Circulating IL-6 Concentrations and Abdominal Adipocyte Isoproterenol-stimulated Lipolysis in Women

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Objective: To examine the association of plasma interleukin-6 (IL-6) concentrations with adiposity and fat cell metabolism in women.
Methods and Procedures: Omental (OM) and subcutaneous (SC) adipose tissue samples were obtained from 48 healthy women (age: 47 ± 5 years, BMI: 26.9 ± 5.3 kg/m^2) undergoing gynecological surgeries. Total and visceral adiposity were assessed by dual-energy X-ray absorptiometry and computed tomography, respectively. Measures of adipocyte lipolysis (basal, isoproterenol-, forskolin-, and cyclic dibutyryl-adenosine monophosphate (AMP)-stimulated) and adipose tissue lipoprotein lipase (LPL) activity were obtained. Plasma IL-6 was measured by radioimmunoassay.
Results: Plasma IL-6 was positively correlated with total body fat mass ($r = 0.32$, $P < 0.05$), SC adipose tissue area ($r = 0.35$, $P < 0.05$), SC adipocyte diameter ($r = 0.30$, $P < 0.05$), and a trend was observed with visceral adipose tissue area ($r = 0.20$, $P < 0.07$). Plasma IL-6 was positively correlated with glycerol released in response to isoproterenol (10^{-5} to 10^{-8} mol/l) by isolated SC ($0.31 \leq r \leq 0.65$, $P < 0.05$) and OM ($0.36 \leq r \leq 0.40$, $P < 0.02$) adipocytes, independent of menopausal status. No correlation was found with LPL activity. A subsample of women with high plasma IL-6 ($n = 10$) was matched with women with low plasma IL-6 ($n = 10$) for total body fat mass. OM adipocyte glycerol release in response to isoproterenol (10^{-5} to 10^{-8} mol/l) was higher in the subsample of women with elevated plasma IL-6 ($P \leq 0.07$).
Discussion: We observed that OM lipolysis was significantly higher in women with elevated plasma IL-6 for a similar body fat mass and menopausal status. These results suggest that higher circulating IL-6 concentrations are associated with increased isoproterenol-stimulated lipolysis especially in OM abdominal adipocytes in women.

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INTRODUCTION

Adipose tissue is known to release a number of cytokines including tumor necrosis factor-α, interleukin-6 (IL-6), adiponectin and resistin, which have been shown to modulate local or systemic metabolism (1,2). Higher circulating concentrations of IL-6 have been positively associated with BMI, waist circumference, visceral fat accumulation as well as elevated triglycerides, plasma-free fatty acid levels, and systemic insulin (1,3,4). Obesity is also associated with increased plasma C-reactive protein (5) possibly through modulation by IL-6 (4–6). Interestingly, weight loss is associated with a reduction in the macrophage infiltration of adipose tissue and an improvement of the inflammatory profile including reduction in plasma IL-6 and C-reactive protein concentrations (5,7,8).

IL-6 has been postulated to have a local impact on adipocyte metabolism. Lipoprotein lipase (LPL) activity was reduced by half in adipose tissue of mice *in vivo* after IL-6 injection (9). This effect was also observed *in vitro* in human adipocytes cultured with dexamethasone and insulin, in which IL-6 decreased LPL activity by 56% in omental (OM) adipose tissue and by 68% in the SC depot (1). Furthermore, when human recombinant IL-6 was added to adipose tissue organ cultures under basal conditions, lipolysis was stimulated (1). Low- or high IL-6 infusions in femoral arteries also stimulated fatty acid release in 18 healthy men, even 8 h after the infusion (10). Another study showed that IL-6 infusion increased fatty acid turnover in type 2 diabetic and healthy patients (11). In isolated adipocytes from mammary adipose tissue incubated with 10 mmol/l of IL-6 for 6 h, Path *et al.* (12) observed no effect on lipolysis, whereas 24-h exposure increased basal and isoproterenol-induced glycerol release.

The objective of this study was to examine the association of plasma IL-6 concentrations with adiposity and fat cell

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metabolism in women. Furthermore, we wanted to study the association of systemic IL-6 concentrations with lipolysis-stimulated downstream of the β-adrenergic receptor by using postreceptor-acting agents forskolin- and dibutyryl cyclic adenosine monophosphate (AMP). We tested the hypothesis that higher circulating IL-6 concentrations are associated with increase in isoproterenol-stimulated lipolysis in OM and SC abdominal adipocytes. On the basis of the fact that isoproterenol-stimulated lipolysis responsiveness is higher in the visceral fat depot (13) and that visceral adipose tissue secretes higher levels of IL-6 (2), we hypothesized that higher circulating IL-6 concentrations are associated with an enhanced isoproterenol-stimulated lipolysis in OM abdominal adipocytes compared to isoproterenol-stimulated lipolysis in SC adipocytes.

METHODS AND PROCEDURES

Subjects

Women for this study were recruited through the elective surgery schedule of the Gynecology Unit of the Laval University Medical Center. This study included 48 healthy women undergoing abdominal gynecological surgery. Women of the study elected for total (n = 22) or subtotal (n = 2) abdominal hysterectomies, some with salpingo-oophorectomy of one (n = 8) or two (n = 16) ovaries. Reasons for surgery included one or more of the following: menorrhagia/menometrorrhagia (n = 24), myoma/fibroids (n = 18), incapacitating dysmenorrhea (n = 7), pelvic pain (n = 2), benign cyst (n = 9), endometriosis (n = 6), adenomyosis (n = 2), pelvic adhesions (n = 4), benign cystadenoma (n = 1), endometrial hyperplasia (n = 3) or polyp (n = 1) and ovarian thecoma (n = 1). Menstrual status was obtained for 47 women based on plasma follicle-stimulating hormone measures, a menstrual history questionnaire and from medical files (menopausal status could not be determined with the information obtained for one participant). Thirty-six women were pre- or perimenopausal, five of whom were using hormone therapy. Eleven women were menopausal, three of whom used a gonadotropin-releasing hormone agonist and one of whom used hormone therapy. Before their inclusion in the study, all subjects provided written informed consent. This study was approved by the medical ethics committees of Laval University and Laval University Medical Center.

Body fatness and body fat distribution measurements

Measurements of body fatness and body fat distribution were performed on the morning of or within a few days before or after the surgery. Measures of total body fat mass, fat percentage, and fat-free mass were determined by dual-energy X-ray absorptiometry using a Hologic QDR-2000 densitometer and the enhanced array whole-body software V5.73A (Hologic, Bedford, MA). Abdominal SC and visceral adipose tissue cross-sectional area measurements were analyzed by computed tomography using a GE Light Speed 1.1 CT scanner (General Electric Medical Systems, Milwaukee, WI). Subjects were examined in the supine position, with arms stretched above the head. The scan was performed at the L4–L5 vertebrae level. Quantification of visceral adipose tissue area was done by delineating the intra-abdominal cavity at the internal-most aspect of the abdominal and oblique muscle walls surrounding the cavity and the posterior aspect of the vertebral body using the ImageJ 1.33u software (National Institutes of Health). Adipose tissue was highlighted and computed using an attenuation range from −190 to −30 Hounsfield units. The coefficient of variation (CV) between two measures from the same observer (n = 10) were 0.0, 0.2, and 0.5 for total, SC, and visceral adipose tissue areas, respectively.

Adipose tissue sampling

SC adipose tissue was collected at the site of surgical incision (lower abdomen) and OM adipose tissue was collected from the greater omentum (epiploon). Samples collected during the surgical procedure were immediately carried to the laboratory in 0.9% saline preheated at 37 °C. Adipocyte isolation was performed with a portion of the fresh biopsy. The remaining tissue was immediately frozen and stored at −80 °C in liquid nitrogen for other analyses.

Adipocyte isolation, lipolysis, and LPL activity

Tissue samples were digested for 45 min at 37 °C with collagenase type I in Krebs-Ringer-Henseleit buffer supplemented with 5 mmol/l glucose, 0.1 μmol/l adenosine, 0.1 mg/ml ascorbic acid and 4% electrophoresis grade, delipidated bovine serum albumin according to a modified version of the Rodbell method (14). Adipocyte suspensions were filtered through nylon mesh and washed three times with the buffer. For cell size measurements, mature adipocyte suspensions were visualized using a contrast microscope attached to a camera and computer interface. Pictures of cell suspensions were taken and the Scion Image software was used to measure the size (diameter) of 250 adipocytes for each tissue sample.

Lipolysis experiments were performed by incubating isolated cell suspensions for 2 h at 37 °C in Krebs-Ringer Henseleit buffer, with or without β-adrenergic receptor agonist isoproterenol in concentrations ranging from 10^{-10} to 10^{-5} mol/l, or postreceptor-acting agents dibutyryl cyclic AMP (10^{-3} mol/l) and forskolin (10^{-5} mol/l). Glycerol release in the medium was measured by bioluminescence using the nicotinamide adenine dinucleotide-linked bacterial luciferase assay (15), a Berthold Microlumat plus bioluminometer (LB 96 V) and the WinGlow software (EG&G, Bad Wildbad, Germany). Intra- and interassay CVs for glycerol release measurements were 5.9 and 13.9%, respectively. Lipid weight used was measured by performing Dole's extraction. Average adipocyte weight and cell number in the suspensions were calculated using lipid weight, average cell volume and the density of triolein. Lipolysis results were expressed in μmoles/10^6 cells/2 h.

Heparin-releasable LPL activity was determined in 30–50 mg adipose tissue samples by the method of Taskinen *et al.* (16). Tissue eluates were obtained by incubating the sample in Krebs-Ringer phosphate buffer and heparin at 37 °C for 90 min. The eluates were then incubated with excess concentrations of unlabeled and ^{14}C-labeled triolein in a Tris-albumin buffer emulsified with ultrasound. The reaction was carried out at 37 °C for 60 min with agitation. Free fatty acids were isolated by the Belfrage extraction procedure. Porcine plasma was used as a source of Apo-CII to stimulate LPL activity, and unpasteurized cow's milk as an internal LPL activity standard for interassay variations. Activity results were expressed in nmol oleate/10^6 cells/h.

Plasma cytokine levels

Blood samples were drawn after a 12-h fast on the morning of the surgery. Plasma adiponectin, resistin, and IL-6 levels were measured by enzyme-linked immunosorbent assay (B-Bridge International, Sunnyvale, CA; Linco Research, St-Charles, MO; and R&D Systems, Minneapolis, MN, respectively). Intra-assay CVs for IL-6 measurements ranged from 6.9 to 7.4% and interassay CVs ranged from 6.5 to 9.6%.

Statistical analyses

Spearman rank correlation coefficients were computed to quantify associations between circulating IL-6 concentrations and characteristics of the 48 women, adiposity variables and adipocyte measurements. A subsample of women with high plasma IL-6 (n = 10) was matched with women with low plasma IL-6 (n = 10) for similar total body fat mass values. Characteristics of the women and lipolysis measures were compared in these two groups using paired t-tests. Covariance analyses were performed to assess the effects of menopausal status, fat mass, and adipocyte size on the association between plasma IL-6 and lipolysis measures. All statistical analyses were performed using the JMP statistical software (SAS Institute, Cary, NC).

RESULTS

Characteristics of the sample of 48 women are shown in Table 1. Women were 47.5 years old and were slightly overweight with an average BMI of 26.9 kg/m^2. Adiposity values covered a wide spectrum, with body fat percentages ranging from 19.6 to 47.5%.

Table 2 shows Spearman rank correlation coefficients between physical characteristics of the 48 women and circulating IL-6 concentrations. Body fat-free mass, SC abdominal adipose tissue area, and resistin levels ($P \leq 0.01$) were positively associated with plasma IL-6; so were body weight, BMI, total body fat mass, and total abdominal adipose tissue areas ($P \leq 0.05$). We also observed a trend for a positive correlation between circulating IL-6 concentrations and visceral abdominal adipose tissue area.

Table 3 shows Spearman rank correlation coefficients between circulating IL-6 concentrations and adipocyte size, LPL activity as well as lipolysis measures in SC and visceral adipose tissue or adipocytes. SC adipocyte size was positively and significantly correlated ($P \leq 0.05$) with circulating IL-6 concentrations while OM adipocyte size and LPL activity were not. Isoproterenol-stimulated (10^{-5} mol/l to 10^{-8} mol/l) lipolysis measures in OM adipocytes were significantly correlated with plasma IL-6 concentrations ($P \leq 0.01$), and so were SC adipocyte isoproterenol-stimulated (10^{-5} mol/l to 10^{-8} mol/l) lipolysis measures ($P \leq 0.05$). When adjusting for adipocyte size, isoproterenol-stimulated lipolysis was still positively associated with circulating IL-6 in both fat depots with minor differences. Statistical adjustment for total body fat mass slightly decreased the correlation coefficients, but trends for correlations between plasma IL-6 concentrations and isoproterenol-stimulated lipolysis were still observed in both fat depots, especially in OM adipocytes (not shown).

Dibutyryl cyclic AMP- and forskolin-stimulated lipolysis in OM adipocytes were positively correlated with plasma IL-6 ($P \leq 0.05$). Forskolin-stimulated lipolysis was positively correlated with circulating IL-6 concentrations in SC adipocytes ($P \leq 0.05$).

Table 1 Characteristics of the study sample of 48 women

Variable	Mean ± s.d.	Range
Age (years)	47.5 ± 5.0	39.6–61.7
Weight (kg)	70.5 ± 15.8	48.5–110.5
Height (cm)	161 ± 6	151–174
BMI (kg/m^2)	26.9 ± 5.3	17.2–41.3
Total body fat mass (kg)	25.1 ± 9.7	10.0–50.8
Body fat-free mass (kg)	43.1 ± 6.9	32.5–63.0
Percent body fat (%)	34.6 ± 6.6	19.6–47.5
Abdominal adipose tissue areas (cm^2)		
Total	417 ± 184	128–991
Visceral	95 ± 47	34–233
Subcutaneous	322 ± 143	94–759
Cytokines		
Adiponectin (ng/ml)[a]	47.2 ± 21.2	1.6–103.4
Resistin (ng/ml)[b]	23.2 ± 6.4	13.0–48.3
IL-6 (pg/ml)	2.0 ± 1.2	0.6–5.5

Menopausal status: Thirty-one pre- or perimenopausal women, five pre -or perimenopausal women using hormone therapy, eight postmenopausal women, three postmenopausal women using GnRH agonist and one postmenopausal woman using HRT.
GnRH, gonadotropin-releasing hormone; HRT, hormone replacement therapy; IL-6, interleukin-6.
[a]*n* = 46. [b]*n* = 47.

Table 2 Spearman rank correlation coefficients between principal characteristics of the 48 women and circulating IL-6

Variables	Circulating IL-6	P value
Age	0.04	NS
Weight	0.35	≤0.05
Height	0.17	NS
BMI	0.31	≤0.05
Total body fat mass	0.32	≤0.05
Body fat-free mass	0.37	≤0.01
Percent body fat	0.27	≤0.07
Abdominal adipose tissue areas (cm^2)		
Total	0.35	≤0.05
Visceral	0.2	≤0.07
Subcutaneous	0.35	≤0.01
Cytokines		
Adiponectin (ng/ml)[a]	–0.29	NS
Resistin (ng/ml)[b]	0.39	≤0.01

IL-6, interleukin-6; NS, nonsignificant.
[a]*n* = 46. [b]*n* = 47.

Table 3 Spearman rank correlation coefficients between adipocyte size, LPL activity, lipolysis measures, and circulating IL-6 in subcutaneous and visceral adipose tissue

	Circulating IL-6	
Variables	Subcutaneous adipocytes	Omental adipocytes
Adipocyte size	0.30*	0.20
LPL activity	0.03	0.03
Lipolysis		
Basal	0.14	0.19**
Isoproterenol (10^{-5} mol/l)	0.35*	0.36**
Isoproterenol (10^{-6} mol/l)	0.33*	0.40**
Isoproterenol (10^{-7} mol/l)	0.35*	0.38**
Isoproterenol (10^{-8} mol/l)	0.31*	0.38**
Isoproterenol (10^{-9} mol/l)	0.17	0.19
Isoproterenol (10^{-10} mol/l)	–0.02	0.26
Dibutyryl cyclic AMP (10^{-3} mol/l)	0.10	0.28*
Forskolin (10^{-5} mol/l)	0.29*	0.35*

AMP, adenosine monophosphate; IL-6, interleukin-6; LPL, lipoprotein lipase.
LPL activity expressed in nmol oleate/10^6 cells/h. Lipolysis expressed in μmol/10^6 cell/2 h.
*$P \leq 0.05$. **$P \leq 0.01$.

Table 4 Characteristics of women with high or low plasma IL-6, but individually matched for total body fat mass

Variables	Low IL-6 (*n* = 10)	High IL-6 (*n* = 10)	*P* value
Age (years)	46.5 ± 3.7	46.0 ± 5.9	NS
Weight (kg)	64.0 ± 10.0	66.0 ± 13.0	NS
BMI (kg/m²)	25.5 ± 4.4	25.7 ± 4.9	NS
Total body fat mass (kg)	21.5 ± 7.5	21.7 ± 8.2	NS
Body fat-free mass (kg)	40.4 ± 3.5	42.6 ± 6.2	NS
Abdominal adipose tissue areas (cm²)			
Total	356 ± 143	339 ± 135	NS
Visceral	82 ± 31	76 ± 30	NS
Subcutaneous	277 ± 114	263 ± 111	NS
Cytokines			
IL-6 (pg/ml)	0.89 ± 0.21	3.58 ± 1.02	<0.01
Adiponectin (ng/ml)	54.1 ± 15.6	46.1 ± 28.6	NS
Resistin (ng/ml)	19.5 ± 2.6	23.3 ± 4.9	0.07

Data are means ± s.d. Menopausal status: Twelve pre- or perimenopausal women, three pre- or perimenopausal women using HRT, two postmenopausal women, two postmenopausal using GnRH agonist, one postmenopausal woman using HRT and one woman with undefined status are included and equally distributed in each group.
GnRH, gonadotropin-releasing hormone; HRT, hormone replacement therapy; IL-6, interleukin-6; NS, nonsignificant.

Characteristics of women with high or low circulating IL-6 concentrations individually matched for total body fat mass are shown in **Table 4**. All variables including age, BMI, weight, total body fat mass, and menopausal status were similar among the two groups, except for plasma IL-6 concentrations, which were significantly different in the group with low plasma IL-6 concentrations when compared with the group with high plasma IL-6 concentrations ($P \leq 0.01$). Plasma resistin concentrations also tended to be higher in the high IL-6 subgroup ($P = 0.07$). Subcutaneous and OM adipocyte size and LPL activity were not significantly different between the two groups (**Figure 1**). However, isoproterenol-stimulated (10^{-5} mol/l to 10^{-8} mol/l) lipolysis in OM adipocytes was significantly higher in the group with high circulating IL-6 concentrations ($0.05 \leq P \leq 0.07$). A similar trend was found for some isoproterenol doses in SC fat cells, although it did not reach significance (P values of 0.12, 0.11, 0.13, 0.09, 0.17, and 0.52 for isoproterenol, 10^{-5}, 10^{-6}, 10^{-7}, 10^{-8}, 10^{-9}, and 10^{-10} mol/l, respectively). In OM adipocytes of women with low vs. high plasma IL-6, we observed nonsignificant trends for differences in responsiveness to dibutyryl cyclic AMP (0.84 ± 0.09 vs. 1.54 ± 0.28, respectively, $P = 0.14$) and forskolin (0.34 ± 0.05 vs. 0.96 ± 0.24, respectively, $P = 0.09$). In SC adipocytes of women with low vs. high plasma IL-6 concentrations, we observed no difference in responsiveness to dibutyryl cyclic AMP (1.55 ± 0.22 vs. 2.18 ± 0.41, respectively, $P = 0.21$) and forskolin (1.78 ± 0.24 vs. 2.46 ± 0.32, respectively, $P = 0.78$).

Plasma IL-6 concentrations were higher in postmenopausal than in pre- or perimenopausal women not taking hormone therapy (2.78 ± 1.35 pg/ml vs. 1.78 ± 0.95 pg/ml, $P = 0.02$).

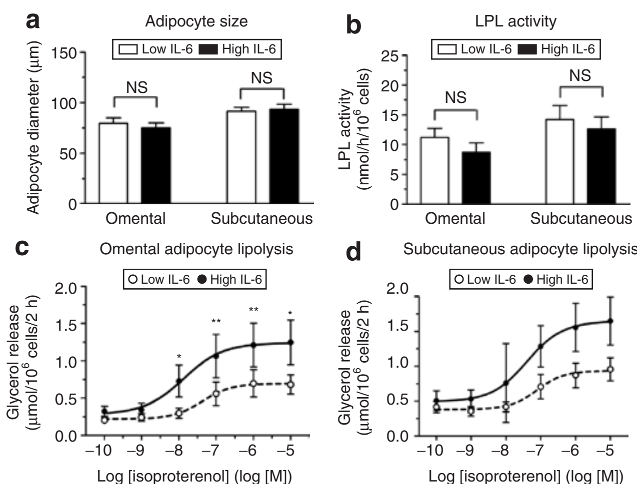

Figure 1 Adipocyte size, lipoprotein lipase activity, and lipolysis in omental and subcutaneous adipose tissue/cells in women with high or low plasma interleukin-6 matched for total body fat mass. IL-6, interleukin-6; LPL, lipoprotein lipase. $*P \leq 0.05$. $**P \leq 0.07$.

When these two groups were compared, we also observed that postmenopausal women were significantly older ($P \leq 0.001$), had higher total body fat mass and percent body fat ($P \leq 0.05$ for both), and higher abdominal adipose tissue areas ($P \leq 0.05$). When excluding postmenopausal women and women on hormone replacement therapy (pre-, peri-, or postmenopausal), correlations between IL-6 and adiposity measures, adiponectin, and resistin levels were unchanged. Furthermore, when menopausal status was considered as a covariate in our analyses, we observed that, except for percent body fat, associations between circulating IL-6 concentrations and weight, total body fat mass, body fat-free mass, and abdominal adipose tissue areas were not affected (not shown).

A trend for a higher isoproterenol-stimulated lipolysis was observed in postmenopausal women (10^{-5} mol/l and 10^{-6} mol/l, $P \leq 0.08$) in fat cells from the SC depot. However, when excluding postmenopausal women and women on hormone replacement therapy (pre-, peri-, or postmenopausal), correlations between plasma IL-6 concentrations and SC isoproterenol-stimulated lipolysis remained unaltered. Furthermore, when menopausal status was considered as a covariate in our analyses, similar results were observed: plasma IL-6 concentrations were significantly correlated with isoproterenol-stimulated lipolysis independent of menopausal status in both depots.

DISCUSSION

The aim of this study was to examine the association between circulating IL-6 concentrations and abdominal SC and OM adipocyte metabolism in women. Some studies had already shown a lipolytic effect of IL-6, either directly or on isoproterenol-induced lipolysis (1,10–12). We aimed to test the hypothesis that higher circulating IL-6 concentrations would be associated with increased isoproterenol-stimulated lipolysis, especially in OM fat cells. As expected, our results show that most measures of adiposity such as BMI, weight, and fat mass were positively correlated with circulating IL-6 concentrations.

Isoproterenol-stimulated lipolysis was also positively correlated with plasma IL-6. Moreover, in women with elevated plasma IL-6 concentrations, OM adipocytes were more responsive to isoproterenol compared to women with low plasma IL-6 concentrations, despite the fact they were matched for adiposity. Covariance analyses confirmed this finding. These results show for the first time that circulating IL-6 concentrations are related to high OM fat cell isoproterenol-stimulated lipolysis independent of adiposity.

Regarding the correlations between adiposity measures and circulating IL-6 concentrations, several studies are concordant with our results. In a study by Fencki *et al.* in overweight or obese postmenopausal women, plasma IL-6 concentrations were positively correlated with BMI, waist circumference, and visceral fat (3). Furthermore, Khaodhiar *et al.* found that across a broad range of obesity values, circulating IL-6 concentrations always correlated with BMI (17). Park *et al.* studied nonobese and obese subjects from both sexes and found that plasma IL-6 concentrations were positively correlated with weight, BMI, waist circumference, hip circumference, and waist-to-hip ratio in all subjects (18). In the same study, especially in obese subjects, circulating IL-6 concentrations were positively associated with BMI, waist circumference, and visceral adipose tissue area (18). All these results are concordant with our findings and confirm that in several populations, adiposity is positively associated with plasma IL-6.

As previously discussed, adipose tissue is an important source of proinflammatory cytokines (19). It is now clear that adipocyte metabolism is influenced by these cytokines, and especially by IL-6. Lipolysis responsiveness to IL-6 has already been studied by many authors. IL-6 stimulates lipolysis in human adipocyte or preadipocyte cultures (1,12). Path *et al.* observed that a 24-h exposure to IL-6 increased basal and isoproterenol-induced glycerol release in isolated adipocytes (12). Moreover, infusion of IL-6 to healthy humans also increased lipolysis (10). In this study, consistent with previous observations, we found positive correlations between high circulating IL-6 concentrations and isoproterenol-stimulated lipolysis. A novel aspect of this study was the use of postreceptor-acting agents. Trujillo *et al.* suggested that high levels of IL-6 within adipose tissue may contribute to alterations in gene or protein expression that influence basal lipolysis (1). In addition, Path *et al.* postulated that IL-6 augments catecholamine-induced stimulation of lipolysis through a direct effect on β-adrenergic receptors (12). Our results indirectly suggest that this effect may include other components of the lipolytic cascade. Indeed, we found that forskolin and dibutyryl cyclic AMP responsiveness were positively correlated with IL-6, similar to isoproterenol responsiveness, especially in the OM fat component.

This study allowed us to examine both OM and SC adipocyte lipolysis. Consistent with our hypothesis, we observed that the difference in isoproterenol-stimulated lipolysis between women with high circulating IL-6 concentrations compared with women with low plasma IL-6 was most pronounced in OM adipose tissue. OM adipose tissue secretes three times more IL-6 than SC adipose tissue (2). In contrast, in response to direct IL-6 treatment, Trujillo *et al.* found that there was no significant depot difference in lipolysis in adipose tissue organ cultures under basal conditions from six obese subjects (1). The fact that we examined isoproterenol-stimulated lipolysis and circulating levels of the cytokine could possibly explain this discrepancy. Second, our study was conducted in a group of lean to moderately obese subjects. Examination of fat cells form morbidly obese individual may have minimized depot differences in glycerol release in response to IL-6.

Since obesity is positively associated with both adipocyte size and plasma Il-6 (17,20), one could hypothesize that elevated circulating IL-6 concentrations are found in subjects with large adipocytes, and consequently consider cell size to explain our association between circulating IL-6 concentrations and lipolysis. Accordingly, plasma IL-6 concentrations were previously found to be positively associated with adipocyte size (21). Cell size also correlated with IL-6 mRNA and IL-6 secretion by SC tissue in healthy men (22). The same positive correlation between IL-6 secretion and SC cell size was also observed in healthy men and women (23). In our study, circulating IL-6 concentrations were positively correlated with adipocyte size but only in SC cells, which is consistent with other studies. However, when women were matched for the same level of fat mass, we did not observe any difference in adipocyte size between women with high circulating IL-6 concentrations and women with low circulating IL-6 concentrations. Yet, isoproterenol-stimulated lipolysis was still elevated in high IL-6 women, suggesting an association that is independent of fat cell size. Covariance analyses with adipocyte size also confirmed that circulating IL-6 level was an independent predictor of isoproterenol-stimulated lipolysis. Based on these results, adipocyte size cannot be considered to explain the elevated lipolysis in women with high circulating IL-6 concentrations. Because 90% of adipose tissue IL-6 release originates from cells other than adipocytes (2,24,25), we suggest that adipocyte size is not a strong determinant of the potential stimulatory effect of IL-6 on isoproterenol-induced lipolysis.

It is important to note that there are some limitations to the use of a cross-sectional design, particularly with respect to cause and effect relationships. Also, we used plasma IL-6 concentrations instead of measuring tissue IL-6 levels or stimulating lipolysis with IL-6 directly like other groups (1,10). However, we know that adipose tissue IL-6 secretion may account for a third of circulating level of the cytokine (5,26). The finding of significant correlation at the systemic level indirectly suggest that stronger associations would have been found with tissue measures.

In conclusion, despite similar adiposity levels, we can speculate that elevated plasma IL-6 concentrations may reflect a more dysfunctional adipose tissue including increased macrophage infiltration and production of inflammatory cytokines. Such profile would be associated with increased isoproterenol-stimulated lipolysis, especially in OM fat cells, independent of adiposity and fat cell size.

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DISCLOSURE

The authors declared no conflict of interest.

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