcherot & Issanchou, 1998; Lévy & Köster, 1999; Stubenitsky *et al.*, 1999). The main purpose of this trial was therefore to determine long-term changes in hedonic responses under a normal domestic eating environment. A second purpose was to determine the effect of freedom of choice on long-term acceptance of foods. Three different variety groups were designed: a *monotony* group, an *imposed variation* group, and a *free choice* in variation group. As the food product, we chose a meat sauce for dinner with three different flavours.

Materials and methods

Subjects

Subjects were 105 consumers recruited by posters at student-flats and university buildings and by advertisements in local papers. After stratification for sex and age, subjects were randomly allocated to one of the three variety groups. Exceptions were cohabiting subjects, who were placed in the same group, and subjects who did not like a flavour, who were placed in the group which did not receive that specific flavour. In total, 6 subjects did not like a specific flavour: 3 subjects did not like curry, 1 subject did not like saté and 2 subjects did not like sweet-and-sour. Table 5.1 shows some characteristics of the subjects classified by group. Subjects were told that the study consisted of tasting and eating different meat sauces for dinner over a 10 week period at home. The study was approved by the Medical Ethical Committee of the Division of Human Nutrition & Epidemiology, and subjects gave their informed consent prior to the study.

Table 5.1 Subject characteristics classified by group (mean ± SD).

	Monotony (*n*=45)	*Imposed variation* (*n*=30)	*Free choice* (*n*=30)
Age (years)	29.1 (11.2)	27.2 (11.4)	27.2 (11.8)
Height (cm)	176.5 (9.2)	177.6 (8.5)	178.4 (10.0)
Weight (kg)	70.5 (12.6)	72.2 (11.3)	70.3 (10.8)
BMI (kg/m^2)	22.5 (30)	22.8 (3.1)	22.0 (2.4)
Men/women (*n*)	22/23	16/14	15/15

Food product

A meat sauce for dinner was used as the food product. We selected three meat sauces of the same brand that varied in flavour: curry, saté and sweet-and-sour. The meat sauces were supplied in branded one-person portions (285 gram/jar). The meat sauces were from the same batch and were commercially available in supermarkets at the time of the study. Subjects were randomly assigned to three groups to consume the meat sauce once a week at dinner at home across 10 weeks:

1. ***Monotony*** group consistently received the same meat sauce; curry (*n*=11), saté (*n*=11) or sweet-and-sour sauce (*n*=23). The latter group comprised twice as

much subjects since this group was also part of another study, of which the results are not relevant to this study.

2. ***Imposed variation*** group received the curry, saté and sweet-and-sour sauce in random order (*n*=30). Different random orders were generated for each subject, with the restriction that each subject received each flavour at least three times.
3. ***Free choice** in variation* group was on every single consumption occasion allowed to choose among the curry, saté and sweet-and-sour sauce (*n*=30).

Procedure

Subjects consumed the meat sauce once a week at dinner at home for a period of 10 weeks. Subjects came to the department every other week. The *monotony* and *imposed variation* group got two jars each time they came to the department. A label on the cap of the jar indicated in which week the meat sauce had to be consumed. The *free choice* group received 6 jars (2 jars of each flavour) every other week, so that they were able to freely choose a flavour each week. The 4 remaining jars had to be returned. The three groups were instructed that these meat sauces should be the only meat sauces consumed over the period of the study, and that the meat sauces were solely for their own personal consumption and should not be shared with others.

Prior to the study subjects were instructed how to cook the meat sauce. Instructions about preparation were also printed on the label of the jar. The meat sauce was simple to prepare. One needed only to add the sauce to meat, and to cook the sauce together with the meat for 20 minutes. Thus, preparation of the meat sauce in the home environment was likely to be well controlled. Subject were allowed to consume the meat sauce on any day of the week, and also to use the meat sauce in combination with all different kinds of meal components (e.g. meat/meat replacer, vegetables, rice). Subjects could choose how much of the jar they wanted to use, with the restriction that they would eat at least a quarter of the jar contents.

Measurements

Subjects gave their answers on 10-point category scales.

Just before consumption of the meat sauce subjects rated:

1. Probability of choosing the meat sauce (If the choice was yours, what is the probability that you would choose this sauce for this occasion?) from 'not at all

probable' to 'extremely probable' (except for the *free choice* group to whom this question was irrelevant);
2. Subjective feelings of hunger from 'not at all hungry' to 'extremely hungry';
3. Desire-to-eat the meat sauce from 'not at all strong' to 'extremely strong'.

While consuming the meat sauce subjects rated:
1. Interestingness (How interesting do you think the taste of the sauce is?) from 'not at all interesting' to 'extremely interesting';
2. Perceived sweetness intensity from 'not at all sweet' to 'extremely sweet';
3. Perceived flavour intensity from 'not at all strong' to 'extremely strong'.

At the point of meal completion subjects rated:
1. Pleasantness from 'not at all pleasant' to 'extremely pleasant';
2. Boredom (To what extent did you get bored with the flavour of the sauce?) from 'not at all bored' to 'extremely bored';
3. Consumption (How much of the sauce did you eat?) was self-reported on a labelled 4-point category scale (1=about a quarter of the jar, 2=about half the jar, 3=about three quarters of the jar, 4=the whole jar).

Statistical analyses

All data analyses were performed with an SAS statistical software package (SAS Institute Inc., 1990). A value of $p < 0.05$ was used as criterion for statistical significance.

The influence of treatment effects on product ratings (both sensory and acceptance ratings) was analysed by repeated-measures analysis of variance (GLM-SAS). We used models with each product rating as dependent variable and group, week and the interaction (group)x(week) as independent variables, with Tukey's test to determine a main group effect. Hunger was added as covariate since hunger significantly contributed to the model.

In order to evaluate changes in product ratings over time, individual-level regression analyses were performed to calculate individual slopes over time, with week and hunger as independent variables and each product rating as dependent variable. Analyses of variance were then used to test possible differences among the three groups in changes in mean slopes over time, with a Tukey-test for group comparisons.

Results

Inspection of the boredom ratings in Figure 5.1 shows that the boredom effect was the largest for the *monotony* group and was least pronounced in the *free choice* group, with the *imposed variation* group in between. Analysis of variance of the boredom ratings showed a significant effect of week [F(9,1017)=5.4; p=0.0001] and group [F(2,1017)=73,1; p=0.0001]. Analysis of variance on the individual slopes confirmed the differences in changes over time among the three groups [F(2,102)=6.19; p=0.003] (Table 5.2).

Table 5.2 Mean slopes (SEM) reflecting changes in acceptability of meat sauces as a function of week and degree of variety.

	Monotony (*n*=45)	*Imposed variation* (*n*=30)	*Free choice* (*n*=30)
Choice probability	-0.17 (0.05)‡	-0.14 (0.04)‡	-
Interestingness	-0.19 (0.04)‡	-0.08 (0.03)‡	-0.01 (0.03)§
Desire to eat	-0.23 (0.04)‡	-0.11 (0.03)‡	-0.08 (0.03)‡, §
Pleasantness	-0.23 (0.04)‡	-0.11 (0.04)‡	-0.07 (0.04)§
Boredom	0.29 (0.05)‡	0.13 (0.04)‡, §	0.09 (0.04)‡, §
Perceived sweetness intensity	0.02 (0.03)	-0.02 (0.04)	0.003 (0.03)
Perceived flavour intensity	-0.07 (0.03)‡	0.01 (0.03)	0.03 (0.04)
Relative amount eaten	-0.02 (0.004)‡	-0.02 (0.004)‡	-0.01 (0.004)‡

‡ Significantly different from zero, p<0.05

§ Significantly different from the *monotony* group, p<0.05

Pleasantness ratings followed an inverse pattern of boredom (*r*=-0.69; p<0.0001), with a decline in ratings over time for the *monotony* group and *imposed variation* group but not for the *free choice* group (Figure 5.2). The results from the repeated measures analysis showed a significant main effect of group [F(2,1016)=75.44; p=0.0001] and week [F(9,1016)=5.31; p=0.0001]. The mean of the individual slopes of pleasantness ratings over time of the *monotony* group was lower than the *imposed variation* group, and the mean of the individual slopes of pleasantness ratings of the *imposed variation* group was lower than the *free choice* group [F(2,102)=4.97; p=0.009].

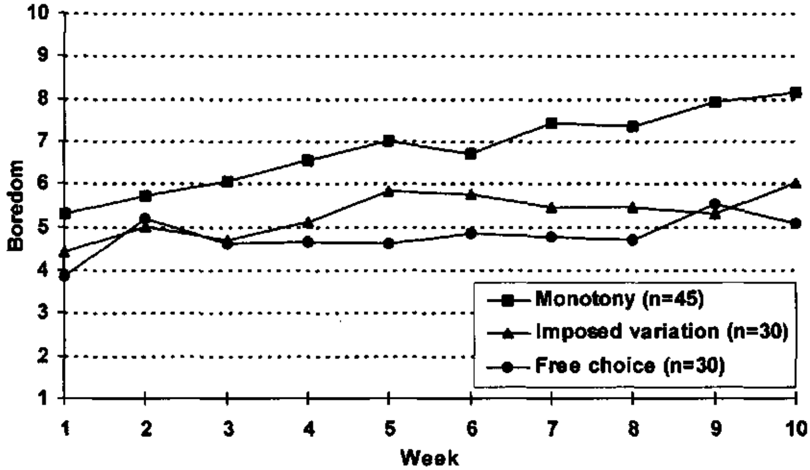

Figure 5.1 Mean boredom ratings over time of monotony group (n=45), imposed variation group (n=30) and free choice group (n=30).

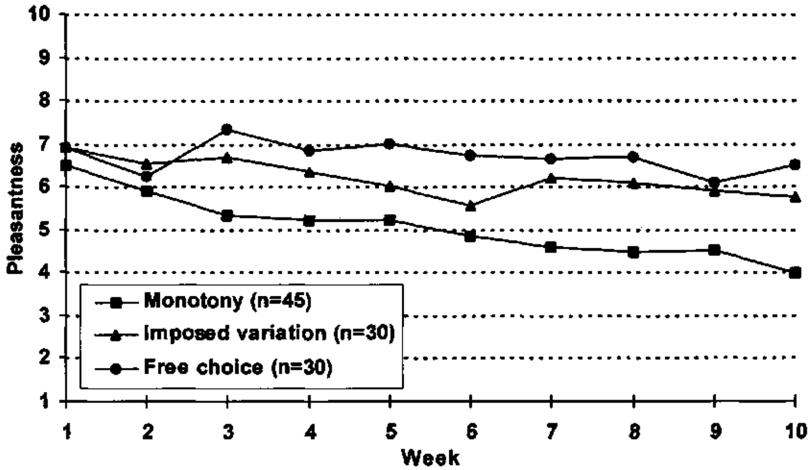

Figure 5.2 Mean pleasantness ratings over time of monotony group (n=45), imposed variation group (n=30) and free choice group (n=30).

Similar differences in slopes were found for interestingness and desire-to-eat [F(2,102)>4.7; p<0.01]. The high spearman correlations between pleasantness and these variables concur with this finding (*r*=0.83 for interestingness; *r*=0.69 for desire-to-eat).

For amount eaten a significant main effect of group [F(2,1016)=14.59; p=0.0001] and week [F(9,1016)=2.94; p=0.002] was found, but the mean slopes over time did not differ among the three groups [F(2,102)=1.29; p=0.28] (Figure 5.3).

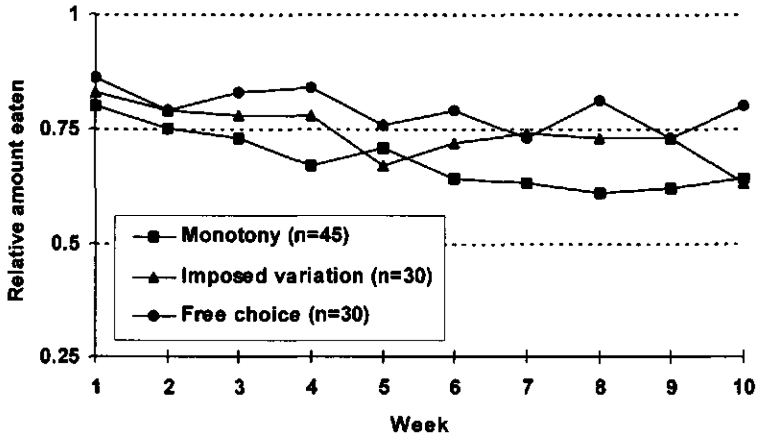

Figure 5.3 Mean (self-reported) relative amounts of sauce consumed over time of monotony group (n=45), imposed variation group (n=30) and free choice group (n=30).

Discussion

Repeated consumption of a meat sauce once a week at dinner at home over a 10-week period resulted in a substantial increase in boredom and decline in acceptance ratings. This effect was the largest for the *monotony* group and was least pronounced in the *free choice* group, with the *imposed variation* group in between. Consumption data were in line with these acceptance ratings; the *monotony* group consumed less of the food than the *free choice* group over that time.

The substantial increase in boredom and decrease in desire-to-eat and pleasantness of the *monotony* group is remarkable given the low frequency of consumption. The results of the *monotony* group are consistent with two early studies which found that repetitive eating results in a persistent decrease in preference and consumption. In these two studies subjects received respectively two alternate daily menus for 22 days (Siegel & Pilgrim, 1958) and 41 items in 4 menus for 37 days (Schutz & Pilgrim, 1958). A reduction in preference of foods which were eaten daily for 6 months was also found in a study at an Ethiopian refugee camp where a limited variety of foods was available (Rolls & De Waal, 1985). The frequency of consumption in these previous studies was much higher compared to the present study in which one single food (within an otherwise regular diet) was eaten only once a week during a 10-week period. A possible explanation for the boredom effect in our study may lie in the type of food product used. Staple foods, for example, are more resistant to boredom than other foods. In the two early studies repeated consumption did not change the palatability of staple foods such as dairy products, bread, or coffee, whereas other foods such as meat and vegetables followed a decline in preference (Siegel & Pilgrim, 1958; Schutz & Pilgrim, 1958). The meat sauce in the present study is a non-staple food that might have been perceived as inappropriate for weekly consumption (Schutz, 1988).

Self-control had a major impact on product acceptance after repeated exposure to foods: subjects were least bored when they were allowed to choose among three alternatives compared to those whose serving order was determined by someone else. In line with this finding, Kamen and Peryam (1961) found a higher overall satisfaction among those who were allowed to plan their own menu compared to those who received preplanned menus. In the psychological literature, the work of Deci on intrinsic motivation (cognitive evaluation theory: (Deci, 1975; Deci & Ryan, 1987)) may provide the theoretical framework for the larger boredom effect in the *imposed variation* group than in the *free choice* group (Van Trijp *et al.*, 1996). Cognitive evaluation theory (Deci, 1975) gives a central role to humans' needs for autonomy as underlying mechanism for intrinsic motivation. It argues that the effects of extrinsic motivation (e.g. rewards) on the initiation and maintenance of intrinsically motivated behaviours depends on the psychological meaning persons give to the contextual factors (Deci, 1975; Deci & Ryan, 1987). Deci and Ryan (1987) categorised the contextual factors as to whether they were generally perceived as 'controlling' or 'autonomy supportive'. Controlling contexts are those that are experienced by persons as controlling their behaviour (e.g. pressuring them toward particular outcomes), whereas autonomy supportive contexts are experienced as supporting autonomy behaviour (e.g. providing and encouraging them to make their own choices). Controlling contexts tend to undermine intrinsic motivation and autonomy supportive contexts tend to enhance intrinsically motivated behaviours. In consumer choice contexts, variety has especially a motivated effect in choice contexts that are perceived as autonomy supportive (e.g. providing freedom of choice) rather than controlling (e.g. lack of choice or imposed choice). In the present study, the *imposed variation* group had similar stimulation levels in variety as the *free choice* group, but the controlled choice context of the *imposed variation* group resulted in a less boredom-reducing effect than the autonomy supportive choice context of the *free choice* group. The reader is referred to Van Trijp (1995) for a more elaborate discussion on the relevance of this theory for variety seeking in food choice behaviour.

In marketing, product boredom or need for variety has been described in a choice behaviour model on variety seeking (Van Trijp & Steenkamp, 1992; Van Trijp, 1994; Van Trijp *et al.*, 1996). The intensity of variety-seeking in product choice behaviour is influenced by both person-related and product-related determinants (Van Trijp, 1994). Three underlying mechanisms for variety-seeking behaviour are curiosity (cognitive response), boredom (with the choice process) and attribute satiation (sensory perceptual response) (Van Trijp *et al.*, 1996). In the latter case, changes in preference over repeated consumption are caused by satiation with specific attributes repeatedly delivered by the consumed product. In the *monotony* group, we found an increase in boredom and a decline in acceptance ratings which supports the idea that specific product attributes can change long-term consumer preferences. However, subjects in the *imposed variation* group were more bored with the food product than in the *free choice* group. This would indicate that boredom with the product (i.e. sensory specific satiety) is not only influenced by specific product attributes, but also by psychological and situational factors beyond sensory and hedonic determinants (Stubenitsky *et al.*, 1999; Hetherington *et al.*, 1998).

In the present study subjects were allowed to eat the meat sauce at home with all different kinds of meal components and to eat as much of the sauce as they wished. These (realistic) uncontrolled factors could have caused unequal levels of exposure to the sauce among subjects and might have biased the results. Nevertheless, an advantage of testing under a normal domestic eating environment is that the results are more applicable to the 'real world'.

In conclusion, extended home-use resulted in a significant boredom effect over time. Subjects were least bored when they were allowed to choose among three alternatives compared to those whose serving order was determined by someone else.

Acknowledgements

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6

Effects of learned flavour cues on short-term regulation of food intake in a realistic setting*

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Abstract

The present study examined the effects of repeated mid-morning consumption of novel-flavoured low and high energy yoghurt beverages on subsequent energy intake at lunch, in 69 adults under actual use conditions. Subjects consumed 200 ml of low and high energy yoghurt beverages (67 and 273 kcal/200 ml, respectively), with 20 exposures to each drink on alternate days. Analyses focused on the development of compensation for the differences in energy content of the beverages, due to learned satiety. Results revealed incomplete energy compensation for the beverages, both at first exposure and also after 20 exposures. Relative to the no yoghurt condition, energy intake compensation (mean ± SEM) averaged 39 ± 36% for the low energy yoghurt and 17 ± 9% for the high energy version, with no evidence of any change in compensation with repeated exposures. When the flavours of the yoghurt beverages were covertly switched after 20 exposures, subjects increased their energy intake after the high energy yoghurt beverage containing the flavour that was previously coupled with the low energy yoghurt beverage. *Vice versa*, however, when subjects switched to the low energy yoghurt beverage containing the high energy flavour, subjects ignored the flavour cue and ate the same lunch size regardless of the energy in the yoghurt beverage. We conclude that adults do not readily learn or express adjustments for a reduced energy content of a food after repeated exposures. The long-term consumption of lower energy versions of commercially foods might therefore assist in decreasing total energy intake over longer periods of time.

* Submitted

Introduction

The marketing of a wide range of reduced-energy foods in the western world should theoretically enable a lower consumption of fat and/or sugar in the diet, and improve weight control, in line with current dietary recommendations (Voedingscentrum, 1998). However, relatively little is known about how these reduced-energy foods influence patterns of food choice and consumption, and whether they are indeed effective in reducing energy intake in real life situations over extended periods of time.

Energy intake compensation is the tendency to adjust food intake after ingesting foods with varying energy densities. There are many laboratory studies on energy intake compensation in humans, but these studies have shown inconsistent results. Some of these studies have shown complete compensation (Rolls *et al.*, 1991; Birch *et al.*, 1993; Birch & Deysher, 1985) but the majority of these studies have reported incomplete energy intake compensation using single meal preloading designs (Hulshof *et al.*, 1995; Blundell *et al.*, 1993; De Graaf *et al.*, 1992; Rolls *et al.*, 1994; Zandstra *et al.*, 2000). Although the majority of these studies reported an incomplete energy intake compensation, it might be that repeated experience is needed to allow regulatory mechanisms to operate. However, longer-term laboratory studies (2 days-11 weeks) have also shown both precise (Foltin *et al.*, 1992; Louis-Sylvestre *et al.*, 1989; Foltin *et al.*, 1988; Mattes *et al.*, 1988) and very poor (Mattes, 1996; Louis-Sylvestre *et al.*, 1994; Lissner *et al.*, 1987; Kendall *et al.*, 1991; Stubbs *et al.*, 1995; Stubbs *et al.*, 1996) compensation of energy intake for a reduction of the energy or macronutrient content of the diets.

Only few studies in humans have focused on the development of energy intake compensation based on learned cues ('conditioned satiety') (Birch *et al.*, 1990; Birch & Deysher, 1985; Booth *et al.*, 1982; Booth *et al.*, 1976; Specter *et al.*, 1998). The development of compensatory eating with repeated experience results from learned associations between internal (e.g. satiety) and sensory cues (e.g. flavour) (Booth *et al.*, 1976). Results of some studies suggest that young children (3-5 y) can learn to adjust their food intake perfectly within a few exposures in an anticipatory way, based on learned associations between foods' sensory cues and the consequences of ingesting the foods (Birch *et al.*, 1990; Birch & Deysher, 1985; Johnson *et al.*, 1991). In contrast to children, two studies on conditioned satiety in adults found incomplete energy intake compensation (Booth *et al.*, 1982; Booth *et al.*, 1976), while a recent study with adults found no energy intake compensation following repeated exposure (Specter *et al.*, 1998). One tentative explanation for the incomplete compensation with adults is that these studies have not been of sufficient duration to allow more precise learning to occur. The study period in the adults' studies was at maximum 5 exposures. It might therefore be necessary to examine development of compensation over longer periods of time, especially under realistic rather than tightly controlled experimental conditions.

The objectives of the present study were twofold. The first objective was to examine energy compensation over an extended period of time (20 exposures) under actual use conditions. We investigated adjustments in short-term regulation of food intake following repeated exposure to a fixed low energy and high energy yoghurt beverage. The yoghurt beverages were paired with two novel tropical fruit flavours 'cupuacu' and 'cherimoya' instead of familiar flavours to avoid preexisting flavour-postingestive consequence associations that might impede the formation of new associations. Food intake was assessed by measuring food intake at lunch in response to covert changes in energy content of the two yoghurt beverages. The second objective was to assess adjustments in energy intake that might occur when the coupling of the flavours with the low and high energy yoghurt beverage was reversed after 20 exposures.

Materials and methods

Subjects

Sixty-nine healthy adults were recruited by public advertisements, according to two selection criteria: 18-65 years old of age and subjects had to like yoghurt beverages. Twenty-two subjects participated in the study at the Institute of Food Research (Reading, UK) and forty-seven subjects participated at Wageningen University (Wageningen, the Netherlands). The two test sites followed identical protocols, and the data were combined for all analyses. Table 7.1 shows the main characteristics of all sixty-nine subjects.

Table 7.1 Characteristics of the subjects (mean (SD)).

	Total (*n*=69)	Men (*n*=31)	Women (*n*=38)
Age (years)	29.6 (13.4)	30.0 (15.4)	29.2 (11.7)
BMI (kg/m^2)	23.0 (3.3)	23.5 (3.1)	22.6 (3.5)
Restraint score[a]	2.3 (0.9)	2.0 (0.9)	2.5 (0.8)

[a] The restraint score was assessed with Van Strien's Dutch Eating Behaviour Questionnaire (1986). The minimum value is 1 (no restraint), the maximum value is 5 (high restraint).

The study was approved by both the Human Research Ethics Committee of the Institute of Food Research and the Medical Ethical Committee of the Department of Human Nutrition & Epidemiology of Wageningen University. Subjects were not informed about the true purpose of the study, but were told that the experiment was to assess sensory perception and feelings of hunger and satiety. All subjects gave their informed consent before participation in the study.

Study design

Subjects consumed high or low energy yoghurt beverages as a mid-morning snack (between 10-00 and 11-00 h) over a period of 9 weeks (5 days a week) in the usual setting (e.g., at home or work). The study consisted of two parts: a conditioning period of 8 weeks and a flavour reversal period of 1 week (Table 7.2). Energy intake compensation was assessed by measuring food intake at lunch.

Table 7.2 Design of the study.

	Conditioning[a]			Flavour reversal[b]
	Week 1	Week 2	Week 3 *etc.*	Week 9
Flavour subgroup 1 (*n*=37)[c]	ABBAB[d]	BABAA	ABABB *etc.*	DCDC
Flavour subgroup 2 (*n*=32)[c]	DCCDC[d]	CDCDD	DCDCC *etc.*	ABAB

[a] Completion of lunch diaries for measuring food/energy intake compensation at exposure 0, 1, 5, 10, 15 and 20 of each yoghurt beverage

[b] Completion of lunch diaries at exposure 1 and 2 of each yoghurt beverage

[c] The presentation order of the low and high energy yoghurt beverages was randomised within subjects and within each week under the condition that subjects were exposed a maximum of three days in a row.

[d] A = High energy yoghurt beverage paired to novel flavour 'cupuacu'
B = Low energy yoghurt beverage paired to novel flavour 'cherimoya'
C = High energy yoghurt beverage paired to novel flavour 'cherimoya'
D = Low energy yoghurt beverage paired to novel flavour 'cupuacu'

Yoghurt beverage

The composition of the yoghurt beverages is shown in table 7.3. The energy difference of the low energy and the high energy yoghurt beverage was approximately 200 kcal per 200 ml serving. Subjects consumed 200 ml of the yoghurt beverage. Based on this portion size, the low energy yoghurt beverage provided 67 kcal (≈ 280 kJ) and the high energy yoghurt beverage provided 273 kcal (≈ 1148 kJ).

Table 7.3 Composition of the high energy and low energy yoghurt beverage.

g/100 g serving	High energy yoghurt beverage	Low energy yoghurt beverage
Skimmed milk (3.5% protein, 4.6% lactose)	69.81	69.81
Cream (40% fat)	16.25	-
Sugar	12	-
Stabilizer (pectin)	0.30	0.75
Emulsifier (mono-diglycerides)	0.50	-
Water	1.14	25.41
Aroma	0.03-0.1	0.017-0.07
Aspartame	-	0.014
Acesulfame K	-	0.014
Maltodextrin	-	2
Inulin	-	2
Total energy	573.6 kJ (= 136.6 kcal)	139.7 kJ (= 33.3 kcal)

The yoghurt beverages were developed and prepared especially for this experiment by Friesland Coberco Dairy Foods (Research Centre Deventer, the Netherlands). The yoghurt beverages had a long shelf life of 3 months, and were packed in 200 ml single serve plain cans. The low energy and high energy yoghurt beverages were composed to be as similar as possible in pleasantness and sensory characteristics such as sweetness, taste and texture.

Two novel tropical fruit flavours 'cupuacu' and 'cherimoya' were chosen on the basis of a pilot study. In the pilot study 6 yoghurt beverages with novel flavours (nashi, pitahaya, cupuacu, cherimoya, pumpkin and popcorn) were ranked by 60 subjects from most to least preferred. The two middle preferred ones -cupucacu and cherimoya- were chosen as target flavours, to allow for increases and decreases in acceptance following exposure. Novel flavours were chosen to avoid possible associations between a particular energy load and a familiar flavour (e.g. chocolate = 'loaded with calories'). Cupuacu and cherimoya are tropical fruits with flavours unfamiliar to Western European consumers.

Conditioning period

Half of the subjects received one flavour coupled with the high energy yoghurt beverage, and the other flavour coupled with the low energy yoghurt beverage. The remaining subjects received the alternative coupling of flavours with energy (Table 7.2). Subjects consumed the yoghurt beverages 5 times a week (Monday to Friday) for 8 weeks. The presentation order of the high and low energy yoghurt beverages was randomised within subjects and within each week under the condition that subjects were exposed a maximum of three days in a row to the same version. Subjects were exposed to each flavoured yoghurt beverage 20 times overall. Before the start of the conditioning period, there was a 'no yoghurt' experimental day as well (exposure 0).

Flavour reversal period

In the last week of the experiment, the flavours of the yoghurt beverages were covertly switched. Subjects received the low energy yoghurt beverage with the flavour previously coupled with the high energy yoghurt beverage, and the high energy yoghurt beverage with the flavour previously coupled with the low energy yoghurt beverage (Table 7.2). The presentation of the yoghurt beverages was randomised within subjects within each set of two days. Subjects were exposed to each flavoured yoghurt beverage 2 times.

Energy intake at lunch

Energy intake at lunch was recorded by means of a food diary. In the conditioning period, subjects filled in their lunch diaries after consuming the yoghurt beverage on days corresponding to 0, 1, 5, 10, 15 and 20 exposures to the low and high energy yoghurt beverage. In the flavour reversal period, subjects filled in their lunch diary on each of the four experimental days.

Subjects received detailed instructions for completing the food diaries before the start of the study. These instructions were included in each food diary as well. The food diaries were checked and coded by experienced dieticians. In case of unclear food records subjects were asked about this within a week. Energy intake at lunch was calculated using Dutch and British food composition tables for the Dutch and British subjects, respectively (Stichting Nederlands Voedingsstoffenbestand, 1993; Holland *et al.*, 1991).

Procedure

On each experimental day, subjects were instructed to consume their normal breakfast but nothing other than water and their usual 1 cup of coffee or tea between breakfast and the consumption of the yoghurt beverage, and between the consumption of the yoghurt beverage and lunch.

Subjects were instructed to consume the yoghurt beverage between 10-00 and 11-00 h, and their lunch between 12-00 and 13-00 h in the usual home or work setting. Subjects came in every week to collect the yoghurt beverages for one week, and a sticker on each can indicated on which day it should be consumed.

At the end of the study subjects filled in a short debriefing question regarding contingency awareness. Subjects were asked to identify which yoghurt (flavours) they believed were high and low energy. In total 43 out of 69 subjects identified the correct yoghurt beverage as high energy, which is not quite significantly better than chance (χ=5.7; p=0.06). Of these 43 subjects, 1 indicated he was "completely sure", 17 were "rather sure", 18 were "rather unsure" and 7 were "completely unsure".

Statistical analyses

Data were analysed using the statistical program SAS (SAS Institute Inc., 1990). Results were considered statistically significant if $p < 0.05$, and data are described as mean ($\pm$ SEM).

For the conditioning period, a repeated-measures analysis of variance (excluding the 'no yoghurt beverage' condition) followed by Tukey tests for post hoc comparisons were used to analyse the effects of repeated exposure to the yoghurt beverages on total energy intake at lunch. We tested time effects separately for the low and high energy yoghurts. This was done using a model with energy intake at lunch as dependent variable, and exposure (1, 5, 10, 15 and 20), gender, restrained eating, awareness and flavour subgroup (see Table 7.2 in text) as independent variables.

To assess trends in energy intake at lunch over time (8 weeks), individual-level regression analyses were performed for lunch intakes following each yoghurt beverage (excluding the 'no yoghurt' condition) to calculate individual slopes over time, with exposure as independent variable and energy intake at lunch as dependent variable. Paired two tailed t-tests were then used to compare the mean of the individual slopes over time of the low and high energy yoghurt beverage.

Energy compensation at lunch for the low vs. the high energy yoghurt beverage was calculated in two ways. The first method assessed the response relative to the 'no yoghurt beverage' condition. The percent compensation at lunch was calculated as the difference between the energy intake at lunch in the 'no yoghurt beverage' condition and the energy intake for each of the yoghurt beverage conditions, divided by the energy of the yoghurt beverage, multiplied by 100. A positive value indicates that subjects (partial) compensate at lunch, while a negative value indicates that subjects overeat at lunch. The second method assessed to what extent subjects compensated for the difference in energy between the yoghurt beverages. The difference of energy intake at lunch in the low energy yoghurt beverage condition and the high energy yoghurt beverage condition was divided by the difference in energy between the yoghurt beverages (868 kJ) and multiplied by 100. For this calculation, values > 100% indicate overcompensation for the energy difference between the yoghurt beverages and values < 100% indicate undercompensation.

During the flavour reversal period, mean energy intake of exposure 1 and 2 were compared with the energy intake of the mean for exposure 1-20 of the conditioning period using paired two-tailed t-tests.

Results

Conditioning period

Subjects showed incomplete energy compensation for the low and high energy yoghurt beverage (Table 7.4 and Figure 7.1). For the low and high yoghurt beverage, mean energy intakes at lunch over the whole experimental period did not differ from the 'no yoghurt' condition [($T=1.1$; $p=0.29$) and ($T=1.9$; $p=0.06$), respectively], or between the low and high yoghurt beverage (2395 ± 94 kJ vs. 2313 ± 78 kJ, respectively) [$T=0.67$; $p=0.5$]. Relative to the 'no yoghurt' condition, energy intake compensation averaged 39 ± 36% for the low yoghurt beverage and 17 ± 9% for the high energy version. Percent compensation at the 1st and 20th exposure, respectively, was 38 ± 45% and 4 ± 53% for the low energy yoghurt condition and 16 ± 10% and 21 ± 10% for the high energy yoghurt condition.

Analysis of variance on repeated exposure revealed that energy intake at lunch (excluding the no yoghurt beverage condition) was not affected by repeated exposure to the low energy yoghurt beverage [$F(4,333)=0.23$; $p=0.86$] or high energy yoghurt beverage [F(4,330)=0.60; p=0.67]. As expected, men had overall higher intakes at lunch than women (2884 ± 93 kJ vs. 1982 ± 58 kJ and 2753 ± 82 kJ vs. 1956 ± 47 kJ for men vs. women, and after low and high energy yoghurt beverage, respectively) [gender effect F(1,333)>77.2; p=0.0001]. Restrained eating, flavour subgroup and awareness of energy manipulation (determined in post-trial debriefing) were not associated with energy intake at lunch after either the low or high energy yoghurt beverage ([F(1,333)<3.0; p>0.09] and [F(1,330)<3.7; p>0.07] for the low and high energy yoghurt beverage, respectively).

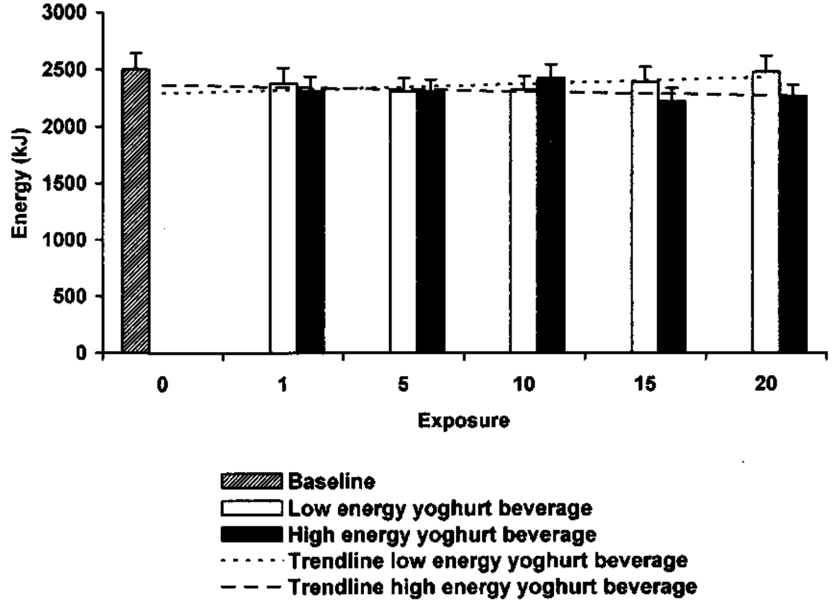

Figure 7.1 Mean (+ SEM) energy intake at lunch (expressed in kJ) on days corresponding to 0 (= baseline), 1, 5, 10, 15, and 20 exposures to the low and high energy yoghurt beverage (280 and 1148 kJ/200 ml, respectively) (n=69).

Energy intake at lunch at exposures 1 and 20, respectively, increased from 2382 ± 139 kJ to 2481 ± 137 kJ with the low energy yoghurt beverage and decreased from 2317 ± 122 kJ to 2271 ± 98 kJ with the high energy yoghurt beverage. However, the positive mean individual slopes over time of the low energy yoghurt beverage did not significantly differ from the negative mean individual slopes over time of the

Table 7.4 Mean energy intake at lunch ± SEM and mean percent compensation[a,b,c] ± SEM for the low and high energy yoghurt (280 and 1148 kJ/200ml, respectively) during A) the conditioning period and B) the flavour reversal period (n=69).

A	Low energy yoghurt beverage/ Low energy flavour		High energy yoghurt beverage/ High energy flavour		
Exposure	Lunch intake (kJ)	Compensation compared to no yoghurt beverage (%)[a]	Lunch intake (kJ)	Compensation compared to no yoghurt beverage (%)[a]	Intake compensation low energy yoghurt beverage compared to high energy yoghurt beverage (%)[c]
0	2505 ± 137	-	2505 ± 137	-	-
1	2382 ± 139	38 ± 45	2317 ± 122	16 ± 10	11 ± 15
5	2309 ± 120	46 ± 47	2313 ± 103	17 ± 12	4 ± 13
10	2324 ± 120	65 ± 45	2434 ± 117	6 ± 13	-9 ± 15
15	2395 ± 131	23 ± 54	2218 ± 118	25 ± 11†	21 ± 16
20	2481 ± 137	4 ± 53	2271 ± 98	21 ± 10†	25 ± 16
Means (1-20)	2395 ± 94	39 ± 36	2313 ± 78	17 ± 9	10 ± 7

B	Low energy yoghurt beverage/ High energy flavour		High energy yoghurt beverage/ Low energy flavour		
Exposure	Lunch intake (kJ)	Compensation compared to means (1-20) (%)[b]	Lunch intake (kJ)	Compensation compared to means (1-20) (%)[b]	Intake compensation low energy yoghurt beverage compared to high energy yoghurt beverage (%)[c]
1	2557 ± 137	-48 ± 40	2509 ± 129	-20 ± 8†	-4 ± 15
2	2539 ± 149	-56 ± 40	2440 ± 127	-11 ± 9	14 ± 16
Means (1-2)	2542 ± 129	-52 ± 33	2496 ± 110‡	-16 ± 7†, ‡	6 ± 11

† Significantly different from zero, $p < 0.05$

‡ Significantly different from mean of exposure 1-20 in the conditioning period, $p<0.05$ (paired t-test)

[a] Formula: ((kJ consumed after 'no yoghurt' - kJ consumed after yoghurt beverage)/kJ in yoghurt beverage)*100

[b] Formula: ((kJ consumed at mean of exposure 1-20 - kJ consumed after yoghurt beverage)/kJ in yoghurt beverage)*100

[c] Formula: ((kJ consumed after low energy yoghurt beverage - kJ consumed after high energy yoghurt beverage)/868 kJ)*100

high energy yoghurt beverage [T=1.41; p=0.16], which is in line with the analysis of variance on repeated exposure (Table 7.5). None of the mean individual slopes over time were significantly different from zero [T<1.59; p>0.12]. Amongst men only, comparisons of the mean individual slopes indicate adjustment of energy intake at lunch via increased and decreased intakes after repeated exposure to the low and high energy yoghurt beverage, respectively [T=2.23; p=0.03]. With respect to the other characteristics, there were no differences between the mean individual slopes for the low and high energy yoghurt beverage (women [T=0.52; p=0.61], unrestrained eaters [T=0.98; p=0.32], restrained eaters [T=1.45; p=0.17], subjects who were aware of energy manipulation [T=1.19; p=0.25], subjects who were not aware of energy manipulation (determined in post-trial debriefing) [T=0.87; p=0.39]).

Table 7.5 Mean individual slopes ± SEM of energy intake at lunch over time in kJ (excluding the 'no yoghurt' beverage condition) for the low and high energy yoghurt beverage during the conditioning period.

	Change in energy intake over time in kJ	
	Low energy yoghurt beverage	High energy yoghurt beverage
Yoghurt beverage type (*n*=69)	11.8 ± 12.6	-8.5 ± 7.8
Men (*n*=31)	36.1 ± 22.8	-18.9 ± 15.1[†]
Women (*n*=38)	-8.0 ± 12.7	0.1 ± 6.8
Unrestrained eater (*n*=54)	10.1 ± 15.5	-7.0 ± 8.6
Restained eater (*n*=15)	17.8 ± 15.8	-13.8 ± 18.3
Aware of energy manipulation[a] (*n*=18)	26.5 ± 28.9	-12.7 ± 18.7
Not aware of energy manipulation (*n*=51)	6.6 ± 13.7	-7.0 ± 8.3

[†] Significant difference between low- and high energy yoghurt beverage, p<0.05

[a] Only subjects who were "rather sure" or "completely sure" about the energy manipulation (determined in post-trial debriefing)

Flavour reversal period

Compared to the mean of exposure 1-20 of the conditioning period, subjects overate in response to the high energy yoghurt beverage containing the flavour that was previously associated with the low energy yoghurt beverage [T=2.84; p=0.006] (Table 7.4). Subjects did not adjust their energy intake at lunch when subjects switched from the low energy yoghurt beverage containing the low energy flavour to the low energy yoghurt beverage containing the flavour that was previously associated with the high energy yoghurt beverage ([T=1.16; p=0.25].

Discussion

The results of the present study indicate that on average subjects had an incomplete energy compensation for low and high energy beverages (67 and 273 kcal/200 ml, respectively), both at first exposure and also after 20 exposures. Relative to the 'no yoghurt' condition, energy intake compensation averaged 39 ± 36% for the low energy yoghurt and 17 ± 9% for the high energy version, with no evidence of any change in compensation with repeated exposures. When the flavours of the yoghurt beverages were covertly switched after 20 exposures, subjects increased their energy intake after the high energy yoghurt beverage containing the flavour that was previously coupled with the low energy yoghurt beverage. *Vice versa*, however, when subjects switched to the low energy yoghurt beverage containing the high energy flavour, subjects ignored the flavour cue and ate the same lunch size regardless of the energy in the yoghurt beverage.

The principal finding in the present study is that subjects had an incomplete energy intake compensation for each flavoured yoghurt beverage, both with no prior experience and conditioned after 20 exposures. The finding of incomplete compensation confirms the results of other studies on learning of satiety with adult humans (Booth *et al.*, 1982; Booth *et al.*, 1976; Specter *et al.*, 1998; Louis-Sylvestre *et al.*, 1994). Two early studies showed some compensation for an 200 kcal difference in soup and 280 kcal difference in dessert following four exposures (Booth *et al.*, 1982; Booth *et al.*, 1976), and two more recent studies found no compensation for changes in fat in ice cream and a lunch dish following five and four exposures, respectively (Specter *et al.*, 1998; Louis-Sylvestre *et al.*, 1994).

Studies with younger age groups, however, have reported accurate learned caloric adjustments after repeated experience. Children (3-5 years old) adjusted their food intake after high energy preloads within eight exposures (Birch *et al.*, 1990; Birch & Deysher, 1985; Kern *et al.*, 1993; Johnson *et al.*, 1991), and adolescent males had a precise energy compensation for an 200 kcal difference in a snack after five exposures (Louis-Sylvestre *et al.*, 1989). In response to energy density in the diet, children may respond to more internal physiological cues (e.g. satiety), whereas older subjects may respond to more external factors other than hunger and satiety (e.g. time of day, presence of other people) (Birch & Fisher, 1997). Consequently, age-related impairments might account for the discrepancies found previously and in the present study in the ability to learn to associate reduced satiety with a decrease in energy density, and to adjust the energy intake accordingly. Future studies are required to confirm this age-related impairment in the ability to condition caloric adjustments after repeated exposure.

During the flavour reversal period, subjects increased their energy intake when they switched to the high energy yoghurt beverage containing the flavour that was previously coupled with the low energy yoghurt beverage. This presumably reflects a learned anticipated adjustment to the caloric content of the yoghurt beverage based on the sensory properties rather than the true energy content of the yoghurt beverage. Subjects in other studies were similarly misled by the flavour cue when it did not match the caloric consequence (i.e. when the flavours were covertly switched) (Shaffer & Tepper, 1994; Tepper *et al.*, 1991). In addition, energy intake at lunch remained stable when subjects switched to the low energy yoghurt beverage containing the flavour that was previously associated with the high energy yoghurt beverage. This is consistent with previous studies which suggested that, under conditions of unlimited access to food, humans compensate more readily for decreases than for increases in energy intake (Foltin *et al.*, 1988; Mattes *et al.*, 1988). The latter illustrates an appetite control system that contains regulatory mechanisms which prevent undereating, but has few defences against overeating (passive overconsumption) (Blundell & Green, 1996; Blundell *et al.*, 1993; Caputo & Mattes, 1992; Mattes *et al.*, 1988). However, during the flavour reversal period in the present study, subjects were only exposed to each flavoured yoghurt beverage 2 times. Tepper *et al.* (1991) reported that the flavour-switch effect was reversed within five days. This suggests a flexibility of the learning or regulatory mechanisms following repeated exposures that warrants further attention.

Only 18 out of 69 subjects were rather sure (n=17) or completely sure (n=1) about the energy manipulation of the yoghurt beverages. Previous studies reported that compensatory eating behaviour might be further enhanced by knowledge of the manipulation (Aaron *et al.*, 1994; Shide & Rolls, 1995; Caputo & Mattes, 1993). In the present study awareness of the energy manipulation did not enhance the strength of the conditioned response. As the sample size of subjects who were aware about the energy manipulation was quite limited, it is possible that the influence of awareness on the strength of conditioning might have been underestimated (Baeyens *et al.*, 1996; Baeyens *et al.*, 1990). Ideally, the results of overt and covert manipulations should be compared.

It should be noted that the present study had some limitations. We investigated the short-term effects of consumption of low and high energy yoghurt beverages as mid-morning snack on energy intake at the subsequent lunch. Consequently, we do not know whether compensatory changes occurred later in the day. Subjects might have adjusted their energy intake not only in the single lunch meal, but maybe also on a longer-term basis (24 h). Previous studies found that the flavour cue had the greatest effect on the very next meal than over the next 24 hours (Shaffer & Tepper, 1994; Specter *et al.*, 1998). We therefore assume that in the present study the compensatory effects would not have been more profound later in the day. Another point is the methodology used to measure the energy intake at lunch in free-living subjects. Subjects were instructed to complete food diaries and on return to the laboratory a trained dietician reviewed the records in the presence of the subject. Absolute honesty and accuracy cannot be guaranteed in this type of measurement. In addition, this method might lack the sensitivity to detect small conditioned adjustments given the large standard deviations in estimated energy intake. Thus, this methodology is clearly a compromise between the restrictions in eating behaviour of laboratory studies and the difficulties associated with the measurement of spontaneous behaviour in free living situations (Gatenby *et al.*, 1995).

We conclude that adults do not acquire accurate conditioned adjustments for the energy content in food after repeated experience with the food in free living situations. The use of lower energy commercial versions of foods might therefore be helpful in decreasing total energy intake over longer periods of time.

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7

No evidence of energy-based conditioned food preferences in human adults under realistic eating conditions*

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Abstract

This study investigated whether adult humans alter their liking for flavours associated with higher- and lower-energy versions of a food eaten regularly within natural eating situations. Subjects (*n*=108) consumed higher- and lower-energy (HE and LE, 273 and 67 kcal per 200 ml serving, respectively) versions of initially novel-flavoured yoghurt drinks 20 times each over a period of 8 weeks, in a within-subjects design. A conditioning group consumed 200 ml at home or at work, at mid-morning each day, while a 'mere exposure' group consumed 10 ml. Pairings of HE and LE with 2 novel target flavours were fully counterbalanced. Flavour liking and ranking tests were carried out at home and in the laboratory, and subjects were debriefed after the trial to assess their recognition of the experimental manipulations (contingency awareness). There was no evidence for changes in flavour liking or ranking based on the energy content of the yoghurts. Data from the in-home and laboratory tests were uniform in showing no greater increase in liking or preference for the HE-paired flavour than for the LE-paired flavour. Ratings for both target flavours increased similarly over time amongst both the conditioning and the 'mere exposure' groups, especially compared to other, non-target flavours. There was no evidence that either group was aware of the manipulations or pairings. The results suggest that robust energy-based conditioned flavour preferences are not readily acquired or expressed by adult humans under normal eating conditions. Evaluative conditioning (via flavour-flavour or other non-postingestive effects) may offer a more plausible explanation for the observed results, and is perhaps dominant in influencing acquisition of food liking in realistic eating situations. Energy content *per se* in foods may not greatly influence development of flavour preference or liking in adult humans; however, further evaluative conditioning research with food stimuli under realistic eating conditions is needed to confirm this conclusion.

* Submitted

Introduction

The question how liking (affective evaluations) of specific sensory attributes of foods are acquired has been the interest of many researchers in different disciplines. Although robust theories have been established for the acquisition of food dislikes, aversions, and disgust (Rozin, 1989; Rozin, 1990; Rozin & Zellner, 1985), the area of the acquisition of food preferences and likes is still very much under investigation, and various theories have been suggested to explain the possible learning processes and mediators involved.

An apparently simple explanation of how food preferences and likes are acquired has been that of 'mere' exposure (Zajonc, 1968). This effect has received considerable empirical support in studies using a wide range of visual and auditory stimuli, and also in small number of food-related studies using laboratory animals (Hill, 1978) and humans (Pliner, 1982; Stang, 1975). Although 'mere' exposure has been mentioned as an explanation for learned food preferences (Pliner, 1982; Zellner *et al.*, 1983; Rozin, 1989; Rozin, 1990; Capaldi, 1996), no robust theory has been developed as to why the changes in affect occur (Bornstein, 1989; Zajonc & Markus, 1991). Although some form of exposure is critical in order for other processes to operate to influence liking (Rozin, 1989), 'mere' exposure may often be used as a description rather than specifying any real mechanism or effectors, as exposure to foods via consumption is not really 'mere', in that it provides opportunities for many other types of potentially reinforcing events to occur.

One of the mechanisms most widely advocated to explain the acquisition of food preference and likes is flavour-nutrient conditioning (Sclafani, 1995; Baeyens *et al.*, 1996b; Capaldi, 1996). In this case, the postingestive consequences of a nutrient or food act as the reinforcer in the learning process, so that liking or preference for a flavour increases if the flavour is associated with the 'beneficial' nutritional consequences of normal eating (Sclafani, 1995; Booth *et al.*, 1982; Capaldi, 1996; Booth, 1985; Rozin & Zellner, 1985). In the basic experimental paradigm, 'cue' flavours (conditioned stimulus, CS) are repeatedly paired (contingently associated) with the nutrient or food under investigation (unconditioned stimulus, UCS), and alternative flavours with a control food. During the conditioning period, the association between the 'cue' flavour and the postingestive consequences of the nutrient is facilitated (Sclafani, 1995; Capaldi, 1996), and changes in preference can be attributed to an acquired response to the cue flavour (Birch *et al.*, 1990; Kern *et al.*, 1993; Johnson *et al.*, 1991; Sclafani, 1995).

Flavour-nutrient conditioning has been successfully demonstrated in many studies, particularly in rats, using specific macronutrients such as starches, sugars and fat (Sclafani, 1997; Sclafani, 1995; Pérez *et al.*, 1996; Pérez *et al.*, 1995; Pérez *et al.*, 1994; Sclafani, 1992; Elizalde & Sclafani, 1990a; Elizalde & Sclafani, 1988b), or energy (Bolles *et al.*, 1981; Warwick *et al.*, 1997; Mehiel & Bolles, 1988, 1989). Human flavour-nutrient conditioning has been observed in children using calories from sugar (Birch *et al.*, 1990) or fat (Kern *et al.*, 1993; Johnson *et al.*, 1991) as the UCS, and in adults using protein (Gibson *et al.*, 1995) and starch (Booth *et al.*, 1976, 1982; Booth, 1985), and also non-nutrients such as caffeine (Richardson *et al.*, 1996; Yeomans *et al.*, 1998; Rogers *et al.*, 1995). However, we are aware through personal communications and experience that many more 'unsuccessful' (and therefore largely also unpublished) attempts to demonstrate flavour-nutrient conditioning in humans have been undertaken. This and the small number of 'successful', published flavour-nutrient conditioning studies in humans indicate the difficulty of demonstrating the phenomenon in human subjects. One of the main reasons for this is the complex design issues required to properly control all aspects of conditioning trials (Sclafani, 1995; Gorn, 1991). Another explanation is that (adult) humans typically enter the experiments with a large and well-established set of pre-existing affective and cognitive responses to foods and flavours.

Food preference and liking clearly are not acquired solely through postingestive consequences of the food alone, and Rozin (1977, 1989, 1990) and Rozin & Zellner (1985) have suggested that, rather than flavour-nutrient conditioning, other processes or forms of reinforcement such as cultural and social influences, may play a major role in influencing the acquisition of food likes. 'Evaluative' conditioning is an example of such a process and has been systematically investigated by Baeyens *et al.* (1990b, 1992, 1995b; 1996a, b, c, d) and Rozin *et al.* (1998). In evaluative conditioning, the contingent pairing of a neutral stimulus (CS) with a liked or disliked stimulus (UCS) may result in the neutral stimulus itself acquiring positive or negative hedonic valence (Baeyens *et al.*, 1996b, c; Rozin & Zellner, 1985). 'Flavour-flavour' conditioning is a form of evaluative conditioning in which a novel food flavour (CS) is paired with an already established liked or disliked flavour (UCS), resulting in a change in preference for the CS (Capaldi, 1996; Sclafani, 1992). Flavour-flavour conditioning has been successfully used in humans (Zellner *et al.*, 1983; Baeyens *et al.*, 1996a; Baeyens *et al.*, 1995b; Fanselow & Birk, 1982; Baeyens *et al.*, 1990b).

There have been both successful (Sclafani, 1995; Zellner *et al.*, 1983; Baeyens *et al.*, 1996b, c), and unsuccessful (Rozin *et al.*, 1998) demonstrations of evaluative conditioning procedures. However, as in flavour-nutrient conditioning, the design parameters are complex. One of those parameters is contingency awareness; i.e. whether subjects are in some way aware of the association between the CS and UCS (Baeyens *et al.*, 1990a, 1992, 1993; Baeyens & De Houwer, 1995a). In 'classical' conditioning, contingency awareness is often viewed as a necessary feature for development of a conditioned response to a novel CS (McSweeney & Bierley, 1984; Gorn, 1991). However, Baeyens *et al.* (1990a, 1992, 1993, 1996a, b, c) have shown that awareness is unrelated to the success of evaluative conditioning, and similar conclusions were drawn by Rozin *et al.* (1998). Arguably, contingency awareness could be problematic in interpreting studies of nutrient-based flavour preference conditioning with human adults, because of the potential introduction of further cognitively-based attitudes into the stimulus evaluations. Nevertheless, studies on the acquisition of caffeine-reinforced flavour preferences show that those can also occur in the absence of contingency awareness.

Despite extensive research in the area, and the great theoretical and practical importance that has been placed on it (Rozin *et al.*, 1998), few studies have investigated the acquisition of changes in affective responses to foods within real life situations (Rozin *et al.*, 1998; Birch *et al.*, 1990; Kern *et al.*, 1993; Johnson *et al.*, 1991). These studies used flavour-nutrient conditioning with children in a school situation (Birch *et al.*, 1990; Kern *et al.*, 1993; Johnson *et al.*, 1991), or evaluative conditioning with non-food (social) stimuli and students in home or activity situations (Rozin *et al.*, 1998). The present research focused on whether adults come to prefer flavours associated with higher-energy versions of foods under realistic eating conditions (e.g. at home or at work). We included a 'mere exposure' group to control for changes in response to the flavour CS due to effects other than postingestive events. Contingency awareness was assessed in order to identify whether this could be an important factor influencing the development of conditioned responses.

Materials and methods

The study was carried out jointly at the Institute of Food Research in Reading (now located in Norwich), UK and the Wageningen University in Wageningen, the Netherlands. Both test sites followed identical protocols for the conditioning trial, with minor differences in some of the related evaluation procedures (indicated in text), and data were combined for the purposes of most analyses. The protocol was approved by both the Human Research Ethics Committee of the Institute of Food Research and the Medical Ethics Committee of the Department of Human Nutrition & Epidemiology of Wageningen University.

Study design

Subjects (*n*=108) consumed high and low energy yoghurt drinks as a mid-morning snack over a period of 8 weeks (Monday-Friday) at home. The yoghurt drinks (described below) were paired with two target flavours 'cupuacu' and 'cherimoya'. The subjects were allocated to two groups: 'conditioning' and 'mere exposure'.

Conditioning group

In the conditioning group (*n*=89), half the subjects received 200 ml of a higher-energy (HE) yoghurt drink paired with one flavour, and 200 ml of a lower-energy (LE) yoghurt drink with another flavour. For the other half of the subjects, the pairing of the specific flavours and the energy content of the yoghurt drinks was reversed. In addition to the in-home conditioning procedure, subjects took part in 3 laboratory sessions before, during and after the conditioning period.

'Mere exposure' group

The 'mere exposure' group (*n*=19) followed exactly the same procedures as the conditioning group, but consumed only a spoonful (10 ml) of the yoghurt drinks.

Yoghurt drinks and flavours

Yoghurt drinks

Flavour-nutrient conditioned preferences are suggested to be larger when a cue flavour is mixed in a food rather than given before a food (Capaldi, 1996), and is also more consistent with the real-life situation of interest. We therefore chose a combined UCS-CS vehicle (yoghurt drinks). Yoghurt drinks were chosen because they were considered suitable as a common mid-morning snack in both the UK and the Netherlands, and they would not be considered novel or unusual in the eyes of the subjects. In addition, a yoghurt vehicle was used in successful demonstrations of flavour-nutrient conditioning in children (Kern *et al.*, 1993; Johnson *et al.*, 1991). The yoghurt drinks were developed and manufactured especially for this experiment by Friesland Coberco Dairy Foods (Research Centre Deventer, the Netherlands).

Each 200 ml serving of the HE contained 273 kcal (1148 kJ), and LE version 67 kcal (280 kJ). The additional energy in the HE version came equally from fat and carbohydrate, and the composition of the drinks is reported elsewhere (Zandstra *et al.*, 2000). The HE and LE drinks were developed to be similar in taste, flavour, texture and appearance, based on pilot tests with subjects who did not take part in the main study (data not shown). For the conditioning group, the yoghurt drinks were packed in 200 ml single-portion unlabelled cans with a shelf life of 3 months. The yoghurt drinks for the 'mere exposure' group (10 ml) were packed into small transparent tubs with a lid.

All cans and tubs were unlabelled except for the day & date of consumption that was printed on the side of the can. A coloured sticker on the can indicated to the subjects that a questionnaire had to be completed that day. The subjects collected the yoghurt drinks from the researchers every week, and kept them refrigerated until use.

Flavours

The two target flavours were novel to ensure that subjects had no previous experience or established liking for them. The target flavours were selected out of 6 novel flavours before the start of the study in a ranking and preference test with 60 subjects in the UK and the Netherlands who did not take part in the main study. Four of the 6 flavours (cupuacu, cherimoya, nashi, pitahaya) were tropical fruit flavours from South American origin, not known to consumers in Europe; the other 2 flavours (pumpkin and popcorn) were novel to yoghurt.

Table 7.1 Results (mean ± SD) of the 6 novel flavours in the pilot study (n=60).

Flavour	Overall liking[a]	Rank from liking score	Rank score[b]	Rank from ranking
Nashi	4.3 ± 2.0	6	174	5
Pitahaya	5.8 ± 1.7	2	227	3
Cupuacu	4.7 ± 2.2	4	182	4
Cherimoya	4.5 ± 1.6	5	173	6
Pumpkin	5.8 ± 2.0	3	238	2
Popcorn	6.0 ± 2.2	1	263	1

[a] On a 9-point scale from 'like not at all' to 'like extremely'

[b] Ranking from '1 = most liked' to '6 = least liked', with rank number = (6xrank1) + (5xrank2) + (4xrank3) + (3xrank4) + (2xrank5) + (1xrank6)

The two middle preferred flavours ('cupuacu' and 'cherimoya', both ranked and scored 4-5 on a 9-point hedonic scale) were chosen as the target flavours to enable an increase or decrease in preference during the conditioning period. The 4 remaining flavours were used in the 3 laboratory sessions (non-target flavours, see below). The results from the pilot test with the 6 novel flavours are presented in Table 7.1.

Subjects

The subjects were recruited through an advertisement in the local paper (UK and the Netherlands) or from the local university (the Netherlands). Subject inclusion criteria were: between 18-65 years, regular yoghurt eaters, not lactose intolerant or pregnant, not on a diet, not having a condition that might affect taste or smell, being able to come to the research site every week to collect the yoghurt drinks, and being available during the time of the study. Subjects were not informed about the true purpose of the study, but were told that the experiment assessed hedonic quality of the yoghurt drinks over time. Subjects completed an informed consent form before participation, and were paid for their participation at the end of the study.

Protocol

Table 7.2 Presentation design of the yoghurt drinks (n=108).

Group	*n*	Subgroup	Week 1	Week 2 *etc.*
Conditioning[a]	46	1	ABBAB[b]	BABAA *etc.*
	43	2	DCCDC[b]	DCDCC *etc.*
Mere exposure[a]	10	3	ABBAB[b]	BABAA *etc.*
	9	4	DCCDC[b]	DCDCC *etc.*

[a] The presentation order of the low- and high energy yoghurt drinks was randomised within subjects and within each week under the condition that subjects were exposed to each yoghurt drink a maximum of three days in a week

[b] A = High energy yoghurt drink paired with flavour 'cupuacu'
B = Low energy yoghurt drink paired with flavour 'cherimoya'
C = High energy yoghurt drink paired with flavour 'cherimoya'
D = Low energy yoghurt drink paired with flavour 'cupuacu'

1. In-home

Every weekday (Monday-Friday) over 8 consecutive weeks (i.e., total of 40 days), subjects in the conditioning group consumed a yoghurt drink as a mid-morning snack between 10 -11 am. Subjects in the 'mere exposure' group followed the same protocol but only consumed a spoonful (10 ml) of the yoghurt drink each day. The subjects were allowed to have their habitual breakfast and coffee/tea intake in the morning, but were instructed to consume only 1 cup of coffee or tea during the rest of morning. Each subject consumed each version of the yoghurt drink 20 times in total. The presentation order of HE and LE yoghurts was randomised for each subject during the 8-week period, with the provision that each subject received one version no more than 3 times in a week. Table 7.2 shows the protocol for the conditioning and 'mere exposure' groups.

In-home conditioning assessment

Subjects completed a set of questionnaires after 0, 1, 5, 10, 15 and 20 exposures of each version of yoghurt drink during the experimental period. The questionnaires (in English or Dutch) assessed the following:

1. Liking ratings

Subjects rated 'overall liking' for the yoghurt drink whilst consuming the yoghurt drink on a 9-point hedonic category scale with the endpoints labelled (from 'like not at all' to 'like extremely').

2. Hunger and satiety ratings

Only the conditioning group completed a hunger- and satiety questionnaire before and after consumption of the yoghurt drink, and just before lunch. The questionnaire assessed hunger, thirst and desire to eat on 9-point category scales, with the endpoints labelled (from 'not at all' to 'extremely hungry/thirsty/desire to eat').

2. Laboratory sessions

All subjects took part in the laboratory sessions before (day 0), during (after 10 exposures), and at the end of the conditioning period (after 20 exposures). Before the start of the study (at day 0), subjects also completed the Dutch Eating Behaviour Questionnaire (Van Strien *et al.*, 1986), and indicated their age, gender, height and weight. At the end of the study, their weight was measured again.

Laboratory conditioning assessment

In each laboratory session, subjects completed 3 tests:

1. Two-choice preference test: liking ratings

Subjects rated each of the 2 target flavours for 'overall liking' on a 9-point category scale with the endpoints labelled (from 'not at all' to 'extremely'). The two target flavours were presented in an iso-caloric version of the drink (half LE and half HE), and were coded with 3-digit numbers in a presentation order randomised for each subject. Subjects were also asked to choose which of the yoghurt drinks they preferred, replicating the two-choice preference test procedure of Birch and colleagues (Birch *et al.*, 1990; Kern *et al.*, 1993; Johnson *et al.*, 1991). However, these results are not presented as they were in agreement with and did not add to the interpretation and conclusions of the study.

2. Hedonic and sensory ratings of target and non-target flavours

Subjects rated the 2 target flavours and the 4 non-target flavours for 'overall liking', 'pleasantness', 'sweetness' and 'creaminess' on 9-point category scales with endpoints labelled (from 'not at all' to 'extremely'). All flavours were presented in the HE version of the drink. The drinks were coded with 3-digit numbers in a presentation order randomised for each subject.

3. Ranking test

Subjects ranked the 2 target flavours and the 4 non-target flavours from '1 = most liked' to '6 = least liked'. All flavours were presented in the HE version of the drink. The drinks were coded with 3-digit numbers in a presentation order randomised for each subject.

In the laboratory sessions, subjects completed the ratings and ranking test first, and finished with the two-choice preference test. The UK group completed all three tests on the same day, whereas the Dutch group had a day between the rating and ranking tests and the two-choice preference test.

Contingency awareness

Subjects completed a short questionnaire that consisted of the following 5 questions:

1. What do you believe was the purpose of the study?;
2. What do you believe were the 2 flavours of the drinks?;

3. Apart from their flavour, what other difference do you believe there was between the 2 yoghurt drinks? (answer options: a) none, b) texture (i.e. creaminess, thickness), c) sweetness, d) both texture and sweetness, and e) other);
4. During the study, did you find one of the yoghurt drinks more filling than the other one? (answer options: a) yes, namely yoghurt drink (fill in name), b) yes, but I don't know which one, c) no, both equally filling);
5. During the study, one of the yoghurt drinks had more energy. Which of the yoghurt drinks do you think it was? (answer option: the yoghurt drink with flavour (fill in name)).

For each question, subjects were asked how sure they of their answer were on a 4-point scale (from 'completely unsure', 'rather unsure', 'rather sure', to 'completely sure').

Statistical analyses

1. In-home conditioning assessment

Because of the number of different types of evaluation, and range of possible comparisons, a variety of statistical analytical approaches were required. The main statistical procedure used in the data analyses was repeated measures ANOVA (using SPSS version 4.0 for the Macintosh). A problem associated with the use of ANOVA for analysis of repeated measures experiments is that the assumption of sphericity is often violated where there are more than two levels on a within-subjects factor (as there are for exposure in this experiment). This problem was addressed by following the procedures recommended by Schlich (1993). Accordingly, in some of the analyses the degrees of freedom were reduced, and p-values increased, using the Greenhouse-Geisser correction (giving fractional degrees of freedom in some instances).

The hypothesis is a energy-by-exposure interaction effect for the hedonic evaluations for the conditioning group, but not for the 'mere exposure' group.

The data for the conditioning group and the 'mere exposure' group were analysed separately because the numbers of subjects differed greatly between the two groups (89 and 19, respectively; see Table 7.3). In most of these analyses there were two within-subjects factors and one between-subjects factor as described below.

Energy effect

There were two levels of this factor, corresponding to LE and HE versions of the yoghurt drink. For the laboratory measures, the subjects actually rated yoghurt drinks of the same caloric value, and energy then refers only to the flavour which had been paired with HE versus the flavour paired with LE for a subject during the experimental period. In some of the analyses (see below) the data for the non-target flavours were included in the energy factor, which was then labelled 'energy/other'.

Exposure effect

Measurements were made after 1, 5, 10, 15 and 20 exposures for the in-home test and 0, 10 and 20 exposures for the laboratory tests.

Group effect

This is the between-subjects factor. It refers to the energy-flavour pairing. Effects involving group would indicate an interaction between flavour and energy density, which might occur if one flavour was more highly preferred.

Hunger, thirst, and desire to eat

The factors included in this analysis were group, energy, time and exposure. Time had two levels, corresponding to the ratings made immediately before and immediately after consumption of the yoghurt drink. The same analysis was applied to the data for thirst and desire to eat.

2. Laboratory conditioning assessment

Laboratory measures of preference for the drinks were analysed in the following ways.

<u>1. Two-choice preference test: liking ratings</u>

Liking ratings for the HE versus LE paired flavours were analysed using ANOVA.

<u>2. Hedonic and sensory ratings for target and non-target flavours</u>

Hedonic and sensory rating data were collected for the 2 target flavours and the 4 non-target flavours. The dependent variables were liking, pleasantness, creaminess, and sweetness. The factors in the analyses were group, energy and exposure. These analyses were then repeated with ratings averaged across the non-target flavours included as a further level in the energy factor. This gave the comparison high versus low versus other (this new three-level factor is called energy/other), in addition to the group comparison.

3. Ranking test

Rank numbers for the HE and LE paired flavour were analysed using ANOVA (with group, energy and exposure as factors).

Contingency awareness

Contingency awareness was measured by calculating the percentage of subjects in each group (conditioning and mere exposure) that was aware of the CS/UCS relationship, i.e. correctly identified the relationship between the flavour (CS) and the drink (UCS). In addition, the sureness of each subject on answering the questions was calculated.

Results

Subject characteristics

The subjects involved in this study (n_{total} = 133; 50 males and 83 females) had a mean body mass index (BMI) of 23.7 kg/m^2 (SD ± 3.7; range 17.0 - 36.9) and a mean age of 30 years (SD ± 13.4; range 18 – 65) (Table 7.3).

Table 7.3 Subject characteristics (n=108; BMI=Body Mass Index; DEBQ=Dutch Eating Behaviour Questionnaire).

Mean ± SD	Conditioning group		'Mere exposure' group	
Subgroup[a]	1	2	3	4
n	46	43	10	9
Age	30.9 ± 1.9	31.0 ± 2.1	31.5 ± 3.9	44.0 ± 5.3
BMI (kg/m^2)	23.5 ± 0.5	23.2 ± 0.4	24.6 ± 1.7	25.4 ± 3.0
Sex (m/f)	21/25	19/24	1/9	0/9
DEBQ - restraint	2.5 ± 0.2	2.3 ± 0.2	2.6 ± 0.3	3.0 ± 0.3
DEBQ – external	3.1 ± 0.1	3.1 ± 0.1	3.0 ± 0.2	2.9 ± 0.2
DEBQ - emotional	2.5 ± 0.1	2.4 ± 0.1	2.5 ± 0.2	2.9 ± 0.3

[a] 1 = High energy yoghurt drink paired with flavour 'cupuacu' and low energy yoghurt drink with flavour 'cherimoya'
2 = High energy yoghurt drink paired with flavour 'cherimoya' and low energy yoghurt drink with flavour 'cupuacu'
3 = High energy yoghurt drink paired with flavour 'cupuacu' and low energy yoghurt drink with flavour 'cherimoya'
4 = High energy yoghurt drink paired with flavour 'cherimoya' and low energy yoghurt drink with flavour 'cupuacu'

Age and sex ratio differed significantly between the ME and the conditioning subjects, but not between the 2 conditioning groups, which were closely matched on all measured subject characteristics.

1. In-home conditioning assessment

Liking ratings

In-home liking ratings were not significantly affected by energy content (Figure 7.1). This was true for both the conditioning [$F(1,87)=1.82$; $p=0.180$] and the 'mere exposure' [$F(1,17)=0.85$; $p=0.369$] groups. The energy-by-exposure interaction was also not significant [$F(3.62,315.25)=0.43$; $p=0.770$ and $F(4,68)=1.23$; $p=0.306$, for conditioning and 'mere exposure' groups, respectively]. Furthermore, the main effect of exposure was also non-significant, and there were no significant main or interaction effects involving group (all $p>0.1$).

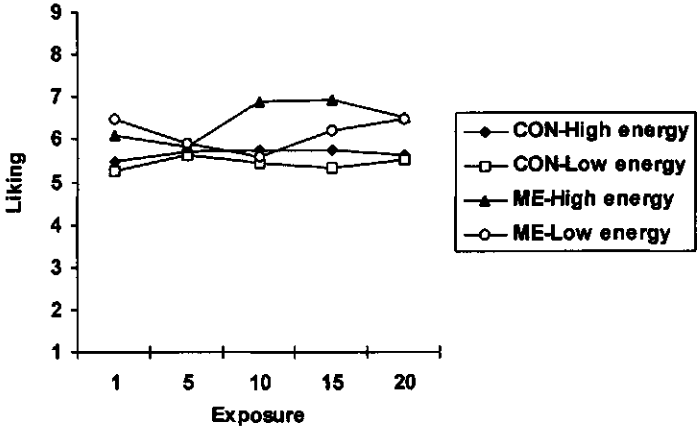

Figure 7.1 In-home liking ratings of high and low energy yoghurt drinks of conditioning (CON, n=89) and mere exposure (ME, n=19) group.

Hunger, thirst, and desire to eat

There were significant main effects of time and exposure for hunger [$F(1,86)=61.8$; $p<0.001$ and $F(37.66,312.37)=8.59$; $p<0.001$], due to a decrease in hunger after consumption of the yoghurt drink and an increase in hunger across exposures (data not shown in Figures). There was also a significant energy-by-exposure-by-time interaction [F(3.45,297.01)=3.46; p<0.012] that could be explained mainly by the difference in hunger for the low versus high energy yoghurt before consumption on the first exposure.

For thirst there were also significant effects of time and exposure [F(1,86)=52.0; p<0.001 and F(4,344)=8.59; p<0.001], but no significant main effect of group and no significant interaction effects (all p>0.1) (data not shown in Figures). Results of separate analyses of the pre-lunch data showed highly significant main effects of time (p<0.001) for hunger, thirst and desire to eat, and weaker main effects of exposure (p=0.029, p=0.073, and p=0.046, respectively). Crucially, however, there were no significant effects involving energy (all p>0.1)(data not shown in Figures).

2. Laboratory conditioning assessment

1. Two-choice preference test: liking ratings

There were significant main effects of both energy and exposure for the conditioning group [F(1,84)=4.39; p=0.039; F(2,168)=7.72; p<0.001], but the energy-by-exposure interaction effect was not significant [F(2,168)=0.44; p=0.644]. Figure 7.2 shows that the HE version of the yoghurt was overall slightly better liked than the LE version and, although liking also increased during conditioning, it did so equally for both versions. There were no significant effects involving group (all p>0.1).

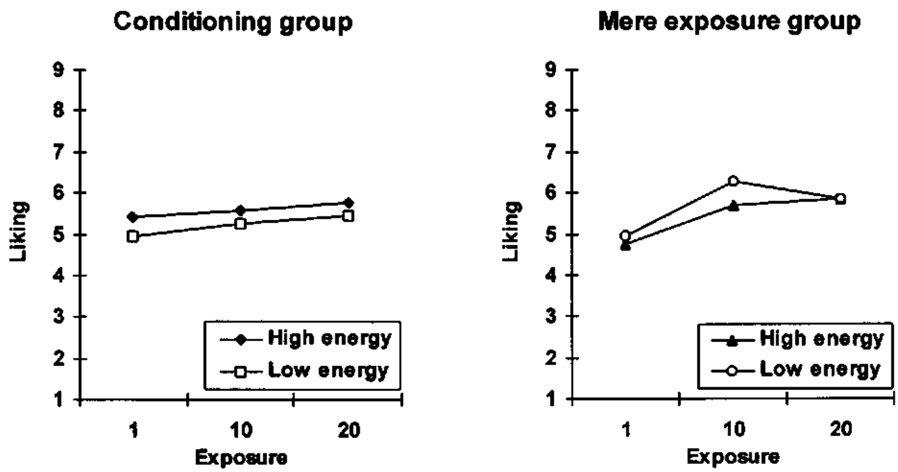

Figure 7.2 Two-choice preference test – liking ratings of high and low energy yoghurt drinks of conditioning (n=89) and mere exposure (n=19) group.

There were also no significant main or interaction effects for the 'mere exposure' group (all $p>0.1$), except for an effect of exposure [$F(2,34)=4.95$; $p=0.013$], showing that liking increased with exposure in this group as well (Figure 7.2).

2. Hedonic and sensory ratings for target and non-target flavours

Conditioning group

Liking ratings

There were significant main effects of energy and exposure for liking [$F(1,85)=6.23$; $p=0.015$ and $F(2,170)=6.32$; $p=0.002$, respectively], but no significant energy-by-exposure interaction effect, nor any significant effects involving group (all $p>0.1$). When energy/other replaced energy in the analysis, there was a significant energy/other-by-exposure interaction effect [$F(3.27,277.68)=7.33$; $p<0.001$]. Figure 7.3 shows that the high energy paired flavour was liked more than the low energy paired flavour, and that this effect was present from the first exposure. There was also an increase in liking for these flavours of the yoghurt after the exposures. In contrast, liking for the other non-target flavours of yoghurt decreased after exposures.

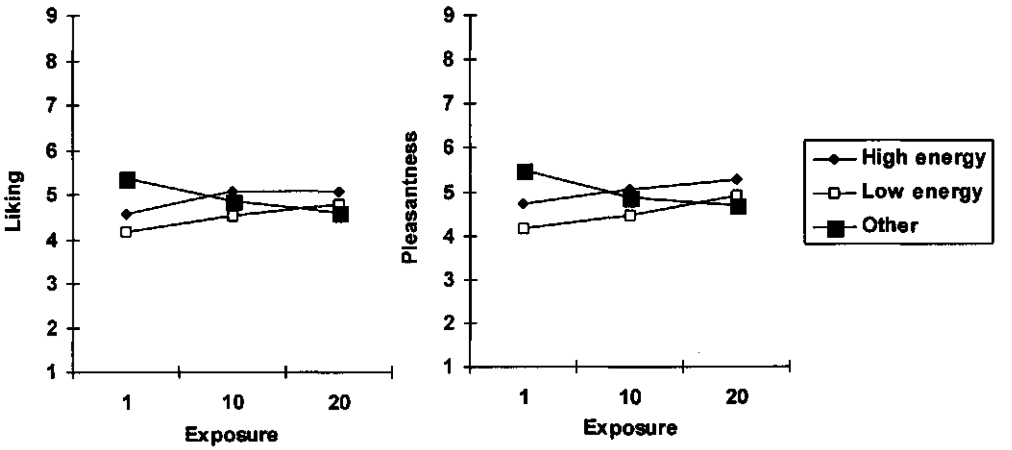

Figure 7.3 Laboratory liking and pleasantness ratings – conditioning group (n=89).

Pleasantness ratings

The results, including means and levels of statistical significance, obtained for the pleasantness ratings were very similar to the results for liking (Figure 7.3).

Creaminess ratings

The group by energy-by-exposure analysis for creaminess revealed a significant energy-by-exposure interaction effect [F(2,170)=5.83; p=0.022] (Figure 7.4). After exposure, there was an increase in rated creaminess of the high energy paired flavour, but a decrease in rated creaminess of the low energy paired flavour. Neither the main effect of energy [F(1,85)=0.78; p=0.378] nor the main effect of exposure was significant [F(2,170)=0.15; p=0.864]. None of the effects involving group were significant (all p>0.1). When energy/other replaced energy in the analysis there was a significant main effect of energy/other [F(2,170)=12.76; p<0.001] and a significant energy/other by exposure interaction effect [F(3.32,282.88)=2.64; p=0.044].

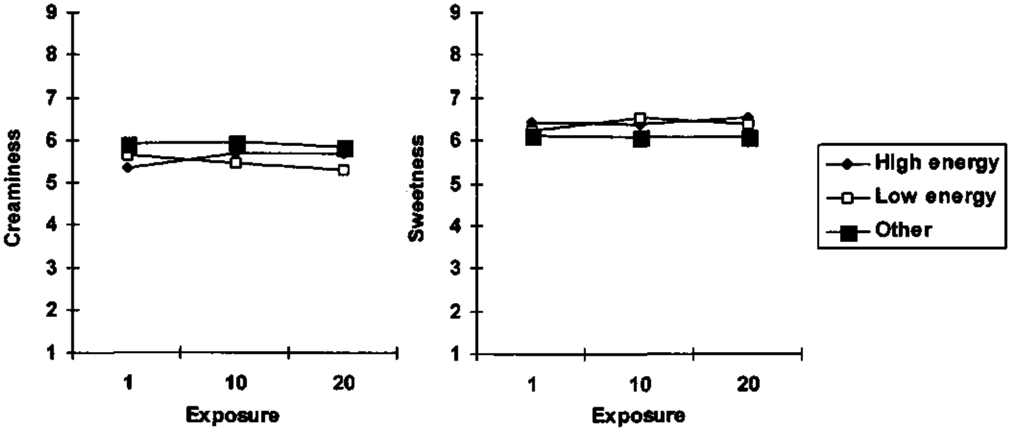

Figure 7.4 Laboratory creaminess and sweetness ratings – conditioning group (n=89).

Sweetness ratings

There was a significant energy/other main effect for sweetness [F(2,170)=6.96; p=0.001) (Figure 7.4). The non-target flavours of yoghurt were rated as slightly less sweet than the target flavours of the yoghurt consumed during conditioning. All other main and interaction effects were non-significant (all p>0.1).

Mere exposure group

Liking ratings

There was significant energy-by-exposure interaction effect for liking [$F(2,34)=4.82$; $p=0.014$], and a significant main effect of exposure [$F(1,17)=8.15$; $p=0.001$]. Liking for the high energy paired flavour increased markedly across exposure. This was in contrast to liking for the low energy paired flavour that changed very little across exposure (Figure 7.5).

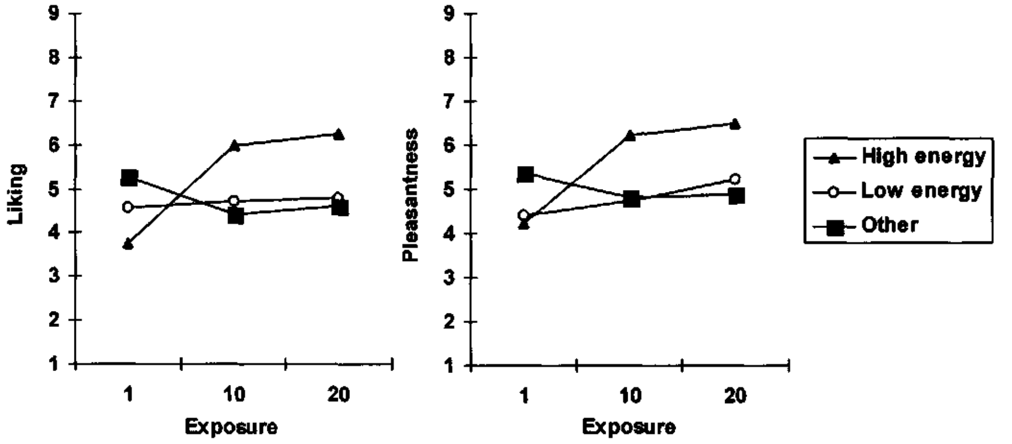

Figure 7.5 Laboratory liking and pleasantness ratings – mere exposure group (n=19).

Pleasantness ratings

A similar pattern of results was observed for pleasantness ratings (Figure 7.5), although the energy-by-exposure interaction was only marginally significant [$F(2,34)=2.63$; $p=0.087$]. When energy/other replaced energy in the analysis of the pleasantness ratings the energy/other by exposure interaction effect was highly significant [$F(4,68)=7.03$; $p<0.001$].

Creaminess and sweetness ratings

For creaminess and sweetness there were no significant effects of energy, exposure, or energy-by-exposure (all $p>0.1$), except exposure for creaminess ($p=0.053$) (data not shown in Figures).

3. Ranking test

The results for the conditioning group showed a significant main effect of energy and exposure [$F(1,84)=6.33$; $p=0.014$ and $F(2,168)=17.43$; $p<0.001$, respectively], but the energy-by-exposure interaction was non-significant [$F(2,168)=0.87$, $p=0.423$]. Figure 7.6 shows that the high energy paired flavour was ranked higher than the low energy paired flavour, and that both were ranked higher on later exposures. For the 'mere' exposure group only the effect of exposure was significant [$F(2,168)=4.54$, $p=0.018$] (Figure 7.6).

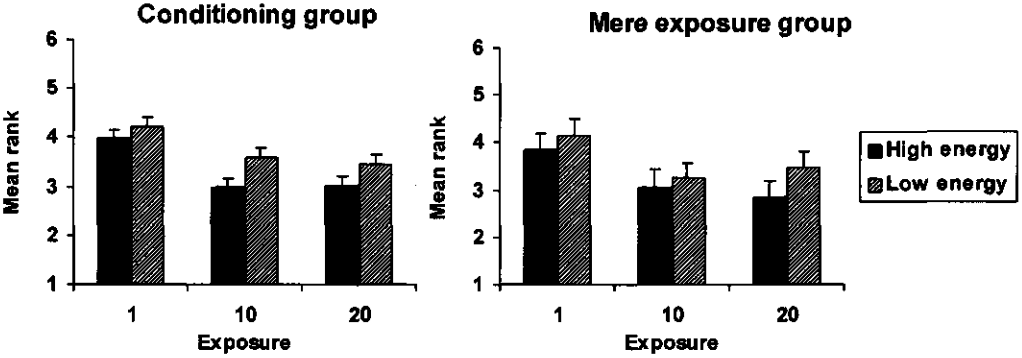

Figure 7.6 Ranking test – mean rank (+ SEM) (1=most preferred, 6=least preferred) of high and low energy yoghurt drinks of conditioning (n=89) and mere exposure (n=19) group.

Contingency awareness

Conditioning group

As a group, there was little evidence of contingency awareness. The proportion of subjects (60%) correctly identifying the HE drink was similar to random chance, and equal proportions of the subjects (54%) correctly or incorrectly identifying the HE drink expressed feeling 'rather' or 'completely' sure of their answer. Furthermore, the proportions of subjects believing the HE and LE version of the yoghurt drinks was the most filling was also similar (40% and 60%, respectively).

Only 18% of the subjects in the conditioning group correctly identified the purpose of the study (investigating changes in preference over time); however, most expressed a view that the study was to do with differences in drink composition, since the other guessed purposes of the study were: preference of new weight loss drink or high-calorie and low-calorie yoghurt drinks (12%), to measure sensory differences (i.e.

sweetness, texture) between the two yoghurt drinks (31%), to measure appetite/fillingness/hungriness over time (9%), or other (7%).

As expected, none of the subjects could correctly identify the novel flavours of the drink. However, the most common description in terms of familiar flavours was that cupuacu had a 'mango/tropical' flavour, and cherimoya 'pineapple/tropical'. Based on this, 13% of the subjects in the conditioning group 'identified' cupuacu, and 17% 'identified' cherimoya. However, the majority of the subjects were completely or rather unsure of this (64% and 53%, respectively).

Mere exposure group

In the 'mere' exposure group, 16% of the subjects correctly identified the purpose of the study (investigating preference changes over time), and 53% of the subjects correctly identified the high energy version of the yoghurt drinks as the one having more calories (80% were rather or completely sure of this). Similar results to the conditioning group were found for the fillingness of the yoghurt drinks (16% of the subjects believed the high energy version to be most filling, whereas 21% believed the low energy version of the yoghurt drink to be most filling).

Similar to the conditioning group, none of the subjects correctly identified the novel flavours of the drink. Considering the taste of cupuacu as mango/tropical, and cherimoya as pineapple/tropical, 11% and 16% 'identified' the respective flavours (with no difference in expressed certainty).

DISCUSSION

Flavour-energy conditioning

The present results in human adults show no evidence of a learned flavour preference based on energy content of foods consumed under normal eating conditions. The data from the in-home and laboratory tests were uniform in showing that there was no greater increase in liking or preference measures for the HE- versus the LE-paired flavour.

These results contrast with those of Birch *et al.* (1990), Johnson *et al.* (1991), and Kern *et al.* (1993), where flavour-nutrient conditioning was demonstrated with energy from sugar or fat as the unconditioned stimulus in a realistic setting. However, these studies used young children as subjects. Children may have a smaller or more flexible range of pre-existing flavour preferences, and be more sensitive to postingestive differences between foods. There are several other possible explanations why we did not observe a conditioning effect of energy in this study.

The energy difference between the yoghurts in this study was derived equally from fat and carbohydrate. Some studies suggest that carbohydrate might be more robust than equi-energetic amounts of fat as a reinforcing agent in nutrient-based conditioning (Kern *et al.*, 1993; Lucas & Sclafani, 1999; Warwick *et al.*, 1997). However, data from Pérez *et al.* (1995), and Capaldi (1996) indicate that energy density is more important than source of energy in macronutrient or energy-based conditioning.

It is also possible that the yoghurt drinks were not energy-dense enough, or the difference in energy content between versions was too small to generate differential effect on affect. However, the differences in energy content between the HE and LE were similar to those used in the successful conditioning study of Birch *et al.* (1990). In addition, the present design was intended to maximise the likelihood that the differences between the foods would be both realistic and salient. The ≈200 kcal/serving difference between the HE and LE version of yoghurts is considerably greater than is commonly seen between differing versions of commercial food products, and represents about 6-8% of typical total daily energy intakes for a sedentary adult. Placement at mid-morning (rather than within a meal) should have further helped to isolate the effects of these foods from others.

The results suggest that, although the LE and HE versions of the yoghurt were initially very closely comparable, there were nevertheless small but perceptible differences between them that could have influenced the conditioning results. However, it is technically impossible to precisely match the exact taste, aroma, and texture of foods with such markedly different energy contents. The fact that the LE and HE versions were initially perceived as similar demonstrates that the study provided a fair test of energy contents effects, and that the manufacturer did very well in developing the yoghurt drinks. The problem of sensory differences is controlled in many animal studies through direct delivery of nutrients to the stomach (e.g., Lucas & Sclafani, 1999). Analogous procedures in humans (though invasive and unpleasant) are possible to do as well, and would provide a more direct test of the effect of the nutrient effects. However, this is far removed from realistic situations where nutrient composition inevitably influences sensory qualities.

In contrast to the 2 flavours consumed in the conditioning trial, the hedonic ratings of the non-target flavours decreased over time, and the non-target flavours were also rated as less sweet compared to the target flavours. These effects may be due to reinforcing effects of attributes of the target yoghurt other than their energy content, e.g. sweetness. This is similar to results of Pliner (1982), where the flavours of juices became associated with sweetness over time. Also, ratings of sweetness may be a 'halo' effect, reflecting greater liking for the target flavours. Rozin (1990) suggested that sweetness is a strong reinforcer in acquisition preferences and likes, and therefore the pairing with sweetness may be in many circumstances a more important reinforcer for the acquisition of preferences or likes than energy. Several other studies on rats and humans have shown an increased preference or liking for flavours that are paired with sweetness (Zellner *et al.*, 1983; Sclafani & Ackroff, 1994; Rozin, 1990). Therefore, a flavour-sweetness association (as in flavour-flavour conditioning) might explain the long-term increase in liking, and would reasonably explain any 'mere exposure' effects with the novel-flavoured yoghurts.

Many studies have shown that a successful outcome of flavour-nutrient conditioning is dependent on the satiety status of the subjects (rats or humans); that is, conditioning appears to be more robust when subjects are in a relevant need state (i.e. food-deprived) (Kern *et al.*, 1993; Gibson & Desmond, 1999; Booth *et al.*, 1982; Rozin, 1989; Gibson *et al.*, 1995; Sclafani & Ackroff, 1993; Capaldi, 1996; Capaldi *et al.*, 1991; Booth, 1985). Such 'state-dependency' is also a feature of caffeine reinforced flavour preferences (Richardson *et al.*, 1996; Yeomans *et al.*, 1998; Rogers *et al.*, 1995). In these studies conditioned preferences were only acquired (or expressed) when the caffeine reversed a caffeine 'need' state (caffeine consumers deprived of caffeine overnight). Therefore, the lack of a conditioning effect in the present study could be due to the negligible impact the energy difference might have had on the subjects' motivational state (as the subjects were well-nourished and minimally deprived). In turn, such strong state-dependency would suggest that flavour-nutrient conditioning is unlikely to be a dominant process for the acquisition or maintenance of food preferences, because in the real setting of developed nations, where food is always abundant, individuals rarely eat in response to marked energy depletion (see Mela & Rogers, 1998). This adds further weight to the suggestion that evaluative (e.g. flavour-flavour) conditioning might be a more dominant process for acquisition of food preferences under these conditions.

Mere exposure

The findings of the mere exposure group suggest a 'mere exposure' effect for the hedonic evaluations, as liking and pleasantness showed exactly the pattern of changes in liking across exposure that were hypothesised (but not obtained) for the conditioning group, while sensory measures (creaminess and sweetness) did not show this pattern. In addition, similar to the conditioning group, the HE-paired flavour was repeatedly ranked higher than the LE-paired flavour, although this effect was not significant.

Although an increase in liking for foods with simple repeat exposure has been demonstrated under controlled conditions for adults (Pliner, 1982; Lévy & Köster, 1999; Stang, 1975), it may in practice be impossible to completely isolate 'mere exposure' effects from sensory or postingestive effects. Indeed, Bornstein (1989) suggested that exposure to some gustatory and olfactory stimuli may be reinforcing in and of itself, and that 'mere exposure' alone is not sufficient to explain the acquisition of food preferences. The 'mere exposure' effect can be explained as an unconscious (implicit) learning process mediated by subliminal stimulus exposures or by simply unattended stimuli (Bornstein, 1989). Zajonc & Markus (1991) confirmed these findings: 'When asked about the reasons for preferring one stimulus to another, participants in [mere] exposure studies never mention familiarity, subjective recognition or frequency of exposure. Instead, they point to stimulus features' (Zajonc & Markus, 1991). Since then, other researchers have further investigated this area, and have shown that, because 'mere exposure' is expressed through implicit memory, awareness, recognition or familiarity between the unconditioned and conditioned stimulus are not necessary or a prerequisite (Seamon *et al.*, 1995). In addition, subliminal stimuli produce stronger exposure effects than clearly recognised stimuli (Ye & Van Raaij, 1997). Thus, 'mere exposure' effects with subtle or minimal stimuli could just be another form of evaluative conditioning, as suggested by recent research of De Houwer *et al.* (1997a, b). This adds further support to our suggestion that evaluative conditioning might be a more plausible explanation than 'mere exposure' or flavour-nutrient conditioning effects in the acquisition of food preferences and likes in realistic eating situations.

Contingency awareness

Both the conditioning group and the 'mere exposure' group showed little evidence of contingency awareness. Several studies have shown that evaluative learning and awareness can be dissociated, and that evaluative learning can be found even when the CS is presented subliminally (De Houwer *et al.*, 1997a, b). For example, Baeyens *et al.* (1990a, 1992, 1993, 1996a, b, c) and Rozin *et al.* (1998) have clearly demonstrated that awareness by the subject of the contingencies is unrelated to the robustness of affect acquisition.

Conclusions

This study tested the development of energy-based acquisition of flavour preferences using a relatively large sample of adults, real food stimuli, natural eating conditions, and multiple outcome measures. The results indicate that flavour-nutrient conditioning is not readily acquired or expressed under these conditions. The data from both the conditioning and 'mere exposure' groups suggest that evaluative conditioning (via flavour-flavour or other non-postingestive effects) may offer a more plausible explanation for the observed results, and is perhaps dominant in influencing acquisition of food liking in realistic eating situations. Energy content *per se* in foods may not greatly influence development of flavour preference or liking in adult humans; however, further evaluative conditioning research with food stimuli under realistic eating conditions is needed to confirm this conclusion.

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8

General discussion

Introduction

The studies described in this thesis concern short- and long-term trials on food acceptance, appetite control and food intake. The objectives of these studies are stated in the general introduction (**Chapter 1**). We first assessed the predictive validity of laboratory sensory tests on food consumption. Secondly, we studied the effects of repeated food consumption, variety and changes in pleasantness on long-term food acceptance and food intake. And thirdly, we investigated the effects of macronutrient and energy content manipulations on long-term food acceptance and appetite control. In this chapter the main findings of these studies are summarized and discussed in relation to the results of other research. The conclusions, implications and recommendations for further research are presented at the end of this chapter.

Predictive validity of laboratory sensory tests on consumption

The methodological study described in **Chapter 2** examined sensory perceptions and preferences of sweetened yoghurts in a taste-and-spit test, a taste-and-swallow test, a fixed quantity test in which an amount of 300 g was consumed, and an *ad libitum* consumption test. The study in **Chapter 2** indicates that laboratory hedonic ratings collected after the taste-and-swallow test give a better prediction of consumption than the taste-and-spit test. The taste-and-spit procedure tends to overestimate the optimal preferred concentration of taste substances/flavours in foods. Thus, hedonic ratings determined in taste-and-spit tests should be interpreted cautiously when used to predict consumption.

The study reported in **Chapter 2** had a fixed-order of presentation of two out of the four sensory test conditions. Specifically, the first session included the taste-and-spit test followed by taste-and-swallow test, always in this order; however, the subsequent ten sessions included the fixed quantity test and *ad libitum* consumption test in random order. This fixed-order pairing of the taste-and-spit test and taste-and-swallow test might have affected the hedonic judgements, in a similar way as contextual effects of repeated tasting can alter hedonic judgements within the first 10 trials of a session (Stang, 1975; Schifferstein & Kuiper, 1997; Schifferstein, 1995). Both the sensory test condition (spitting vs. swallowing) and subjects' previous experience with the test stimuli are therefore important factors to be considered in the interpretation of our hedonic ratings. Taste adaptation, a decline in taste intensity with prolonged stimulation (Theunissen *et al.*, 2000), was probably not an important influence on the outcome, since stimulation was not continuous and perceived sweetness intensity did not change as function of sensory test used.

In **Chapter 2** we found that the optimal sucrose concentration as determined in the taste-and-spit test (10% w/w) was higher than that determined from the taste-and-swallow test, the fixed quantity test and the *ad libitum* consumption test (5.9% w/w). In the taste-and-spit test we also found a shift from 10% w/w in the laboratory sample to 5.9% w/w in the same sample but considered as a large quantity, a shift shown earlier by Mattes and Mela (1986). Rating a small sample while anticipating consuming a larger amount of this sample might therefore be an option to improve the predictive power on consumption of the taste-and-spit test. However, this question has not been validated and needs further research, for example by asking people to actually consume larger quantities of the food samples in a realistic setting and compare this rating with the rating in the laboratory context.

Laboratory sensory testing methods focus mainly on short-term effects, out of the context of normal consumption situations. The hedonic ratings from laboratory tests are, as a consequence, poor indicators of long-term food acceptance and consumption in free living situations (Vickers & Holton, 1998). However, small-scale laboratory tests are desirable for their greater efficiency, time and cost savings, relative to larger samples or in-home trials. Therefore, we need better tools and more insight in sensory methodology in order to utilize such tests to predict longer-term food acceptance and consumption in realistic settings.

Effects of repeated food consumption, variety and changes in pleasantness on long-term food acceptance and food intake

Repeated exposure to foods may lead to changes in food acceptance or liking over time (Porcherot & Issanchou, 1998; Zajonc, 1968; Zellner *et al.*, 1983; Rozin, 1990). Food acceptance or liking ratings can either increase (**Chapter 4 and 7**) or decrease (**Chapter 5**) following repeated consumption. This shift in pleasantness might occur through 'mere exposure' effects, whereby repeated exposure leads to an increase in liking (**Chapter 4**) (Pliner, 1982; Stang, 1974). However, high frequency or extended exposure to foods can also lead to a decrease in liking (**Chapter 5**) (Stubenitsky *et al.*, 1999). A shift in pleasantness might also occur by reinforcing peri- or postingestive effects (**Chapter 7**), in which a previously neutral or even unpleasant taste becomes preferred (Rozin *et al.*, 1998; Zellner *et al.*, 1983). The direction of the shift in pleasantness probably depends on the frequency of exposure, attitudes or beliefs regarding the food, the eating context, the type of food product and its physiological effects, the choice context, and the initial pleasantness of the foods consumed.

The study described in **Chapter 4** was, as far as we know, the first study that focused on longer-term relationships between pleasantness and food intake. In a balanced cross-over design, we investigated the effects of pleasantness on *ad libitum* food intake, liking and appetite within single meals over five successive days. As food product, we chose bread consumed as lunch that varied in sodium chloride concentrations. The results indicate that the relationship between pleasantness and food intake can change over time. The most important result was that with repeated exposure to the less preferred food, the pleasantness of that food remained unaltered, while the desire-to-eat and actual intake of that food increased. In our opinion, however, a prerequisite is that the pleasantness of the less preferred food starts above a certain acceptance level, in order to overcome the possibility that subjects would not eat or choose the food at all. Furthermore, the difference in results between desire-to-eat and pleasantness strengthens the importance to clarify and subdivide for subjects questions about 'liking' and 'desire-to-eat' (= wanting) a food. If subjects had only rated liking, they might have used the ratings of liking in an attempt to indicate an underlying change in desire-to-eat, rather than a real change in liking (Rogers, 1990; Gibson & Desmond, 1999).

The study described in **Chapter 5** investigated the effect of extended exposure and variety on long-term product acceptance and consumption in a home-use situation. Subjects in three different variety groups consumed a meat sauce once a week at dinner at home for a period of ten weeks. The *monotony* group consistently received the same flavour of meat sauce across ten weeks; the *imposed variation* group received three different flavours of the meat sauce in random order; and the *free choice* in variation group was allowed to choose among three flavours of the meat sauce. Results showed a substantial increase in boredom, which was in line with the study by Stubenitsky *et al.* (1999) using chocolate bars and sausages. Two early studies found similar results for non-staple food products, such as meat and vegetables, but not for staple food products, such as bread and coffee, which were consumed daily over a period of 3-4 weeks (Siegel & Pilgrim, 1958; Schutz & Pilgrim, 1958). Food products may therefore differ in their capacity to produce boredom, e.g. staple foods are more resistant to boredom than other foods, and may differ in their appropriateness for consumption frequency. The foods consumed in our study and in the study by Stubenitsky *et al.* (1999) were non-staple foods of which the weekly consumption frequency might have been perceived as too high.

The boredom effect was smaller for subjects who could choose among three different flavours of the meat sauce than for subjects whose serving order was fixed. In line with this finding, Kamen and Peryam (1961) found a higher overall satisfaction among those who were allowed to plan their own menu compared to those who received preplanned menus. Combined with our results, this indicates that lack of variety and above all lack of choice can have a substantial influence on boredom, and hence on long-term food acceptance and food intake.

The study described in **Chapter 7** investigated whether adults alter their liking for flavours associated with higher- vs. lower-energy versions of a drink yoghurt eaten regularly in-home in a within-subject design. A conditioning group consumed 20 times 200 ml of the drink yoghurt at home or work, as mid-morning snack, while a 'mere exposure' group consumed 10 ml. The results suggest that adults do not readily form or learn a conditioned flavour preference based on energy content of foods consumed in a realistic home-use situation. Liking and preference increased with exposure for both the conditioning and 'mere exposure' groups, regardless of the flavour or the energy in the drink yoghurt. In contrast, other studies have shown that children (2-5 y old) acquired relative increases in preference for the flavours associated with higher energy content (Birch *et al.*, 1990; Kern *et al.*, 1993; Johnson *et al.*, 1991). A possible explanation for the discrepancy in results is that children may have a smaller and/or more flexible range of pre-existing flavour preferences, and may be more sensitive to postingestive differences in foods than adults. The former is also the reason why we chose novel flavours as stimuli in our study in adults. An advantage of the use of novel flavours over common flavours is that it ensures that subjects do not hold an initial preference; the less well-learned the stimulus is initially, the more learning can take place, and hence the greater the enhancement of affect. However, a disadvantage is that the results may not be 'applicable' to the real world. Another point is the use of a natural environment (at home) which brings uncontrolled variation to the experiment, but on the other hand, laboratory results may not reflect the true situation (time, place, other people, context etc.).

Some studies suggested that increases in liking over time might be acquired via flavour-flavour conditioning, with sweetness as reinforcer, rather than via flavour-nutrient conditioning, with energy as reinforcer (Zellner *et al.*, 1983; Rozin, 1990). Evaluative conditioning (via flavour-flavour or other non-postingestive effects) may therefore offer an explanation for the observed results in **Chapter 7**, and is perhaps dominant in influencing acquisition of food likes in normal eating situations. Thus, energy content *per se* in foods may not greatly influence development of flavour liking by adults.

Effects of macronutrient and energy content manipulations on long-term food acceptance and appetite control

One strategy to decrease total energy intake over longer periods of time is the consumption of lower energy versions of commercially foods. The effectiveness of this approach depends on the precision of physiological and behavioural mechanisms that regulate food intake. On the one hand, if the precision of these mechanisms is high, then people will eat more of these or other foods to compensate for the missing energy, rendering this strategy unsuccessful. On the other hand, if regulatory mechanisms are not very precise, then the consumption of low energy versions of foods will indeed lead to a decrease in total energy intake (Kendall *et al.*, 1991). In our studies we found an imprecise food intake regulation, both at first exposure with no prior experience in children, young and elderly adults (**Chapter 3**), and conditioned after repeated exposures in adults (**Chapter 6**).

The study in **Chapter 3** contrasted the responses of children, young and elderly adults to the consumption of food preloads varying in fat, carbohydrate and energy content, eaten 90 min prior to a test lunch meal. The study did not show a decline in the ability to regulate the food intake with increasing age, although compared to younger adults, the hunger ratings from the elderly subjects suggested they may be less sensitive to the energy content of the preloads. All age groups exhibited an incomplete energy compensation (i.e. the energy intake in a later test meal was not suppressed by an amount equal to the energy in the preload). We did not find an impairment in energy compensation in elderly compared to younger adults, as was reported earlier by Rolls *et al.* (1995) and Roberts *et al.* (1994). In children, we found an incomplete energy compensation similar to that of younger and older adults. This finding confirmed the findings of studies with children using a preload-test meal design with a 90 min time interval (Birch *et al.*, 1993; Johnson *et al.*, 1991), but contrasts with studies with children that found complete energy compensation using a 20 min time interval (Birch & Deysher, 1986; Birch & Deysher, 1985; Birch *et al.*, 1989). The discrepancy found in energy compensation in our younger and older age groups compared with other studies may partly be explained by the length of the time interval used between preload and test meal

(Rolls *et al.*, 1991; Houniet *et al.*, 1997). Rolls *et al.* (1991) reported that the accuracy of energy intake compensation diminished with increasing time intervals. We chose a 90 min time interval since we believe that a shorter time interval of 20 minutes mainly measures volume and weight effects of the preload, whereas a longer time interval includes physiological postingestive and postabsorptive effects as well.

The young adults showed larger differences in their appetite ratings than the elderly, indicating that elderly may be less sensitive to the preload manipulations than young adults. De Castro (1993) indicated as well that food intake did not affect hunger to the same extent in elderly as it did in younger subjects. Food intake in the elderly may, as a result, primarily be influenced by external factors. Therefore, some elderly individuals may require an increased conscious control over food intake than younger adults in order to ensure adequate dietary intakes.

The results of the study in **Chapter 3** were limited to a single exposure to the preload. It might be that the precision of energy compensation improves with repeated exposure to the preload due to the development of learned associations between satiety and the flavour characteristics of the food (Booth *et al.*, 1982; Booth *et al.*, 1976; Specter *et al.*, 1998). In **Chapter 6**, we conducted a study in adults with 20 exposures to the preload to offer the opportunity for the development of more precise energy compensation. Results revealed incomplete energy compensation for the food preloads at first exposure, with no evidence of any change in compensation with repeated exposures. This finding confirms the results of the few other relevant studies, which largely indicate that adults do not readily learn or express adjustments for a reduced energy content of a food after repeated exposures (Booth *et al.*, 1982; Booth *et al.*, 1976; Specter *et al.*, 1998; Louis-Sylvestre *et al.*, 1994). Contrary to our study and previous studies with adults, studies with children and adolescents have reported accurate learned energy adjustments following repeated experience (Birch *et al.*, 1990; Birch & Deysher, 1985; Kern *et al.*, 1993; Johnson *et al.*, 1991). In response to energy-density in the diet, children may respond to more internal physiological cues (e.g. satiety), whereas older subjects may respond to more external factors other than hunger and satiety (e.g. time of day, presence of other people) (Birch & Fisher, 1997). An age-related impairment might therefore account for the discrepancies found previously and in **Chapter 6** in the ability to learn (e.g. children may learn more quickly and readily than adults) to associate reduced satiety with a decrease in energy density, and to adjust energy intake accordingly.

Main conclusions and implications

The main conclusions from the studies and their implications are the following:

• Adults do not readily learn or express compensation for a reduced-energy food after repeated experience with the food in free living situations. From a public health perspective the use of lower energy versions of foods might therefore be an useful instrument in the prevention of obesity and nutrition related diseases, and in consumers' effort to control their body weight.

• Repeated exposure to foods can lead to changes in liking over time. Liking can either increase or be enhanced, or decrease following repeated consumption. Whether liking increases or decreases with exposure depends on the sensory properties of foods, the type of food product, the availability of different varieties of foods, and the context in which the foods are consumed. This implies that reduced-energy foods with initial good acceptance may, over time, sustain or even increase in their acceptance. The development of palatable reduced-energy foods should therefore be encouraged in order to increase their initial acceptance and, as a consequence, their use and acceptance on the longer-term.

Recommendations for further research

A number of suggestions for further studies on food preference and appetite control are set out below:

• There is a need for long-term studies in the area of food preferences and food acceptance. Both methodological and long-term experiments should be continued and extended to obtain more insight in factors that influence consumers' food choice and food acceptance on the longer-term. For example, studies could be performed on the identification of how and to what extent various factors contribute to the long-term acceptance or rejection of a food product. These studies should not only focus on the usual consumer's hedonic ratings and descriptions of sensory properties of foods, but also on other factors, such as, consumer attitudes and expectations regarding the food product, and the context in which the food product is consumed.

• Methodological studies should focus on the validity of laboratory and in-home sensory tests for predicting food acceptance and consumption after extended use in free-living situations. For example, repeated responses for the same products in laboratory and in-home settings (e.g. different contexts, labeling etc.) may be compared. For instance, with the increasing use of sensory and health claims, both in advertising and on food packaging, future laboratory and in-home trials may focus on the impact of these claims on food acceptance using products with and without these claims over extended periods of time.

- Product development research is increasingly recognizing the importance of understanding the variation in consumer perceptions, motivations, and behaviours; hence there is a trend toward increasing segmentation, and using this knowledge to identify new opportunities. It is therefore important to improve and develop food products that are in accordance with divergent consumer demands. A rather new development in sensory analysis is the approach of improving and developing food products from the consumer perspective, rather than starting with the food product *per se*. However, so far, we still do not know exactly how to translate consumer wishes into specific product characteristics. It is therefore of great interest to validate and improve sensory methods that translate consumer wishes and demands into specific product specifications. In addition, more insight is required in the identification of consumer groups, the preferences and wishes of these consumer groups, and their interactions with consumer attitudes, product types, information (e.g. health claims) and choice contexts.
- A study has recently been published which suggests that very early experience with a specific flavour may exert an influence on food preferences later in life: adults who were as newborn bottle-fed with milk that was flavoured with vanilla had a greater preference for vanilla-flavoured foods than adults who were breast-fed (Haller *et al.*, 1999). It is of interest to determine further the extent to which there may be 'critical periods' in development, when early exposure to certain sensory qualities might exert a particularly significant influence upon later food likes. It might, for example, be possible to use this knowledge to orient preferences toward foods with a lower energy density or other desirable nutritional qualities, such as specific vegetables and fruits.
- The study in **Chapter 7** on conditioned flavour preferences showed increases in liking over time in both the conditioning and 'mere exposure' group. Whether the increases in liking are produced via flavour-flavour conditioning with sweetness as reinforcer or via other non-postingestive effects needs to be unravelled and investigated further. For example, to elucidate the possible role of sweetness as reinforcer, studies on conditioned flavour preferences using sweetness and other taste components (non-sweet stimuli) might be worthwhile to conduct. The lack of a conditioning effect in our study might also be due to the negligible impact the energy difference had on the subjects motivational (hunger) states. Previous experiments found that acquisition and expression of caffeine-reinforced flavour

preferences is highly sensitive to the caffeine need state (deprived vs. non-deprived) (Richardson *et al.*, 1996; Yeomans *et al.*, 1998). In our study, the energy difference, although apparently substantial, might be irrelevant (in motivational terms) because all subjects were well-nourished and minimally deprived. A learned preference for the high-energy-paired flavour might be expressed in a similar study but with subjects tested in a food deprived situation.

- During a 6 month multi-centre study, free living subjects had acces to a special shop with similar ranges of food products which only varied in carbohydrate and fat content (Westerterp *et al.*, 1996; De Graaf *et al.*, 1997). In this study weight maintenance was observed on a high-carbohydrate/reduced fat diet versus an increase in body weight on a low carbohydrate/full-fat diet. Although the effects on body weight were modest, these effects can have potentially important consequences if they were cumulative over periods of years. To learn more about the causes of obesity, it might be interesting to conduct similar long-term trials (a) studying the effect of label information relating to the nutritional properties of foods on responses such as satiety and subsequent food intake, and (b) comparing specific groups, such as lean versus obese individuals and individuals with versus without a genetic predisposition for obesity.

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Summary

This thesis described experiments studying the impact of nutritionally modified foods on food acceptance and appetite control. The major outcomes of the studies related to (1) predictive validity of laboratory sensory tests on food consumption (**Chapter 2**), (2) effects of repeated food consumption, variety and changes in pleasantness on long-term food acceptance and food intake (**Chapter 4 and 5**), and (3) effects of macronutrient and energy content manipulations on long-term food acceptance and appetite control (**Chapter 3, 6 and 7**).

We first investigated the predictive validity of laboratory sensory tests on consumption (**Chapter 2**). Thirty-six adults rated pleasantness and perceived sweetness intensity of yoghurts varying in sucrose concentration in a taste-and-spit test, a taste-and-swallow test, a fixed quantity test in which an amount of 300 grams was consumed, and an *ad libitum* consumption test. The optimal sucrose concentration as determined by the taste-and-spit test (10% w/w) was higher than that determined from the taste-and-swallow test, the fixed quantity test and the *ad libitum* consumption test (5.9% w/w). The perceived sweetness intensity did not change as function of the sensory test used. We concluded that pleasantness ratings collected after the taste-and-swallow test give a better prediction of consumption than the taste-and-spit test.

In an other study the effects of pleasantness on *ad libitum* food intake, liking and appetite were examined over 5 successive days (**Chapter 4**). Thirty-five students consumed *ad libitum* sandwiches for lunch, made with bread varying in salt level and evaluated as low, medium or high in pleasantness, in a balanced cross-over design. On the first day, the students ate less of the least pleasant bread than of the medium and most pleasant bread, whereas on the fifth day consumption of all the breads was similar. For the least pleasant bread, energy intake at lunch, desire-to-eat and fullness all increased over days, whereas these variables remained constant for the medium and most pleasant bread. Mean pleasantness ratings for all breads remained unaltered across the days. We concluded that, with repeated exposure, the desire-to-eat, fullness and intake of a less preferred food can increase over time. Thus, the relationship between pleasantness and food intake can change over a longer time period.

We also investigated the effect of variety on long-term product acceptance and consumption in a home-use situation (**Chapter 5**). The 105 adults in three different variety groups consumed a meat sauce once a week at dinner at home for a period of 10 weeks. The *monotony* group (n=45) consistently received the same flavour of meat sauce across all 10 weeks; the *imposed variation* group (n=30) received one

from three different flavours of the meat sauce in random order, and the *free choice* (*n*=30) in variation group was allowed to choose among three flavours of the meat sauce. Results showed a remarkable increase in boredom over time. The boredom effect was the largest for subjects who consistently received the same food, and was least pronounced for subjects who were allowed to choose among three different flavours of the food.

In a subsequent study (**Chapter 7**), we investigated whether adults alter their liking for flavours associated with higher- and lower-energy versions of a food eaten regularly under realistic eating conditions. Subjects (*n*=108) consumed higher- and lower-energy (273 and 67 kcal per 200 ml serving, respectively) versions of two initially novel-flavoured yoghurt drinks 20 times each over a period of 8 weeks, in a within-subjects design. A conditioning group consumed 200 ml at home or at work, at mid-morning each day, while a 'mere exposure' group consumed 10 ml. There was no evidence for changes in flavour liking or ranking based on the energy content of the yoghurts. Ratings for both flavours increased similarly over time amongst both the conditioning and the 'mere exposure' groups. Evaluative conditioning (via flavour-flavour or other non-postingestive effects) may offer a more plausible explanation for this result, and is perhaps dominant in influencing acquisition of food liking in realistic eating situations. Energy content *per se* in foods may not greatly influence development of flavour preference or liking among adults; however, further evaluative conditioning research with food stimuli under realistic eating conditions is needed to confirm this conclusion.

We also investigated age-associated changes in the short-term regulation of food intake (**Chapter 3**). Therefore 30 children, 33 young adults and 24 elderly were asked to participate in lunch sessions with or without a preload. The preloads consisted of four different strawberry yoghurt preloads that varied in energy and macronutrient content. Children, young and elderly adults consumed 200, 340, and 300 g of the preload, respectively. One yoghurt was low-fat, low-carbohydrate and low in energy (= the control; 0.7 MJ/500 g serving), one yoghurt was high-fat and medium in energy (71 en% of fat; 2 MJ/500 g serving), one yoghurt was high-carbohydrate and medium in energy (87 en% of carbohydrate; 2 MJ/500 g serving) and the fourth yoghurt was high-fat & high-carbohydrate and high in energy (42 en% of fat and 53 en% of carbohydrate; 3 MJ/500 g serving). Ninety minutes after preload consumption subjects had an *ad libitum* lunch-buffet. Results showed evidence for an incomplete energy compensation after consumption of the preloads compared to the no-preload condition. The energy compensation observed in the children ranged between -21% and 34%, in the adults between 15% and 44% and in the elderly between 17% and 23%. Hunger responses were clearly different between the young and elderly adults. The young adults showed larger differences in their appetite ratings than the elderly, indicating that the elderly were less sensitive to the energy content of the preload than the young adults. However, based on the food intake data, we concluded that the ability to regulate the food intake within a preload -90 min test meal paradigm did not differ among children, young and elderly adults.

Finally, we examined the effects of repeated mid-morning consumption of novel-flavoured low and high energy yoghurt beverages on subsequent energy intake at lunch under actual use conditions (**Chapter 6**). Adults (n=69) consumed 200 ml of low and high energy yoghurt beverages (67 and 273 kcal/200 ml, respectively), with 20 exposures to each drink on alternate days. Results revealed incomplete energy compensation for the beverages, both at first exposure and also after 20 exposures. Relative to the no yoghurt condition, energy intake compensation (mean ± SEM) averaged 39 ± 36% for the low energy yoghurt and 17 ± 9% for the high energy version, with no evidence of any change in compensation with repeated exposures. Therefore, we concluded that adults do not readily learn or express adjustments for a reduced energy content of a food after repeated exposures. The long-term consumption of lower energy versions of commercially foods might therefore assist in decreasing total energy intake over longer periods of time.

In **Chapter 8** the main findings of the studies described in this thesis are discussed in relation to the findings of other research. The conclusions, implications and recommendations for further research are given. On the basis of the studies in this thesis we conclude that adults do not readily learn or express compensation for a reduced-energy food after repeated experience with the food in free living situations. Also, repeated exposure to foods can lead to changes in liking over time. Liking can either increase or decrease following repeated consumption. Whether liking increases or decreases with exposure depends on the sensory properties of foods, the type of food product, the availability of different varieties of foods, and the context in which the foods are consumed. This implies that reduced-energy foods with initial good acceptance may, over time, sustain or even increase in their acceptance. The development of palatable reduced-energy foods should therefore be encouraged in order to increase their initial acceptance and, as a consequence, their use and acceptance on the longer-term.

Samenvatting

Deze Nederlandse samenvatting heb ik geschreven voor familie, vrienden en andere belangstellenden buiten het gebied van de voeding en sensoriek, om uit te leggen waar ik me de afgelopen 4 jaar mee heb bezig gehouden.

De studies in dit proefschrift beschrijven de invloed van de smaak en samenstelling van voedingsmiddelen op de voedselacceptatie, voedselinneming en gevoelens van eetlust. Allereerst hebben we in een methodologische studie onderzocht in hoeverre sensorische testen in het laboratorium voorspellen hoeveel er uiteindelijk van een voedingsmiddel wordt gegeten (Hoofdstuk 2). Ten tweede onderzochten we de effecten van het herhaald consumeren van voedingsmiddelen variërend in aangenaamheid, smaak en samenstelling op de lange-termijn voedselacceptatie en voedselinneming (Hoofdstuk 4, 5 en 7). Uiteindelijk hebben we korte- en lange-termijn effecten bestudeerd van verschillende hoeveelheden energie in voedingsmiddelen op de daaropvolgende voedselinneming en eetlustgevoelens (Hoofdstuk 3 en 6).

Smaakonderzoek wordt veelvuldig gebruikt tijdens de ontwikkeling van nieuwe producten en het verbeteren van bestaande producten. De meeste smaakonderzoeken vinden echter plaats in het laboratorium en het is de vraag of de resultaten van die onderzoeken goed vertaalbaar zijn naar de praktijk. In sensorische laboratoria wordt vaak gebruik gemaakt van testen waarin proefpersonen wordt gevraagd een klein monster van een voedingsmiddel kort te proeven en vervolgens de aangenaamheid daarvan te noteren. Daarbij wordt aangenomen dat de antwoorden die hierbij verkregen worden een goede voorspeller zijn voor hoe vaak en ook hoeveel mensen van de geteste voedingsmiddelen gaan eten. Deze testen zijn naar verhouding snel, goedkoop en makkelijk uit te voeren. Maar het nadeel van deze testen is dat de testsituatie niet vergelijkbaar is met de situatie waarin voedingsmiddelen normaal gesproken (bijvoorbeeld thuis) worden gegeten. In zogenaamde *ad libitum* consumptie testen wordt getracht de normale situatie te benaderen door proefpersonen zelf te laten bepalen hoeveel ze van het aangeboden voedingsmiddel proeven. Deze testen hebben echter als nadeel dat minder monsters in één sessie aangeboden kunnen worden en het testen daardoor tijdsintensiever en duurder uitpakt dan de korte sensorische testen. In **Hoofdstuk 2** hebben we de resultaten van drie verschillende korte sensorische testen vergeleken met een *ad libitum* consumptie test. Daarbij hebben we bij 36 proefpersonen de sensorische waarneming van en voorkeur voor vijf yoghurts variërend in suikerconcentratie (1%, 5.9%, 10%, 17.6% en 30%

g/100g) gemeten. Ter vergelijking: commerciële yoghurts bevatten ongeveer 10 gram suiker per 100 gram yoghurt. De korte sensorische testen waren: (1) een test waarin het monster werd geproefd en uitgespuugd, (2) een test waarin het monster werd geproefd en doorgeslikt en (3) een test waarin een voorgeschreven (grote) hoeveelheid van 300 gram gegeten werd. Bij de *ad libitum* consumptie test kregen de proefpersonen ook 300 gram aangeboden, maar mochten ze zelf bepalen hoeveel ze ervan aten. Bij alle testen werd bij iedere yoghurt gevraagd hoe lekker en hoe zoet de yoghurt was. De hoeveelheid yoghurt die gegeten werd in de *ad libitum* consumptie test werd nagewogen. Het bleek dat bij de test waarbij de yoghurt moest worden uitgespuugd de proefpersonen een hogere concentratie suiker in de yoghurt het lekkerst vonden (10% g/100g) dan bij de testen waarbij de yoghurt werd doorgeslikt (5.9% g/100g). In de *ad libitum* consumptie test werd het meest gegeten van de 5.9% g/100g yoghurt. We vonden verder geen verschil tussen de testen in hoe zoet de yoghurt was. Dit suggereert dat de aangenaamheid gemeten in tests waarin het monster wordt doorgeslikt een betere voorspeller is voor de consumptie dan de test waarbij het monster wordt uitgespuugd.

In een volgende studie hebben we de effecten onderzocht van verschillen in aangenaamheid op de voedselinneming en eetlust gedurende 5 opeenvolgende dagen (**Hoofdstuk 4**). Vijfendertig studenten kregen bij de lunch in willekeurige volgorde 3 keer 5 dagen achter elkaar brood (met beleg) met een lage, middel of hoge concentratie zout erin. Het brood werd daardoor beoordeeld als 'laag', 'midden' en 'hoog' in aangenaamheid. Eén groep kreeg bijvoorbeeld 5 dagen brood beoordeeld als hoog, 5 dagen als midden en 5 dagen als laag in aangenaamheid en een andere groep weer andersom. De studenten mochten zoveel eten van het brood als ze zelf wilden. We vonden dat op de eerste dag de studenten minder aten van het minst aangename brood dan van het midden en meest aangename brood, terwijl op de vijfde dag evenveel werd gegeten van alle drie de broden. Voor het minst aangename brood nam de energie-inname bij de lunch, de trek in de boterhammen en de verzadiging toe over de 5 dagen, terwijl deze variabelen constant bleven voor het midden en meest aangename brood. De gemiddelde responsies op aangenaamheid bleven echter onveranderd voor de drie broden gedurende deze periode. Met herhaalde blootstelling kan de trek in, verzadiging en inneming van een minder aangenaam voedingsmiddel toenemen over de tijd. De relatie tussen de aangenaamheid en voedselinneming kan dus veranderen over een langere tijdsperiode. Tevens kwam naar voren dat het belangrijk is om in sensorisch onderzoek onderscheid te maken tussen 'trek in' en 'aangenaamheid van' een voedingsmiddel.

We hebben ook onderzoek gedaan naar de invloed van beschikbare variatie in smaak op het aspect verveling bij het herhaald consumeren van een voedingsmiddel in de normale gebruikssituatie (**Hoofdstuk 5**). In drie verschillende variatie groepen aten 105 volwassenen 1 keer per week, gedurende 10 weken, thuis een saus bij de warme maaltijd. We maakten gebruik van 3 verschillende smaken: zout-zuur, kerrie en saté. De monotone groep kreeg 10 weken lang steeds dezelfde smaak saus te eten (*n*=45); de opgelegde variatie groep kreeg in willekeurige volgorde 1 van de 3 smaken sauzen aangeboden (*n*=30), en de vrije keuze groep mocht kiezen uit de drie verschillende smaken sauzen (*n*=30). In alle drie de groepen trad over de 10 weken heen verveling met het product op. Dit vervelingseffect was het grootst voor de monotone groep en het minst sterk voor de vrije keuze groep.

De meeste smaakpreferenties en -aversies zijn aangeleerd. Enkele smaakvoorkeuren zijn bij de geboorte al aanwezig, zoals de voorkeur voor zoet en de afkeer van bitter. Echter, de grote meerderheid van voorkeuren en aversies wordt aangeleerd. Dit aanleren vindt plaats via leerprocessen die nauw samenhangen met de gevolgen van de voedselinname, zoals het vullen van de maag en de opname van energie uit de voeding. Voedselaversies kunnen bijvoorbeeld ontstaan door negatieve consequenties van het eten van voedsel, zoals ziekte of misselijkheid. Voedselvoorkeuren zouden kunnen ontstaan en veranderen door pòsitieve effecten van voedsel. Mensen zouden bijvoorbeeld een voorkeur kunnen ontwikkelen voor smaken die gekoppeld zijn aan energierijke producten (hoog in vet en/of koolhydraten). In **hoofdstuk 7** bestudeerden we de rol van postingestieve factoren in de ontwikkeling van deze zogeheten 'geconditioneerde' smaakvoorkeuren. Leren volwassenen sneller een voorkeur aan voor smaken die gekoppeld zijn aan hoog-energetische voedingsmiddelen dan voor smaken die gekoppeld zijn aan laag-energetische voedingsmiddelen? Om dit te onderzoeken dronken 89 volwassenen 8 weken lang iedere dag 's ochtends (thuis of op de werkplek) 200 ml van een hoog-energetische drink-yoghurt (1148 kJ/200 ml) of 200 ml van een laag-energetische drink-yoghurt (280 kJ/200 ml). Als smaken werden nieuwe, onbekende zuid-tropische vruchtensmaken 'cupuacu' en 'cherimoya' gebruikt. Dit hebben we gedaan omdat de voorkeur voor nieuwe smaken makkelijker kan verschuiven dan de voorkeur voor bekende, 'vastgeroeste' smaken. De helft van de groep kreeg een hoog-energetische cupuacu en laag-energetische cherimoya en de andere helft kreeg een laag-energetische cupuacu en hoog-energetische cherimoya. De proefpersonen dronken in willekeurige volgorde beide smaken twee à drie keer per week, maar wel op verschillende dagen, met in totaal 20 blootstellingen aan iedere smaak. Het resultaat van de studie liet een vergelijkbare stijging zien in voorkeur voor de smaken die gekoppeld waren aan de hoog- en de laag-energetische drink-yoghurt. Op basis hiervan concluderen we dat het energiegehalte in voedingsmiddelen niet van grote invloed is op de ontwikkeling van smaakvoorkeuren bij volwassenen.

In hoofdstuk 3 en hoofdstuk 6 hebben we de korte-termijn regulatie van de voedselinneming bestudeerd. Met andere woorden: of mensen na een hoog-energetische maaltijd compenseren door bij de daaropvolgende maaltijd minder te eten, en andersom, of mensen na het eten van een laag-energetische maaltijd meer eten bij de daaropvolgende maaltijd. In **Hoofdstuk 3** hebben we de korte-termijn regulatie van de voedselinneming onderzocht bij kinderen, jong volwassenen en ouderen. In de proef dronken 30 kinderen, 33 jong volwassenen en 24 ouderen 's ochtends een drink-yoghurt en aten zij 90 minuten later een broodmaaltijd waarvan de proefpersonen zoveel kon eten als zij zelf wilden. We hadden in totaal 4 verschillende drink-yoghurts die varieerden in energie-, vet-, en koolhydraatgehalte. Er was ook een conditie waarin de proefpersonen geen drink-yoghurt kregen. De kinderen, jong volwassenen en ouderen kregen respectievelijk 200, 340 en 300 gram van de drink-yoghurts. Eén drink-yoghurt was laag-vet, laag-koolhydraat en laag-energie (= de controle; 0.7 MJ/500 g), één drink-yoghurt was hoog-vet, laag-koolhydraat en midden-energie (71 en% vet; 2 MJ/500 g), één drink-yoghurt was laag-vet, hoog-koolhydraat en ook midden-energie (87% koolhydraat; 2 MJ/500 g) en de vierde drink-yoghurt was hoog-vet, hoog-koolhydraat en hoog-energie (42% vet en 53% koolhydraat; 3 MJ/500 g). Uitkomstmaten waren de hoeveelheid energie ingenomen tijdens de broodmaaltijd en gevoelens van honger en verzadiging gemeten vlak voor en op verschillende tijdstippen na de consumptie van de preloads. Alle drie de leeftijdsgroepen, de kinderen, jong volwassenen en ouderen, vertoonden een incomplete energetische compensatie na de consumptie van de drink-yoghurts vergeleken met de conditie waarin geen drink-yoghurt werd gedronken. Dit wil zeggen dat wanneer de proefpersonen bijvoorbeeld een hoog-energetische drink-yoghurt kregen, zij hiervoor niet compenseerden door minder te eten bij de daaropvolgende lunch in vergelijking met de conditie waarin geen drink-yoghurt werd gedronken. Gevoelens van honger waren wel duidelijk verschillend tussen de jong volwassenen en de ouderen. De drink-yoghurts hadden meer effect op de gevoelens van eetlust bij de jong volwassenen dan bij de ouderen, hetgeen erop wijst dat ouderen minder gevoelig zijn voor het energie-gehalte in de drink-yoghurts dan de jong volwassenen.

In hoofdstuk 3 werden proefpersonen eenmalig blootgesteld aan een drink-yoghurt. Mogelijk zijn meerdere blootstellingen over een langere tijdsperiode nodig om het effect van de energie in de drink-yoghurts in te kunnen schatten. In **Hoofdstuk 6** onderzochten we daarom bij volwassenen de lange-termijn effecten van de consumptie van drink-yoghurts met verschillende energie-gehaltes op de voedselinneming bij de lunch in een thuis-situatie. In totaal dronken 69 volwassenen 's ochtends 200 ml van een laag-energetische drink-yoghurt (280 kJ/200 ml) of een hoog-energetische drink-yoghurt (1148 kJ/200 ml), met in willekeurige volgorde 20 blootstellingen aan iedere drink-yoghurt. We vonden een incomplete energetische compensatie na de consumptie van de laag- en de hoog-energetische drink-yoghurts, zowel bij de eerste blootstelling als na 20 blootstellingen. We concluderen daarom dat volwassenen niet leren te compenseren voor verschillen in energie-inhoud in voedingsmiddelen na herhaalde consumptie.

In **Hoofdstuk 8** worden de belangrijkste resultaten besproken in relatie tot bevindingen van andere onderzoekers. De conclusies, implicaties en aanbevelingen voor verder onderzoek worden weergegeven.

We kunnen concluderen dat:

- Volwassenen niet leren te compenseren voor verschillen in energie-inhoud in voedingsmiddelen.
- Herhaalde consumptie van voedingsmiddelen kan leiden tot veranderingen in aangenaamheid over de tijd. De aangenaamheid van een voedingsmiddel kan toenemen of afnemen hetgeen afhangt van de sensorische eigenschappen van het voedingsmiddel, het type voedingsmiddel, de beschikbare variatie en de context waarin het voedingsmiddel gegeten wordt. Dit impliceert dat laag-energetische voedingsmiddelen, met een initiële goede acceptatie, op de lange termijn geaccepteerd kunnen blijven of zelfs in acceptatie kunnen toe nemen. Ter voorkoming van overgewicht verdient het aanbeveling de ontwikkeling van smakelijke laag-energetische producten voort te zetten. Dit verhoogt de initiële acceptatie en daarmee ook het gebruik en acceptatie op de langere termijn.

Nawoord

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Liesbeth

About the author

Elizabeth Hendrika Zandstra was born on June 20, 1971, in Woerden, The Netherlands. In 1989, she passed comprehensive school (A levels) at the 'RSG F.A. Minkema' in Woerden and started the study 'Human Nutrition' at the Wageningen Agricultural University. In June 1995, she obtained her M.Sc. degree in Human Nutrition with main topics in human nutrition and epidemiology. From May 1995 till January 1996, she was appointed as a research associate on two projects, entitled 'Age-associated changes in sensory perception and pleasantness' and 'Effects of consumption of fructo-oligosaccharides on blood glucose and appetite', at the department of Human Nutrition, Wageningen Agricultural University (now: Human Nutrition & Epidemiology, Wageningen University). From February 1996 till February 2000 she was appointed as a Ph.D.-fellow at the same department. During this appointment she conducted the research described in this thesis. She participated in an EU-project, entitled 'Understanding and improving the selection and acceptance of foods for health promotion' and, as part of this project, she collaborated with research groups from the Institute of Food Research, Reading (UK), Department of Food Technology, University of Helsinki (Finland), and Norwegian Food Research Institute MATFORSK, Ås (Norway). She joined the education program of the Graduate School VLAG (advanced courses in Food Technology, Agrobiotechnology, Nutrition and Health Sciences). She was a member of the executive board of the department of Human Nutrition & Epidemiology, member of the departmental 'postgraduate education' committee, and member of the Ph.D.-excursion committee that organized a two-week study-tour to Scandinavia in 1997. She attended the annual international 'Erasmus Epidemiology Summer Program' in Rotterdam, and participated in the international course 'Regulation of food intake, and its implications for nutrition and obesity'. She obtained the *Cerealia* Award for outstanding oral presentation at the Sixth Food Choice Conference (1997) in Uppsala (Sweden). She was selected to participate in the Sixth Seminar of the European Nutrition Leadership Programme in Luxembourg in June 2000.

Over de auteur

Elizabeth Hendrika Zandstra werd op 20 juni 1971 geboren in Woerden. In 1989 behaalde zij haar VWO-diploma op de Rijksscholengemeenschap F.A. Minkema te Woerden en begon zij met de studie Voeding van de Mens aan de Landbouwuniversiteit in Wageningen (LUW). In juni 1995 rondde zij deze studie af met doctoraalonderzoeken in de voeding en epidemiologie. Van mei 1995 tot januari 1996 was zij aangesteld als toegevoegd onderzoeker op twee projecten getiteld 'Invloed van de leeftijd op de sensorische perceptie en preferentie' en 'Effect van fructo-oligosacchariden op bloed-glucose en eetlust' op de voormalige vakgroep Humane Voeding aan de LUW (nu: afdeling Humane Voeding & Epidemiologie aan Wageningen Universiteit). Van februari 1996 tot en met januari 2000 was ze aangesteld als assistent in opleiding (AIO) op dezelfde afdeling. Tijdens deze aanstelling voerde zij de onderzoeken beschreven in dit proefschrift uit. Ze nam deel aan een EU-project, getiteld 'Inzicht verkrijgen in en verbeteren van de selectie en acceptatie van gezondheidsbevorderende voedingsmiddelen', en als onderdeel van dit project werkte zij samen met onderzoeksgroepen uit Engeland (Institute of Food Research), Finland (universiteit van Helsinki) en Noorwegen (Norwegian Food Research Institute MATFORSK). Ze nam deel aan het onderwijsprogramma van de onderzoeksschool VLAG. Ze was bestuurslid van het dagelijks bestuur van de afdeling Humane Voeding & Epidemiologie, lid van de 'postgraduate education' commissie en lid van de AIO-excursie commissie die een twee-weekse studie-reis naar Scandinavië in 1997 organiseerde. Ze participeerde in de internationale 'Erasmus Epidemiology Summer Program' in Rotterdam en in de internationale cursus 'Regulation of food intake, and its implications for nutrition and obesity'. In 1997 won zij de *Cerealia* prijs voor het geven van een mondelinge presentatie tijdens het 6de Voedselkeuze congres in Uppsala (Zweden). Zij werd uitgekozen om deel te nemen aan het 6de European Nutrition Leadership Programme in Luxemburg in juni 2000.

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