Journal Pre-proof

ROLE OF BETA2 ADRENERGIC RECEPTOR POLYMORPHISM (RS1042714) ON BODY WEIGHT AND GLUCOSE METABOLISM RESPONSE TO A MEAL-REPLACEMENT HYPOCALORIC DIET

Daniel de Luis , Olatz Izaola , David Primo , J López Gómez

PII: S0899-9007(23)00198-3
DOI: https://doi.org/10.1016/j.nut.2023.112170
Reference: NUT 112170

To appear in: *Nutrition*

Received date: 24 February 2023
Revised date: 13 June 2023
Accepted date: 18 July 2023

Please cite this article as: Daniel de Luis , Olatz Izaola , David Primo , J López Gómez , ROLE OF BETA2 ADRENERGIC RECEPTOR POLYMORPHISM (RS1042714) ON BODY WEIGHT AND GLUCOSE METABOLISM RESPONSE TO A MEAL-REPLACEMENT HYPOCALORIC DIET, *Nutrition* (2023), doi: https://doi.org/10.1016/j.nut.2023.112170

Highlights

- A significant interaction effects was found between *rs1042714* and pMR induced changes on body weight, BMI, fat mass and waist circumference.
- A significant interaction was found between *rs1042714* and pMR induced changes (CC vs CG+GG) on glucose, fasting insulin levels and HOMA-IR.
- A significant interaction was found between *rs1042714* and pMR induced changes (CC vs CG+GG) on Total cholesterol, LDL- cholesterol and triglyceride levels.
- The odds ratio to improve alteration in glucose metabolism was (OR= 0.26, 95% CI=0.07-0.95; p=0.02) in G allele carriers.

ROLE OF BETA2 ADRENERGIC RECEPTOR POLYMORPHISM (RS1042714) ON BODY WEIGHT AND GLUCOSE METABOLISM RESPONSE TO A MEAL-REPLACEMENT HYPOCALORIC DIET

Daniel de Luis, Olatz Izaola, David Primo, J López Gómez

Endocrinology and Nutrition Research Center, School of Medicine, Department of Endocrinology and Nutrition, Hospital Clinico Universitario, University of Valladolid, Valladolid, Spain.

Corresponding author. MDPh Daniel de Luis Endocrinology and Nutrition Research Center. School of Medicine, Valladolid University, C/Los perales 16 Simancas, 47130 Valladolid Spain. Tel.: 34 983420400; fax: 34983331566.

E-mail address: dadluis@yahoo.es (D.A. de Luis).

Running title: rs1042714 polymorphism and meal replacement Hypocaloric diet

Word count 3564

ABSTRACT

Background and aims: The Beta2-adrenergic receptor (ADRB2) is involved in energy balance regulation. The objective of our study was to evaluate the role of rs1042714 genetic variant of *ADRB2* gene on weight loss, body composition and metabolic changes secondary to partial Meal replacement (pMR) hypocaloric diet in women with obesity.

Methods*:* We realized an interventional study in 95 premenopausal women with body mass index (BMI) $\geq$ 35 kg/m^2. The subjects received two intakes per day of a normocaloric hyperproteic formula during 12 weeks in a pMR diet. Body weight, BMI, fat mass, waist circumference, lipid profile, fasting insulin levels and HOMA-IR were determined. All patients were genotyped rs1042714 and evaluated in a dominant model (CC vs. CG+GG).

Results: Genotype frequencies were 31 (37.3%), 38 (45.8%) and 14 (16.9%) for the CC, CG and GG genotypes, respectively. We found significant interaction effects between *ADRB2* variant a pMR induced changes (CC vs CG+GG) on body weight (-7.1$\pm$0.3 kg vs. -13.5$\pm$0.5 kg: p=0.03), BMI (-0.9$\pm$0.1 kg/m^2 vs. -1.2$\pm$0.2 kg/m^2: p=0.03), fat mass (-4.9$\pm$0.5 kg vs. -10.2$\pm$1.2 kg: p=0.01), waist circumference (-5.1$\pm$0.2 cm vs. -10.1$\pm$1.9 cm: p=0.03), glucose (-5.1$\pm$1.3 mg/dl vs. -12.5$\pm$2.5 mg/dl: p=0.03), Total cholesterol (-18.1$\pm$9.3 mg/dl vs. -33.5$\pm$4.5 mg/dl: p=0.03), LDL- cholesterol (-9.1$\pm$5.3 mg/dl vs. -24.5$\pm$4.1 mg/dl: p=0.04), triglyceride levels (-6.1$\pm$5.3 mg/dl vs. -31.5$\pm$9.5 mg/dl: p=0.04), fasting insulin levels (-1.8$\pm$0.3 UI/L vs. -6.3$\pm$0.5 UI/L: p=0.03) and HOMA-IR (-0.6$\pm$0.3 units vs. -1.9$\pm$0.5 units: p=0.03). The odds ratio to improve alteration in glucose metabolism adjusted by age and weight loss throughout the study was (OR= 0.26, 95% CI=0.07-0.95; p=0.02) in G allele carriers.

Conclusions*:*

G allele of rs *rs1042714* predicts the magnitude of weight loss resulting from a pMR diet. These adiposity improvements produce a better improvement of insulin resistance and percentage of impaired glucose metabolism in G allele carriers.

Key words: rs1042714, partial meal replacement, *ADBR2* gene, body weight.

INTRODUCTION

A successful weight-loss intervention is an essential treatment of obesity and related comorbidities [1]. In Western countries, obesity is the main causes of mortality and morbidity including cardiovascular events, diabetes mellitus type 2 and malignant tumours [2]. The most important goal of dietary treatment is to achieve a weight loss of at least 5-10% in a short-term period [1]. One possibility for the treatment of subjects with obesity is the diets of partial meal replacements (pMRs). In the meta-analysis of Heymsfield et al. [3], they demonstrated that pMRs diets produced superior weight loss than conventional diets, 7% vs. 3% in a short-term period comparing with traditional energy restricted food-based diets.

Otherwise, genetic factors play a key role in the risk to develop obesity and the response of weight management treatment such as hypocaloric diets [4]. In this context, the Beta2-adrenergic receptor (ADRB2) is involved in energy balance regulation through the stimulation of both lipid mobilization in adipose tissue and thermogenesis [5] and finally, in catecholamine-induced lipolysis in muscles [6]. Some single nucleotide polymorphisms (SNPs) have been described within the coding region of *ADRB2* gene. The most frequent SNP occur at codon 16 (GLn27GLu; rs1042714). Some investigations have evaluated the association of the Gln27Glu variant with obesity and related traits [7-8]. Some studies failed to identify any association and others found significant relationships, yet there is insuficient evidence regarding the effect of this genetic variant with the response to hypocaloric diets and its related metabolic changes. For example, Ruiz et al [9] reported that rs1042714 modulated effect on diet-induced changes on body weight in women with obesity. Moreover, Ramos-Lopez et al [10] demonstrated a similar weight loss after two different hypocaloric diets in allelic risk carriers and non-carriers of this SNP, with different

secondaries lipid response. Recently, Coletta et al [11] reported no association of this SNP with weight response after an exercise and weight loss program in women with obesity. Finally, Dos Santos et al [12] demonstrated a lack of association between excessive gestational weight gain and rs1042714 in pregnant women with pregestational diabetes mellitus. The effect of this relevant genetic variation secondary to the pMR diet has not been investigated before.

Considering the existing controversy, the objective of our study was to evaluate the role of rs1042714 genetic variant of *ADRB2* gene on weight loss, body composition and metabolic changes secondary to pMR hypocaloric diet in women with obesity.

METHODS AND PROCEDURES

Participants and design

A total of 105 women with obesity from North West of Spain, aged 30-55 years volunteered to participate in the study from January 2019 to December 2020. Finally, 95 patients were included in the study (figure 1) and 10 were excluded. Participants were premenopausic and showed a body weight stability over the last 6 months with obesity, body mass index (BMI > 35 kg/m^2). We prescribed to these women a pMR hypocaloric diet and it was supplemented with a normocaloric hyperproteic formula. Exclusion criteria included history of cardiovascular events (heart attack or stroke), severe renal or hepatic dysfunction, active alcoholism, malignant tumor and pregnancy. Women with medication for hyperlipidemia, hyperuricemia, hypertension and diabetes mellitus were not included, too. The exclusion criteria were questioned during follow-up. The exclusion criteria of the 10 patients were (severe renal dysfunction n=5, severe hepatic dysfunction n=4 and malignant tumor n=1.

Diet Intervention

This intervention was designed as a 3-month controlled body weight loss study. Body weight reduction was produced by a meal-replacement hypocaloric diet (pMR). This pMR was distributed in 6 meals, as follows; breakfast, morning snack, lunch, afternoon snack, dinner, after dinner snack. The meals (lunch and dinner) were substituted by a normocaloric hyperproteic formula ~~(VEGESTART Complete®),~~ whose composition are reported in table 1. A dietitian gave reinforcement by phone call twice per week and all patients reported their dietary intakes of 72 hours in order to estimate the daily intakes of calories and macronutrients, before and after 3 months of dietary

intervention. The dietary registrations (2 weekdays and one weekend day) were evaluated with a software (Dietsource ®, Nestlé, Geneve, Switzerland). During the dietary intervention, the only physical activity allowed was the following; aerobic physical activities at least 3 times per week (30 minutes each) and the allowed exercises were running, walking and cycling. Physical activity was reported through a self-registration.

Outcome parameters

Body weight, height, body mass index (BMI), waist circumference and fat mass by electrical impedance was measured before and after dietary intervention. Blood pressure was determined at same times, too. The height was estimated with the patient in an upright, using a stadiometer (Omrom, LA, Ca, USA). The corporal weight was measured without clothing with an accuracy of 10 gr. (Omrom, LA, Ca, USA). The BMI was calculated using the above-mentioned parameters with the following equation: Weight (kg)/Height x Height (m^2). The difference in relative weight was determined by the percentage of weight loss (%PP) with the next formula: (Weight before intervention - Weight after intervention (kg) / Initial weight(kg)) x 100. A loss of more than 7.5% of the initial weight was considered a success, taking into account our usual clinical practice. Waist circumference was determined with the patient standing in the narrowest diameter between xiphoid process and iliac crest using an extendable tape measure (Omrom, LA, Ca, USA). A bioelectrical impedance analysis (BIA) was realized, too. The parameters analysed with the BIA was total fat mass (kg) and ft free mass (kg) [12].

In each subjects and in both times, arterial blood pressure was determined three times after a 10 minutes rest with a random zero mercury sphygmomanometer, and averaged (Omrom, LA,CA, USA). In order to diagnose the presence of diabetes mellitus or impaired fasting blood glucose, the criteria were glucose $\geq$ 126 mg/dl or $\geq$ 110 mg/dl, respectively [13].

In both times fasting blood samples were collected into tubes containing EDTA, for analysis of basal fasting (8 hs) glucose, fasting insulin, insulin resistance calculated (HOMA-IR) and lipid profile. Biochemical measurements, including glucose, insulin, total cholesterol, HDL-cholesterol, and triglyceride levels were measured using the COBAS INTEGRA 400 analyser (Roche Diagnostic, Basel, Switzerland). LDL cholesterol was determined using Friedewald formula (LDL cholesterol= total

cholesterol-HDL cholesterol-triglycerides/5) [14]. Based on these parameters, homeostasis model assessment for insulin resistance (HOMA-IR) was calculated using the equation (glucose x insulin/22.5) [15].

Genotyping

Genomic DNA was isolated from oral mucosa cells using a commercial kit as indicated by manufactured (Applied Biosystems, Foster City, CA, LA) from. Genotyping (rs1042713) was performed by using commercial assays with the TaqMan® OpenArray™ Genotyping platform (Thermofisher, Pittsburg, Pens). Samples of DNA were loaded using the AccuFill system, and amplification realized on the QuantStudio 12K Flex Real-Time qPCR instrument (Thermofisher, Pittsburg, Pens). A volume of 10 µl with 2.5 µl TaqMan Open Array Master Mix (Applied Biosystems, Foster City, CA, LA) and 2.5 µl human DNA sample were used and amplified on arrays following the manufacturer's instructions, too. Sample clustering and genotype calling for Open Array assays was performed in TaqMan Genotyper (LifeTechnologies, Carlsbad, CA). Genotyping success rate was 100% and no discordant genotypes were observed in duplicate samples.

Statistical analysis

We analyzed differences in changes (after 3 months energy-restricted diet) on adiposity parameters and biochemical variables between *ADBRB2* rs1042714 genotypes using a univariate analysis of covariance (ANCOVA) with Bonferroni test post Hoc. We tested the dominant model (CC vs CG+GG). The genotype distribution was studied for deviation from Hardy-Weinberg equilibrium by a Chi-square. Sample size was calculated to detect differences over 5 kg during dietary treatment with 90% power and 5% significance (n=90), with an effect size (d=2.1). The results were reported as average+/- standard deviation. In within-groups, we used paired t-Student test for biochemical parameters at baseline and after 3 months of pMR. In between-groups, independent t-test was utilized to compare the differences. The Mann-Whitney U test and the Wilcoxon signed rank test were utilized in non-parametric variables. Categorical variables were evaluated with Chi-Square test, with Yates correction as necessary. Logistic regression analyses were used to calculated odds ratio (OR) and 95% confidence interval (CI) to estimate the association of the rs1042714 variant with weight loss as dichotomic (7.5% weight loss) and presence of alteration of glycemic

metabolism (diabetes mellitus or impaired fasting glucose). Both models were adjusted for a well-known variable that influences biological models: age. The first model was adjusted for basal weight and the second for weight loss, which influences metabolic improvement. Number necessary to treat (NNT) was calculated. NNT was obtained from the following formula NNT= 100/ARR = NNT, in which ARR is the absolute risk reduction. A *p*-value <0.05 was considered significant. Statistical analysis was realized with SPSS version 23.0 (Chicago, IL. USA).

RESULTS

We recruited 95 premenopausal women with obesity. Genotype frequencies were 36 (37.3%), 44 (45.8%) and 15 (16.9%) for the CC, CG and GG genotypes, respectively. The variant of *ADBRB2* gene was in Hardy Weinberg equilibrium (p=0.37). All obese subjects completed the 12 weeks follow-up period without dropouts and no adverse effects secondary to the dietary intervention were observed.

The average age of the all patients was 45.8±3.9 years (range: 31-54 years), the average age was similar in both genotype groups (wild type (CC) *vs*. mutant type (CG+GG)) (46.1±3.9 years *vs*. 43.9±4.1 years: ns) as a dominant model.

In this ~~controlled~~ dietary interventional study, subjects with both genotypes (CC vs. CG+GG) showed a significant decrease in daily intakes of energy, carbohydrate, fat and protein (table 2). However, the percentage of calories provided by protein after the dietary intervention was higher than at baseline. The final distribution of type of fats in both genotype groups was similar (CC vs. CG+GG); 32.0% vs. 33.2% of saturated fats, a 50.5% vs. 49.7% of monounsaturated fats and a 17.5% vs. 18.2% of polyunsaturated fats. Dietary fibre remained unchanged throughout the study in both groups. Physical exercise time was similar in the two groups at baseline and after the intervention (table 2). In subjects with CC genotype, 95% of all the prescribed ~~VEGESTAR ®~~ bricks were taken and in patients with the CG+GG genotype 97%.

As shown in table 3, we found significant interaction effects between *ADRB2* variant and pMR induced changes (CC vs CG+GG) on body weight (-7.1±0.3 kg vs. -13.5±0.5 kg: p=0.03), BMI (-0.9±0.1 kg/m^2 vs. -1.2±0.2 kg/m^2: p=0.03), fat mass (-4.9±0.5 kg vs. -10.2±1.2 kg: p=0.01) and waist circumference (-5.1±0.2 cm vs. -10.1±1.9 cm: p=0.03). Therefore, all these anthropometric parameters were lower after

3 months of nutritional intervention in carriers of the G allele than in non-G allele carriers. The percentage of patients who achieved 7.5% weight loss was higher in the G carriers (51.2% vs 27.1%). The odds ratio to achieved 7.5% of weight loss adjusted by age and initial weight was (OR= 1.91, 95% CI=1.07-3.44; p=0.03). Systolic and diastolic blood pressure levels were similar in both genotypes at baseline. The improvement in blood pressures were similar in both groups, too.

Table 4 showed all biochemical parameters. Glucose (-5.1±1.3 mg/dl vs. -12.5±2.5 mg/dl: p=0.03), Total cholesterol (-18.1±9.3 mg/dl vs. -33.5±4.5 mg/dl: p=0.03), LDL-cholesterol (-9.1±5.3 mg/dl vs. -24.5±4.1 mg/dl: p=0.04), triglyceride levels (-6.1±5.3 mg/dl vs. -31.5±9.5 mg/dl: p=0.04), fasting insulin levels (-1.8±0.3 UI/L vs. -6.3±0.5 UI/L: p=0.03) and HOMA-IR (-0.6±0.3 units vs. -1.9±0.5 units: p=0.03) improved in G allele carriers. Therefore, all these biochemical parameters were lower after 3 months of nutritional intervention in carriers of the G allele than in non-G allele carriers.

In both genotypes, presence of diabetes mellitus or impaired fasting glucose didn´t show statistical differences ((CC vs CG+GG): 24.3% vs 22.4%;p=0.61). Moreover, the decrease after dietary intervention in the percentage of diabetes mellitus or impaired fasting glucose percentage was statistically significant in G allele carriers ((CC+CG+GG):21.6% vs 6.9%;p=0.02). The odds ratio to improve alteration in glucose metabolism adjusted by age and weight loss was (OR= 0.26, 95% CI=0.07-0.95; p=0.02). It would be necessary to treat (number needed to treat with pMR diet), a total of 6.74 patients with the G allele for the disappearance a case of diabetes mellitus or impaired blood glucose in the fasting state NNT 6.74 (95% CI: 3.38-91.2; p=0.03).

DISCUSSION

This study shows a relationship between rs1042714 polymorphism in the *ADRB2* gene and pMR diet on body weight and metabolic response in premenopausal women with obesity, so that women carrying G allele had a larger reduction in body weight, fat mass, insulin resistance and LDL-cholesterol after a 3-month of energy restriction compared with non-G allele carriers.

We believe that these results provide the potential modulating effect of this genetic variant to this type of weight reduction therapy. There are few studies that have evaluated the relationship of this SNP with the response to hypocaloric diets.

Preliminary studies [16] showed that overfeeding induced a greater gain in weight and fat mass in non-G allele carriers than G allele carriers. These findings are in agreement with previous interventional studies [9]. Ruiz et al [9] reported that women with obesity carrying G allele had a greater reduction in body weight than non-G allele carriers. Moreover, lean mass decreased more in carriers of the G allele than in non-carriers. On the other hand, this work did not evaluate the metabolic modifications after weight loss. There are important differences between that study and ours that may explain the differences in the results. First, although the population was middle-aged, our sample was overweight with a mean BMI of 39 kg/m^2 versus 33 kg/m^2 in the previous study [9]. Second, the dietary intervention had a lower protein content (15% total caloric value) compared to 23% in our design. This increase in protein intake is due to the use of a hyperproteic non-caloric formula in our rMP diet that allows a greater protein intake despite dietary restriction, and presumably a better preservation of lean mass. The hypothesis to explain this greater weight loss may be related to a lower decrease in the resting metabolic rate in patients who carry the G allele, as it has been indicated in previous studies [9,16]. As above-mentioned, there is a lack of studies evaluating the effect of this SNP with dietary intervention designs. There is another study in the literature [17] that demonstrated the relationship between the presence of the G allele in obese women and a higher intake of carbohydrates and a higher risk of obesity. In this study, a higher intake of carbohydrates may increase the obesity risk in women carrying G allele, which may be associated with changes in the carbohydrates/fat proportions oxidized as a consequence of a different Beta2 receptor function. In our study we did not detect this relationship, however the population of the study by Martinez et al [17] is different from ours, both in age, presence of males in the sample, a design of case-control study and a lower mean BMI. Ethnicity may also have effects, for example in Zhang's meta-analysis [18] demonstrated that rs1042714 might be a significant risk factor for obesity in Asians American Indians, and Pacific Islanders, but not in Europeans, as our data shows. Other factors may also explain these differences found in the literature is the socioeconomic status. For example, Saliba et al [19] in a 7-week dietary intervention study with a standard hypocaloric diet did not detect differences between both genotypes, however, patients with low socioeconomic status showed lower BMI than carriers of the G allele, suggesting a protective effect of the polymorphism.

Other interventional study was developed by Ramos-Lopez et al [10]. Moreover, Ramos-Lopez et al [10] reported a similar weight loss after two different hypocaloric diets (low fat diet) and (moderately high-protein diet) in allelic risk carriers and non-carriers of this SNP. However, the genetic variant is related to a decrease in LDL-cholesterol and total cholesterol with a ~~the~~ moderately high-protein diet branch. This diet has a proportion of calories from macronutrients very similar to our intervention (40% carbohydrates, 30% proteins and 30% fats). The ADRB2 receptor is related in the regulation of lipolysis in adipose tissue and energy expenditure [20]. Moreover, molecular interactions between ADRB2 and cholesterol dynamics have been elucidated by nano techniques [21]. Finally, Coletta et al [11] reported no association of this SNP with weight response after an exercise and weight loss program of 24 weeks in women with obesity. This is the study with the longest duration, however the caloric restriction achieved was much lower than in our present study and the protein intake was much lower.

Finally, the effect found in our study on the improvement in the prevalence of diabetes mellitus and/or changes in fasting blood glucose during treatment does not have a clear explanation. It may be that this risk allele is processed in imbalance with other unknown alleles that mark the response of resistance to insulin and the development of diabetes. Recently, K dos Santos [22] has shown an effect on early weight gain in women with gestational diabetes and the presence of a variant of the *ADRB2* gene (rs1042713). Perhaps, these SNPs are associated with the pathogenesis of insulin resistance as it inhibits the insulin-induced translocation of GLUT4 and reduces glucose uptake via the cAMP-dependent protein kinase A-dependent pathways [23].

pMR diet is the first time that it has been evaluated with this working hypothesis and this is a strength of the study, of this work. Well-controlled nutritional intervention is also a strength of our design. Our study has some limitations. First, the inclusion in the trial of our premenopausal women with obesity and with a low cardiovascular risk that does not allow the generalization of the results beyond a population of obese without comorbidities. Second, we only analysed one SNP of *ADRB2* gene, so other variants could be associated with our findings. Third, many other uncontrolled factors could influence our results (epigenetic, hormonal status and timing of food, for example). Fourth, the absence of determination of resting metabolic ratio might be a

bias. ~~Fifth, we did not measure RMR~~. Finally, the self-reported dietary intake might include bias of under- or over-reporting energy.

In summary, the results of the present study suggest that the ADRB2 (rs1042714) polymorphism has a modulating effect on pMR diet -induced changes on body weight, lipid metabolism and insulin resistance. The pMR diet provides anthropometric benefits in premenopausal obese women without comorbidities, with or without the G allele in the relevant genetic structure. That is, the presence of the G allele increases the metabolic benefit of the pMR diet. These modifications produce a significant decrease in the prevalence of impaired glucose metabolism. A personalized nutrition and a genetic approach to obesity intervention is expected in the near future. In this context, a premenopausal woman with obesity and the presence of the G allele would need a more aggressive therapeutic strategy to achieve metabolic improvements associated with weight improvement.

Statement of Ethics

This study protocol was reviewed and approved by [HCVUA Commitee], approval number (HVUVA committee 2/2018) Written Informed consent was obtained from all individual participants included in the study.

Conflict of Interest Statement

The authors have no conflicts of interest to declare

Funding Sources

The authors have no funding sources to declare

Author Contributions

Daniel Antonio de Luis and Juan Jose Lopez designed the study an wrote the article

Olatz Izaola, and Jose Lopez realized nutritional evaluation

David Primo and Daniel de Luis realized biochemical evaluation

Data Availability Statement

All data generated or analyzed during this study are included in this article. Further enquiries can be directed to the corresponding author.

REFERENCES_

1. Gutiérrez-Fisac JL, Guallar-Castillón P, León-Muñoz LM, Graciani A, Banegas JR, Rodríguez-Artalejo F. Prevalence of general and abdominal obesity in the adult population of Spain, 2008-2010: the ENRICA study. Obes Rev 2012;13 :388-392
2. Finucane MM, Stevens GA, Cowan MJ, Danaei G, Lin JK, Paciorek CJ, Singh GM, Gutierrez HR, Lu Y, Bahalim AN, Farzadfar F, Riley LM, Ezzati M. National, regional, and global trends in body-mass index since 1980: systematic analysis of health examination surveys and epidemiological studies with 960 country-years and 9,1 million participants. Lancet 2011; 377: 557-567.
3. Heymsfield SB, van Mierlo CA, van der Knaap HC, Heo M, Frier HI. Weight management using a meal replacement strategy: meta and pooling analysis from six studies. Int J Obes Relat Metab Disord. 2003;27:537-49.
4. Izaola O, Primo D, de Luis D. Dietary Intervention during 9 Months with a Hypocaloric Diet, Interaction of the Genetic Variant of Adiponectin Gene rs822393 with Metabolic Parameters. Dis Markers. 2022 Jan 25;2022:7058389. doi: 10.1155/2022/7058389.
5. Lafontan M, Barbe P, Galitzky J *et al*. Adrenergic regulation of adipocyte metabolism. Hum Reprod 1997;12 Suppl 1:6–20.
6. Jocken JW, Blaak EE. Catecholamine-induced lipolysis in adipose tissue and skeletal muscle in obesity. Physiol Behav 2008;94:219–23
7. Ordovas JM. Genetic interactions with diet influence the risk of cardiovascular disease. Am J Clin Nutr 2006;83:443S–446S.
8. Bantle JP, Wylie-Rosett J, Albright AL *et al*.; American Diabetes Association. Nutrition recommendations and interventions for diabetes: a position statement of the American Diabetes Association. Diabetes Care 2008;31Suppl 1:S61–S78
9. Ruiz JR, Larrarte E, Margareto J, Ares R, Labayen I. Role of β_2-adrenergic receptor polymorphisms on body weight and body composition response to

energy restriction in obese women: preliminary results. Obesity (Silver Spring). 2011;19:212-5.

10. Ramos-Lopez O, Riezu-Boj JI, Milagro FI, Goni L, Cuervo M, Martinez JA. Differential lipid metabolism outcomes associated with ADRB2 gene polymorphisms in response to two dietary interventions in overweight/obese subjects. Nutr Metab Cardiovasc Dis. 2018;28:165-172.
11. Coletta AM, Sanchez B, O'Connor A, Dalton R, Springer S, Koozehchian MS, Murano PS, Woodman CR, Rasmussen C, Kreider RB. Alignment of diet prescription to genotype does not promote greater weight loss success in women with obesity participating in an exercise and weight loss program. Obes Sci Pract. 2018;4:554-574.
12. Lukaski H, Johson PE. Assessment of fat-free mass using bioelectrical impedance measurements of the human body. Am J Clin Nutr 1985;41:810-7.
13. American Diabetes Association. 2. Classification and Diagnosis of Diabetes: Standards of Medical Care in Diabetes-2021. Diabetes Care. 2021 Jan;44(Suppl 1):S15-S33.
14. Friedewald WT, Levy RJ, Fredrickson DS. Estimation of the concentration of low-density lipoprotein cholesterol in plasma without use of the preparative ultracentrifuge. Clin Chem 1972; 18: 499-502
15. Mathews DR, Hosker JP, Rudenski AS, Naylor BA, Treacher DF: Homeostasis model assessment: insulin resistance and beta cell function from fasting plasma glucose and insulin concentrations in man. Diabetologia 1985;28:412-414
16. Ukkola O, Tremblay A, Bouchard C. Beta-2 adrenergic receptor variants are associated with subcutaneous fat accumulation in response to long-term overfeeding. Int J Obes Relat Metab Disord 2001;25:1604–1608
17. Martínez JA, Corbalán MS, Sánchez-Villegas A, Forga L, Marti A, Martínez-González MA. Obesity risk is associated with carbohydrate intake in women carrying the Gln27Glu beta2-adrenoceptor polymorphism. J Nutr. 2003;133:2549-54.
18. Zhang H, Wu J, Yu L. Association of Gln27Glu and Arg16Gly polymorphisms in Beta2-adrenergic receptor gene with obesity susceptibility: a meta-analysis. PLoS One. 2014;9(6):e100489.
19. Saliba LF, Reis RS, Brownson RC, Hino AA, Tureck LV, Valko C, de Souza

RL, Furtado-Alle L. Obesity-related gene ADRB2, ADRB3 and GHRL polymorphisms and the response to a weight loss diet intervention in adult women. Genet Mol Biol. 2014;37:15-22.

20. Rosado EL, Bressan J, Martínez JA. Environmental factors and beta2-adrenergic receptor polymorphism: influence on the energy expenditure and nutritional status of obese women. Lipids 2015;50:459e67

21. Zocher M, Zhang C, Rasmussen SG, Kobilka BK, Müller DJ. Cholesterol increases kinetic, energetic, and mechanical stability of the human b2-adrenergic receptor. Proc Natl Acad Sci U S A 2012;109(50):E3463e72.

22. Santos KD, Rosado EL, da Fonseca ACP, Belfort GP, da Silva LBG, Ribeiro-Alves M, Zembrzuski VM, Martínez JA, Saunders C. FTO and ADRB2 Genetic Polymorphisms Are Risk Factors for Earlier Excessive Gestational Weight Gain in Pregnant Women with Pregestational Diabetes Mellitus: Results of a Randomized Nutrigenetic Trial. Nutrients. 2022;14(5):1050.

23. Mangmool S, Denkaew T, Phosri S, Pinthong D, Parichatikanond W, Shimauchi T, Nishida M. Sustained βAR Stimulation Mediates Cardiac Insulin Resistance in a PKA-Dependent Manner. Mol Endocrinol. 2016;30:118-32.

Figure1 Flow chart of patients

Legend: Flow chart with analyzed patients

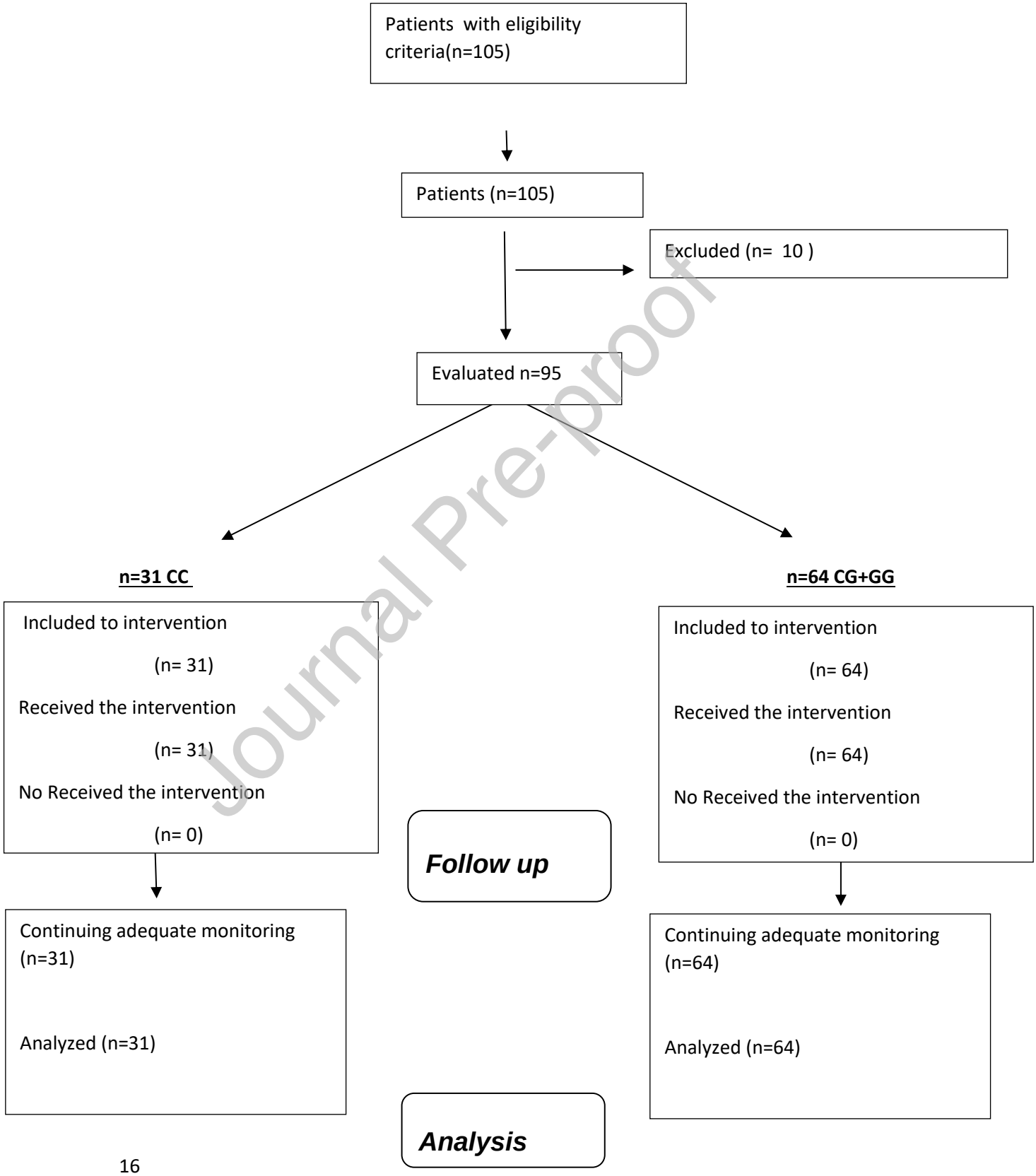


Table 1: Energy and macronutrients in the partial Meal replacement diet (four intakes as natural food and two intakes as normocaloric hyperproteic formula)

DATA	ORAL DIET + FORMULA	NORMOCALORIC HYPERPROTEIC FORMULA (200 ml per brick)
Calories(kcal)	1035	200
Proteins (g (%TCV))	64.4 (25%)	15.4(31%)
Fats (g(%TCV))	19.1 (17%)	5.2(23%)
Carbohydrates (g(%TCV))	151.6 (59%)	21(42%)
Dietary Fiber (g)	15.9	4.2

Normocaloric hyperproteic formula has ~~VEGESTART~~® (%TCV: % Total Caloric Value)

Table 2. ADRB2 rs1042714 polymorphism and Energy/macronutrients intakes and physical activity changes after 3 months of intervention (mean ± SD).

Daily intakes	**n=95**						
	CC (n=31)			**CG+GG (n=64)**			
	Basal	**3 months**	**P Time**	**Basal**	**3 months**	**P time**	**P Basal Genotype** **P Post treatment Genotype** **P Changes between Genotypes**
Calorie intake (kcal/day)	1721.9±71.8	1018.8±21.1*	P=0.01	1791.1±48.9	1099.1±38.1*	P=0.01	P=0.39 P=0.41 P=0.52
Carbohydrate intake (g/day) (PTC%)	171.8±51.2 (41.4%)	131.8±21.1$ (63.4%)	P=0.01	169.9±23.9 (41.0%)	130.0±21.1$ (63.0%)	P=0.02	P=0.40 P=0.57 P=0.41
Fat intake (g/day) (PTC%)	58.8±20.3 (39.1%)	27.1±10.1# (22.6%)	P=0.01	59.1±18.1 (39.4%)	27.1±8.1# (22.8%)	P=0.01	P=0.48 P=0.35 P=0.40
Protein intake (g/day) (PTC%)	74.1±14.1 (19.5%)	55.1±10.3& (23.0%)	P=0.02	74.9±13.0 (20.6%)	58.1±12.2& (23.2%)	P=0.03	P=0.43 P=0.52 P=0.29
Fiber intake (g/day)	17.2±6.1	17.9±4.2	P=0.29	16.8±6.2	17.7±3.2	P=0.41	P=0.24 P=0.51 P=0.19
Physical activity (min/week)	125.2±17.1	129.3±17.9	P=0.22	126.8±12.9	130.1±11.2	P=0.41	P=0.28 P=0.41 P=0.39

PTC: Percentage of total calorie; Statistical differences $P<0.05$, in each genotype group (* Daily Calorie intake, $ Daily Carbohydrate intake, # Daily fat intake, & Daily protein intake).

Table 3. ADRB2 rs1042714 polymorphism and adiposity parameters and arterial pressure changes after 3 months of intervention (mean ± SD).

Parameteres	n=95						
	CC (n=31)			CG+GG (n=64)			
	Basal	3 months	P time	Basal	3 months	P time	P Basal Genotype P Post treatment Genotype P Changes between Genotypes
BMI	39.5±2.1	38.6±2.0*	P=0.02	39.4±1.9	38.2±2.1*	P=0.02	P=0.22 P=0.03 P=0.03
Weight (kg)	103.4±2.1	96.4±3.1$	P=0.02	102.1±3.1	89.6±2.0$	P=0.01	P=0.32 P=0.03 P=0.03
Fat mass (kg)	50.4±4.1	45.5±6.1#	P=0.03	49.9±3.0	39.4±2.1#	P=0.01	P=0.23 P=0.02 P=0.01
Fat free mass (kg)	56.6±3.1	54.8±2.1	P=0.13	55.9±3.0	54.1±2.1	P=0.21	P=0.23 P=0.42 P=0.41
WC (cm)	120.1±4.0	115.0±3.9&	P=0.01	119.1±3.9	109.0±2.8&	P=0.002	P=0.31 P=0.02 P=0.03
SBP (mmHg)	131.3±4.1	124.1±3.1* *	P=0.02	132.2±3.3	124.3±4.0* *	P=0.02	P=0.34 P=0.29 P=0.23
DBP (mmHg)	83.8±3.0	77.1±2.1***	P=0.03	81.7±4.1	76.9±3.0 ***	P=0.03	P=0.25 P=0.37 P=0.26

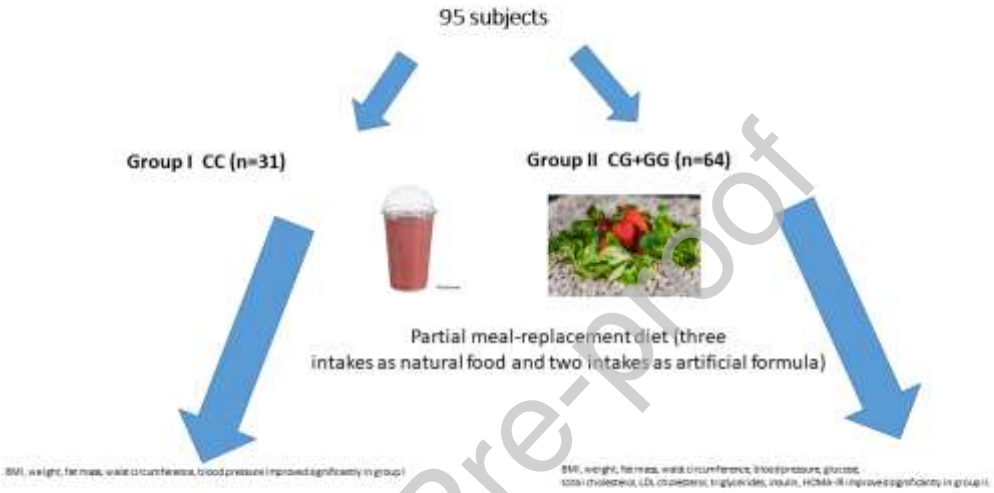
BMI: body mass index DBP, diastolic blood pressure; SBP, systolic blood pressure; WC, waist circumference; Statistical differences $P<0.05$, in each genotype group (* BMI, $ Weight, #fat mass, & WC, **SBP, ***DBP)

Table 4. ADRB2 rs1042714 polymorphism and biochemical parameters changes after 3 months of intervention (mean ± SD).

Biochemical parameters	n=95						**P Basal Genotype** **P Post treatment Genotype** **P Changes between Genotypes**
	CC (n=31)			**CG+GG (n=64)**			
	Basal	3 months	P time	Basal	3 months	P time	
Glucose (mg/dl)	103.3±4.2	98.6±2.1	P=0.27	103.9±3.1	91.8±3.0	P=0.03	P=0.12 P=0.01 P=0.01
Total cholesterol (mg/dl)	210.1±8.7	192.1±8.1	P=0.21	213.1±4.0	180.8±5.2 *	P=0.01	P=0.44 P=0.02 P=0.03
LDL-cholesterol (mg/dl)	136.1±4.3	127.1±4.0	P=0.02	138.9±4.1	114.2±3.9$	P=0.03	P=0.61 P=0.03 P=0.04
HDL-cholesterol (mg/dl)	56.3±3.1	55.9±3.0	P=0.41	56.2±4.0	54.9±3.1	P=0.51	P=0.31 P=0.60 P=0.34
Triglycerides (mg/dl)	115.1±10.9	109.2±8.9	P=0.21	113.1±10.2	89.1±8.1#	P=0.03	P=0.11 P=0.03 P=0.04
Insulin (mUI/l)	15.5±1.9	13.7±1.1	P=0.23	16.1±1.2	9.8±1.2&	P=0.01	P=0.42 P=0.02 P=0.03
HOMA-IR	4.5±0.5	3.9±0.9	P=0.13	4.7±0.4	2.8±0.4**	P=0.01	P=0.51 P=0.03 P=0.02

HOMA-IR (Homeostasis model assessment).. Statistical differences $P<0.05$, in each genotype group (*Total cholesterol, $ LDL-cholesterol, # Triglycerides, &insulin, **HOMA IR).

Graphical Abstract



We send the credit author statement of the article

Daniel Antonio de Luis and Juan Jose Lopez designed the study an wrote the article

Olatz Izaola, and Jose Lopez realized nutritional evaluation

David Primo and Daniel de Luis realized biochemical evaluation