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Original Article

The Association of Transcript Levels of Proinflammatory Cytokines in Adipose Tissues with Various Adiposity Indices in Women with Obesity

Masoume Aliabadi[1], Sadra Samavarchi Tehrani[1], Mehrnoosh Shanaki[2*]

1. Department of Clinical Biochemistry, School of Medicine, Tehran University of Medical Sciences, Tehran, Iran.
2. Department of Medical Laboratory Technology, School of Allied Medical Sciences, Shahid Beheshti University of Medical Sciences, Tehran, Iran.

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Abstract

Background and Aim: To investigate the association of adipose tissue transcript levels of IL-1β, IL-6, TNF-α, and MCP-1 with various adiposity indices in obese women.
Methods: Real-time PCR was carried out to investigate the mRNA expression level of the mentioned genes in VAT and SAT from all participants.
Results: The results presented higher mRNA levels of IL-6 and MCP-1 in SAT and VAT of obese women, compared to normal-weight women. As well, results showed a positive correlation of IL-6 and MCP-1 with HOMA-IR. Obesity indices including BMI, hip, and WHtR were considerably higher in the obese group in comparison with the control group. More importantly, we observed a positive correlation of mRNA expression of these pro-inflammatory factors in adipose tissues with some obesity indices.
Conclusion: We have shown here that adipose tissue transcript levels of pro-inflammatory cytokines were significantly higher in obese participate than non-obese participants. In obese individuals, this proinflammatory molecules was significantly correlated with various obesity indices. These results suggest that targeting obesity and adipose tissue could prevent the high expression of cytokine.

Keywords: Adipose Tissue; Pro-inflammatory Cytokines; Adiposity Indices; Anthropometric Index; Obesity.

[*]**Corresponding Author:** Dr. Mehrnoosh Shanaki; **Email:** shanaki_m@sbmu.ac.ir

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Introduction

Obesity, as an inflammation-related disease, is a result of either overeating or of inadequate exercise; that is, it is associated with an imbalance between energy intake and expenditure, as well as it is usually defined as a condition of low-grade chronic inflammation (1, 2). Obesity, as a public health problem, affect different physiological roles of the body. Compelling evidence demonstrates that obesity elevates the risk for progression of several diseases, including type 2 diabetes mellitus (T2DM), non-alcoholic fatty liver disease (NAFLD), cardiovascular diseases (CVD), osteoarthritis and cancer, which in turn all of which have adverse effects on the personal satisfaction, work productivity, and medical services costs(3, 4). In spite of enormous effort to prevent, obesity prevalence rates have increased in each age and both male and female genders. Epidemiologically, it is reported that by 2030, 38% and 20% of the world's adult population will be overweight and obese, respectively (5). Moreover, the prevalence of obesity in Iranian population above the age of 18 is 21.7%, and there is a positive correlation between the percentage of obesity and age, especially in women (6). However, the body mass index (BMI) is as simple anthropometric index that used for determining obesity, as proposed by the WHO, the BMI have a few limitations, such as low sensitivity

and inter-individual variability, which might lead to substantial mistakes in the diagnosis of the obesity. To improve these limitations, there are other anthropometric tools that could well add further evidence to BMI; such as waist circumference (WC), waist-to-hip (WHR), waist-to-height ratio (WHtR), Body adiposity index (BAI), The abdominal volume index (AVI) and Weight-adjusted waist index (WWI). In this way, there is increasing data that reveal these novel anthropometric indexes closely correlated with obesity, insulin resistance, and metabolic syndrome (7); however, there are contrary findings with these results (8, 9). Hence, investigation of these anthropometric parameters in obesity-related studies is necessary to receive the information about management of obesity and its related disorders.

It is mostly known that white adipose tissue (WAT) functions as the primary reserve for extra energy in the body, storing excess nutrients as triglyceride, which is categorized into the two types: I) subcutaneous adipose tissue (SAT) and II) visceral adipose tissue (VAT). These two classes of WAT have differences in cellular, molecular, and clinical properties (1, 10). Strategies for precisely assessing SAT and VAT comprise computed tomography (CT) and magnetic resonance imaging (MRI). Although, exposure to radiation, high cost, and discomfort restrict their utilization for clinician screening (11). Hence, a great number of studies are trying to explore low cost, noninvasive and suitable approaches for determining VAT and SAT.

Obesity is associated with chronic inflammation in WAT, which participates in insulin resistance and T2DM. WAT is regarded as a main resource of cytokines and chemokines like interleukin (IL)-1β, interleukin (IL)-6, tumor necrosis factor (TNF)-α and Monocyte chemoattractant protein-1 (MCP-1) (3).

Accumulating evidence indicated that these cytokines play a critical role in the progression and development of obesity. For instance, TNF-α, as a mediator of inflammation, implicated in regulation of growth and differentiation cells. It was reported that this pro-inflammatory cytokine has correlation with insulin resistance. In addition, overexpression of cytokines and chemokines led to enhanced endothelial cell permeability. IL-6 is secreted from different cells such as activated macrophages and lymphocytes, which can elevate the status of TNF-a (12, 13). One of the key pro-inflammatory cytokines, IL-1β, has physiological and pathological functions; it additionally intensifies damage through chronic disease (14, 15). MCP-1, as a crucial chemokine, regulate infiltration of monocytes/macrophages that is essential for response to inflammation and has a vital function in the pathogenesis of obesity-induced inflammation and a variety of metabolic diseases (16, 17). Circulating levels of these cytokines increased in obese subjects in comparison with normal ones (18, 19). Although the relationship between inflammation and obesity is progressively being discovered, the recent understanding of the associations between inflammatory cytokines and novel obesity parameters is still in its infancy. Therefore, in the present study, we aimed to investigate the correlation between some inflammatory cytokines and new obesity-related indexes in obese women compared to normal-weight individuals.

Methods

Study population

The present case-control study was conducted 20 obese (BMI≥30 kg/m^2) and 90 non-obese (BMI <25 kg/m^2) women, aged 20–53 years. The obese patients were selected from Iranian women who undergone bariatric surgery (vertical sleeve gastrectomy and Roux-en-Y gastric bypass) at the bariatric surgery center of Erfan hospital.

Non-obese group was chosen from women who experienced elective cholecystectomy or inguinal hernia at the center of advanced laparoscopic surgeries at Sina and Loqman Hakim hospitals, Tehran, Iran. Exclusion criteria of the present study were women with a prior history of other malignancies, chronic inflammatory diseases, infectious diseases, renal or hepatic disease, T2DM, pregnancy, thyroid and hormonal dysfunctions, as well as subjects who underwent surgery or hospitalizations throughout the last six months. Furthermore, the participants used medication including hypoglycemic agents, anti-hyperlipidemic

drugs; metformin and anti-obesity drugs were excluded. This study was approved by the ethics committee of Iran University of Medical Sciences (Tehran, Iran).

Measurement of Anthropometric indices

All subjects were evaluated for anthropometric measurements. WC was calculated at a level midway between the lowest rib and the iliac crest. In addition, hip was determined at the maximum circumference of the buttocks using inelastic strap. Obesity indices were calculated as follows:

BMI= weight/height^2 (kg/m^2)

WHR= waist circumference (cm)/hip circumference (cm)

WHtR = waist (cm)/height(cm)

$$(BAI) = \frac{hip\ circumference(cm)}{height(m)^{1.5}} - 18$$

Given the gender differences in VAT estimates, the best approach seems the VAI that states the function of VAT and is evaluated based on WC, BMI, triglycerides and high-density lipoprotein cholesterol (HDL-C) via the subsequent formula:

$$AVI = \frac{[2\ cm \times (WC(cm))^2 + 0.7cm \times (WC(cm) - hip(cm))^2]}{1000}$$

WWI for evaluating adiposity by standardizing WC for weight was calculated as WC in cm divided by the square root of weight in kg (cm/√kg). Conicity index (CI) was calculated via the subsequent formula:

$$\text{Conicity index (CI)} = \frac{WC(m)}{0.109\sqrt{weight(kg)/height(m)}} - 18$$

Collecting Types of Adipose Tissue

SAT and VAT samples were collected during the Surgery of obese and non-obese women. Briefly, 0.5-1 g of visceral fat derived from the omentum was eviscerated by a professional surgeon during the bariatric or elective surgery. Furthermore, by cutting a small aperture under the skin, subcutaneous fat (~0.5 g) was isolated with a scalpel blade. To remove extra blood, the AT samples were quickly cleaned in cold and sterile phosphate-buffered saline (PBS) and snap-frozen in liquid nitrogen. Then, the biopsy specimen was kept at a -80°C for molecular analysis.

Quantitative PCR analysis

First of all, frozen VAT and SAT samples were homogenized in liquid nitrogen. Total RNA was isolated using Gene All Hybrid-R RNA purification kit (Gene All, Seoul, Korea). The purity of extracted total RNA was assessed using Nano Drop based on the A260/280 ratios of ~ 2.0. RNA integrity was evaluated by agarose gel electrophoresis.

Then, cDNA synthesis was performed using the Prime Script 1st Strand cDNA Synthesis kit (Takara, Japan). Finally, for determining the gene expression, quantitative reverse transcription-polymerase chain reaction (qRT-PCR) was accomplished using Bio FACT™ 2X Real-Time PCR Master Mix (For SYBR Green I) in a Step-One-Plus TM real-time (ABI Applied Biosystems). Meanwhile, each gene's relative mRNA expression levels were normalized using β-actin and glyceraldehyde 3-phosphate dehydrogenase (GAPDH) expression level.

It is necessary to mention that the standard curves were made for all target and reference genes to determine the linear range of the real-time PCR. for calculating relative levels of target genes in obese women compared to the controls $2^{-\Delta CT}$ was used. The sequences of primer pairs in RTqPCR are denoted in Table 1 of the supplementary file.

Statistical analysis

In the present study, anthropometric parameters were presented as mean±standard deviation (SD), and variables with non-normal distribution was illustrated as median. All data in the graphs were presented as the Mean±SEM. Log-transformation was employed for variables without normal distribution. Anthropometric measurements and gene expression levels of the obese women and the control groups were compared using independent Student's t-test. Subsequently, to get rid of the effects of potential confounders, ANCOVA analysis was carried out. Correlation coefficients were calculated using Pearson's two-tailed correlation analysis.

All data analysis was performed test using SPSS Version 25 (SPSS, Chicago, IL). Moreover, differences with P values <0.05 were considered significant.

Table 1. Anthropometric and laboratory characteristics of participants

Characteristics	Normal-weight participants (n=19)	Obese women (n=20)	Difference p-value
Age (years)	37.68 ± 9.06	34.91± 6.61	0.115
VAI	1.80 ± 0.70	2.27 ± 1.12	0.02
LAP	25.87 ± 13.12	79.68± 36.86	0.000
BMI (kg/m^2)	23.15 ± 1.45	42.11 ± 5.43	0.000
Waist	82.72 ± 7.76	116.66 ± 8.33	0.000
Hip (cm)	93.54 ± 6.02	128.37 ± 10.25	0.000
WHR	0.88 ± 0.05	0.91 ± 0.06	0.808
BAI	27.81 ± 3.21	43.89 ± 6.79	0.000
WWI	10.66 ± 0.70	11.08 ± 0.84	0.3
CI	0.89 ± 0.09	1.09± 0.11	0.6
AVI	13.89 ± 2.34	27.49 ± 4.06	0.001
WHtR	0.51 ± 0.04	0.71 ± 0.06	0.03
SBP (mmHg)	116.09 ± 11.44	121.04 ± 14.52	0.973
DBP (mmHg)	75.22 ± 7.63	77.62 ± 12.53	0.695
FBS (mg/dL)	85.20 ± 7.28	88.99± 8.71	0.529
Creatinine (mg/dL)	0.58 ± 0.15	0.73 ± 0.11	0.001
Uric acid	4.00 ± 0.83	5.50 ± 1.06	0.02
HDL (mg/dL)	44.01 ± 7.33	44.95 ± 7.29	0.001
LDL (mg/dL)	88.69± 28.92	113.41 ± 19.65	0.001
Total Cholesterol (mg/dL)	147.40 ± 37.60	180.40 ± 25.48	0.002
TG (mg/dL)	114.50± 64.98	115.03 ± 61.70	0.291
hs-CRP (mg/L)	1.87 ± 0.96	7.49 ± 5.25	0.003
HOMAIR	1.68 ± 0.74	4.25 ± 1.07	0.000
Insulin (µU/mL)	8.02 ± 3.56	19.41 ± 4.70	0.000

Abbreviations: VAI: Visceral Adiposity Index; LAP: Lipid Accumulation Product; BMI: body mass index; WHR: Waist to Hip Ratio; BAI: Body Adiposity Index; WWI: weight-adjusted-waist index; CI, conicity index; AVI, Abdominal Volume Index; WHtR: waist-to-height ratio; SBP: Systolic Blood Pressure; DBP: Diastolic Blood Pressure; FBS: Fasting Blood Sugar; HDL-C, High-Density Lipoprotein Cholesterol; LDL-C: Low-Density Lipoprotein Cholesterol; TG: Triglyceride; hs-CRP: high sensitive C-Reactive protein; HOMAIR: Homeostatic Model Assessment for Insulin Resistance.
Data are reported as mean±standard error mean (SEM).
p-value <0.5 shows significant differences between the obese and normal-weight groups.

Results

Anthropometric and laboratory characteristics

The laboratory and anthropometric characteristics of the obese and nonobese individuals are shown in Table 1. Women in the obese and normal-weight group had a mean age of 34.91± 6.61 years and 37.68±9.06 years, respectively (p=0.07). Obesity indices including BMI, hip, and WHtR were considerably higher in the obese group in comparison with the control group, though, according to the WHR, there was no significant variation between the two studied groups.

The obese women had greater plasma insulin and HOMA-IR levels than the normal-weight individuals. Laboratory parameters including LDL-c, total cholesterol, hs-CRP, TG, creatinine, and uric acid were higher in the obese group compared to another group. However, no difference in the level of HDL and FBS was identified between the two groups.

Expression of IL-1β, IL-6, TNF-α, and MCP-1 in the normal-weight and obese women

We analyzed the gene expression of IL-1β, IL-6, TNF-α, and MCP-1 as the inflammatory cytokines and chemokine in SAT and VAT of normal-weight (n=19) and obese (n=20) women (Figure 1).

In SAT and VAT of the obese group, the gene expression of IL-6 was significantly increased (P=0.009 and P<0.0001, respectively) compared to those in SAT and VAT of the normal-weight group.

VAT of the normal-weight group indicated the lower expression levels of IL-1β (P= 0.04) compared with the obese group; However, IL-1β gene expression in SAT was not significantly different between the two groups (Figure 1).

In addition, MCP-1 expression levels were significantly higher in the SAT (P=0.005) and VAT (P=0.001) from obese subjects, in comparison to the normal weight ones. However, our data indicated that no significant difference was revealed in the transcript levels of TNF-α in VAT and SAT deposits between non-obese and obese groups (Figure 1).

The correlation analysis of IL-1β, IL-6, TNF-α, and MCP-1 mRNA levels with anthropometric and laboratory parameters

Bivariate correlation analysis of IL-1β, IL-6, TNF-α and MCP-1 mRNA levels with anthropometric and laboratory parameters in the whole population study is shown in Table 2 and 3. As represented in this table, IL-1β mRNA expression in SAT directly correlated with obesity indices; LAP (r = 0.389; p=0.014), BMI (r=0.292; p=0.071), BAI (r=0.359; p=0.025), CI (r = 0.378; p=0.018), AVI (r = 0.357; p = 0.026) and WHtR (r = 0.015; p = 0.388). The SAT IL-6 mRNA expression showed a significant direct correlation with VAI (r=0.440; p=0.005), LAP (r=0.609; p=0.000), BMI (r=0.482; p=0.002), BAI (r=0.497; p=0.001), CI (r=0.444; p=0.005), AVI (r= 0. 463; p=0.003), WHtR (r= 0.534; p=0.000) and HOMAIR (r = 0.393; p = 0.01).

The mRNA expression of MCP-1 in the SAT had a positive correlation with VAI (r = 0.440; p=0.005), LAP (r = 0.528; p = 0.001), BMI (r = 0.281; p = 0.083), BAI (r=0.418; p=0.008), CI (r = 0.408; p=0.010), AVI (r= 0. 344; p=0.032), WWI (r=0.341; p = 0.034), WHtR (r = 0.441; p = 0.005) and HOMAIR (r = 0.360; p = 0.02). The correlation of SAT TNF-α mRNA level with anthropometric and laboratory parameters was not significant.

As summarized in Table 4, VAT mRNA expression of IL-1β positively correlated with obesity indices; LAP (r = 0.322; p=0.046), BMI (r=0.291; p=0.073), BAI (r=0.414; p=0.009), WWI (r = 0.393; p=0.013), CI (r = 0.494; p=0.001), AVI (r=0.454; p=0.004) and WHtR (r = 0.454; p = 0.004), hs-CRP (r=0.326; p=0.04) and HOMAIR (r=0.356; p=0.03).

The VAT IL-6 mRNA expression showed a significant positive correlation with LAP (r = 0.503; p=0.001), BMI (r=0.548; p=0.000), BAI (r=0.557; p=0.000), CI (r = 0.492; p=0.001), AVI (r= 0. 577; p=0.000), WHtR (r= 0.585; p=0.000), hs-CRP (r = 0.045; p = 0.000) and HOMAIR (r = 0.345; p = 0.01).

The mRNA expression of MCP-1 in the VAT had a positive correlation with BMI (r = 0.380; p = 0.017), BAI (r=0.468; p=0.003), CI (r = 0.420; p=0.008), AVI (r= 0.459; p=0.003), WHtR (r = 0.440; p = 0.005), hs-CRP (r = 0.31; p = 0.05) and HOMAIR (r = 0.391; p = 0.01). The VAT TNF-α mRNA expression showed a significant positive correlation with WWI (r = 0.362; p = 0.024), CI (r=0.429; p=0.006), AVI (r = 0.334; p = 0.038).

Bivariate correlation analysis of IL-1β, IL-6, TNF-α, and MCP-1 mRNA levels with each other in the whole population study is summarized in Table 4 and 5. As depicted in this Table, VAT mRNA expression of IL-1β had a positive correlation with transcript level of IL-6 (r = 0.421; p = 0.008) and MCP-1 (r = 0.609; p = 0.000) in VAT of a whole population study. Also, VAT mRNA levels of IL-6 positively correlated with mRNA expression of MCP-1 (r = 0.797; p = 0.00) in VAT of whole population study.

SAT mRNA expression of IL-1β had a positive correlation with transcript level of IL-6 (r = 0.732; p = 0.00) and MCP-1 (r = 0.619; p = 0.00) and TNF-α (r = 0.668; p = 0.00) in SAT of a whole population study.

Furthermore, SAT mRNA levels of IL-6 positively correlated with mRNA expression of MCP-1 (r = 0.759; p = 0.00) and TNF-α (r = 0.270; p = 0.097) in SAT of whole population study.

Table 2. Pearson correlation of IL-1β, IL-6, MCP-1 and TNF-α genes expression in subcutaneous of whole participants with metabolic related indices.

Variable	IL-1β in SAT	IL-6 in SAT	MCP-1 in SAT	TNF-α in SAT
VAI	r=0.222 p=0.174	r=0.440 p=0.005	r=0.388 p=0.015	r=0.161 p=0.328
Log LAP	r=0.389 p=0.014	r=0.609 p=0.000	r=0.528 p=0.001	r=0.173 p=0.291
Log BMI	r=0.292 p=0.071	r=0.482 p=0.002	r=0.281 p=0.083	r=0.013 p=0.939
WHR	r=0.144 p=0.380	r=0.166 p=0.311	r=0.144 p=0.383	r=-0.060 p=0.717
Log BAI	r=0.359 p=0.025	r=0.497 p=0.001	r=0.418 p=0.008	r=0.203 p=0.216
WWI	r=0.301 p=0.062	r=0.293 p=0.071	r=0.341 p=0.034	r=0.263 p=0.106
CI	r=0.378 p=0.018	r=0.444 p=0.005	r=0.408 p=0.010	r=0.221 p=0.177
AVI	r=0.357 p=0.026	r=0.463 p=0.003	r=0.344 p=0.032	r=0.152 p=0.356
WHtR	r=0.388 p=0.015	r=0.534 p=0.000	r=0.441 p=0.005	r=0.165 p=0.316
hs-CRP	r=0.08 p=0.64	r=0.30 p=0.06	r=0.29 p=0.07	r=-0.14 p=0.39
HOMAIR	r=0.24 p=0.15	r=0.393 p=0.01	r=0.360 p=0.02	r=-0.05 p=0.76

Abbreviations: IL-1 β: Interleukin 1 beta; IL-6: Interleukin 6; MCP-1: Monocyte chemoattractant protein-1; TNF-α: Tumor necrosis factor alpha; SAT: Subcutaneous Adipose Tissue; VAT: Visceral Adipose Tissue; Log: Logarithm; VAI: Visceral Adiposity Index; LAP: Lipid Accumulation Product; BMI: body mass index; WHR: Waist to Hip Ratio; BAI: Body Adiposity Index; WWI: weight-adjusted-waist index; CI: conicity index; AVI: Abdominal Volume Index; WHtR: waist-to-height ratio; hs-CRP: high sensitive C-Reactive protein; HOMAIR: Homeostatic Model Assessment for Insulin Resistance.

Table 3. Pearson correlation of IL-1β, IL-6, MCP-1 and TNF-α genes expression in visceral of whole participants with metabolic related indices.

	IL-1β in VAT	IL-6 in VAT	MCP-1 in VAT	TNF-α in VAT
VAI	r=0.055 p=0.739	r=0.160 p=0.330	r=-0.057 p=0.731	r=-0.006 p=0.972
log LAP	r=0.322 p=0.046	r=0.503 p=0.001	r=0.313 p=0.053	r=0.175 p=0.285
log BMI	r=0.291 p=0.073	r=0.548 p=0.000	r=0.380 p=0.017	r=0.121 p=0.462
WHR	r=0.080 p=0.628	r=0.144 p=0.381	r=0.061 p=0.712	r=-0.108 p=0.512
log BAI	r=0.414 p=0.009	r=0.557 p=0.000	r=0.468 p=0.003	r=0.270 p=0.097
WWI	r=0.393 p=0.013	r=0.166 p=0.313	r=0.190 p=0.247	r=0.362 p=0.024
CI	r=0.494 p=0.001	r=0.492 p=0.001	r=0.420 p=0.008	r=0.429 p=0.006
AVI	r=0.454 p=0.004	r=0.577 p=0.000	r=0.459 p=0.003	r=0.334 p=0.038
WHtR	r=0.454 p=0.004	r=0.585 p=0.000	r=0.440 p=0.005	r=0.284 p=0.080
Hs-CRP	r=0.326 p=0.04	r=0.045 p=0.000	r=0.31 p=0.05	r=0.00 p=0.98
HOMAIR	r=0.356 p=0.03	r=0.345 p=0.01	r=0.391 p=0.01	r=0.18 p=0.29

Abbreviations: IL-1 β: Interleukin 1 beta; IL-6: Interleukin 6; MCP-1: Monocyte chemoattractant protein-1; TNF-α: Tumor necrosis factor alpha; SAT: Subcutaneous Adipose Tissue; VAT: Visceral Adipose Tissue; Log: Logarithm; VAI: Visceral Adiposity Index; LAP: Lipid Accumulation Product; BMI: body mass index; WHR: Waist to Hip Ratio; BAI: Body Adiposity Index; WWI: weight-adjusted-waist index; CI: conicity index; AVI: Abdominal Volume Index; WHtR: waist-to-height ratio; hs-CRP: high sensitive C-Reactive protein; HOMAIR: Homeostatic Model Assessment for Insulin Resistance.

Table 4. Bivariate correlation analysis of IL-1β, IL-6, TNF-α, and MCP-1 mRNA levels with each other in subcutaneous AT of the whole population study.

		mRNA Expression in SAT			
	Variable	**IL-1β**	**IL-6**	**MCP-1**	**TNF-α**
mRNA expression in SAT	IL-1 β	r=1.000	r=0.732 p=0.00	r=0.619 p=0.00	r=0.668 p=0.00
	IL-6	r=0.732 p=0.00	r=1.000	r=0.759 p=0.00	r=0.270 P=0.097
	MCP-1	r=0.619 p=0.00	r=0.759 p=0.00	r=1.000	r=0.392 p=0.013
	TNF-α	r=0.668 p=0.00	r=0.270 p=0.097	r=0.392 p=0.013	r=1.000

Table 5. Bivariate correlation analysis of IL-1β, IL-6, TNF-α, and MCP-1 mRNA levels with each other in visceral AT of the whole population study.

		mRNA expression in VAT			
	Variable	IL-1β	IL-6	MCP-1	TNF-α
mRNA expression in VAT	IL-1 β	r=1.000	r=0.421 p=0.008	r=0.609 p=0.00	r=0.281 p=0.083
	IL-6	r=0.421 p=0.008	r=1.000	r=0.797 p=0.00	r=0.194 p=0.238
	MCP-1	r=0.609 p=0.00	r=0.797 p=0.00	r=1.000	r=0.272 p=0.094
	TNF-α	r=0.281 p=0.083	r=0.194 p=0.238	r=0.272 p=0.094	r=1.000

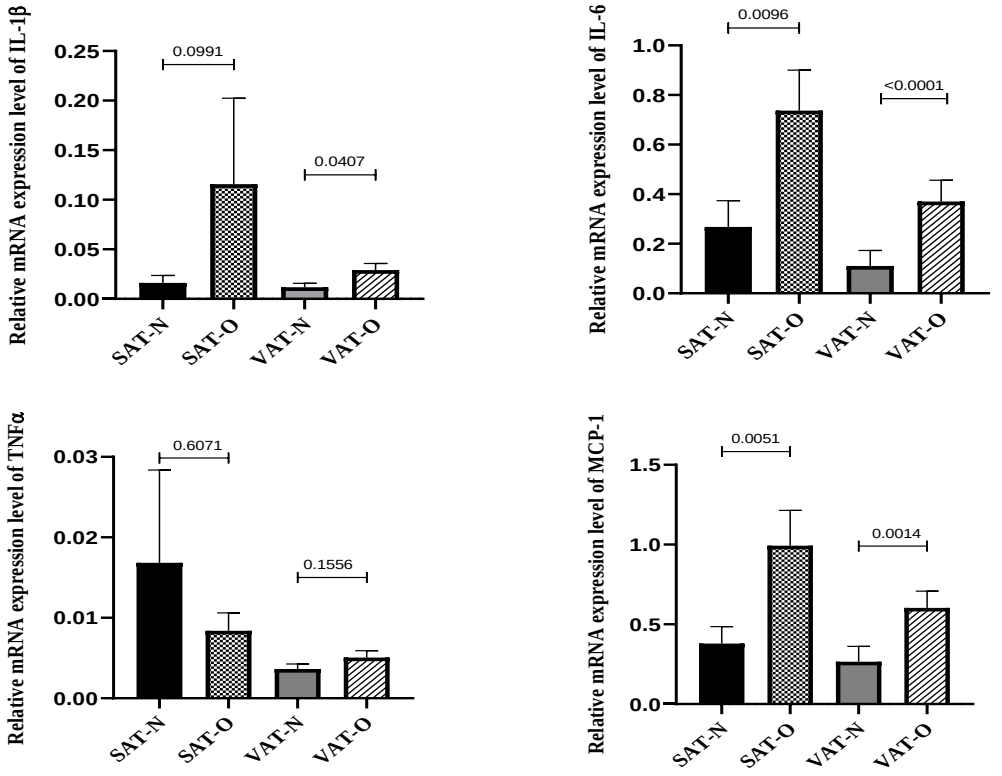

Figure 1. The expression level of IL-1β (a), IL-6 (b), TNF-α (c), and MCP-1 (d) in VAT and SAT from obese and non-obese groups. SAT, subcutaneous adipose tissue; VAT, visceral adipose tissue; N, non-obese; O, obese. Data were shown as the mean ± standard error of the mean (SEM).

Discussion

Emerging evidence declares a noticeable correlation between obesity and low-grade chronic inflammation which is observed in CVD, T2DM, or metabolic syndrome. Cytokines secreted by adipose tissue (AT) play a critical role in the development and progression of obesity. Although the relationship between inflammation and obesity is being majorly recognized, to the best of our knowledge, investigations evaluating the affiliations between AT inflammatory cytokines and novel anthropometric indices are rarely conducted.

In this study, we aimed to assess the possible link between the transcript levels of these molecules and new obesity-related indexes in obese women compared to normal-weight individuals.

We also gathered the results obtained from a set of markers associated with the inflammation in adipose tissue of obese and non-obese women. The mRNA level of IL-6 and MCP-1 was significantly increased in both VAT and SAT of obese women while the non-obese controls have shown the opposite. Furthermore, the mRNA levels of IL-1β in VAT were higher in obese subjects than controls. The differential expression of pro-inflammatory factors has also been observed in individuals suffering from obesity or its other subsequent disorders. For instance, Skurk et al. demonstrated an elevation in IL-6 and MCP-1 circulating levels in obese people and the patients suffering from IR, T2DM, and CVD(20). According to the estimations, roughly 35% of circulating obesity-related IL-6 is secreted from AT(21). Unanimously, previous reports suggested that AT could produce an extensive amount of IL-6 and MCP-1 when obesity emerges(22-24).

As mentioned above, the correlation between inflammation and new obesity-related indexes remains to be fully understood. To investigate the potential association of transcript levels of AT inflammatory cytokines with anthropometric indices, patients were subjected to anthropometric measurements and IL-1β, IL-6, TNF-α, and MCP-1 gene expression in SAT and VAT of obese and normal-weight women was appraised. The results obtained in our study demonstrate a considerable cohesion between the expression of these molecules and various anthropometric indices in SAT and VAT as well.

Based on the evidence found from recent studies, the adiposity is expressed by BAI which is associated with chronic inflammation. Furthermore, WHtR, a novel indicator of visceral obesity, can predict obesity-associated inflammation more efficiently compared to classical indexes like WHR and WC. In obese men, VAI is a new important index indicating the chronic inflammatory process resulted from visceral obesity(25, 26). In our study, we also observed a remarkable correlation between AT mRNA expression of IL-1β and IL-6 and some newer indexes representing obesity such as WHtR, LAP, AVI, and CI. Likewise, the SAT IL-6 and MCP-1 mRNA expression showed a positive relationship with VAI. Besides, the results obtained in our study expressed a great potential in clinical use because VAI could act as a cardiometabolic risk factor predicting the development of overt metabolic syndrome. In addition, this index is a valuable tool reflecting the visceral adiposity degree, adipokine secretion, dyslipidemia, AT dysfunction, and insulin resistance(27); therefore in usual practice, the evaluation of VAI could be applied as an extra indicator of visceral fat dysfunction and might provide early detection of glucose metabolism dysfunction and development of T2DM(28).

Using various analyses, we found that TNF-α levels were not significantly dependent on anthropometric parameters in any group of Iranian women that had been studied. Alongside these findings, Agraval et al conducted an investigation on the North Indian healthy general population from which no momentous correlation between TNF-α with BMI and WC was realized while our results suggested a higher TNF-α level in obese individuals compared to non-obese ones (29). Contrary to our results, Khan et al. observed a substantial correspondence between TNF-α and classical obesity parameters like BMI(30). We presume that the lack of striking links between several cytokines and obesity indexes such as WWI, VAI, and WHR in our study might have resulted from the small size of the groups studied.

In the group of obese women, we observed higher BAI, BMI, and WHtR. Based on the data reported by previous studies, increased rates of BMI, WHtR, and BAI were the indicators predicting cardiovascular risk (30-32). In comparison with BMI and WC, WHtR had a considerably greater discriminatory power; hence, the assessment of WHtR is recognized as a preferred screening tool determining the cardiometabolic risk in adults (33).

Conclusion

All in all, we have found higher expression of IL-1β, IL-6, and MCP-1 in adipose tissues from obese individuals in comparison with normal-weight women. Moreover, our results affirmed a positive correlation between IL-6 and MCP-1 with HOMA-IR. More importantly, a positive relevance

of the mRNA expression of these pro-inflammatory factors in adipose tissues with some obesity indices has also been suggested.

The accumulation of VAT and SAT plays a key role in the up-regulation of obesity-related low-grade inflammation. These results suggest that targeting obesity and adipose tissue could prevent the high expression of cytokine as well as atherosclerosis. Additionally, a combination of classical and novel obesity indexes might exert the greatest potential of clinical utility in the detection of patients having cardiovascular risk factors.

Acknowledgments

Note declared.

Conflict of Interest

The authors declared that they have no conflict of interest associated with this study.

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Ethics

This study was approved by the ethics committee of Iran University of Medical Sciences (Tehran, Iran). **Code:** IR. SBMU RETECH. REC.1397.1175.

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