



Trabajo Original

Development and validation of a smartphone application to analyze subjective appetite variables

Desarrollo y validación de una aplicación para smartphones para analizar variables subjetivas de apetito

Juan José Hernández-Morante, Joaquín Santiago Galindo-Muñoz and María del Carmen Barberá-Ortega

Research Unit of Eating Disorders. Faculty of Nursing. Universidad Católica de Murcia. Murcia, Spain

Abstract

Introduction: Nowadays dietary treatment for obesity is not effective in most cases, probably because it is designed taking into account patient's total energy expenditure while other important circumstances such as hunger/satiety daily rhythms are usually not considered. New electronic devices may help in this regard.

Objective: The objective of the present work was to develop an accurate and reliable smartphone application to analyze subjective appetite variables.

Methods: Subjective appetite sensations were evaluated in 15 healthy volunteers by two different methods: paper and pen (P-P) and a new Android and iOS application. Appetite scores were completed every 30 minutes for 4 hours in two conditions: high-carbohydrate (HC) or high-fat (HF) breakfast. Secondly, subjects completed the new application every 30 minutes in free-living conditions.

Results: Our repeated measures ANOVA revealed no significant differences regarding method (P-P or new application) or meal (HC or HF) ($p > 0.05$ in all cases) for hunger, desire to eat or fullness scores. Correlation coefficients showed a high agreement between both methods (R^2 from 0.94-0.98). In addition, Bland-Altman test also revealed a high concordance between both methods. The application was also able to measure daily variation of subjective sensations under free-living conditions.

Conclusion: The smartphone application was able to accurately determine subjective appetite scores in both laboratory and free-living conditions. The application was able to detect the effect of meal and recovery of appetite during two different conditions, following HC or HF breakfasts, which confirm the suitability of this application to future studies conducted to evaluate appetite regulation in humans.

Key words:

Appetite. Electronic health record. Smartphone. Feeding behavior.

Resumen

Introducción: actualmente el tratamiento dietético de la obesidad no suele ser efectivo en muchos casos, probablemente porque se diseña fundamentalmente teniendo en cuenta el gasto energético mientras que otros aspectos relevantes, como los ritmos de hambre/saciedad, apenas son considerados. Los nuevos dispositivos móviles (*smartphones*) podrían ayudar a este respecto.

Objetivo: desarrollar una aplicación para *smartphones* con el fin de analizar variables subjetivas relacionadas con el apetito de forma precisa y fiable.

Métodos: se analizaron las sensaciones subjetivas de apetito de 15 voluntarios sanos mediante dos métodos: papel-lápiz (P-P) y una nueva aplicación para *smartphones*. Las sensaciones de apetito se analizaron cada 30 minutos durante 4 horas en dos situaciones: desayuno alto en carbohidratos (HC) y alto en grasas (HF). Además, los sujetos completaron la aplicación cada 30 minutos en condiciones ambientales.

Resultados: los datos derivados del ANOVA indican que no existieron diferencias respecto al método (P-P vs. aplicación) o comida (HC vs. HF) ($p > 0.05$) para las puntuaciones de hambre, deseo de comer y saciedad. Los coeficientes de regresión mostraron una elevada concordancia (R^2 : 0.94-0.98). El test de Bland-Altman también reveló una alta reciprocidad entre ambos métodos. Además, la aplicación fue capaz de medir las variaciones subjetivas de apetito en condiciones ambientales.

Conclusión: la aplicación para *smartphones* fue capaz de determinar de forma fiable y precisa las variables subjetivas de apetito tanto en condiciones de laboratorio como ambientales, lo que confirma la idoneidad de esta aplicación para llevar a cabo futuros estudios en los que se evalúe la regulación del apetito en humanos.

Palabras clave:

Apetito. Registro electrónico. Smartphone. Comportamiento alimentario.

Received: 03/09/2015
Accepted: 21/01/2016

Hernández-Morante JJ, Galindo-Muñoz JS, Barberá-Ortega MC. Development and validation of a smartphone application to analyze subjective appetite variables. Nutr Hosp 2016;33:415-421

Correspondence:

Juan José Hernández Morante. Facultad de Enfermería. Universidad Católica de Murcia. Campus de Los Jerónimos, s/n. 30107 Guadalupe, Murcia. Spain
e-mail: jjhernandez@ucam.edu

INTRODUCTION

Obesity has become the main community health problem in Spain and in most developed countries (1). So far, treatment options are scarce and frequently unsuccessful. For subjects with a BMI > 40, surgical and pharmacological therapies are available but, on the other hand, overweight (BMI: 25-29.9) and moderate obese (BMI: 30-39.9) subjects must follow a behavioral modification therapy, which consist of a hypocaloric diet with an increase of physical activity energy expenditure (2). However, although great advances have been developed in the last years regarding dietary treatments, including group therapy or cognitive-behavioral therapies (3), most patients still fail to achieve the weight loss originally intended (4). This situation might be a consequence of the diet design in itself, since nowadays diet compositions are primarily designed on the base of subject's daily calorie expenditure and patient's food preferences. Furthermore, appetite sensations are not usually taken into account (2). In addition, other important issues like hunger/satiety circadian rhythm are not considered (5) probably because these variables are difficult to assess.

Nevertheless, it is important to keep in mind that although appetite (or hunger/fullness variations) is determined by a complex and reciprocal interaction of genetic, neuroendocrine, environmental and psychological factors (6), it is ultimately a subjective variable and thus it is difficult to assess. These subjective variables, such as pain or sadness, are evaluated in clinical and research settings through visual analogue scales (7), providing valuable information on sensations. However, one important limitation of this method lies in the fact that patients must be closely monitored, so there are important limitations to obtain data on free-living conditions. Fortunately, new electronic devices or smartphones may help in this regard.

Therefore, the objective of the present work was to develop and validate a new Android and iOS application for smartphones to easily assess both single and rhythmical appetite subjective sensations, helping patients and health professionals understand their appetite daily rhythms.

MATERIAL AND METHODS

SUBJECTS

Fifteen healthy volunteers (10 women and 5 men) with a mean body mass index (BMI) of 23.6 (95% IC: 20.4-26.8) were recruited by means of e-mail and advertisements. Table I summarizes the general characteristics of the population studied.

Subjects with clinically significant illness, including type 2 diabetes, kidney or hepatic diseases, and those taking any medication known to affect body weight (thyroid hormone, corticosteroids, etc.) were excluded. Subjects who were under dietary treatment prior to participation in the present study were also excluded.

Body weight was measured to the nearest 0.1 kg with the subject in underwear, and height was measured to the nearest centimeter. From this data, BMI was calculated. Body fat percent-

tage was also calculated by bioimpedance with a Tanita® BC-418 (Tanita Corporation, Tokyo, Japan) in order to exclude possible subjects with normal BMI but excess of body fat.

The survey was carried out from June to December 2014, with previous written authorization from the Catholic University of Murcia Ethics Committee. Patients were informed about the design of this study orally and in written form. An explanation of the research project with regard to ethics was also provided, specifying the aim of the results obtained, ensuring confidentiality and anonymity of the data and conforming to the Helsinki Declaration Agreement.

DESIGN

The present work comprises 2 different experiments. First, a within-subject crossover study was conducted, using time (11 points), meal (HC and HF) and method (P&P and the new application) as independent factors. The 15 subjects were evaluated in 2 experimental days, separated by 7 days. All participants performed two identical experimental procedures using both high carbohydrates (HC) and high fat (HF) meal manipulations, in alternative sequence as previously designed by a Latin square procedure (8). Secondly, these subjects followed a free-living experiment in which they were invited to assess their appetite sensations every 30 minutes during waking hours.

Sample size was estimated according to the variance observed in previous works (8,9). A minimum sample size of 12 subjects

Table I. General characteristics of the population studied

	Population studied (n = 15)	CI (95%)
<i>Clinical characteristics</i>		
Age	24 ± 2	(20-28)
Height (cm)	169 ± 2	(166-173)
Weight (kg)	68,2 ± 5,3	(56,9-79,5)
BMI	23,6 ± 1,5	(20,4-26,8)
Body fat (%)	24,8 ± 2,1	(20,3-29,2)
RMR (Kcal/day)	1,555 ± 101	(1,318-2,134)
<i>Three factor eating questionnaire</i>		
Cognitive Restriction of eating	7,0 ± 1,2	(4,2-9,8)
Hunger	9,8 ± 1,7	(6,1-13,6)
<i>Eating attitudes test</i>		
Diet	7,1 ± 1,7	(3,3-10,9)
Bulimic symptoms	0,8 ± 0,6	(- 0,6-2,1)
Oral control	3,8 ± 1,2	(1,2-6,3)
Total Score	11,6 ± 2,2	(6,7-16,4)

Mean ± sd. CI: confidence interval. BMI: body mass index. RMR: resting metabolic rate.

was estimated. Finally, our sample size yielded > 80% power that allowed us to detect true within-group differences with an effect size of partial $\eta^2 \geq 0.5$.

SUBJECTIVE APPETITE SCORES

Subjective appetite sensation was analyzed by two different methods. Classical pen and paper (P-P) method consists on a visual analogue scale (VAS) of 100 mm of length (9). The new application (Dietavisa[®]) was presented in a BQ Aquarius 5 smartphone (supplemental data 1; <http://www.nutricionhospitalaria.org/nutricion-hospitalaria-vol-33-n2-contenido-adicional/>) with three different screens for every question. Participants were invited to read the question and then move a cursor along a horizontal line. The cursor may be also displayed by a single selection. Once the participants have confirmed that the cursor was in the “real” subjective appetite sensation, they might press a “continue” button to confirm their responses. The horizontal dimension of the new application scale was of 100 pixels of length (99 mm).

Participants were presented with a series of questions accompanied by horizontal lines anchored at each end by the words “Not at all” (translated from the Spanish “*Nada en absoluto*”) and “Extremely” (translated from “*Extremadamente*”). The order and wording of the sentences was identical in the two methods: “How hungry do you feel now?” (“*¿Cuánta hambre sientes ahora?*”), “How full do you feel now?” (“*¿Cómo te sientes de saciado ahora?*”) and, finally, “How strong is your desire to eat now?” (“*¿Cómo de fuerte es tu deseo de comer ahora?*”).

Volunteers completed the two different methods alternatively attending to the Latin square and one method was removed from the sight to ensure they could not compare their responses with the previous method. Each of the three questions was presented individually to the volunteers who were not able to progress to the next question until the current VAS was correctly completed.

For the free-living study, volunteers were asked to complete the smartphone application immediately before breakfast (considered as baseline) and every 30 minutes during waking hours. The smartphone alarm was programmed to alert volunteers to respond to the smartphone application.

HIGH-CARBOHYDRATE (HC) AND HIGH-FAT (HF) BREAKFAST COMPOSITION

The subjects were interviewed at 08:00 after an overnight fast to analyze baseline appetite sensations. Immediately, participants were invited to eat a fixed breakfast meal consisted on commercially available foods. High-carbohydrate (HC) breakfast had a mean energy content of 506 Kcal (2073 KJ), with a relative contribution of energy from carbohydrates, fat and protein of 71%, 20% and 9% respectively. On the other hand, high fat (HF) breakfast was of 770 Kcal (3164 KJ), with a composition of 31% carbohydrates, 56% fat and 13% protein. Calories were estimated to approximately deliver a 20% of their daily energy expenditure (as estimated as RMR x 1.6).

Four hours after the subjects had finished their breakfast, volunteers were provided *ad libitum* with a fixed meal. Average meal composition was of 41% carbohydrates, 50% fat and 9% proteins (average weight 270 g). The meal was also composed by commercially available products, with a mean energy content of 610 Kcal (2,501 KJ). Subjects were instructed to eat as much as they felt comfortable. The precise composition of breakfasts and fixed meal is available as supplemental data 2 (<http://www.nutricionhospitalaria.org/nutricion-hospitalaria-vol-33-n2-contenido-adicional/>).

EATING ATTITUDES

To evaluate the subjects' eating habits and to exclude possible patients with eating disorders symptoms, eating attitudes test-26 (EAT-26) and three factors eating questionnaire (TFEQ) test were performed.

The Eating Attitudes Test-26 (EAT-26) is a standardized measure of symptoms and characteristics of eating disorders (10). For most of the questions, participants indicated the extent to which items were true for them (“I find myself preoccupied with food”) using a Likert-scale ranging from (0) “never” to (3) “always”. This questionnaire includes three subscales (Diet, Bulimic symptoms and Oral control). Overall scores were calculated using a standardized system of summing responses ($\alpha = 0.83$) (10).

The Three Factor Eating Disorder Questionnaire (TFEQ) is a 51-item self-report questionnaire that assesses dietary restraint, loss of control over eating, and subjective feelings of hunger. This measure comprises three subscales (Cognitive Restraint, Disinhibition and Susceptibility to Hunger). This measure demonstrates adequate internal consistency (Cronbach's alpha ranging from 0.79 to 0.92) and good convergent and discriminant validity (11).

STATISTICAL ANALYSIS

General characteristics data are presented as mean \pm sem and the 95% confidence interval. Pearson correlation coefficients were calculated between the AUC of P-P and EVA-APP methods for “hunger”, “fullness” and “desire to eat” variables. Repeated measures ANOVA with a within-subjects factor design (time x method x meal) was conducted for the subjective appetite variables studied. A *post-hoc* Bonferroni's correction test was performed for time factor. The same procedure was performed using gender, age and BMI as covariates in an ANCOVA analysis, but our data reflected no association or effect of these variables on the VAS scores, probably because the homogeneity of the subjects studied. Bland & Altman test (12) was also used to evaluate the concordance degree between P-P and EVA-APP methods. This test was calculated for every time evaluated and every appetite variable (“hunger”, “fullness” and “desire to eat”). All statistical analyses were carried out using SPSS for windows (release 21.0; SPSS Inc, Chicago, US).

RESULTS

As shown in figure 1, our data indicated that there were no significant differences regarding method (P-P or new application) or meal (high carbohydrates or high fat) ($p < 0.05$ in all cases) for hunger score. There was a similar trend for “desire to eat” scores (data not shown). Furthermore, figure 1 illustrates the profile of “fullness”, which showed the same situation, specifically, there were no differences between methods or meals. In all subjective appetite variables studied, there was a significant effect of time, with higher ratings of “hunger” and “desire to eat” and lower ratings of “fullness” as subjects moved from baseline (after breakfast).

Table II shows the correlation coefficients (r^2) among AUCs for the variables studied. Attending to data obtained, both methods showed an extremely high correlation, since the coefficients vary from 0.94 for “hunger” in HF and 0.98 for “desire to eat” in HC. Correlation coefficients were very similar between HF and HC.

To further analyze the validity of our smartphone application, Bland-Altman test was carried out (Fig. 2). In this regard, our data reflected a high agreement degree between both methods. Specifically, only baseline “fullness” score slightly rely below the confidence interval ($\pm 1.96 \times SD$).

A brief evaluation questionnaire was conducted to analyze the ease of use and preference of the new application. All participants ($n = 15$) considered more attractive the smartphone application. All subjects except one also preferred the new app. Moreover, 80% of subjects ($n = 12$) considered the smartphone application easier to use and more reliable.

On the other hand, figure 3 represents the daily rhythm of the three appetite related variables studied in 4 volunteers. There was a large drop in subjective “hunger” and an elevation of “full-

ness” immediately after a meal, which seems to indicate that the smartphone application was able to measure daily variations on subjective appetite sensations.

Finally, data derived from the different tests used confirmed that no subjects showed eating disorder symptoms, since EAT 26 score was not higher than 20 points in any subject. Finally, a positive and significant correlation was observed between AUC_{APP} for “hunger” and the dimension “susceptibility to hunger” of the TFEQ test ($r = 0.600, p = 0.039$), whereas a negative correlation was observed with the AUC_{APP} for “fullness” ($r = -0.598, p = 0.040$), which taken as a whole seems to support the validity of the obtained data through the new EVA-APP application.

DISCUSSION

The present work was carried out with the objective of developing a new tool to analyze subjective appetite variables by administering a visual analogue scale (VAS) electronically with

Table II. Correlation coefficients between P-P and EVA-APP for ‘hunger’, ‘fullness’ and ‘desire to eat’ AUC

	High carbohydrates		High fat	
	R^2	p value	R^2	p value
Hunger	0.96	< 0.001	0.94	< 0.001
Fullness	0.94	< 0.001	0.98	< 0.001
Desire to eat	0.98	< 0.001	0.95	< 0.001

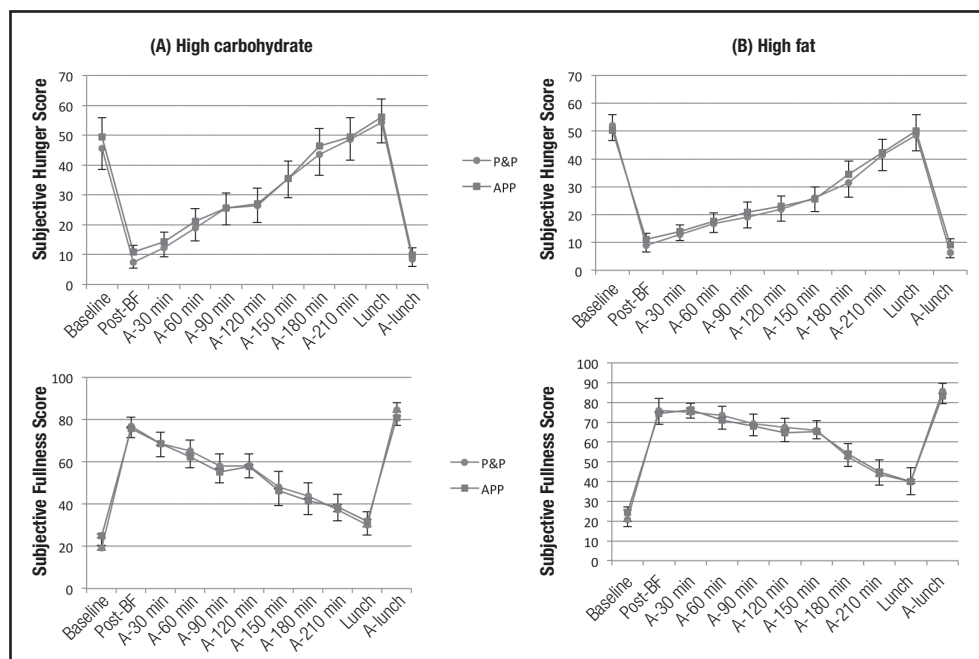


Figure 1. Comparison of subjective hunger ratings using P-P and EVA-APP on the high-carbohydrate (A) and high-fat (B) breakfast days. Data show mean \pm sem.

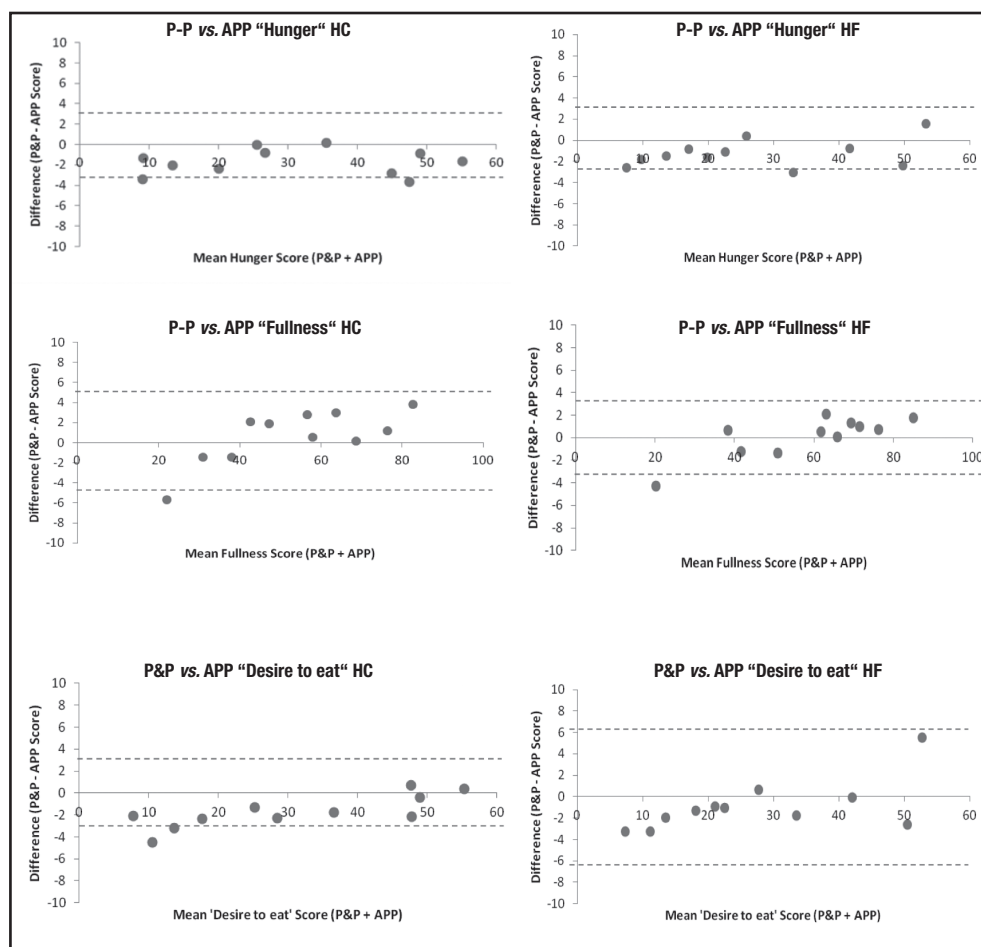


Figure 2.

The Bland-Altman analysis showing the bias, limits of agreement, and $\pm 1.96 \times \text{SD}$ values of the estimated hunger ratings on the high carbohydrate (HC) and high fat (HF) days by P-P and EVA-APP. Y-axis represents the difference between the measured and the predictive values. X-axis represents the mean of the measured and predictive values.

a friendly and easy to use graphical environment and, what is most important, an application that might be able to measure patients' appetite rates without being subjected to direct laboratory observation.

The first step in the development of any new methodology to obtain any data is to measure its reliability and validity. To this regard, our correlation data, and specially, the data derived from our Bland & Altman analysis allow us to confirm that the data obtained through our application is, at least, as reliable as data obtained through the classical paper and pen method.

Nowadays, modern lifestyle have produced drastic changes in the way we eat, which has led to an increase in the number of subjects suffering from overweight or obesity (13). For this reason, the appetite regulation system has been intensely studied by the scientific community and great advances have been performed. Special attention deserves the role of certain hormones, including hunger (ghrelin) and satiety signals (CCK, GLP-1) originated for the gastrointestinal tract, or other central effectors like the neuropeptidergic (NPY, AgRP), monoaminergic (dopamine) and endocannabinoid systems (14), among others.

At first, it might seem that measuring these hormones would be enough to analyze the appetite of these subjects. However, we

have been able to recently demonstrate a hormone-resistance state of anorexigenic signals in overweight/obese subjects (15). Therefore, our previous study has revealed that although subjects with an excess of body weight have a higher plasma level of anorexigenic hormones, they eat a higher amount of food and calories (15). This issue reflects the difficulty of measuring appetite, as it depends on numerous factors, including physiological, psychological, educational and even social factors.

The evaluation of these subjective variables such as pain or appetite was traditionally conducted through VAS and data were collected with the use of classical paper and pen (P-P) (9). However, several authors have previously pointed several drawbacks of this methodology. For instance, the integrity of data cannot be checked or verified, questionnaires may be mislabeled and tabulation of data is especially time-consuming (8,9,16). But in our opinion, the most important caveat of P-P methodology is that in long studies (involving more than 24 h) and/or in free-living conditions, subjects often forget to complete the questionnaires at certain times, which imply the lack of relevant data.

To try to avoid these caveats, several authors and especially the group of Blundell et al. have brought great advances about the development of electronic appetite rating systems (8,9) but, to

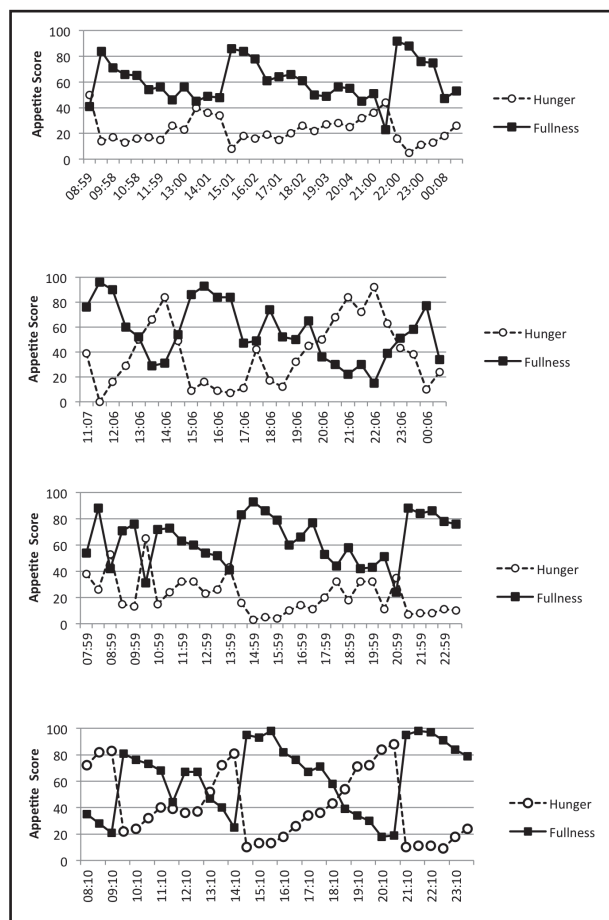


Figure 3.

Subjective “hunger”, “fullness” and “desire to eat” tracked on the smartphone application EVA-APP during waking hours in 4 volunteers in free-living conditions. Y-axis represents VAS scores and X axis represents daytime. X-axis varies among subjects.

our knowledge, the greatest inconvenience of these previous applications relies on the fact that they are not compatible with present smartphones. Thus, although these applications are quite accurate and reliable they have a disadvantage, which is related to the fact that subjects under study must stay in the laboratory or otherwise carry and external device (like a palmtop) to obtain data. While this situation is quite interesting and have brought a great amount of relevant bibliography regarding appetite regulation (17,18), it could be also interesting to evaluate the appetite rhythms in free living conditions, which may reflect, in a more realistic way, the daily variation of hunger and fullness sensations of the evaluated subjects. This is a key point, since it is tempting to hope that knowing the subjects’ daily appetite rhythms, particularly of overweight and obese patients, will allow us to design better hypocaloric diets and increase the effectiveness and adherence of hypocaloric treatment for these subjects.

For this reason, Brunger et al. have recently developed an iPad visual analogue rating system for assessing appetite and satiety

similar to the application of the present study (19). However, access to iPad devices is not as widespread as the use of smartphones so, in our opinion, the development of our application may be much more accessible to the general population.

The present work is not absent from limitations. For instance, we did not perform a test-retest analysis to assess reliability of the new application on different time-points. However, this situation may be offset taking into account that the two different experimental days showed strong similarities. Furthermore, it might seem that the sample size was small but previous studies have demonstrated that a sample size of $n = 12$ is enough to detect a within-subject difference > 10 mm, a criterion appropriate to detect variations in appetite ratings due to technical or physiological differences (20). Particular attention should be paid to potential biases due to the subjects and the design of this study. For instance, our sample was composed by healthy volunteers and only two of them were overweight. Therefore, the validity of our application to evaluate subjective appetite ratings in overweight or obese subjects is still uncertain. Moreover, all subjects were young and familiar with smartphones. In older age groups with less knowledge of new technologies prior training may be necessary to effectively assess appetite sensations in these subjects.

In conclusion, the new smartphone application developed in the present study was able to accurately determine subjective appetite scores in both laboratory and free-living conditions. In addition, the new application was able to detect the effect of meal and recovery of appetite during two different conditions, after a high carbohydrate or a high fat breakfast, which confirms the suitability of this application for further studies conducted to evaluate appetite regulation in humans. Our data also showed no differences between the gold standard P-P method and our smartphone application. Taking into account that our application is easily installable in any iOS or Android smartphone, it is logical to expect that the data collected will allow us and other researchers to easily evaluate patients’ appetite sensations.

ACKNOWLEDGEMENTS

This work was partially supported by the PMAFI 14/02 Project of the Catholic University of Murcia. We would like to thank Rafael Ruiz San Martín for his assistance on the smartphone application development. We also like to thank all volunteers that participated in the present study. Finally, we would like to thank KFTO-Vending S.L. for their support with commercial foods.

REFERENCES

1. Franco Sassi. Obesity and the economics of fat: fit not fat. Paris: OCDE Publishing; 2010.
2. Gargallo-Fernández M, Breton-Lesmes I, Basulto-Marset J, Quiles-Izquierdo J, Formiguera-Sala X, Salas-Salvadó J. Evidence-based nutritional recommendations for the prevention and treatment of overweight and obesity in adults (FESNAD-SEEDO consensus document). The role of diet in obesity treatment (III/III). *Nutr Hosp* 2012;27(3):833-64.

3. Rieger E, Treasure J, Swinbourne J, Adam B, Manns C, Caterson I. The effectiveness of including support people in a cognitive behavioural weight loss maintenance programme for obese adults: study rationale and design. *Clin Obes* 2014;4(2):77-90.
4. Calleja-Fernández A, Vidal-Casariago A, Cano-Rodríguez I, Ballesteros-Pomar MD. One-year effectiveness of two hypocaloric diets with different protein/carbohydrate ratios in weight loss and insulin resistance. *Nutr Hosp* 2012;27(6):2093-101.
5. Hernández-Morante JJ, Gómez-Santos C, Milagro F, Campión J, Martínez JA, Zamora S, et al. Expression of cortisol metabolism-related genes shows circadian rhythmic patterns in human adipose tissue. *Int J Obes (Lond)* 2009;33(4):473-80.
6. Sohn JW, Elmquist JK, Williams KW. Neuronal circuits that regulate feeding behavior and metabolism. *Trends Neurosci* 2013;36:504-12.
7. Whybrow S, Stephen JR, Stubbs RJ. The evaluation of an electronic visual analogue scale system for appetite and mood. *Eur J Clin Nutr* 2006;60(4):558-60.
8. Gibbons C, Caudwell P, Finlayson G, King N, Blundell J. Validation of a new hand-held electronic data capture method for continuous monitoring of subjective appetite sensations. *Int J Behav Nutr Phys Act* 2011;8:57.
9. Stubbs RJ, Hughes DA, Johnstone AM, Rowley E, Ferris S, Elia M, et al. Description and evaluation of a Newton-based electronic appetite rating system for temporal tracking of appetite in human subjects. *Physiol Behav* 2001;72(4):615-9.
10. Garner DM, Olmsted MP, Bohr Y, Garfinkel PE. The eating attitudes test. Psychometric features and clinical correlates. *Psychological Medicine* 1982;12:871-8.
11. Allison DB, Kalinsky LB, Gorman BS. A comparison of psychometric properties of three measures of dietary restraint. *Psychological Assessment* 1992;4:391-8.
12. Bland JM, Altman DG. Statistical methods for assessing agreements between two methods of clinical measurement. *Lancet* 1986;50:72-92.
13. Berthoud HR. The neurobiology of food intake in an obesogenic environment. *Proc Nutr Soc* 2012;71:478-87.
14. Valassi E, Scacchi M, Cavagnini F. Neuroendocrine control of food intake. *Nutr Metab Cardiovasc Dis* 2008;18:158-68.
15. Galindo-Muñoz JS, Jiménez-Rodríguez D, Hernández-Morante JJ. Diurnal rhythms of plasma GLP-1 levels in normal and overweight/obese subjects: lack of effect of weight loss. *J Physiol Biochem* 2015;71(1):17-28.
16. Stubbs RJ, Hughes DA, Johnstone AM, Rowley E, Reid C, Elia M, et al. The use of visual analogue scales to assess motivation to eat in human subjects: a review of their reliability and validity with an evaluation of new hand-held computerized systems for temporal tracking of appetite ratings. *Br J Nutr* 2000;84(4):405-15.
17. Caudwell P, Gibbons C, Hopkins M, Naslund E, King N, Finlayson G, et al. The influence of physical activity on appetite control: an experimental system to understand the relationship between exercise-induced energy expenditure and energy intake. *Proc Nutr Soc* 2011;70(2):171-80.
18. Gibbons C, Caudwell P, Finlayson G, Webb DL, Hellström PM, Näslund E, et al. Comparison of postprandial profiles of ghrelin, active GLP-1, and total PYY to meals varying in fat and carbohydrate and their association with hunger and the phases of satiety. *J Clin Endocrinol Metab* 2013;98(5):E847-55.
19. Brunger L, Smith A, Re R, Wickham M, Philippides A, Watten P, et al. Validation of an iPad visual analogue rating system for assessing appetite and satiety. *Appetite* 2015;84:259-63.
20. Flint A, Raben A, Blundell JE, Astrup A. Reproducibility, power and validity of visual analogue scales in assessment of appetite sensations in single test meal studies. *Int J Obes Relat Metab Disord* 2000;24(1):38-48.