Elucidating the mechanisms of individual variation in fat perception and preference

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Abstract

Fat is perceived through three modalities; mouthfeel, odour and, also taste where free fatty acids are the stimuli generating this sensation. Individuals showed variation in oral fat taste perception which significantly influenced fat intake and preference. The mechanisms causing differences in individual oral fat taste perception could be explained by genetic variation of the fatty acid translocase CD36 and lipase activity. Individuals had different lipase activities in saliva to release free fatty acids from dietary fat, which influenced oral fat perception. In addition, CD36 genotypes influenced oral fat perception and the influence of CD36 genotype on oral fat perception differed between subjects with high salivary lipase activity and those with low lipase activity.

Introduction

Taste is one of the influential determinants driving individual food preference and consumption. Fat contributes to the unique texture and odour of foods, and recent studies proposed that fat can also be perceived through taste. Fatty acids are proposed as the effective stimuli in generating fat taste sensation due to the discovery of fatty acid receptors and activation of transduction pathways in mouth [1]. Individuals present diverse oral fat taste perception and these variations could contribute to individual differences in food liking and consumption [2]. As triglyceride is the major component in dietary fat and free fatty acids are only present in foods at very low levels, recent study report that salivary lipase can hydrolysis triglyceride in the mouth into free fatty acids [3]. This may cause an increase in free fatty acid in the mouth and hence result in a greater oral taste sensation. However, it is still unclear whether differences in individual salivary lipase activity may be a potential cause of individual differences in oral fat perception. CD36, as the fatty acid translocase, has been regarded as a putative candidate for the oral fat sensor. Genetic variations in CD36 have been proposed as another potential factor that could influence individual differences to oral fat taste perception.

This study aims to understand how fat is perceived in mouth. It also aims to understand inter-subject variability in fatty acid sensitivity, fat perception, fat preference and choice of high fat foods, and to elucidate the impact of CD36 genotype and salivary lipase activity on individual variation in fat perception.

Materials and methods

Participant

Ninety participants of age range 18-55 years were recruited and self-reported to be healthy. Three participants dropped out before they had completed the sessions, and two participants were excluded from the data analysis due to the incomplete questionnaires.

Fat intensity rating

The samples and method for the intensity test were developed as described in the study of Zhou et al., 2016 [4]. Non-fat skimmed milk, single cream and double cream (Tesco, UK) were used to generate seven samples of different fat levels: 0%, 2.5%, 5%,

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7.5%, 10%, 15% and 20%. Mouthfeel masked samples (containing thickener (Nestlé Nutrition Resource ThickenUp Clear, UK) and liquid paraffin (Care, Thornton & Ross, UK)) and mouthfeel non-masked samples were prepared. Participants were asked to rate the perceived fat intensity on a generalised labelled magnitude scale (gLMS). Nose-clips were provided for mouthfeel-masked samples to obtain intensity ratings under the "taste" modality in isolation from odour and mouthfeel.

Fatty acid sensitivity test

Samples and methods for threshold sensitivity were developed as described by Zhou et al. (2016). Food-grade oleic acid (Sigma, UK) was chosen and the concentration of oleic acid ranged from 0.098 to 55.9 mM (0.0028 to 1.58% w/v), with dilution differing by 0.25 log units. The rapid 3 alternative forced choice (3AFC) approach was used to measure fatty acid sensitivity. During the whole process, participants were asked to wear nose-clips to avoid any olfactory effects.

Dietary intake and preference

A Food Frequency Questionnaire (FFQ) and a Food Preference Questionnaire (FPQ, adapted from Deglaire et al., 2012 [5]) were used to collect recalled food intake and food preference data.

Salivary lipase activity measurement

The lipase activity in saliva was reflected by the free oleic acid generation in expectorated almond samples. Participants were asked to chew one almond (15s) without swallowing and the expectorated almond was collected. The free oleic acid in expectorated almond samples was analysed using Folch extraction of fat from expectorated almond samples, separation of free oleic acids from fat (using solid phase extraction with aminopropyl cartridges (ISOLUTE® NH2, Biotage)), derivatization of free oleic acids to oleic acid methyl ester at room temperature for 5 min (by using 1.5% H_2SO_4 in methanol) and gas chromatography analysis (using gas chromatography flame ionization detector (Hewlett Packard 5890 Series II) with non-polar capillary Agilent J&W DB-5 column (60 m × 0.25 mm × 0.25 μm)). The total fat in the expectorated almond sample was measured by using Folch extraction to calculate the free oleic acid in total fat (g/ml).

CD36 genotype measurement

Participants were asked to swab the inside of their cheek 7 times to collect buccal cells using sterile Omni swabs (Whatman, UK). The buccal swab samples were collected in duplicate. Genotyping of three CD36 SNPs (*rs1761667, rs1527483, rs3840546*) was carried out at iDNA genetics Ltd (Norwich, UK).

Statistical analysis

Fat intensity ratings were collected by Compusense at-hand (Canada). Data was analysed by XLSTAT (version 2016.8, Addinsoft). Latent cluster analysis was conducted to classify participants into different liking groups based on their recalled liking ratings of foods. Multivariate ANOVA with Bonferroni pairwise comparison was conducted to examine the difference in perceived fat intensity between groups (e.g. liking groups, sensitivity groups or genotypes). Significance was set at 0.05.

Results and Discussion

Taste modality on oral fat perception

Significant differences were found between fat levels under the "taste" condition ($p<0.05$), which implies that fat can be distinguished by taste. The perceived fat intensity rated in the "taste" modality was significantly higher than in the "overall" modality ($p<0.0001$), which was due to the addition of paraffin and thickener in the mouthfeel-masked samples. This confirms that mouthfeel, such as thickness and lubrication, is an important indicator of oral fat perception.

Food liking, food intake and oral fat perception

Based on liking results collected by FPQ, high fat likers (HFLs) and high fat dislikers (HFDs) were established. HFLs (n=34) displayed higher liking scores in most food items (42 out of 46) which was significant for 25 items ($p<0.05$). HFLs showed higher fat intake (as % total energy) ($p=0.004$), which implies that high liking to foods rich in fat could stimulate the consumption of these foods. In addition, HFLs showed significantly lower perceived fat intensity under the "taste" modality compared to HFDs (n=51, Figure 1A). Fat taste generated by fatty acids has been reported to be an unpleasant taste sensation [6], so this might explain why the HFDs do not like foods rich in fat.

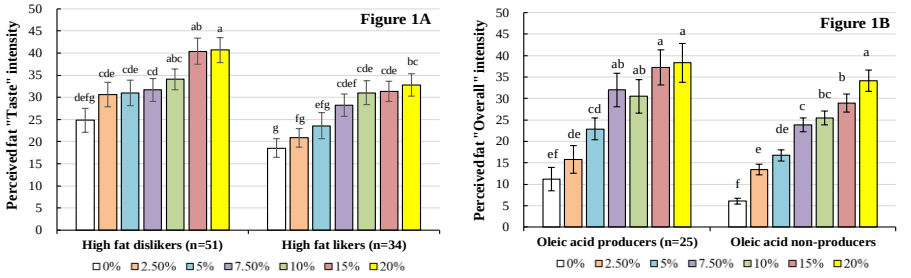

Figure 1: Perceived fat "taste" intensity between HFLs and HFDs (A, left) and perceived fat "overall" intensity between oleic acid producers/non-producers (B, right). Bars not sharing a common letter differ significantly ($p<0.05$) between fat levels and between groups. Error bars represent standard error of the mean.

Fatty acid sensitivity and oral fat perception

Individual threshold sensitivity to oleic acid varied, and participants were divided into high (n=47) / medium (n=19) / low (n=21) sensitivity groups (grouping approach was developed by Zhou et al. (2016)). No significant difference in the perceived fat intensity rating was found between fatty acid sensitivity groups ($p=0.46$). However, the high sensitivity group could discriminate more pairs of fat levels in the fat intensity rating, which implies that individual sensitivity to oleic acid influenced the ability to distinguish fat levels in the real food model.

Salivary lipase activity and oral fat perception

Free oleic acid as a percentage of total fat in expectorated almond samples ranged from 0.024% to 3.75%w/w. Compared with free oleic acid (as % of total fat) in whole ground almond (0.027% to 0.26%), participants with expectorated free oleic acid (%) above 0.26% (n=20) were grouped as "oleic acid producers", and those below 0.26% (n=65) were "oleic acid non-producers". No significant difference in fat "taste" intensity ratings was found between producers and non-producers ($p=0.39$), however, under the "overall" condition, oleic acid producers rated oral fat perception higher than non-

producers (p<0.0001, Figure 1B). Therefore, to some extent, subjects who could produce free oleic acid in the mouth perceived a stronger oral sensation of fat in the cream model.

CD36 genotype and oral fat perception

CD36 *rs1761667* genotype influenced perceived fat "taste" intensity rating (p=0.003), where A/A carriers (n=27) had significantly higher perceived fat "taste" intensity than G/G (n=21) and A/G (n=35). No significant effect of *rs1527483* nor *rs3840546* was found on perceived fat intensity (p=0.22, p=0.14, respectively).

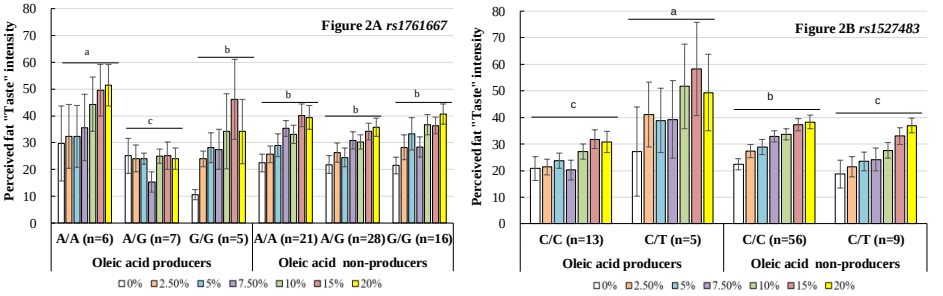

Figure 2: Perceived fat "taste" intensity between oleic acid producers and non-producers with different CD36 genotypes at *rs1761667* (A) and at *rs1527483* (B). Bars not sharing a common letter differ significantly (p<0.05) between groups. Error bars represent standard error of the mean.

A significant interaction between *rs1761667* genotypes and oleic acid producers/non-producers, and between *rs1527483* genotypes and producers/non-producers on perceived fat "taste" intensity was observed (p=0.007, p<0.0001, respectively). In oleic acid producers, *rs1761667* A/A carriers (n=6) showed higher perceived fat "taste" intensity than G/G carriers (n=5, p=0.014) and A/G carriers (n=7, p<0.0001, Figure 2A). In addition, *rs1527483* C/T carriers (n=5) presented higher perceived fat "taste" intensity than C/C carriers (n=13) (p<0.0001, Figure 2B). Regarding non-producers, no significant difference in perceived fat "taste" intensity was found between *rs1761667* A/A (n=21), G/G (n=28) and A/G (n=16); however, *rs1527483* C/C carriers (n=56) showed higher perceived fat "taste" intensity than C/T carriers (n=9) (p=0.035, Figure 2B). This implies that the influence of CD36 genotypes on oral fat perception varied between subjects according to their ability to generate free fatty acids.

Conclusion

Fat can be perceived through taste. Individual preference to high-fat foods varies. High-fat likers had significantly lower fat taste perception in dairy model. Both lipase activity and CD36 genotype influence oral fat perception. The influence of CD36 genotypes on oral fat perception varies between subjects with high or low lipase activity.

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