Fat intake modulates cerebral blood flow in homeostatic and gustatory brain areas in humans[1–4]

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ABSTRACT

Background: The hypothalamus is the central homeostatic control region of the brain and, therefore, highly influenced by nutrients such as glucose and fat. Immediate and prolonged homeostatic effects of glucose ingestion have been well characterized. However, studies that used stimulation with fat have mainly investigated immediate perceptional processes. Besides homeostatic processes, the gustatory cortex, including parts of the insular cortex, is crucial for the processing of food items.

Objective: The aim of this study was to investigate the effect of high- compared with low-fat meals on the hypothalamus and the insular cortex.

Design: Eleven healthy men participated in a single-blinded, functional MRI study of high- and low-fat meals on 2 measurement days. Cerebral blood flow (CBF) was measured before and 30 and 120 min after intake of high- and low-fat yogurts. Hunger was rated and blood samples were taken before each CBF measurement.

Results: High-fat yogurt induced a pronounced decrease in CBF in the hypothalamus, and the corresponding CBF change correlated positively with the insulin change. Furthermore, insular activity increased after 120 min in the low-fat condition only. The CBF change in both regions correlated positively in the high-fat condition.

Conclusions: The decrease in hypothalamic activity and the interaction with the insular cortex elicited by fat may contribute to an efficient energy homeostasis. Therefore, fat might be a modulator of homeostatic and gustatory brain regions and their interaction. This trial was registered at clinicaltrials.gov as NCT01516021. *Am J Clin Nutr* 2012;95:1342–9.

INTRODUCTION

The hypothalamus is the major homeostatic region of the brain and controls food intake and energy homeostasis (1). Hypothalamic nuclei receive inputs in response to food intake. Neurons in these nuclei sense gut hormones, glucose, insulin, or leptin and, in response, regulate appetite, hunger, and food intake again (2). Several studies have described the homeostatic effect of glucose action in the hypothalamus. For example, oral glucose ingestion elicits decreased hypothalamic activity immediately after the intake of a glucose drink (3–5). Moreover, it has been shown that glucose but not a nonsweet glucose dimer results in such hypothalamic reactions, which indicated that both sweet taste and energy content are required for the hypothalamic response (6). Glucose-related responses of the hypothalamus are also influenced by sex (7) and altered glucose tolerance (ie, type 2 diabetes) (8). In addition to these glucose-related findings, the hypothalamus is affected by other nutrients such as fat, which is, besides glucose, one of the most important energy sources. There is evidence for direct effects of free (ie, nonesterified) fatty acids (FFAs)[5] on the hypothalamus, primarily at the level of the arcuate nucleus (9). Specific effects of blood-derived FFAs on the hypothalamus were first shown in rats (10). In addition, brain insulin sensitivity is associated with plasma FFAs, which points toward the effects of FFAs on insulin signaling in the brain (11).

Neuroimaging studies after stimulation with fat are very rare and have mainly investigated the immediate fat representation in the mouth with focus on viscosity, texture, taste, and pleasantness. The oral representation of fat in the mouth revealed an immediate increase in hypothalamic activity (12, 13) and the anterior cingulate cortex, insular cortex (12, 14), and amygdala (13, 14). Viscosity differences of the solutions had no effect on the hypothalamus but mainly on the insular cortex (12, 15). The findings of these studies are particularly interesting because these immediate reactions were sensory-based rather than homeostatic-based effects.

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[3] Supported by the German Research Foundation (PAK347 FR 1561/4-1 and BI 195/62-1), the Arbeitskreis Industrielle Forschung (15960 N), the Zentrum für Ernährungsmedizin (26.A I-08), and the German Center for Diabetes Research.

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[5] Abbreviations used: CBF, cerebral blood flow; FFA, free fatty acid; PASL, pulsed arterial spin labeling; ROIs, regions of interest; TE, echo time; TI, inversion time; TR, repetition time.

Received November 25, 2011. Accepted for publication March 20, 2012.
First published online May 9, 2012; doi: 10.3945/ajcn.111.031492.

On a behavioral level, the intake of different fat emulsions with equivalent fat content leads to a differential subsequent food intake for several hours after ingestion as well as to differences in hunger, desire to eat, and perceived fullness between lean (16) and obese subjects (17).

The aim of the current study was to investigate the effect of a high-fat compared with low-fat meal (operationalized by high- and low-fat yogurts) on the hypothalamus and on the insular cortex, which are both areas involved in food processing. Because sex differences were observed in earlier studies that targeted the hypothalamus (7), only men were included in a study that used pulsed arterial spin labeling (PASL). We used the resting cerebral blood flow (CBF) as a measure of brain activity because we were interested in the absolute quantification of possible differences in brain perfusion because of the fat content of the yogurt without any task. We measured the CBF within the hypothalamus, the insular cortex, and several control regions before and after (30 and 120 min) intakes of 500 mL high- and low-fat yogurts.

SUBJECTS AND METHODS

Subjects

For this study, 11 healthy men were included [age: 28.81 ± 0.57 y; BMI (in kg/m^2): 24.17 ± 0.71; 10 subjects were right handed].

Before the first scanning day, all subjects underwent a medical screening (including blood sampling and an examination by a physician) and filled out psychological and psychiatric questionnaires as well as eating-behavior questionnaires. To address psychiatric disorders, the Patient Health Questionnaire (18) and the Beck Depression Inventory (19) were used. To ensure a normal eating behavior for all subjects, the German versions of the Three-Factor Eating Questionnaire (20) and the Eating-Disorder Examination (21) and the trait version of the Food Craving Questionnaire (22) were applied. None of the subjects showed any kind of physiologic or psychiatric disorder or other diseases as assured by a physician.

The study protocol was approved by the ethics committee of the medical faculty of the University of Tübingen, and all participants gave written informed consent.

Study design

After an overnight fast ≥10 h, subjects completed 3 PASL measurements on each of 2 separate days. Before each PASL measurement, blood samples were taken, and subjects rated their subjective hunger on a 0–100 (0 = not at all hungry; 100 = very hungry) visual analog scale. After the first measurement, subjects were instructed to eat 500 mL of a high-fat (8%) or low-fat (<0.1%) yogurt in ≤10 min. The order of the yogurt was counterbalanced and single-blinded. The first postmeasurement (CBF 2) took place 30 min after each subject started to eat the yogurt, and the second postmeasurement started at the time point of 120 min. We chose these time points because metabolic and endocrine reactions in response to food intake are well known for these time points. An overview of the study design is given in **Figure 1**.

Blood sampling and analysis

At 6 time points (pre and 30, 60, 90, 120, and 150 min), blood samples were taken to determine insulin, glucose, and FFAs.

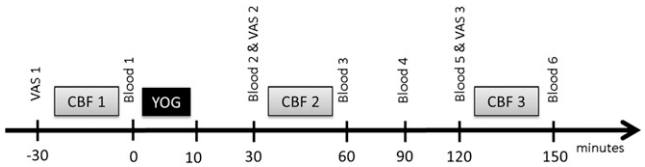

FIGURE 1. Study design. CBF, cerebral blood flow; VAS, visual analog scale; YOG, yogurt.

Blood glucose was determined by using the glucose dehydrogenase method. Plasma insulin concentrations were measured by using a chemiluminescence assays for ADVIA Centaur (Siemens Medical Solutions). Total FFA concentrations were determined by using an enzymatic method (NEFAC kit; WACO Chemicals).

Yogurt production

Low- and high-fat yogurts were produced at the Institute of Food Science and Biotechnology (University of Hohenheim). Bovine raw milk was obtained freshly from the Dairy Research Station Meiereihof (University of Hohenheim), separated (fat <0.1% wt:wt), and pasteurized in house at 74°C for 30 s. Subsequently, the protein content was set to 3.4 ± 0.1% (wt:wt). Because the subjects should not have noticed a difference in respect to textural properties of the low- and high-fat yogurts, 2 processing ways were performed. For the production of the low-fat yogurt (<0.1% fat), skim milk was concentrated with a crossflow membrane filtration device (Membralox, cutoff: 0.1 μm; Pall Seitz Schenk) until a concentration factor of i = 1.8 was reached. For the production of the high-fat yogurt, skim milk was adjusted to a fat content of 8.0 ± 0.1% (wt:wt) by using cream (35 ± 0.1% wt:wt) from the separation process. Subsequently, both standardized milk types were heated (95°C for 4.3 min) and subsequently cooled to 35°C in the tubular heating equipment of a pilot heating plant (Asepto GmbH). The content of protein, fat, and lactose of heated yogurt milk types were analyzed by using LactoScope FTIR Advanced (Fourier Transform InfraRed Spectroscopy; Delta Instruments). After fermentation at 35°C to a pH of 4.4 ± 0.1 with the freeze-dried culture Freeze-Dried Direct Vat Set Yo-Flex 812, which contained *Lactobacillus delbrueckii* subspecies *bulgaricus* and *Streptococcus thermophilus* (Chr Hansen GmbH), the milk gel was manually broken with a stainless-steel bored disk by up-and-down movements for 60 s and sheared with a needle valve. Fat contents of yogurt products were measured by using the Gerber standard method (23). On the basis of the Dumas method DIN 10467, the protein content was determined with an FP-528 protein and nitrogen analyzer (Leco Instruments GmbH). The dry matter was determined at 90°C by using an infrared dryer (Moisture Analyzer MA30; Sartorius) (23). On the basis of the energy value of protein (4 kcal/g), fat (9 kcal/g), and carbohydrate (3 kcal/g) (24), the caloric value of low- and high-fat yogurts was calculated as follows:

$$\text{Caloric value yogurt} = (C_F \times H_F) + (C_P \times H_P) + (C_L \times H_L) \quad (1)$$

where C_F is the fat content, H_F is the energy value of fat, C_P is the protein content, H_P is the energy value of protein, C_L is the lactose content, and H_L is the energy value of lactose. The main carbohydrates in the yogurt were lactose. The energy value of

organic acids and minerals was discarded. The determined macronutrients and calculated caloric value of yogurt products are shown in **Table 1**.

After 24 h of storage at 10°C, both yogurt types were analyzed by means of rheologic measurements (25). Results of the large deformation test revealed no difference in terms of viscosity properties of the low- and high-fat yogurts (*see* Figure S1 under "Supplemental data" in the online issue).

Data acquisition

Scanning was performed on a 3T (Tesla) scanner (Tim Trio; Siemens) equipped with a 12-channel transreceiver head coil. PASL images were obtained with FAIR-QUIPSS II (flow-sensitive alternated inversion recovery–quantitative imaging of perfusion using a single subtraction sequence) using a hyperbolic secant pulse for inversion and an echo planar imaging readout for acquisition. A total of 8 axial slices with a slice thickness of 6 mm (no gap) positioned around the hypothalamus were acquired in ascending order. Two presaturation pulses were applied in the imaging planes directly before the inversion tag to minimize the impact of the static tissue. Each measurement consisted of 200 alternating tag and control images (overall: 8 min 27 s) with the following after-imaging variables: inversion time $(TI)_1$ = 700 ms, TI_2 = 1400 ms, repetition time (TR) = 2500 ms, echo time (TE) = 13 ms, in-plane resolution = 4×4 mm^2, field of view = 256 mm, and flip angle = 90°. The same sequences were used to estimate the equilibrium magnetization of the blood for absolute CBF quantification with the same sequence variables as mentioned except that TR and TI_2 were chosen to be 10 and 4 s, respectively, to allow for T1 (spin-lattice relaxation) recovery and avoid saturation effects (26). In addition, a high-resolution T1-weighted anatomical image was acquired (Magnetization Prepared Rapid Gradient Echo; matrix size: 256×256; 176 slices; $1 \times 1 \times 1$-mm isotropic voxels; TR = 1900 ms; TE = 2.26 ms; TI = 900 ms).

Image processing

Image processing was performed as described previously (27). Functional data were analyzed with FMRIB's Software Library software (FSL, version 4.1.1; http://www.fmrib.ox.ac.uk/fsl/) (28) and additional custom-made MATLAB (Mathworks) and Linux shell script routines. Images were motion-corrected using the MCFLIRT module of FSL (version 4.1.1) (28) by using the mean volume of the corresponding session as a reference. Time series of all functional sessions were high-pass filtered to remove low-frequency baseline drifts that were potentially caused by scanner instability, subject motion, and physiologic noise. Resulting mean volumes of each session and day were coregistered separately to the mean image of the first session in the first day by using a 6-variable rigid-body transformation and sinc interpolation. Estimated transformation variables for each session and day were applied for all other images of the same session. In addition, the individual anatomical image was coregistered to the mean functional reference image. Finally, the functional images were smoothed with a Gaussian kernel (full width at half maximum: $4 \times 4 \times 6$ mm). Perfusion images were generated by calculating the control-tag differences by using surround subtraction (ie, computing the difference between each image and the average of its 2 nearest neighbors), which minimized blood oxygen level–dependent signal weighting of CBF (29). Absolute perfusion quantification was performed by using the general kinetic model (30)

$$\Delta M = (2 \times \alpha \times M_{0B} \times f \times \mathrm{TI}_1 \times e^{-\mathrm{TI}_2/\mathrm{T1}_B})/\lambda \qquad (2)$$

where f denotes the CBF (mL · 100 g^{-1} · min^{-1}), $T1_B$ is the longitudinal relaxation time, and M_{0B} denotes the equilibrium magnetization of the arterial blood [details for the absolute quantification of CBF are shown in Cavuşoğlu et al (26)]. The value of $T1_B$ at 3T was estimated to be 1684 ms (31). Moreover, a water partition coefficient between blood and gray matter of λ = 0.9 g/mL and an inversion efficiency of α = 0.95 were used for absolute CBF quantification (32). The delay time TI_2 in the ascending imaging slices (transit delay time of each slice) was adjusted. We used an M_{0B} map instead of a global value to quantify the perfusion on each voxel, which corrected for the spatial sensitivity profile of the head coil (26). The FAIR-QUIPSS II ASL sequence we used in the study showed the highest signal-to-noise ratio compared with other PASL methods (26). In addition, the voxel-specific local values of the equilibrium magnetization of the brain (M_{0B}) revealed the most accurate CBF quantification compared with global values.

Data analyses

Individual masks for the hypothalamus for each subject on the coregistered anatomical image were created. Starting from the anterior commissure (y = 0 mm), a mask (by using the create mask option in the FSLVIEW module of FSL, version 3.1; http://www.fmrib.ox.ac.uk/fsl/fslview/index.html) was manually drawn in the posterior direction with the following boundaries: y = 0 to −16, x = −10 to 10, and z = 0 to −12, after the description of Matsuda et al (33). The individual mask was transformed into the dimensions of the functional images that

TABLE 1
Macronutrient contents and calculated energy values of low- and high-fat yogurts[1]

	C_F	C_P	C_L	DM	Energy value
	Percentage of wt:wt	*Percentage of wt:wt*	*Percentage of wt:wt*	*%*	*kcal/100 g*
Low-fat yogurt	0.08 ± 0.05[2]	6.13 ± 0.05	4.7 ± 0.04	12.9 ± 0.13	~40
High-fat yogurt	8.04 ± 0.12	3.42 ± 0.06	4.6 ± 0.06	15.8 ± 0.34	~100

[1] C_F, fat content; C_L, lactose content; C_P, protein content; DM, dry matter.
[2] Mean ± SE of 3 measurements (all such values).

comprised 5 × 5 × 3 voxels, and CBF values of each voxel in the mask were extracted for all days and sessions. Beside the hypothalamus, the insular cortex (anterior and medial) and additional control regions of interest (ROIs) (thalamus, temporal, and visual cortex) were selected. All ROIs were completely covered by the field of view and could easily be defined anatomically (the selection of ROIs in one representative subject is depicted in red in Figure 3; also *see* Figure S2 under "Supplemental data" in the online issue). The average CBF of all voxels in the ROIs was calculated separately for each day and measurement.

Statistical analyses were performed with SPSS 18 software (SPSS). To account for interindividual and intraindividual differences in the resting CBF, the percentage of CBF change from the premeasurement (−30 min) to both post measurements (30 and 120 min) was calculated. Thereafter, repeated-measurement ANOVAs were calculated for each region of interest with the factor fat (high- and low-fat) and time (before and 30 and 120 min after intakes) and hunger as a covariate. In case of a significant time × fat interaction, post hoc paired *t* tests were calculated, and the significance level was Bonferroni corrected for multiple comparisons (corrected α level for paired *t* tests, $P = 0.016$). The association of hypothalamic and insular activity was investigated by the correlation of the percentage of CBF change of these 2 regions. For the examination of the hormonal influence to hypothalamic changes, correlation analyses were performed with the hypothalamic percentage of CBF change and the corresponding changes in insulin, glucose, and FFA concentrations.

RESULTS

Subject characteristics

Scores of the eating-behavior questionnaires resulted in very low scores for food craving, restraint eating, disinhibition during eating, or generally experienced hunger. In addition, subjects reported low eating-related, weight, or shape concerns (*see* Table S1 under "Supplemental data" in the online issue).

Hunger rating

As expected, yogurt intakes led to a significant time effect of subjective hunger ($F_{[1,20]} = 9.544$, $P < 0.05$). However, no difference between high- and low-fat yogurts was observed (**Figure 2**).

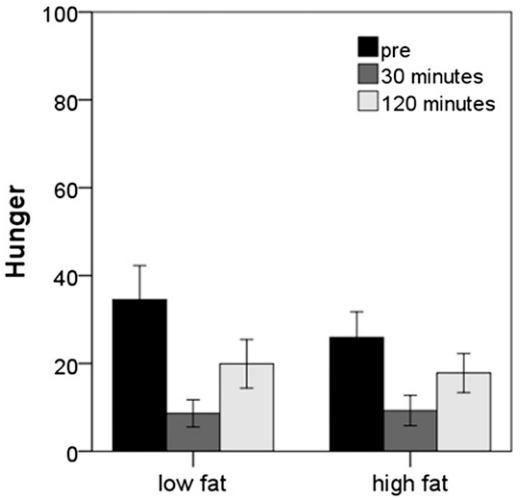

FIGURE 2. Mean (±SEM) hunger ratings at 3 time points for the high- and low-fat condition. Bar plots represent hunger rating on a 0–100 (0 = not at all hungry, 100 = very hungry) visual analog scale ($n = 11$). No significant differences were observed between high- and low-fat conditions in an ANOVA.

CBF

A regional CBF analysis of yogurt consumption revealed a main effect of fat ($F_{[1,20]} = 8.185$, $P = 0.01$) in the hypothalamus, which implied that the percentage of CBF change 30 and 120 min after consumption of yogurt with an 8% fat emulsions was significantly more reduced than that with the low-fat condition (**Figure 3**A). In addition, a significant time effect of yogurt ingestion on CBF was observed in the hypothalamus ($F_{[1,20]} = 5.880$, $P < 0.05$) that was independent of the fat content.

Time-dependent changes were also observed in the insular cortex ($F_{[1,20]} = 19.277$, $P < 0.001$). The insula revealed no main effect of fat but a fat × time interaction trend ($F_{[1,20]} = 5.346$, $P = 0.06$). Post hoc paired *t* tests revealed significant effects of the percentage of CBF change before to 120 min after low-fat yogurt ingestion to all other fat × time conditions [$t_{LF30}(10) = -4.53$, $P = 0.01$; $t_{HF30}(10) = -2.187$, $P = 0.05$; $t_{LF120}(10) = 5.096$, $P < 0.001$, with HF = high-fat and LF = low-fat to the time point 30 or 120 min] (Figure 3B).

To ensure that the observed differences in the hypothalamus were not due to unspecific or global effects, several control regions were also analyzed. In contrast to the hypothalamus, we found no fat specific modulation in the adjacent thalamus (*see* Figure S2A under "Supplemental data" in the online issue). Only a time trend was observed ($F_{[1,20]} = 3.831$, $P = 0.06$) with a decrease on both days. Similarly, we showed a time effect in the visual ($F_{[1,20]} = 21.282$, $P < 0.001$) and temporal cortex ($F_{[1,20]} = 12.296$, $P < 0.05$) that was due to a transient CBF decrease after 30 min and a later increase after 120 min that was independent of the fat content (*see* Figure S2B and C under "Supplemental data" in the online issue).

Interaction of hypothalamic and insular CBF

Both the hypothalamus and the insular cortex responded differentially to the fat content of the yogurts. Correlation analyses were performed to determine possible functional interactions between these areas. No significant correlations were observed for the low-fat condition. However, for the high-fat yogurt, a significant correlation between the percentage of CBF change in the insular cortex (pre to 120 min) and the hypothalamus (pre to 30 min) was detected ($R^2 = 0.486$, $P < 0.05$). Furthermore, a trend toward an association between the change in CBF in the insular cortex (pre to 120 min) and the hypothalamus (pre to 120 min) was observed ($R^2 = 0.280$, $P = 0.094$) (**Figure 4**).

Interaction of hypothalamic CBF and insulin concentrations

To evaluate possible associations between the hypothalamus and plasma insulin concentrations, correlations of the percentage of CBF change after 30 and 120 min with the change in insulin

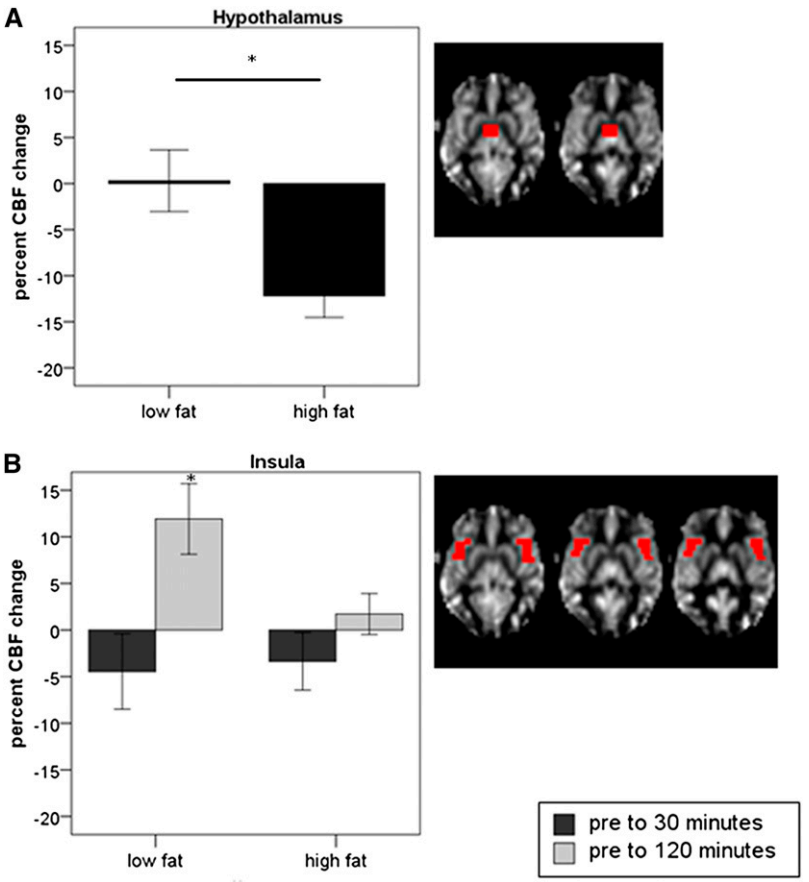

FIGURE 3. A (left): Percentage (±SEM) changes in hypothalamic CBF for consumption of low- and high-fat yogurts before (pre) and after fat intakes (percentage change from pre to 30 min and pre to 120 min combined). *Significant effect of fat, $P \leq 0.05$ (ANOVA; $n = 11$). B (left): Percentage (±SEM) changes in insular CBF for consumption of low- and high-fat yogurts. *Significant results of paired t tests, $P \leq 0.05$ (Bonferroni corrected). CBF after 120 min in the low-fat condition was significantly higher than in all other conditions ($n = 11$). A (right) and B (right): Region of interest selection depicted in red on the average CBF image of a representative subject. CBF, cerebral blood flow.

were analyzed. The change in insulin before to 30 min after yogurt ingestion revealed significant positive correlations with the percentage of CBF change of the hypothalamic activity pre to both post time points [$R^2 = 0.253$ ($P < 0.05$) and $R^2 = 0.246$ ($P < 0.05$), respectively] (**Figure 5**). A small increase of insulin after yogurt ingestion was associated with the suppression of CBF in the hypothalamus, whereas a higher increase of insulin was associated with an increase of CBF in the hypothalamus. Neither plasma glucose nor plasma FFAs showed a correlation with the CBF change.

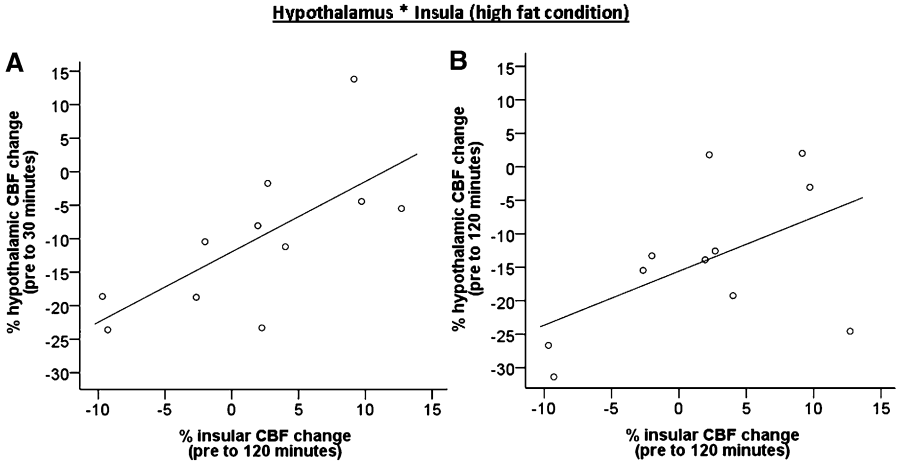

FIGURE 4. Interaction of hypothalamic and insular CBF. A: Scattergram of the percentage CBF changes before (pre) to 120 min after high-fat yogurt ingestion in the insular cortex and the hypothalamus (pre to 30 min). Correlation analysis revealed $R^2 = 0.486$ and $P < 0.05$ ($n = 11$). B: Scattergram of the percentage of CBF changes before (pre) to 120 min after high-fat yogurt ingestion in the insular cortex and the hypothalamus (pre to 120 min). Correlation analysis revealed $R^2 = 0.280$ and $P < 0.09$ ($n = 11$). CBF, cerebral blood flow.

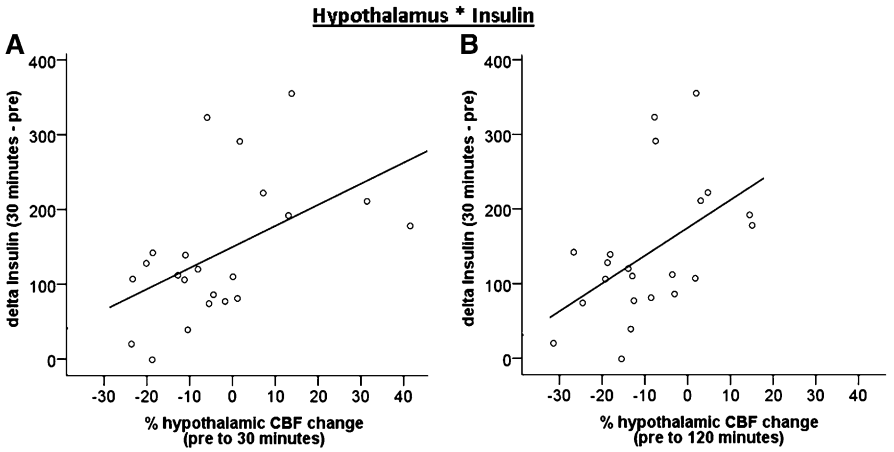

FIGURE 5. Interaction of hypothalamic CBF and insulin concentrations. A: Scattergram of the insulin concentration change (Δ: 30 min − pre) 30 min after yogurt ingestion and CBF changes in the hypothalamus (pre to 30 min). Correlation analysis revealed $R^2 = 0.253$ and $P < 0.05$ ($n = 22$). B: Scattergram of insulin concentration change (Δ: 120 min – pre) 30 min after yogurt ingestion and CBF changes in the hypothalamus (pre to 120 min). Correlation analysis revealed $R^2 = 0.246$ and $P < 0.05$ ($n = 22$). CBF, cerebral blood flow.

DISCUSSION

In this study, we investigated the effect of fat content on the hypothalamic and insular cortex by using CBF measurements. We showed a pronounced reduction of CBF in the hypothalamus in response to the intake of high-fat yogurt, whereas the low-fat condition showed no such effect. Study participants were investigated after an overnight fast; therefore, we assume a homeostatic drive to eat; thus, the homeostatic control region responded stronger to the high-fat yogurt. This effect might have been mediated by metabolic or endocrine changes (34). For example, satiation hormones send vagal afferent information to the brain through the nucleus tractus solitarius and area postrema in the caudal brainstem, dispending into the hypothalamus and forebrain (35). No difference of hunger rating was observed in our study. Furthermore, multivariate analyses were corrected for hunger, and a correlation analysis between the hypothalamic CBF change and the hunger change did not reveal any significant effects (*see* Supplementary Material under "Supplemental data" in the online issue). Therefore, our results for the hypothalamus seem to be independent from hunger. Functional MRI experiments showed an immediate increase in the hypothalamus after the application of a drop of a high-fat solution in the mouth (12, 13). However, Eldeghaidy et al (14) did not find changes in hypothalamic activity elicited by different fat emulsions. Opposed to our study, these results showed an immediate response to fat stimuli. We did not focus on the direct response but on the prolonged temporal dynamics of brain responses related to meal ingestion (ie, in the postprandial state).

We showed significant associations between the hypothalamic response and the major postprandial hormone insulin. Thus, the differential response could be partially mediated by this hormone. The insulin response showed a higher increase in the low-fat condition and a lower increase in the high-fat condition. These effects may have been based on delayed gastric emptying in the high-fat condition (36) or on a fat mediated hypothalamic regulation of insulin concentrations (37). This attenuated response of insulin was associated with a stronger suppression of CBF in the hypothalamus. A higher increase of insulin, which was present in the low-fat condition, was associated with a small increase in CBF in the hypothalamus (Figure 5). Insulin is not only a well-known modulator of homeostatic processes in the hypothalamus (38, 39) but also influences neural responses of other areas involved in food processing such as the fusiform gyrus after stimulation with food pictures (40, 41) as well as activity during rest (42–44).

In their study, Smeets et al (3) showed a strong decrease in the hypothalamic activity ~20 min after glucose ingestion, which depended on the amount of ingested glucose. Therefore, the decrease in the hypothalamic activity was presumably related to the different blood glucose concentrations. The ingestion of a higher amount of glucose also leads to higher insulin concentrations. However, in our study, we showed that a smaller increase in insulin after a yogurt intake correlated with a stronger suppression of response in the hypothalamus. This observation was, at first sight, opposed to the results of Smeets et al (3). A possible explanation could be that increased glucose concentrations, which were present in the study of Smeets et al (3), lead to a decrease in hypothalamic activity independent of insulin concentrations. In the case of normal glucose concentrations, which were obtained in the current study, insulin can become more important for the activity in the hypothalamus; low insulin concentrations lead to a decrease, and high insulin concentrations lead to an increase, in hypothalamic activity. Furthermore, the yogurts did not differ in amounts of carbohydrates. Therefore, we assume that the carbohydrate amounts cannot explain the difference in the insulin release.

In addition, we identified a trend effect in the insular cortex. A marked increase in activity was observed after 2 h, but only in the low-fat condition. The insula is involved in food perception (45–48) and gustation (49) and also shows an immediate response to oral viscosity after the ingestion of a fatty drink (12, 15). These studies measured the immediate effect of food perception. Our results show a delayed difference (120 min), indicating the differential processing of fat also in areas important for gustatory processing. In addition, states of hunger and satiation have been shown to influence insular cortex activity (7). This result is in line with our results. The insular activity correlated negatively with hunger in the low-fat condition. The stronger the decrease in hunger 30 and 120 min after yogurt intake, the stronger the increase in insular CBF after 120 min (*see* Supplementary

Material under "Supplemental data" in the online issue). Therefore, hunger seems to contribute to the prediction of insular activity. In contrast to the results in the hypothalamus, we did not show a significant correlation with insulin for insular activity.

An additional influencing factor might be the signal transmission from fat receptors in the mouth to the brain because, besides the primary tastes, fat is also represented at the tongue (50) and sends signals to the brain (51). Therefore, sensorial fat perception might have an additional pathway to influence brain functions.

No effect of fat was observed in the investigated control regions (ie, thalamus and temporal, and visual cortices). However, these regions showed a time effect after ingestion for both yogurts with a small decrease after 30 min and a subsequent increase after 120 min. We assume that these effects were not specific because the global values of the activity showed a similar pattern over time (*see* Figure S4 under "Supplemental data" in the online issue).

To address the interaction of the hypothalamus and the insular cortex, correlation analyses were performed. Correlations between CBF changes in the hypothalamus and the insular cortex were only significant in the high-fat condition. This result led to the possible interpretation that fat, and not food intake per se, modulates activity patterns and the interplay of the hypothalamus and the insula.

In conclusion, we showed major effects of dietary fat content on the hypothalamus and the insular cortex. Although these regions respond differentially, both regions seem to be generally influenced by ingested fat. Furthermore, dietary fat seems to modulate interactions between these regions. These findings indicate that high-fat and fat-reduced food products may influence the brain differently, which could be of great importance when choosing appropriate diets. Furthermore, our data can help the understanding of physiologic postprandial reactions that may contribute to end a meal and prevent overeating.

We thank Maike Borutta (University of Tübingen) and Luc Mertz (University of Hohenheim) for excellent technical assistance.

The authors' responsibilities were as follows—AF, AK, HP, JH, RV, and SF: design of the experiment; CK, KL, RV, SF, and SK: data collection; RV and SF: analysis of the data; MC, technical advice; AK, RV, and SF: manuscript draft; AF, HP, H-UH, KL, MC, MH, SK: critical revision of the manuscript; and AF, HP, H-UH, and JH: study supervision. None of the authors had a conflict of interest.

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