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Neural representations of food:
Disentangling the unprocessed and processed dimension

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OVERVIEW

Food is fuel for life. Our feeding behaviors are guided by both homeostatic and hedonic (or reward-based) mechanisms. By simply inspecting visually presented food stimuli, our brain extracts information such as edibility or caloric content, as described by the results of the meta-analysis. However, whether such ability extends to the discrimination between unprocessed and processed foods is to date unknown. Therefore, the aim of the present thesis is to understand whether this particular dimension, that has been hypothesized to have a central role in human evolution (*Cooking hypothesis),* has a brain signature and how it affects food preferences and choices. All these aspects are introduced in Chapter 1 of my thesis while in the following ones (Chapters 2-4) I will report original studies in which I used different techniques.

In Study 1, explicit and implicit evaluations towards foods have been investigated using explicit ratings and the Implicit Association Test (IAT), in order to explore whether evaluations differed based on the food type (unprocessed vs processed) (Chapter 2). The results of Study 1 showed that both at the explicit and implicit level normal-weight participants held different evaluations towards the stimuli depending on the food type. Also, participants' hunger level, BMI and gender were found to modulate participants' evaluations, but only at the explicit level. Interestingly, a strong influence of participants' dietary habits was found at the implicit level.

Using electroencephalography (EEG), in Study 2 I aimed at investigating whether the difference between unprocessed and processed foods had a detectable neural signature and whether the brain rapidly discriminates between these food types as an adaptive behavior (Chapter 3).

The spatio-temporal dynamics of the distinction between unprocessed and processed foods in normal-weight individuals showed that as early as 130 ms post-stimulus onset differences in amplitude emerged. Other within-category discriminations involving food stimuli

(i.e. caloric content), as well as other biologically relevant stimuli such as faces or animals, have been observed within this time window. This study is the first to show distinct brain responses to unprocessed and processed foods in a simple food vs non-food categorization task.

In Study 3 I used functional magnetic resonance imaging (fMRI) with the aim of disentangling the brain responses to different foods in the regions which greatly respond to foods compared to other non-edible objects (Chapter 4). Moreover, the results show how different brain regions responded to unprocessed and processed foods while normal-weight individuals were performing a simple one-back task.

In final chapter I discussed the main findings obtained in my studies in the light of the extant literature, with particular emphasis on the processed-unprocessed dimension (Chapter 5).

CHAPTER1

GENERAL INTRODUCTION

1.1 Basic principles in feeding behaviors

Food is necessary for survival of all species. In the animal kingdom, there are specialist or monophage species that eat only a specific food (Rozin, 1976), such as the koala bear that eats and survives depending only on eucalyptus leaves. On the other hand, generalist or omnivore species, among which humans, can potentially eat a huge variety of foods. Choosing what to eat generates what Rozin defined the *omnivore's dilemma* especially in western countries where the availability of palatable foods is abundant and food cues are omnipresent.

In turn, the abundance of food and an excessive caloric intake have led to an epidemic increase in obesity prevalence (World Health Organization, 2014), the worldwide prevalence of obesity doubled between 1980 and 2014, with 13% of world's adult population was obese in 2014 (more than 1.9 billion adults, see figure A1.1 in the Appendix). The World Health Organization (WHO) defined obesity as abnormal fat accumulation that may impair health. In adults, a common way to classify overweight and obese individuals is through the body mass index (BMI), defined as a person's weight in kilograms divided by the square of its height in meters (kg/m^2). The BMI is commonly used to define normal weight (between 18.5-25), overweight (>25) and obese (>30) individuals and it is the most useful population-level measure since it is equal for females and males and for all ages of adults (WHO website fact sheet, section updated June 2016). However, this measure does not take in account different degrees of fatness between individuals. Recently, measures such as the waist-to-hip ratio have been considered as more reliable measures compared to the BMI (WHO report 2013).

The WHO pointed out that the main cause of obesity nowadays is the imbalance between ingested calories and the expended calories, globally an increased intake of highly fat foods and an increased sedentary lifestyle and physical inactivity. Cognitive neuroscience can contribute towards reducing the health consequences associated with obesity such as hypertension, strokes, osteoarthritis, diabetes, endocrine problems, depression (World Health Organization 2000, Wyatt et al., 2006), and the associated increased medical costs (Wyatt et al., 2006), by explaining how the brain in normal-weight individuals processes information regarding foods as well as by identifying the mechanisms that drive such an increase in highly fat foods consumption.

Food intake is a daily necessity that is controlled by the *homeostatic* and the *hedonic* (or reward-based) mechanisms (Lutter et al., 2009; Berthoud 2011; Alonso-Alonso et al., 2015) that do not function independently (De Silva et al., 2012). On one hand, the *homeostatic* system primarily regulates energy balance: hypothetically, caloric input and caloric expense should be balanced in order to maintain a normal weight, the motivation (drive) to eat is heightened after deficiency of energy stores, this mechanism is under the control of hormones that carry information about energetic levels in the periphery to the brain acting primarily on the hypothalamus (Malik et al., 2008). Leptin and ghrelin represent two of the most important peripheral hormones (Lutter et al., 2009). Briefly, ghrelin (which is a stomach-derived peptide; Figure 1.1 for a simple schematics) administration in animals causes an increase in both food intake and adiposity (Nakazato et al., 2001), in humans it has been found that ghrelin plasma levels trigger hunger and promote the initiation of meals (Cummings et al., 2001). Malik et al. (2008) administered ghrelin during an fMRI study in which participants passively viewed pictures of foods and non-foods and found that receiving ghrelin increased the responsiveness to pictures of food in the following areas: amygdala, orbitofrontal cortex (OFC), insula, visual cortex and striatum. Whereas, the adipose tissue hormone leptin concentration circulates in proportion of fat stores (Lutter et al., 2009, Morton et al., 2011). High levels of leptin are associated with food

intake suppression (Krugel et al., 2003) and leptin administration reduces food consumption and body weight (Morton et al., 2011), whereas, an impairment or reduction in neuronal leptin signaling can cause hyperphagia and weight gain (Scarpace & Zhang, 2009). In humans, fMRI data from two patients with congenital deficiency in leptin has been reported (Faroqui et al., 2007), these patients displayed enhanced striatal activity (nucleus accumbens-caudate and putamen-globus pallidus regions) in response to food images, this enhanced activity was normalized after a 7 days therapy of leptin replacement. Additionally, the pancreas secretes the hormone insulin which as leptin circulates in the plasma based on fat levels and acts on the brain reducing food intake (Morton et al., 2007).

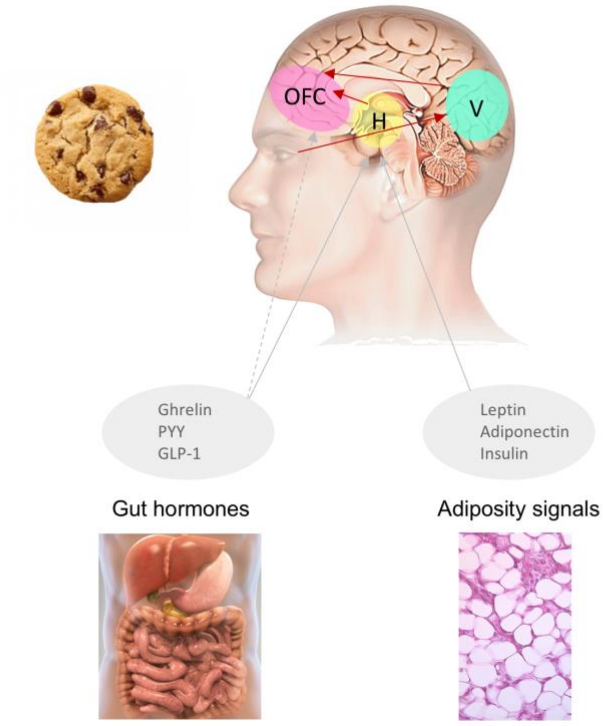


Figure 1.1 Figure readapted from De Silva et al (2012). Highly simplified schematic. Peripheral hormones (produced in the gut and adipose tissue) signal the nutritional status to the hypothalamus (H). The orbitofrontal cortex (OFC) has a central role in the reward encoding network. Gut hormones (such as PYY an GLP-1) attenuate OFC activity. Conversely, ghrelin upregulates OFC activity. External visual food cues processed via the early visual regions (V) modulate OFC activity.

On the other hand, there is the *hedonic* or reward-based contribution to food intake, which induces eating even in the absence of a necessity of energetic intake, especially the consumption of highly palatable foods. The mesocorticolimbic pathway mediates the experience of reward (Alonso-Alonso et al., 2015; Lutter et al., 2009) towards stimuli such as drugs, alcohol, money, smiling faces and foods (Berridge et al., 2009). The reinforcing effects of such stimuli in particular drugs and palatable foods depend on the increase of the midbrain produced neurotransmitter dopamine in the mesocorticolimbic system (Alonso-Alonso et al., 2015). Dopamine has a central role in the nonhomeostatic mechanisms of food consumption since dopaminergic neurons do not code for the stimulus sensory characteristics but for searching behaviors (motivational saliency, Berridge et al., 2009).

The mesocortical circuit, is constituted by the midbrain ventral tegmental area's (VTA) dopaminergic neurons which project in the limbic areas in particular the nucleus accumbens (NA) and prefrontal cortex (PFC; which has descending projections to both to the NA and VTA).

Berridge et al. (2009) analyzed the brain systems associated with reward and dissected reward in 'liking' and 'wanting' components. To measure liking, Berridge et al. (2009) investigated objective reactions (affective facial expressions) to sweet taste rewards both in infants and rodents. Sweets elicited lip licking, rhythmic tongue protrusions whereas bitter tastes elicited 'disliking' expressions (e.g. gapes). Distinct brain substrates for liking and wanting have been reported in this seminal paper, however for the purpose of my thesis it is important to remind that many factors can modulate the rewarding value of a stimulus, especially food, with internal and external factors driving the single food choice we make on a daily basis. Moreover, it is relevant to remind here that the different impact of wanting and liking should be taken in account when investigating food choices.

The neuro-computational basis of decision-making and the homeostatic regulators of feeding have been investigated by Antonio Rangel in a work published in 2013. He argued that

feeding decisions can be triggered by either an internal state (hunger) or an external cue (sight or odor of food), that for any decision (i.e. eating an apple or a piece of cake) a value must be assigned to each option, and that eventually the outcome of ingesting the food must be evaluated (Rangel & Hare, 2010; Plassmann et al., 2010; Hare et al., 2011). Learning is involved in the update of the representation of each option to make future optimal choices. Despite the view of two opposed systems (hedonic vs homeostatic) regulating feeding behaviors, Rangel engages with the idea that the homeostatic systems operate by modulating the decision-making circuitry (Rangel, 2013). Compared to other decisions, food-related choices are made with a high frequency and each decision is driven by the attributes and assigned value to that specific situation, combined with emotional responses, memory retrieval and hedonic evaluations which are based on previous experience. In the following paragraph, I will describe how the brain responds to the sight of food compared to other objects encountered in everyday life.

1.2 "The first taste is with the eyes": brain responses to visually presented food cues

Despite food is a multisensory stimulus in nature, this thesis will focus on visually presented food stimuli. For instance, in a meta-analysis Huerta et al. (2014) tried to identify which sensory modality (vision, taste and smell) produced the most robust results in studies in which food stimuli were presented. The significant clusters of the different food-cue deliveries are displayed in the Appendix (Figure A1.2). The only region active through all three stimuli-deliveries was the anterior insula (BA 13, in the region of the primary gustatory cortex, see also Simmons et al., 2005) confirming the old saying that *"the first taste is with the eyes"*. Importantly this meta-analysis showed how neural responses to food images led to the most robustly convergent activations. The peak in ALE value was in the region of the fusiform gyrus (FG), this finding has been interpreted as a "pop-out" effect of food stimuli, this was explained given the

importance for our evolution to find edible food in the environment in a rapid and reliable way (Huerta et al., 2014; same finding in the FG for other relevant stimuli, such as familiar faces). This is consistent with the knowledge that differently from early aquatic species that rely on taste or other species relying on olfaction, most primates rely on vision for an efficient food discovery (Linne et al., 2002).

Indeed, edibility is a crucial problem that our brain should be able to solve in a rapid and reliable manner. Using magnetoencephalography (MEG), Tsourides et al. (2016) showed how the brain discriminates edible and inedible stimuli as early as 85 ms post-stimulus onset. Several neuroimaging studies unveiled the network of brain regions active while viewing foods compared to other objects (Killgore et al., 2003; Uher et al., 2006; Frank et al., 2010; Garcia-Garcia et al., 2013; Simmons et al., 2016).

1.2.1 Food vs non-food visual stimuli: an ALE meta-analysis

In order to identify the regions active during the presentation of foods compared to non-foods, I have conducted a coordinate-based meta-analysis on the published functional magnetic resonance imaging (fMRI) studies with visually presented food stimuli, applying the activation likelihood estimation (ALE) method (for a review Eickhoff et al., 2012). Most of the papers were already in the Brainmap database named Sleuth (http://brainmap.org/sleuth/) as two meta-analyses of food studies had already been performed using the ALE method. However, additional studies have been incorporated by examining the reference lists of the retrieved articles and through online electronic databases, including Pubmed (https://www.ncbi.nlm.nih.gov/pubmed). To be included papers had to be published in peer reviewed journals and contain whole-brain imaging analysis (that is, studies that published only results from region-of-interest analysis have been excluded).

Sleuth application allows to select for each paper the contrasts of interest. I therefore selected the published fMRI studies on food perception that reported the contrast between foods (both containing high or low caloric content) and non-foods (e.g. tools, flowers, locations). Fourteen papers containing 19 experiments were selected, including data from 205 subjects and 156 locations (Table 1). Ginger ALE Brainmap software 2.3.6 version has been used to analyze the data (http://brainmap.org/ale/), based on the likelihood estimation (ALE) as implemented by Turkeltaub et al. (2002). The convergence of fMRI location results is calculated by statistical contrasts to an ALE null-distribution map, all foci for a given experiment are modelled as Gaussian probability distributions (Eickoff, 2012), in the reported results the Turkeltaub non-additive ALE method (Turkeltaub et al., 2012) has been used, with an additional full width at half maximum (FWHM) of 4 mm, false discovery rate (FDR) correction for multiple comparisons and a $p < .05$ threshold with a minimum volume cluster size of 100 mm^3, all of the reported coordinates (Table 2) are in MNI space. The overlay of results was then superimposed on the Colin27_T1_seg_MNI template available from brainmap.org/ale/ using Mricron (http://people.cas.sc.edu/rorden/mricron/install.html). The coordinates of the clusters with significant ALE values have been inserted in Neurosynth (http://www.neurosynth.org/locations) to review the regions associated in the literature with each cluster.

Table 1 Studies included in the food vs non-food ALE meta-analysis

Author (year)	*Food Stimuli*	*Hours fasted*	*N*	*Foci*	*Non-food stimuli*
1. *Killgore et al. (2003)*	*Low-cal foods (whole grain cereals, Fruits and vegetables); high-cal foods (cheesburgers, hot-dogs, ice-cream, cookies)*	*>1.5*	13	18	*Non-edible objects (shrubs, rocks, trees, flowers); non-edible food related utensils (dishes, forks, spoons)*
2. *Holsen et al. (2005)*	*Food stimuli used in Labar et al. (2001)*	*4*	*9*	*7*	*Animals*
3. *Killgore et al. (2005)*	*Low-cal foods (whole grain cereals, fruits and vegetables); high-cal foods (cheesburgers, hot-dogs, ice-cream, cookies)*	*>1*	*21*	*12*	*Non-edible objects (shrubs, rocks, trees, flowers); non-edible food related utensils (dishes, forks, spoons)*
4. *Simmons et al. (2005)*	*High-calorie foods (hamburgers or cookies)*	*not reported*	*9*	*6*	*Locations, buildings (mall, house, school)*
5. *Beaver et al., (2006)*	*Appetizing foods (chocolate cake), bland foods (uncooked rice)*	*>2*	*12*	*16*	*Objects (videocassettes, iron)*
6. *Santel et al. (2006)*	*High-caloric food*	*>12*	*10*	*3*	*Objects (tools, makeup, pencils)*
7. *Uher et al. (2006)*	*Savory and sweet foods*	*>3*	*18*	*5*	*Objects (brushes, car, flowers)*
8. *Cornier et al. (2007)*	*High hedonic foods (chocolate cake, cookies, eggs and bacon); low hedonic (fruit, bagels, bread)*	*overnight*	*25*	*7*	*Animals, trees, furniture, buildings*
9. *Rothemund et al. (2007)*	*High-cal (hamburgers); low-cal (fruits)*	*>1.5*	*13*	*1*	*Rocks, flowers*
10. *Calder et al. (2007)*	*Appetizing foods (chocolate cake, ice-cream sundae); bland foods (uncooked rice, potatoes)*	*not reported*	*12*	*2*	*Non-foods (videocassets, iron)*
11. *Fuhrer et al. (2008)*	*Ready-to-eat foods (salad, meat with vegetables)*	*14*	*12*	*12*	*Non-related to food objects (screwdriver)*
12. *Malik et al. (2008)*	*Appetizing foods (cherry pie, hamburger, steak, chocolate cheescake)*	*3*	*20*	*34*	*Scenery, landscapes*
13. *Schienle et al. (2009)*	*High-cal foods (French-fries, ice-cream, cake, chips)*	*>12*	*19*	*12*	*Household articles*
14. *Frank et al., (2010)*	*High-cal and low.-cal foods (salad, meat, soup, desserts)*	*14*	*12*	*21*	*Objects (chair, umbrella , toy, money, car)*

Table 2 Locations (MNI) of clusters with significant ALE values for the contrast of food vs non-food

Cluster	*Label*	*BA*	*x*	*y*	*z*	*Volume* (mm^3)	*ALE value* ($\times 10^{-3}$)
1	R Fusiform Gyrus	19	46	-68	-10	4048	11.8
	R Cerebellum	-	38	-64	-16		
2	L Orbitofrontal Cortex	47	-30	28	-14	1124	9.6
3	L Hippocampus	-	-28	-24	-10	1016	9.4
4	L Inferior Occipital Gyrus	17	-16	-98	-2	472	8.6
5	L Fusiform Gyrus	37	-48	-68	-10	440	7.9
6	R Insula	13	38	-6	10	432	8.4
7	R Precuneus	7	24	-64	56	368	8.0
8	R Orbitofrontal Cortex	47	26	26	-16	344	7.8
9	R Middle Temporal Gyrus	-	-58	-32	2	272	7.7
10	L Insula	13	-36	16	8	168	7.5
11	R Fusiform Gyrus	19	36	-80	-8	104	7.2

The contrast food vs non-food stimuli produced 11 significant ALE clusters (Table 2), summing a volume of 8888 mm^3 and a maximum ALE score of 0.012.

The most robust convergence of activation was found bilaterally in the fusiform gyrus (FG, BA19). Moreover, activations were observed bilaterally in the orbitofrontal cortex (OFC, BA 47), insula (BA 13), the right precuneus (BA 7) and part of the left inferior occipital gyrus in the primary visual cortex (BA 17) and hippocampus (HC) in the left hemisphere.

GingerALE's output files allowed to identify the papers that contribute to each cluster of activation as we will report in the following. As reported by Huerta et al. (2014) the fact that the most robust

convergence is found in higher order visual areas and secondly in rewarding areas is of main interest of this thesis.

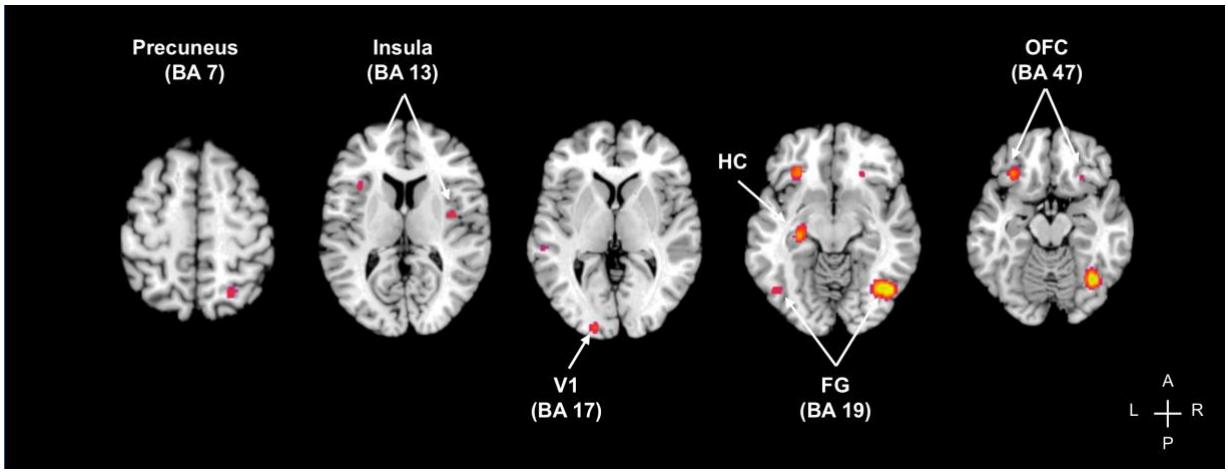


Figure 1.3 Results of the ALE meta-analysis showing clusters with significant ALE maxima ($p < .05$, FDR corrected, cluster size> 100 mm^3) for the contrast food > non-food, overlaid on the MNI Colin template brain.

The regions found in this meta-analysis are in line with the results reported by Huerta et al. (2014). However, while only high-calorie foods were considered in Huerta's paper, I have also considered the contrasts with both, low- and high-calorie foods.

To conclude, foods greatly activate occipital structures in particular the primary visual cortex (V1, BA17; Santel et al., 2006; Fuhrer et al, 2008; Malik et al., 2008; Simmons et al., 2016) and the fusiform gyrus (Killgore et al., 2003; Simmons et al., 2005; Uher et al., 2006; Cornier et al., 2007), parietal areas such as the postcentral gyrus (Killgore et al., 2003) and the precuneus (Cornier et al., 2007; Malik et al., 2008), prefrontal regions such as the orbitofrontal cortex (Killgore et al., 2005; Simmons et al., 2005; Malik et al., 2008; Frank et al., 2010), and limbic structures such as the insula (Simmons et al., 2005; Uher et al., 2006; Malik et al., 2008) and amygdala (Killgore et al., 2003; Simmons et al., 2016). For coronal and sagittal images of the

results see Figure A1.3 in the Appendix (see Simmons et al., 2013; Avery et al., 2015 for insula's functional organization).

Which features of the visually presented foods (e.g. basic features such as shape or color, caloric content, healthiness, level of processing, rewarding properties) are coded within each region and how these features modulate brain activations remain to date unclear.

1.3 Modulations of the food network

Not only is our brain able to rapidly discriminate food from other stimuli, but it is also capable of rapid and efficient within-category discriminations. Using both EEG and fMRI it has been found that a distinction between high and low-energy dense foods occurs as early as 160 ms after stimulus onset (Toepel et al., 2009 and Meule et al., 2013). This aspect will be extensively described in Chapter 3). In addition, greater activation of prefrontal, hypothalamic and striatal responses when viewing high-energy foods compared to low-energy ones has been reported (Killgore *et al*., 2003; Beaver *et al*., 2006).

1.3.1 High calorie vs low calorie foods: an ALE meta-analysis

In order to test for the effect of caloric content on food stimuli perception I have performed a second meta-analysis of the existent literature. Nine papers reporting 11 experiments were selected, with data from 136 subjects and 125 locations (Table 3). The same ALE method described in paragraph 1.2. was used. In the following results, the Turkeltaub non-additive ALE method (Turkeltaub et al., 2012) has been used, with an additional full width at half maximum (FWHM) of 4 mm, false discovery rate (FDR) correction for multiple comparisons with a $p < .05$ threshold did not produce any significant cluster, probably due to the small number of papers and locations. Uncorrected results are reported with $p < .001$ threshold and a minimum volume cluster

size of 100 mm^3, all of the reported coordinates (Table 4) are in MNI space. The overlay of results was then superimposed on the Colin27_T1_seg_MNI template available from brainmap.org/ale/ using Mricron (http://people.cas.sc.edu/rorden/mricron/index.html).

Table 3 Studies included in the high-cal (appetizing) vs low-cal (bland) food ALE meta-analysis

Author (year)	*Food Stimuli: high and low caloric content*	*Hours fasted*	*N*	*Foci*
1. Killgore et al. (2003)	*High-cal foods (cheesburgers, hot-dogs, ice-cream, cookies); low-cal foods (whole grain cereals, fruits and vegetables)*	*>1.5*	*13*	*7*
2. Killgore et al. (2005)	*High-cal foods (cheesburgers, hot-dogs, ice-cream, cookies); low-cal foods (whole grain cereals, fruits and vegetables)*	*>1*	*8*	*5*
3. Beaver et al. (2006)	*Appetizing foods (chocolate cake), bland foods (uncooked rice)*	*>2*	*12*	*16*
4. Cornier et al. (2007)	*High hedonic foods (chocolate cake, cookies, eggs and bacon); low hedonic (fruit, bagels, bread)*	*overnight*	*25*	*7*
5. Rothemund et al. (2007)	*High-cal (hamburgers); low-cal (fruits)*	*>1.5*	*13*	*12*
6. Calder et al. (2007)	*Appetizing foods (chocolate cake, ice-cream sundae); bland foods (uncooked rice, potatoes)*	*not reported*	*12*	*2*
7. Goldstone et al. (2009)	*High-cal (burgers, cakes and chocolate); low-cal (salads, fish, fruits, vegetables)*	*overnight*	*20*	*38*
8. Passamonti et al. (2009)	*Appetizing foods (chocolate cake, ice-cream); bland foods (rice, potatoes)*	*>2*	*21*	*13*
9. Frank et al. (2010)	*High-cal and low.-cal foods (salad, meat, soup, desserts)*	*14*	*12*	*9*

The coordinates of the clusters with significant ALE values have been inserted in Neurosynth (http://www.neurosynth.org/locations) to review the regions associated in the literature with each cluster. This analysis produced 14 significant ALE clusters (Table 4), summing a volume of 8488 mm^3 and a maximum ALE score of 0.007. The following clusters (Figure 1.3) responded greatly to high-calorie (appetizing) compared to low-calorie (bland) stimuli: bilateral

cerebellum, right hypothalamus, the right precuneus (BA 7), right middle frontal gyrus (MFG, BA 6), the right portion of the occipital cortex in correspondence of the cuneus, left medial prefrontal cortex (mPFC, BA 9), the right anterior cingulate (AC, BA32), the left thalamus, bilateral hippocampus, right orbitofrontal cortex (OFC, BA 47) and the left temporal pole (BA38).

Table 4 Locations (MNI) of clusters with significant ALE values for the contrast of food vs non-food (uncorr p< .001)

Cluster	*Label*	*BA*	*x*	*y*	*z*	*Volume* (mm^3)	*ALE value* ($\times 10^{-3}$)
1	R Hypothalamus	25	6	2	-10	1520	6.71
2	R Orbitofrontal Cortex	47	26	34	-12	1344	6.02
3	L Temporal Pole	38	-32	12	-20	1176	5.50
4	L Hippocampus	-	-26	-26	-8	1144	6.15
5	R Hippocampus	-	24	-32	-6	768	5.68
6	L Cerebellum	-	-36	-62	-28	384	5.03
7	L Thalamus	-	-2	-16	6	368	4.75
8	R Cerebellum	-	36	-62	-18	352	5.10
9	R Middle Frontal Gyrus	6	30	-4	48	352	5.07
10	R Precuneus	7	22	-66	54	352	5.07
11	R Cerebellum	-	30	-40	-32	296	4.84
12	L Medial Prefrontal Cortex	9	-26	50	32	200	4.66
13	R Anterior Cingulate	32	14	38	14	120	4.27
14	R Cuneus	18	18	-82	26	112	4.40

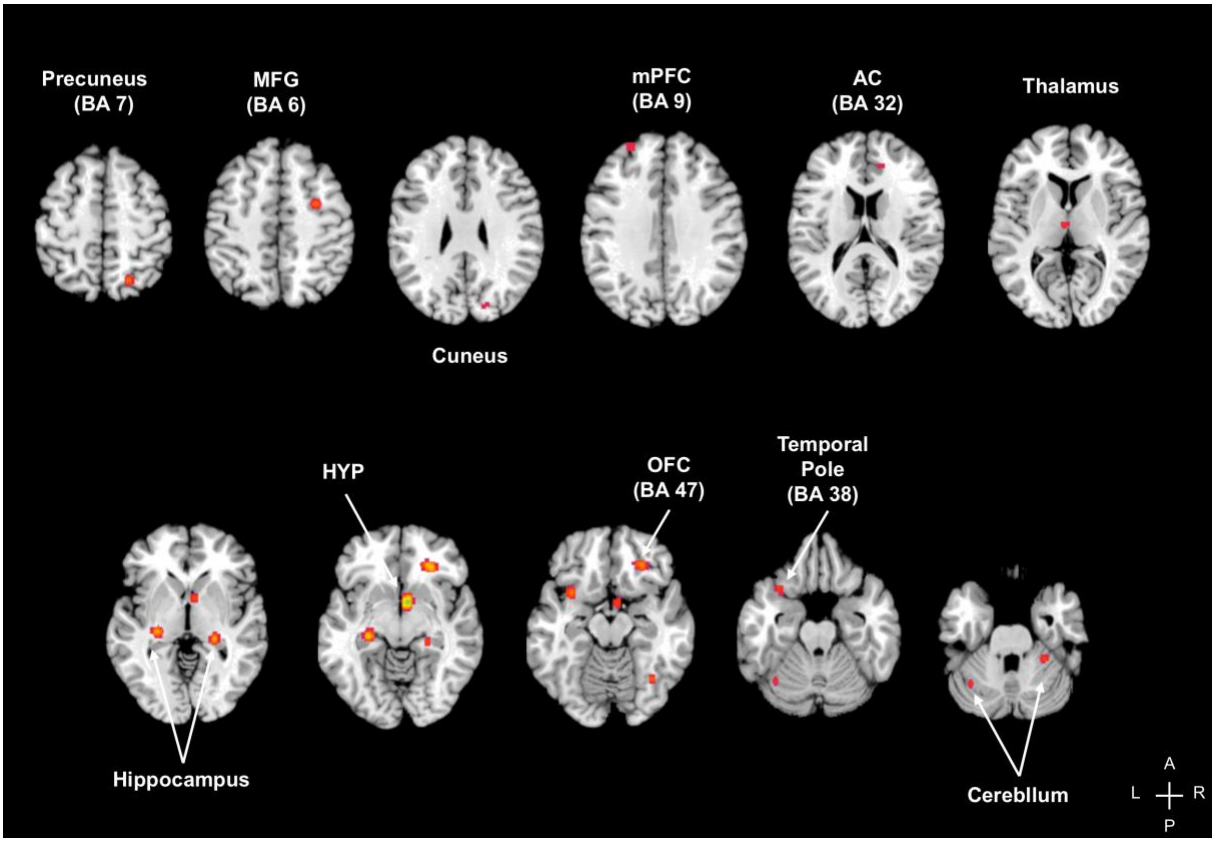


Figure 1.4 Results of the ALE meta-analysis showing clusters with significant ALE maxima ($p < .001$, cluster size> 100 mm^3) for the contrast high-cal (appetizing) > low-cal (bland), overlaid on the MNI Colin template brain.

Appetizing and high-calorie food stimuli activated regions involved in reward such as mPFC (Killgore et al., 2003; Goldstone et al., 2009), AC (Frank et al., 2010) and OFC (Rothemund et al., 2007; Goldstone et al., 2009; Frank et al., 2010) when compared to bland foods with low-caloric content (see Figure A1.4 in the Appendix for coronal and sagittal images). Plassmann, O'Doherty and Rangel (2010) dissociated saliency and reward values of food stimuli and found that saliency is mainly encoded in the OFC. Lateral OFC activity may reflect the expected pleasantness of the food depicted in the image (Van der Laan et al., 2011). Activity in OFC and AC in response to high-calorie stimuli has been found to be inversely related to the BMI (Killgore & Yurgelun-Tood, 2005). The primary and associative visual areas were greatly active for appetizing

foods (Beaver et al., 2006; Calder et al., 2007, Simmons et al., 2016). Importantly, in most of the studies, food stimuli were matched for low-level features, hence greater activity in visual regions has been explained with appetizing stimuli being more salient (Killgore and Yurgelun-Todd, 2007, Stingl et al., 2010; Van der Laan et al., 2011). The role of the hippocampus in food intake has been recently reviewed (Stevenson & Francis, 2017), as appetizing foods might greatly activate this region since they induce episodic recollection (greater enjoyment associated to these hedonic stimuli).

Since the results reported in paragraph 1.3. are uncorrected the same meta-analysis has been performed with a more conservative threshold of $p < .0001$ in order to check for the core regions that are consistently reported in previous literature when contrasting high-calorie foods and low-calorie foods using fMRI. This analysis produced 5 significant ALE clusters (Table 5), summing a volume of 1744 mm^3 and a maximum ALE score of 0.007.

Table 5. Summary of ALE results for the contrast high- vs low-calorie foods (uncorr $p < .0001$)

Cluster	*Label*	*BA*	*x*	*y*	*z*	*Volume* (mm^3)	*ALE value* ($\times 10^{-3}$)
1	R Hypothalamus	25	6	2	-10	672	6.71
2	R Orbitofrontal Cortex	47	26	34	-12	384	6.03
3	L Hippocampus	-	-26	-26	-8	336	6.15
4	L Temporal Pole	38	-32	12	-20	176	5.50
5	R Hippocampus	-	24	-32	-6	176	5.68

Greater activity in response to high-calorie (appetizing) vs low-calorie (bland) foods was found in the following regions (Figure 1.5): right hypothalamus, right OFC (BA47), bilateral hippocampus and left temporal pole (BA38). The role of these regions has been described above,

however it is of great interest that across all of the studies these regions seem to be the core active network in response to appetizing foods.

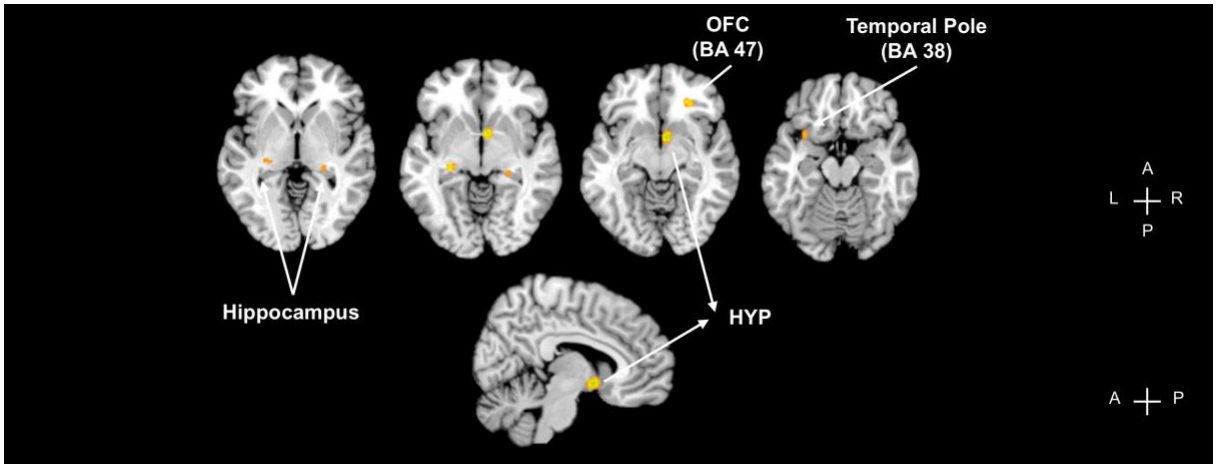


Figure 1.5 Results of the ALE meta-analysis showing clusters with significant ALE maxima ($p < .0001$, cluster size> 100 mm^3) for the contrast high-cal (appetizing) > low-cal (bland), overlaid on the MNI Colin template brain.

Caloric content of foods is not the only intrinsic factor that modulates brain activations. Healthiness of foods has been found to influence the activity in lateral and medial prefrontal cortex (lPFC and mPFC; Hare et al., 2011) while participants examine food stimuli. In everyday life, we hardly just passively view foods we make decisions and choices, Hare et al. (2009) showed how participants' internal factors (successful vs unsuccessful controllers) influence food choices when healthiness and taste of the foods are taken into account. Unsuccessful controllers were found to choose foods which were rated tasty and regardless they were unhealthy, during these choices activity in the dorsolateral prefrontal cortex (dlPFC) and the ventromedial prefrontal cortex (vmPFC) had a central role. These regions are involved in the cognitive control/inhibition of behaviors and the assigned value to a given stimulus. The failure to find other areas may depend

on the fact that in the studies included in the meta-analyses the task was passive viewing of food pictures or simple food vs non-food categorization.

Another internal factor that has a huge impact on the activity of the participants' food network is hunger. Many studies reported that activity in the parahippocampal gyrus and the OFC is greater when participants are hungry compared to when they are satiated (Labar et al., 2001; Holsen et al., 2005; Santel et al., 2006; Fuhrer et al., 2008; Van der Laan et al., 2011). A phenomenon named *sensory-specific satiety* has been reported in literature (Rolls et al., 1981), when participants are fed to satiety to a specific food there is a decrease in BOLD signal to that food in the OFC (result found also in the OFC neurons of macaques, Critchley & Rolls, 1996) but not in the insula and not in OFC in response to other foods (Kringelbach et al., 2003; for a review Rolls et al., 2015). This decrease in brain activity mirrors the decrease in the motivational/hedonic value assigned to the food (di Pellegrino et al., 2010). Participants' hunger level should be controlled during food studies. Moreover, personality and behavioral traits such as reward sensitivity, dietary restraint, external eating behavior represent modulating factors (Beaver et al., 2006; Coletta et al., 2009; Passamonti et al., 2009). Specific questionnaires for each of these traits have been developed and should be used to assess participants during experiments with food (i.e. *Restraint Scale*; Herman and Polivy, 1980, EAT-26, Garner, 1989).

Indeed, the impact of obesity on the food network is well documented in literature (Brooks et al., 2013 and Kennedy et al., 2014 for meta-analysis of fMRI studies, Garcia-Garcia et al., 2013 and Garcia-Garcia et al., 2014 for reviews) and also of the impact of other eating disorders (e.g. *anorexia* and *bulimia nervosa*, Uher et al., 2004; Garcia-Garcia et al., 2013 for a review, Foerde et al., 2015; *binge eating disorder*, Schienle et al., 2008). However, the main focus of this thesis will be on normal-weight individuals (BMI ranging between 18.5 and 25).

1.4 Catching Fire: The Cooking Hypothesis

A particular aspect of food that has received little attention to date is the level of transformation (Rumiati & Foroni, 2016; Foroni & Rumiati, 2017), despite humans ubiquitously transform food items through cooking, aggregation or preservation procedures (Wrangham et al., 2003). Cooking food is a unique and universal behavior which has been claimed to have played a fundamental role in human evolution (Wrangham, 1999). Compared to chimpanzees, our closest living relatives, a reduction in our body structures designated for mastication (e.g. reduced molar dentition, more gracile mandibles and smaller chewing muscles, Carmody et al. 2009b) and digestion suggest that we adapted to a high-quality diet (Carmody et al., 2009b). Great-apes spend on average 4-7 hours per day chewing, since the toughness of foods is reduced both chewing (fewer chewing cycles and shorter time of the food in the mouth) and digestion benefit from cooking, the modifications in body structures and eating behaviors allowed our ancestors to spend more time in activities that were not hunting or eating/chewing.

One of the main effects of cooking food is represented by the increased net energy value (Carmody et al., 2009a) This effect is caused by several factors such as the gross caloric value, digestibility, diet-induced thermogenesis (Carmody et al., 2009b), along with the reduction in toxins in cooked foods.

Cooked starch (e.g. raw potato vs cooked potato) and proteins have an increased digestibility. Raw starches which are not digested in the small intestine (resistant starches) are fermented by microbes in the caecum of the intestine and in the colon and return only 50% of metabolizable energy of the starch (Carmody et al., 2009b), whereas the gain of energy when eating them cooked is quantifiable between 12-35%.

The effects of raw-food diets have been investigated by Koebnick et al. (1999) who reported data of 513 (297 females) raw foodists and studied the impact of this diet on body weight and other body functions. In a population of German individuals, the authors divided their participants

based on the total amount of raw food present in their diets (70, 80, 90, 95 and 100% of raw food), participants were further divided in meat eaters, vegetarians and vegans. Participants were administered with various questionnaires regarding their eating behaviors and their health situation. The mean percentage of raw food consumed by participants was high (M= 91%, s.e. = 0.4%), with participants being on average were on a raw food diet for 3.7 years and only a few participants integrating their diets with mineral or vitamin supplements (7%). An unexpected result was that 14.7% of the males and 25% of females resulted being underweight based on their BMI and 2.6% of males and 5.7% of females suffered of chronic energy deficiency (CED; Ferro-Luzzi et al., 1992). Authors calculated the weight loss from the beginning of the raw diet and males on average lost 9.9 kg and females 12 kg. Diet group differences showed no effects, with vegans having the same percentage of weight loss compared to vegetarians and meat eaters. In female participants, the authors investigated eventual menstrual issues and found that About 70% of females reported a change in menstrual cycle since they began the raw food diet, and 23% of the childbearing age reported a total absence of menstruation (*amenorrhea*). These energy and reproductive function deficiencies in raw-foodists suggested that caloric gains allocated by cooking might be necessary for normal biological functions.

The cooking hypothesis (Wrangham et al., 1999) takes together all of the above-mentioned accounts and formulated the hypothesis that early hominids might have possessed a preference for cooked items prior fire control. To test this hypothesis animal models have been chosen.

Carmody et al. (2011) tried to isolate the consequences of thermal (heat/cooking) processing compared to nonthermal processing (pounding). To do so, she compared the effects of two types of *ad libitum* diets on the *Mus musculus* mouse, this species was chosen since in natural context it ingests both meat and starch rich foods (i.e. tubers). In a within-subjects study design mice were fed in a counterbalanced order with either sweet potato or lean beef in four diets (raw and whole,

raw and pounded, cooked and whole and cooked and pounded). The diets were ad libitum for 4 days with 6 days washout periods between each diet. The dependent variable was represented by changes in the body mass, moreover preference tests were administered to the mice. Preference tests consisted in fasted mice in a naïve (prior to exposure to any tuber) and experienced mouse using first bite and total intake (in grams) as measures.

Results on tuber diets showed that mice on a cooked diet maintained their weight, while mice on a raw diet (both whole and pounded) lost weight. No difference in activity (wheel running) was found across diets. A greater food intake was found for thermal processing diets contributing to the higher energy intake. In the preference test, naïve mice showed a preference for pounded diets, after experiencing the four conditions the same experienced mice selected the cooked diets in all of the cases (17 out of 17 mice). Same effect was found for total intake.

Results on meat diets are less straightforward since pure meat diets were not expected to be beneficial for any omnivore species, and as expected mice lost body mass on all conditions since a diet based only on proteins induces a negative energy balance. As expected, mice lost weight on all diets, however an absolute energy gain on cooked diets was observed. Preference data on total intake, mirrored the results with tubers.

Wobber et al. (2008) investigated whether great apes showed an inherent preference for either raw or cooked foods. Chimpanzees' choices in a preference task (the experimenter held one piece of food in each hand) towards cooked and uncooked tubers (carrots, sweet potatoes, white potatoes) were assessed. Chimpanzees in the wild rarely eat raw tubers therefore these stimuli were relatively unfamiliar. Subjects showed a significant preference for the cooked items. Warneken and Rosati (2015) tested 29 chimpanzees in dichotomous choices between raw and cooked potatoes, using again a preference test. Again, chimpanzees significantly chose the cooked slice, supporting the claim that apes have an intrinsic preference for cooked foods. In a second experiment, temporal preferences towards these food categories were assessed in chimpanzees,

in both delay conditions the animal had to choose between receiving one raw/cooked piece immediately, or to wait one minute and receive three pieces of food (this design resembles the typical experiments with humans on *temporal discounting*, Chapman, 1996). Results showed that chimpanzees more often chose the cooked delayed reward, showing a will to pay temporal costs in this condition.

Taken together, these observations have been interpreted as indirect proof that hominids too might have showed a spontaneous preference for cooked food. The extent to which the level of transformation might differentially shape human perception and evaluations towards food has received little attention to date. My thesis tries to fill this gap.

CHAPTER 2

EXPLICIT AND IMPLICIT EVALUATIONS OF FOODS: AN EXPLORATORY STUDY[1]

ABSTRACT

In western societies choosing what to eat can be a demanding task due to the excessive availability of food, ranging from unprocessed to processed food. To make our feeding decisions more complex, our explicit and implicit evaluations may differ from each other. In the present study, we investigated whether individuals assigned different values to unprocessed and processed food depending on explicit (assessed with ratings) or implicit measures (assessed with Implicit Association Test), this is the first study which uses this type of food stimuli with such task. Additionally, we investigated if factors such as dietary habitudes, hunger or body mass index modulate such evaluations and the relationship between implicit and explicit measures. Our findings reveal differences depending on food-type and a critical role of self-control and goal-directed behavioral aspects in food evaluation depending from the type of food.

[1] A version of this chapter is under review in the *Quarterly Journal of Experimental Psychology*: Coricelli C., Foroni F., Osimo S., Rumiati I. R. Doctor 'Natural food' and Mister 'Transformed': How explicit and implicit evaluations depend on food type.

2.1 INTRODUCTION

Choosing what to eat is a demanding task subjected to internal and external factors (see Chapter 1). Moreover, depending on the situational contingencies, explicit or implicit evaluation may strongly guide our choice, and the divergence between the two may vary depending on the characteristics of the food items.

As pointed out in Chapter 1 a particular aspect of food that has received little attention is the level of transformation (Rumiati & Foroni, 2016; Foroni & Rumiati, 2017). This study was designed in order to investigate whether, and to which extent, this dimension might differentially shape our implicit and explicit evaluations towards food has received little attention to date. The present work tries to fill this gap.

Implicit evaluations are by definition fast and not directly accessible to control or awareness, and have been extensively studied in several domains using the *Implicit Association Test* (IAT; Greenwald, McGhee & Schwartz, 1998). The IAT measures the relative strengths of automatic associations between target concepts and attributes (e.g., positive/negative) and has been exploited also to investigate implicit food evaluations with non-conclusive results (e.g., Houben et al., 2010, Czyzewska et al., 2011 for a review). In contrast, explicit evaluations are built up by individuals more slowly by individuals in a deliberate fashion and subject to biasing factors such as self-presentation strategies (e.g., Fazio & Olson, 2003). Explicit evaluations of food can be explored by asking participants, for instance, to rate the valence or pleasantness of food stimuli on a scale, or to forcedly choose between different options. Food explicit evaluations showed high between-study variability, with more positive evaluations of both high- (Maison et al., 2001; Killgore et al., 2003) and low-calorie content foods (Rothemund et al., 2007, Roefs & Jansen, 2002) being reported. A clear explanation to this between-study variability has not been proposed to date. Overall, explicit and implicit evaluations are often reported to be uncorrelated and investigations of their predictive power returned diverging results. Explicit measures (e.g.,

Karpinski & Hilton, 2001) and the IAT (e.g., Richetin et al., 2007) have both been found to predict participants' behavioral choice between snacks and fruits. The absence of a strong correlation between implicit and explicit measures can be explained by the dual attitudes model (Wilson et al., 2000), according to which different evaluations can coexist for a given object (e.g., an automatic implicit positive evaluation and an explicit negative evaluation for French fries). Furthermore, the Reflective-Impulsive Model of behavior regulation by Strack and Deutsch (2004) proposes the existence of two systems (impulsive vs reflective) which generate behavioral decisions based either on knowledge about facts and values or on motivational orientations.

Thus, according to these two models eating behavior can be guided by two processes or systems. While, implicit and automatic processes or the impulsive system, guide spontaneous approach or avoidance tendencies to food, the explicit processes or the reflective system (see e.g., Foroni et al., 2016a; Strack & Deutsch, 2004) are shaped by individual personal goals and other external pressures. A prediction derived from these models is that when control resources are reduced (e.g. by stress, cognitive load, or fatigue) the functioning of the reflective system is limited and impulsive behaviors are increased (Friese et al., 2008; Czyzewska et al., 2011).

The aim of the present study was to investigate whether participants differentially evaluate food depending on its level of transformation (unprocessed vs processed) and on the task (involving explicit vs. implicit processes).

Participants were never informed of the distinction between unprocessed and processed foods, it was always orthogonal to the task at hand.

A positive evaluation towards unprocessed food was hypothesized when participants were asked to explicitly rate foods. This preference is expected to be driven by participants' reflective system (Strack & Deutsch, 2004), their health concerns, knowledge about food quality, and self-presentation strategies (Bicchieri et al., 2006). However, these aspects are not expected to affect

the implicit level governed by the impulsive system, while a higher evaluation towards processed food is expected because of the relevance of its generally higher energy gain (see e.g., Carmody, Weintraub, & Wrangham, 2011).

Moreover, we expect participants' dietary habitudes, assessed through the Restraint Scale (Herman & Polivy, 1980) and their hunger level (Labar et al., 2001; Santel et al., 2006) to modulate their food evaluations, both at the implicit and the explicit level. Restrained eating (or chronic dieting) has been defined as a chronic restriction of food intake aimed in losing or controlling weight alternated with periods of overeating (Herman & Polivy, 1980) therefore often unsuccessful. Low inhibitory control has been reported in restrained eaters (Meule et al., 2011, Bartholdy et al., 2016, Price et al., 2016) together with enhanced reward sensitivity (Finlayson & Dalton 2012). At the neural level, restrained eaters showed a reduced activity in the dorsolateral prefrontal cortex, involved in the top-down control of appetite (Dong et al., 2014) and hyper responsivity in reward-related regions (Burger and Stice, 2011; Coletta et al., 2009; Ely et al., 2014). Therefore, we expect restrained eaters which lack in inhibitory control to show an enhanced preference for processed foods also at the explicit level.

2.2 MATERIALS AND METHODS

2.2.1 Participants

Forty-eight healthy native-Italian speakers (24 females) participated in the experiment. Participants were recruited with an age comprised between 18-30 years (*M*=23.04, *SD*=3.11) and Body Mass Index (BMI; kg/m^2) in the normal range (18.5<BMI<25.0). Participants were not included in the study if: (i) they showed signs of aberrant eating behavior and/or behavioral symptoms (e.g. binge eating, vomiting, use of diuretics or laxatives) commonly associated with risks of eating disorders based on the *Eating Disorder Inventory-3* Symptom Checklist (EDI-3 Symptom Checklist; Garner et al., 1983); (ii) they acknowledged the consumption of neurotropic substances; or (iii) they had dietary restrictions for medical or religious reasons.

2.2.2 Procedure

Upon arrival, participants signed a written informed consent and sat in front of a computer cubicle at 60 cm distance from the monitor. Stimulus presentation and registration of responses was controlled by *Eprime 2.0* version (Psychology Tools Inc., Pittsburgh, USA; www.pstnet.com/eprime). The experiment lasted approximately 1.5 hours and participants received 15 euros for compensation. The study conformed to the Declaration of Helsinki and was approved by SISSA's Ethic Committee.

The study comprised of four separate phases: (a) *psycho-physical assessment*; (b) *assessment of the implicit attitudes* using two separate IATs (Greenwald et al., 1998); (c) *explicit ratings* of different foods; and (d) *questionnaire session* evaluating dieting and other food-related habits.

2.2.2.1 Psycho-physical assessment

Participants answered five questions regarding their current psycho-physical state by clicking with the mouse on the appropriate point of a *Visual Analog Scale* (VAS). Responses were analyzed by converting distances to a scale ranging from 0 to 100, although this was not explicitly displayed

to the participants. The questions were the following (in brackets the labels at two extremes): (a) 'How hungry are you at the moment?' ('not at all hungry' [0] and 'very hungry' [100]). (b) 'How thirsty are you at the moment?' ('not at all thirsty' [0] and 'very thirsty' [100]). (c) 'How tired are you at the moment?' ('not at all tired' [0] and 'very tired' [100]). (d) 'How much time did pass since your last complete meal?' ('less than an hour' [0] and 'more than 5 hours' [100]). (e) 'How much time did pass since you last have eaten something?' ('less than an hour' [0] and 'more than 5 hours' [100]). Questions were presented to participants in a random order.

2.2.2.2 Assessment of the implicit evaluations (IAT)

Each participant performed two IATs (in counterbalanced order across participants): *UnprocessedFood-IAT (Uf-IAT)* and *ProcessedFood-IAT (Pf-IAT)*.

Each IAT followed the traditional structure consisting of a total of three single-classification practice blocks and two combined test blocks (Greenwald et al., 1998). The evaluative classification involved the classification of words in "positive" and "negative". The target categorization involved the classification of pictures in "food" (unprocessed food or processed food according to the IAT) and "utensils" (i.e., kitchen utensils).

Based on the response key assignment, test blocks were considered either compatible (food/positive-word one key and utensils/negative-word the other key) or incompatible (food/negative-word one key and utensils/positive-word the other key). The IAT effect reflects the relative easiness/difficulty of performing the task in the compatible vs. incompatible block. Response-key assignment and order of the compatible and incompatible blocks were counterbalanced across participants.

The experimental stimuli for the two IATs consisted of 15 images of food-related utensils (e.g., *toaster, slicer,* and *bottle*), and 30 Italian words (of which 15 positive and 15 negative) common to the two IATs. In addition, 15 images of unprocessed food (e.g., *mandarin*, *banana*, and *almond*)

and 15 images of processed food (e.g., *salami*, *potato chips,* and *candies*) were employed in the *Uf*-IAT and *Pf*-IAT respectively (see Figure 2.1). All pictures were color photographs in jpg-format (530x530 pixels) selected from the validated FRIDa Database (Foroni et al., 2013). There were 60 trials in each experimental block. In order to avoid confounding factor and to test directly the distinction between unprocessed and processed foods, stimuli of these two categories were matched on relevant dimensions.

Figure 2.1 Exemplar stimuli used in the Implicit and Explicit tasks. Stimuli consisted of food items both unprocessed (UF) and processed (PF) foods and kitchen utensils.

A separate set of 86 native-Italian speakers (Foroni et al., 2013) with an average age of 23.1 years (SD = 3.3) and a normal-range BMI rated a larger set of images, providing pilot data prior to the present study. Based on the pilot data stimuli were selected to so that unprocessed foods (M = 221.47, SD = 146.92) and processed foods (M = 229.93, SD = 135.69) were not different on calorie content per 100gr ($t(28) = -.164$, $p > .250$). Moreover, unprocessed and processed food images were also matched for arousal, valence, frequency in language and brightness (all $ts(28) < 1.26$, and all $ps > .216$). Critically, unprocessed food images were perceived as low in level of transformation (M = 11.48, SD = 12.31) compared to processed food images (M = 55.66, SD = 14.48; $t(28) = -9.00$, $p < .001$), this served as a manipulation-check. Food images were also matched with utensils for arousal, valence, frequency and brightness (all ts (28) < .853, $p > .250$). Verbal material common to both IATs were selected based on an independent pilot so that positive words (e.g., *star, life, holiday*) and negative words (e.g. *death, accident, poison*) were matched for frequency in language, number of letters, and arousal (all $t(28) < .096$, and all $p > .250$) however, as required by the task they differed on valence ($t(28) = 33.01$, $p < .001$).

2.2.2.3 Explicit ratings

Participants rated all the 30 food images presented in the IATs on different dimensions by clicking with the mouse along a *Visual Analog Scale* (VAS). In order to analyze participants' responses, distances were converted to a scale ranging from 0 to 100, although this was not explicitly displayed to the participants. Participants rated the images of unprocessed food (Uf) and processed food (Pf) on the five dimensions (in brackets the two labels at the extremes of the scale):

(a) *Valence*: 'How negative/positive do you value the content of the picture?' ('Very negative' [0] and 'Very positive' [100]).

(b) *Wanting*: 'How much do you desire in this moment the food represented in the picture?' ('I do not desire it at all' [0] and 'I desire it very much' [100]).

(c) *Health value*: 'How healthy is the food represented in the picture?' ('Not healthy' [0] and 'Very healthy' [100]).

(d) *Hedonic value*: 'How pleasant would it be to experience a mouthful of the food represented in the picture in this moment?' ('Little pleasurable' [0] and 'Very pleasurable' [100]).

(e) *Willingness to pay*: 'How much money are you willing to spend to buy the food represented in the picture?' ('10 euro-cents' [0] and '15 euros' [100] with the middle of the scale labeled '7,50 euro' [50]).

All questions were presented on a computer screen; the centrally presented images measured 530x530 pixels positioned above the VAS scale. The order of presentation of the first two ratings (a, b) was fixed whereas the third and fourth ratings (c, d) were counterbalanced in order, the last rating was always willingness to pay (e), within each block of ratings stimuli were randomized in order.

2.2.2.4 Questionnaires

Participants filled in two standardized questionnaires and answered additional questions assessing their dietary habits and use of psychoactive substances. The order of questions was fixed. The first standardized questionnaire was the *Restraint Scale* (Herman & Polivy, 1980; see *Appendix* 2.1), a 10-question questionnaire used to assess restrained dieting. The second standardized questionnaire was the *EDI-3 Symptoms Checklist* (Garner et al., 1983); this questionnaire was administered in order to identify participants that could be at risk for eating disorders or of aberrant eating behaviors. Moreover, participants were asked to specify if they had religious or medical restrictions to their habitual diet (e.g., allergies or intolerances) and what type of diet they conduct (omnivore, vegetarian or vegan).

2.3 RESULTS

2.3.1 Psychophysical assessment

Participants' psychophysical state assessed through VAS scales was analyzed by converting distances to a scale ranging from 0 to 100. Participants reported on average a low hunger level (*M* = 18.04, *SD* = 22.2), low thirst level (*M* = 35.7, *SD* = 23.6) and low level of tiredness (*M* = 30.18, *SD* = 27.0). Participants had their last snack approximately 2 hours before the experiment (*M* = 33.45, *SD* = 32.8) and last completed meal approximately 3 hours and a half before the experiment (*M* = 59.45, *SD* = 38.8).

2.3.2 Implicit Association Test (IAT)

The IAT effect was calculated, according to the improved scoring algorithm (expressed by Cohen's *d'*, see Greenwald, Nosek & Banaji, 2003). For each participant two IAT-effects were separately computed (i.e., *Uf-IAT* and *Pf-IAT*). The two scores in our sample were positively correlated (*r* = .335, *p* = .020).

The IAT-effect showed no difference for *food-type* (Uf-IAT: *M* = .56, SD = ,05; Pf-IAT: M = .65, SD= .05; paired sample t-test result, t (47) = -1,6, *p* = .12). Irrespective of the type of food, the IAT score showed a significant association between positivity and food relative to kitchen utensils (all *d's* > .56, *t*s (47) > 11.12, *p*s < .001, Figure 2.2).

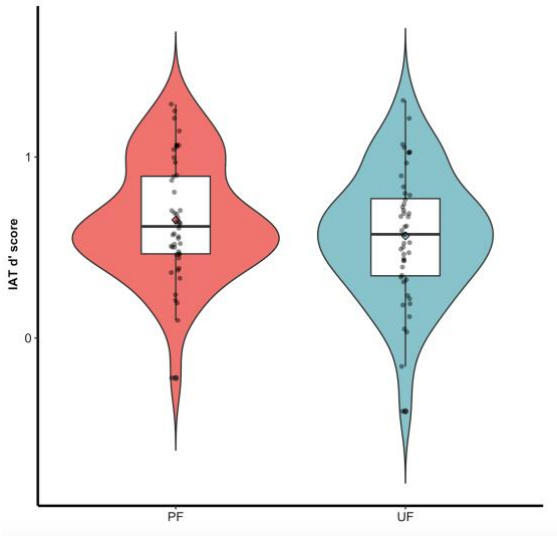


Figure 2.2 IAT d' scores. The black dots represent single data points, the diamonds the means. Each box-plot represents the interquartile range of the distribution with its median. The violin plots represent the smoothed distribution of the data.

2.3.3 Explicit ratings

For each of the five rating scales data were converted to a scale ranging from 0 to 100 and then averaged to create for each question two scores one for unprocessed (e.g., Uf-valence) and for processed food (e.g., Pf-valence) resulting from the average of the valence ratings for all the items in a given category (e.g., Pf). These scores (mean and standard deviations reported in Table 1) were analyzed with *food-type* as a within factor using the non-parametric Wilcoxon for paired samples test. Results of the test are reported in Table 2, significant differences between unprocessed and processed foods were found in the healthiness ratings and the willingness to pay (Figure 2.3), with unprocessed foods rated as healthier and participants willing to pay a higher amount of money for processed foods.

2.3.4 Questionnaires

The total score of the Restraint Scale (Herman & Polivy, 1980) was considered as a continuous variable in the LMM described in the following paragraphs. On average participants scored (M = 11. 33, SD = 5.19) at the RS scale.

The responses to the *EDI-3 symptoms checklist* were used to exclude participants that could be at risk for eating disorders or of aberrant eating behaviors.

Table 1 Explicit Ratings (0-100). Mean values, minima and maxima, standard deviation and standard error are reported.

RATINGS	**Mean**	**Min**	**Max**	**SD**	**SE**
Valence-Uf	69,69	42,33	97,80	14,77	2,13
Valence-Pf	64,48	33,80	90,93	13,32	1,92
Wanting-Uf	38,52	6,60	76,27	17,51	2,53
Wanting-Pf	41,45	1,67	79,00	18,31	2,64
Healthy-Uf	18,09	40,13	88,27	10,38	1,50
Healthy-Pf	27,77	29,40	68,73	8,00	1,15
Hedonic-Uf	72,63	22,53	89,60	16,95	2,45
Hedonic-Pf	45,58	35,33	83,73	10,24	1,48
Willingness-Uf	57,85	3,93	49,33	9,06	1,31
Willingness-Pf	60,17	6,27	59,87	11,87	1,71

Table 2 Wilcoxon paired samples test results z-values and p-values are reported.

Pairs	Z	*p*-value
Valence PF- Valence UF	-1,8	.07
Wanting PF- Wanting UF	-.88	.38
Healthy PF- Healthy UF	-6.01	< .001 ***
Hedonic PF- Hedonic UF	-.92	.36
Willingness to pay PF- Willingness to pay UF	-5.32	< .001 ***

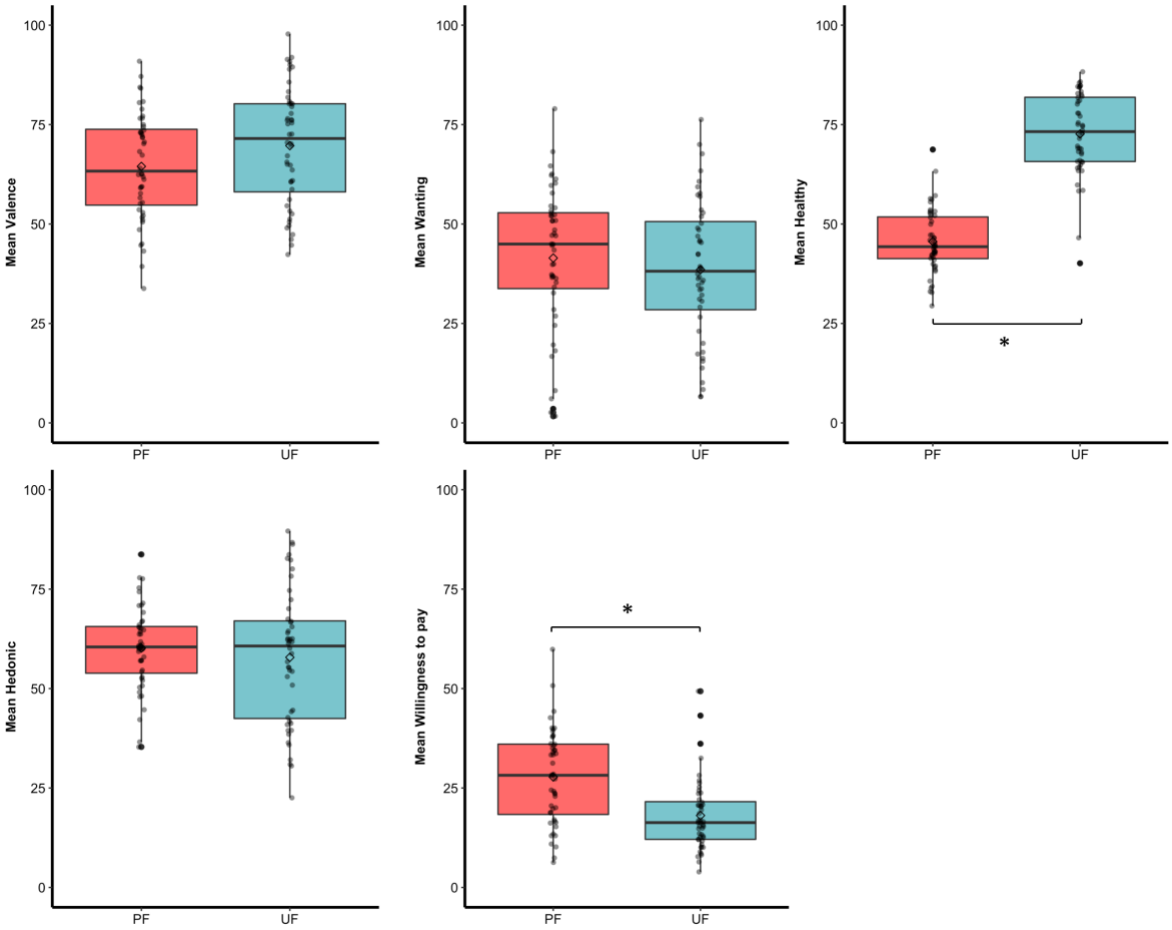


Figure 2.3 The black dots represent single data points, the diamonds the means. Each box-plot represents the interquartile range of the distribution with its median.

2.3.5 Linear Mixed-effects Models (LMM)

Data were analyzed with linear mixed-effects models (LMMs) on Rstudio (version 1.0.44; http://www.rstudio.com/products/rstudio) using the lmer function (lme4 package; http://cran.r-project.org/web/packages/lme4/index.html). A random intercept for participants and for images was included in every model, to account for individual differences and variability related to the different images.

In order to investigate the experimental factors influence on participants' RTs in the IAT task a stepwise procedure was followed. Keeping RTs as our dependent variable we added one factor at a time (*stimulus category* (*food* and *non-food*), *food-type* (*UF* and *PF*), *congruency* (*Food-Positive* and *Food-Negative* trials), *BMI*, *hunger*, *RS*, *explicit ratings* (*Valence, Wanting, Healthy, Hedonic and Willingness to pay*). Models were compared using the Anova function and factors (and interactions) were kept in the model only if they caused a significant increase of fit (tested by the Akaike Information Criterion, AIC); after having selected the significant factors we included second-level interactions in the model.

In our first model, we tested whether there were significant differences between RTs to foods vs non-foods (stimulus category), and a main effect of stimulus category was found with participants responding faster to food stimuli ($t(308) = 2.243$, $p = .026$). The RTs to non-food items were no longer included in the models since participants did not perform explicit ratings on such stimuli and since adding our modulating factors of interest (i.e. RS, BMI) did not lead to an increase of fit of the model.

Focusing then on the RTs to food stimuli, the model with the best fit included food-type, congruency, RS, valence and willingness to pay as factors (AIC = 42094, BIC = 42302, $p = .001$; Table 3); whereas adding any other factor (*hunger*, *BMI*, *gender*, *wanting*, *healthiness*, *hedonic ratings*) did not produce an increase of fit of the model. Only two and three-way interactions will be reported and discussed since higher-order interactions are hard to disentangle and interpret.

Table 3 Summary of the best fitting LMM for RTs, only significant effects are reported.

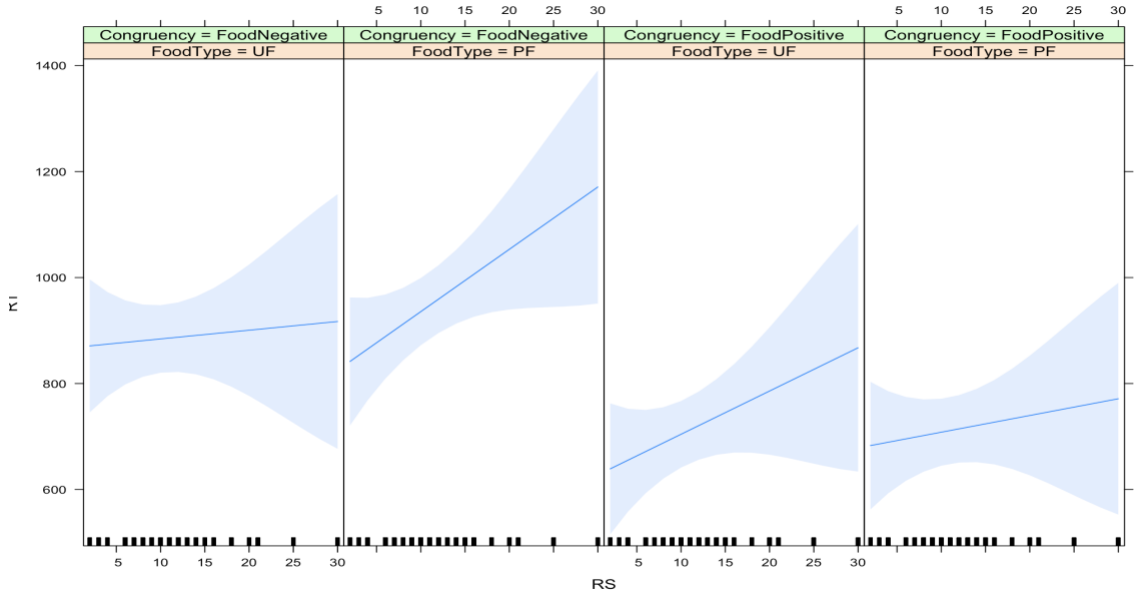
RTs	*β*	*SE*	*t*-value	*p*-value	*95% CI*	
Fixed Effects					*Lower*	*Upper*
Foodtype	-775.6	227.1	-3.41	< .001 ***	-1217.3	-331.6
*Foodtype*Congruency*	909.3	313.2	2.90	.004 **	298.7	1520.3
*Foodtype*Willingness to pay*	24.7	9.4	2.63	.009 **	6.3	43.0
*Foodtype*Valence*	10.9	3.1	3.49	< .001 ***	4.8	17.0
*Foodtype*RS*	94.7	17.0	5.53	< .001 ***	61.2	127.9
*Foodtype*Congruency*Valence*	-11.9	4.3	-2.77	.006 **	-20.4	-3.53
*Foodtype*Willingness to pay*Valence*	-3.7	.12	-3.18	.001 **	-.60	-.14
*Foodtype*Congruency*RS*	-104.3	23.5	-4.44	< .001 ***	-150.1	-58.5
*Foodtype*Willingness to pay*RS*	-2.5	.82	-3.11	.002 **	-4.14	-9.44
*Foodtype*Valence*RS*	-1.19	.23	-5.08	< .001 ***	-4.14	-.94

We will start reporting the effects regarding food-type; we will then report the impact of dietary habits, and finally the relation between implicit and explicit measures. As reported in Table 3, a main effect of food-type was found, as participants were significantly slower in categorizing processed food images as foods compared to unprocessed foods (t(2658) = -3.41, p < .001). A significant interaction between food-type and congruency was found (t(2624) = 2.90, p = .004), as participants were slower in categorizing processed foods as food when they were responding to food images and negative words with the same key (food-negative trials), while no difference was found between types of foods when they were responding to food images and positive words with the same key (food-positive trials). A significant three-way interaction between food-type, congruency and RS score was found (t(2623) = -4.44, p < .001). This shows how both restrained and unrestrained eaters are faster in categorizing processed foods as foods in the food-positive trials (as shown by the food-type by congruency interaction); the slopes of the regression lines indicate that only restrained eaters are significantly slower in categorizing processed foods as foods in food-negative trials (Figure 2.4a). The influence of the explicit factors

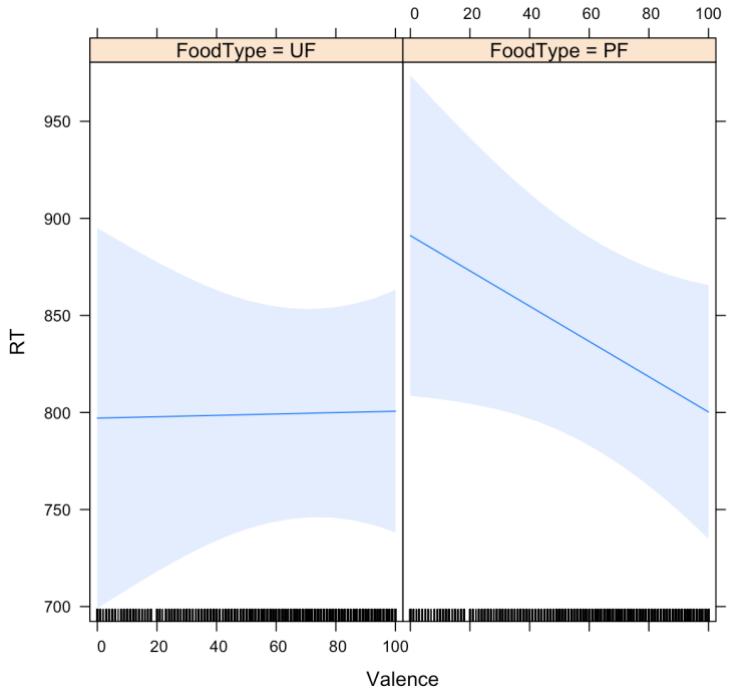
on the RTs is shown by the two-way interaction between food-type and valence ratings ($t(2669) = 3.49$, $p < .001$) and food-type and willingness to pay ratings ($t(2848) = 2.63$, $p < .008$), showing that valence does not influence the RTs in response to unprocessed foods (Figure 2.4b), whereas low valence ratings lead to slower RTs in categorizing processed foods as foods; and that willingness to pay influences RTs to processed foods with participants being faster when they are willing to pay a high amount of money.

Furthermore, the three-way interaction between food-type, congruency and valence ($t(2624) = -2.76$, $p = .005$) shows that overall participants are faster in the food-positive trials, and that participants are significantly slower in categorizing processed foods with low valence as food in the food-negative trials. The effects of the three way interaction between food-type, RS and respectively valence ($t(2624) = -5.08$, $p < .001$; Figure 2.4c) and willingness to pay ($t(2654) = -3.11$, $p = .002$) also indicate that explicit measures interact with subjects' dietary habits, the differences in RTs influenced by valence are present in subjects with higher scores at the RS (Figure 2.4c).

a)


Congruency = FoodNegative
FoodType = UF
Congruency = FoodNegative
FoodType = PF
Congruency = FoodPositive
FoodType = UF
Congruency = FoodPositive
FoodType = PF
RT
RS

b)


FoodType = UF
FoodType = PF
RT
Valence

c)

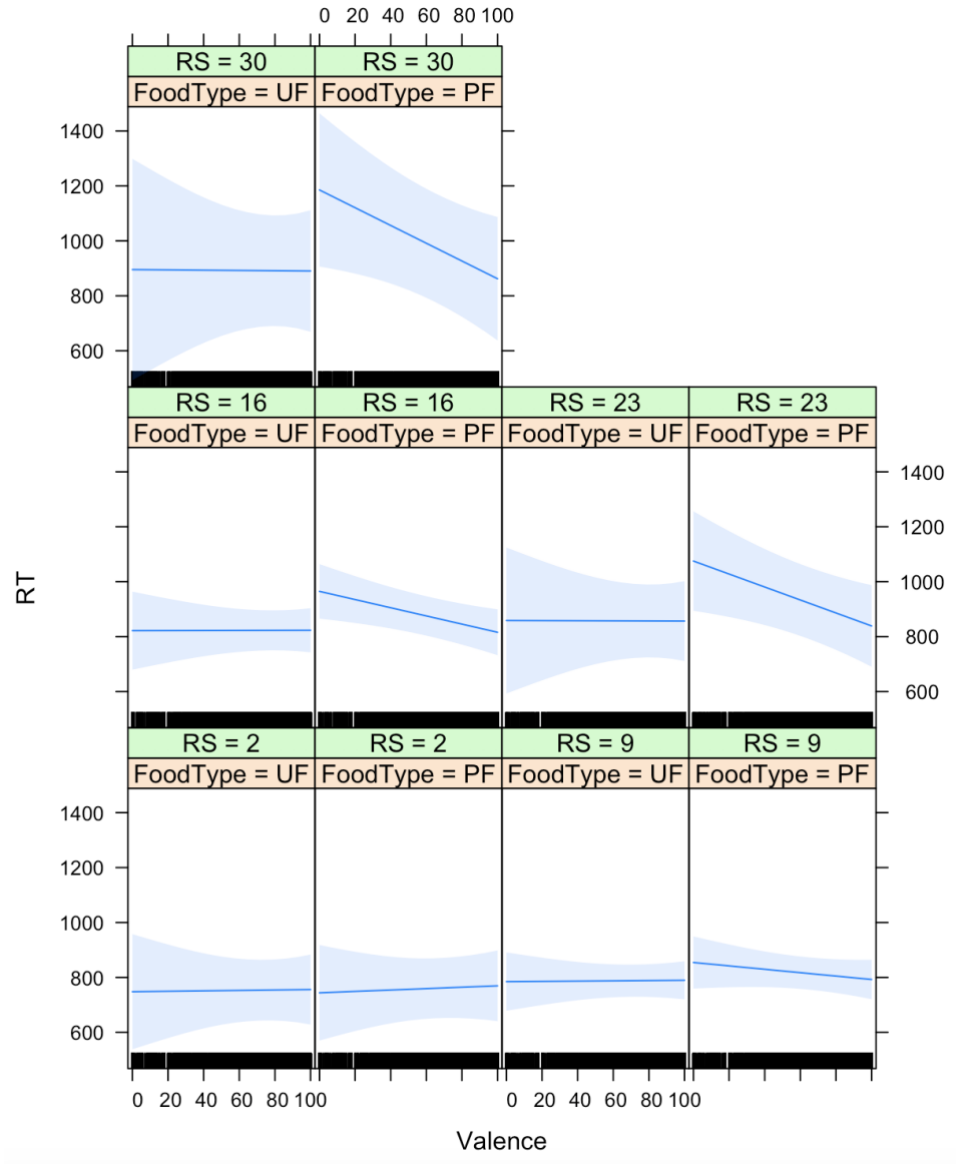


Figure 2.4 Results of the best fitting LMM for RTs a) Three-way interaction of FoodType*Congruency*RS; b) Two-way interaction of FoodType*Valence; c) Three-way interaction of Foodtype*Valence*RS.

In order to investigate which factors modulate participants' explicit ratings, we tested a model with participants' *valence* ratings as dependent variable, following the same stepwise procedure followed for RTs to decide which factors to include. The following factors were significant when added one at a time: *food-type* (*UF* and *PF*), *BMI*, *hunger*, *RS*, *gender***.** Models were compared using the Anova function and factors (and interactions) were kept in the model only if they caused a significant increase of fit (tested by the Akaike Information Criterion, AIC). After having selected the significant factors we included second-level interactions in the model.

The final model included all of the factors we tested (AIC = 26466, BIC = 26544, $p < .001$; Table 4). Only two and three-way interactions will be reported and discussed since higher-order interactions are hard to disentangle and to interpret.

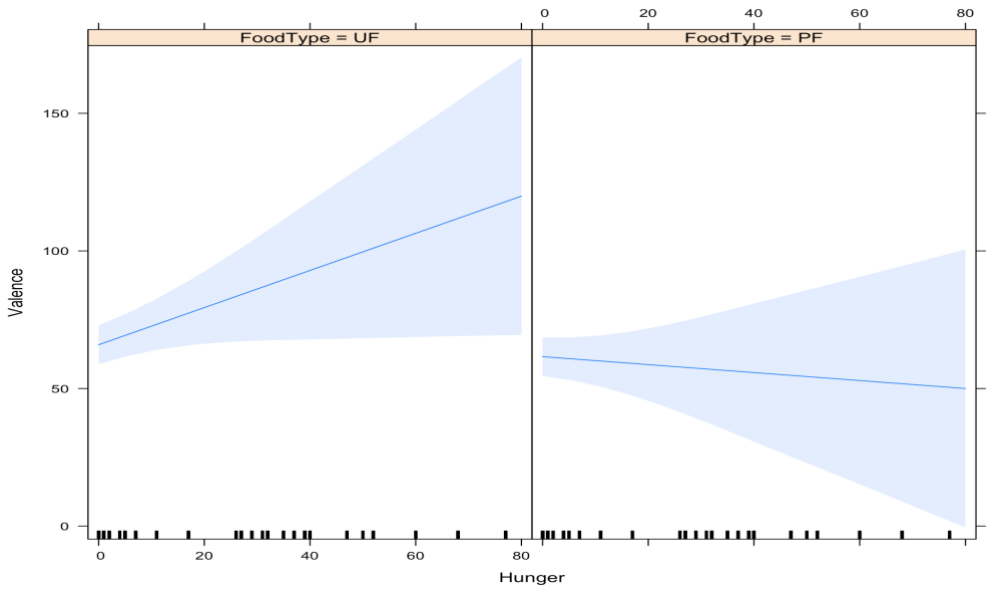
After accounting for interaction effects, no main effect was found to be significant in this model. Interestingly, food-type interacted with both hunger and gender, whereas such factors did not modulate responses at the implicit level, as measured by the RTs.

The interaction effect between food-type and hunger ($t(2304) = 2.30$, $p = .021$; Figure 2.5a) showed that participants' valence ratings of food stimuli were significantly higher when they were hungry, but only for unprocessed foods. The interaction effect of food-type and gender ($t(2681) = 2.60$, $p = .009$; Figure 2.5b) showed that male participants did not differ in their valence ratings, while females showed a significantly enhanced positivity associated to unprocessed foods. Moreover, these effects of gender were found to be affected by participants' BMI, as shown by the three-way interaction between food-type, BMI and gender ($t(2681) = -2.92$, $p = .003$; Figure 2.5c). This indicates that females with higher BMI tended to rate unprocessed food more highly positive.

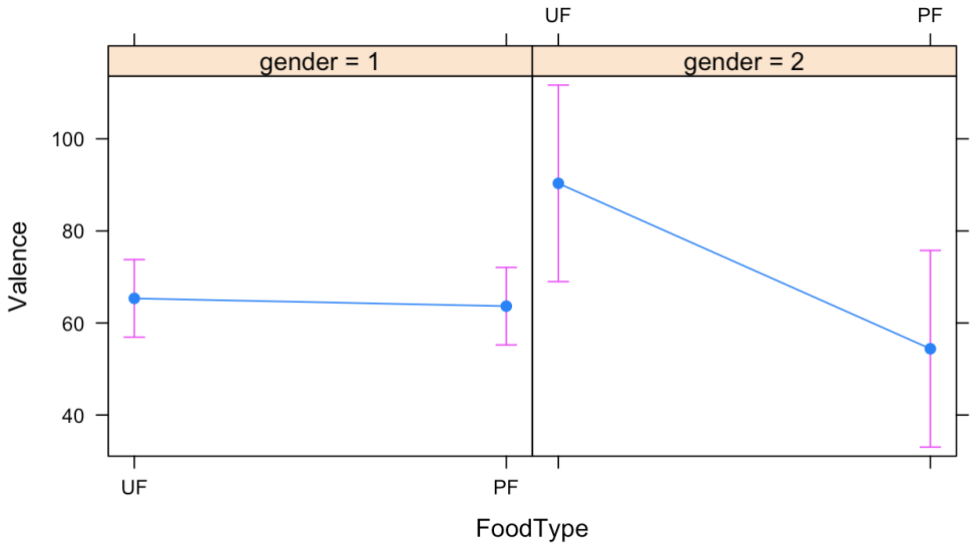
Table 4 Summary of the best fitting LMM for valence ratings. only significant effects are reported.

VALENCE	*β*	*SE*	*t*-value	*p*-value	*95% CI*	
Fixed Effects					*Lower*	*Upper*
*Foodtype*hunger*	5.81	2.52	2.30	.021 *	.87	10.74
*Foodtype*gender*	243.7	93.7	2.60	.009 **	60.45	426.98
*Foodtype*BMI*hunger*	-.26	.11	-2.31	.021 *	-49	-.04
*Foodtype*BMI*gender*	-13.0	4.46	-2.92	.003 **	-21.73	-4.29
Foodtype hunger*gender*	-46.8	8.85	-5.30	< .001 ***	-64.17	-29.57

a)



b)



c)

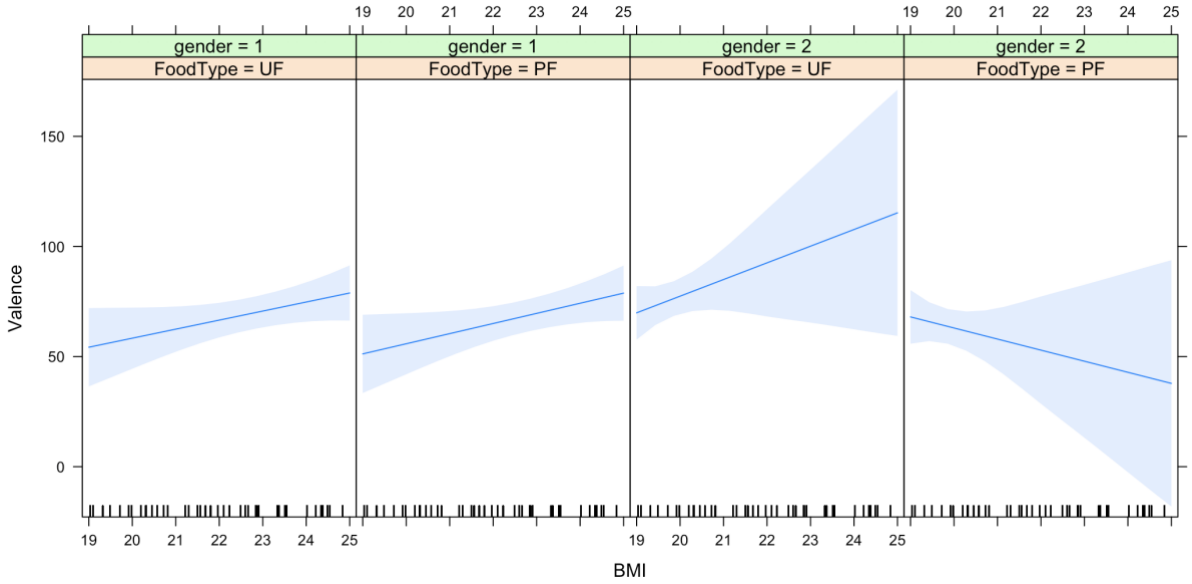


Figure 2.5 Results of the best fitting LMM for valence ratings a) Two-way interaction of FoodType*Hunger; b) Two-way interaction of FoodType*gender (gender 1= males, gender 2= females); c) Three-way interaction of FoodType*BMI*gender.

Finally, we tested a model with the *willingness to pay* explicit ratings as dependent variable, following the same stepwise procedure to add factors (factors considered: *foodtype* (*UF* and *PF*), *BMI*, *hunger*, *RS*, *gender).* Models were compared using the Anova function and factors (and interactions) were kept in the model only if they caused a significant increase of fit (tested by the Akaike Information Criterion, AIC). After having selected the significant factors we included second-level interactions in the model.

The model that was found to explain the willingness to pay ratings included only food-type and hunger (AIC = 26466, BIC = 26544, $p < .001$; Table 5).

Table 5 Summary of the best fitting LMM for willingness to pay ratings. only significant effects are reported here.

WILLINGNESS TO PAY	β	*SE*	*t*-value	*p*-value	*95% CI*	
Fixed Effects					*Lower*	*Upper*
Hunger	.17	.063	2.69	.009**	.04	.31
*Foodtype*hunger*	-.13	.03	-4.61	< .001 ***	-.19	-.08

A main effect of hunger emerged ($t(51) = 2.69$, $p = .009$) with participants willing to pay more when their level of hunger were higher. The interaction between food-type and hunger resulted to be significant ($t(2695) = -4.61$, $p < .001$; Figure 2.6), with participants willing to pay a higher amount of money for processed foods when their hunger level was high and no significant difference with unprocessed foods.

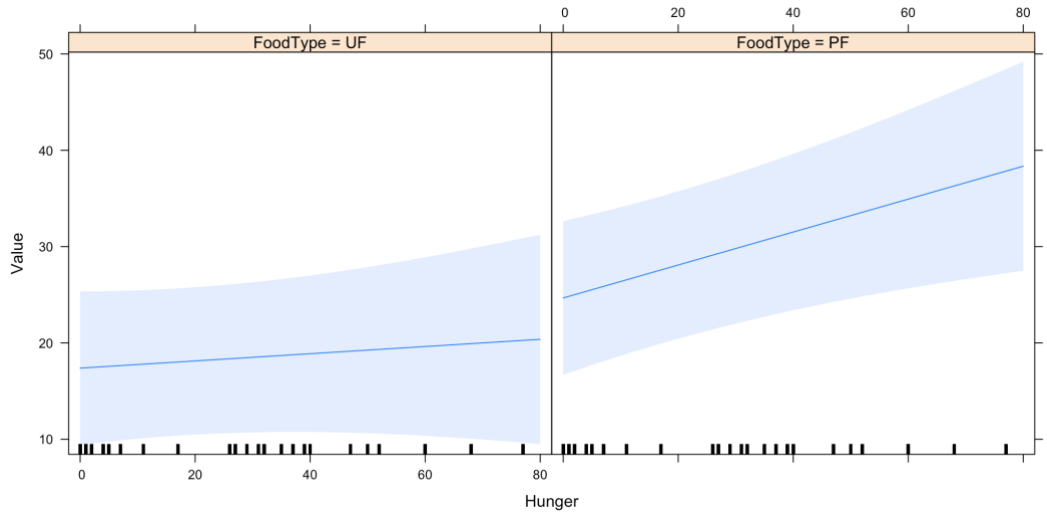


Figure 2.6 Results of the best fitting LMM for willingness to pay ratings. Two-way interaction between FoodType*hunger.

2.4 GENERAL DISCUSSION

In this study, we investigated how unprocessed and processed foods are evaluated by normal weight participants at the implicit and explicit level, and how these levels relate to each other. The main results are the following.

At the implicit level, participants overall held a strong positive evaluation for food compared with food-related objects, revealing positive associations for both unprocessed and processed foods. The RTs in the IAT task were analyzed using LMM which revealed the factors which modulated participants responses at the implicit level. Differences in the RTs to our food categories emerged, with participants slower in categorizing processed foods as foods when the response key was shared with the categorization of negative words. Participants' eating habits assessed through the restrained scale were found to modulate participants RTs, restrained eaters were found to be slower in categorizing processed foods as foods when this category was associated with negative words, as if it was harder for them to associate images of processed foods with negative attributes,

this could explain why these individuals tend to be unsuccessful dieters and could tap into the lack of inhibitory control reported in restrained eaters (Meule et al., 2001, Bartholdy et al., 2016).

Also, the explicit measures were found to modulate participants RTs with shorter RTs in response to processed foods when these were rated as higher in valence, this result is in line with Bielser et al. (2015) finding in which participants were faster in responding to images of foods when their liking ratings were higher for those images, however the task in Bielser et al. was different (choice between two images).

At the explicit level, on average participants rated unprocessed foods as healthier, and were willing to pay more for processed foods. It is important to stress that in our set of stimuli processed foods represent transformed/cooked foods but not necessarily unhealthy foods. Nevertheless, the explicit healthiness rating shows a consistent bias in favor of unprocessed food.

A critical aspect is represented by information time processing between implicit and explicit measurements, with the former having time constrains and the latter allowing participants a long time to make their decisions. When performing explicit ratings, participants might be influenced by aspects such as food healthiness (Sullivan *et al*., 2015) and self-control guiding the best choice among different options (Hare *et al*., 2009). However, when they perform implicit evaluation, the time pressure is more unlikely to allow these factors to guide their evaluations.

In our experiment, participants had no time pressure during explicit ratings, and therefore they might have taken the time to evaluate aspects not only related to food taste or reward but also to how its healthiness. This interpretation is based on the differences in time pressure of our tasks and the fact that evaluating healthiness attributes requires more time than other aspects like tastiness (Rangel *et al.*, 2013; Sullivan *et al*., 2015).

We disentangled these general effects with LMM and we found that factors such as participants dieting habitudes, assessed through the RS, participants BMI, hunger level and gender shaped their explicit evaluations in the following way.

Participants' with a high hunger level were found to rate unprocessed foods as higher in valence, this effect was driven by females and in particular by females with a higher BMI. Hungry females with higher BMI rated processed foods as lower in valence.

Evidence in literature suggests that women have greater activity within prefrontal regions in response to food with high-calorie content suggesting possible gender differences in motivation towards food and self-control mechanisms (Del Parigi, 2002; Killgore 2010; Toepel *et al*., 2012). Our results suggest higher self-control in females, since they seem to be able to suppress their liking for processed foods at an explicit level. An alternative interpretation is that females respond in this way to adhere to normative expectations (Bicchieri *et al*., 2006), this is in line with the fact that gender differences were found only at the explicit level.

Hunger level modulated also participants' willingness to pay ratings, individuals were prone to spend more for processed food when they were hungry.

The impact of hunger state on food perception is well documented in the literature, showing greater activity of the food network when participants are hungry compared to when they are satiated (LaBar *et al*., 2001, Santel et al., 2006; Furher et al., 2008). However, no impact of hunger on the implicit level and the direction of the modulation of hunger at the explicit level was quite surprising since a higher positivity (valence) was associated to unprocessed foods in hungry individuals.

Taken together our results revealed how normal-weight participants evaluate differently unprocessed and processed food, a distinction that received little attention so far (but see Foroni et al., 2016a; 2016b; Rumiati & Foroni, 2016; Rumiati et al., 2016). We found that participants held different evaluations depending on the type of food and that, depending on the amount of time they had available to build up their preference. We speculate that participants responses are guided either by implicit evaluations or explicit evaluation based on the time available and the motivation to take into account abstract and less rewarding attributes of foods.

CHAPTER 3

DISTINCT BRAIN REPRESENTATIONS OF PROCESSED AND UNPROCESSED FOODS: AN ELECTROENCEPHALOGRAPHY (EEG) INVESTIGATION[1]

ABSTRACT

Cooking as a mean for food processing is an exclusively human ability. Cooked diets, have also been argued to have made possible for our ancestors to turn into Homo sapiens. Being able to rapidly distinguish between unprocessed and processed foods might therefore be an adaptive behavior that, as such, should have left a detectable trace in the brain. In order to find such a neural signature with specific spatio-temporal brain dynamics, we applied an electrical neuroimaging analysis framework to visual evoked potentials (VEPs) recorded from volunteers who viewed color images of unprocessed and processed foods adapted in caloric content.

Our results revealed that VEPs to unprocessed vs. processed foods differed as early as 130ms after image onset, driven by changes in topography. From 200ms, these differences were also reflected in the strength of the electric field showing a greater response to processed foods. Source estimations revealed stronger activity within occipital cortical regions, in response to processed foods, whereas stronger premotor and inferior frontal activity was observed in response to unprocessed foods. This is the first evidence of substantial differences in brain responses between unprocessed and processed foods, independent of caloric content.

[1] A version of this chapter is (*in preparation*): Coricelli C., Toepel U., Bielser M. L., Murray M. M., Rumiati I. R. *Distinct brain representations of processed and unprocessed foods*.

3.1 INTRODUCTION

Even though different sensory modalities contribute to food perception (Rolls, 2015), visual recognition alone provides a great deal of information about the intrinsic and rewarding characteristics of food (Linné *et al*., 2002). As reported in the introduction, a brain network subserving human food perception includes occipital visual areas (i.e., fusiform gyrus), orbitofrontal cortex, insula and amygdala (Garcia-Garcia *et al*., 2013; Van der Laan *et al*., 2011, Simmons et al., 2016), as well as fronto-striatal circuits supporting reward evaluation of food stimuli and self-control behaviors (Hare *et al*., 2009; Shur *et al*., 2009; Siep *et al*., 2012).

In addition to having dedicated neural structures, our brains can distinguish between edible and non-edible visually-presented items as early as at 85 ms post-stimulus onset (Tsourides *et al*., 2016) as well as between low and high fat foods within 160ms post-stimulus presentation (Toepel *et al*., 2009; Meule *et al*., 2013), with higher prefrontal, hypothalamic, and striatal activation when viewing high-energy foods (Killgore *et al*., 2003; Beaver *et al*., 2006).

Of particular interest here, is the study by Meule and colleagues (2013) in which participants performed a *Regulation of Craving* (ROC) consisting in evaluating the immediate or later effects of a food target based on a given cue (a word displaying NOW or LATER) and then rating the immediate craving for such food image. Stimuli consisted of 34 high-calorie density foods (HC) and low-calorie density foods (LC) stimuli matched for low-level features (RGB brightness and contrast) and palatability. However, the authors did not take into account the level of processing of the images and 34 out of the 34 HC consisted of processed foods and 33 out of the 34 LC stimuli consisted of unprocessed foods. The results at the electrodes level are displayed in in the Appendix (Figure A3.1), and show how in posterior electrodes the conditions elicited the classical components P1, N1 and P2, with significant differences in the negative amplitude of the N1 in response to low-calorie stimuli irrespective of the condition of the task (now vs later).

Another EEG study of particular importance for the present study is the study of Toepel and colleagues (2009) in which participants had to perform a simple food/non-food categorization task while viewing colored images presented at the center of the screen. This paradigm has been readapted in this study using different images as stimuli. Toepel et al. (2009) showed how distinct responses to low- and high-fat foods emerged in the electric field emerged at the electrode level, using global measures and in the underlying sources, the authors concluded that the implicit discrimination between these food subcategories takes place in two different time windows: around 160 ms post-stimulus onset and around 300 ms. The main results of Toepel are displayed in the Appendix (Figure A3.2).

As pointed out in the introduction chapter, a particular aspect of food recognition that has received little attention to date is the level of processing imposed on food by humans (Rumiati and Foroni, 2016). Thus, given the central role that food processing had in human lineage could have left a neural signature in the brain.

The aim of the study reported in this chapter was to determine whether and how the spatio-temporal brain dynamics in normal-weight participants reveal discrimination between visually presented images of unprocessed vs. processed foods which were on average equal in caloric content. This objective is based on the following considerations. First, as previously found (see Toepel *et al*., 2009; Meule *et al*., 2013), we expected that the human brain is capable of an early discrimination of food stimuli given their relevance for survival. Second and more importantly, this ability to discriminate between processed and unprocessed food is expected to have a neural signature. Different strands of evidence suggest that such ability might have boosted human evolution and shaped food preferences. Moreover, the distinction between processed and unprocessed foods is likely to parallel the distinction between non-living things and living things (see Capitani *et al.*, 2003; Rumiati & Foroni, 2016).

Abundant evidence exists for rapid discrimination between categories of objects (i.e. images of animals vs. non-animals within 100-150 ms post-stimulus onset; Thorpe *et al*., 1996; Antal *et al*., 2000; Proverbio *et al*., 2007 and sounds of living vs. man-made objects within 70-120ms; Murray *et al*., 2006; De Lucia *et al.*, 2010). Given the relevance of food stimuli and the shared properties with both natural and artificial entities, we expected the brain to track the difference between our two food categories in the same time windows previously reported in the literature for natural and artificial stimuli.

3.2 MATERIALS AND METHODS

3.2.1 Participants

Twenty (10 women) healthy Italian speakers participated in the experiment. Participants were aged between 18-35 years (*M*=24.8, SE= .88) and their Body Mass Index (BMI) was in the normal range (18.5 kg/m^2<BMI<25.0 kg/m^2; *M*=21.61, *SD*=1.80, *range*= 18.61-24.93). Participants were not included in the study if: (i) they showed signs of aberrant eating behavior and/or behavioral symptoms in the three months previous testing (e.g. binge eating, vomiting, use of diuretics or laxatives, at least once); (ii) they acknowledged prior neurological or psychiatric illness or the consumption of neurotropic substances; (iii) they had severe dietary restrictions for medical or religious reasons or (iv) they presented traits of a severe depressed mood assessed by the Beck Depression Inventory (BDI-II; Beck et al., 1996). All participants reported being right-handed (confirmed by the Edinburgh Handedness Inventory; Oldfield, 1971) and had normal or corrected-to-normal vision. The experiment began either at 9:00am or 2:00pm, and participants were asked to eat a meal two hours before the beginning of the recordings (upon arrival they reported at what time and the quantity of food eaten) in order to moderate the impact of the circadian modulations of hunger.

3.2.2 Procedure and Stimuli

Upon arrival, participants signed a written informed consent, completed the Edinburgh Handedness Inventory (Oldfield, 1971) and the Beck Depression Inventory (BDI-II; Beck et al., 1996). They were then prepared for the EEG recordings and sat in front of a computer inside a sound attenuated EEG cabin at 60 cm distance from the monitor. Stimulus presentation and registration of responses was controlled by *Eprime 2.0* version (Psychology Tools Inc., Pittsburgh, USA; www.pstnet.com/eprime). Experiment completion required approximately 2 hours, and participants received 30 CHF in compensation. The study conformed to the Declaration of Helsinki and was approved by the Ethics Committee of the Vaudois University Hospital Center (CHUV) of Lausanne.

Before EEG recordings, participants also answered four questions regarding their current psycho-physiological state via *Visual Analog Scales* (VAS). The questions were the following (in brackets the anchored labels): (a) 'How hungry are you at the moment?' ('Not at all hungry' - 'very hungry'). (b) 'How much would you like to eat at the moment?' ('Not at all' - 'a lot'). (c) 'How thirsty are you at the moment?' ('Not at all thirsty' - 'very thirsty'). (d) 'How tired are you at the moment?' ('Not at all tired' - 'very tired').

During the EEG recordings, participants performed a categorization task (Figure 3.1a) involving full-color images. After a central cross appeared for a randomized period between 250 ms and 750 ms images were presented centrally for 500 ms on a 21" CRT monitor. After image presentation, a question mark ("?") appeared, informing participants that they should respond whether the previously viewed image was a food or a non-food item via button press. The question mark disappeared as soon as participants responded or if a maximum period of 1500ms elapsed. This food versus non-food categorization was orthogonal to the discrimination between unprocessed and processed foods of interest for the behavioral and EEG analyses. During the EEG recordings three blocks of trials of 350 color photographs each were presented in pseudo-

randomized order. The 350 images per block consisted of 200 food images and 150 non-food images (Figure 3.1b, for the complete list of stimuli Figure A3.3 of the Appendix). The 200 food stimuli were selected from three different food image databases: images used by Toepel and colleagues (Toepel *et al*., 2009), images from the FRIDa database (Foroni *et al*. 2013) and images from the FoodPics database (Blechert *et al*., 2014). Images were resized (300x300 pixels) and placed on a white background (plates in the background were removed) using the GNU Image Manipulation Program (GIMP; https://www.gimp.org). The 200 food images were equally divided in 100 unprocessed foods (UF; e.g. *avocado*, *strawberry*, *raw prawns*) and 100 processed foods (PF; e.g. *grilled zucchini*, *blue cheese*, *ice cream*). Within the 100 unprocessed, foods 50 images comprised low calorie foods and 50 high calorie foods; the same for the processed foods. Overall, food images were matched on various dimensions that were statistically tested with independent samples t-tests. In particular, the unprocessed and processed images were matched in *caloric density* (kcal per 100g portion; UF: M = 194.98 kcal/100g SE = 21.37; PF: *M* = 201.66 kcal/100g, *SE* = 16.15; $t(198) = -.250$, $p = .803$), *valence* (UF: *M* = 60.61, *S*E = 1.04, PF: M = 59.74, SE = .84, $t(198) = .650$, $p = .516$, and *arousal* (UF: *M* = 42.44, SE = 1.66, PF: M = 42.18, SE = 1.21; $t(198) = .124$, $p = .901$). Non-food images were represented by three categories consisting of kitchen utensils (e.g., *knife, pan, cup*), common objects (e.g., *dice, balloon, book*) and natural entities that are not edible (e.g., *rose, coral, leaf*). The images were matched in luminance and spatial spectral power following the analyses described by Knebel *et al.* (2008).

After the end of the EEG recordings, participants again viewed all of the food images on the PC screen in pseudo-randomized order and rated them on VAS scales via a computer mouse. The images were rated along the following dimensions: *valence* 'How negative/positive do you value the content of the picture?' ('Very negative' [0] and 'Very positive' [100]), *wanting* 'How much do you desire in this moment the food represented in the picture?' ('I do not desire it at all'

[0] and 'I desire it very much' [100]) and *frequency of consumption* 'How frequently do you eat the food represented in the picture?' ('Never' [0] and 'Every week' [50], 'Every day' [100]).

Finally, participants filled in paper-pencil questionnaires regarding their health state (e.g. hours slept, time and quantity of food ingested, quantity of caffeine, cigarettes and alcohol, habitual drugs and other factors that could have influenced their performance). Moreover, participants filled in the EDI-3 (Garner *et al*., 1983) questionnaire, which is informative of individuals' dieting and eating habits.

3.2.3. Electroencephalography (EEG) acquisition and preprocessing

A 128-channel Biosemi ActiveTwo system (Biosemi, Amsterdam, The Netherlands; http://www.biosemi.com) acquired continuous EEG at 512 Hz sampling rate and referenced to the common mode sense (CMS; active electrode) and grounded to the driven right leg (DRL; passive electrode), which functions as a feedback loop driving the average potential across the electrode montage to the amplifier zero. Data preprocessing and the reported analyses were all performed using Cartool software (http://www.fbmlab.com/cartool-download; Brunet *et al*., 2011).

Single participant's raw EEG data were inspected trial-by-trial using epochs from 98 ms pre- to 488 ms post-stimulus onset (corresponding to 50 data points pre- and 250 data points post-stimulus onset) using an artifact rejection criterion of ±80 μV at each channel. Epochs containing eye blinks and other movement or non-stereotypic artifacts were rejected. During averaging, data were baseline corrected using the 98 ms pre-stimulus period, filtered (0.1 HZ high-pass, 40 Hz low-pass, and 50 Hz notch), and recalculated against the average reference. Data at artifact electrodes were interpolated using 3-D splines (Perrin *et al*., 1987). On average 6.4% (SE= 1.12) of trials were rejected during single subject preprocessing. Furthermore, only trials on which participants were accurate were further analyzed, such that in the end the average percentage of

excluded trials was 9.15% (SE=1.80) in individuals. Visual Evoked Potentials (VEPs) were first calculated for each participant and stimulus condition (i.e. unprocessed food and processed food images). Second, VEPs to each stimulus condition were group-averaged.

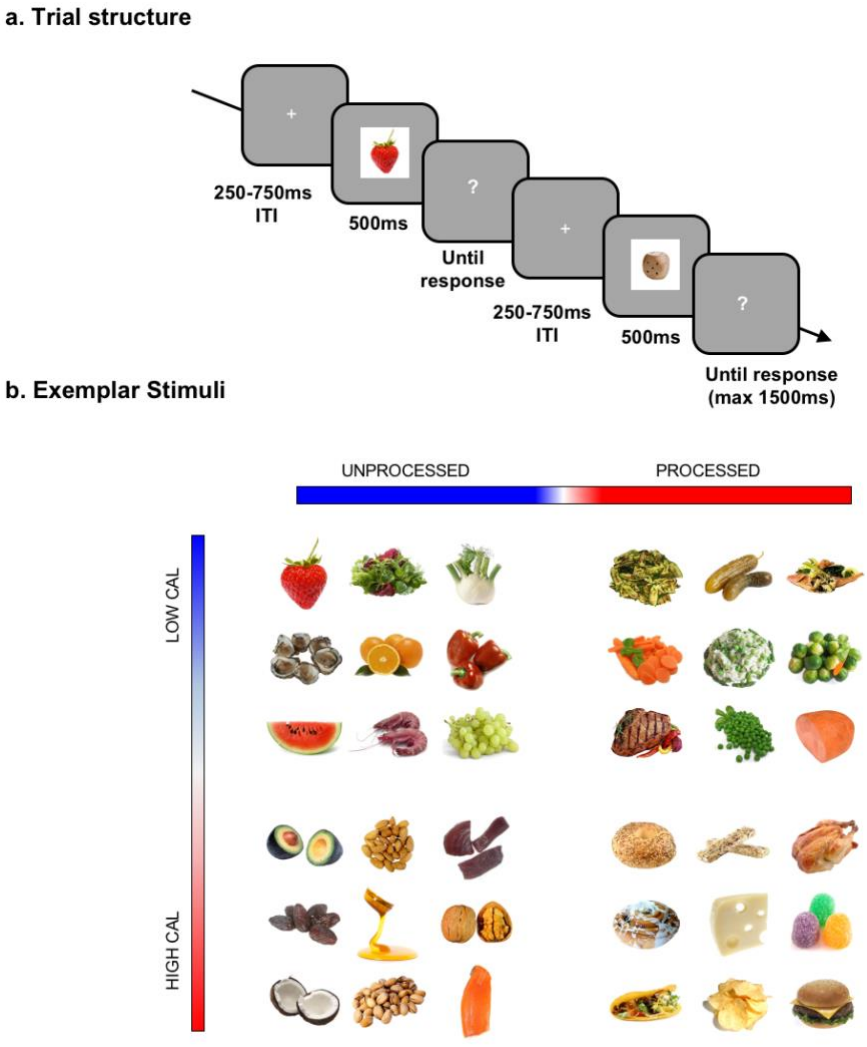


Figure 3.1 a) Exemplar trial structure. Participants had to perform a food/non-food categorization task of each image responding via button press after the "?" appeared on the screen. Inter-trial interval (ITI) varied randomly between 250-750 ms; b) Exemplar images of unprocessed foods (UF), processed foods (PF).

3.3 EEG DATA ANALYSIS

3.3.1 General analysis strategy

In order to investigate whether VEPs to unprocessed and processed food images differed, we conducted VEP analyses at the single waveform level, on the global strength of the electric field at the scalp (viz. global field power, GFP), on the topography of the VEP (viz. global map dissimilarity, GMD, as well as topographic clustering), and on the estimated intracranial sources of the VEP responses (viz. LAURA source estimations). Details of the analyses will be described in the following paragraphs; however, these methods have been extensively described elsewhere (Murray *et al*., 2008; Brunet *et al*., 2011; Michel and Murray, 2012). All analyses presented are based on paired contrasts between unprocessed and processed food viewing conditions.

3.3.2 Analysis of VEPs waveform modulations

As first level of analysis, VEP group-averaged waveform data from all 128 electrodes were analyzed as a function of time. At each time point (millisecond-by-millisecond; Murray et al., 2004) pairwise comparisons (t-tests) between the two conditions (unprocessed vs processed) at each of the scalp electrodes were performed. Effects that did not last at least 20ms (11 contiguous data-points) were rejected in order to correct for temporal auto-correlation at individual electrodes (as done in Toepel *et al*., 2009). Results of this analysis give a visual impression of the distribution of significant differences in time and space; minimizing the possibility of missed effects (type II errors).

3.3.3 Global electric field analyses

The second level of analyses first assessed the modulations in the strength of the electric field at the scalp using GFP (Lehmann and Skrandies, 1980) over the post-stimulus period for each participant and stimulus condition using paired t-tests. GFP is calculated as the square root of the mean of the squared value recorded at each electrode (vs. the average reference) and is equal to

the spatial standard deviation across electrodes at a given instant in time (Michel and Murray, 2012). Larger GFP values are associated with greater synchronized neural activity.

Differences in the topography between electric fields in response to each stimulus condition were assessed through the analysis of GMD (Lehmann and Skrandies, 1980). Strength-normalized data were compared (by dividing potentials at each electrode of a given map by its GFP). GMD is calculated as the root mean square of the difference between two normalized maps and can range from 0 to 2, where 0 indicates identical maps and 2 maps topographies inverted. A Monte Carlo nonparametric bootstrapping procedure (5000 permutations per time point) referred to as "topographic ANOVA" or "TANOVA" (Murray *et al*., 2008), identified statistical differences in the GMD between two conditions by comparing the observed GMD with the empirical distribution obtained from the bootstrapping. Effects that lasted at least 11 contiguous data points (~20ms; with a *p-value* below 0.05) were considered reliable (as in Toepel et al., 2009). Topographic differences are indicative of differences in the underlying neural generators. Time intervals in which the GMD differed between stimulus conditions were therefore used as time intervals of interest for the source estimation analysis (LAURA). Of note, analyses of GMD and GFP are independent, observations of time intervals with GFP modulations with a lack of differences in map topographies are interpreted as the modulation of statistically indistinguishable generators across stimulus conditions.

3.3.4 Intracranial source estimations of electric activity

A distributed linear inverse solution was applied using the local autoregressive average (LAURA) regularization approach (Grave de Peralta *et al*., 2001; for a review, see Michel *et al*., 2004a for a review). Distributed source models reconstruct the brain electric activity in each point of a 3D grid of solution points. LAURA selects the sources that better mimic the behavior of the electric vector fields using a realistic head model, which included 3005 solution point nodes of a 6x6x6 mm grid equally distributed within the gray matter of the Montreal Neurological Institute's

average brain. The time intervals in which differences in the GMD were found were used as time periods of interest regarding modulations in neural source activity between conditions. Paired t-tests were calculated at each solution point node using the variance across participants. Only nodes with *p*-values < 0.05 and within clusters of at least 10 contiguous nodes were considered significant. The results were rendered on the MNI brain with the Tailarach and Tournoux (1988) coordinates.

3.4 RESULTS

3.4.1 Psychophysical state

Participants' psychophysical state assessed via VAS scales ranging from 0 to 100. Participants reported a low hunger level (*M*=39.26, *SD*=6.41), a low desire to eat (*M*=37.04, *SD*=5.71), low thirst (*M*=42.93, *SD*=5.21), and a low level of tiredness (*M*=25.62, *SD*=5.55). As requested, participants reported that they ate a meal 2 hours before the beginning of the recordings.

3.4.2 Behavioral results: Accuracy and RTs

Participants' performance was at ceiling during the categorization between food and non-food images. Accuracy was calculated as the percentage of correct answers. On average (± s.e.m) participants correctly categorized 98.20 ± 0.2% of foods and 98.61 ± 0.2% of non-food images (paired t-test: t(19) = -2.13; p = .046). Within the food images, 98.30 ± 0.2% of unprocessed foods and 98.03 ± 0.3% of processed foods were correctly categorized (paired t-test: t(19) = 1.38; p = .184, see Appendix Figure A3.4a).

As participants were instructed to respond only after the question mark appeared that followed the images presentation, reaction time data (RTs) are less informative. Participants were equally fast in categorizing food and non-food images, on average (± s.e.m); 342 ± 20 ms to categorize food images and 331 ± 20 ms for non-foods (paired t-test: t(19) = 1.71; p = .103). A significant

difference in RTs emerged within the food category; 352 ± 20 ms for unprocessed food and 332 ± 20 ms for processed food (paired t-test: t(19) = 3.01; p = .007, see Appendix Figure A3.4a).

3.4.3 Behavioral results: Ratings

Participants' ratings of the images were analyzed by averaging data of each subject and each image category viewed, results revealed that on average (± s.e.m) participants' ratings did not differ between unprocessed and processed foods in terms of valence (UF: 70.06 ± 3.04; PF: 65.83 ± 2.61; t(19) = 1.66; p = .112, see Appendix Figure A3.4b), wanting (UF: 50.89 ± 3.15; PF: 50.65 ± 3.75; t(19) = .074; p = .941) nor frequency of consumption (UF: 49.43 ± 3.28; PF: 48.60 ± 2.77; t(19) = .315; p = .756).

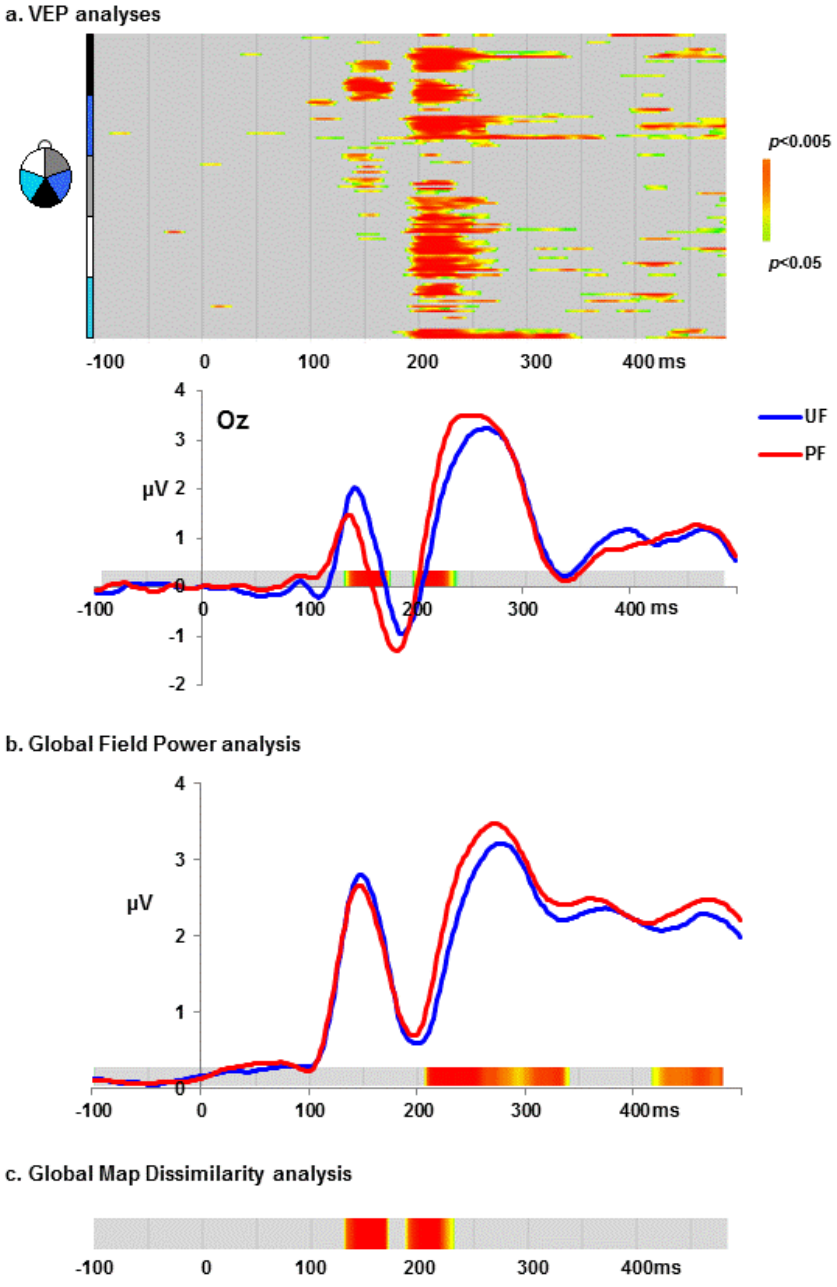
3.4.4 Results of VEP data analysis: Waveform modulations at individual electrodes

Single electrode level analyses revealed VEP response differences between unprocessed and processed food images. Differences arose as early as around 130 ms at posterior electrodes and more wide-spread around 200 ms (Figure 3.2a). Group-averaged VEP waveforms per each condition at an exemplar midline occipital electrode (O_z) revealed a first peak with positive amplitude greater for unprocessed foods in the 130-175 ms interval and a second positive peak significantly greater for processed foods in the 195-240 ms interval (Figure 3.2a). These VEP peaks correspond to the traditional series of ERP components, including the P1, N1 and P2. Significant differences in amplitudes emerged in the posterior electrodes in both P1 and P2, whereas N1 component amplitude differences between our conditions were not significant in the central occipital sites.

3.4.5 Results of VEP data analysis: Global measures of the electric field

Significant modulations in response strength between conditions, assessed via global field power (GFP), were observed over the 207-341 ms interval and >400ms (Figure 3.2b) post-stimulus onset, wherein processed foods elicited stronger responses than unprocessed foods. Significant

modulations in response topography between unprocessed and processed foods assessed via global map dissimilarity (GMD) were observed over the 130-171 ms and 187-232 ms intervals post-stimulus onset (Figure 3.2c), indicating different configurations of brain sources over these intervals.



* Caption of Figure 3.2 in the following page

Figure 3.2 a) Results of the electrode-wise and millisecond-by-millisecond paired t-tests between unprocessed and processed conditions. Temporally sustained differences emerged around 150 ms post stimulus onset. Exemplar midline occipital electrode (O_z) group-average waveforms. b) Modulations in response strength assessed through Global Field Power waveforms, differences between conditions emerged in the 207-341 ms and >400 ms post-stimulus onset; c) Modulations in response topography assessed through Global Map Dissimilarity emerged in the 130-171 ms and 187-232 ms intervals.

3.4.6 Results of source estimations analysis

For the time intervals revealed by the global map dissimilarity (GMD) analysis (130-171 ms and 187-232 ms) distributed source estimations were calculated for each condition and each individual participant. The reported coordinates represent the maximal t-values within a cluster and are based on the Tailarach and Tournoux (1988) system, the corresponding Brodmann areas (BA) are reported (Figure 3.3). Over the 130-171 ms post-stimulus period, several brain regions showed different responses to unprocessed vs. processed foods. The occipital cortex (BA 17/18/19; x= 9, y= -69, z= 23), and the lateral portion of the right inferior temporal cortex (BA 20; x=60, y=-7, z=-18) showed stronger responses to processed foods than unprocessed foods.

In contrast, the left lateral premotor cortex (BA 6, x= -31, y= 7, z= 58) and left inferior frontal gyrus (BA 47, x= -60, y= -1, z= 23) were significantly stronger in response to unprocessed foods than processed foods. Over the 187-232 ms post-stimulus onset distributed brain regions showed differential responses to unprocessed and processed foods.

The occipital cortex (BA 17/18/19; x= 33, y= -75, z= 35) extending to the parietal cortex, responded significantly stronger to processed foods than unprocessed foods. In contrast, the left precentral gyrus (BA 4, x= 40, y= 12, z= -27) and right medial portion of the inferior temporal cortex (BA 20, x= -9, y= -27, z= 71) showed stronger responses to unprocessed foods than processed foods.

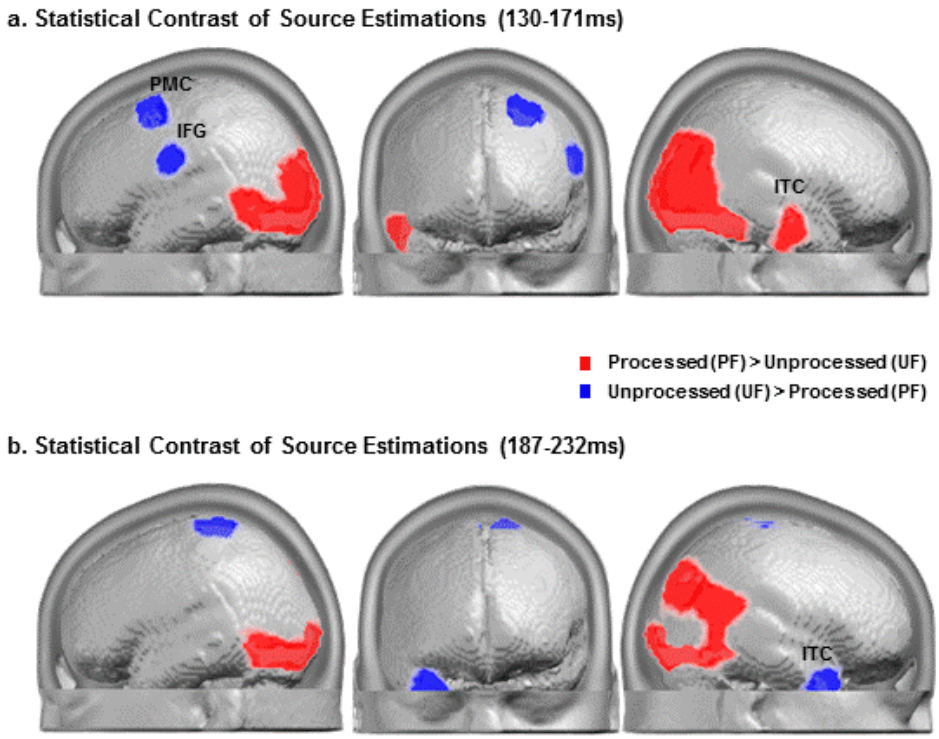


Figure 3.3 Source estimation in the 130-171 ms and 187-232 ms post-stimulus onset intervals (panels a and b, respectively). Sources are displayed on the MNI template brain.

3.5 GENERAL DISCUSSION

We investigated whether and how the brain differentially responds to processed and unprocessed foods. To this end, normal-weight participants viewed images of processed and unprocessed foods that were matched for physical characteristics such as brightness and spatial frequency, as well as for valence, arousal and, most importantly, for caloric density. Behaviorally, no differences between the two food categories were found either in response accuracy or ratings. However, a

significant difference in reaction times emerged; participants were faster in categorizing processed foods.

The spatio-temporal brain dynamics revealed the following differential responses. First, differences between processed and unprocessed foods were observed as early as 130 ms post-stimulus onset. This timeframe was reported by Toepel *et al*. (2009) and Meule *et al.* (2013) in studies in which participants showed an implicit within-category discrimination of food caloric content. Our data thus suggest that the brain not only discriminates food's caloric content, but also other food properties. Furthermore, the ability to discriminate food items on the basis of their degree of processing is adaptive given that processed foods are generally associated with higher net energy gain after ingestion (Carmody *et al*., 2011), and that some transformation procedures, such as cooking at high temperatures or boiling, reduce the risk of food infections.

The time interval in which the processed-unprocessed discrimination occurs is in common with other biologically salient stimuli such as faces (Michel *et al*., 2004b) or animals (Antel *et al*., 2000; Proverbio *et al*., 2007), although the dedicated and distinct brain network in response to food stimuli (Van der Laan *et al*., 2011) suggests that foods may represent a separate object category. Discriminating between processed and unprocessed foods efficiently and rapidly might well be the neural signature that developed through human evolution (Wrangham *et al*., 2003).

The VEPs inspection revealed the traditional series of ERP components, including the P1, N1 and P2. Significant differences in amplitudes emerged in the posterior electrodes in both P1 and P2, whereas no significant difference between our conditions emerged in the N1 in the central occipital sites. Previous studies showed independent P1 component phases, with the later portions (peak around 100-130 ms post-stimulus onset) arising from the fusiform gyrus (see Di Russo et al., 2002) compared to the early portions (60-90 ms) that instead arise from the dorsal extrastriate cortex. In our results, this component's amplitude at peak was significantly greater in response to unprocessed foods. Moreover, the P1 wave has been found to be sensitive to subject's

state of arousal (Vogel & Luck, 2000). Greater negativity of the N1 component was found in the literature at temporal and parietal sites for non-animals (fruits, flowers, buildings) compared to animals by Antal and colleagues (2000), and for low calorie foods by Meule and colleagues (2013), in our results, although it did not reach a significant difference, a greater negativity was found for processed foods at the posterior central midline electrodes (Oz). However, it is not possible to directly compare these results since the tasks differed across studies.

Differences at the scalp were supported by distinct activity of distributed brain areas. Wide activations within the occipital cortex were found to be higher in response to processed foods. Greater activation in visual areas has been observed, in response, for instance, to high-calorie foods (Toepel *et al*., 2010; Killgore *et al*., 2003; Simmons *et al.*, 2005), as well as to other highly relevant stimuli such as monetary rewards (Small *et al*., 2005) or emotional faces (Eger *et al.,* 2003; Moratti *et al*., 2004). The difference in occipital activation we observed is not likely due to differences in visual complexity of the stimuli (as in Stingl *et al.*, 2010), since our stimuli were matched for low-level features; however, it cannot be excluded that processed food composition might be more complex (see also Meule *et al*., 2013). For instance, using multivariate pattern classification on MEG data, Cichy *et al*. (2014) found that in the ventral visual cortex objects were discriminated along the dimension of naturalness with a peak at 122 ms, whereas the animacy dimension peaked later at 157 ms. This particular finding on the naturalness dimension of objects is mirrored in our study with food stimuli within the same time window, at 130ms. Moreover, our source estimation results showed that the right inferior parietal lobule and right temporo-parietal junction were more strongly active in response to processed foods. These regions have been suggested to be part of the human attentive system (Posner, 1990) whose key function is to allow individuals to detect relevant stimuli in the environment (Corbetta and Shulman, 2002). Processed foods likely fall into this category.

Whether the discrimination between our stimuli might be a particular instance of the more general living vs. man-made entities discrimination remains to be understood fully. Previous electrical neuroimaging studies have already successfully revealed the time intervals and the underlying sources of this discrimination when stimuli were visually (Michel *et al*., 2004b) or auditorily (Murray *et al*., 2006, DeLucia *et al*., 2010) presented. In our study, the lateral portion of the inferior temporal cortex was found to be more strongly active in response to processed foods, whereas the medial portion of the temporal cortex was observed to be more active in response to unprocessed foods. This lateral-medial gradient within the temporal cortex parallels the gradient reported in previous studies using man-made (Chao *et al*., 1999, Grill-Spector et al., 2014) or natural entities (Martin and Chao, 2001). Even though the critical role of the inferior temporal cortex in visual object recognition is well documented (Kriegeskorte et al., 2008a; for a review see Grill-Spector and Weiner, 2014), its engagement in the discrimination of food categories had hitherto remained unknown.

On the other hand, unprocessed foods activated areas in the frontal and parietal lobes. More specifically, the left lateral premotor cortex was found greatly activated in response to this category of foods. This activation is consistent with action preparation given that unprocessed foods are likely perceived as requiring some actions in order to be consumed (e.g., a banana needs to be peeled off), whereas processed foods have already undergone substantial transformation and, as such, are more likely "ready" to be eaten. This interpretation is in line with the study by Foroni *et al.* (2013) in which unprocessed foods were perceived by participants as more distant from edibility than processed foods, which were perceived as ready to be consumed. The inferior frontal gyrus and the precentral gyrus were both found to be more strongly activated by the unprocessed foods. Using the Representational Similarity Analysis on fMRI data both regions have recently been found to represent food words similarly to action words associated with the mouth

and to tool words (Carota *et al*., 2017), suggesting that the associated actions are mapped similarly in these regions.

Our study suggests that processed foods like other highly relevant stimuli (high caloric foods, monetary rewards, or emotional faces) greatly activate the ventral visual pathway. In contrast, unprocessed foods activate premotor and inferior frontal regions since they require some actions in order to be eaten. This is the first study that reported differential brain responses towards unprocessed and processed food stimuli, demonstrating how the brain keeps track of this dimension early as 130 ms post-stimulus onset.

CHAPTER 4

NEURAL CODING OF BRAIN REGIONS INVOLVED IN FOOD PERCEPTION IN HUMANS: AN fMRI INVESTIGATION

ABSTRACT

No animal can live without food. Food choices pervade our everyday life, however to date the way in which food stimuli are represented in the brain is not fully understood. With the present fMRI study, we wanted to extend knowledge regarding food perception and categorization by investigating brain responses to diverse food sub-categories, in particular focusing on the unprocessed/processed dimension. Our preliminary results show how brain activations are modulated by our different categories of food types, with processed foods greatly activating the basal ganglia involved in the reward system.

4.1 INTRODUCTION

As described in Chapter 1, the functional magnetic resonance imaging (fMRI) was employed in several studies to identify the neural basis of food perception. The food network that is active when participants viewed foods compared to non-foods led to brain activations in response to food compared to non-food in the following regions: anterior insula, fusiform gyrus, hippocampus, orbitofrontal cortex and cerebellum (Huerta et al., 2014, for a meta-analysis; see Chapter 1). This network has been replicated several times and it has been shown to be modulated by several factors, the most relevant of them being the caloric content. High-calorie foods greatly activated the dorsolateral prefrontal cortex, thalamus, hypothalamus and the cerebellum, whereas low-calorie foods activate greatly the superior temporal gyrus and the ventromedial prefrontal cortex

(Killgore et al., 2003). The prefrontal cortex has been found to play a central role in food choices, as found for other rewarding stimuli (i.e., money), with the ventromedial prefrontal cortex (vmPFC) being involved in encoding the value of food, and the dorsolateral prefrontal cortex (dlPFC) playing a central role in self-control (Hare et al., 2009). The ventral visual stream in the occipitotemporal cortex is of particular interest when addressing the issue of how food is represented in the brain. In fact, along this stream areas have been found to be activated in response to biologically relevant categories such as faces, visual word forms or places as stimuli have often been reported in the literature (Deheane & Cohen, 2011; Grill-Spector & Weiner, 2014). Therefore, it is highly probable that these regions might as well respond to food.

However, as foods share properties with both natural and man-made objects it might be hard to single out dedicated neural correlates of food category as a whole. Patient studies too suggest that food as a category appears to violate the living/non-living dichotomy (for a review Capitani et al., 2003).

Recently, the decoding of object representations in the ventral visual stream has been carried out in imaging studies (Kriegeskorte et al., 2008a; Konkle & Caramazza, 2013; Grill-Spector & Weiner, 2014; Cichy et al., 2014). The results confirmed the early finding that the representation of *animacy* play a central role in object representation, with a medial-to-lateral organization along the ventral surface of the cortex (Chao et al., 1999; Martin, 2007; Konkle & Caramazza, 2013). On top of this, the important role of real-world object *size* has been shown, with size representations also showing a medial-to-lateral gradient (Konkle & Oliva, 2012; Konkle & Caramazza, 2013, see Appendix Figure A4.1): this factor should be taken into account when comparing object categories as they include stimuli that are heterogeneous in their real-world size and as such they might lead to confounding results.

Interestingly, Kriegeskorte et al. (2008a) presented 92 colored images of real-world objects to monkeys and humans, and recorded single neurons activity in the primate brain and high-

resolution fMRI in humans. Patterns of responses for each stimulus were compared through representational dissimilarity matrices (RDM; Kriegeskorte et al., 2008). They found a striking match between monkey and human RDMs, with the main grouping factor of the stimuli being *animacy*. Multidimensional Scaling (MDS) was chosen to plot the RDM results in a two-dimensional space, objects that are placed close together in the MDS elicited similar response patterns, and stimuli placed far apart elicited different response patterns (Figure 4.1, for human IT results).

As Figure 4.1 shows, objects belonging to a specific category (e.g., faces, animals, places, body parts) tend to elicit similar response patterns and are clustered close to each other in the MDS. In the human IT food stimuli, however, do not cluster as a category, and are instead sparse in the inanimate portion of the MDS, with no clear boundary between natural and artificial objects., Specifically food stimuli do not tend to cluster for type, shape or color.

As suggested in previous Chapters 1-3, the distinction between unprocessed and processed foods has received little attention to date (Foroni & Rumiati, 2017). In Chapter 3, I reported the results from the EEG study that suggest how the brain tracks the difference between these two food-subcategories as early as 130 ms post-stimulus onset, and that this discrimination is supported by several regions.

The EEG study addressed mainly the temporal dynamics of our visually presented food categories, with the present study I chose fMRI to get a better spatial resolution in order to respond to a couple of questions regarding food perception. First, I further investigated brain responses to foods as a category, while carefully controlling for factors such as the real-world size. Second, I disentangled brain responses to unprocessed and processed foods by exploiting fMRI's whole brain coverage and spatial resolution while accounting for foods caloric content.

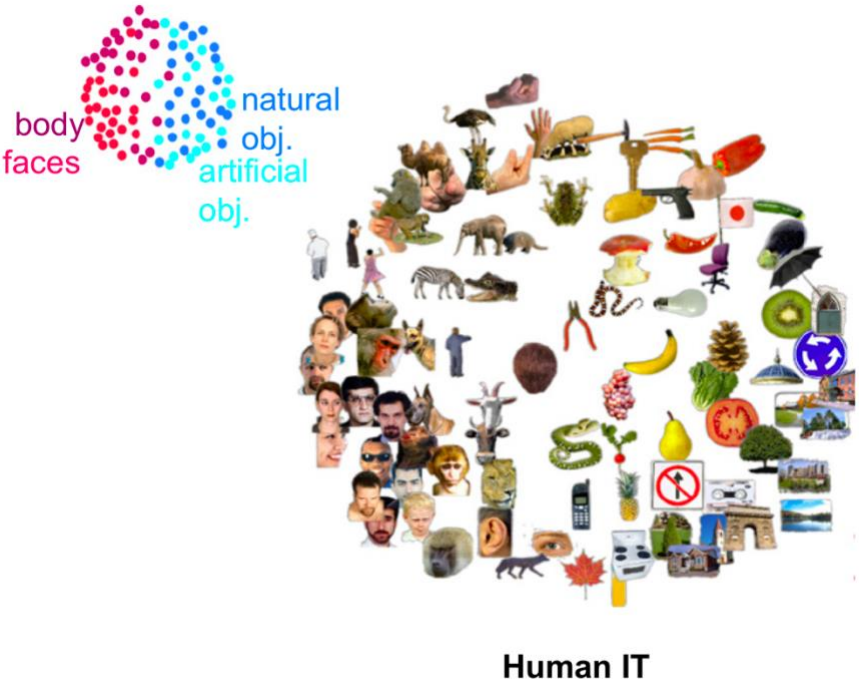


Figure 4.1 Kriegeskorte et al. (2008a) Multidimensional Scaling results in human IT.
*Figure from: "Matching categorical object representations in inferior temporal cortex of man and monkey". *Neuron*, *60*, p. 1130.

4.2. MATERIALS AND METHODS

4.2.1 Participants

Thirteen (6 females) healthy right-handed English speakers participated in the experiment. In this chapter, preliminary data collected from 10 (5 females) right-handed English speakers are reported. Participants were aged between 18-35 years (M=27.7, SE= 2.2) and their Body Mass Index (BMI) was in the normal range (18.5 kg/m^2<BMI<25.0 kg/m^2; M= 21.55, SD= 1.9). Participants were not included in the study if: (i) they showed signs of aberrant eating behavior

(ii) they acknowledged prior neurological or psychiatric illness or the consumption of neurotropic substances; (iii) they had severe dietary restrictions for medical, religious or personal reasons. All participants had normal or corrected-to-normal vision and were asked to eat a meal two hours before the beginning of the recordings (upon arrival they reported at what time and the quantity of food eaten), in order to moderate the impact of the circadian modulations of hunger. This study was conducted at the Brain and Mind Institute, London (Ontario, Canada). The study conformed to the Declaration of Helsinki and was approved by the Ethics Committee of the Brain and Mind Institute.

4.2.2 *Stimuli*

Participants in the scanner viewed colored photographs of foods and non-food items. In the block design localizer runs participants viewed 32 pictures belonging to four different categories: animals (*bat, squirrel, frog*), objects (*tennis-ball, cd-rom, light bulb*), foods (*donut, orange, ice-cream*), and scrambled images, 8 per each category (Figure 4.1, for the complete list of stimuli). Images sized 700 x 700 pixels were centered and placed on a white background. Images in the localizer run were matched for real world object size (see Konkle et al., 2012), adding a control that hasn't been taken into account in most of the studies present in literature. Scrambled images of the food stimuli were generated on Matlab using the *box-scramble* method in which images are broke into boxes and then such boxes are randomly shifted around, the scrambled images preserve the color of the original image.

Figure 4.2 Complete set of stimuli used in the localizer runs.

During the event related experimental runs participants viewed pictures of 32 food stimuli which differed along level of processing and caloric content dimensions (Figure 4.2, for the complete list of stimuli used) resulting in the following four categories of stimuli unprocessed foods low-calorie (UF_Low), unprocessed foods high-calorie (UF_High), processed foods low-calorie (PF_Low), unprocessed foods high-calorie (PF_High). Caloric content of each food item was calculated on the exact quantity (in grams) of the image instead of using the caloric density (kcal per 100g; as done in previous studies). On average our stimuli did not differ in the *actual caloric content* between unprocessed (M = 362.3; SD = 115.2) and processed foods (M = 250.5; SD = 46.08; paired t(15) = 1,19; p = .25). In addition, the stimuli two stimulus types were equally *recognizable* and *familiar* (*ps* > .59). Each food type consisted of both sweet and salty stimuli, and foods with various shapes (i.e elongated, round) and of different colors. Images sized 700 x 700 pixels were centered and placed on a white background. An analysis of the low-level features was carried out on luminance, spatial frequency and silhouette using Matlab functions.

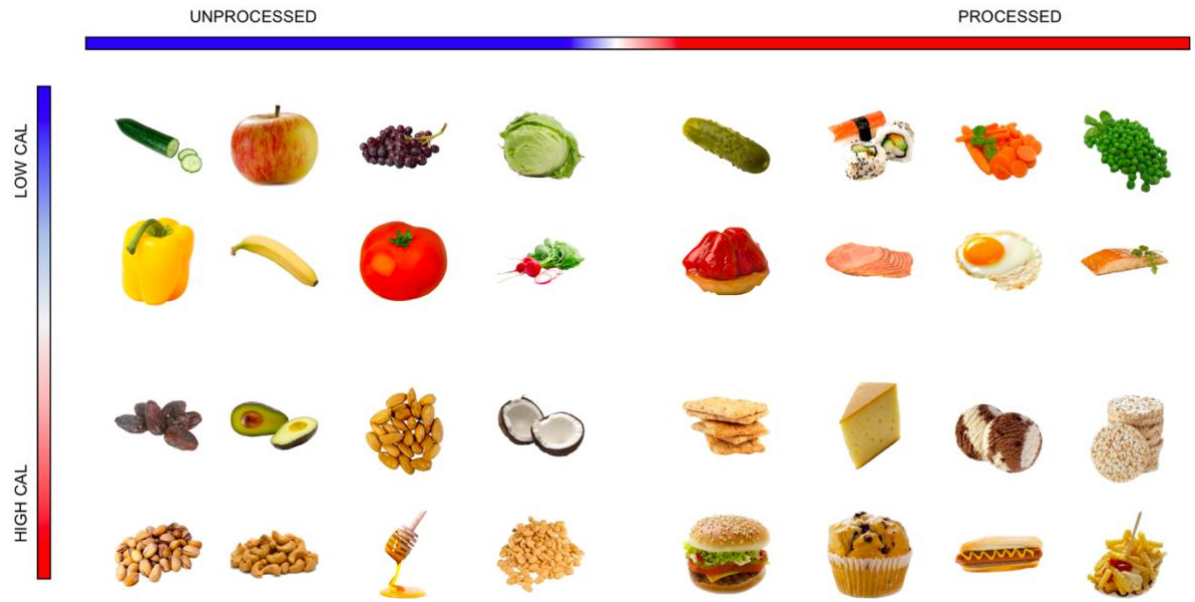


Figure 4.3 Complete set of stimuli used in the experimental runs.

4.2.3 *Procedure*

The study comprised three separate phases: questionnaires, scanning session and image ratings. Participants were pre-screened with a questionnaire on their dietary habits. Once participants were selected and agreed to take part in the fMRI study, upon arrival at Robarts Research Institute they filled in the *Eating Attitude test* - 26 items (EAT-26; Garner et al., 1982) and responded to questions regarding their hunger level and what they have eaten before their testing session through visual analogue scales (VAS scales). Then, participants were positioned in the 3T scanner and viewed on a screen positioned behind them through a mirror system positioned above the head coil and responded through a button box that was positioned in their hand, the scanning session consisted of two localizer runs, eight experimental runs and a structural (MPRAGE) acquisition, in total participants were scanned for about 1 hour and a half. When they exited the scanner, they completed image ratings on a lab laptop and were questioned again on their hunger

level. The experiment lasted overall about 2 hours and participants received 50 CAD for their participation.

4.2.3.1 Localizer runs

Participants performed two localizer runs in which they passively viewed images (Figure 4.3 for a schematics of the run). Stimuli were presented in a block design, and each image was presented at the center of the screen for 2 s, so that each category block (animals, food, objects, scrambled) lasted 16 s for a total of 64 s per block interleaved with 16 s of null event (blank screen). The 64 s images blocks were repeated four times (with a different order of the categories, two localizer runs of about 6 minutes per run were performed.

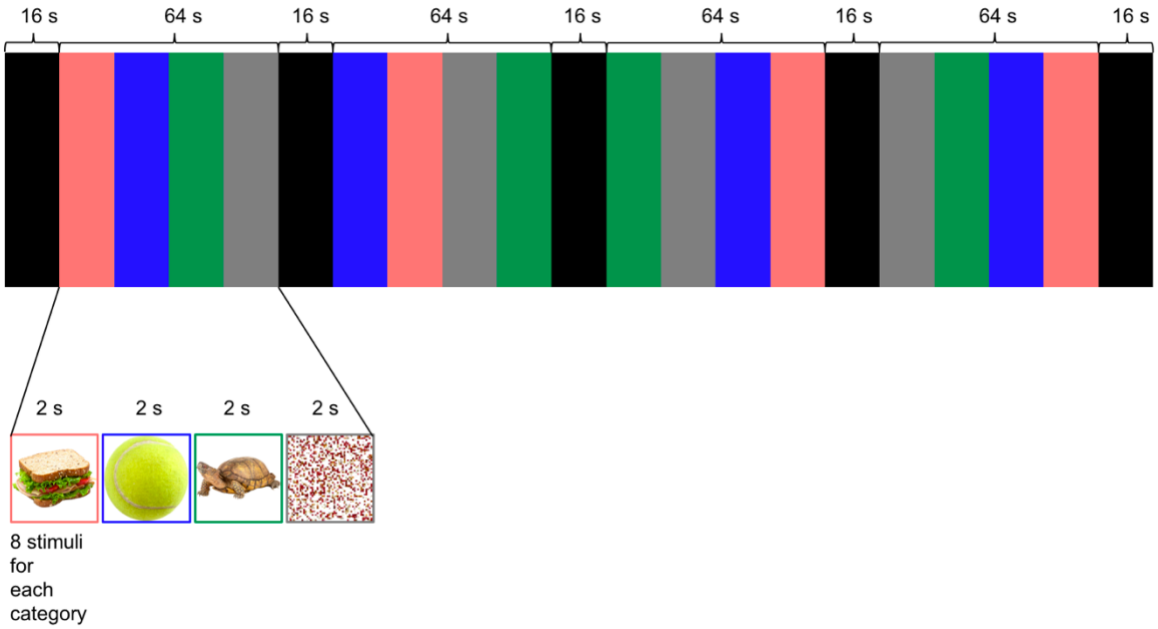


Figure 4.4 Schematics of the localizer run.

4.2.3.2 Experimental runs

After the two localizer runs, participants were presented with eight experimental runs with a fast event-related design. Each block began with a 16s fixation block, each image was

presented at the center of the screen superimposed by a "*bull-eye"* fixation for 1s, random inter-trial intervals (ITI) of 4s each allowed us to jitter the presentation of our experimental stimuli (see Figure 4.5 for a schematics of the run), participants had to perform a one-back task responding via button press each time the exact stimulus was presented back-to-back. This task was chosen in order to maintain participants' attentive level high; however, such trials were excluded from my data analysis (only four trials for each experimental run were repetitions). In each run, the 32 food pictures were presented twice taking into account the null events and the repetitions each experimental run consisted of 106 events (of 4s each) for a total of about 7 minutes per run.

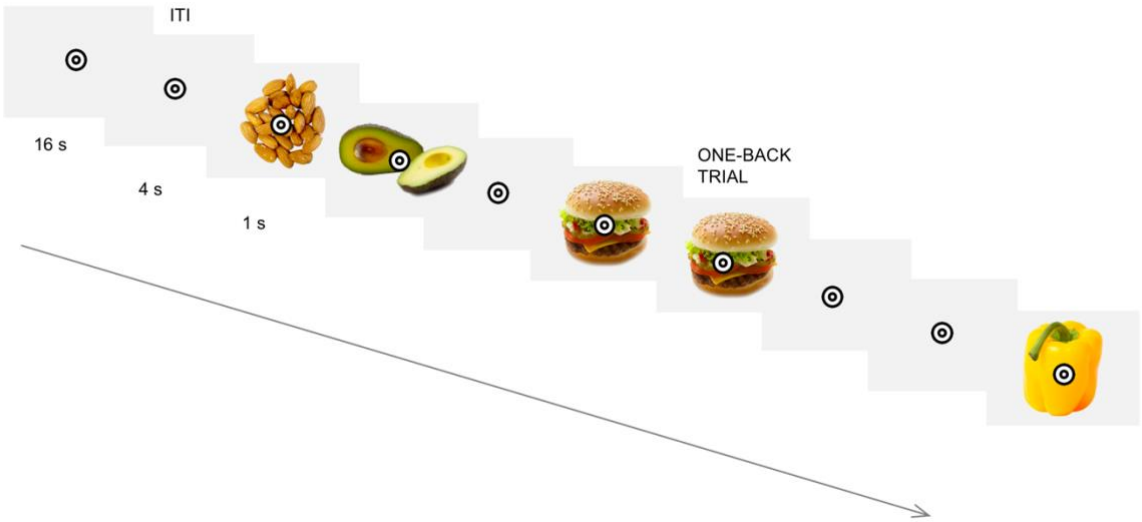


Figure 4.5 Schematics of the Experimental Runs.

4.2.3.3 Ratings

Once participants exited the scanner they had to rate each of the images they viewed in the scanner on the following questions, questions were presented in a block manner while images were presented randomly.

(a) *Tastiness*: 'How tasty is this food?' ('Not at all' [0] and 'Very [100]);

(b) *Healthiness*: 'How healthy is this food?' ('Not at all' [0] and 'Very [100]);

(c) *Liking*: 'How much do you like is this food?' ('Not at all' [0] and 'Very [100]);

(d) *Frequency of consumption*: 'How often do you eat this food?' ('Not at all' [0] and 'Very [100]).

4.2.4 Data acquisition

Data collection was performed at Robarts Research Institute at the University of Western Ontario (London, Ontario, Canada) using a 3T Siemens MAGNETOM Prisma fit MRI scanner with a 32-channel head coil. Functional experimental volumes were collected using a T2*weighted, echo-planar imaging (EPI) acquisition sequence with a multiband accelerator factor of 4, time to repetition (TR) 1000 ms; 2.5 x 2.5 x 2.5 mm isotropic slice thickness, time to echo (TE) 28 ms; field of view 210 x 210 mm^2, matrix sixe 84 x 84, flip angle 40°. Each functional volume comprised 52 slices oriented at a 30° angle to the AC-PC in order to acquire a near to whole-brain coverage and optimize the BOLD signal in the orbitofrontal cortex. Each functional run consisted of 336 volumes in the localizer runs and 424 volumes for the experimental runs. The T1-weighted structural volumes were acquired using a MPRAGE sequence; TR 2300 ms, TE 2.98 ms, field of view 192x240x256 mm, matrix size 256x256 mm^2, flip angle 9°, 1 x 1x 1 mm isotropic slice thickness.

4.2.5 Data Analysis

Data analysis was performed using Brain Voyager QX 2.8.465 (Brain Innovation; http://www.brainvoyager.com/).

4.2.5.1. Preprocessing

Data of each subject was preprocessed by applying slice scan-time correction, volumes were 3D motion corrected, and high-pass temporal filtered (cutoff of frequencies of 3 cycles per run). Data was smoothed using a Gaussian kernel with full width at half maximum (FWHM) of 4 mm.

Functional runs were realigned to the first volume of the first experimental run after the MPRAGE structural acquisition which was also used for functional-to-anatomical coregistration. Functional and structural volumes were aligned to the ACPC plane and transformed into Talairach space using sinc interpolation.

4.2.5.2 General linear model

Both, localizer and experimental runs data was analyzed with a fixed-effects (FFX) general linear model that included one predictor for each of the 4 categories of stimuli (for localizer runs: animals, foods, objects, scrambled; for experimental runs: UF_Low, UF_HighPF_High, PF_Low foods), the conditions were convolved with the "two-gamma" hemodynamic response function (Friston et al., 1998) as default for Brain Voyager. As predictors of no interest, we included the 6 motion parameters resulted from the 3D motion correction analysis (x, y, z, for translation and rotation). Results are overlaid on the Talairach Colin brain template.

4.3 RESULTS

4.3.1 Localizer runs

Whole-brain analyses were thresholded at $p < .05$, false discovery rate (FDR) corrected at the threshold level, with a cluster threshold of 50 voxels. Preliminary results on 10 subjects will be reported. The first contrast that was tested as a sanity check was the contrast between *food* and *scrambled* conditions. Scrambled images as expected greatly activated the primary visual cortex (V1) while foods greatly activated the extrastriate cortex (see Figure A4.2 in the Appendix). When the second contrast between *food* and *animal* category was carried out, the *animacy* gradient from medial-to-lateral (see Konkle et al., 2012) was found: animals greatly activated the lateral portion of the occipitotemporal cortex (ranging from the fusiform gyrus to the lateral occipital cortex, LOC), while food stimuli greatly activated the medial portion of the occipitotemporal cortex (parahippocampal gyrus, see Figure 4.6 and Table 1).

Consistently with what has been found in the literature (Huerta et al., 2014), when contrasting food stimuli and objects, we found a greater activation in a large portion of the occipital cortex, bilateral posterior insula, left hippocampus, bilateral anterior cingulate in response to foods (see Figure A4.3 in the Appendix). Surprisingly, in this preliminary analysis objects did not greatly activate any region more than foods.

Finally, foods were contrasted with all the other three categories (animals, objects and scrambled) matched for real world object size. The results show that foods greatly activated bilaterally the occipital cortex, bilateral parahippocampal gyrus, left anterior cingulate and cerebellum (see Figure 4.7 and Table 2), again in line with what found in previous studies.

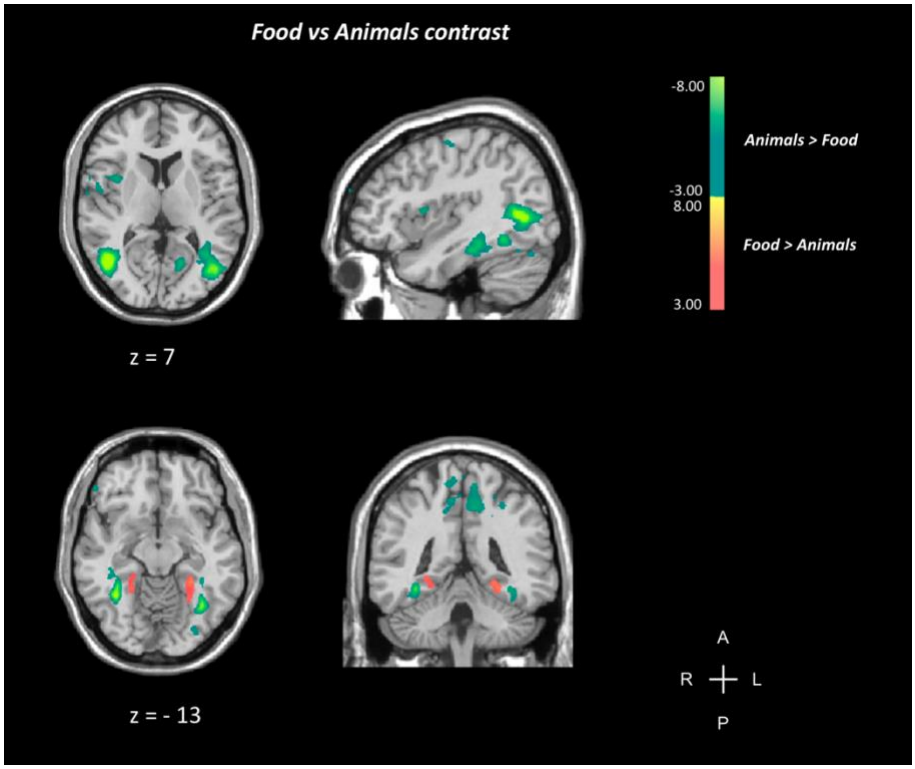


Figure 4.6 Results of the whole-brain contrast between food and animal categories, FDR corrected at $p < .05$ and a cluster threshold of 50 mm^3.

Table 1 Regions exhibiting differences between food and animal stimuli, coordinates are in Talairach space.

	Peak Coordinates				
Location / Side	**x**	**y**	**z**	**Volume (mm^3)**	**t - value**
Food > Animal					
Parahippocampal gyrus **L**	-27	-41	-12	457	4.66
Parahippocampal gyrus **R**	27	-41	-6	326	4.35
Supplementary Motor Area **R**	-17	23	62	248	5.01
Animal > Food					
LOC **R**	45	-67	6	988	- 10.98
LOC **L**	-45	-75	2	979	-10.67
Inferior temporal gyrus **L**	-17	-65	8	615	-4.97
Inferior parietal lobule **L**	-11	-47	50	591	-4.86
Intraparietal sulcus **L**	-27	-53	46	580	-5.35
Insula **R**	56	8	-4	481	-4.69
Cuneus **R**	15	-77	20	463	-5.04
Fusiform gyrus **L**	-37	-43	-16	424	-5.13
Premotor **R**	21	-5	64	363	-5.49

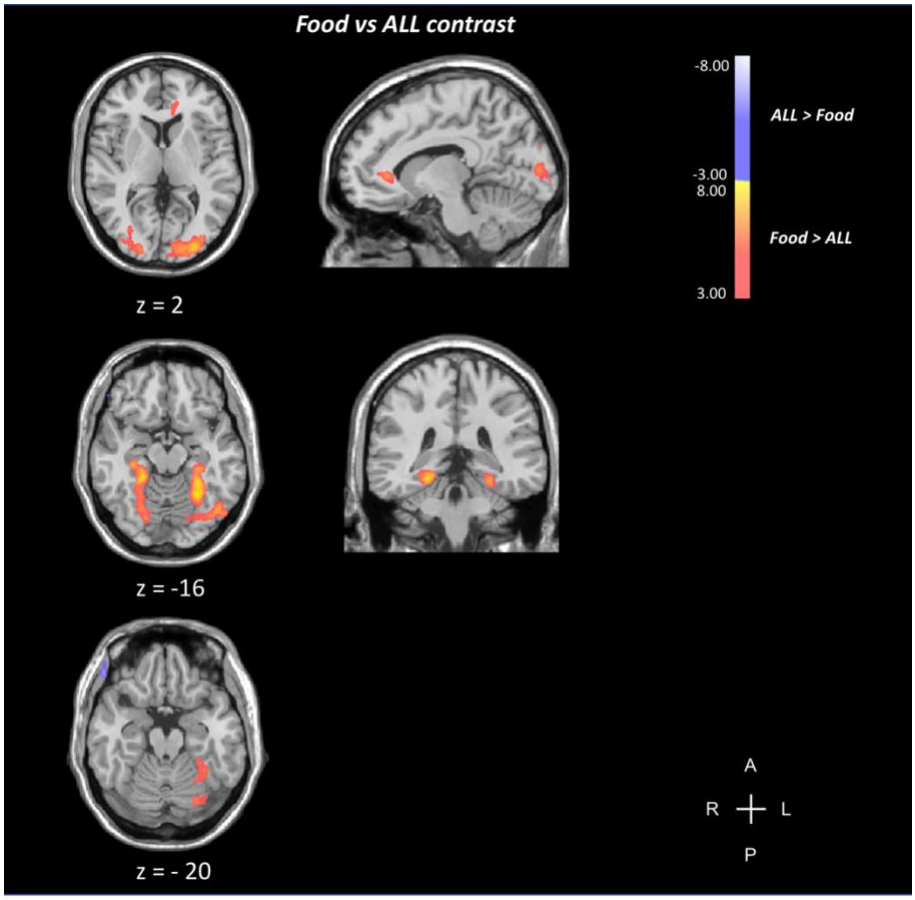


Figure 4.8 Results of the whole-brain contrast between food and ALL (animal, object, scrambled), FDR corrected at $p < .05$ and a cluster threshold of 50 mm^3.

Table 2 Regions exhibiting differences between food and ALL (animal, object, scrambled), coordinates are in Talairach space.

	Peak Coordinates				
Location / Side	**x**	**y**	**z**	**Volume (mm^3)**	**t - value**
Food > ALL					
Parahippocampal gyrus **L**	-27	-55	-14	823	7.47
Occipital cortex **R**	29	-89	8	746	5.81
Occipital cortex **L**	-29	-93	4	673	6.44
Fusiform gyrus **L**	-47	-69	-16	585	5.42
Parahippocampal gyrus **R**	23	-39	-14	543	6.88
Anterior Cingulate **L**	-11	33	2	332	5.39

4.3.2 Experimental runs

Whole-brain analyses were thresholded at $p < .05$, preliminary uncorrected results on 10 subjects will be reported with a cluster threshold of 100 voxels. High-calorie foods (both processed and unprocessed; PF_high and UF_high) and low-calorie foods (both PF_low and UF_low) were contrasted. High-calorie foods greatly activated a wide portion of the occipital visual cortex, the striatum and vmPFC (Figure 4.9). Low calorie foods greatly activated regions extending from the cuneus, intraparietal sulcus, premotor cortex and the bilateral insula (Figure 4.9 and Table 3).

The contrast between processed foods (both with a low and a high caloric content; PF_low and PF_high) and unprocessed foods (both UF_low and UF_high) was tested. Processed foods greatly activated the occipital cortex, the OFC, and two subcortical basal ganglia the nucleus accumbens and the the striatum (Figure 4.10 and Table 4). These regions are central in the dopaminergic pathway (described in Chapter 1). Unprocessed foods greatly activated regions extending from the FG, motor cortex, postcentral gyrus and prefrontal cortex (Figure 4.10). Moreover, the extrastriate region V5 was found greatly activated by unprocessed foods.

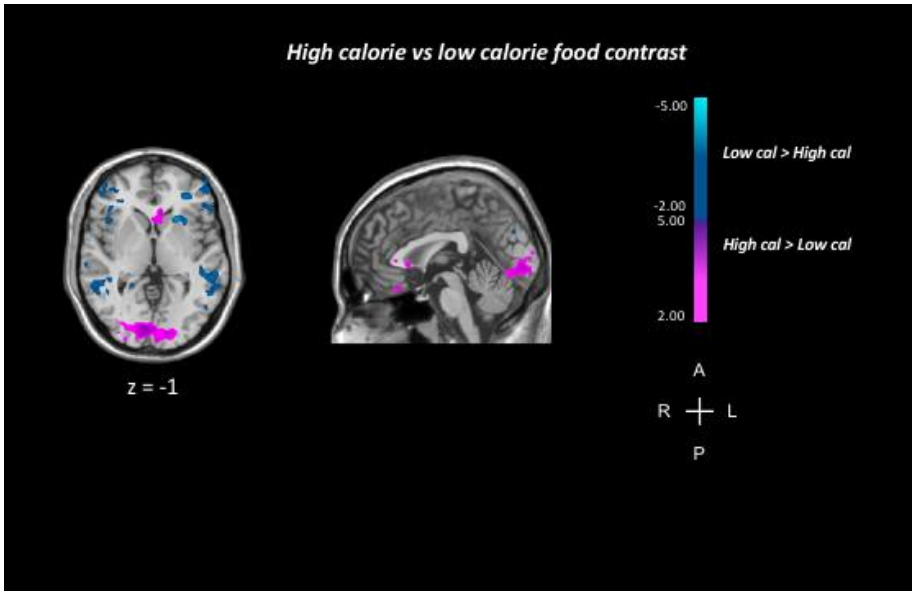


Figure 4.9 Results of the whole-brain contrast between high-calorie and low-calorie food uncorrected at $p < .05$ and a cluster threshold of 100 mm^3.

Table 3 Regions exhibiting differences between high-calorie and low-calorie food, coordinates are in Talairach space.

Location / Side	Peak Coordinates x	y	z	Volume (mm^3)	t - value
High-cal > Low-cal					
Occipital cortex **R**	13	-93	4	976	4.71
Caudate **L**	-3	19	2	551	3.42
Ventral striatum **L**	-9	21	-16	424	3.75
Low-cal > High-cal					
Intraparietal sulcus **L**	-35	-47	44	643	-3.66
Cuneus **L**	-9	-75	30	567	-3.45
Sensorimotor cortex **L**	-35	-25	44	517	-3.15
Premotor **R**	53	7	18	369	-3.13
Posterior Insula **L**	-43	-7	-10	368	-4.04
Anterior Insula **R**	47	-5	0	354	-3.16

Table 4 Regions exhibiting differences between processed and unprocessed food, coordinates are in Talairach space.

Location / Side	Peak Coordinates x	y	z	Volume (mm^3)	t - value
Processed > Unprocessed					
OFC **R**	13	25	-16	460	3.59
Nucleus accumbens **L**	-17	19	-10	275	3.10
Ventral striatum **R**	5	7	-10	251	3.10
OFC **R**	23	25	-16	209	3.58
Occipital cortex **R**	37	-97	10	101	2.98
Unprocessed > Processed					
dlPFC **R**	31	7	58	634	-3.40
Motor cortex **R**	29	-25	66	623	-4.14
Intraparietal sulcus **L**	-31	-55	62	548	-4.26
Occipital cortex **L**	-45	-75	-4	432	-3.53
Postcentral gyrus **R**	21	-45	66	390	-3.31
Premotor cortex **R**	53	-3	42	357	-3.05
mPFC **R**	9	37	46	352	-3.06
Fusiform gyrus **L**	-41	-31	-14	302	-3.16
Insula **R**	49	-3	2	282	-3.09
Intraparietal sulcus **R**	45	-31	36	221	-3.08
Middle temporal gyrus **R**	43	-31	-12	205	-3.20

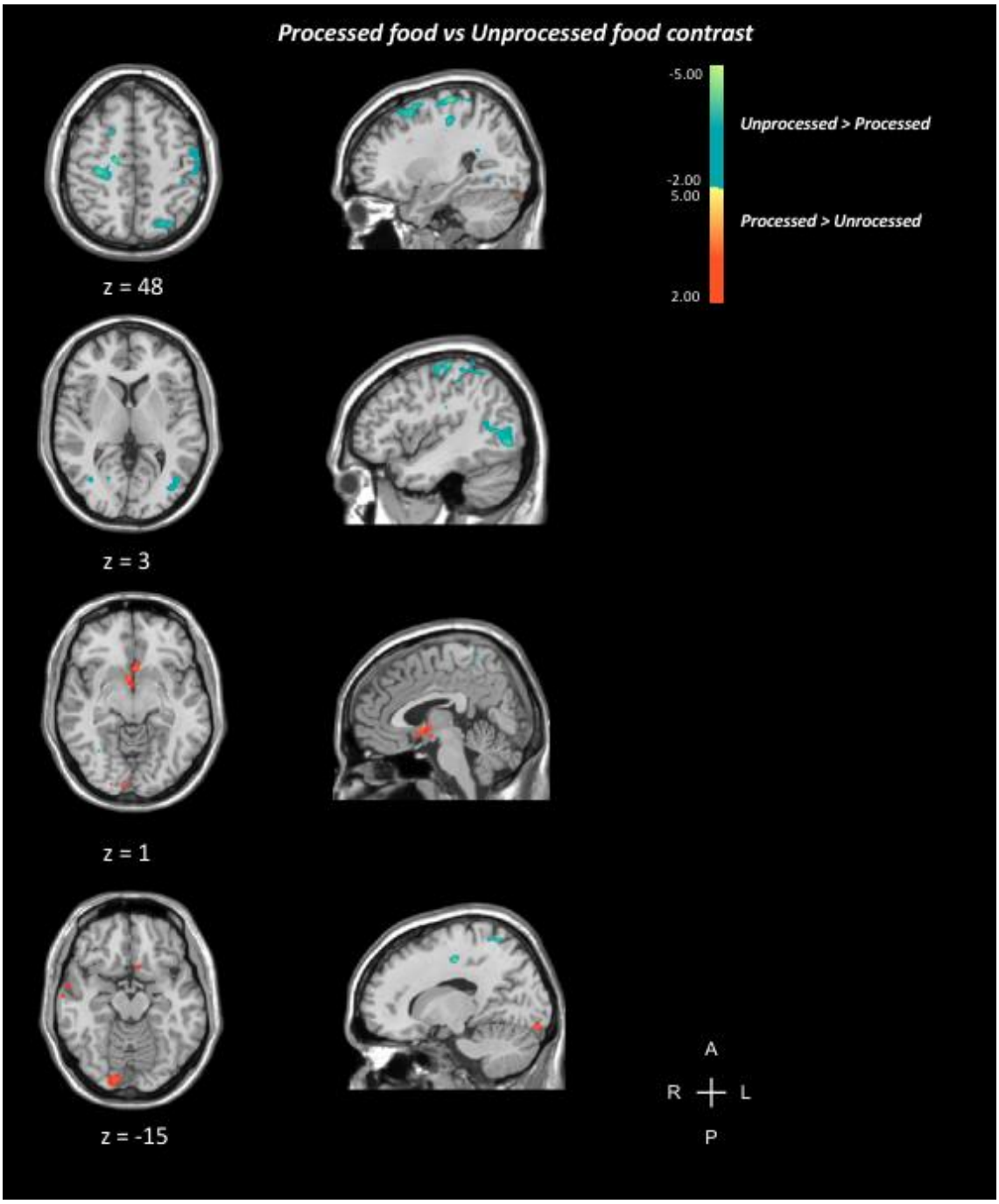


Figure 4.10 Results of the whole-brain contrast between processed and unprocessed food, uncorrected at $p < .05$ and a cluster threshold of 100 mm^3.

4.4 GENERAL DISCUSSION

In the present study, brain responses to foods were investigated using functional magnetic resonance imaging (fMRI) with the aim to disentangle different aspects of this category. As a first step, foods were contrasted to other non-edible objects, such as natural entities (*animals*) and man-made objects used in everyday life. We carefully controlled for real-world object size since this was found to modulate the activations in the occipitotemporal cortex (Konkle & Caramazza, 2013). Therefore, no big animals or big objects were included in our stimuli set since we were interested in food stimuli that are small in the real world.

Preliminary results, showed that the contrast of foods and animals stimuli, produced a medial-to-lateral gradient of responses consistently with previous results (Martin, 2007; Grill-Spector & Weiner, 2014), with animate stimuli activating the lateral portion of the occipitotemporal cortex and the inanimate stimuli the medial portion. The distinction between animate and inanimate objects along the ventral visual stream is here confirmed as a strong predictor of differences in brain responses (Kriegeskorte et al., 2008a; Konkle & Caramazza, 2013).

Interestingly, when contrasting foods to man-made objects, this medial portion (same region of food > animals above) was found to be greatly active for foods compared to objects. This parahippocampal region has already been found to interact with participants' motivational state, being greatly active in response to visual food stimuli in hungry participants (Labar et al., 2001). Additionally, foods greatly activated the posterior insula, the hippocampus and the anterior cingulate, core regions of the food network (Van der Laan et al., 2011; Huerta et al., 2014) as described in the meta-analysis in Chapter 1. On the other hand, objects did not produce any significant cluster in this analysis.

Bilateral parahippocampal gyrus, occipital cortex, anterior cingulate and cerebellum were found to be active when foods were then contrasted with the remaining non-food items (animals, objects and scrambled images), in line with what has been described above. Note that the anterior cingulate has been found to be involved in reward anticipation with food taste stimuli (O'Doherty et al., 2002).

Moving to food sub-categories, stimuli with high- and low-caloric content were contrasted (equally balanced for the level of transformation of the stimuli). A wide portion of the occipital visual cortex, the striatum and vmPFC were greatly active in response to high-calorie foods. While, low calorie foods greatly activated regions extending from the bilateral temporal gyrus, inferior frontal gyrus and the bilateral anterior insula.

In the last analysis, I contrasted processed and unprocessed foods equal in caloric content and I found that processed foods greatly activated the occipital cortex, nucleus accumbens and the striatum. These basal ganglia regions play a central role in the dopamine mesocorticolimbic system (Alonso-Alonso et al., 2015) and, since dopaminergic neurons code for motivational saliency (Berridge et al., 2009), they underlie non-homeostatic mechanisms of food consumption. Therefore, processed foods seem to strongly activate the rewarding system.

On the other hand, unprocessed foods greatly activated the motor cortex, precentral gyrus and prefrontal cortex. This finding is in line with my EEG study result (Chapter 3) in which unprocessed foods elicited greater activity in premotor regions (an additional second fMRI session was designed, and tested on the same participants, see Appendix A4.4, in order to test whether the action involved in food consumption affects brain representations of such foods). To my knowledge this is the first imaging evidence that the brain of healthy normal-weight individuals responds differently to processed and unprocessed food stimuli.

Taken together these preliminary results show how different properties of food stimuli (level of processing and caloric content) differentially modulate brain activations and therefore should

both be taken into account while investigating food perception. As hypothesized for my previous two studies (Chapters 2-3) the ability to discriminate between processed and unprocessed stimuli was expected given the role that cooking had in our evolution (Wrangham et al., 2003). The results of the present study further support the hypothesis that a preference for processed foods is present also in humans (Carmody et al., 2011; Wobber et al., 2008; for animal models) since it greatly activated the reward system compared to unprocessed foods.

For the future, I plan to carry out further analyses on the present data as follows. First, regions of interest (ROIs) will be determined by the food vs ALL contrast; in such regions activations in response to our food categories will be analyzed using Representational Similarity Analysis (RSA, Kriegeskorte et al. 2008b) in order to investigate which factors modulate the activity in each region. Using both, data driven and model driven data, representation dissimilirty matrices (RDMs) and multidimensional scaling (MDS) as in Kriegeskorte (2008a), it will be possible to show which factors (i.e. caloric content, shape, color, level of processing) drive food representations in each region, ranging from early perceptual occipital regions to higher order prefrontal regions.

CHAPTER 5

GENERAL DISCUSSION

Food represents one of the most rewarding stimulus present in nature since it is necessary for our survival. In Western societies, we are overwhelmed by the abundance and the availability of a huge variety of food types ranging from unprocessed to heavily processed foods. Everyday, we front decisions regarding foods guided by an energetic necessity (homeostatic mechanisms) or a pleasure drive (hedonic or reward-based mechanisms). Evolution shaped our brain in order to respond to food cues in a time in which food was scarse, and this would explain why we rapidly and efficiently extract information regarding foods by visually inspecting them. However nowadays the abundance of food cues obliges us to refrain ourselves from ingesting an excessive daily caloric intake. The epidemic increase in the prevalence of obesity should encourage neuroscientists to contribute to the study of the mechanisms underlying this excessive consume of highly fat foods. I argue that to identify the mechanisms that guide food perception in normal-weight individuals represents the first step towards the understanding of what goes wrong in obesity. To date, there are several studies that aimed at identifying regions that are active while participants perceive, smell and taste foods, and factors that modulate these activations (see e.g. Huerta et. al, 2014). In my thesis, I focused on the mechanisms that guide us in extracting information about foods from visual food cues.

In Chapter 1, I conducted a meta-analysis on the imaging studies in which participants viewed foods compared to other non-edible objects. Results showed how simply viewing food pictures activates brain regions extending from the early visual cortex to the prefrontal cortex and basal ganglia. Moreover, studies conducted using EEG, showed that humans are able to extract

information about food properties such as edibility and caloric content, very rapidly and efficiently (see Tsourides et al., 2016 and Toepel et al., 2009).

However, one dimension of food that has received little attention to date is the unprocessed/processed food distinction, although animal research (Carmody et al., 2011; Wobber et al., 2008) suggests a central role of such dimension in our evolution (Wrangham et al., 2003). The *Cooking Hypothesis* (Wrangham et al., 1999) maintains that a cooked diet was responsible for morphological changes of the human body during evolution, it increased the energy gain and food value, and it freed time for the hominids to engage in activities other than gathering and chewing food. Moreover, it reduces the probability of infections while it increases food palatability and consumption. Chimps, bonobos, gorillas and orangutans tend to prefer their food cooked, from tubers to meat (Wobber et al., 2008) and early hominids may have cooked their food soon after controlling of fire, as they were already inclined to prefer items prepared in this way. Whether this preference left a neural signature in the human brain is to date unknown and my thesis tried to fill this gap. With the goal of answering this question, I have conducted three studies using different techniques investigating *when*, *how* and *where* the brain detects the processed/unprocessed distinction in foods. Moreover, processed/cooked foods lead to a higher net energy gain once ingested and therefore it is of great interest to investigate how such dimension interacts and differentiates from the largely studied high/low calorie food dimension.

In my studies, I have employed the food images FRIDa (Foroni et al., 2013) that have been validated with participants rating each image on various dimensions (i.e. valence, arousal, perceived caloric content etc.). Most importantly participants were asked to which extent they perceived the processing level of each stimulus, thus sorting all foods in unprocessed and processed. Therefore, in my first behavioral study I could select a set of images with differed levels of transformation, but balanced for caloric content, valence, arousal and brightness, with the aim to investigate *how* the unprocessed/processed dimension affected participants' implicit and

explicit evaluations of food (Chapter 2). This study showed how different aspects of food evaluation (i.e. valence, healthiness) kick-in depending on the time available to perform the task, and on the task itself, being implicit or explicit (see Sullivan et al., 2015). I found that participants expressed different evaluations depending on the type of food and on the amount of time they had available to build up their preference. Perceivers' characteristics such as their dieting habits (assessed through the Restraint Scale) and their body mass index (BMI) were also found to interact with the food-type both at the implicit and explicit level.

Results from my study are accommodated within the general theory according to which explicit attitudes reflect deliberate behaviors (affected by self-presentation strategies) while implicit attitudes are linked to impulsive actions (see Fazio & Olson, 2003). This is a new finding since previous studies on implicit and explicit components of behavior (Czyzewska et al., 2011 for a review) addressed only the distinction between high- and low-calorie foods (Roefs & Jansen, 2002; Czyzewska & Graham, 2008).

The resulting differences between unprocessed and processed foods of this first behavioral study, led to the development of an electroencephalography (EEG) study that aimed at investigating whether the human brain is able to track the difference between unprocessed and processed foods (Chapter 3). The EEG technique allowed me to respond to the question of *when* (i.e. in which time window) this dimension becomes relevant in food perception. The original experimental design of Toepel and colleagues (2009a), who found that the brain tracks the difference between high and low-fat foods as early as at 160 ms post-stimuli presentation during a simple food/non-food categorization task, was adapted to our set of stimuli. Results from my EEG study showed that differences between unprocessed and processed foods emerged at the electrode level as early as 130 ms post stimulus-onset. This result extends previous findings by Toepel et al. (2009a) and Meule et al. (2013) on an implicit within-category discrimination of foods based on fat/caloric content around 150 ms post-stimulus presentation. Interestingly the time window in which the

discrimination between unprocessed and processed foods emerged in my study overlaps with the time window in which the natural vs artificial discrimination has been observed in a recent MEG study (Cichy et al., 2014). Whether the discrimination between processed/unprocessed food might be a particular instance of the more general living vs. man-made entities discrimination remains to be understood fully.

Moreover, I observed differences in the strength of the electric field in response to the processed/unprocessed foods, as well as differences in the topographies in two distinct time windows. These differences in topographies were generated by different activations in distributed sources. Results suggest that processed foods, like other highly relevant stimuli, greatly activated the ventral visual pathway. In contrast, unprocessed foods activated premotor and inferior frontal regions. This latter finding, can be explained in light of the fact that such foods require to plan and perform some actions in order to be eaten. This, triggers an interesting and novel question in food perception, namely whether the actions that are required in order to eat a food are somewhat represented in the food concept. To my knowledge, this was the first study in which differential brain responses have been reported when categorizing unprocessed and processed food stimuli.

Similarly, Pergola and colleagues (2017) presented a "prime" sentence describing a sensory property (i.e. "it tastes sweet") or a functional property (i.e. "it is suitable for a wedding") of food, followed by a target picture of unprocessed or processed food. Prime-target pairs were judged as congruent or incongruent by participants. The authors found that prime-target incongruency modulated brain responses (N400 component) consistently with the sensory-functional theory (SFT, Warrington & Shallice, 1984; Warrington & McCarthy, 1987) and that these modulations interacted with participants' BMI. Taken together, the two EEG studies addressed different aspects, elicited by the differences in the tasks, of the neural signature of the unprocessed and processed foods dimension, the common ground being a modulation of brain responses that depend of the level of processing of the food stimuli.

With the third and final study, I aimed to answer to the question about *where* the distinction between processed and unprocessed foods takes place in the brain using fMRI (Chapter 4). The main results of this study are the following. First, in line with the results of my EEG study (Chapter 3), unprocessed foods seem to activate regions involved in motor production and motor imagery. This finding could be driven by the fact that actions have to be performed on such foods in order to be eaten (i.e., peeling). As discussed above, this result deserved to be further investigated as it opens up to interesting questions concerning the way in which action representations and food concepts are related to each other.

Second, processed foods were found to greatly activate basal ganglia as part of the dopaminergic system which codes for motivational saliency (Berrige et al., 2009) and, as such, strongly associated with the reward system. This finding is in line with the hypothesis that the preference for processed/cooked foods has been left also in the human brain, consistently with what has been found in mice (Carmody et al., 2011) and apes (Wobber et al., 2008).

Interestingly, the results of my fMRI study pointed out once again that the processed/unprocessed dimension is distinct from the high/low calorie dimension. Therefore, the level of food transformation should no longer be neglected in future studies using food as stimuli.

Taken together, the results from my three studies suggest that the unprocessed/processed dimension is relevant in building implicit/explicit evaluations of foods and in extracting information from visually presented foods (Chapters 3 and 4).

In the studies I have carried out and included in my thesis, all participants were omnivores, normal-weight, and with no signs of aberrant eating behaviors. However, it would be of great interest to extend my investigation to populations that violate the normal weight body mass index (underweight, overweight and obese). Likewise, it would also be interesting to test deliberate raw-foodists who do not include cooked foods in their diets in order to investigate to which extent the neural signature of the processed/unprocessed is present.

To conclude, the findings of the present thesis suggest that the level of processing is a key factor in developing food representation. From a theoretical point of view these studies can enrich the present literature on food processing. Further research will be necessary to better understand the relationship between our food sub-categories and other semantic categories.

APPENDIX

A1 CHAPTER 1

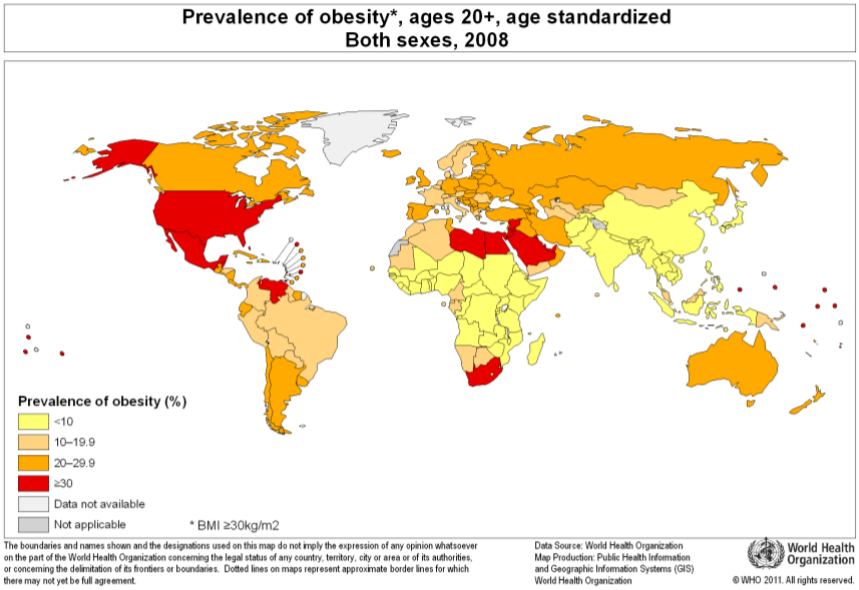


Figure A1.1 *Prevalence of obesity worldwide, from WHO report 2008.* For an interactive map of the report of the data of 2014: http://www.who.int/gho/ncd/risk_factors/overweight/en/ .

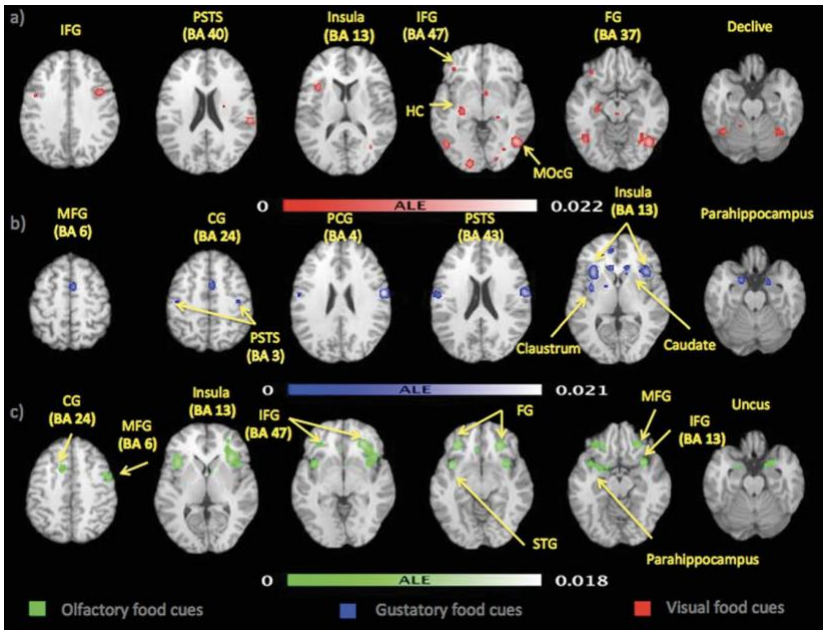


Figure A1.2 Results of the meta-analysis of Huerta et al. (2014). * Figure from: "Neural bases of food perception: Coordinate-based meta-analyses of neuroimaging studies in multiple modalities". *Obesity*, *22*(6).

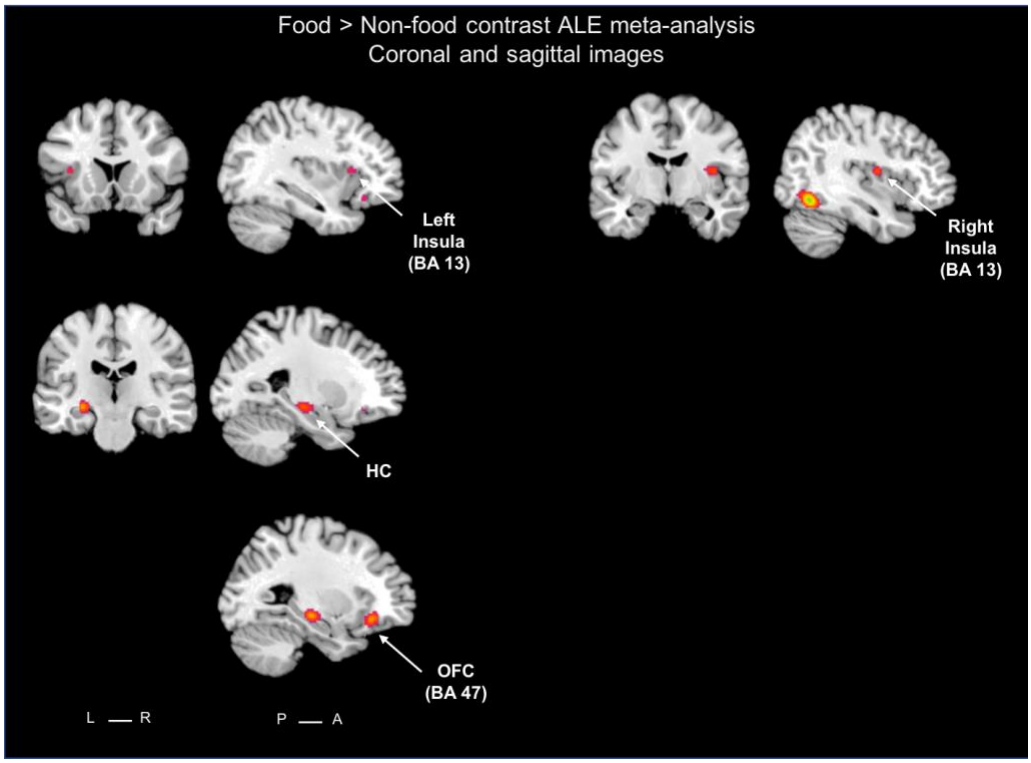


Figure A1.3 Coronal and sagittal images of the ALE meta-analysis results of the food vs non-food contrast.

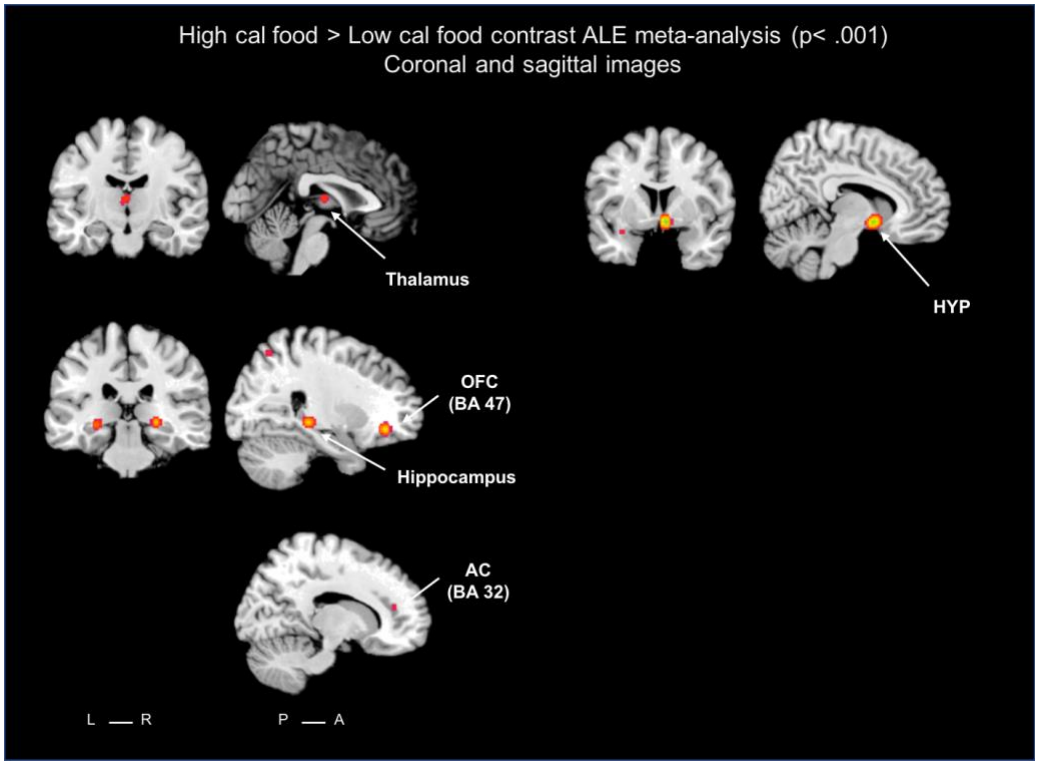


Figure A1.4 Coronal and sagittal images of the ALE meta-analysis results of the high-calorie vs low-calorie food contrast ($p < .001$).

A2 CHAPTER 2

The Restraint Scale

1. How often are you dieting?
Never; rarely, sometimes, often, always (Scored 0–4)
2. What is the maximum amount of weight (in kilos) you have ever lost within 1 month?
(0–2.5; 2.5–5; 5–7.5; 7.5–10; 10+ (Scored 0–4))
3. What is the maximum amount of weight gain (in kilos) within a week?
(0–0.5; 0.5–1; 1–1.5; 1.5–2.5; 2.5+ (Scored 0–4))
4. In a typical week, how much does your weight fluctuate?
(0–0.5; 0.5–1; 1–1.5; 1.5–2.5; 2.5+ (Scored 0–4))
5. Would a weight fluctuation of 2.5 kilos affect the way you live your life?
Not at all; slightly, moderately; very much (Scored 0–3)
6. Do you eat sensibly in front of others and splurge alone?
Never; rarely, often, always (Scored 0–3)
7. Do you give too much time and thought to food?
Never, rarely, often; always (Scored 0–3).
8. Do you have feelings of guilt after overeating?
Never, rarely, often, always (Scored 0–3).
9. How conscious are you what you are eating?
Not at all; slightly, moderately, extremely (Scored 0–3)
10. How many kilos over your desired weight were you at your maximum weight?
(0–0.5; 0.5–3; 3–5; 5–10; 10+ (Scored 0–4).

A2.1 Restraint Scale (Herman & Polivy, 1980)

A3 CHAPTER 3

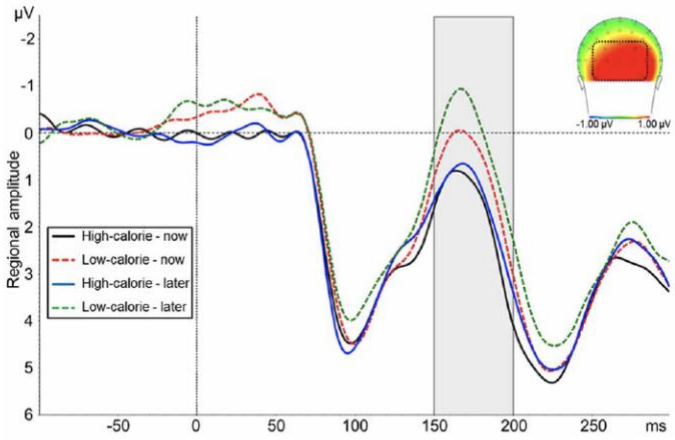


FIGURE 5 | Mean amplitude of pooled ERPs of a posterior cluster (PO9, PO7, PO3, POz, PO4, PO8, PO10, O1, Oz, O2). The headplot shows the difference between trials with high-calorie minus low-calorie food pictures in a time window between 150–200 ms.

Figure A3.1 Results at the electrode level of Meule et al., (2013). *Figure from: "Time course of electrocortical food-cue responses during cognitive regulation of craving". *Frontiers in Psychology*, *4.*

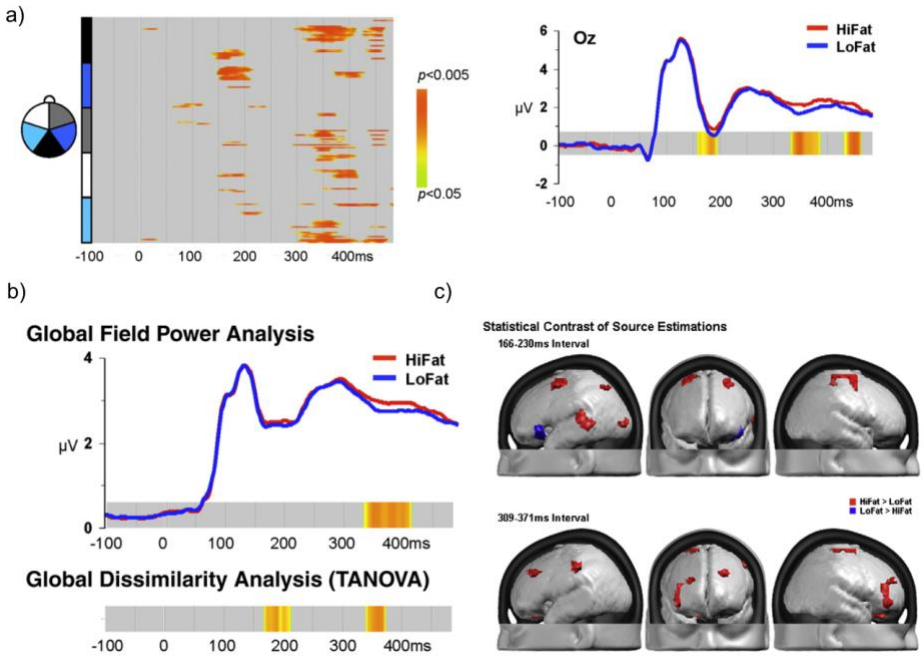
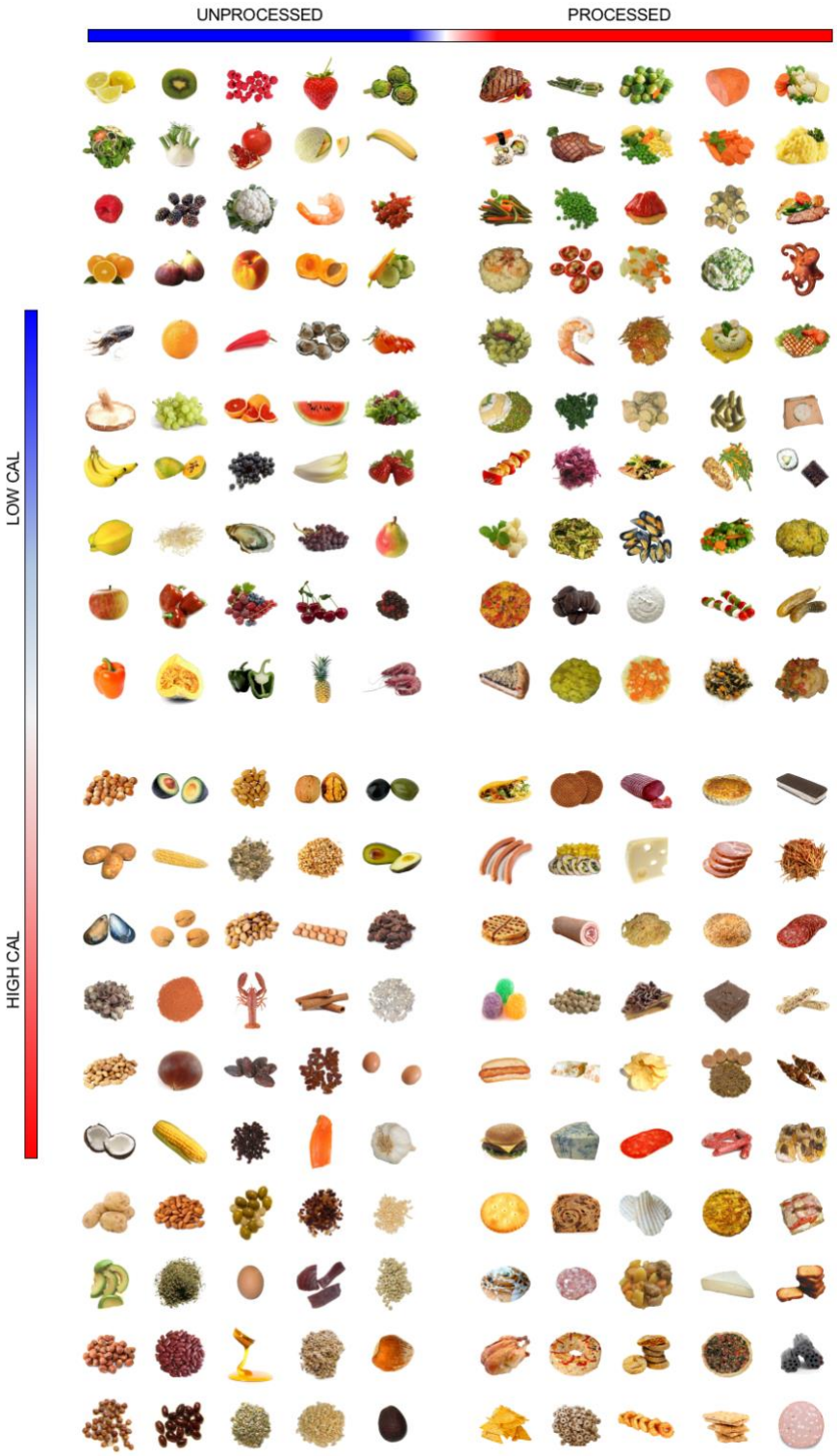


Figure A3.2 Results a) at the electrode level; b) GFP and GMD;c) Source estimation of ,Toepel et al. (2009). Figure from:

a)



b)

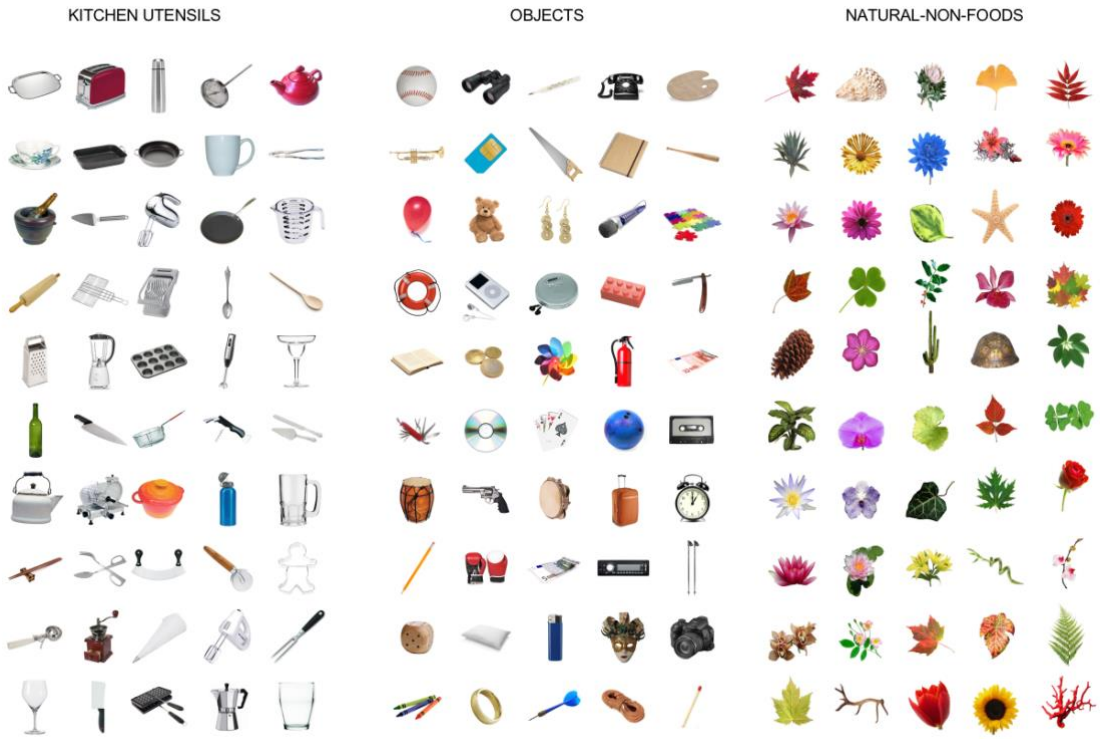
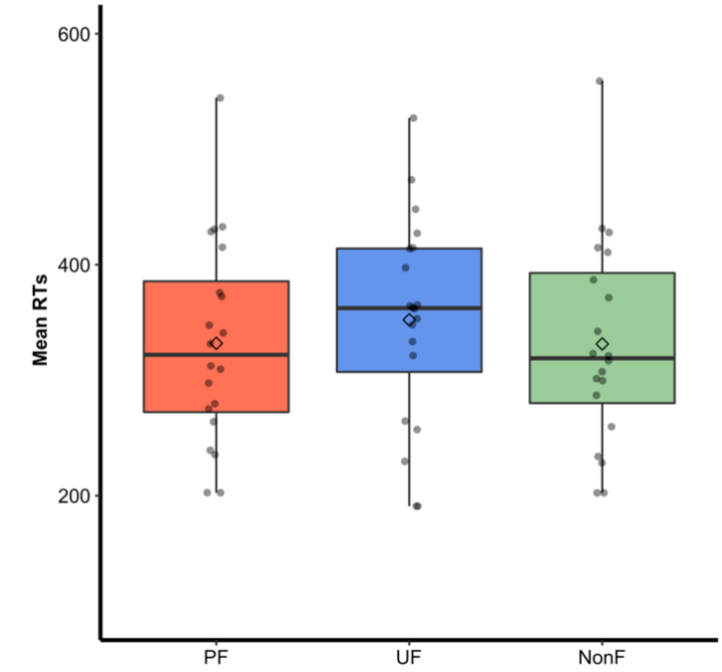
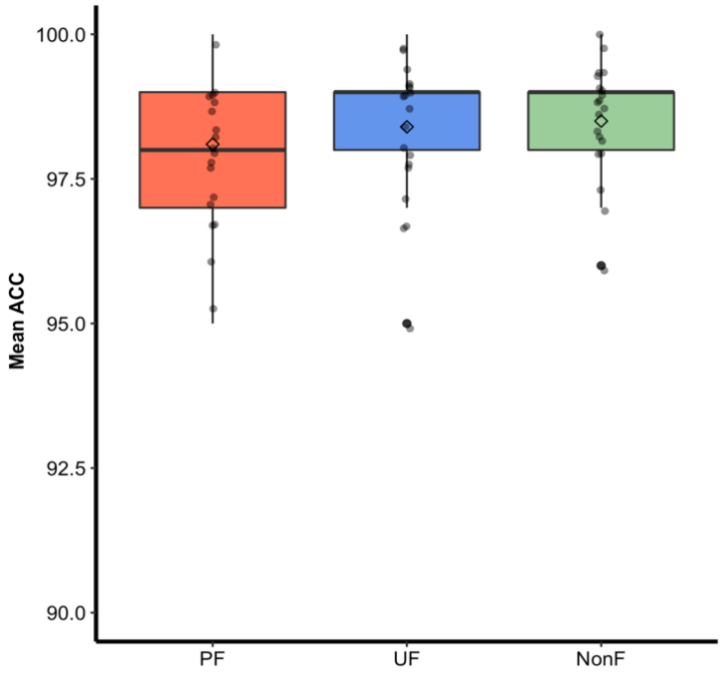


Figure A3.3 The complete set of stimuli used in the experiment are displayed here. a) Food stimuli b)Non-food stimuli

a)


600
400
200
Mean RTs
PF
UF
NonF


100.0
97.5
95.0
92.5
90.0
Mean ACC
PF
UF
NonF

b)

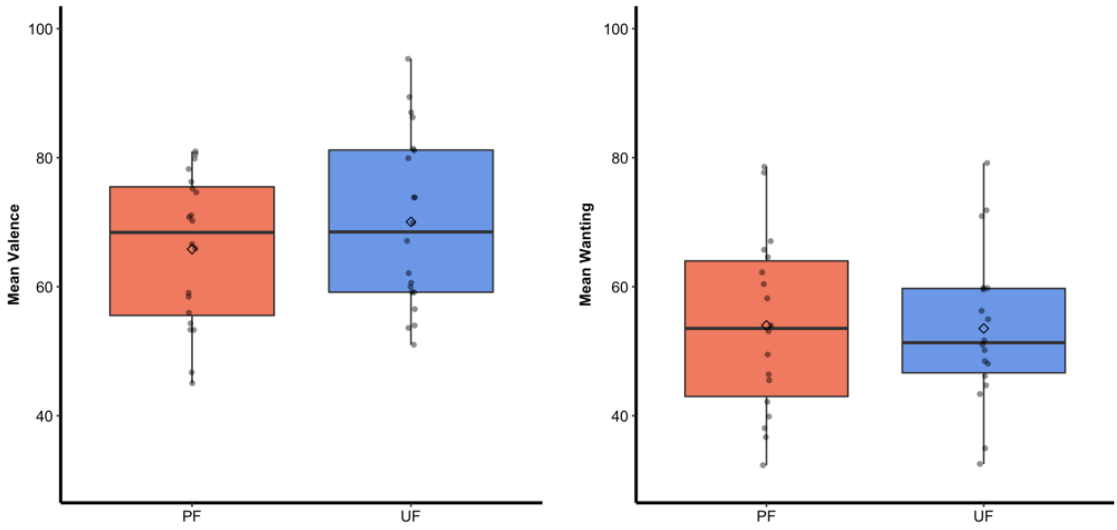


Figure A3.4 Behavioral results a) Mean RTs and Accuracy b) Ratings

A3.5 Global electric field analyses: Topographic clustering analysis

The post-stimulus periods of the VEPs in response to each stimulus condition were collectively submitted to a hierarchical clustering algorithm (Murray et al., 2008). This analysis is based on the observation that topographic maps remain stable for several milliseconds and then switch to a novel configuration that remains stable again (functional microstates, Lehmann et al., 1971; Brunet et al., 2011). The optimal number of maps (i.e. the minimal number of maps that accounts for the greatest variance of the data) was determined using a modified Krzanowski-Lai criterion (Murray et al. 2008). Differences in the patterns of maps observed for the group-averaged data were tested statistically by comparing each map with the moment-by-moment scalp topography of individual participant's VEPs ("fitting" procedure, see Toepel et al., 2009). As output, the occurrence and duration of template maps over post-stimulus periods were assessed and statistically analyzed.

To better understand the topographic differences, the cumulative group-averaged data across conditions were analyzed with a hierarchical cluster analysis. Thirteen different scalp topography maps over the post-stimulus period accounted for 99.04% of the global explained variance (GEV). The sequence of maps was generally identical between the two conditions, except over the 158-207 ms time period, when different maps were identified in the group-average VEP data from each condition (Figure 3.6a). Single time-point data over this time interval were submitted to a fitting procedure based on the spatial correlation with the map identified in the group-averaged data as described above (see also Murray et al., 2008). The dependent measure was the duration over which a given map (arbitrarily named Map A, Map B and Map C) identified in the group-averaged data better correlated spatially with an individual subject's data over the 158-207ms time period (Figure 3.6b). A significant interaction was observed between map and condition over the 158-207 ms time interval ($F(1, 19) = 4.24$, $p = 0.030$). Paired t-tests showed

that Map C lasted longer (is more representative) for processed foods while Map A is more representative for unprocessed foods.

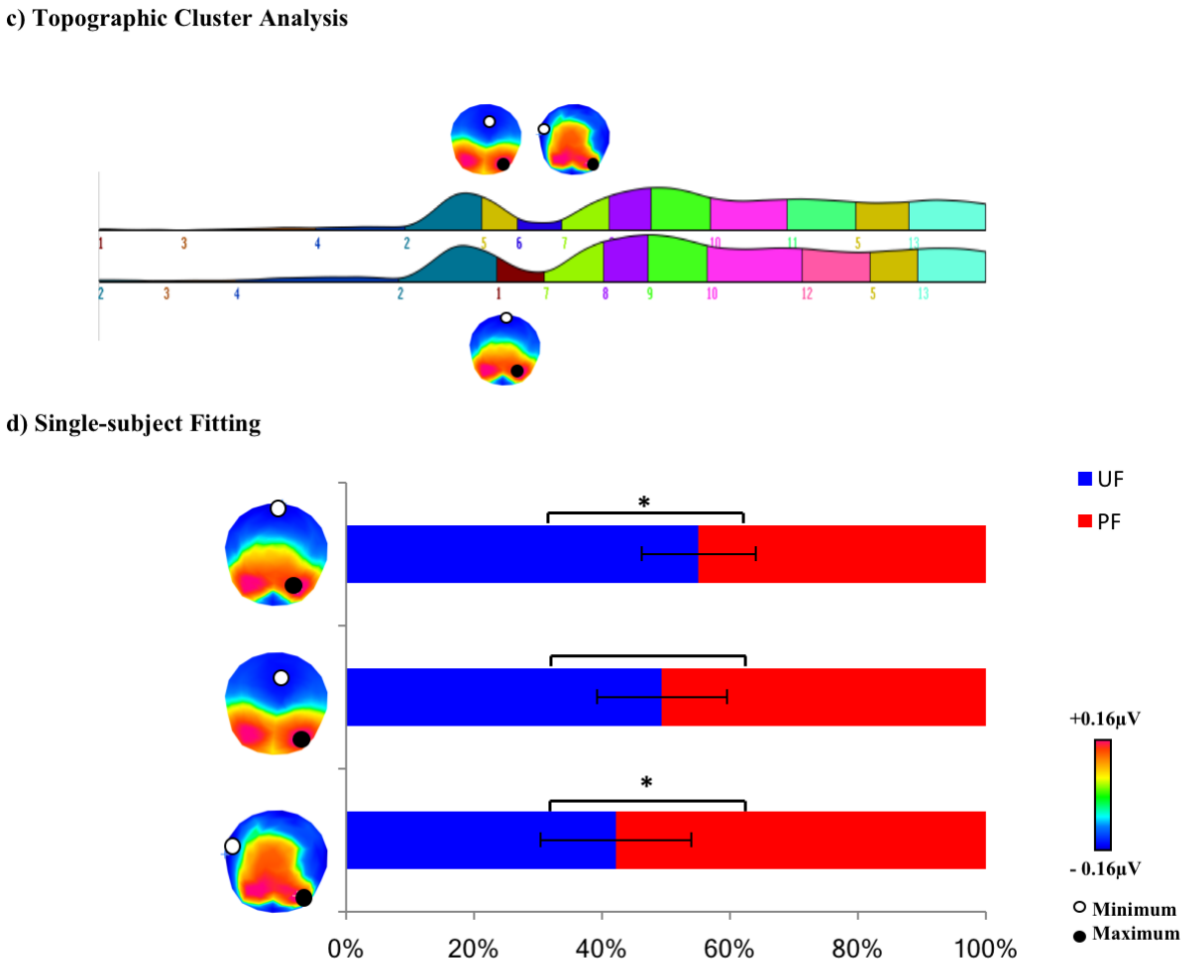


Figure A3.5 Topographic clustering analysis revealed thirteen time periods of stable electric field topographies in the group-averaged VEPs. Results of the fitting procedure on the x-axis map durations are expressed in percentages.

APPENDIX

A4 CHAPTER 4

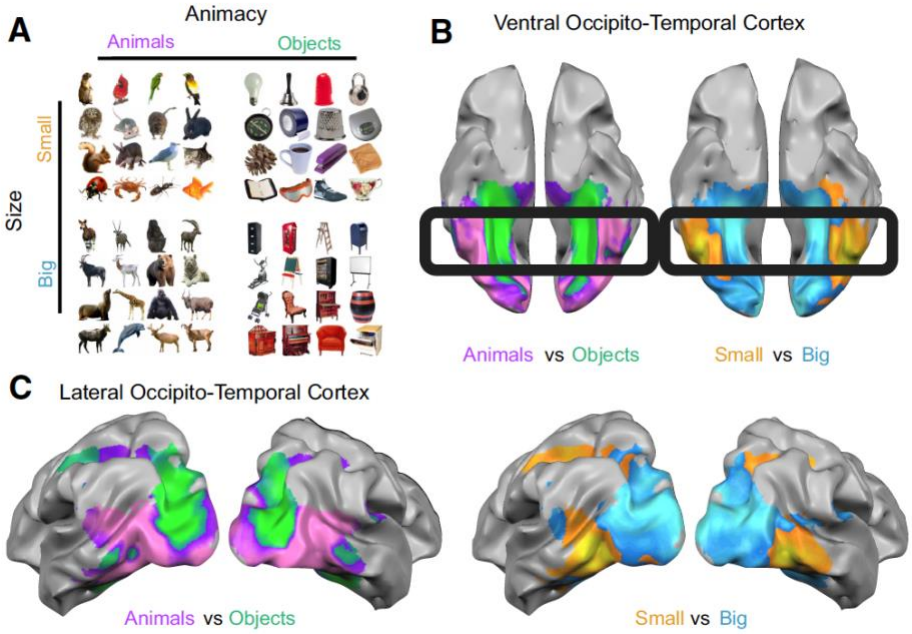


Figure A4.1 Konkle & Caramazza (2013) stimuli and results.

* Figure from: "Tripartite organization of the ventral stream by animacy and object size". *Journal of Neuroscience*, *33*(25) p. 10237.

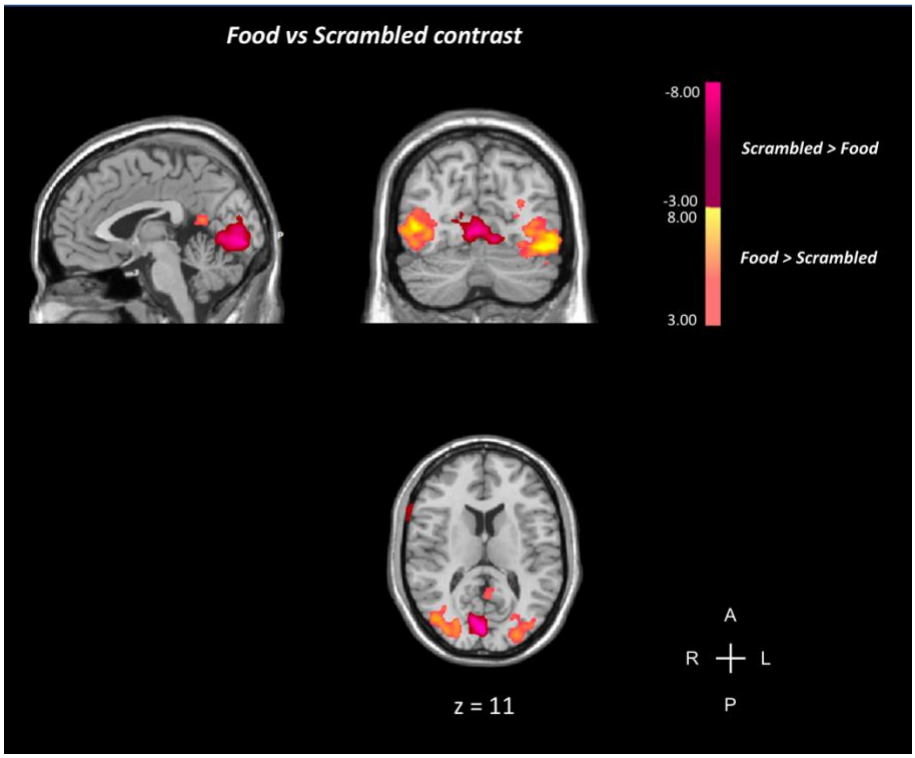


Figure A4.2 Whole-brain results of the contrast Food vs Scrambled in the localizer runs, FDR corrected results at $p < .05$ and a 50 mm^3 cluster threshold.

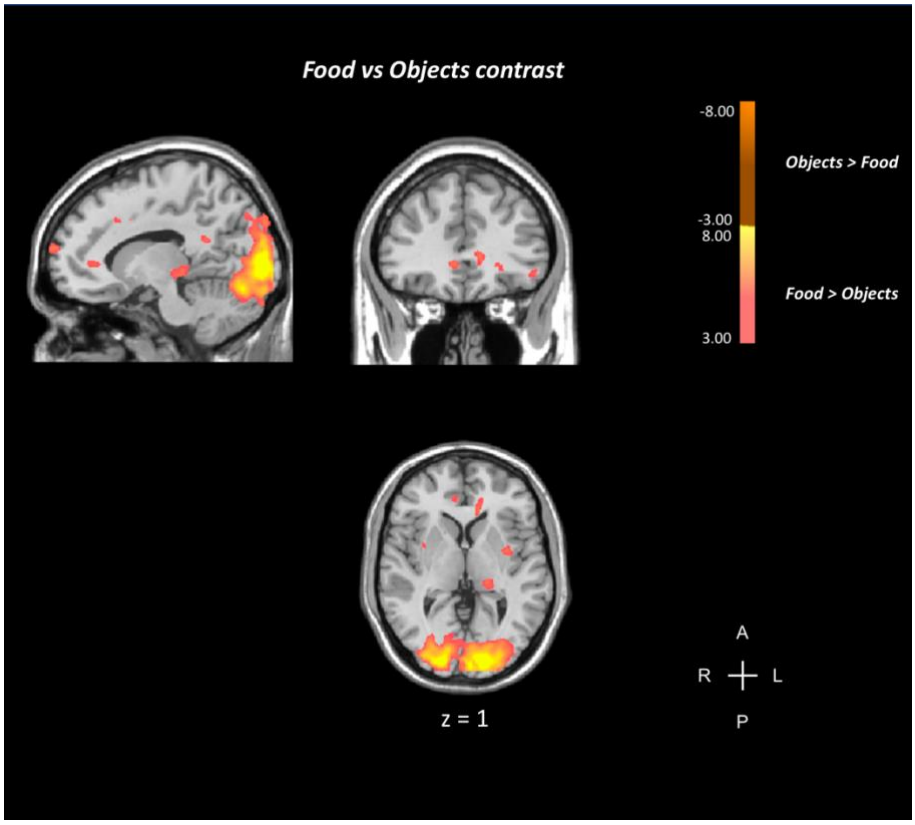


Figure A4.3 Whole-brain results of the contrast Food vs Objects in the localizer runs, FDR corrected results at $p < .05$ and a 50 mm^3 cluster threshold.

A4.4 Session 2 fMRI

The same participants agreed to take part in a second fMRI session, which aim was to investigate whether the way we eat foods (action performed) modulates our brain representations towards such foods. This study was inspired by the finding of our EEG study (Chapter 3) in which we found that the premotor cortex was active in response to natural foods on which more actions were necessary in order to be eaten, and by the study of Bracci et al. (2012) in which they found overlapping representations of hands and hand objects in lateral occipital cortex (LOC).

To respond to our experimental question, we selected images of foods that are generally ate in a particular way (1 hand, 2 hands, spoon, fork, chopsticks or knife, see Figure A4.3) we also

presented body parts, objects and kitchen utensils to subjects in a rapid event-related design. Participants in the scanner performed a one-back task.

Figure A4.4 Experimental stimuli used during the fMRI scanning session.

REFERENCES

Alonso-Alonso, M., Woods, S. C., Pelchat, M., Grigson, P. S., Stice, E., Farooqi, S., & Beauchamp, G. K. (2015). Food reward system: current perspectives and future research needs. *Nutrition reviews*, *73*(5), 296-307.

Antal, A., Keri, S., Kovacs, G., Janka, Z., & Benedek, G. (2000). Early and late components of visual categorization: an event-related potential study. *Cognitive Brain Research*, *9*, 117-119.

Avery, J. A., Kerr, K. L., Ingeholm, J. E., Burrows, K., Bodurka, J., & Simmons, W. K. (2015). A common gustatory and interoceptive representation in the human mid-insula. Human brain mapping, 36(8), 2996-3006.

Bartholdy, S., Dalton, B., O'Daly, O. G., Campbell, I. C., & Schmidt, U. (2016). A systematic review of the relationship between eating, weight and inhibitory control using the stop signal task. *Neuroscience & Biobehavioral Reviews*, *64*, 35-62.

Beaver, J. D., Lawrence, A. D., van Ditzhuijzen, J., Davis, M. H., Woods, A., & Calder, A. J. (2006). Individual differences in reward drive predict neural responses to images of food. *The Journal of Neuroscience*, *26*, 5160-5166.

Beck, A. T., Steer, R. A., & Brown, G. K. (1996). Beck depression inventory-II. *San Antonio, TX*, 78204-2498.

Berridge, K. C., Robinson, T. E., & Aldridge, J. W. (2009). Dissecting components of reward:'liking','wanting', and learning. *Current opinion in pharmacology*, *9*(1), 65-73.

Berthoud, H. R. (2011). Metabolic and hedonic drives in the neural control of appetite: who is the boss?. *Current opinion in neurobiology*, *21*(6), 888-896.

Bicchieri, C. (2006). *The Grammar of Society: the Nature and Dynamics of Social Norms*, New York: Cambridge University Press.

Blechert, J., Meule, A., Busch, N. A., & Ohla, K. (2014). Food-pics: an image database for experimental research on eating and appetite. *Frontiers in Psychology, 5*, 40-49.

Bracci, S., Cavina-Pratesi, C., Ietswaart, M., Caramazza, A., & Peelen, M. V. (2012). Closely overlapping responses to tools and hands in left lateral occipitotemporal cortex. *Journal of neurophysiology*, *107*(5), 1443-1456.

Brooks, S. J., Cedernaes, J., & Schiöth, H. B. (2013). Increased prefrontal and parahippocampal activation with reduced dorsolateral prefrontal and insular cortex activation to food images in obesity: a meta-analysis of fMRI studies. *PloS one*, *8*(4), e60393.

Brunet, D., Murray, M. M., & Michel, C. M. (2011). Spatiotemporal analysis of multichannel EEG: CARTOOL. *Computational Intelligence and Neuroscience*, *2011*, 1-15.

Burger, K. S., & Stice, E. (2011). Relation of dietary restraint scores to activation of reward-related brain regions in response to food intake, anticipated intake, and food pictures. *Neuroimage*, *55*(1), 233-239.

Calder, A. J., Beaver, J. D., Davis, M. H., Van Ditzhuijzen, J., Keane, J., & Lawrence, A. D. (2007). Disgust sensitivity predicts the insula and pallidal response to pictures of disgusting foods. *European Journal of Neuroscience*, *25*(11), 3422-3428.

Capitani, E., Laiacona, M., Mahon, B., & Caramazza, A. (2003). What are the facts of semantic category-specific deficits? A critical review of the clinical evidence. *Cognitive Neuropsychology*, *20*, 213-261.

Carmody, R. N., & Wrangham, R. W. (2009a). Cooking and the human commitment to a high-quality diet. In *Cold Spring Harbor symposia on quantitative biology*. Cold Spring Harbor Laboratory Press.

Carmody, R. N., & Wrangham, R. W. (2009b). The energetic significance of cooking. *Journal of Human Evolution*, *57*(4), 379-391.

Carmody, R. N., Weintraub, G. S., & Wrangham, R. W. (2011). Energetic consequences of thermal and nonthermal food processing. *Proceedings of the National Academy of Sciences*, *108(48)*, 19199-19203.

Carota, F., Kriegeskorte, N., Nili, H., & Pulvermüller, F. (2017). Representational Similarity Mapping of Distributional Semantics in Left Inferior Frontal, Middle Temporal, and Motor Cortex. *Cerebral Cortex*, *27*, 294-309.

Chao, L. L., Haxby, J. V., & Martin, A. (1999). Attribute-based neural substrates in temporal cortex for perceiving and knowing about objects. *Nature Neuroscience*, *2*, 913-919.

Churchland, M. M., Byron, M. Y., Ryu, S. I., Santhanam, G., & Shenoy, K. V. (2006). Neural variability in premotor cortex provides a signature of motor preparation. *Journal of Neuroscience*, *26*, 3697-3712.

Cichy, R. M., Pantazis, D., & Oliva, A. (2014). Resolving human object recognition in space and time. *Nature neuroscience*, *17*(3), 455-462.

Coletta, M., Platek, S., Mohamed, F. B., van Steenburgh, J. J., Green, D., & Lowe, M. R. (2009). Brain activation in restrained and unrestrained eaters: an fMRI study. *Journal of abnormal psychology*, *118*(3), 598.

Corbetta, M., & Shulman, G. L. (2002). Control of goal-directed and stimulus-driven attention in the brain. *Nature Reviews Neuroscience*, *3*, 201-215.

Cornier, M. A., Von Kaenel, S. S., Bessesen, D. H., & Tregellas, J. R. (2007). Effects of overfeeding on the neuronal response to visual food cues. *The American journal of clinical nutrition*, *86*(4), 965-971.

Critchley, H. D., & Rolls, E. T. (1996). Hunger and satiety modify the responses of olfactory and visual neurons in the primate orbitofrontal cortex. *Journal of neurophysiology*, *75*(4), 1673-1686.

Cummings, D. E., Purnell, J. Q., Frayo, R. S., Schmidova, K., Wisse, B. E., & Weigle, D. S. (2001). A preprandial rise in plasma ghrelin levels suggests a role in meal initiation in humans. *Diabetes*, *50*(8), 1714-1719.

Czyzewska, M., & Graham, R. (2008). Implicit and explicit attitudes to high-and low-calorie food in females with different BMI status. *Eating behaviors*, *9*(3), 303-312.

Czyzewska, M., Graham, R., & Ceballos, N. A. (2011). Explicit and implicit attitudes to food. In *Handbook of Behavior, Food and Nutrition* (pp. 673-692). Springer New York.

Dehaene, S., & Cohen, L. (2011). The unique role of the visual word form area in reading. *Trends in cognitive sciences*, *15*(6), 254-262.

De Lucia, M., Clarke, S., & Murray, M. M. (2010). A temporal hierarchy for conspecific vocalization discrimination in humans. *Journal of Neuroscience*, *30*, 11210-11221.

De Silva, A., Salem, V., Matthews, P. M., & Dhillo, W. S. (2012). The use of functional MRI to study appetite control in the CNS. *Experimental diabetes research*, *2012*.

Del Parigi, A., Chen, K., Gautier, J. F., Salbe, A. D., Pratley, R. E., Ravussin, E., & Tataranni, P. A. (2002). Sex differences in the human brain's response to hunger and satiation. *The American Journal of Clinical Nutrition*, *75(6)*, 1017-1022.

di Pellegrino, G., Magarelli, S., & Mengarelli, F. (2010). Food pleasantness affects visual selective attention. *The Quarterly Journal of Experimental Psychology*, *64*(3), 560-571.

Di Russo, F., Martínez, A., Sereno, M. I., Pitzalis, S., & Hillyard, S. A. (2002). Cortical sources of the early components of the visual evoked potential. *Human brain mapping*, *15*(2), 95-111.

Eger, E., Jedynak, A., Iwaki, T., & Skrandies, W. (2003). Rapid extraction of emotional expression: evidence from evoked potential fields during brief presentation of face stimuli. *Neuropsychologia*, *41*, 808-817.

Eickhoff, S. B., Bzdok, D., Laird, A. R., Kurth, F., & Fox, P. T. (2012). Activation likelihood estimation meta-analysis revisited. *Neuroimage*, *59*(3), 2349-2361.

Ely, A. V., Childress, A. R., Jagannathan, K., & Lowe, M. R. (2014). Differential reward response to palatable food cues in past and current dieters: A fMRI study. *Obesity*, *22*(5).

Farooqi, I. S., Bullmore, E., Keogh, J., Gillard, J., O'rahilly, S., & Fletcher, P. C. (2007). Leptin regulates striatal regions and human eating behavior. *Science*, *317*(5843), 1355-1355.

Fazio, R. H., & Olson, M. A. (2003). Implicit measures in social cognition research: Their meaning and use. *Annual Review of Psychology*, *54(1)*, 297-327.

Ferro-Luzzi, A., Sette, S., Franklin, M., & James, W. P. (1992). A simplified approach of assessing adult chronic energy deficiency. *European Journal of Clinical Nutrition*, *46*(3), 173-186.

Finlayson, G., & Dalton, M. (2012). Hedonics of food consumption: are food 'liking' and 'wanting' viable targets for appetite control in the obese?. *Current Obesity Reports*, *1(1)*, 42-49.

Foerde, K., Steinglass, J. E., Shohamy, D., & Walsh, B. T. (2015). Neural mechanisms supporting maladaptive food choices in anorexia nervosa. *Nature neuroscience*, *18*(11), 1571-1573.

Foroni, F., Pergola, G., Argiris, G., & Rumiati, R. I. (2013). The FoodCast research image database (FRIDa). *Frontiers in Human Neuroscience*, *7*, 1-19.

Foroni, F., Rumiati, R.I., Coricelli, C. & Ambron, A. (2016a). A bait that we cannot avoid: Food-induced motor distractibility. *Brain and Cognition*,*110*, 74-84.

Foroni, F., Pergola, G., & Rumiati, R.I. (2016b). Food color is in the eye of the beholder: the role of human trichromatic vision in food evaluation. *Scientific Reports*, 6, 37034.

Foroni, F., & Rumiati, R. I. (2017). Food Perception and Categorization: From Food/No-Food to Different Types of Food. In *Handbook of Categorization in Cognitive Science (Second Edition)* (pp. 271-287).

Frank, S., Laharnar, N., Kullmann, S., Veit, R., Canova, C., Hegner, Y. L., & Preissl, H. (2010). Processing of food pictures: influence of hunger, gender and calorie content. *Brain research*, *1350*, 159-166.

Friese, M., Hofmann, W., & Wänke, M. (2008). When impulses take over: Moderated predictive validity of explicit and implicit attitude measures in predicting food choice and consumption behaviour. *British Journal of Social Psychology*, *47*(3), 397-419.

Friston, K. J., Fletcher, P., Josephs, O., Holmes, Rugg, M. D., & Turner, R. (1998). Event-related fMRI: characterizing differential responses. *Neuroimage*, *7*(1), 30-40.

Führer, D., Zysset, S., & Stumvoll, M. (2008). Brain activity in hunger and satiety: an exploratory visually stimulated FMRI study. *Obesity*, *16*(5), 945-950.

García-García, I., Narberhaus, A., Marqués-Iturria, I., Garolera, M., Rădoi, A., Segura, B., & Jurado, M. A. (2013). Neural responses to visual food cues: insights from functional magnetic resonance imaging. *European Eating Disorders Review*, *21*(2), 89-98.

García-García, I., Horstmann, A., Jurado, M. A., Garolera, M., Chaudhry, S. J., Margulies, D. S., & Neumann, J. (2014). Reward processing in obesity, substance addiction and non-substance addiction. *obesity reviews*, *15*(11), 853-869.

Garner, D.M., Olmsted, M.P., Bohr, Y., & Garfinkel, P.E. (1982). The eating attitudes test: Psychometric features and clinical correlates*. Psychological Medicine, 12*, 871-878*.*

Garner, D. M., Olmstead, M. P., & Polivy, J. (1983). Development and validation of a multidimensional eating disorder inventory for anorexia nervosa and bulimia. *International Journal of Eating Disorders*, *2(2),* 15-34.

Goldstone, A. P., Prechtl de Hernandez, C. G., Beaver, J. D., Muhammed, K., Croese, C., Bell, G., & Bell, J. D. (2009). Fasting biases brain reward systems towards high-calorie foods. *European Journal of Neuroscience*, *30*(8), 1625-1635.

Grave de Peralta Menendez, R., Andino, S. G., Lantz, G., Michel, C. M., & Landis, T. (2001). Noninvasive localization of electromagnetic epileptic activity. I. Method descriptions and simulations. *Brain Topography*, *14*, 131-137.

Greenwald, A. G., McGhee, D. E., & Schwartz, J. L. (1998). Measuring individual differences in implicit cognition: the implicit association test. *Journal of Personality and Social Psychology*, *74(6)*, 1464.

Greenwald, A. G., Nosek, B. A., & Banaji, M. R. (2003). Understanding and using the implicit association test: I. An improved scoring algorithm. *Journal of Personality and Social Psychology*, *85(2)*, 197.

Grill-Spector, K., & Weiner, K. S. (2014). The functional architecture of the ventral temporal cortex and its role in categorization. *Nature Reviews Neuroscience*, *15*, 536-548.

Hare, T. A., Camerer, C. F., & Rangel, A. (2009). Self-control in decision-making involves modulation of the vmPFC valuation system. *Science, 324(5927*), 646-648.

Hare, T. A., Malmaud, J., & Rangel, A. (2011). Focusing attention on the health aspects of foods changes value signals in vmPFC and improves dietary choice. *Journal of Neuroscience*, *31*(30), 11077-11087.

Herman, C. P., & Polivy, J. (1980). Restrained eating. *Obesity*, 208-225.

Holsen, L. M., Zarcone, J. R., Thompson, T. I., Brooks, W. M., Anderson, M. F., Ahluwalia, J. S., & Savage, C. R. (2005). Neural mechanisms underlying food motivation in children and adolescents. *Neuroimage*, *27*(3), 669-676.

Houben, K., Roefs, A., & Jansen, A. (2010). Guilty pleasures. Implicit preferences for high calorie food in restrained eating. *Appetite*, *55(1)*, 18-24.

Huerta, C. I., Sarkar, P. R., Duong, T. Q., Laird, A. R., & Fox, P. T. (2014). Neural bases of food perception: Coordinate-based meta-analyses of neuroimaging studies in multiple modalities. *Obesity*, *22*(6), 1439-1446.

Karpinski, A., & Hilton, J. L. (2001). Attitudes and the Implicit Association Test. *Journal of Personality and Social Psychology*, *81(5)*, 774.

Kennedy, J., & Dimitropoulos, A. (2014). Influence of feeding state on neurofunctional differences between individuals who are obese and normal weight: a meta-analysis of neuroimaging studies. *Appetite*, *75*, 103-109.

Kiefer, M. (2001). Perceptual and semantic sources of category-specific effects: Event-related potentials during picture and word categorization. *Memory & Cognition*, *29*, 100-116.

Killgore, W. D., Young, A. D., Femia, L. A., Bogorodzki, P., Rogowska, J., & Yurgelun-Todd, D. A. (2003). Cortical and limbic activation during viewing of high-versus low-calorie foods. *Neuroimage*, *19*, 1381-1394.

Killgore, W. D., & Yurgelun-Todd, D. A. (2005). Body mass predicts orbitofrontal activity during visual presentations of high-calorie foods. *Neuroreport*, *16*(8), 859-863.

Killgore, W. D., & Yurgelun-Todd, D. A. (2007). Positive affect modulates activity in the visual cortex to images of high calorie foods. *International Journal of Neuroscience*, *117*, 643-653.

Killgore, W. D., & Yurgelun-Todd, D. A. (2010). Sex differences in cerebral responses to images of high vs low calorie food. *Neuroreport*, *21(5*), 354.

Knebel, J. F., Toepel, U., Hudry, J., Le Coutre, J., & Murray, M. M. (2008). Generating controlled image sets in cognitive neuroscience research. *Brain Topography*, *20*, 284-289.

Koebnick, C., Strassner, C., Hoffmann, I., & Leitzmann, C. (1999). Consequences of a long-term raw food diet on body weight and menstruation: results of a questionnaire survey. *Annals of Nutrition and Metabolism*, *43(2)*, 69-79.

Konkle, T., & Caramazza, A. (2013). Tripartite organization of the ventral stream by animacy and object size. *Journal of Neuroscience*, *33*(25), 10235-10242.

Kriegeskorte, N., Mur, M., Ruff, D. A., Kiani, R., Bodurka, J., Esteky, H., Tanaka, K., & Bandettini, P. A. (2008a). Matching categorical object representations in inferior temporal cortex of man and monkey. *Neuron*, *60*, 1126-1141.

Kriegeskorte, N., Mur, M., & Bandettini, P. (2008b). Representational similarity analysis–connecting the branches of systems neuroscience. Frontiers in systems neuroscience, 2.

Kringelbach, M. L., O'Doherty, J., Rolls, E. T., & Andrews, C. (2003). Activation of the human orbitofrontal cortex to a liquid food stimulus is correlated with its subjective pleasantness. *Cerebral cortex*, *13*(10), 1064-1071.

Krügel, U., Schraft, T., Kittner, H., Kiess, W., & Illes, P. (2003). Basal and feeding-evoked dopamine release in the rat nucleus accumbens is depressed by leptin. *European journal of pharmacology*, *482*(1), 185-187.

LaBar, K. S., Gitelman, D. R., Parrish, T. B., Kim, Y. H., Nobre, A. C., & Mesulam, M. (2001). Hunger selectively modulates corticolimbic activation to food stimuli in humans. *Behavioral Neuroscience*, *115(2),* 493.

Lehmann, D. (1971). Multichannel topography of human alpha EEG fields. *Electroencephalography and Clinical Neurophysiology*, *31*, 439-449.

Lehmann, D., & Skrandies, W. (1980). Reference-free identification of components of checkerboard-evoked multichannel potential fields. *Electroencephalography and Clinical Neurophysiology*, *48*, 609-621.

Linné, Y., Barkeling, B., Rössner, S., & Rooth, P. (2002). Vision and eating behavior. *Obesity Research*, *10*, 92-95.

Lutter, M., & Nestler, E. J. (2009). Homeostatic and hedonic signals interact in the regulation of food intake. *The Journal of nutrition*, *139*(3), 629-632.

Maison, D., Greenwald, A. G., & Bruin, R. (2001). The Implicit Association Test as a measure of implicit consumer attitudes. *Polish Psychological Bulletin*, *32 (1)*, 1-9.

Malik, S., McGlone, F., Bedrossian, D., & Dagher, A. (2008). Ghrelin modulates brain activity in areas that control appetitive behavior. *Cell metabolism*, *7*(5), 400-409.

Martin, A., & Chao, L. L. (2001). Semantic memory and the brain: structure and processes. *Current Opinion in Neurobiology*, *11*, 194-201.

Martin, A. (2007). The representation of object concepts in the brain. *Annu. Rev. Psychol.*, *58*, 25-45.

Meule, A., Kübler, A., & Blechert, J. (2013). Time course of electrocortical food-cue responses during cognitive regulation of craving. *Frontiers in Psychology*, *4*, 1-11.

Michel, C. M., Murray, M. M., Lantz, G., Gonzalez, S., Spinelli, L., & de Peralta, R. G. (2004a). EEG source imaging. Clinical neurophysiology, 115(10), 2195-2222.

Michel, C. M., Seeck, M., & Murray, M. M. (2004b). The speed of visual cognition. *Supplements to Clinical Neurophysiology*, *57*, 617-627.

Michel, C. M., & Murray, M. M. (2012). Towards the utilization of EEG as a brain imaging tool. *Neuroimage*, *61*, 371-385.

Moratti, S., Keil, A., & Stolarova, M. (2004). Motivated attention in emotional picture processing is reflected by activity modulation in cortical attention networks. *Neuroimage*, *21*, 954-964.

Morton, G. J. (2007). Hypothalamic leptin regulation of energy homeostasis and glucose metabolism. *The Journal of physiology*, *583*(2), 437-443.

Morton, G. J., & Schwartz, M. W. (2011). Leptin and the central nervous system control of glucose metabolism. *Physiological reviews*, *91*(2), 389-411.

Murray, M. M., Michel, C. M., de Peralta, R. G., Ortigue, S., Brunet, D., Andino, S. G., & Schnider, A. (2004). Rapid discrimination of visual and multisensory memories revealed by electrical neuroimaging. *Neuroimage*, *21*, 125-135.

Murray, M. M., Camen, C., Andino, S. L. G., Bovet, P., & Clarke, S. (2006). Rapid brain discrimination of sounds of objects. *Journal of Neuroscience*, *26*, 1293-1302.

Murray, M. M., Brunet, D., & Michel, C. M. (2008). Topographic ERP analyses: a step-by-step tutorial review. *Brain Topography*, *20*, 249-264.

Nakazato, M., Murakami, N., Date, Y., Kojima, M., Matsuo, H., Kangawa, K., & Matsukura, S. (2001). A role for ghrelin in the central regulation of feeding. *Nature*, *409*(6817), 194-198.

O'Doherty, J. P., Deichmann, R., Critchley, H. D., & Dolan, R. J. (2002). Neural responses during anticipation of a primary taste reward. *Neuron*, *33*(5), 815-826.

Ogden, C. L., Carroll, M. D., & Flegal, K. M. (2014). Prevalence of obesity in the United States. *Jama*, *312*, 189-190.

Oldfield, R. C. (1971). The assessment and analysis of handedness: the Edinburgh inventory. *Neuropsychologia*, *9*, 97-113.

Passamonti, L., Rowe, J. B., Schwarzbauer, C., Ewbank, M. P., Von Dem Hagen, E., & Calder, A. J. (2009). Personality predicts the brain's response to viewing appetizing foods: the neural basis of a risk factor for overeating. *Journal of Neuroscience*, *29*(1), 43-51.

Perrin, F., Pernier, J., Bertnard, O., Giard, M. H., & Echallier, J. F. (1987). Mapping of scalp potentials by surface spline interpolation. *Electroencephalography and clinical neurophysiology*, *66*, 75-81.

Plassmann, H., O'Doherty, J., & Rangel, A. (2007). Orbitofrontal cortex encodes willingness to pay in everyday economic transactions. *The Journal of neuroscience*, *27*(37), 9984-9988.

Plassmann, H., O'Doherty, J. P., & Rangel, A. (2010). Appetitive and aversive goal values are encoded in the medial orbitofrontal cortex at the time of decision making. *Journal of neuroscience*, *30*(32), 10799-10808.

Posner, M. I., & Petersen, S. E. (1990). The attention system of the human brain. *Annual Review of Neuroscience*, *13*, 25-42.

Power, M. L., Schulkin, J. (2013). *The Evolution of Obesity*. JHU Press.

Price, M., Lee, M., & Higgs, S. (2016). Food-specific response inhibition, dietary restraint and snack intake in lean and overweight/obese adults: a moderated-mediation model. *International Journal of Obesity*, *40*(5), 877-882.

Proverbio, A. M., Del Zotto, M., & Zani, A. (2007). The emergence of semantic categorization in early visual processing: ERP indices of animal vs. artifact recognition. *BMC Neuroscience*, *8*-24.

Rangel, A. (2013). Regulation of dietary choice by the decision-making circuitry. *Nature neuroscience*, *16*(12), 1717-1724.

Rangel, A., & Hare, T. (2010). Neural computations associated with goal-directed choice. *Current opinion in neurobiology*, *20*(2), 262-270.

Richetin, J., Perugini, M., Prestwich, A., & O'Gorman, R. (2007). The IAT as a predictor of food choice: The case of fruits versus snacks. *International Journal of Psychology*, *42(3)*, 166-173.

Roefs, A., & Jansen, A. (2002). Implicit and explicit attitudes toward high-fat foods in obesity. *Journal of abnormal psychology*, *111*(3), 517.

Roefs, A., Herman, C. P., MacLeod, C. M., Smulders, F. T. Y., & Jansen, A. (2005). At first sight: how do restrained eaters evaluate high-fat palatable foods?. *Appetite*, *44(1)*, 103-114.

Rolls, B. J., Rolls, E. T., Rowe, E. A., & Sweeney, K. (1981). Sensory specific satiety in man. *Physiology & Behavior*, *27*(1), 137-142.

Rolls, E.T. (2015). Taste, olfactory, and food reward value processing in the brain. *Progress in Neurobiology*, *127*, 64-90.

Rothemund, Y., Preuschhof, C., Bohner, G., Bauknecht, H. C., Klingebiel, R., Flor, H., & Klapp, B. F. (2007). Differential activation of the dorsal striatum by high-calorie visual food stimuli in obese individuals. *Neuroimage*, *37*(2), 410-421.

Rozin, P. (1976). The selection of foods by rats, humans, and other animals. *Advances in the Study of Behavior*, *6*, 21-76.

Rumiati, R. I., & Foroni, F. (2016). We are what we eat: How food is represented in our mind/brain. *Psychonomic bulletin & review*, *23*(4), 1043-1054.

Rumiati, R.I., Foroni, F., Pergola, G., Rossi, P., & Silveri, M.C. (2016). Lexical-semantic deficits in processing food and non-food items. *Brain and Cognition, 110*, 120-130.

Santel, S., Baving, L., Krauel, K., Münte, T. F., & Rotte, M. (2006). Hunger and satiety in anorexia nervosa: fMRI during cognitive processing of food pictures. *Brain research*, *1114*(1), 138-148.

Scarpace, P. J., & Zhang, Y. (2009). Leptin resistance: a prediposing factor for diet-induced obesity. *American Journal of Physiology-Regulatory, Integrative and Comparative Physiology*, *296*(3), R493-R500.

Schienle, A., Schäfer, A., Hermann, A., & Vaitl, D. (2009). Binge-eating disorder: reward sensitivity and brain activation to images of food. *Biological psychiatry*, *65*(8), 654-661.

Schur, E. A., Kleinhans, N. M., Goldberg, J., Buchwald, D., Schwartz, M. W., Maravilla, K. (2009). Activation in brain energy regulation and reward centers by food cues varies with choice of visual stimulus. *International Journal of Obesity*, *33*, 653-661.

Siep, N., Roefs, A., Roebroeck, A., Havermans, R., Bonte, M., Jansen, A. (2012). Fighting food temptations: the modulating effects of short-term cognitive reappraisal, suppression and up-

regulation on mesocorticolimbic activity related to appetitive motivation. *Neuroimage*, *60*, 213-220.

Simmons, W. K., Martin, A., & Barsalou, L. W. (2005). Pictures of appetizing foods activate gustatory cortices for taste and reward. *Cerebral Cortex*, *15*(10), 1602-1608.

Simmons, W. K., Rapuano, K. M., Kallman, S. J., Ingeholm, J. E., Miller, B., Gotts, S. J., & Martin, A. (2013). Category-specific integration of homeostatic signals in caudal but not rostral human insula. Nature neuroscience, 16(11), 1551-1552.

Simmons, W. K., Burrows, K., Avery, J. A., Kerr, K. L., Bodurka, J., Savage, C. R., & Drevets, W. C. (2016). Depression-related increases and decreases in appetite: dissociable patterns of aberrant activity in reward and interoceptive neurocircuitry. *American Journal of Psychiatry*, *173*(4), 418-428.

Small, D. M., Gitelman, D., Simmons, K., Bloise, S. M., Parrish, T., Mesulam, M. M. (2005). Monetary incentives enhance processing in brain regions mediating top-down control of attention. *Cerebral Cortex*, *15*, 1855-1865.

Stevenson, R. J., & Francis, H. M. (2017). The hippocampus and the regulation of human food intake. *Psychological Bulletin*, *143*(10), 1011.

Stingl, K. T., Rogić, M., Stingl, K., Canova, C., Tschritter, O., Braun, C., & Preissl, H. (2010). The temporal sequence of magnetic brain activity for food categorization and memorization—an exploratory study. *Neuroimage*, *52*(4), 1584-1591.

Strack, F., & Deutsch, R. (2004). Reflective and impulsive determinants of social behavior. *Personality and Social Psychology Review, 8(3),* 220-247.

Sullivan, N., Hutcherson, C., Harris, A., & Rangel, A. (2015). Dietary self-control is related to the speed with which attributes of healthfulness and tastiness are processed. *Psychological Science*, *26(2)*, 122-134.

Tailarach, J., & Tournoux, P. (1988). Co-planar stereotaxic atlas of the human brain: 3-dimensional proportional system-an approach to cerebral imaging. *Stuttgart and New York: Thieme Verlag*.

Thorpe, S., Fize, D., & Marlot, C. (1996). Speed of processing in the human visual system. *Nature*, *381*(6582), 520.

Toepel, U., Knebel, J. F., Hudry, J., le Coutre, J., & Murray, M. M. (2009a). The brain tracks the energetic value in food images. *Neuroimage*, 44, 967-974.

Toepel, U., Knebel, J.F., Hudry, J., Le Coutre, J., & Murray, M.M., (2009b). Advantageous object recognition for high-fat food images. In: Montmayeur, J.P., Le Coutre, J. (Eds.), *Fat Detection: Taste, Texture, and Post-Ingestive Effects*. Taylor and Francis Group.

Toepel, U., Knebel, J. F., Hudry, J., le Coutre, J., & Murray, M. M. (2012). Gender and weight shape brain dynamics during food viewing. *PloS one*, *7(5)*, e36778.

Tsourides, K., Shariat, S., Nejati, H., Gandhi, T. K., Cardinaux, A., Simons, C. T., Cheung, N. M., Pavlovic, V., & Sinha, P. (2016). Neural correlates of the food/non-food visual distinction. *Biological Psychology*, *115*, 35-42.

Turkeltaub, P. E., Eickhoff, S. B., Laird, A. R., Fox, M., Wiener, M., & Fox, P. (2012). Minimizing within-experiment and within-group effects in activation likelihood estimation meta-analyses. *Human brain mapping*, *33*(1), 1-13.

Uher, R., Murphy, T., Brammer, M. J., Dalgleish, T., Phillips, M. L., Ng, V. W., & Treasure, J. (2004). Medial prefrontal cortex activity associated with symptom provocation in eating disorders. *American Journal of Psychiatry*, *161*(7), 1238-1246.

Uher, R., Treasure, J., Heining, M., Brammer, M. J., & Campbell, I. C. (2006). Cerebral processing of food-related stimuli: effects of fasting and gender. *Behavioural brain research*, *169*(1), 111-119.

Van der Laan, L. N., De Ridder, D. T. D., Viergever, M. A., Smeets, P. A. (2011). The first taste is always with the eyes: a meta-analysis on the neural correlates of processing visual food cues. *Neuroimage*, *55*, 296-303.

Vogel, E. K., & Luck, S. J. (2000). The visual N1 component as an index of a discrimination process. *Psychophysiology*, *37*(2), 190-203.

Warneken, F., & Rosati, A. G. (2015). Cognitive capacities for cooking in chimpanzees. *Proceedings of the Royal Society B*, *282*, 20150229.

Wilson, T. D., Lindsey, S., & Schooler, T. Y. (2000). A model of dual attitudes. *Psychological Review*, *107(1)*, 101.

Wobber, V., Hare, B., & Wrangham, R. (2008). Great apes prefer cooked food. *Journal of Human Evolution*, *55(2)*, 340-348.

World Health Organization (WHO. (2013). Obesity and Overweight factsheet from the WHO). http://www.who.int/mediacentre/factsheets/fs311/en

Wrangham, R. W., Jones, J. H., Laden, G., Pilbeam, D., & Conklin Brittain, N. (1999). The raw and the stolen. *Current Anthropology*, *40(5*), 567-594.

Wrangham, R. W., Jones, J. H., Laden, G., Pilbeam, D., Conklin-Brittain, N., Brace, C. L., ... & Blurton Jones, N. G. (1999). The raw and the stolen: cooking and the ecology of human origins. *Current anthropology*, *40*(5), 567-594.

Wrangham, R., & Conklin-Brittain, N. (2003). Cooking as a biological trait. *Comparative Biochemistry and Physiology Part A: Molecular & Integrative Physiology*, *136*(1), 35-46.

Wrangham, R. (2013). The evolution of human nutrition. *Current Biology*, *23*, 354-355.

Wyatt, S. B., Winters, K. P., & Dubbert, P. M. (2006). Overweight and obesity: prevalence, consequences, and causes of a growing public health problem. *The American journal of the medical sciences*, *331*(4), 166-174.

Zink, K. D., & Lieberman, D. E. (2016). Impact of meat and Lower Palaeolithic food processing techniques on chewing in humans. *Nature*, *531*, 500–503.