

# Influence of food images with different macronutrient compositions on serum ghrelin levels: Analysis in healthy males

Jan-Gerd Rademacher<sup>1</sup>  | Dirk Raddatz<sup>2</sup> | Thomas Ellrott<sup>3</sup>

<sup>1</sup>Department of Nephrology and Rheumatology, University Medical Center Göttingen, Göttingen, Germany

<sup>2</sup>Department of Gastroenterology, Gastrointestinal Oncology and Endocrinology, University Medical Center Göttingen, Göttingen, Germany

<sup>3</sup>Institute for Nutrition and Psychology, University Medical Center Göttingen, Göttingen, Germany

## Correspondence

Jan-Gerd Rademacher, Department of Nephrology and Rheumatology, University Medical Center Göttingen, Robert-Koch-Str. 40, 37075 Göttingen, Germany.  
Email: [jan-gerd.rademacher@med.uni-goettingen.de](mailto:jan-gerd.rademacher@med.uni-goettingen.de)

## Funding information

Open access funding enabled and organized by Projekt DEAL

## Abstract

**Objective:** Serum concentrations of the orexigenic hormone ghrelin fluctuate in anticipation of food intake. Moreover, presentation of food images causes an increase in serum ghrelin levels. Thus, the visual system may have a quantifiable role in the development of hunger via the endocrine system. The influence of macronutrient visualization on ghrelin has not yet been investigated.

**Methods:** In four separate sessions, ghrelin concentrations, insulin, and glucose levels were compared before and after the presentation of different pictures to 14 male participants. Pictures included neutral, non-food-related items or isocaloric dishes whose macronutrient composition corresponded predominately to protein/fat, simple carbohydrates, or complex carbohydrates.

**Results:** While pre/post ghrelin concentrations numerically increased in all sessions, significant increases were only observed following neutral and protein/fat pictures. The differences were not significant between food groups and compared to neutral images. Insulin levels decreased in all groups, but no significant differences were observed between sessions. The glucose concentrations were within the euglycemic range.

**Conclusion:** The results did not reproduce the induction of ghrelin secretion in different food images. Therefore, it is unclear whether the visual perception of food influences ghrelin secretion or whether separation into macronutrients changes the hormone response. Further research is required to differentiate the interactions of sensory-specific satiety.

## KEYWORDS

ghrelin, macronutrient composition, nutrition

This is an open access article under the terms of the Creative Commons Attribution-NonCommercial License, which permits use, distribution and reproduction in any medium, provided the original work is properly cited and is not used for commercial purposes.

© 2021 The Authors. Obesity Science & Practice published by World Obesity and The Obesity Society and John Wiley & Sons Ltd.

## 1 | INTRODUCTION

The absorption of different macronutrients leads to optimal regulation of endocrine messengers.<sup>1</sup> The orexigenic peptide hormone ghrelin plays an essential role in the development of hunger. Ghrelin is mainly secreted by the gastric fundus.<sup>2</sup> In humans, the activation of a peptide hormone by esterification with octanoic acid in acyl-ghrelin via the enzyme ghrelin O-acyltransferase (GOAT) occurs.<sup>3,4</sup> In contrast to the non-acylated derivative, acylated ghrelin can activate the growth hormone secretagogue receptor (GHS-R) due to its membrane permeability.<sup>5</sup> The physiological effects of ghrelin include numerous mechanisms that result in a positive energy balance.<sup>6,7</sup> Intravenous application of ghrelin increases food intake in humans, and chronic application causes weight gain in mice.<sup>8,9</sup> Physiological blood levels are subject to pulsatile fluctuations. During food restriction and before food intake, ghrelin concentrations rise and fall again after meal ingestion.<sup>10,11</sup> Thus, the subjectively perceived feeling of hunger and the expectation of a forthcoming meal correlate positively with ghrelin.<sup>12</sup> After weight loss, increased hormone levels correlate negatively with body mass index (BMI).<sup>13,14</sup>

Ghrelin and insulin demonstrate antagonistic effects. An injection of ghrelin leads to insulin suppression even after glucose infusion.<sup>15</sup> Ghrelin unfolds this effect directly in insulin-producing pancreatic  $\beta$ -cells.<sup>16</sup> In contrast, ghrelin-producing gastric cells express insulin receptors on their surfaces and are inhibited by insulin.<sup>17</sup> While ghrelin increases insulin sensitivity, obesity-associated insulin resistance with elevated insulin levels leads pathophysiologically to lower circulating ghrelin levels.<sup>18</sup> Furthermore, insulin lowers blood glucose, while ghrelin prevents hypoglycemia during extreme fasting periods through the release of growth hormone (GH).<sup>19</sup>

Visual perception of high-calorie food images leads to increased activity in the prefrontal cortex, reward centers, and limbic system.<sup>20,21</sup> In humans, intravenous application of ghrelin increases activity in these brain regions. Moreover, it was found that subjects could also remember food pictures better than a placebo group.<sup>22,23</sup> Schüssler et al. showed that the visual presentation of food induces endocrine secretion of total ghrelin levels. They examined the ghrelin levels of male volunteers before and after the presentation of food images and compared them to non-food-related photos. Significant differences were observed in the increase in ghrelin concentrations 30 min before and after the presentation. Insulin and leptin levels were measured simultaneously, but no significant differences were observed.<sup>24</sup>

Macronutrients have a specific influence on postprandial ghrelin levels, and ingestion of carbohydrates (CH) causes a pronounced short-term drop in ghrelin. Moreover, proteins result in long-term suppression, and lipid intake leads to a weak hormone reduction.<sup>25,26</sup>

Whether the visualization of different macronutrients affects ghrelin concentration has not been investigated. To specify the visual impact, this study divided the food pictures into meals that consisted predominantly of fat/protein, short-chain carbohydrates (SCCH), or long-chain carbohydrates (LCCH). The control session included

neutral pictures. Insulin and glucose levels were determined to evaluate glucose homeostasis and analyze the ghrelin interaction.

Based on solid ghrelin suppression after ingestion of CH, it was hypothesized that ghrelin is increased most by visualization of CH, whereas a minor increase was expected after the presentation of fat/protein.

## 2 | SUBJECTS AND METHODS

### 2.1 | Participants

Healthy male volunteers (college students) were recruited from various locations at Georg-August-University in Göttingen, Germany. The sex preference resulted from the pilot study protocol<sup>24</sup> and was followed to avoid additional variables. The test participants were informed in advance that they would be confronted with pictures, but they were not aware of the content. Each participant received an expense allowance.

A total of 18 males that were in the normal weight range (BMI 18.5–25 kg/m<sup>2</sup>) were included. All were aged between 20 and 30 years and had an omnivorous diet corresponding to the local culture. Followers of specific diets (e.g., low carb) or alternative diets (e.g., vegetarian, vegan) were excluded. All study participants were emmetropic or corrected to emmetropia. With the help of an anamnesis curve, it was confirmed that the participants did not suffer from food allergies, eating disturbances, or chronic illnesses. Participants were not exposed to shift work/sleep deprivation, drug abuse, or excessive physical training, as shown in Table S1.

The ethics committee of Georg-August-University of Göttingen approved the project. All participants provided written informed consent.

### 2.2 | Experimental paradigm

Each of the 18 volunteers underwent four different study sessions and were subjected to a photo presentation during each. Each presentation consisted of 50 different photos demonstrated three times on a computer screen for 15 min in random order. The four separate sessions were arranged in random order for each participant.

Study condition A: Non-food photos (control condition, e.g., office supplies).

Study condition B: Fat-protein meal (lipid/protein, e.g., steak with salad and dressing).

Study condition C: Short-chain carbohydrates (SCCH, e.g., dessert).

Study condition D: Long-chain carbohydrates (LCCH, e.g., pasta dish).

The dishes shown in study conditions B–D had a similar energy content of approximately 420 kcal. More than 50% of the energy

content was delivered by complex carbohydrates (starches) in study condition D. In study condition C, at least 50% of the energy content consisted of simple carbohydrates, such as sucrose, glucose fructose, lactose, and maltose. The macronutrients were distributed in this way for practicability: meals containing lipids are often combined with protein (e.g., nuts), while animal proteins often contain appropriate amounts of fat (e.g., red meat, fish). The presentation of predominantly fat-containing foods, such as oil or butter, as a distinct group would be questionable because they are not consumed in their pure form without a side dish and are probably considered less appetizing on their own. The meals were based on the current local food culture, for example, a sandwich, sausage with mustard and ketchup, or snacks like gummy bears, nuts, or a piece of cake.

Dishes were prepared and photographed in an appetizing manner based on the requirements of macronutrient composition. The neutral images in group A depicted non-food items such as clothing, office supplies, or musical instruments and were purchased from the collection of [www.fotolia.com](http://www.fotolia.com). The images are available from the authors.

### 2.3 | Pre-test

To prevent different image quality of the food presented, 15 male volunteers rated the food photos in terms of their attractiveness using a scale from one (very tasty) to seven (not tasty). The pre-test participants rated all 150 food images, which were viewed in a mixed order. In this way, an equal attractiveness of the food shown could be guaranteed. Since the volunteers were aware of the images, all 15 participants in the pre-test were excluded from further study. This procedure was adopted in the pre-test of the pilot study.<sup>24</sup>

### 2.4 | Study procedure

The volunteers were fasted at 8:15 AM at all four appointments. Before breakfast, participants had to confirm that they had not eaten or consumed caloric beverages since the previous evening. At 8:30 AM, a traditional breakfast was served in the form of two buns with two portions of butter, a serving of jam, and two servings of cold cuts, alternately consisting of cheese or sausage so that the total calorie content of the meal was approximately 650 kcal. The consumption of water, tea, and coffee do not affect ghrelin concentrations.<sup>27-29</sup> Therefore, tea and water drinking were allowed during the morning, with an additional cup of coffee at breakfast.

Since the blood draws by Schüssler et al. prior to 10:00 AM were not considered in their statistical results, blood samples were collected eight times between 10:00 and 11:20 AM (10:00, 10:15, 10:30, 10:40, 10:50, 11:00, 11:10, and 11:20). The interval between each of the appointments was between three and eight days. The time schedule is shown in Figure 1.

Between 9:30 and 9:45 AM, an intravenous catheter was placed into the elbow vein of each participant to perform blood sampling at

the given time. Blood was drawn via an attached three-way stopcock. Moreover, 5 ml of blood was aspirated and discarded before each measurement to avoid dilution of the samples. One sample intended for ghrelin and insulin measurement (7 ml) and one sample for glucose level determination (3.1 ml) were taken.

At 10:30 AM, the participants watched one of the four photo presentations on a 24 in. (61 cm) diagonal screen. The 50 images of each session were randomly presented three times over 15 min and changed every 6 s.

The test room was free of confounding factors, such as visual or acoustic stimuli, and the participants could read only neutral, non-food-related text material before and after the presentation. For this purpose, magazines were provided for entertainment after the removal of food-related content.

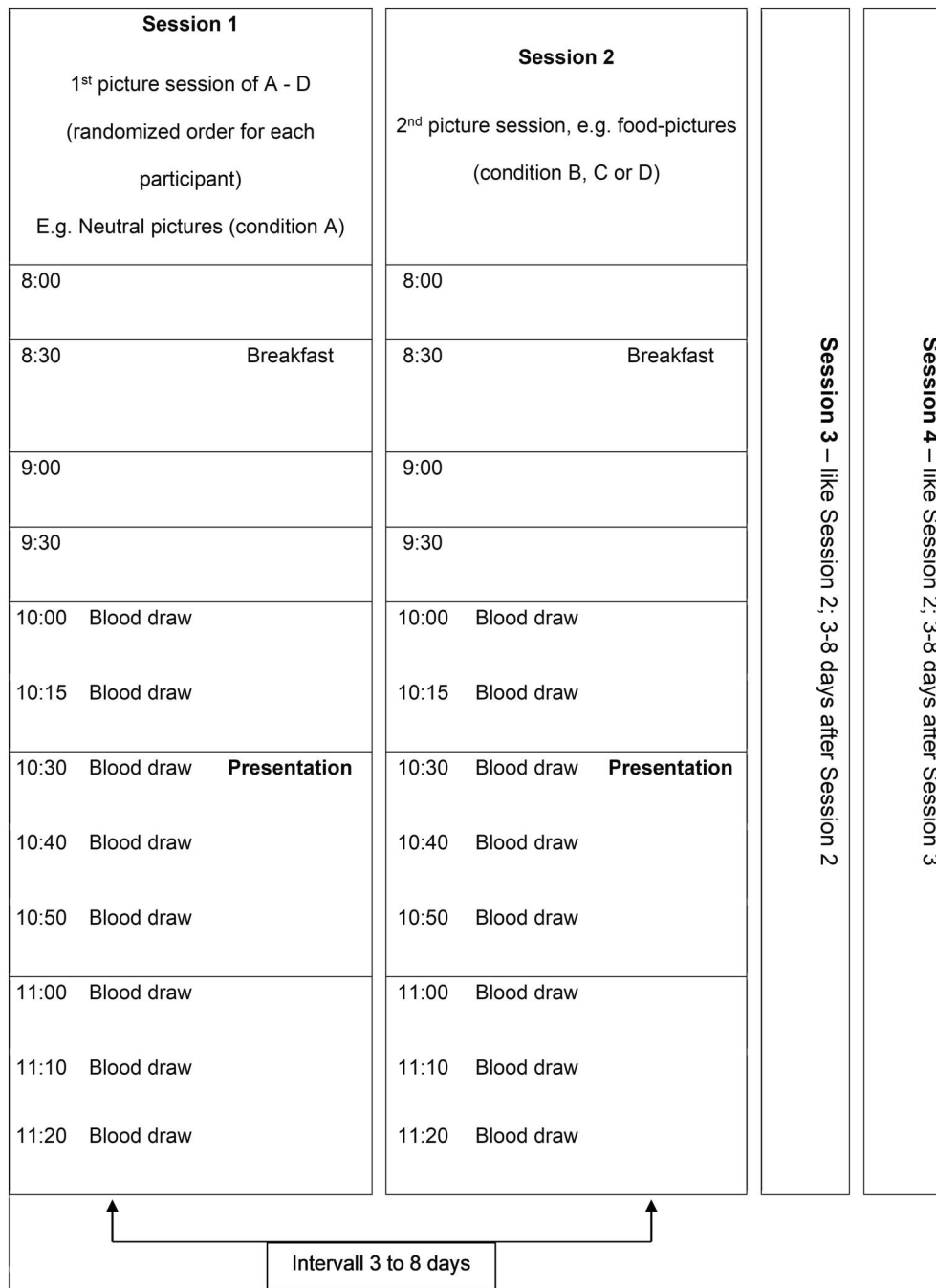
### 2.5 | Determination of laboratory parameters

After labeling with the subject number, session type, and time of collection, serum samples intended for hormone level measurement were transported on ice and stored at  $-80^{\circ}\text{C}$  until the final sessions. The determination of glucose levels was performed in the central laboratory of the University Medical Center Göttingen after each session and the analysis of ghrelin and insulin in a dedicated laboratory for gastrointestinal hormones at the University Medical Center Göttingen. The total ghrelin levels were measured using serum samples with a radioimmunoassay (Mediagnost "Ghrelin RIA R90") based on a polyclonal antiserum. The inter- and intra-assay coefficients of variation (CV) were 8.2% and 5.3%, respectively. Insulin levels were measured using an immunoradiometric assay (Beckmann Coulter "Insulin(e) IRMA kit IM3210"). The inter- and intra-assay CVs were 3.4% and 4.3%, respectively. Plasma glucose levels were stabilized in Sarstedt's "GlucoEXACT" samples that contained a fluoride citrate solution.

### 2.6 | Statistical analysis

The sample size was calculated based on a pilot study.<sup>24</sup> A certain drop-out rate was considered. Therefore, 18 participants were recruited at the beginning of the study, who appeared on all study dates. However, a defective laboratory material damaged the serum samples of six participants. Thus, complete blood results required for the area under the curve (AUC) determination were available for 14 volunteers, while the measurement at 10:40 AM, which was not included in the evaluation of the pre-post interval, could only be determined in 12 of the 14 participants. The data were analyzed using Excel (Office 365, Microsoft Corp.), SPSS version 25.0 (SPSS Inc., IBM Corp.), and Prism version 9.0 (GraphPad Software Inc.).

In addition to the mean, the AUC for ghrelin and insulin was determined according to the trapezoidal rule. Therefore, the difference between the concentrations at the beginning and end of the interval multiplied by the time interval defines the AUC. The mean



**FIGURE 1** Procedure of study days. Participants received a traditional breakfast at 8:30 AM hormone measurements were performed using repeated blood draws from 10:00 to 11:20 AM. Between 10:30 and 10:45 AM, one of the four picture collections was presented to the participants. After three to eight days, the remaining photo presentations were conducted. The order of the presentations was randomized for each participant

values and AUC were collected for the period before the presentation (AUC 1: 10:00–10:30) and after the presentation (AUC 2: 10:50–11:20). The quotient of the AUC 2 and AUC 1 defined the change in hormone concentration after visual stimulation as a dimensionless value of the interindividual serum levels.

The data obtained were analyzed for normal distribution using the Kolmogorov-Smirnov and Shapiro-Wilk tests. Pre/post

comparisons of the parametric data were performed using a paired *t*-test. For non-parametric values, the Wilcoxon signed-rank test was used. To test for differences between groups, a repeated-measures analysis of variance (ANOVA) with Tukey's multiple comparison test or Friedman test as post hoc tests were performed, as appropriate. Differences between groups were considered statistically significant at an alpha level of  $p < 0.05$ .

### 3 | RESULTS

#### 3.1 | Study cohort

The 14 male participants had a mean age of  $23.40 \pm 2.65$  years. The mean height was  $1.85 \pm 0.06$  m, and the mean weight was  $79.40 \pm 8.83$  kg. This equated to a mean BMI of  $23.20 \pm 1.42$  kg/m<sup>2</sup> (Table 1).

#### 3.2 | Pre-test

The attractiveness of the pictures was tested in advance. The letters A-D were used for the respective study conditions. The scale ranged from one (very tasty) to seven (not tasty). The mean value was  $3.39 \pm 1.10$  for study condition B,  $3.41 \pm 0.83$  for study condition C, and  $3.74 \pm 0.94$  for photo group D. The ANOVA showed no differences between the food sessions ( $F(2, 147) = 2.018, p = 0.137$ ). Therefore, an equivalence of the attractiveness of the pictures was assumed.

## 4 | HORMONE MEASUREMENTS

#### 4.1 | Ghrelin

Ghrelin concentrations increased in all sessions, and maximum levels were measured at 11:20 AM. The lowest values of total serum ghrelin were in group B-D at 10:15 and in group A at 10:30 AM. Figure 2A shows the mean values over the four sessions. Few measurements in different participants and sessions showed lower levels after the picture sessions (Figure S2). Except for the measurements at 10:40 AM ( $n = 12$ ), the results of all 14 participants were included in the diagram. The largest ghrelin increase with a pre/post ratio of  $1.049 \pm 0.07$  was observed in the non-food condition A, followed by the fat/protein photos with  $1.042 \pm 0.07$ . The presentation of carbohydrate pictures was accompanied by an increase of  $1.027 \pm 0.06$  in LCCH and  $1.021 \pm 0.07$  in SCCH. The AUC of the ghrelin concentrations was calculated analogously to the pre-and post-presentation mean values. Similar to the mean values, increasing AUC1/AUC2 quotients were observed in all groups. The ghrelin-AUC 2 in session B increased by  $1.049 \pm 0.07$  compared to AUC 1, slightly more than in group A ( $1.047 \pm 0.06$ ). In addition, the ratios of LCCH ( $1.029 \pm 0.07$ ) and SCCH ( $1.021 \pm 0.06$ ) represented the lowest increase in the AUC. All values and pre-/post-quotients were distributed parametrically. The paired *t*-test showed a significant increase in ghrelin levels in the non-food group (mean:  $p = 0.02$ ; AUC:  $p = 0.15$ ) as well as in the fat/protein group (mean:  $p = 0.03$ ; AUC  $p = 0.019$ ). The carbohydrate groups showed no significant increase in ghrelin concentration with respect to the mean value and AUC changes (Table 2). RM-ANOVA did not indicate any differences in the mean quotients ( $F(3, 39) = 0.8581, p = 0.47$ ) or AUC quotients ( $F(3, 39) = 0.8642,$

TABLE 1 Cohort statistics

	Mean	Range	SD	N
Age (years)	23.43	(20–29)	2.65	14
Weight (kg)	79.43	(69–100)	8.83	14
Height (cm)	1.85	(1.75–2.00)	0.06	14
BMI (kg/m <sup>2</sup> )	23.20	(20.94–25)	1.42	14

Note: The age and bodyweight-specific characteristics of the study population. All participants were in the average weight range.

$p = 0.47$ ) between the sessions. A *t*-test comparison of the average food quotients A-C versus non-food quotients did not detect any statistically significant differences.

#### 4.2 | Insulin

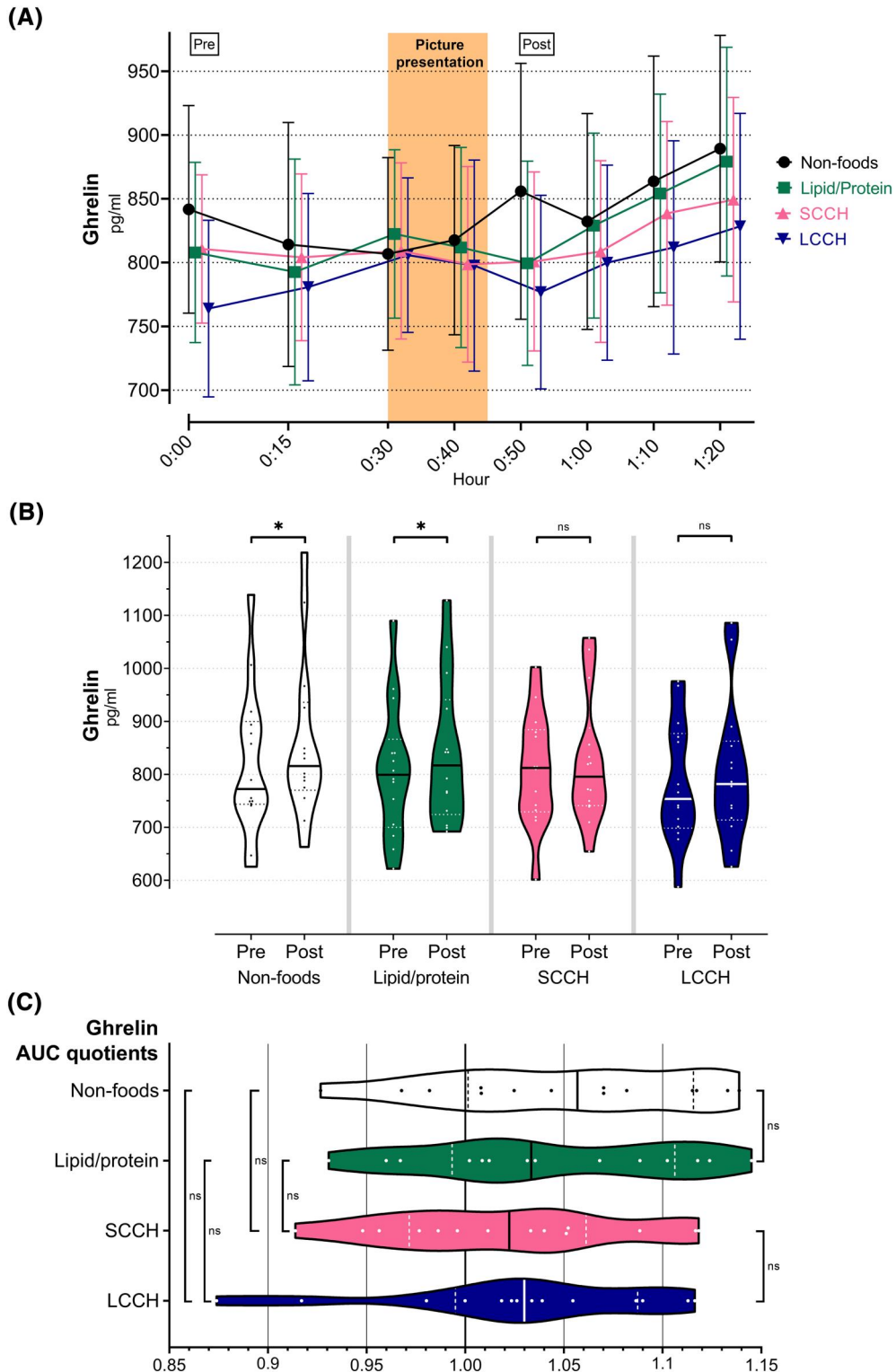
Insulin concentrations dropped in all groups over the measurement time, as demonstrated in Figure 3A. The lowest values were thus determined at 11:20. The quotients of the insulin mean values pre- and post-presentation showed decreasing values in each group (quotients <1), with maximum decreases in LCCH of  $0.428 \pm 0.14$  and minimum decreases in SCCH of  $0.623 \pm 0.10$  compared to baseline (Figure 3B). The decreases in the AUC quotients for insulin were comparable to those of the mean values. Group C had the smallest reduction in the AUC (Quot.  $0.61 \pm 0.38$ ), group D showed a decrease in the AUC 2 to  $0.41 \pm 0.14$  of the initial value. In sessions A and B, an approximate halving of the AUC after photo presentation was calculated (Quot. A  $0.51 \pm 0.31$ ; Quot. B  $0.49 \pm 0.27$ ).

The data were non-parametric, and the Wilcoxon signed-rank test indicated a significant insulin decrease in the mean and AUC post scores in all sessions ( $p = 0.001$ , Table 2). The Friedman test of differences compared the insulin quotients between sessions. Although the mean rank was highest in group C, the testing showed a non-significant Chi-square value of 3.857 for AUC (Figure 3C,  $p = 0.277$ ) and 5.74 for the mean scores ( $p = 0.125$ ) between A-D.

The correlations between insulin and ghrelin were not statistically significant (Table 2).

#### 4.3 | Glucose

Glucose levels were consistently in the normoglycemic range and fell moderately after an initial rise (Figure 3D). The lipid-protein images had a pre/post blood sugar quotient of  $0.896 \pm 0.03$  ( $T = 3.4, p = 0.005$ ). Sessions A and D were  $0.920 \pm 0.03$  ( $T = 2.39, p = 0.033$ ) and  $0.923 \pm 0.03$  ( $T = 2.72, p = 0.17$ ), respectively. The lowest decreases in glucose concentration were in SCCH with a quotient of  $0.952 \pm 0.03$  ( $T = 1.9, p = 0.079$ ). The mean quotients had a non-parametric distribution. Friedman's test did not detect differences between the scores of the mean quotients A-D.



**FIGURE 2** (A) The mean ghrelin concentrations with 95% CI between the pre (0:00–0:30) and post (0:50–1:20) presentation interval. The ghrelin levels increased in all the sessions. Picture presentations were conducted between 0:30 and 0:45. Non-food and lipid/protein hormone changes after pictures were significant, while ghrelin response was minor after carbohydrate sessions. (B) Violin plots of the ghrelin concentrations before and after the presentation of food pictures. Clustered values lead to an increasing curvature; the absence of values results in plot thinning. In contrast to the arithmetic mean (Table 2), the median ghrelin concentration in the SCCH group was lower after the presentation. (C) Inter-group comparison via ANOVA with repeated measurements found no differences among the AUC quotients. A  $p$  value of  $<0.05$  was considered statistically significant (\*). Abbreviations: ANOVA, analysis of variance; AUC, area under the curve; CI, confidence interval; LCCH, long-chain carbohydrates; ns, not significant; SCCH, short-chain carbohydrates

TABLE 2 Ghrelin and insulin concentrations and correlation before and after the picture presentations

Session	Variables	Ghrelin		Insulin		Correlation Ghrelin-Insulin Quot
		Mean	SD	Mean	SD	
A-Non-foods	Mean A1	820.93 pg/ml	±138.92	32.31 mIU/L	±18.45	$r = -0.43, p = 0.125$
	Mean A2	860.29 pg/ml	±153.70	15.72 mIU/L	±11.17	
		$t(13) = -2.74,$ $p = 0.02$		$Z = -3.23,$ $p = 0.001$		
	Quot Mean A	1.049	±0.07	0.513	±0.27	
B - Lipid/ Protein	Mean B1	807.69 pg/ml	±127.16	30.33 mIU/L	±15.36	$r = -0.12, p = 0.69$
	Mean B2	840.46 pg/ml	±135.45	13.38 mIU/L	±7.20	
		$t(13) = -2.37,$ $p = 0.03$		$Z = -3.23,$ $p = 0.001$		
	Quot Mean B	1.042	±0.07	0.48	±0.26	
C - SCCH	Mean C1	807.98 pg/ml	±105.73	29.85 mIU/L	±19.39	$r = -0.12, p = 0.678$
	Mean C2	824.39 pg/ml	±121.63	16.03 mIU/L	±9.54	
		$t(13) = -1.15,$ $p = 0.27$		$Z = -2.86,$ $p = 0.001$		
	Quot Mean C	1.021	±0.06	0.623	±0.37	
D - LCCH	Mean D1	783.52 pg/ml	±114.72	33.09 mIU/L	±12.03	$r = -0.44, p = 0.115$
	Mean D2	804.29 pg/ml	±133.97	13.61 mIU/L	±5.16	
		$t(13) = -1.40,$ $p = 0.18$		$Z = -3.30,$ $p = 0.001$		
	Quot Mean D	1.027	±0.07	0.428	±0.14	

Note: Comparison of the ghrelin concentrations before (1) and after (2) the picture presentations. The Wilcoxon signed-rank test was used for the non-parametric insulin values. Pearson's correlation coefficient was used to calculate the ghrelin/insulin quotients of each session. A  $p$  value of  $<0.05$  was considered statistically significant.

Abbreviations: LCCH, long-chain carbohydrates; Quot, quotient; SCCH, short-chain carbohydrates; SD, standard deviation.

## 5 | DISCUSSION

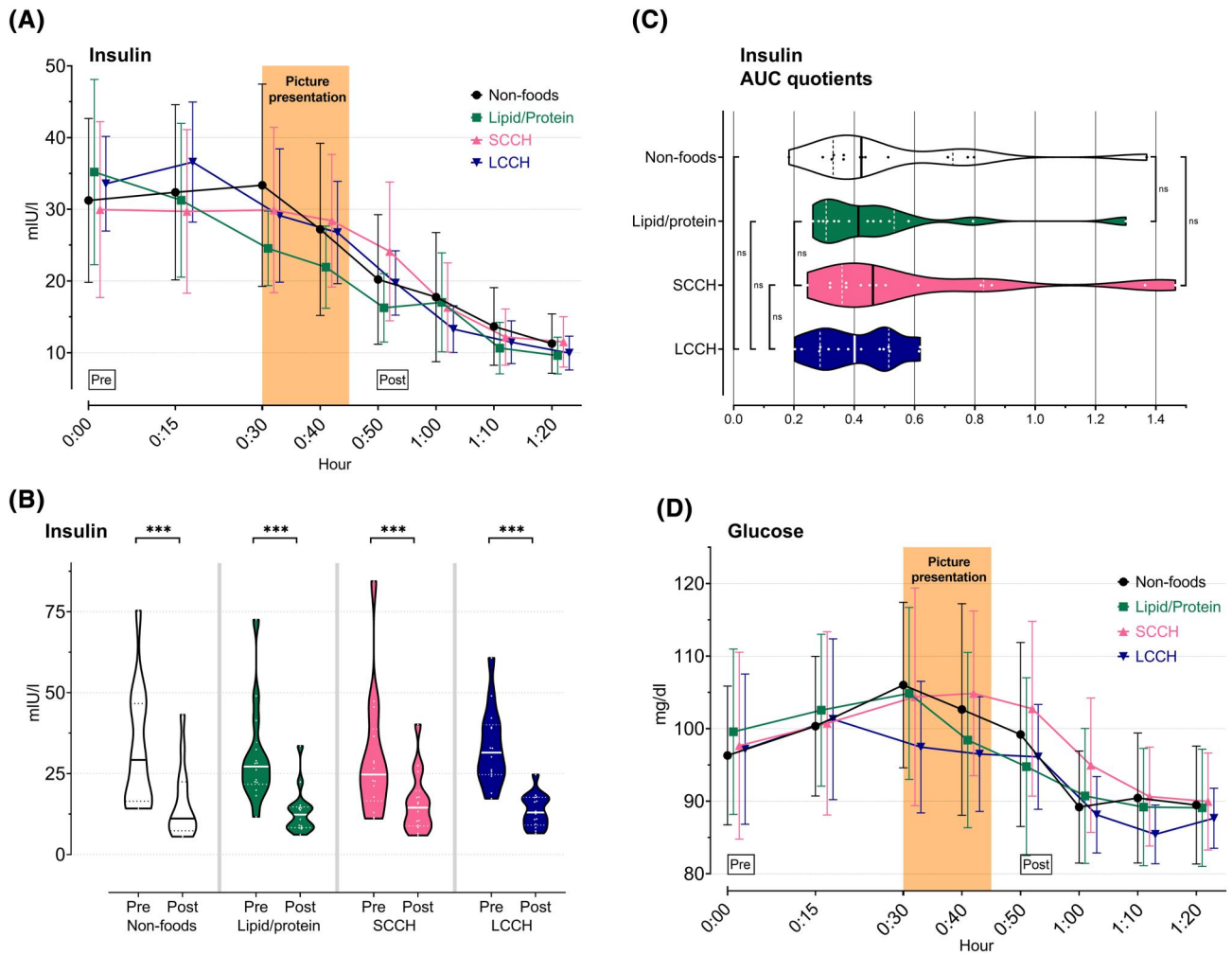
Ghrelin concentrations increased in all sessions, which was plausible considering the food-free interval after the previous breakfast. The differences pre- and post-presentation were only statistically significant in the neutral session A and lipid/protein group B. Unexpectedly, there was no difference in ghrelin levels between non-food and macronutrients or within the three food groups observed.

For insulin, a significant decrease in hormone concentration was noted after image presentation. These concentrations reflect a physiological postprandial decrease. The comparison of the pre- and post-presentation quotients between the sessions showed no significant differences. The non-parametric distribution of the insulin and glucose concentrations resulted from the small study cohort. Although not significant, there was a physiological trend toward a negative correlation between ghrelin and insulin.<sup>15</sup> The blood glucose levels were in the normoglycemic range and decreased throughout the study. As expected in the late postprandial phase, 90 min after the initiation of breakfast, the glucose concentrations showed a slight decrease in the macronutrient and non-food groups, and no significant differences were found between the groups. However, the

values were within the physiological range, and insulin concentrations decreased reactively.

Since non-foods resulted in the most substantial increase in ghrelin concentrations, the results, therefore, allow two primary antithetical considerations: either there is no increase in ghrelin after viewing meal-based pictures, or the division into nutrient groups biases the induction of ghrelin secretion after "balanced" food cues.

Taste seems to be decisive in the choice of food. Associations with expediency, health promotion, and pleasure through food are of secondary importance.<sup>30</sup> There have been some indications that consuming these favorite, good-tasting foods leads to greater satisfaction and satiety.<sup>31</sup> Test subjects showed lower ghrelin levels after consuming a presumably high-calorie milkshake than after consuming a supposedly "healthier" shake with an identical calorie content.<sup>32</sup> An evaluation of the food photos after the presentations by the participants would have made it possible to comprehend the subjective palatability of the pictures and thus categorize personal favorites or antipathies. In the context of hunger and anticipation of an upcoming meal, fasting experiments showed how the serum concentrations of ghrelin change pulsatile over the day and increase before the usual



**FIGURE 3** Insulin (A) and glucose (D) concentrations decreased after the picture presentations. The insulin values were significantly different in the intra-group comparison using the Wilcoxon signed-rank test (B). Friedman's test demonstrated no differences between the insulin AUC quotients of the four sessions (C). Abbreviations: AUC: area under the curve; LCCH: long-chain carbohydrates; ns: not significant; SCCH: short-chain carbohydrates

mealtimes. Hence, there is a conditioning of the brain hormone axis.<sup>12,33</sup> This study was conducted 2 h after breakfast and approximately 2 h before habitual lunch. The influence of the usual mealtime on the measurements could be considered a confounder, even though the time was comparable to that of the pilot study.

Regarding the influence of food intake on ghrelin serum levels, there are different patterns after ingesting different macronutrients: protein is a potent long-term suppressor of ghrelin over 6 h, while lipid intake leads to a slight decrease. The temporal dimension is particularly relevant for carbohydrates as it causes a biphasic course of the ghrelin level: a more pronounced drop in hormone levels is followed by an early and more substantial increase than after uptake of proteins or lipids.<sup>25,34</sup> Ghrelin suppression after CH is thought to be caused by hyperglycemic insulin secretion.<sup>7</sup> Some authors have found no relevant changes in ghrelin levels after lipid ingestion.<sup>35,36</sup> Among the food pictures, group B showed the most substantial increase in ghrelin levels. Arguably, proteins and lipids were visualized together, which may obscure opposing or attenuated ghrelin

stimulation. Therefore, it is impossible to predict the influence of pictures of one of the two macronutrients on ghrelin secretion. Overcoming this issue is challenging because regional food and common grocery items often contain both of these nutrients. In addition, the attractiveness of the food would probably be different. For example, a low-fat piece of meat may appear more appetizing than a predominantly high-fat or oily food.

The CH (sessions C and D) presentation had the most negligible influence on the ghrelin level, which speaks against a transfer of the findings of postprandial hormone changes to the visually induced ghrelin concentrations. Furthermore, the lower ghrelin levels in all food groups compared to the non-food images are conflicting. A more recent study observed an increase in total ghrelin, but not acyl-ghrelin, 30 min after presenting different food images. These pictures were of mixed foods with different calorie contents and macronutrient composites.<sup>37</sup>

Irrespective of its macronutrient composition, an ingested meal loses its attractiveness in taste or appearance in favor of another



food.<sup>38–40</sup> This sensory-specific satiety (SSS) is probably an evolutionary protective mechanism that prevents unbalanced nutrition. On the other hand, this phenomenon leads to increased food intake due to more significant variations in food and taste, whereas the presentation of homogeneous foods and flavors results in reduced consumption.<sup>41</sup> This is in line with the findings of hedonic regulation, which are modulated by sensory influences. It is unknown whether SSS exerts an influence on the ghrelin system. Although we aimed to compare different macronutrients, the images may have been perceived in one-sided taste categories. Images of homogeneous food groups repeated three times for 15 min could have led to a saturated perception. Ghrelin concentrations did not change noticeably. The low ghrelin increase in CH groups C and D was conspicuous, which could be considered similar in taste in this context. Session C, with the simple CH, could all be considered sweet foods. The protein/lipid images generally were of savory foods, but these were characterized by compositions of meat/sausage, fish, eggs, nuts, legumes, milk, and potato products, which could lead to a feeling of greater diversity.

There are limitations to this study. First, the sample size was small, and the study only included males. Second, the degree of saturation was also not systematically examined after breakfast, but it should be noted that the meal quantity and type of food provided were typical for the local culture. Furthermore, meals consumed at night or directly before the study could not be verified.

This study also has a number of strengths. First, repeated measurements were performed on a strict schedule. Second, hormone concentrations were compared among randomized non-food and different food picture sessions.

Our findings question the absolute picture-induced ghrelin response and contribute to a better understanding of the hormone response to isolated or balanced macronutrients. Thus, protein-based images might be more stimulating than CH for ghrelin secretion. The influence of SSS on ghrelin response should be examined in future studies.

## 6 | CONCLUSION

The induction of ghrelin secretion after the presentation of food pictures could not be verified in the current study. However, the induction of ghrelin secretion may have been changed due to the division into macronutrients. The extent to which SSS influences serum ghrelin concentration remains to be determined. Thus, further research is required to examine this phenomenon.

## ACKNOWLEDGMENTS

Thomas Ellrott and Jan-Gerd Rademacher conceived and carried out the experiments, and Thomas Ellrott and Jan-Gerd Rademacher conceived the experiments and analyzed the data. Jan-Gerd Rademacher performed the experiments. All authors were involved in the data interpretation, literature search, writing of

the paper, and final approval of the submitted and published versions.

Open access funding enabled and organized by Projekt DEAL.

## CONFLICT OF INTEREST

The authors do not have any conflicts of interest.

## ORCID

Jan-Gerd Rademacher  <https://orcid.org/0000-0001-9764-3421>

## REFERENCES

- González Olmo BM, Butler MJ, Barrientos RM. Evolution of the human diet and its impact on gut microbiota, immune responses, and brain health. *Nutrients*. 2021;13:196. <https://doi.org/10.3390/nu13010196>
- Date Y, Kojima M, Hosoda H, et al. Ghrelin, a novel growth hormone-releasing acylated peptide, is synthesized in a distinct endocrine cell type in the gastrointestinal tracts of rats and humans. *Endocrinology*. 2000;141:4255–4261.
- Gutierrez JA, Solenberg PJ, Perkins DR, et al. Ghrelin octanoylation mediated by an orphan lipid transferase. *Proc Natl Acad Sci U S A*. 2008;105:6320–6325.
- Yang J, Brown MS, Liang G, Grishin NV, Goldstein JL. Identification of the acyltransferase that octanoylates ghrelin, an appetite-stimulating peptide hormone. *Cell*. 2008;132:387–396.
- Staes E, Absil P-A, Lins L, et al. Acylated and unacylated ghrelin binding to membranes and to ghrelin receptor: towards a better understanding of the underlying mechanisms. *Biochim Biophys Acta*. 2010;1798:2102–2113.
- Müller TD, Nogueiras R, Andermann ML, et al. Ghrelin. *Mol Metab*. 2015;4:437–460.
- Mani BK, Shankar K, Zigman JM. Ghrelin's relationship to blood glucose. *Endocrinology*. 2019;160:1247–1261. <https://doi.org/10.1210/en.2019-00074>
- Wren AM, Seal LJ, Cohen MA, et al. Ghrelin enhances appetite and increases food intake in humans. *J Clin Endocrinol Metab*. 2001;86:5992.
- Wren AM, Small CJ, Abbott CR, et al. Ghrelin causes hyperphagia and obesity in rats. *Diabetes*. 2001;50:2540–2547.
- Cummings DE, Purnell JQ, Frayo RS, Schmidova K, Wisse BE, Weigle DS. A preprandial rise in plasma ghrelin levels suggests a role in meal initiation in humans. *Diabetes*. 2001;50:1714–1719.
- Tschöp M, Wawarta R, Riepl RL, et al. Post-prandial decrease of circulating human ghrelin levels. *J Endocrinol Invest*. 2001;24:RC19–RC21.
- Frecka JM, Mattes RD. Possible entrainment of ghrelin to habitual meal patterns in humans. *Am J Physiol Gastrointest Liver Physiol*. 2008;294:G699–G707.
- Cummings DE, Weigle DS, Frayo RS, et al. Plasma ghrelin levels after diet-induced weight loss or gastric bypass surgery. *N Engl J Med*. 2002;346:1623–1630.
- Shiyya T, Nakazato M, Mizuta M, et al. Plasma ghrelin levels in lean and obese humans and the effect of glucose on ghrelin secretion. *J Clin Endocrinol Metab*. 2002;87:240–244.
- Tong J, Prigeon RL, Davis HW, et al. Ghrelin suppresses glucose-stimulated insulin secretion and deteriorates glucose tolerance in healthy humans. *Diabetes*. 2010;59:2145–2151.
- Dezaki K, Kakei M, Yada T. Ghrelin uses Galphai2 and activates voltage-dependent K<sup>+</sup> channels to attenuate glucose-induced Ca<sup>2+</sup>-signaling and insulin release in islet beta-cells: novel signal transduction of ghrelin. *Diabetes*. 2007;56:2319–2327.

17. Gagnon J, Anini Y. Insulin and norepinephrine regulate ghrelin secretion from a rat primary stomach cell culture. *Endocrinology*. 2012;153:3646-3656.
18. McLaughlin T, Abbasi F, Lamendola C, Frayo RS, Cummings DE. Plasma ghrelin concentrations are decreased in insulin-resistant obese adults relative to equally obese insulin-sensitive controls. *J Clin Endocrinol Metab*. 2004;89:1630-1635.
19. Goldstein JL, Zhao T-J, Li RL, Sherbet DP, Liang G, Brown MS. Surviving starvation: essential role of the ghrelin-growth hormone axis. *Cold Spring Harb Symp Quant Biol*. 2011;76:121-127.
20. De Silva A, Salem V, Matthews PM, Dhillon WS. The use of functional MRI to study appetite control in the CNS. *Exp Diabetes Res*. 2012;2012:1-13. <https://doi.org/10.1155/2012/764017>
21. Stoeckel LE, Weller RE, Cook EW, Twieg DB, Knowlton RC, Cox JE. Widespread reward-system activation in obese women in response to pictures of high-calorie foods. *Neuroimage*. 2008;41:636-647.
22. Goldstone AP, Prechtl CG, Scholtz S, et al. Ghrelin mimics fasting to enhance human hedonic, orbitofrontal cortex, and hippocampal responses to food. *Am J Clin Nutr*. 2014;99:1319-1330.
23. Malik S, McGlone F, Bedrossian D, Dagher A. Ghrelin modulates brain activity in areas that control appetitive behavior. *Cell Metab*. 2008;7:400-409.
24. Schüssler P, Kluge M, Yassouridis A, Dresler M, Uhr M, Steiger A. Ghrelin levels increase after pictures showing food. *Obes*. 2012;20:1212-1217.
25. Foster-Schubert KE, Overduin J, Prudom CE, et al. Acyl and total ghrelin are suppressed strongly by ingested proteins, weakly by lipids, and biphasically by carbohydrates. *J Clin Endocrinol Metab*. 2008;93:1971-1979.
26. Koliaki C, Kokkinos A, Tentolouris N, Katsilambros N. The effect of ingested macronutrients on postprandial ghrelin response: a critical review of existing literature data. *Int J Pept*. 2010;2010:1-9. <https://doi.org/10.1155/2010/710852>
27. Gavrieli A, Yannakoulia M, Fragopoulou E, et al. Caffeinated coffee does not acutely affect energy intake, appetite, or inflammation but prevents serum cortisol concentrations from falling in healthy men. *J Nutr*. 2011;141:703-707.
28. Haghghatdoost F, Nobakht MGhBF, Hariri M. Effect of green tea on plasma leptin and ghrelin levels: a systematic review and meta-analysis of randomized controlled clinical trials. *Nutrition*. 2018;45:17-23.
29. Tschöp M, Smiley DL, Heiman ML. Ghrelin induces adiposity in rodents. *Nature*. 2000;407:908-913.
30. Phan UTX, Chambers E. Motivations for choosing various food groups based on individual foods. *Appetite*. 2016;105:204-211.
31. Mattes MZ, Vickers ZM. Better-liked foods can produce more satiety. *Food Qual Prefer*. 2018;64:94-102.
32. Crum AJ, Corbin WR, Brownell KD, Salovey P. Mind over milkshakes: mindsets, not just nutrients, determine ghrelin response. *Health Psychol*. 2011;30:424-429.discussion 430-431.
33. Natalucci G, Riedl S, Gleiss A, Zidek T, Frisch H. Spontaneous 24-h ghrelin secretion pattern in fasting subjects: maintenance of a meal-related pattern. *Eur J Endocrinol*. 2005;152:845-850.
34. Tannous dit El Khoury D, Obeid O, Azar ST, Hwalla N. Variations in postprandial ghrelin status following ingestion of high-carbohydrate, high-fat, and high-protein meals in males. *Ann Nutr Metab*. 2006;50:260-269.
35. Tentolouris N, Kokkinos A, Tsigos C, et al. Differential effects of high-fat and high-carbohydrate content isoenergetic meals on plasma active ghrelin concentrations in lean and obese women. *Horm Metab Res*. 2004;36:559-563.
36. Poppitt SD, Leahy FE, Keogh GF, et al. Effect of high-fat meals and fatty acid saturation on postprandial levels of the hormones ghrelin and leptin in healthy men. *Eur J Clin Nutr*. 2006;60:77-84.
37. Duszka K, Gregor A, Reichel MW, Baierl A, Fahrngruber C, König J. Visual stimulation with food pictures in the regulation of hunger hormones and nutrient deposition, a potential contributor to the obesity crisis. *PLoS One*. 2020;15:e0232099. <https://doi.org/10.1371/journal.pone.0232099>
38. Rolls BJ, Rolls ET, Rowe EA, Sweeney K. Sensory specific satiety in man. *Physiol Behav*. 1981;27:137-142.
39. Rolls BJ, Rowe EA, Rolls ET. How sensory properties of foods affect human feeding behavior. *Physiol Behav*. 1982;29:409-417.
40. Johnson J, Vickers Z. Effects of flavor and macronutrient composition of food servings on liking, hunger and subsequent intake. *Appetite*. 1993;21:25-39.
41. Rolls BJ, Rowe EA, Rolls ET, Kingston B, Megson A, Gunary R. Variety in a meal enhances food intake in man. *Physiol Behav*. 1981;26:215-221.

#### SUPPORTING INFORMATION

Additional supporting information may be found in the online version of the article at the publisher's website.

**How to cite this article:** Rademacher J-G, Raddatz D, Ellrott T. Influence of food images with different macronutrient compositions on serum ghrelin levels: analysis in healthy males. *Obes Sci Pract*. 2022;8(3):328-337. <https://doi.org/10.1002/osp4.577>