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Effect of Specific Allergen Inhalation on Serum Adiponectin in Human Asthma

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Abstract

Background—Adiponectin is associated with asthma. The direction of this association is not known in humans. In mice, this association is bidirectional - allergen inhalation affects serum adiponectin and exogenous adiponectin administration affects asthma. We sought to evaluate whether allergen inhalation affects serum adiponectin in human asthma.

Methods—This study included eight sensitized mild asthmatics and six healthy controls. Asthmatics were challenged with inhaled specific allergen (positive allergen skin test), methacholine, and irrelevant allergen (negative allergen skin test). Controls were challenged with irrelevant allergen. Sequential serum samples were obtained before and nine times after each challenge. Serum adiponectin (primary outcome), leptin, adiponectin-to-leptin ratio, eotaxin, and tumor necrosis factor- α - response curves, area under the curves, baseline and peak concentrations, were evaluated. Statistical analysis used repeated measures ANOVA and paired t-tests.

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Results—There were no significant differences in outcome measures among the challenges in asthmatics or when compared to controls. Type II error is an unlikely explanation for these findings since the study was adequately powered to detect changes in serum adiponectin, as reported in the literature. Further, pooled data showed that serum adiponectin diurnal variation curves were lower in asthma than in controls.

Conclusions—Serum adiponectin concentrations are lower in asthma than controls. Specific allergen inhalation in asthma does not acutely affect serum adiponectin concentrations. The reverse association *i.e.* effect of adiponectin on asthma needs further study. If future studies prove adiponectin to be a protective factor for asthma, modulating adiponectin may open a new approach towards managing asthma.

Keywords

Adipokine; Adiponectin; Allergen inhalation challenge; Asthma; Leptin

INTRODUCTION

Adipokines, a diverse group of proteins secreted from adipose tissue, include various hormones, cytokines, chemokines, and acute-phase proteins. These may be anti-inflammatory, such as adiponectin or pro-inflammatory, such as leptin, eotaxin, and tumor necrosis factor-alpha (TNF- α). Some investigators have hypothesized that adipokines may cause asthma¹⁻³. If this hypothesis is correct, modulating adipokines may open a new approach towards managing asthma.

Recent studies have confirmed that serum adiponectin is associated with asthma^{4,5}. Exogenous adiponectin administration in sensitized mice attenuates allergen-induced increase in airway hyper-responsiveness⁵. In addition, serum adiponectin concentrations decrease following allergen challenge in sensitized mice⁵. The adiponectin-asthma relationship is therefore bidirectional in mice, whereby allergen inhalation affects serum adiponectin and exogenous adiponectin administration affects asthma.

Our recent cross sectional study shows that high serum adiponectin concentrations are associated with decreased risk for asthma in women⁶. However, cross-sectional studies are unable to determine the direction of association and hence the need for longitudinal and interventional studies in this field. Therefore, we determined whether specific allergen inhalation affects serum adipokines in humans with asthma, similar to mice. The primary aim was to determine whether serum concentrations of adiponectin decrease following specific allergen inhalation in asthma. The secondary aim was to determine whether serum concentrations of leptin, eotaxin, and TNF- α increase and whether the ratio of serum adiponectin-to-leptin decreases following specific allergen inhalation.

METHODS AND MATERIALS

Study Design

This was an interventional study including eight sensitized subjects with asthma and six healthy controls. Asthma was defined by the presence of all the following criteria - physician diagnosis of asthma, confirmed skin test 'atopy', presence of nonspecific airway hyper-reactivity (methacholine PC₂₀ of ≤ 16 mg/mL), and presence of specific airway reactivity (*i.e.* FEV₁ decline of $\geq 20\%$) to either inhaled Juniper-Mountain cedar or Bermuda grass allergen on screening evaluation. Controls were defined as those that met none of the first three criteria. All subjects with asthma had mild disease (intermittent or mild persistent in severity). This phenotype was chosen to minimize serious adverse reactions to allergen inhalation and to minimize the confounding effect of asthma medications. The study

inclusion and exclusion criteria are outlined in the Online Supplement. The study protocol was approved by the local institutional review board. Informed consent was obtained from all study participants.

Exposures

All subjects underwent one to three inhalational challenge/s in random order on separate days (as outlined in Table 1). All subjects inhaled incremental concentrations of an irrelevant allergen to which they were not sensitive *i.e.* associated with a negative skin prick test. Two additional inhalational challenges were similarly performed on all subjects with asthma - with a non-immunological stimulus (methacholine) ^{7,8}, and with specific allergen to which the subjects were sensitive (*i.e.* associated with a positive skin prick test) ⁹. The specific allergens used were again Bermuda grass (n = 3) and Juniper-Mountain Cedar (n = 5). Additional details of these procedures are in the Online Supplement.

Outcome Measures

The primary outcome measure was the post-challenge change in concentration of serum adiponectin from baseline. Secondary outcome measures included similar changes in serum concentrations of leptin, adiponectin-to-leptin ratio, eotaxin, and TNF- α . Multiple venous blood samples were obtained in a fasting state at 15 minutes and just before the start of the challenge, just after the completion of the challenge (0 hours), and at 1, 2, 3, 4, 5, 6, 9, and 23 hours after the challenge. Baseline (or 'pre-test') levels were derived from the mean of the first two levels obtained. Additional details are provided in the online data supplement.

Statistical Analysis

All outcome measures, other than serum adiponectin-to-leptin ratio, were logarithmically transformed, due to their non-normal distribution. Statistical analysis used repeated measures (RM) ANOVA, with both the three challenges and time as repeated factors in subjects with asthma, using SAS PROC-MIXED (SAS 9.1.3, Cary, NC). Outcomes between subjects with asthma and controls were similarly compared using case status as the grouping factor and time as the repeated factor. If there were significant differences, post-hoc testing with paired t-tests was performed. Challenge response curves were plotted as geometric means for logarithmically transformed outcome measures, and as arithmetic means for serum adiponectin-to-leptin ratio. Additionally, area under the curves, baseline and peak concentrations of outcome measures were compared. The covariates studied included measures of obesity and insulin resistance which may affect serum adiponectin concentrations in humans ^{10,11}. These covariates are further described in the Online Supplement. Statistical significance was accepted as $p < 0.05$.

Sample size and Power estimates

Based on a previous study ¹² and assuming a modest correlation of 0.7 between baseline and follow-up values, the S.D. of the difference was estimated at 4.4 $\mu\text{g/mL}$ for serum adiponectin (as detailed in Table E4 in the Online Supplement). Therefore, our sample size of eight subjects with asthma was adequate to detect a difference of 5.1 $\mu\text{g/mL}$ for serum adiponectin with $\alpha = 0.05$ and $\beta = 0.2$. This study was therefore adequately powered since the published absolute difference in serum adiponectin in a human intervention study was 6.7 $\mu\text{g/mL}$ ¹². Further, power estimate based on mice allergen challenge studies was also adequate ⁵. The actual power of the study was in fact greater than the above estimation, since data was logarithmically transformed.

RESULTS

Fourteen subjects, eight with asthma and six controls, were studied - mostly pre-menopausal overweight women (see Table 2). Among all challenges, the decline in FEV₁ following specific allergen inhalation was greatest in magnitude ($28.8 \pm 5.3\%$) and duration (Tables 2 in text and E3 in Online Supplement).

Following any inhalational challenge (in both asthma and controls), there was an immediate rise in serum adiponectin concentration above baseline, lasting about an hour (Figure 1, Table E2 in Online Supplement). In addition, there was an immediate decline in serum leptin concentration below baseline, lasting several hours, returning to baseline by the next morning (Figure 2, Table E2 in Online Supplement). Despite these changes, there was no significant change in serum adiponectin-to-leptin ratio as compared to baseline (Figure 3). This lack of significant change was attributed to large standard deviations associated with this measure (Table E2 in Online Supplement). While serum eotaxin concentration (Figure 4) was unaffected by any inhalational challenge, a small transient decline in serum TNF- α concentration 2-3 hours after the specific allergen and methacholine challenges in subjects with asthma was suggested (Figure 5).

Repeated measures analysis of outcome measures among subjects with asthma

Comparison of the three challenges among subjects with asthma showed no significant differences with respect to the response curves, area under the curves, baseline and peak concentrations of serum adiponectin, leptin, adiponectin-to-leptin ratio, and TNF- α (Figures 1-3 and 5, Table E1 in Online Supplement). A difference in the main effect of the eotaxin response curves between specific allergen on one hand and methacholine and irrelevant allergen challenges on the other hand was noted. However, no significant difference at individual time points in (geometric) mean serum eotaxin level was seen among the various curves (all $p > 0.30$). Furthermore, there were no significant differences among the three challenges with respect to area under the curve and baseline and peak concentrations of serum eotaxin (Figure 4, Table E1 in Online Supplement).

Repeated measures analysis of outcome measures between subjects with asthma and controls

Comparison of the specific allergen challenge in subjects with asthma and irrelevant allergen challenge in controls again showed no significant differences with respect to the above outcome measures, except for a difference in main effects between the two eotaxin response curves (Figures 1-5, Table E1 in Online Supplement). Again, no significant differences in (geometric) mean serum eotaxin concentrations at individual time points, including the baseline, between the two curves were noted (all $p > 0.18$).

Further, when data from the three challenges in subjects with asthma were pooled, serum adiponectin curves during the 24-hour period were significantly lower in subjects with asthma, compared to controls ($p = 0.02$). Adjustment for covariates *i.e.* obesity (BMI, DEXA-assessed percent body fat or percent truncal fat separately) and insulin resistance did not explain this observation and in fact further strengthened the association of adiponectin diurnal variation curves with asthma status ($p = 0.01$ for all analyses). Similar results were seen when serum adiponectin-to-leptin ratio was used instead of adiponectin.

DISCUSSION

This study of human subjects with mild asthma does not support the hypothesis established in mice that specific allergen inhalation affects serum adiponectin concentrations. We postulate the following explanations for this discrepancy. First, mice are not the same as

humans and mouse asthma is not the same as human asthma¹³. Quite like the current study, past studies investigating adipokines in insulin resistance have often produced discrepant results between humans and mice (adipsin, resistin, and IL-6)¹⁴. Second, our results may be negative because of subject characteristics. Our subjects were predominantly overweight pre- and post-menopausal women and men with a mild asthma phenotype. The choice of subjects was partly based on safety of challenge testing. Further, the relevant animal studies were done on lean (BALB/cJ) mice of both sexes^{5,15}. Third, the severity of intervention in this study could not match that of the mice allergen challenge studies. Fourth, the results may be negative because of a small sample size (Type II statistical error). Despite the small sample size, our power analysis was adequate to detect changes in serum adiponectin, as reported in the referenced human interventional study as well as in the mice allergen challenge experiments^{5,12}. A careful post-hoc evaluation of the power analysis showed that our assumptions for standard deviations used to calculate effect size were in fact correct (Table E4 in Online Supplement). Furthermore, logarithmic transformation of data resulted in a greater effect size than was postulated in the prior power analysis.

Serum concentrations of adiponectin, an anti-inflammatory adipokine, are reduced among obese subjects^{16,17}. High serum adiponectin concentrations are associated with reduced risk of asthma in women, after adjusting for obesity⁶. In sensitized mice, exogenous adiponectin administration attenuates allergen-induced airway hyper-responsiveness⁵. Further, adiponectin mRNA production in mice decreases following allergen challenge⁵. The adiponectin-asthma relationship in mice is therefore bidirectional. However, the direction of the adiponectin-asthma association is not known in humans.

This study demonstrates transient changes in serum adiponectin and leptin concentrations (Figures 1 and 2) following any inhalational challenge in both asthma and controls. There are two possible explanations for these findings. First, multiple spirometry maneuvers during the challenge test may itself affect the release of adipokines through a mechanism that is independent of the nature of the inhalant. Second and more likely, the change is due to the superimposed diurnal variations in serum adiponectin and leptin concentrations^{18,19}. The diurnal variation in serum adiponectin concentration is characterized by a nocturnal decline with a nadir in the early morning and a subsequent peak in the late morning. After peak levels are reached, there is minimum daytime variation¹⁸. Since all challenges were performed between 0700 and 0800 hours in this study, the subsequent increase in adiponectin in Figure 1 may represent the described diurnal peak. Similarly, leptin shows diurnal variation with a peak between 2200-0300 (median 0120) and a nadir between 0800 and 1740 (median 1033) in one study¹⁹ with an additional delay if subjects were fasting. The decline in serum leptin in Figure 2 may therefore also be explained by normal diurnal variation¹⁹.

Although we found a difference in main effects between the eotaxin response curves (Figure 4, Table E1 in Online Supplement), no significant differences at individual time points in (geometric) mean serum eotaxin concentration were seen among the various curves on post-hoc analysis. We therefore postulate that the statistical difference between the eotaxin curves, as detected by the powerful RM-ANOVA analysis, does not have any clinical significance.

If specific allergen inhalation does not affect serum adiponectin concentrations in human asthma, how can we explain our previous cross-sectional observation that high concentrations of serum adiponectin are associated with decreased risk for asthma⁶? One explanation is that this association is determined by the chronic effects of asthma on adipose tissue. If correct, the acute experimental study conditions may not reflect the chronic effects of asthma on adipose tissue. However, we believe that the more plausible explanation is that

serum adiponectin affects asthma status in humans (*i.e.* a reverse direction of the adiponectin-asthma association than was hypothesized in this study). Thus, high serum concentration of adiponectin may be a protective factor for human asthma^{6,20}. However, this still remains to further investigated.

Additionally, we found that the diurnal variation curves of serum adiponectin concentrations in asthma were lower than controls. This finding is consistent with the result of our previous large cross-sectional study where serum adiponectin measure at a single time point was studied⁶. Adiponectin inhibits mitogen-induced proliferation and migration of cultured murine vascular smooth muscle cells^{17,21} and may have similar effects on airway smooth muscle⁵. Given that adiponectin receptors are expressed in human airway smooth cells²², it is possible that continually decreased serum adiponectin may contribute towards increased airway smooth muscle mass in asthma¹.

The strengths of this study include careful characterization of subjects, multiple (within-group and between-group) comparison of specific allergen challenge, randomized and blinded administration of challenges, and extensive time-sensitive sampling of outcome measures in a fasting state. Further, this is the first human study translated from the novel theory formulated on mice experiments^{5,15,23}.

Potential limitations include possibly lower level of severity of allergen intervention and truncated timing of the last serum sample drawn post-challenge in this human study, as compared to the relevant mice study. Our abbreviated time strategy (23 hours compared to 48 hours in the mice study⁵) was influenced by the limits we could safely impose on fasting human volunteers to be interned in controlled conditions. It is also possible that airway adipokines may have been more relevant than serum adipokines in this study. However, the study objective was to replicate the mouse experiment that measured serum adipokines⁵. Further, measurement of airway adipokines is technically difficult and multiple measurements would not have been possible. Another potential weakness may be the inclusion of men and post-menopausal women in this study. However, post-hoc analysis of the four pre-menopausal women in each group revealed similar results. It should be noted that this study was not designed to evaluate the interaction of sex, menopausal status, or obesity on adiponectin response to allergen challenge. The relevant animal study (that this human study was translated from) was done on lean mice of both sexes⁵.

To summarize, this study is the first to report lower diurnal variation curves of serum adiponectin in subjects with asthma as compared to healthy controls. Further, unlike mice, specific allergen inhalation in humans with mild asthma does not acutely affect serum adiponectin concentrations. We instead hypothesize the reverse association *i.e.* serum adiponectin may affect asthma status in humans⁶. If future studies prove that serum adiponectin is a protective factor for human asthma, modulating adiponectin may open a unique and innovative approach towards managing asthma.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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References

1. Shore SA. Obesity and asthma: lessons from animal models. *J Appl Physiol.* 2007; 102:516–528. [PubMed: 17053103]
2. Beuther DA, Weiss ST, Sutherland ER. Obesity and asthma. *Am J Respir Crit Care Med.* 2006; 174:112–119. [PubMed: 16627866]
3. Shore SA. Obesity and asthma: implications for treatment. *Curr Opin Pulm Med.* 2007; 13:56–62. [PubMed: 17133126]
4. Sood A, Cui X, Qualls C, et al. Association between Asthma and Serum Adiponectin Concentration in Women. *Thorax.* 2008
5. Shore SA, Terry RD, Flynt L, et al. Adiponectin attenuates allergen-induced airway inflammation and hyperresponsiveness in mice. *J Allergy Clin Immunol.* 2006; 118:389–395. [PubMed: 16890763]
6. Sood A, Cui X, Qualls C, et al. Association between asthma and serum adiponectin concentration in women. *Thorax.* 2008 Apr 4. Epub ahead of print.
7. Chai H, Farr RS, Froehlich LA, et al. Standardization of bronchial inhalation challenge procedures. *J Allergy Clin Immunol.* 1975; 56:323–327. [PubMed: 1176724]
8. Allen ND, Davis BE, Hurst TS, et al. Difference between dosimeter and tidal breathing methacholine challenge: contributions of dose and deep inspiration bronchoprotection. *Chest.* 2005; 128:4018–4023. [PubMed: 16354875]
9. Gerblich AA, Campbell AE, Schuyler MR. Changes in T-lymphocyte subpopulations after antigenic bronchial provocation in asthmatics. *N Engl J Med.* 1984; 310:1349–1352. [PubMed: 6232459]
10. Matthews DR, Hosker JP, Rudenski AS, et al. Homeostasis model assessment: insulin resistance and beta-cell function from fasting plasma glucose and insulin concentrations in man. *Diabetologia.* 1985; 28:412–419. [PubMed: 3899825]
11. Mather KJ, Hunt AE, Steinberg HO, et al. Repeatability characteristics of simple indices of insulin resistance: implications for research applications. *J Clin Endocrinol Metab.* 2001; 86:5457–5464. [PubMed: 11701722]
12. Fallo F, Scarda A, Sonino N, et al. Effect of glucocorticoids on adiponectin: a study in healthy subjects and in Cushing's syndrome. *Eur J Endocrinol.* 2004; 150:339–344. [PubMed: 15012619]
13. Mestas J, Hughes CC. Of mice and not men: differences between mouse and human immunology. *J Immunol.* 2004; 172:2731–2738. [PubMed: 14978070]
14. Arner P. Resistin: yet another adipokine tells us that men are not mice. *Diabetologia.* 2005; 48:2203–2205. [PubMed: 16193286]
15. Shore SA, Schwartzman IN, Mellema MS, et al. Effect of leptin on allergic airway responses in mice. *J Allergy Clin Immunol.* 2005; 115:103–109. [PubMed: 15637554]
16. Wulster-Radcliffe MC, Ajuwon KM, Wang J, et al. Adiponectin differentially regulates cytokines in porcine macrophages. *Biochem Biophys Res Commun.* 2004; 316:924–929. [PubMed: 15033490]
17. Ouchi N, Kihara S, Arita Y, et al. Novel modulator for endothelial adhesion molecules: adipocyte-derived plasma protein adiponectin. *Circulation.* 1999; 100:2473–2476. [PubMed: 10604883]
18. Gavrilu A, Peng CK, Chan JL, et al. Diurnal and ultradian dynamics of serum adiponectin in healthy men: comparison with leptin, circulating soluble leptin receptor, and cortisol patterns. *J Clin Endocrinol Metab.* 2003; 88:2838–2843. [PubMed: 12788897]
19. Saad MF, Riad-Gabriel MG, Khan A, et al. Diurnal and ultradian rhythmicity of plasma leptin: effects of gender and adiposity. *J Clin Endocrinol Metab.* 1998; 83:453–459. [PubMed: 9467557]
20. Sood A, Camargo CA Jr, Ford ES. Association between leptin and asthma in adults. *Thorax.* 2006
21. Okamoto Y, Kihara S, Ouchi N, et al. Adiponectin reduces atherosclerosis in apolipoprotein E-deficient mice. *Circulation.* 2002; 106:2767–2770. [PubMed: 12451000]
22. Shore SA, Johnston RA. Obesity and asthma. *Pharmacol Ther.* 2006; 110:83–102. [PubMed: 16297979]

23. Yeatts K, Sly P, Shore S, et al. A brief targeted review of susceptibility factors, environmental exposures, asthma incidence, and recommendations for future asthma incidence research. *Environ Health Perspect.* 2006; 114:634–640. [PubMed: 16581558]

Abbreviation list

ATS	American Thoracic Society
DEXA	Dual energy X-ray absorptiometry
ELISA	Enzyme-linked immunosorbent assay
FEV₁	Forced expiratory volume in one second
HIV	Human immunodeficiency virus
HOMA	Homeostasis model assessment
PC₂₀	Provocative concentration (of methacholine) causing 20% decline in FEV ₁
RM ANOVA	Repeated measures analysis of variance
SD	Standard deviation
TNF-α	Tumor necrosis factor- alpha

Figure 1.

Serum adiponectin (geometric) mean response curves to inhalational challenge. Following any challenge, there was an immediate transient rise in serum adiponectin concentration. Comparison of the three response curves among the eight subjects with asthma using RM-ANOVA showed no significant differences. Similar results were noted when specific allergen challenge in asthma and irrelevant allergen challenge in controls were compared. Logarithmically transformed data shown in Table E2 in the Online Supplement.

Figure 2.

Serum leptin (geometric) mean response curves to inhalational challenge. Following any challenge, there was an immediate transient decline in serum leptin concentration. Comparison of the three response curves among the eight subjects with asthma using RM-ANOVA showed no significant differences. Similar results were noted when specific allergen challenge in asthma and irrelevant allergen challenge in controls were compared. Logarithmically transformed data shown in Table E2 in the Online Supplement.

Figure 3.

Mean response curves of serum adiponectin to leptin ratio to inhalational challenge. Comparison of the three response curves among eight subjects with asthma using RM-ANOVA showed no significant differences. Similar results were noted when specific allergen challenge in asthma and irrelevant allergen challenge in controls were compared. Raw data shown in Table E2 in the Online Supplement.

Figure 4.

Serum eotaxin (geometric) mean response curves to inhalational challenge. A main effect difference in eotaxin response curves was seen in the eight subjects with asthma using RM-ANOVA, between specific allergen challenge on one hand and methacholine and irrelevant allergen challenges on the other hand. However, no significant differences at individual time points were seen between the various curves. Similar results were noted when specific allergen challenge in asthma and irrelevant allergen challenge in controls were compared. Logarithmically transformed data shown in Table E2 in the Online Supplement.

Figure 5.

Serum TNF- α (geometric) mean response curves to inhalational challenge. Comparison of the three response curves among eight subjects with asthma using RM-ANOVA showed no significant differences. Similar results were noted when specific allergen challenge in asthma and irrelevant allergen challenge in controls were compared. Logarithmically transformed data shown in Table E2 in the Online Supplement.

Table 1

List of inhalational challenges performed in random order

Sensitized subjects with asthma	Non-atopic healthy controls
Irrelevant allergen inhalation (allergen tested negative on skin prick test on screening evaluation)	Irrelevant allergen inhalation (allergen tested negative on skin prick test on screening evaluation)
Methacholine inhalation (non-immunological stimulus)	
Specific allergen inhalation (allergen tested positive on skin prick test on screening evaluation and starting challenge dose subsequently evaluated on titration skin allergen prick test)	

Table 2

Baseline characteristics of study subjects

Characteristic	Asthma (n = 8)	Controls (n =6)
Women *	5	4
Pre-menopausal *	4/5	4/4
Age (in years) *	31.4 ± 8.5	39.9 ± 9.7
White race *	7	4
Ex-smokers *	2	3
Age of onset of asthma (years)	12.4 ± 5.4	N/A
Use of inhaled corticosteroids	1	0
Confirmed skin test atopy to common aeroallergens	8	0
Body composition measurements		
Body mass index (kg/m ²) *	26.4 ± 4.9	29.4 ± 6.6
Percent body fat (DEXA) *	35.1 ± 6.3	38.8 ± 12.3
Percent truncal fat (DEXA) *	37.9 ± 7.2	42.3 ± 11.4
Insulin Resistance (HOMA units) *	2.4 ± 1.2	2.5 ± 1.7
Baseline FEV ₁ (in ls. prior to irrelevant challenge) *	3.1 ± 0.5	3.3 ± 0.8
Inhalational challenge test results		
Methacholine PC ₂₀	10.5 ± 9.9	N/A
Percent drop in FEV ₁ following methacholine challenge	26.8 ± 8.9	N/A
Percent drop in FEV ₁ following specific challenge	28.8 ± 5.3	N/A
Percent drop in FEV ₁ following irrelevant challenge *	-3.0 ± 5.2	-2.8 ± 5.7

* There was no significant difference between the two groups with respect to these variables ($p > 0.05$ for all analyses).

Abbreviations - DEXA: Dual energy X-ray absorptiometry; HOMA: Homeostasis model assessment; N/A: Not applicable