‘Salt taste detection thresholds and pleasantness of test meals varying in salt content in obese persons’

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Abbreviations

AFC: alternative forced choice
ASTM: American Society for Testing and Materials
BET: Best-estimate threshold
BMI: body mass index
CD36: Cluster of Differentiation 36
CHC: chronic hepatitis C
CI: confidence interval
ENaC: epithelial sodium channel
GPCRs: G-protein-coupled receptors
HCN: Hyperpolarization-activated cyclic-nucleotide-gated
ISO: International Organization for Standardization
LA: Linoleic acid
MG: megajoule
MSG: monosodium glutamate
RYGB: Roux-en-Y gastric bypass
SD: Standard Deviation
SDT: salt detection threshold
TBC: taste buds cells
TRCs: taste-receptor cells
VLCD: very low calorie diet
WHO: World Health Organization
1 Aim of the study

This study addresses the question whether obese persons have different salt taste detection thresholds and different preferences in salt content of test meals compared to non-obese persons.

Obesity is a leading preventable cause of death worldwide, with increasing prevalence in adults and children particularly in the Western world. Comorbidities associated with excess body weight are especially type 2 diabetes and cardiovascular diseases.

The most common causes for obesity are an excessive food energy intake combined with a lack of physical activity [Lau et al., 1997]. Besides the increased food energy intake the food selection also plays a role in increasing prevalence of overweight in Western societies, where the trend leads to a higher intake of refined sugars and fat [Brand-Miller et al., 2002]. Another reason for different food selections of obese persons could be differences in taste and smell perception of these individuals [Drewnowski, 1997]. Therefore the aim of this study is to investigate possible differences of the salt taste perception of obese persons compared to non-obese individuals.

There are few studies about relationships between taste perception of various tastes and body mass index (BMI) among obese and normal-weight persons. Pasquet et al. reported higher taste sensitivity in massively obese adolescents than non-obese control persons, especially for sucrose and salt [Pasquet et al., 2007]. Simchen et al. reported a significant reduction of bitter and sour taste perception in subjects with BMI greater than 28, but not for sweet and salty tastes [Simchen et al., 2006].

Investigations of the relations between taste perception and obesity have concentrated mostly on sweet and bitter tastes, with little work on salt taste. However, the literature is scarce in determining relationships between salt taste perception and BMI.
1.1 Hypothesis:

Previous studies suggest that several diseases (e.g. obesity, type 2 diabetes) and micronutrient deficiencies can modify taste (see also chapter ‘2 - Literature’). Moreover, salt recognition thresholds were also reported as being lower in massively obese adolescents compared to lean individuals [Pasquet et al., 2007]. The study of Donaldson et al. provided some inconsistent results with the overweight women in their study liked salt taste more than normal-weight or obese women, but the converse was true for overweight men [Donaldson et al., 2009]. There are also inconsistent studies like the one of Simchen et al., which results in no changes in salty and sweet taste perception, but in bitter and sour tastes [Simchen et al., 2006]. These few studies about changes in taste acuity initiates following hypotheses:

1. Therefore we hypothesize that salt acuity may be modified in obese persons compared to non obese persons.

2. As a second hypothesis we hypothesize that salt taste acuity of obese test persons with diseases like type 2 diabetes and hypertension is more likely altered than the salt taste acuity of healthy test persons.

3. The third hypothesis is that obese persons prefer meals with higher salt content compared to normal-weight persons.
2 Literature

This chapter is aimed to give a short overview of current literature about obesity and taste acuity especially salt taste. Basic knowledge about obesity, taste and salt is described in this chapter. There exists a huge amount of literature about obesity and its comorbidities, especially type 2 diabetes, but only a few studies about obesity and (salt) taste. This overview narrows down to the influences of obesity and other factors on taste acuity, plus the relation of salt consumption and obesity. Literature search was performed on www.pubmed.com and www.sciencedirect.com.

2.1 Obesity

The definition of obesity of the World Health Organization (WHO) is:

‘Overweight and obesity are defined as abnormal or excessive fat accumulation that may impair health.’ [WHO, 2013]

The most common method to classify the grade of overweight and obesity is the calculation of the body mass index (BMI). The BMI was defined by Adolphe Quetelet and describes the individual’s body mass divided by the square of their height.

\[
\text{BMI} = \frac{\text{mass}(\text{kg})}{\left(\text{height}(\text{m})\right)^2}
\]

Table 1 presents the BMI classification of the WHO, where a BMI from 30 upwards implies obesity.
Recent data from the Austrian Nutrition Report 2012 (‘Österreichischer Ernährungsbericht’) reveal that 40 percent of Austrian adults aged 18-64 years are overweight (52% of men and 28% of women), thereof 12 percent obese (15% of men and 10% of women). [Elmadfa et al., 2012]

Overweight and obesity arises when the food energy intake chronically exceeds the energy expenditure. Apart from physiological factors, including sex, ethnic origin and age, environmental and behavioral factors play a role in developing obesity. Socioeconomic status for example is a major factor, as well as parenting, peer group, stress, lack of exercise, rapid eating, frequent fast food consumption or food choice in general and many more. Also excessive consumption of sugar-sweetened beverages, called ‘soft-drinks’, has been linked to obesity in the United States. [Bellisle et al., 2012]

Obesity increases the risk of several medical disorders such as diabetes mellitus type 2, hypertension, hypercholesterolemia, hypertriglyceridemia, certain cancers and reduced life expectancy, and these complications may account for 5-10% of all health costs in EU countries. The combination of abdominal obesity, hypertension, dyslipidemia and insulin resistance is also called the metabolic syndrome, which highly increases the risk of developing cardiovascular diseases and type 2 diabetes. [Astrup, 2001]
Obesity affects especially people with lower educational and income levels. Drewnowski (2007) describes a link between obesity and low-cost foods consumption. Low-cost foods, consisting of refined grains, added sugars and added fats are inexpensive, good tasting and convenient. Energy-dense foods cost less than nutrient-dense foods, therefore lower-income consumer unsurprisingly prefer the cheaper, energy-dense foods. [Drewnowski, 2007]

Also because of the soothing properties of sweet-tasting foods, people with psychological vulnerability have a higher intake of sweet products. Furthermore, sweet-tasting foods have been reported to alleviate depression, premenstrual symptoms or responses to stress. [Drewnowski et al., 2012]

### 2.1.1 Obesity and taste

Obesity may also influence the olfactory and gustatory sense. The study of Richardson et al. (2004) implies that morbidly obese individuals have an altered olfactory acuity compared to leaner test persons [Richardson et al., 2004]. Not only olfactory acuity can be altered by obesity and its comorbidities, but also taste acuity.

Studies have examined taste perception and obesity, with a specific emphasis on sweet taste. Populations which are more prone to obesity, such as African Americans, seem to have an elevated desire for sweet diets. The greater desire for intense sweet tastes may be a factor in the elevated incidence of obesity and diabetes in African Americans. [Schiffman et al., 2000]

Sartor et al. (2011) examined differences in sweet and salty taste perception in overweight/obese subjects compared to normal-weight controls. The obese test persons reported perceived sweet and salty tastes as less intense (-23% and -19% respectively) compared to the lean control group. [Sartor et al., 2011]

However, not much evidence exists suggesting that either sweet taste perception or sucrose preference may be a causal factor in obesity. Several contradictory studies show people with high BMI reporting lower pleasantness on eating sweet foods compared
with people with lower BMI [Felsted et al., 2007] on the one hand, but also no differences in sweet taste perception in different BMI groups on the other hand [Grinker, 1978]. These differences may result out of different measuring techniques for sweet taste perception. Besides sucrose or sweet diet in general, fat preference may have an even greater influence on body mass. For example obese women preferred foods that were less sweet but higher in fat compared with lean women. [Donaldson et al., 2009]

Several studies show that body weight does not affect the perception of sweet taste. There were neither threshold nor suprathreshold differences across body weight. Bartoshuk et al. (2006) describe that for nearly 50 years there were no studies about neither sensory nor hedonic differences for sweetness between obese and normal-weight individuals. However, higher BMI is associated with lower perceived sweetness. [Bartoshuk et al., 2006]

Then again the study of Simchen et al. suggests that overweight was associated with a lower taste perception ability in adults (<65 years). Differences in taste perception were found in sour (P=0.015) and bitter (P=0.026) taste perception, but not in salty or sweet taste perception. A strong effect of age and gender on olfactory and gustatory abilities was also observed. Generally, gustatory and olfactory abilities decreased with increasing age and were higher in females than in males. [Simchen et al., 2006]

Dietary surveys have reported that the diets of overweight or obese persons are not particularly rich in sugar or sweet products, but they are rather high in fat [van Baak and Astrup, 2009]. Van Baak and Astrup also suggest a possible relationship between consumption of sugar-sweetened beverages and body weight, but there is currently insufficient evidence from randomized controlled trials of sufficient size and duration. [van Baak and Astrup, 2009] [Drewnowski et al., 2012]

Based on the 1986-1987 Dietary and Nutrition Survey of British Adults Macdiarmid et al. (1998) found a positive relationship between BMI and dietary fat intake for men. A negative relationship was found between sugar intake and BMI in men, but not women. In women the only sugar source associated with BMI was high fat sweet products, where higher intakes were associated to higher BMIs. The reverse relationship was found for men. [Macdiarmid et al., 1998]
A recent animal study of Abdoul-Azize et al. (2013) indicates that obese rodents exhibit a strong preference for lipid solutions in a two-bottle test. In this study the expression of CD36 (Cluster of Differentiation 36), a lipid-receptor, in taste buds cells (TBC), was decreased at mRNA level, but remained unaltered at protein level, in obese rodents. In addition to the increased lipid intake, obesity was also associated with an altered Calcium signaling in TBC. Linoleic acid (LA) induced increases in free intracellular calcium concentrations, largely via CD36. [Abdoul-Azize et al., 2013]

Several studies have shown that especially after weight loss through gastric bypass surgery the preference for sweet taste alters [Bueter et al., 2011] [Hajnal et al., 2010] [Burge et al., 1995]. Bueter et al. (2011) conclude that bypass patients showed increased taste sensitivity to low sucrose concentrations compared with controls (p<0.05). However, a study of Scruggs et al. indicates increased acuity for bitter and sour tastes and a reduction in salt and sweet detection after gastric bypass, but failed to reach significance. [Scruggs et al., 1994]

2.2 Taste

Taste plays an important role in human development since taste is a protective sense, evolved to drive food intake and aid in the avoidance of poison. The taste system detects five basic tastes: sweet, sour, bitter, salty, and umami. [Chandrashekar et al., 2006]

Human newborns and animal infants already show aversions to bitter because many poisonous fruits tend to have a bitter taste. On the other hand sweet taste is often associated with high energy content and fast energy food source. Salt taste ensures the proper dietary electrolyte balance and umami enables the recognition of amino acids [Chandrashekar et al., 2006].

Figure 1 shows examples of positive hedonic and aversive components of facial expression to taste on animal infants and human newborns. [Steiner et al., 2000]
Apart of the protective sense, taste contributes to the overall pleasure and enjoyment of a meal. The anatomical units of taste detection are taste-receptor cells (TRCs). Taste-receptor cells are assembled into taste buds (see Figure 2), which are composed of 50 to 150 TRCs. Taste buds are distributed across different papillae. Those papillae are circumvallate papillae, foliate papillae and fungiform papillae, which are located differently on the human tongue. Circumvallate papillae are found at the back of the tongue and contain thousands of taste buds. Foliate papillae are located at the posterior lateral edge of the tongue and contain hundreds of taste buds. Fungiform papillae are present in the anterior two-thirds of the tongue and contain only a few taste buds. The taste pore, where the interaction with tasting agents takes place, consists of TRCs projecting microvillae to the apical surface of the taste bud. Saliva and dissolved tasting agents accumulate in the taste pore. Tasting agents bind to the taste receptors in the microvillae membrane. These bindings lead to a depolarization of the receptor potential in the taste cells. If the potential exceeds the threshold of stimulation, transmitter signals are released. [Chandrashekar et al., 2006]

The famous tongue ‘map’ of David Hänig’s publication in 1901 was refuted by recent molecular and functional data, which revealed the presence of the five basic modalities in all areas of the tongue (see Figure 2).
There are two models for encoding taste qualities at the periphery: the labeled-line model and the across-fiber model (see Figure 3). In the labeled line model, receptor cells are tuned to respond to single taste modalities – sweet, bitter, sour, salty or umami – and are innervated by individually tuned nerve fibers. In this case, each taste quality is specified by the activity of non-overlapping cells and fibers. The across-fiber-model states that either individual TRCs are tuned to multiple taste qualities, and the same afferent fiber carries information for more than one taste modality (Figure 3b), or that TRCs are still tuned to single taste qualities but the same afferent fiber carries information for more than one taste modality (Figure 3c). [Chandrashekar et al., 2006]
Sweet and umami taste modalities are mediated by three G-protein-coupled receptors (GPCRs). These GPCRs are T1R1, T1R2 and T1R3. The GPCRs assemble into either homodimeric or heterodimeric receptor complexes. Their long amino-terminal extracellular domains are believed to mediate ligand recognition and binding. Studies in heterologous cells revealed that T1R3 combines with T1R2 to form a sweet taste receptor. Therefore T1R2+3 cells are the sweet-sensing TRCs.

Umami taste is evoked by the amino acids monosodium glutamate (MSG) and aspartate. The umami taste was discovered by Kikunae Ikeda only in the beginning of the twentieth century in 1908 [Yamaguchi and Ninomiya, 2000]. The Japanese meaning of umami is ‘delicious flavor’. The main feature of umami taste is the potentiation by purine nucleotides (such as inosine monophosphate (IMP) and guanosine monophosphate (GMP)), which is commonly used by the food industry to enhance the flavor of a wide range of products. In vivo studies of T1R1- and T1R3-knock-out mice proofed that T1R1+T1R3-expressing cells are umami-sensing cells. The review of Chandrashekar et al. (2006) implies that sweet and amino-acid (umami) taste, both attractive tastes for human, share a common receptor repertoire.

Bitter taste has the task to prevent the ingestion of toxic compounds. Since there is very limited discrimination between all bitter chemicals, bitter taste is mediated by a large family of around 30 highly divergent GPCRs (the T2Rs). Chandrashekar et al. (2006)
explain that there is no need for animals to distinguish between many bitter compounds, although they must be able to detect them.

A wide range of receptors and mechanisms have been proposed to be responsible for sour taste. Hyperpolarization-activated cyclic-nucleotide-gated (HCN) channels are one of these proposals. [Chandrashekar et al., 2006]

Various receptors have been proposed for salty tastes, along with the possible taste detection of lipids and complex carbohydrates. The sodium taste receptor epithelial sodium channel (ENaC) had been hypothesized as a candidate to salty sensation for 25 years. Only the report of Chandrashekar et al. (2010), which was published in January 2010, validates the salt taste receptor (ENaC). It was the breakthrough of the team of Chandrashekar et al. to generate mice that had the ENaC receptor gene in most of their cells, but not in their tongues. If the ENaC gene was deleted entirely, the animals died early since the ENaC channel play a critical role in the kidneys, lungs and other organs. The engineered mice responded normally to sweet, umami, sour and bitter flavors, but they had no salt taste. Even after depletion of salt, mice without ENaC in their receptor cells showed little interest in saline solutions. Chandrashekar et al. (2010) also created taste cells that light-up when activated to visualize the taste cells in action (see Figure 4). With this method it was possible to highlight salt-tasting cells which contain ENaC channels whereas receptors for the other four tastes were not displayed. [Chandrashekar et al., 2010]
Sodium chloride is the only compound which evokes pure salt taste. Other chemical salts are for example ammonium chloride, potassium sulphate or magnesium chloride. Ammonium chloride tastes sour-salty, potassium sulphate sour-bitter and magnesium chloride salty-bitter. All salty compounds are crystalline, water-soluble salts, which dissociate in positive and negative ions in liquid solution. Both cations and anions contribute to the flavor intensity. Salty taste is produced by decay of chemical salts to ions, therefore dissociation of ions is the requirement for salty taste. The molecular mechanism is based on cation-permeable ion channels, which enables the influx of sodium ions into the cell after salt consumption. These ion channels (especially the ENaC) are proteins which form pores in the cell surface. The positive sodium ions can enter the TRCs through those pores. This leads to a depolarization of the cell and a subsequent charge movement.

Sweet, sour, bitter and umami are mediated by separate taste-receptor cells (TRCs) each tuned to a single taste modality. Chandrashekar et al. (2010) concluded that sodium sensing is also mediated by a dedicated population of TRCs. These taste cells express the epithelial sodium channel ENaC (see Figure 5).
Signaling cascades downstream of taste receptors have been speculated over years. Recent results have demonstrated that the receptors for sweet, bitter and umami taste, although expressed in separate subsets of cells, all signal through a common pathway to transduce tastant recognition into cell activation. Chandrashekar et al. (2006) suggest that tastant binding to T1Rs or T2Rs activates the heterotrimeric G proteins gustducin leading to the release of the Gβγ subunits and the subsequent stimulation of phospholipase Cβ2 (PLC-β2). Activation of PLC-β2 hydrolyses phosphatidylinositol-4,5-bisphosphate to produce the two intracellular messengers inositol-1,4,5-trisphosphate and diacylglycerol, and ultimately leads to the gating of the taste-transduction channel (the transient receptor potential (TRP) protein TRPM5). Mouse knockouts of gustducin, PLC-β2 or TRPM5 have major deficits in sweet, umami and bitter tastes, while salty and sour tastes remain unimpaired. This finding shows that these two modalities use a different signaling pathway. [Chandrashekar et al., 2006]
2.2.1 Influences on taste acuity

Most determinants of taste threshold are genetic, but various factors may influence taste thresholds. Diseases and environmental changes for example can be influencing factors.

For instance, taste impairment is one effect of oral manifestations in type 2 diabetes. Bajaj et al. observed 50 cases of type 2 diabetes with oral manifestations. Twenty percent of the individuals showed taste impairment. Other complications were periodontal diseases, oral candidiasis, tooth loss, dental caries and salivary gland hypofunction. [Bajaj et al., 2012]

Gondivkar et al. (2009) compared the gustatory function of 40 controlled and 40 uncontrolled diabetes mellitus type 2 patients to the gustatory function of 40 age- and gender-matched healthy control persons. Hypogeusia, reduced ability to taste things, was found among 80% and 45% of uncontrolled and controlled diabetic patients, whereas only 12.5% of the control persons had hypogeusia. There was a significant decreased sensitivity for sweet (P=0.00001), sour (P=0.00002) and salty (P=0.001) taste from healthy to controlled diabetic individuals with the uncontrolled diabetic persons showing the least sensitivity. The decreased sensitivity for sweet followed by sour and salt tastes may influence the choice of nutrients. Therefore the diabetic patients may evolve a preference for sweet-tasting foods, thereby possibly inducing hyperglycemia. [Gondivkar et al., 2009]

Furthermore, Khobragade et al. (2012) evaluated the physiological taste thresholds in 70 cases of type 1 diabetic and 70 non diabetics. The taste threshold was evaluated using 7 different serially half diluted concentrations of glucose (2.00 M-0.031 M), sodium chloride (1.00 M-0.0156 M), citric acid (0.05 M-0.0007 M) and quinine sulphate (0.001 M-0.000015 M). The investigators observed a significant increase in taste threshold for sweet (P<0.0001), salt, sour and bitter (P<0.001) in type 1 diabetic individuals. Other factors like age, local and systemic diseases, alcohol consumption or smoking could also influence taste threshold. Taste sensation for sweet and bitter was significantly reduced in type 1 diabetics. [Khobragade et al., 2012]
Obesity, a primary cause of type 2 diabetes, and its influence on taste acuity is described above in chapter 2.1.1.

Anti-Hypertensive drugs may also influence taste sensitivity in patients with primary hypertension. Suliburska et al. (2012) concluded higher taste sensitivity in healthy people compared to patients suffering from primary arterial hypertension. In this study taste sensitivity was evaluated by chemosensory and electrogustometric method on 84 patients and 20 healthy persons, who represented the control group. [Suliburska et al., 2012]

In this regard it is well known that hypertensive persons are more likely to be salt-sensitive compared with normotensive persons [Elias et al., 2011].

Taste alterations, especially umami and sweet taste disorders, were also found in chronic hepatitis C (CHC) patients. There are only a few studies of alterations in appetite in the case of patients with chronic liver diseases, but taste disturbances affect up to 40% of patients. The aetiology remains unknown but investigators assume that zinc deficiency or a low serum level of some vitamins could be a cause for taste disturbances. Musialik et al. (2012) observed taste recognition thresholds and taste intensity with hedonic perception of forty CHC patients and 110 age- and gender-matched healthy volunteers. The recognition threshold of umami taste of chronic hepatitis C patients was increased (P<0.01) and the intensity of sweet taste perception was higher (P<0.05) in CHC patients compared to the healthy control persons. Those taste alterations, especially umami and sweet taste disorders, may alter real food perception and lead to a reduction in food intake in some CHC patients. [Musialik et al., 2012]

Patients with cirrhosis are often malnourished. Cirrhotic patients were also shown to have impaired gustatory function for detection of salt, sweet and sour and for recognition of bitter, salt, sweet and sour, together with a higher overall median gustatory score (P<0.0001). Moreover the circulating concentrations of magnesium, zinc, vitamin A, and alpha- and beta-carotene were significantly lower in the patient population. Serum magnesium was also significantly negatively associated with the detection of salt (P=0.02) and gustatory score, which were evaluated through using a rinsing technique in 75 cirrhotic patients and 75 comparable healthy individuals. Zinc and vitamin A are both important in maintaining taste integrity, although their exact
roles have not been elucidated. Zinc depletion is commonly reported in individuals with cirrhosis. [Madden et al., 1997]

Micronutrient deficiencies, especially zinc deficiency, are also playing a role in taste changes. Asano et al. for example studied taste threshold and serum zinc level changes after chemotherapy. The outcome was consistent with the results of Musialik et al (2012) and Madden at el. (1997). Asano et al. (2012) also found a correlation between high serum zinc level and more sensitivity to salty taste. [Asano et al., 2012]

Furthermore, Nagai et al. evaluated taste acuity in Japanese young women and the relationship with micronutrients status. Serum concentrations and dietary intakes of zinc, iron, copper and selenium were measured in 38 healthy young women. A significant coherence between serum iron concentration and hypogeusia for salty taste was described. However, dietary micronutrient intake had no influence on the four taste acuities. In spite of that, iron status should, next to zinc status, also be considered in the study of taste acuity. [Nagai et al., 2012]

Tabuchi et al. studied the influence of levels of dietary protein on taste sensitivity for sodium chloride and zinc status in rats. A previous study of Ohara et al. (1994) already reported an impaired taste preference by protein malnutrition in juvenile rats [Ohara et al., 1994]. The results of Tabuchi et al. suggested that 8% purified egg protein seems to be an optimal dietary protein level to normalize taste sensitivity for sodium chloride. Another conclusion was that the increase of protein consumption had also increased the serum zinc concentration. [Tabuchi et al., 1997]

Rats from a protein free diet group showed significantly lower taste sensitivity than other protein containing diet groups, while serum zinc concentrations were not affected by dietary protein levels. [Ohara et al., 1995]

Therefore the studies conclude that dietary protein level affects not only growth but also taste preference in rats.

Heath et al. (2006) concluded that taste thresholds may also be modulated by serotonin and noradrenaline. In this study the enhancement of serotonin lead to a reduction of sucrose taste threshold by 27% and of quinine taste threshold by 53%, whereas enhancing noradrenaline lead to a significantly reduction of bitter taste (by 39%) and sour taste (by 22%) reduction. Furthermore there was a positive correlation of the
anxiety level with bitter and salt taste thresholds. [Heath et al. 2006]

Changes in salt preference also appear through different phases of menstrual cycle. The preference for salted popcorn was most during the luteal phase and correlated with the strength of the salt solution used in one study. [Verma et al., 2005]

After summing-up the main factors, which can influence taste acuity, the next chapter focuses on salt, salt taste and the influences on salty taste perception.

### 2.3 Salt

Salt, consisting of sodium and chloride ions, is involved in regulation processes of the body water. Sodium is also essential for electrical signaling in the nervous system. Within the body, sodium also regulates extracellular volume, maintains acid-base balance, renal function, cardiac output and myocytic contraction [Liem et al., 2011].

The minerals sodium and chloride are major elements. Major elements are essential minerals and each is present in the human body at a ratio of at least 50 mg per kg of the body weight. The D-A-CH (Germany, Austria and Switzerland) recommended intake (estimated value) for adults for sodium is 550 mg/day and 830 mg chloride per day. One gram salt (NaCl) consists of 17 mmol sodium and 17 mmol chloride. Therefore the equation results in:

\[
NaCl \text{ (g)} = Na \text{ (g)} \times 2.54
\]

\[
1g \ NaCl = 0.4g \ Na
\]

[D-A-CH, 2000]

Salt taste is one of the basic human tastes (sweetness, sourness, saltiness, bitterness and umami) and the most important food seasoning. Nevertheless the evidence of an association between dietary salt intakes and blood pressure has increased [Karppanen et al., 2005]. It has been estimated that 62% of stroke and 49% of coronary heart disease is caused by high blood pressure. Besides high blood pressure, excess sodium
consumption is also associated with other negative health effects like gastric cancer, decreased bone mineral density and possibly obesity. [Liem et al., 2011]

The WHO recommendation is 5g dietary salt per day. The Austrian Nutrition Report 2012 exposes that 25% of Austrian women and 36% of men exceed a daily intake of more than 10g salt per day. The estimated mean of daily salt consumption is 7g for women and 8.3g for men in Austria. [Elmadfa et al., 2012]

The average US sodium intake is estimated to be 8.2-9.4 g sodium chloride per day. In the United Kingdom the intake is around 9.4 g sodium chloride per day and in Asian countries the intake is even higher than 12.0 g sodium chloride per day. These data illustrate consumption levels well in excess of sodium required for optimum health. In industrialized countries, about 75% of sodium in the diet comes from manufactured foods. Increase of saltiness is associated with palatability of foods, though high consumption of sodium is related to negative health effects such as hypertension and cardiovascular disease including stroke. [Liem et al., 2011]

Palatable foods may lead to overeating and furthermore to obesity. Most studies investigated correlations between sweet taste and obesity, only a few studies examines salt taste, although there is evidence of an indirect correlation between BMI and liking for salty and fatty food [Keskitalo et al., 2008].

He and MacGregor describe an indirect relation of salt intake and obesity through soft drink consumption, associated with renal stones, osteoporosis and stomach cancer [He and MacGregor, 2008].

Moreover, Pasquet et al. investigated taste perception in massively obese and in non-obese adolescents. Pasquet et al. compared taste sensitivity and hedonic responses of 39 obese adolescents (mean BMI: 39.5 kg/m²; Range: 30.9-51.6) and 48 non-obese adolescents (mean BMI: 21.0 kg/m²; Range: 16.5-27.9) of both sexes. Recognition thresholds for fructose, sucrose, citric acid and sodium chloride were measured. Supra-threshold perceived intensity and hedonic responses were quantified for sucrose and sodium chloride solutions. Besides, the occurrence of the metabolic syndrome was also assessed by measuring blood pressure and taking blood samples. The results concluded that massively obese individuals had a higher sensitivity to sucrose and sodium chloride than non-obese test persons. Obese test persons showed significantly lower recognition
thresholds and higher perceived intensities at supra-threshold levels for sucrose and sodium chloride, whereas the hedonic responses were significantly lower for sodium chloride. Gender-specific differences were also observed among obese girls only, where a significant positive correlation between taste responsiveness and the number of obesity-related metabolic disturbances existed. [Pasquet et al., 2007]

The study of Donaldson et al. (2009) provided some inconsistent results with the overweight women in their study liked salt taste more than normal-weight or obese women, the converse was true for overweight men. The cohort study contained 69 men and women (BMI range 18.6-36.3). Recognition thresholds, intensity and liking of sodium chloride and monosodium glutamate solutions were measured. The liking of the 1-mol/L solutions were measured with a generalized-labeled magnitude scale and suggest that overweight women like salt taste more than normal-weight or obese individuals. Figure 6 displays the mean values showing that salt liking was altered in overweight women and normal-weight men compared with the other groups. Overweight women (n = 14) and normal-weight men (n = 10) liked salt, whereas the other groups (n = 11 overweight men, 23 normal-weight women, 8 obese women) did not (*P, 0.05 compared with normal-weight women). [Donaldson et al., 2009]

![Figure 6: Mean salt liking, x-axes: groups, y-axes: mean salt liking (mm), * significance P<0.05 compared with normal-weight women [Donaldson et al., 2009]](image_url)
Frequent fast food consumption may also lead to an enhanced preference for salt taste [Kim and Lee, 2009].

An older study of Desor et al. (1975) states that younger test persons preferred greater sweetness and saltiness compared to adult individuals. The investigators also observed race and sex differences in preferences in the younger groups but not among the adults. [Desor et al., 1975]

Taste perception is a minor factor in the complex cause of obesity, but little is known about correlations between salt taste and eating behavior, since most studies investigated the relations between sweet taste and body mass.
3 Materials and methods

This chapter describes the project team, test persons, testing methods, samples and organizational matters, especially the peculiarities of working with patients of the Vienna General Hospital. Instructions of the sample preparations are also included in this chapter. It also includes calculations and sensory evaluation methods.

This project has been submitted to the ethics commission of the Vienna Medical University and has been approved in June 2013. Therefore a declaration of obligation has been signed, since the study includes medical patient data.

3.1 Project team

Supervisor of this study is Prof. Dr. Cem Ekmekcioglu of the Vienna Medical University, who has assessed the topic and aim of this study. Cooperation partners are Dr. Karin Schindler of Vienna Medical University and Assoc.-Prof. Dr. Klaus Dürrschmid of the University of Natural Resources and Life Sciences in Vienna. Assoc.-Prof. Dr. Klaus Dürrschmid also provided most of the materials used for sensory evaluations for this study. Materials such as Sodium Chloride (industrial salt NaCl 99%) and deionized water were provided by Prof. Dr. Cem Ekmekcioglu and the Institute of Environmental Health.

Further participants are Dr. Gerhard Prager and Dr. Felix Langer, surgeons of the Medical University, as well as Prof. Dr. Bernhard Ludvik, Dr. Renate Kruschitz, Dr. Soheila Shakeri-Leidenmühler and Maria Luger, all from the Vienna Medical University. Furthermore the dietician of the obesity patients was Melanie Walker of the Vienna General Hospital.
Nutritional science student Julia Petzenka, Bsc, of the Vienna University assisted throughout the sensory tests of the healthy control persons and several obesity patients of the Vienna General Hospital.

3.2 Test persons

Test persons are obese patients undergoing bariatric surgery at Vienna General Hospital. The control group consists of age- and gender-matched healthy test persons with a BMI between 16.1 kg/m² and 32.8 kg/m² (Mean BMI: 24.1 kg/m²). The purpose of the control group is to identify any general differences in salt detection threshold and pleasantness of test meals between persons with a high and low BMI.

Patients of the Vienna General Hospital are under medical surveillance. Medical conditions including diseases (for example type 2 diabetes, hypertension) are documented and considered in this study.

The tests were performed one day before the patients’ gastric bypass surgery. This project is going to be extended beyond this master thesis, therefore the test persons consist of patients undergoing gastric bypass surgery, who will be tested again in the further future to observe possible changes of salt taste acuity after weight loss.

Generally the patients have a follow-up appointment in the obesity clinic 3 month after bypass surgery, where the second test will take place.

Age- and gender-matched, healthy test persons were recruited for the control group from the Institute of Environmental Health and from the Institute of Pathophysiology, both departments of the Vienna Medical University, as well as at the company SGS Austria.

Table 2 shows an overview of the test persons and performed tests of this study.
Table 2: Test persons and performed tests

<table>
<thead>
<tr>
<th>Test persons</th>
<th>Controls</th>
<th>Performed tests</th>
</tr>
</thead>
<tbody>
<tr>
<td>30 obese (gastric bypass patients)</td>
<td>23 non obese (BMI between 16.1 and 32.8)</td>
<td>1. 3-alternative forced choice (3-AFC) test for sodium chloride detection</td>
</tr>
<tr>
<td></td>
<td></td>
<td>2. 9-point hedonic scale for pleasantness of test meal (soup)</td>
</tr>
</tbody>
</table>

The sample size of 53 (30 test persons and 23 controls) derives from similar studies in obese persons, where taste acuity was tested before and after bariatric surgery in different time intervals (Table 3). Parameters (recognition thresholds of salt taste) were estimated statistically (ANOVA, paired- and unpaired tests) by including published values from similar studies and the sample size is computed with the sample size calculator ($p < 0.05$, Power = 0.80, 10% drop out rate). [SISA, 2013]

Table 3: Estimated means and sample size in similar studies

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Mean group 1, Mean group 2 (unit)</th>
<th>Sigma (SD)</th>
<th>Sample Size</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Recognition threshold (sucrose)</td>
<td>0.05, 0.03 (mol/L)</td>
<td>0.02</td>
<td>10</td>
<td>Bueter et al., 2011</td>
</tr>
<tr>
<td>Recognition threshold (sucrose)</td>
<td>0.047, 0.024 (mol/L)</td>
<td>0.02</td>
<td>14</td>
<td>Burge et al., 1995</td>
</tr>
</tbody>
</table>

For example, in a similar study of Bueter et al. (2011) sucrose threshold was investigated in obese patients before and after Roux-en-Y gastric bypass (RYGB) and a mean difference of approximately 0.02 mol/l with a SD of 0.02 was calculated. The estimated sample size for this study would be $n = 10$. In another comparable study by Burge et al. (1995), where changes in taste acuity were examined, the sample size was
14. Therefore, after assuming a drop-out rate of 10 % and a safety margin, we decided to set the sample size to at least 18 per group (30 patients, 23 control persons).

<table>
<thead>
<tr>
<th>Comparable study</th>
<th>RYGB patients</th>
<th>Controls</th>
<th>Test methods</th>
</tr>
</thead>
</table>
| Bueter et al., 2011 | 9 obese (8 female, 1 male) | 9 normal weight (7 female, 2 male) | • ‘corrected hit rate” for sucrose detection  
• Hedonic visual analogue scale test (VAS) |
| Burge et al., 1995 | 14 obese (8 female, 6 male) | 4 obese, non controlled, consuming VLCD diet | • Cornsweet’s staircase (taste detection and recognition threshold) |

Bueter et al. investigated that gastric bypass patients (n=9) detected lower concentrations of sucrose when compared to normal weight controls (n=9) after surgery [Bueter et al., 2011].

Taste recognition thresholds for sucrose of the 14 RYGB patients decreased significantly (P<0.05) postoperatively. There were no significant changes in the taste detection thresholds for sucrose and urea in the patients consuming a very low calorie diet (VLCD). [Burge et al., 1995]

There are no expected health benefits for the test persons from their participation in this study. However, the test persons contributed to the scientific understanding and to possible improvements for the salt problem. The benefit for the test persons is a free counseling interview on the advantages of salt reduction for human health after participation (performed by Prof. Dr. Cem Ekmekcioglu). There were no risks or side effects for the participants.
3.3 Sensory evaluation

Sensory evaluation is defined as:

‘A scientific discipline used to evoke, measure, analyze and interpret those responses to products that are perceived by the senses of sight, smell, touch, taste and hearing.”
[Stone and Sidel, 1993]

For the assessment of the salt taste threshold the ‘Standard Practice for Determination of Odor and Taste Thresholds By a Forced-Choice Ascending Concentration Series Method of Limits’ had been chosen. This standard practice was developed by ASTM International, known until 2001 as the American Society for Testing and Materials (ASTM) and belongs to the fundamentals of sensory. The 3-alternative forced choice (3-AFC) presentation, a set consisting of one test sample and two blank samples, where the test person has to select one sample, was used for this study. Further information about the 3-AFC test is described in chapter 3.4. [ASTM E679 – 04, 2011]

Another commonly used method of threshold estimation is the Cornsweet’s Staircase method or also called up-down technique. With this method, the test person’s responses determine the concentrations tested: if the test person indicates the presence of a stimulus (e.g. salt), then the concentration of the test stimulus is decreased on the next trial while if the test person fails to indicate a stimulus, the concentration is increased. The first reversal is usually discarded and the six subsequent reversals are averaged in order to determine the threshold. [Cornsweet, 1962][Bartoshuk, 1978]

The frequent reversals in the Staircase method makes odor and taste threshold testing a lenghty procedure of unpredictable duration. Since the test persons’ ability of willingness to concentrate strongly influences the quality of the test results, standardized test duration is desirable. [Lötsch et al., 2004]

In the procedure of the study of Scruggs et al. (1994), a single drop was placed on the anterior tongue three times. One of those drops was a taste solution and the other two were water and the test person was asked to report whether there was a difference
between the three drops and this was considered the detection threshold. The concentration at which the test persons could correctly identify the taste quality of the stimulus was considered the recognition threshold. However, only a limited number of taste buds are stimulated with this technique. [Scruggs et al., 1994]

Bueter et al. (2011) used the method of constant stimuli in which taste stimuli were presented randomly and performance was assessed across a set of concentrations. Seven sucrose concentrations were used in this study. Concentrations were tested in eight blocks with each block consisting of seven sucrose and seven water stimuli. Sucrose and water samples were presented in random order without replacement. Each of the seven sucrose concentrations was presented once within a block. Test persons then sampled the stimulus in the mouth, expectorated the sample and had to indicate whether the stimulus was water or not. The disadvantage of this method is, same as with the staircase procedure, the duration of the test, since this method contained 112 samples per test person.

Since the duration of the taste threshold estimation is an important factor, the ‘Standard Practice for Determination of Odor and Taste Thresholds By a Forced-Choice Ascending Concentration Series Method of Limits’ of ASTM was judged as the most reasonable and appropriate method for the obese patients of the Vienna General Hospital and therefore used in this study.

3.4 Test 1: Assessment of salt taste threshold

Thirty obese and 23 non obese test persons, representing the control group, were investigated for sodium chloride detection threshold. Detection tests were performed in the afternoon approximately 2 hours after lunch at the Vienna General Hospital. Room temperature had been kept constant at 21 ± 1 °C for all test sessions. All solutions were presented at room temperature.
The testing method of the sodium chloride detection threshold was the ‘alternative forced choice (AFC) test”, which is defined by ASTM International as:

‘Alternative forced choice (AFC) test, n—method in which 2, 3, or more stimuli are presented, and assessors are given a criterion by which they are required to select one stimulus. Typical examples include 2-AFC (directional different test) and 3-AFC (selecting the one stimulus among a set of three that differs in a defined attribute).’

[ASTM E679 – 04, 2011]

The 3-alternative forced choice test was used to assess the sodium chloride detection threshold.

Figure 7: 3-AFC test with salt and water stimuli

Eight sodium chloride concentrations (0.003 to 0.034 mol/L, Table 5) are tested in eight blocks with each block consisting of one sodium chloride (20ml) and two distilled water stimuli (20ml) in ascending concentration levels (see Figure 7 and Figure 8). The blank and test samples are encoded with random three-digit numbers so that there is no visible difference between the samples (see appendix for Questionnaire). The test person starts with the first block, which consists of two water cups and one cup with the lowest salt concentration (0.16 g/L). The whole content of the cup should be taken into the mouth, rinse in the oral cavity and spit out again. A cup of tap water was provided for rinsing the mouth between the different stimuli. The study participant indicates which of the three samples is different from the other two. A choice has to be made, even if no difference is noted, so that all data can be utilized for further analysis. All participants have to test all dilutions (8 blocks), even if the salt solution is detected correctly earlier. Individual best-estimates values of threshold are derived from the pattern of correct/incorrect responses produced separately by each test person. Group thresholds
are derived by geometrical averaging of the individual best-estimate thresholds. [ASTM E679 – 04, 2011]

Salt solutions were prepared under supervision of Assoc.-Prof. Dr. Klaus Dürrschmid at the Vienna University of Natural Resources and Life Sciences. The sodium chloride (industrial salt NaCl 99%) amounts for 500ml solution were weighed on a calibrated scale, filled in 500ml DURAN® laboratory glass bottles, filled with distilled water and were agitated. After each test session the salt solutions were stored in a +4°C fridge. The solutions were renewed every six weeks.
Table 5: Preparation of the salt solutions [ISO 3972 Standard]

<table>
<thead>
<tr>
<th>Dilution</th>
<th>Sodium Chloride concentration</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>ρ [g/l]</td>
</tr>
<tr>
<td>D1</td>
<td>2</td>
</tr>
<tr>
<td>D2</td>
<td>1.4</td>
</tr>
<tr>
<td>D3</td>
<td>0.98</td>
</tr>
<tr>
<td>D4</td>
<td>0.69</td>
</tr>
<tr>
<td>D5</td>
<td>0.48</td>
</tr>
<tr>
<td>D6</td>
<td>0.34</td>
</tr>
<tr>
<td>D7</td>
<td>0.24</td>
</tr>
<tr>
<td>D8</td>
<td>0.16</td>
</tr>
</tbody>
</table>

For the data evaluation the series of each panelist’s judgments was expressed by writing a sequence containing (0) for an incorrect choice or (1) for a correct choice arranged in the order of judgments of ascending concentrations of the salt solution. The best-estimate threshold concentration for the panelist is then the geometric mean of that concentration at which the last miss (0) occurred and the next higher concentration designated by a (1). [ASTM E679 – 04, 2011]

An example of the best-estimate threshold calculation is shown in Table 6.
Table 6: Examples of best-estimate thresholds

<table>
<thead>
<tr>
<th>Test-persons</th>
<th>NaCl Concentrations increase → [mg/L]</th>
<th>Judgments</th>
<th>Best-estimate threshold (BET)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0.16</td>
<td>0.24</td>
<td>0.34</td>
</tr>
<tr>
<td>1</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>2</td>
<td>1</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>3</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

Details of calculation are as follows:

For test person 1, the best-estimate threshold is: \[\sqrt{0.16 \times 0.24} = 0.58 \text{ mg/L NaCl}\]

For test person 2, the BET is: \[\sqrt{0.69 \times 0.98} = 0.82 \text{ mg/L NaCl}\]

All other values follow these same calculations.

3.5 Test 2: Assessment of preference for different concentrations of salt in soup

After the first assessment of the sodium chloride detection threshold, 5 cups of creamed soup with different salt concentrations in ascending concentration levels are offered for each test person. The test persons are asked to rate their liking of the soup on a 9-point hedonic scale that ranged from 1 (dislike extremely) to 9 (like extremely). The soup is made out of tab water, sour cream, flour and salt in different concentrations (see Table 7).
Table 7: Salt concentrations in cream soup

<table>
<thead>
<tr>
<th>Dilution</th>
<th>$\rho$ [g/200ml]</th>
<th>$\rho$ [g/l]</th>
<th>Mol/L</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0.6</td>
<td>3.0</td>
<td>0.051</td>
</tr>
<tr>
<td>2</td>
<td>0.9</td>
<td>4.5</td>
<td>0.077</td>
</tr>
<tr>
<td>3</td>
<td>1.2</td>
<td>6.0</td>
<td>0.103</td>
</tr>
<tr>
<td>4</td>
<td>1.5</td>
<td>7.5</td>
<td>0.128</td>
</tr>
<tr>
<td>5</td>
<td>1.8</td>
<td>9.0</td>
<td>0.154</td>
</tr>
</tbody>
</table>

The preparations of the soup were made under supervision of Assoc.-Prof. Dr. Klaus Dürrschmid at the Vienna University of Natural Resources and Life Sciences. For one liter soup one sour cream jar (250g, brand ‘milfina’, 15% fat) and 35g of cake flour (brand ‘Clever’) were mashed together, filled with approximately 250ml of water, mixed thoroughly and transferred in a 1L volumetric flask through a funnel. The residues were transferred with tap water into the volumetric flask. Afterwards water was filled up to the 1L mark. The volumetric flask was agitated thoroughly and divided into five 200ml glass bottles. One magnetic rod was in each of these closed bottles for the magnetic stirrer, so that the soup does not clump or burn when the soup in the bottles was heated to 60°C (see Figure 9). The closed glass bottles prevent evaporation of the cream soup. After the soup reached the temperature, five different amounts of salt (brand ‘Bad Ischler Tafelsalz’) were added (see Table 7) and the bottles were agitated. These five salt concentrations were assessed in a pre-test with 15 random students of the University of Natural Resources and Life Sciences. The concentration steps were 1.5 per step, starting with a concentration of 3.0 g/l to the highest concentration, which was 9.0 g/l.
Accordingly the soups were filled in 20ml plastic cups (see Figure 10) with numbered plastic lid and deep frozen at -20°C. The portioned cream soups were reheated in a microwave right before testing to 50°C ± 1°C for all test sessions. The test persons’ liking scores of the cream soups were assessed with a 9-point hedonic scale (see Table 8).

### Table 8: Liking scores for cream soup

<table>
<thead>
<tr>
<th>9-Point Hedonic Scale</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>9</td>
<td>Like Extremely</td>
<td>Liking area</td>
</tr>
<tr>
<td>8</td>
<td>Like Very Much</td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>Like Moderately</td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>Like Slightly</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>Neither Like nor Dislike</td>
<td>Neutral</td>
</tr>
<tr>
<td>4</td>
<td>Dislike Slightly</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>Dislike Moderately</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>Dislike Very Much</td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>Dislike Extremely</td>
<td></td>
</tr>
</tbody>
</table>
The 9-point hedonic scale is the most commonly used scale in sensory evaluation. It is suitable for acceptance testing and was developed by Jones et al. and Peryam and Pilgrim. Originally the scale was developed for the purpose of measuring food preferences of soldiers at the U.S. Armed Forces, but was quickly adopted by the food industry. [Jones et al., 1955][Peryam and Pilgrim, 1957] Because of the easy understanding composition and the simple application the scale is particularly eligible for sensory evaluation.

3.6 Test Environment

The International Organization for Standardization (ISO) standard for the design of test rooms for sensory analysis of foods states the following:

‘Conduct sensory evaluation under constant controlled conditions with minimum distractions to reduce the effects psychological factors and physical conditions can have on human judgment.’ [ISO 8589]

Since the test persons consisted of obesity patients of the Vienna General Hospital, the test room had to be as close to the ward as possible. The patients were at the Vienna General Hospital to go under a gastric bypass (Roux-en-Y or omega loop bypass) surgery the next day. The sensory evaluation was made on the day of the admission to the hospital, approximately 1-2 hours after lunch (1:00-2:00 PM). The test persons were picked up from their room and taken to the test room, the conference room at the control point on level 21 (general surgery department). The room conditions were the same over all tests with minimum distractions (see Figure 11). Room temperature was kept constant at 21°C ± 1°C for all test sessions. Preparations were done in the anteroom, which also included a microwave for heating up the test soup and a sink.
The tests and purpose of the study was explained to the test persons and the declaration of consent was signed. The participation was voluntary therefore the test persons were able to quit the study at any time. There were no expected health benefits for the test persons from their participation in this study. There were no risks or side effects for the participants.

The control group consisted of healthy persons with a BMI between 16.1 and 32.8. Healthy test persons were recruited from the Institute of Environmental Health and from the Institute of Pathophysiology, both departments of the Vienna Medical University, as well as at the company SGS Austria. Methods of recruitment were made across circular emails within the departments, where healthy volunteers within 30 and 65 years, without diabetes and hypertension and with no impaired sense of taste (also no intake of medication which can influence taste) were demanded. The test persons were asked not to eat or smoke at least one hour before the sensory test.

Test rooms of the control persons were the conference room of the Institute of Environmental Health, the seminar room in the Institute of Pathophysiology and the break room of SGS Austria. These test rooms had the same testing conditions as the test room of the Vienna General Hospital patients.
4 Results and discussion

Statistical analysis was performed on IBM© SPSS© Statistics Version 20.

4.1 Test persons

The patients group consisted of 30 (18 female and 12 male), obese patients with a mean BMI of 43.4 kg/m² (standard deviation (SD): 5.9 kg/m²). From those 30 patients 13 had hypertension and 8 had diabetes. The healthy control group is made up of 23 (9 female and 14 male) persons with a mean BMI of 24.0 kg/m² (SD: 3.5 kg/m²) (see Table 9).

<table>
<thead>
<tr>
<th>Group</th>
<th>gender</th>
<th>age [years]</th>
<th>BMI [kg/m²]</th>
<th>Major comorbidities</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patients</td>
<td>Female: 18</td>
<td>Mean ± SD: 44.9 ± 12.7</td>
<td>Mean ± SD: 43.4 ± 5.9</td>
<td>Hypertension: 13 Diabetes: 8</td>
</tr>
<tr>
<td></td>
<td>Male: 12</td>
<td>Range: 17-66</td>
<td>Range: 29.8-51.3</td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>Female: 9</td>
<td>Mean ± SD: 42.9 ± 14.6</td>
<td>Mean± SD: 24.0 ± 3.5</td>
<td>No hypertension, no diabetes</td>
</tr>
<tr>
<td></td>
<td>Male: 14</td>
<td>Range: 22-62</td>
<td>Range: 16.6-32.8</td>
<td></td>
</tr>
</tbody>
</table>

The age distribution of all test persons, divided in 7 age categories, is shown in Figure 12.

Table 10 contains the age categories, age in years and the frequency of all test persons.
The distribution of the BMI in the control and patients group is shown in Figure 12.
Thirteen of the 30 patients (43.3%) had hypertension and eight patients (26.7%) had diabetes (see Table 9). These data suggest that obesity patients apparently often might have comorbidities like hypertension or diabetes. Those diseases may also act as a confounder in this study. With a sample size of n=30 in the patients group, no convincing conclusion about the influences of these diseases on potential changes in salt taste can be made.

4.2 Salt detection threshold

The test on normal distribution was executed to test if the data (salt detection thresholds) is normally distributed. The result was that the distribution of salt detection thresholds (SDT) is significantly (Kolmogorov-Smirnov test: p=0.00, Shapiro-Wilk test: p=0.00) normally distributed. One single salt detection threshold value lies at 1.67 g NaCl/l with a z-score of 3.48. This value is an outlier and the distribution would be more normal if there was no outlier, but we decided to include this value to the statistical analysis since the value is a valid test result.
Andy Field explains that in a normal distribution it is expected that 5% of the absolute value (z-score) is greater than 1.96, and 1% to have absolute values greater than 2.58, and none to be greater than about 3.29. Table 11 illustrates the frequency of z-scores of salt taste detection threshold outliers. [Field, 2009]

<table>
<thead>
<tr>
<th>Outliers</th>
<th>Frequency</th>
<th>Percent</th>
</tr>
</thead>
<tbody>
<tr>
<td>Absolute z-score less than 2</td>
<td>48</td>
<td>90.57</td>
</tr>
<tr>
<td>Absolute z-score greater than 1.96</td>
<td>4</td>
<td>7.55</td>
</tr>
<tr>
<td>Absolute z-score greater than 2.58</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Absolute z-score greater than 3.29</td>
<td>1</td>
<td>1.88</td>
</tr>
</tbody>
</table>

Since the test for normal distribution was significant, parametric tests were performed for the normal distributed salt detection thresholds.

The SPSS descriptive analysis revealed that the mean (± SD) salt detection threshold for the total sample was 0.49 ± 0.33 g NaCl per liter.

In the patients group, the mean (± SD) salt detection threshold was 0.45 ± 0.28 g/l, whereas the mean (± SD) in the control group was 0.55 ± 0.40 g/l (see Table 12).

<table>
<thead>
<tr>
<th>Sample</th>
<th>Salt detection threshold [g/l]</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total</td>
<td>Mean (± SD) 0.49 ± 0.33</td>
</tr>
<tr>
<td>Patients</td>
<td>Mean (± SD) 0.45 ± 0.28</td>
</tr>
<tr>
<td>Control</td>
<td>Mean (± SD) 0.55 ± 0.40</td>
</tr>
</tbody>
</table>

The t-test for independent variables implies that there is no significant difference in the mean salt detection threshold between the two groups (p=0.097, F=2.859). Figure 14
patients, right: controls) and Figure 15 show the salt detection thresholds in the patients and the control group.

![Figure 14: boxplot of salt detection thresholds in the 2 groups (SPSS explorative data analysis, x-axes: patients (left), controls (right), y-axes: salt detection threshold [g NaCl/l])](image)

![Figure 15: bar chart of the frequency of assorted salt detection thresholds (blue=patients, green=controls, x-axes: salt detection threshold [g NaCl/l], y-axes: frequency)](image)

The 95% confidence interval (CI) of the salt detection threshold in the patients group lies between 0.35 and 0.56 g NaCl/l and in the control group the 95% CI goes from 0.38 to 0.72 g NaCl/l.

We also used the Mann-Whitney-Test to see if it would reveal different results, since the data includes one outlier. Normally the Mann-Whitney-Test is performed for non parametric data. The asymptotic significance (2-sided) of the mean salt detection threshold of the two groups would be $P=0.517$. Therefore a non parametric method would also reveal that there is no significant difference between the mean salt detection threshold of the patients and the control group.
The main hypothesis of this study, that the salt taste detection may be modified in obese persons compared to non-obese persons could not be verified. The study results suggest no significant differences in the mean salt detection thresholds in obese test persons compared to non-obese individuals.

An explanation for those not significant results may be that the ability of (salt) taste is influenced by many factors such as genetic variations, environmental influences and diseases, but apparently the results of this study suggest that obesity does not have a significant relation with salt taste detection thresholds. Also the relatively low sample size of 30 patients and 23 control test persons may hide potential relations between a high BMI and salt taste acuity, although similar studies used similar or lower sample sizes.[Bueter et al., 2011] [Burge et al., 1995]

Our study results match with the results of Simchen et al., which results in no significant differences in salty and sweet taste perception, but in bitter and sour tastes [Simchen et al., 2006].

The study results of Pasquet et al. (2007) display significant lower recognition thresholds for sucrose and sodium chloride in obese adolescents compared to non-obese adolescents. Possible explanations for those different findings could be the use of different sensory evaluation methods or the fact that Pasquet et al. observed adolescents only whereas the test persons in this study consisted of adults with a mean age of 44.15 ± SD 12.5 years.

4.2.1 Gender differences in salt detection threshold

Further t-tests were performed to test if there were gender differences in the mean salt detection thresholds.

There were neither gender differences of the salt detection thresholds in the total sample (p=0.776) nor in the patients (p=0.939) and control group (p=0.955) (see Table 13).
Table 13: t-test for independent samples of mean salt detection threshold in female and male test persons and significance

<table>
<thead>
<tr>
<th>Mean (± SD) salt detection threshold (SDT)</th>
<th>Total sample</th>
<th>patients</th>
<th>controls</th>
</tr>
</thead>
<tbody>
<tr>
<td>Female</td>
<td>0.48 (± 0.31)</td>
<td>0.46 (± 0.28)</td>
<td>0.51 (± 0.37)</td>
</tr>
<tr>
<td>Male</td>
<td>0.51 (± 0.37)</td>
<td>0.45 (± 0.29)</td>
<td>0.57 (± 0.43)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Significance (t-test for independent samples)</th>
<th>P-value</th>
<th>F-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>P-value</td>
<td>0.776</td>
<td>0.939</td>
</tr>
<tr>
<td>F-value</td>
<td>0.082</td>
<td>0.006</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.003</td>
</tr>
</tbody>
</table>

The results of this study show that there are no significant differences of the salt detection threshold between women and men in all groups. From this data we can conclude that women and men probably have analogous salt taste acuity.

4.2.2 Influences of diabetes and hypertension on salt taste detection

Furthermore, there were no significant differences between the mean salt detection threshold of hypertensive patients and patients without hypertension in the total sample (P=0.263) and in the patients group (P=0.854). Also no significant mean SDT differences of diabetics and non diabetics were observed in the total sample (P=1.73), as well as in the patients sample (P=0.520) (see Table 14).
Table 14: Mean SDT of test persons with/without hypertension or diabetes and significance (t-test for independent samples)

<table>
<thead>
<tr>
<th>Diseases</th>
<th>Total sample</th>
<th>Patients</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean salt detection threshold (± SD)</td>
<td>P-value (F-value)</td>
<td></td>
</tr>
<tr>
<td>No hypertension</td>
<td>0.48</td>
<td>0.46</td>
<td>0.263 (1.281)</td>
</tr>
<tr>
<td>Hypertension</td>
<td>0.51</td>
<td>0.45</td>
<td></td>
</tr>
<tr>
<td>No diabetes</td>
<td>0.49</td>
<td>0.41</td>
<td>0.173 (1.914)</td>
</tr>
<tr>
<td>Diabetes</td>
<td>0.57</td>
<td>0.57</td>
<td></td>
</tr>
</tbody>
</table>

Because of the results of the non significant differences of the mean salt detection thresholds between test persons with diabetes, hypertension and the healthy test persons, our second hypothesis was also rejected. Therefore our study results show no significant relation between diseases like type 2 diabetes and hypertension and an altered salt taste perception. In this study the results reveal that diseases like type 2 diabetes and hypertension did not modify salt taste acuity.

4.3 Preference for different concentrations of salt in soup

Test persons were instructed to evaluate their likings for 5 soups with different salt concentrations on a 9-point hedonic scale (see Table 8). The lowest rating (‘Dislike Extremely’) was rated with number 1 and the highest rating (‘Like Extremely’) was rated with number 9. For the statistical analysis, all liking rates were recorded with the numbers 1-9.

Mean liking scores and standard deviations of soups 1-5 are listed in Table 15.
Table 15: Mean Liking Scores and SD of soup 1-5 in all groups

<table>
<thead>
<tr>
<th>Soup</th>
<th>Group</th>
<th>Mean Liking Score</th>
<th>Standard Deviation (SD)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Soup 1</strong></td>
<td>Patients (n=30)</td>
<td>2.97</td>
<td>2.14</td>
</tr>
<tr>
<td></td>
<td>Control (n=23)</td>
<td>4.13</td>
<td>2.26</td>
</tr>
<tr>
<td></td>
<td>Total (n=53)</td>
<td>3.47</td>
<td>2.25</td>
</tr>
<tr>
<td><strong>Soup 2</strong></td>
<td>Patients</td>
<td>3.13</td>
<td>2.46</td>
</tr>
<tr>
<td></td>
<td>Control</td>
<td>4.83</td>
<td>1.95</td>
</tr>
<tr>
<td></td>
<td>Total</td>
<td>3.87</td>
<td>2.39</td>
</tr>
<tr>
<td><strong>Soup 3</strong></td>
<td>Patients</td>
<td>3.17</td>
<td>2.49</td>
</tr>
<tr>
<td></td>
<td>Control</td>
<td>4.96</td>
<td>2.21</td>
</tr>
<tr>
<td></td>
<td>Total</td>
<td>3.94</td>
<td>2.52</td>
</tr>
<tr>
<td><strong>Soup 4</strong></td>
<td>Patients</td>
<td>3.57</td>
<td>2.74</td>
</tr>
<tr>
<td></td>
<td>Control</td>
<td>4.00</td>
<td>2.28</td>
</tr>
<tr>
<td></td>
<td>Total</td>
<td>3.75</td>
<td>2.53</td>
</tr>
<tr>
<td><strong>Soup 5</strong></td>
<td>Patients</td>
<td>2.93</td>
<td>2.72</td>
</tr>
<tr>
<td></td>
<td>Control</td>
<td>3.61</td>
<td>2.45</td>
</tr>
<tr>
<td></td>
<td>Total</td>
<td>3.23</td>
<td>2.60</td>
</tr>
</tbody>
</table>

In the total sample, soup 3 has the highest liking score with a mean (± SD) of 3.94 ± 2.52. The obese test persons rated soup 4 highest with a mean (± SD) liking score of 3.57 ± 2.74, whereas the control persons had the highest mean liking score at soup 3 with a mean (± SD) of 4.96 ± 2.21. Soup 5 had the lowest liking scores in all groups (total sample, patients and controls).
Figure 16: Boxplot illustration of soup 1-5 in patients and control group, *significant differences (soup 2 p=0.009, soup 3 p=0.009) in mean soup likings between patients and control groups (univariat analysis of variance), x-axes: groups, y-axes: liking score

In order to be able to assess the statistical significance of the soup liking differences, univariat analyses of variance were performed (see Table 16).

Table 16: Significance of univariat analysis of variance for differences in the mean soup liking scores between patients (n=30) and control group (n=23)

<table>
<thead>
<tr>
<th>Significance</th>
<th>Soup 1</th>
<th>Soup 2</th>
<th>Soup 3</th>
<th>Soup 4</th>
<th>Soup 5</th>
</tr>
</thead>
<tbody>
<tr>
<td>P-value</td>
<td>0.061</td>
<td>0.009</td>
<td>0.009</td>
<td>0.542</td>
<td>0.353</td>
</tr>
<tr>
<td>F-value</td>
<td>3.662</td>
<td>7.352</td>
<td>7.408</td>
<td>0.376</td>
<td>0.877</td>
</tr>
</tbody>
</table>
The univariate analysis of variance results in significant mean liking differences of soup 2 (P=0.009) and soup 3 (P=0.009) between the patients and the control group. No significant mean liking differences of patients and control group were calculated in the other soups (soup 1, soup 4 and soup 5).
Figure 17: Bar graphs of soup 1-5 and quantity of liking scores (blue=patients, green=controls), x-axes: liking scores (1-9) of current soup, y-axes: frequency

Figure 17 shows the bar graphs of the 5 soups and the quantity of liking scores (1-9) of patients (blue bars) and controls (green bars). Figure 17 also shows visible differences of the liking scores between patients and controls. Patients had rated all soups more often with a score of 1 than control persons did.
Figure 18 illustrates line graphs of the liking scores of patients (red) and control persons (green) for soup 1-5. In most of the soups more than 1 peak of the liking scores are shown in both groups, therefore comparisons of means might not be convincing. On account of this a non parametric test (Mann-Whitney-U test) was performed additionally. The result of the Mann-Whitney-U test is shown in Table 17. Results revealed significant differences of soup liking scores in soup 1, 2 and 3 between patients and control group.
Table 17: Asymptotic significance of Mann-Whitney-U test for independent samples in the soup liking scores between patients (n=30) and control group (n=23)

<table>
<thead>
<tr>
<th>Soup</th>
<th>Significance (asymptotic)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Soup 1</td>
<td>0.040</td>
</tr>
<tr>
<td>Soup 2</td>
<td>0.008</td>
</tr>
<tr>
<td>Soup 3</td>
<td>0.009</td>
</tr>
<tr>
<td>Soup 4</td>
<td>0.361</td>
</tr>
<tr>
<td>Soup 5</td>
<td>0.216</td>
</tr>
</tbody>
</table>

Test persons who didn’t like the cream soup at all rated all 5 soups with a score of 1. Therefore an additional analysis of variance was performed without test persons who rated all soups with a score of 1. After exclusion of all test persons with a computed mean soup liking score of 1, the mean liking scores and standard deviations are shown in Table 18.

Table 18: Mean liking scores of patients and controls without test persons with a mean liking score of 1

<table>
<thead>
<tr>
<th>Soup</th>
<th>Group</th>
<th>Mean liking score</th>
<th>SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>Soup 1</td>
<td>Patients (n=22)</td>
<td>3.68</td>
<td>2.079</td>
</tr>
<tr>
<td></td>
<td>Controls (n=22)</td>
<td>4.27</td>
<td>2.208</td>
</tr>
<tr>
<td>Soup 2</td>
<td>Patients (n=22)</td>
<td>3.91</td>
<td>2.448</td>
</tr>
<tr>
<td></td>
<td>Controls (n=22)</td>
<td>5.00</td>
<td>1.799</td>
</tr>
<tr>
<td>Soup 3</td>
<td>Patients (n=22)</td>
<td>3.95</td>
<td>2.478</td>
</tr>
<tr>
<td></td>
<td>Controls (n=22)</td>
<td>5.14</td>
<td>2.077</td>
</tr>
<tr>
<td>Soup 4</td>
<td>Patients (n=22)</td>
<td>4.50</td>
<td>2.632</td>
</tr>
<tr>
<td></td>
<td>Controls (n=22)</td>
<td>4.14</td>
<td>2.232</td>
</tr>
<tr>
<td>Soup 5</td>
<td>Patients (n=22)</td>
<td>3.64</td>
<td>2.871</td>
</tr>
<tr>
<td></td>
<td>Controls (n=22)</td>
<td>3.73</td>
<td>2.434</td>
</tr>
</tbody>
</table>
The univariat analysis of variance reveals no more significant differences in mean soup likings between patients and control group when test persons with a mean soup liking scores of 1 were excluded (see Table 19).

**Table 19: Significance of univariat analysis of variance for differences in the mean soup liking scores between patients (n=22) and control group (n=22) without test persons with a mean liking score of 1**

<table>
<thead>
<tr>
<th>Significance</th>
<th>Soup 1</th>
<th>Soup 2</th>
<th>Soup 3</th>
<th>Soup 4</th>
<th>Soup 5</th>
</tr>
</thead>
<tbody>
<tr>
<td>P-value</td>
<td>0.366</td>
<td>0.100</td>
<td>0.094</td>
<td>0.624</td>
<td>0.910</td>
</tr>
<tr>
<td>F-value</td>
<td>0.835</td>
<td>2.837</td>
<td>2.939</td>
<td>0.244</td>
<td>0.013</td>
</tr>
</tbody>
</table>

A multivariate analysis of covariance of mean liking differences between the patients and control group by correcting for soup means was also executed (see Table 20). The results show that there were significant differences of the mean liking in soup 4 between patients and controls by correcting for the soup means.

**Table 20: Multivariate analysis of covariance of mean liking differences between the 2 groups by correcting for computed soup means**

<table>
<thead>
<tr>
<th>Significance</th>
<th>Soup 1</th>
<th>Soup 2</th>
<th>Soup 3</th>
<th>Soup 4</th>
<th>Soup 5</th>
</tr>
</thead>
<tbody>
<tr>
<td>P-value</td>
<td>0.555</td>
<td>0.088</td>
<td>0.090</td>
<td><strong>0.015</strong></td>
<td>0.153</td>
</tr>
<tr>
<td>F-value</td>
<td>0.353</td>
<td>3.030</td>
<td>2.990</td>
<td>6.293</td>
<td>2.103</td>
</tr>
</tbody>
</table>

Generally the univariat analysis of variance shows that the patients had significant lower mean liking ratings of soup 2 and 3 compared to the non obese control group. The non parametric Mann-Whitney-U test revealed significant differences in soup 1, 2 and 3 in soup likings between patients and control group.

There were no significant differences in mean liking scores of the other soups between patients and control group, except for soup 4 but only when the computed soup mean
was included to the multivariate analysis of covariance. When test persons with a mean liking score of 1 were excluded, there were no significant mean liking differences from the univariate analysis of variance.

Further on a multivariate analysis of covariance with diabetes and hypertension as covariates revealed no significant differences in the mean likings scores of all soups between the patients and the controls.

To observe differences of the soups among themselves within the patients and control group, a contrast analysis in the 2 groups within an analysis of repeated measures was performed. Within the patients group the analysis of repeated measures (with soup 5 as the reference category) showed significant differences between soup 4 and 5 (P=0.047, F=4.310), but no significant differences between soup 1-3 and soup 5 (see Table 21).

<table>
<thead>
<tr>
<th>Soup</th>
<th>Significance (P-value)</th>
<th>F-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Soup 1 vs. 5</td>
<td>0.942</td>
<td>0.005</td>
</tr>
<tr>
<td>Soup 2 vs. 5</td>
<td>0.662</td>
<td>0.195</td>
</tr>
<tr>
<td>Soup 3 vs. 5</td>
<td>0.647</td>
<td>0.214</td>
</tr>
<tr>
<td>Soup 4 vs. 5</td>
<td><strong>0.047</strong></td>
<td>4.310</td>
</tr>
</tbody>
</table>

The analysis of repeated measures revealed significant differences between soup 2 (P=0.002, F=12.949) and soup 5, as well as soup 3 (P=0.001, F=13.675) and soup 5 in the control group (see Table 22).
Table 22: Analysis of repeated measures (with soup 5 as the reference category) in the control group

<table>
<thead>
<tr>
<th>Soup</th>
<th>Significance (P-value)</th>
<th>F-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Soup 1 vs. 5</td>
<td>0.270</td>
<td>1.278</td>
</tr>
<tr>
<td>Soup 2 vs. 5</td>
<td>0.002</td>
<td>12.949</td>
</tr>
<tr>
<td>Soup 3 vs. 5</td>
<td>0.001</td>
<td>13.675</td>
</tr>
<tr>
<td>Soup 4 vs. 5</td>
<td>0.288</td>
<td>1.183</td>
</tr>
</tbody>
</table>

The non parametric Kruskal-Wallis-test results in significant mean liking differences in soup 1 (P=0.040), soup 2 (P=0.008) and soup 3 (P=0.009), but not soup 4 (P=0.361) and soup 5 (P=2.16) between the patients and the control group. These results, significant differences in the mean liking scores of soups 1-3, are similar to the parametric univariate analysis of variance with the exception of soup 1, which was not significant in the parametric test.

In the patients group the highest mean liking score was 3.57 for soup 4. The soup with the highest mean liking score in the control group was soup 3 with a score of 4.96.

Since soup 3 has a lower salt concentration than soup 4, we suspect that the obese patients may prefer meals with higher salt content more than non obese persons. On the other hand soup 5, which has the highest salt concentrations (9g NaCl/l), had the lowest mean liking scores in both groups. Hence the well-known mechanism of an increasing salt concentration transforming into an aversive stimulus is established. The cellular substrate for salt aversion was unknown, but Oka et al. (2013) showed that high salt recruits the two primary aversive taste pathways by activating the sour- and bitter-taste-sensing cells [Oka et al., 2013].

One important confounder in the soup liking ratings is the willingness of the test persons to participate in this study. Whereas the healthy volunteers were all enthusiastic to participate, the obese patients participated voluntarily on the one hand, but on the
other hand patients were waiting for their gastric bypass surgeries and part-way only participated because they had nothing else to do in the hospital. Another reason for the lower interest could be that the patients probably did not want to disappoint hospital staff by denying the test. A greater part of the patients were already overanxious because of the surgery and were not able to eat the cream soup. Those test persons rated all soups with the lowest liking scale of 1. Nevertheless no significant differences in the salt detection thresholds were found, which could imply a similar interest of patients and control persons in the sensory evaluation testing.
4.3.1 Gender differences in cream soup likings

The univariat analysis of variance was performed to test potential differences of soup likings between female and male test persons. Mean liking scores and standard deviations are listed in Table 23 and the significance (P- and F-values) is presented in Table 24.

<table>
<thead>
<tr>
<th>Soup</th>
<th>Group</th>
<th>Mean Liking Score</th>
<th>Standard Deviation (SD)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Soup 1</strong></td>
<td>Female (n=27)</td>
<td>2.56</td>
<td>2.08</td>
</tr>
<tr>
<td></td>
<td>Male (n=26)</td>
<td>4.42</td>
<td>2.04</td>
</tr>
<tr>
<td></td>
<td>Total (n=53)</td>
<td>3.47</td>
<td>2.25</td>
</tr>
<tr>
<td><strong>Soup 2</strong></td>
<td>Female</td>
<td>2.63</td>
<td>1.98</td>
</tr>
<tr>
<td></td>
<td>Male</td>
<td>5.15</td>
<td>2.09</td>
</tr>
<tr>
<td></td>
<td>Total</td>
<td>3.87</td>
<td>2.39</td>
</tr>
<tr>
<td><strong>Soup 3</strong></td>
<td>Female</td>
<td>2.67</td>
<td>1.86</td>
</tr>
<tr>
<td></td>
<td>Male</td>
<td>5.27</td>
<td>2.44</td>
</tr>
<tr>
<td></td>
<td>Total</td>
<td>3.94</td>
<td>2.52</td>
</tr>
<tr>
<td><strong>Soup 4</strong></td>
<td>Female</td>
<td>2.67</td>
<td>2.08</td>
</tr>
<tr>
<td></td>
<td>Male</td>
<td>4.88</td>
<td>2.50</td>
</tr>
<tr>
<td></td>
<td>Total</td>
<td>3.75</td>
<td>2.53</td>
</tr>
<tr>
<td><strong>Soup 5</strong></td>
<td>Female</td>
<td>2.15</td>
<td>2.07</td>
</tr>
<tr>
<td></td>
<td>Male</td>
<td>4.35</td>
<td>2.50</td>
</tr>
<tr>
<td></td>
<td>Total</td>
<td>3.23</td>
<td>2.60</td>
</tr>
</tbody>
</table>
Table 24: Univariate analyses of variance for differences in the mean liking scores between females and males

<table>
<thead>
<tr>
<th>Sample</th>
<th>Significance</th>
<th>Soup 1</th>
<th>Soup 2</th>
<th>Soup 3</th>
<th>Soup 4</th>
<th>Soup 5</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>P-value</td>
<td>0.002</td>
<td>0.000</td>
<td>0.000</td>
<td>0.001</td>
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<tr>
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<td>F-value</td>
<td>10.856</td>
<td>20.333</td>
<td>19.135</td>
<td>12.369</td>
<td>11.360</td>
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<tr>
<td>Patients</td>
<td>P-value</td>
<td>0.045</td>
<td>0.026</td>
<td>0.022</td>
<td>0.072</td>
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<tr>
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<td>F-value</td>
<td>4.398</td>
<td>5.498</td>
<td>5.876</td>
<td>3.508</td>
<td>1.863</td>
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<tr>
<td>Controls</td>
<td>P-value</td>
<td>0.052</td>
<td>0.000</td>
<td>0.002</td>
<td>0.002</td>
<td>0.001</td>
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<tr>
<td></td>
<td>F-value</td>
<td>4.234</td>
<td>17.644</td>
<td>12.031</td>
<td>11.827</td>
<td>15.467</td>
</tr>
</tbody>
</table>

There are significant differences of mean soup likings between female and male test persons in all soups (soup 1: P=0.002, soup 2: P=0.000, soup 3: P=0.000, soup 4: P=0.001, soup 5: P=0.001) in the total sample. Figure 19 shows a boxplot illustration of female and male group in the total sample and their soup likings. Female individuals had significant lower liking ratings for all soups.
Significant gender differences of mean soup likings were found in soup 1 (P<0.045), 2 (P<0.026) and 3 (P<0.022) in the patients group, but not in soup 4 and 5 (see Table 24). In the control group significant mean liking differences between female and males were found in soup 2 (P<0.000), 3 (P<0.002), 4 (P<0.002) and 5 (P<0.001), but not in soup 1. Figure 20 illustrates boxplot images of female and male groups in the patients (left) and control group (right) and their soup liking scores.
A possible explanation for the lower liking ratings in the female groups could be that women generally may have a lower milk and dairy products intake than men.

Liebman et al. (2003) observed gender differences in selected dietary intakes in 1817 adults with the food frequency survey. The study concludes that women reported lower intakes of milk (P=0.0034) and sweetened beverages such as soft drinks compared to men. The frequency of milk consumption was also lower in female individuals. Twentyone percent of the women reported that they ‘never’ consume milk compared to 14% of the men. Women also reported higher intakes of fruits and vegetables. The data of this study suggested that the diets of female persons were more nutrient-dense, with the exception of milk-derived calcium. Liebman et al. (2003) assume that a higher prevalence of low milk intakes in women may result in higher prevalence of osteoporosis among women. Due to that optimal calcium intake plays an important role in the maintenance of skeletal mass during adulthood. Intake of dairy products in general was not analyzed in this study, but Liebman et al. (2003) refers that there is evidence in the literature for the assertion that males typically consume more dairy products. [Liebman et al., 2003]
The recent study of Samara et al. (2013) shows a significantly higher consumption of total dairy products and cheese (as servings per day) in men compared to women, but the total dairy intake expressed per 1 megajoule (MG) of energy was significantly higher in women. In women, total dairy consumption was positively related to weight (P=0.004), BMI (P=0.023), waist circumference (P=0.003), and triglyceride concentration (P=0.004) and negatively to HDL-concentration (P=0.004). Then again a higher consumption of dairy products was associated with positive changes in the metabolic profile in a 5-years period but only in men. [Samara et al., 2013]

Also the review of Kalergis et al. (2013) implies a link between dairy intake and a reduced risk for type 2 diabetes. Potential underlying mechanisms include the role of dairy products, such as calcium, vitamin D, dairy fat and trans-palmitoleic acid. But the role of specific dairy products needs to be clarified and more research is needed. [Kalergis et al. 2013]

No studies about a different liking of dairy products of obese persons compared to normal weight persons were found.

To assess the possible reasons for the distinctions in the liking ratings of our test soup between men and women, further investigations are required.
5 Summary

The study addresses possible differences in salt taste detection between obese test persons and non obese individuals. Furthermore possible influences of typical comorbidities of obesity like type 2 diabetes and hypertension on salt taste detection have been observed in this study. Moreover the potential liking of food with a greater salt content in obese individuals have been tested by evaluating the hedonic liking of 5 cream soups varying in their salt content.

The current available literature is limited regarding obesity and changes in salt taste detection.

In this study 30 obese test persons were recruited from the obesity clinic of the Vienna General Hospital. The patients group consisted of 18 female and 12 male obese test persons with a mean age of 44.9 ± 12.7 years and a mean BMI of 43.4 ± 5.9 kg/m². For the gender and age matched, non obese control group, 23 healthy volunteers were recruited from several departments of the Vienna Medical University and also other companies. Nine female and 14 male volunteers participated in this study. The mean age of the control group was 42.9 ± 15.6 years and the mean BMI was 24.0 ± 3.5 kg/m².

In order to assess the salt detection threshold the 3-alternative forced choice test was performed. For the assessment of pleasantness of test meals varying in salt content 5 different cream soups were offered to the participants and their hedonic ratings were listed in a 9-point hedonic scale.

The results showed no significant differences in the mean salt detection threshold between patients and control persons (p=0.097). Diseases like type 2 diabetes and hypertension also had no significant effect on the salt detection thresholds in our study. There were neither gender differences of the salt detection thresholds in the total sample (p=0.776) nor in the patients (p=0.939) and control group (p=0.955).

The results of the pleasantness of cream soup varying in salt content revealed significant mean liking differences of soup 2 (P=0.009) and soup 3 (P=0.009) between
the patients and the control group. Patients had lower liking ratings for all soups compared to the non obese test persons. Test persons who didn’t like the cream soup at all rated all 5 soups with a score of 1. After exclusion of those test persons with a computed mean soup liking score of 1, there were no significant differences between the soup likings between patients and control group anymore.

The highest mean liking score was 3.57 for soup 4 in the patients group. The soup with the highest mean liking score in the control group was soup 3 with a score of 4.96. Since soup 3 has a lower salt concentration than soup 4, we suspect that the obese persons may prefer meals with higher salt content more than non obese persons.

There are significant differences of mean soup likings between female and male test persons in all soups (soup 1: P=0.002, 2: P=0.000, 3: P=0.000, 4: P=0.001, 5: P=0.001) in the total sample. Female individuals had significant lower liking ratings for all soups compared to male test persons. Significant gender differences of mean soup likings were found in soup 1 (P=0.045), 2 (P=0.026) and 3 (P=0.022) in the patients group. In the control group significant mean liking differences between female and males were found in soup 2 (P=0.000), 3 (P=0.002), 4 (P=0.002) and 5 (P=0.001). A possible explanation for the lower liking ratings in the female groups could be that women generally may have a lower milk and dairy products intake than men. To assess the possible reasons for the distinctions in the liking ratings of our test soup between men and women, further investigations are required.

Die derzeit verfügbare Literatur über Adipositas und Veränderungen des Salzgeschmacks ist limitiert.

In dieser Studie wurden 30 adipöse Testpersonen aus der Adipositas Ambulanz des Allgemeinen Krankenhauses (AKH) Wien rekrutiert. Die Patientengruppe bestand aus 18 weiblichen und 12 männlichen, adipösen Testpersonen mit einem durchschnittlichen Alter von 44,9 ± 12,7 Jahren und einem durchschnittlichem BMI von 43,4 ± 5,9 kg/m². Für die auf das Alter und Geschlecht abgestimmte, nicht adipöse Kontrollgruppe wurden 23 gesunde Freiwillige von verschiedenen Instituten der Medizinischen Universität und auch von anderen Firmen rekrutiert. Neun weibliche und 14 männliche Freiwillige haben an dieser Studie teilgenommen. Das durchschnittliche Alter der Kontrollgruppe beträgt 42,9 ± 15,6 Jahre und der durchschnittliche BMI liegt bei 24,0 ± 3,5 kg/m².

Um die Salzerkennungsschwelle zu bestimmen wurde der „3-alternative-forced choice“ Test ausgeführt. Für die Bestimmung der Beliebtheit von Testmahlzeiten mit unterschiedlichem Salzgehalt wurden 5 verschiedene Rahmsuppen gereicht und die hedonische Bewertung (likert-Skala bzw. likert-Werte) wurde mittels einer 9-Punkt hedonischen Skala ermittelt.

Die Studie ergab keine signifikanten Unterschiede in der durchschnittlichen Salzerkennungsschwelle zwischen der Patienten- und der Kontrollgruppe (p=0,097). Krankheiten wie Typ 2 Diabetes und Hypertonie hatten ebenfalls keinen signifikanten Effekt auf die Salzerkennungsschwelle in unserer Studie. Außerdem gab es keine
geschlechtsspezifischen Unterschiede bei der durchschnittlichen Salzerkennungsschwelle, weder im Gesamtsample (p=0,776) noch in der Patientengruppe (P=0,939) oder in der Kontrollgruppe (p=0,955).


Der höchste durchschnittliche likert-Wert war 3,57 für Suppe 4 in der Patientengruppe. Die Suppe mit dem höchsten durchschnittlichen likert-Wert in der Kontrollgruppe war Suppe 3 mit einem durchschnittlichen Wert von 4,96. Da Suppe 3 eine niedrigere Salzkonzentration als Suppe 4 hat, vermuten wir, dass adipöse Personen Mahlzeiten mit einem höheren Salzgehalt mehr bevorzugen könnten als nicht adipöse Personen.

Es gab signifikante Unterschiede bei den durchschnittlichen Suppen likert-Werten zwischen weiblichen und männlichen Testpersonen bei allen Suppen (Suppe 1 (p=0,002), 2 (p=0,000), 3 (p=0,000), 4 (p=0,001), 5 (p=0,001)) im Gesamtsample. Frauen hatten signifikant niedrigere likert-Werte für alle Suppen verglichen zu den Männern. Signifikante geschlechtsspezifische Unterschiede der durchschnittlichen Suppen likert-Werte wurden in Suppe 1 (p=0,045), 2 (p=0,026) und 3 (p=0,022) in der Patientengruppe gefunden. In der Kontrollgruppe gab es signifikante Unterschiede bei den durchschnittlichen likert-Werten zwischen Frauen und Männern bei Suppe 2 (p=0,000), 3 (p=0,002), 4 (p=0,002) und 5 (p=0,001). Eine mögliche Erklärung für die niedrigeren likert-Werte in der Frauengruppe könnte sein, dass Frauen generell eine niedrigere Milch- und Milchprodukteinnahme haben als Männer. Um die möglichen Gründe für die Unterschiede in den Bewertungen unserer Testsuppe zwischen Männern und Frauen festzustellen, sind weitere Untersuchungen erforderlich.
7 References


Field A. Discovering Statistics using SPSS, SAGE Publications Ltd, 2009


ISO 8589 - Sensory Analysis -- General Guidance for the Design of Test Rooms


8 Appendix

Teilnehmerinformation und Einwilligungserklärung  ii
Questionnaire  vii
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Teilnehmerinformation und Einwilligungserklärung
zur Teilnahme an der klinischen Studie
„Salzerkennungsschwelle und Geschmacksempfindung bei einer Testmahlzeit mit
unterschiedlicher Salzkonzentration bei fettleibigen PatientInnen vor und nach
einer Magenbypass-Operation“

Sehr geehrte Teilnehmerin, sehr geehrter Teilnehmer!

Wir laden Sie ein an der oben genannten klinischen Studie teilzunehmen. Die Aufklärung
darüber erfolgt in einem ausführlichen ärztlichen Gespräch.

Ihre Teilnahme an dieser Studie erfolgt freiwillig. Sie können jederzeit ohne Angabe von
Gründen aus der Studie ausscheiden. Die Ablehnung der Teilnahme oder ein vorzeitiges
Ausscheiden aus dieser Studie hat keine nachteiligen Folgen für Ihre medizinische Betreuung.

Klinische Studien sind notwendig, um verlässliche neue medizinische Forschungsergebnisse zu
gewinnen. Unverzichtbare Voraussetzung für die Durchführung einer klinischen Studie ist
jedoch, dass Sie Ihr Einverständnis zur Teilnahme an dieser klinischen Studie schriftlich
erklären. Bitte lesen Sie den folgenden Text als Ergänzung zum Informationsgespräch mit
Ihrem Arzt sorgfältig durch und zögern Sie nicht Fragen zu stellen.

Bitte unterschreiben Sie die Einwilligungserklärung nur

- wenn Sie Art und Ablauf der klinischen Studie vollständig verstanden haben,
- wenn Sie bereit sind, der Teilnahme zuzustimmen und
- wenn Sie sich über Ihre Rechte als Teilnehmer an dieser klinischen Studie im Klaren
  sind.

Zu dieser klinischen Studie, sowie zur Teilnehmerinformation und Einwilligungserklärung
wurde von der zuständigen Ethikkommission eine befürwortende Stellungnahme abgegeben.

Was ist der Zweck der klinischen Studie?

**Wie läuft die klinische Studie ab?**

Diese klinische Studie wird im Allgemeinen Krankenhaus der Stadt Wien stattfinden. Es werden insgesamt etwa 36 Personen daran teilnehmen.

Sie werden in einem gewissen Zeitrahmen 2 identische, sensorische Tests zur Erkennung der Salzkonzentrationsschwelle und zusätzlich Tests zur Ermittlung der am schmackhaftesten empfundenen Salzkonzentration in einer Testmahlzeit (Suppe) durchführen. Außerdem bitten wir Sie auch einen kurzen Fragebogen zur Salzpräferenz auszufüllen.

Der Test wird 1 Tag vor, sowie 3 Monate nach der bariatrischen Operation durchgeführt. Die normalgewichtigen Personen, die als Kontrolle dienen, werden einmalig getestet.

Der Test für die Salzkonzentrationsschwelle findet morgens statt. Ihnen werden 8 mal 3 Becher angeboten, wobei Sie jedes Mal jeweils 2 Becher eine Wasserlösung und 1 Becher eine Salzlösung, mit unterschiedlichen Salzkonzentrationen, erhalten. Sie müssen sich dann entscheiden, bei welcher der 3 Becher es sich um die Salzlösung handelt und dies auf einem Testformular ankreuzen.

Beim Test für die Vorlieben von verschiedenen Salzkonzentrationen in einer Suppe werden Ihnen 5 Suppenproben angeboten, wobei Sie den Geschmack bzw. das Gefallen an der Suppe auf einem Fragebogen mit einer Skala von 1 („schmeckt mir überhaupt nicht“) bis 9 („schmeckt mir sehr gut“) angeben müssen.

Der Fragebogen zur Salzpräferenz sollte bitte nach dem Geschmackstest ausgefüllt werden. Er beinhaltet 8 Fragen, in denen Sie vor allem nach Ihrer Vorliebe für salzhaltige Speisen und Ihre Kenntnis zu Salz in Lebensmitteln gefragt werden.

**Worin liegt der Nutzen einer Teilnahme an der Klinischen Studie?**
Es ist nicht zu erwarten, dass Sie aus Ihrer Teilnahme an dieser klinischen Studie gesundheitlichen Nutzen ziehen werden. Jedoch können Sie durch Ihre Teilnahme zur wissenschaftlichen Bereicherung beitragen und das Verständnis zur Salzproblematik verbessern. Auf persönlichen Wunsch können Sie am Ende der Studie, nach der Auswertung der Ergebnisse, ein Aufklärungsgespräch über die Salzproblematik erhalten.

**Gibt es Risiken und Nebenwirkungen?**

Es bestehen für Sie keinerlei Risiken und Nebenwirkungen.

**In welcher Weise werden die im Rahmen dieser klinischen Studie gesammelten Daten verwendet?**

Die Weitergabe der Daten erfolgt ausschließlich zu statistischen Zwecken und Sie werden darin ausnahmslos nicht namentlich genannt. Auch in etwaigen Veröffentlichungen der Daten dieser klinischen Studie werden Sie nicht namentlich genannt.

**Entstehen für die Teilnehmer Kosten? Gibt es einen Kostenersatz oder eine Vergütung?**

Durch Ihre Teilnahme an dieser klinischen Studie entstehen für Sie keine zusätzlichen Kosten.

**Möglichkeit zur Diskussion weiterer Fragen**

Für weitere Fragen im Zusammenhang mit dieser klinischen Studie stehen Ihnen Ihr Prüfarzt und seine Mitarbeiter gern zur Verfügung. Auch Fragen, die Ihre Rechte als PatientIn und TeilnehmerIn an dieser klinischen Studie betreffen, werden Ihnen gerne beantwortet.

**Kontaktpersonen:**

Ao. Univ. Prof. Dr. med. Cem Ekmekcioglu  
Ständig erreichbar unter: 40160-34927

Linda Lam, Bakk.rer.nat  
E-mail: linda_lam@gmx.at
Einwilligungserklärung

Name der Patientin/des Patienten in Druckbuchstaben:

...........................................................................


Ich erkläre mich bereit, an der klinischen Studie „Salzerkennungsschwelle und Beliebtheit verschiedener Salzkonzentrationen“ teilzunehmen.


Ich werde den ärztlichen Anordnungen, die für die Durchführung der klinischen Studie erforderlich sind, Folge leisten, behalte mir jedoch das Recht vor, meine freiwillige Mitwirkung jederzeit zu beenden, ohne dass mir daraus Nachteile für meine weitere medizinische Betreuung entstehen.

Ich bin zugleich damit einverstanden, dass meine im Rahmen dieser klinischen Studie ermittelten Daten aufgezeichnet werden. Um die Richtigkeit der Datenaufzeichnung zu überprüfen, dürfen Beauftragte des Auftraggebers und der zuständigen Behörden beim Prüfarzt Einblick in meine personenbezogenen Krankheitsdaten nehmen.

Beim Umgang mit den Daten werden die Bestimmungen des Datenschutzgesetzes beachtet.


.......................................................................................

(Datum und Unterschrift der Teilnehmerin/des Teilnehmers)

.......................................................................................

(Datum, Name und Unterschrift des verantwortlichen Arztes)
Salzstudie

Testperson Nr.: ____________
Datum: ____________

Liebe/r Teilnehmer/in!

Test 1: Salzerkennung
Bitte identifizieren Sie die salzhaltigen Proben!

1. □ 664 □ 176 □ 324
2. □ 312 □ 445 □ 237
3. □ 313 □ 857 □ 874
4. □ 573 □ 149 □ 443
5. □ 929 □ 615 □ 570
6. □ 630 □ 707 □ 485
7. □ 677 □ 294 □ 766
8. □ 180 □ 890 □ 358

Test 2: Beliebtheitstest
Bitte bewerten Sie die Suppen von „Mag ich überhaupt nicht“ bis „Mag ich besonders gern“.

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<th>Mag ich überhaupt nicht</th>
<th>Mag ich sehr wenig</th>
<th>Mag ich wenig</th>
<th>Mag ich nicht besonders</th>
<th>Mag ich weder noch</th>
<th>Mag ich etwas</th>
<th>Mag ich gern</th>
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Vielen Dank für die Teilnahme!
**Curriculum vitae**

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<tr>
<td>Name</td>
<td>Linda Lam</td>
</tr>
<tr>
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</tr>
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### Berufserfahrung

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<td>01/2008 - heute</td>
<td><strong>CIP GmbH, Wien</strong> (freier Dienstnehmer)</td>
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<tr>
<td></td>
<td>adidas und Reebok Produktttrainer</td>
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<tr>
<td>10/2010 - heute</td>
<td><strong>Medizinische Universität Wien</strong> (geringfügig)</td>
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<tr>
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<td>Assistentin beim Universitätslehrgang „Grundlagen und Praxis der TCM“</td>
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<tr>
<td>10/2011 - heute</td>
<td><strong>Vienna International School</strong> (freier Dienstnehmer)</td>
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<tr>
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<td>Mitarbeiterin bei der After School Activity „Playing, Balancing, Climbing and More“ für 8-10-jährige SchülerInnen</td>
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<tr>
<td>08/2011 - 08/2011</td>
<td><strong>MA 38 Lebensmitteluntersuchungsanstalt Wien</strong></td>
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<td>Ferialpraktikantin Labor: Organoleptik, Bakteriologie, Histologie</td>
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<td>09/2010 - 07/2011</td>
<td><strong>Volkshochschule Donaustadt</strong> (freier Dienstnehmer)</td>
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<td>07/2008 – 08/2008</td>
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<td><strong>M&amp;P Werbeagentur GmbH Wien</strong> (freier Dienstnehmer):</td>
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<tr>
<td></td>
<td>Promoterin</td>
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<tr>
<td>12/2007 – 02/2008</td>
<td><strong>Anker Brot</strong> Wien (geringfügig): Verkäuferin</td>
</tr>
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</table>
Universitätsausbildung und Schulbildung

03/2011 - heute
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Ernährungswissenschaften: Master „Public Health Nutrition“
Masterarbeit Thema: „Salt taste detection thresholds and pleasantness of test meals varying in salt content in obese persons“

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HTBLVA Spengergasse Wien (EDV und Organisation)
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Sprachen
- Deutsch (Muttersprache)
- Min-Dialekt (2. Muttersprache)
- Englisch (verhandlungssicher)
- Chinesisch (Grundkenntnisse)

EDV-Kenntnisse
Microsoft Office (sehr gut), Adobe Photoshop (sehr gut), HTML, CSS (gut), Webdesign (gut), Adobe Illustrator (gut), TYPO3 (Grundkenntnisse), SPSS (Grundkenntnisse)

Zusatzqualifikationen
Karate-Übungsleiter Ausbildung, Karate Schwarzgurtin (1. Dan)

Interessen
Kyokushin Karate, Klettern, Laufen, Snowboarden, Kochen