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Original Article

Analysis of urinary ET-1 levels and kidney function in obese and non-obese young adults

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Abstract

Obesity is characterized by abnormal fat accumulation. The onset and morbidity of obesity are common in adults, leading to various complications, including decreased renal function. Endothelin-1, a molecule that functions as a vasoconstrictor, significantly contributes to the pathophysiology of the renal and cardiovascular systems. This cross-sectional analytical study used purposive sampling, involving 30 obese and 26 non-obese young adult subjects. Urine samples were analyzed using the human Endothelin-1 kit with the ELISA method, while blood samples were analyzed for urea and creatinine levels using an enzymatic technique. The results of urinary Endothelin-1 concentrations between the obese group, which had a median of 92.27 ng/L, and the non-obese group, with a median of 74.41 ng/L. Kidney function test showed in the group for urea 16.50 mg/dL, creatinine 0.70 mg/dL, and GFR 128.35 ml/min/1.73 m^2, while the non-obese group showed urea 22.00 mg/dL, creatinine 0.65 mg/dL, and GFR 148.97 ml/min/1.73 m^2. The levels of Urinary Endothelin-1 were notably elevated in obese individuals compared to those who were not obese. However, kidney function, as measured by urea, creatinine, and GFR, was not consistently worse in the obese group.

Keywords: obesity, endothelin-1, creatinine, urea, glomerular filtration rate.

Introduction

Obesity is a health condition defined by an abnormal or excessive accumulation of body fat, which can lead to various health complications. It is a global issue, with the WHO reporting approximately 2.8 million deaths annually due to obesity [1]. The use of urinary biomarkers as a noninvasive screening method to monitor the progression of systemic diseases has recently gained increasing attention. Urinary biomarkers associated with obesity, metabolic disorders, and type 2 diabetes show a significant correlation with kidney damage and vascular injury in adults [2, 3].

Plasma volume is used to evaluate the glomerular filtration rate over a specific period, indicating the kidney's capacity to completely clear a particular substance. GFR is typically measured by serum creatinine levels, especially in individuals at risk of kidney function impairment [4]. Creatinine and urea are primary indicators of kidney function, where elevated urea levels often indicate decreased kidney function, potentially leading to uremia [5].

Endothelin-1 (ET-1) is a peptide that significantly maintains body volume balance and exhibits vasoconstrictive properties, particularly under pathological conditions. ET-1 contributes to renal blood vessel constriction in the kidneys and inhibits sodium and water reabsorption. Studies in obese rats have shown that higher local ET-1 expression can damage the glomeruli and renal tubules, which indicates early chronic kidney disease (CKD) development [6, 7]. Kidney dysfunction in obese individuals can occur even before the onset

of hypertension or diabetes [8]. However, the relationship between urinary ET-1 levels and kidney function in obesity is not fully understood.

This study aims to explore urinary ET-1 levels and kidney function in obese individuals to understand the extent of renal vascular damage and its impact on kidney function in this population.

Material and methods

Study design and patients

This observational study utilized a cross-sectional design and was carried out between October and November 2024. The study involved 58 samples comprising 30 obese and 26 non-obese subjects at the HUMRC UNHAS and LABKESMAS Makassar. Consent was obtained from all participants in the study. The inclusion criteria consisted of adults aged 19–25 years with a BMI categorized as normal weight (18.5–24.9 kg/m^2) or obesity class I (30–34.9 kg/m^2), class II (35–39.9 kg/m^2), and class III (≥40 kg/m^2) based on the WHO Global BMI classification. Exclusion criteria included subjects with diabetes, cardiovascular diseases, chronic kidney disease, and those with alcohol consumption or smoking habits. This study was approved by the Health Research Ethics Committee under approval number 1060/UN4.6.4.5.31/PP36/2024.

Body height and weight measurement

The subjects' height and weight were measured to determine their Body Mass Index (BMI), which is used to classify individuals as having normal weight or obesity. A questionnaire was used to screen subjects and confirm that they met the inclusion and exclusion criteria. Eligible subjects were provided with an explanation of the research procedures, actions to be taken, and potential impacts eligible subjects were provided with. Participants who agreed to participate were required to sign an informed consent document.

Sample collection

Morning urine samples were collected in sterile containers. The urine was then transferred to 10 ml Falcon tubes and centrifuged at 2000 rpm for 15 minutes at 2–8°C. After centrifugation, the samples were divided into aliquots and stored at -80°C to avoid freeze-thaw cycles until further analysis. Venous blood samples were obtained from the median cubital vein using the vacutainer method, placed into tubes without anticoagulant, and left to clot for 30 minutes at room temperature. The blood was then centrifuged at 3000 rpm for 5 minutes.

Urinary ET-1 was examined at the Hasanuddin University Medical Research Center (HUMRC) using the ELISA method with the Human ET-1 ELISA BT-LAB kit (Bioassay Technology Laboratory, Zhejiang, China). Urea and creatinine were analyzed at the Makassar Public Health Laboratory 1 (LABKESMAS 1) using the enzymatic method with reagents obtained from Clinical Diagnostics Finland (Thermo Fisher Scientific, Finland).

Urinary ET-1 measurement

This test uses the sandwich-ELISA method. The ELISA microplate used is coated with a specific antibody for human ET-1. The sample or standard is introduced into the microplate wells, where the ET-1 present binds to the antibody that coats the wells. Subsequently, a biotinylated antibody targeting human ET-1 and streptavidin-horseradish Peroxidase (HRP) is added and incubated. Then, Solutions A and B are sequentially applied to each well. A blue color develops in wells with Human ET-1, the biotinylated antibody, and the streptavidin-HRP conjugate. The enzyme reaction is halted using a stop solution, which changes the color to yellow. The optical density (OD) is subsequently measured at 450 nm with a microplate reader.

Urea and serum creatinine measurement

Urea is measured by hydrolyzing urea into ammonia using urease, followed by a reaction between the resulting ammonia and a reagent to form a colored compound, which is measured spectrophotometrically. Meanwhile, creatinine measurement involves several enzymatic reaction steps that produce hydrogen peroxide as the final product. This hydrogen peroxide then reacts with another reagent to form a colored compound, which is also measured spectrophotometrically.

Statistical analysis

The data were structured according to the research objectives, and statistical analyses were conducted using SPSS software. The normality of data distribution was evaluated using the Kolmogorov-Smirnov test.

Descriptive statistics, such as range, median, mean, standard deviation, and distribution, were calculated. Independent t-tests were used to compare groups with normally distributed data, while Mann-Whitney U tests were utilized for non-normally distributed data. The association between urinary ET-1 levels and kidney function was analyzed using Spearman's rank correlation test. For inferential statistics, a p-value below 0.05 was deemed statistically significant, and 95% confidence intervals were provided when relevant.

Results

A total of 58 participants took part in this study, comprising 30 obese and 28 non-obese individuals who fulfilled the inclusion criteria. The participants were made up of 15 males and 43 females, with an average age of 21 years. Table 1 summarizes the demographic and physical characteristics of the participants.

The statistical analysis showed significant differences in BMI, systolic blood pressure, and diastolic blood pressure between the obese and non-obese groups ($p<0.05$). Additional findings on kidney function and urinary ET-1 levels are summarized in Table 2.

Urinary ET-1 levels were significantly higher in the obese group compared to the non-obese group ($p=0.038$). However, no significant differences were found in urea, creatinine, or eGFR between the two groups ($p>0.05$), as detailed in Table 3.

Figure 1 shows that an increase in obesity class is associated with higher urinary endothelin-1 levels. Additionally, Figure 2 indicates that creatinine levels are higher in obese individuals compared to the control group, while GFR is lower. Urea levels are also higher in obese individuals than in the control group.

Spearman correlation analysis revealed no significant association between urinary ET-1 levels and kidney function markers (urea, creatinine, and eGFR; $p>0.05$). A slight negative correlation was identified for urea and creatinine, while a slight positive correlation was observed for eGFR, as presented in Table 4.

Discussion

The results of this study indicate that urinary ET-1 levels are significantly higher in the obese group. This finding is consistent with previous research showing that obesity is associated with increased ET-1 levels due

Table 1: Characteristics of the study subjects.

Variable	N=58 (%)	Mean±SD	Median (Min–Max)
Age (year)		21.17±1.63	21.00 (19.00–25.00)
Gender			
Woman	43 (74.91%)		
Man	15 (25.90%)		
Group:			
Obesity	30 (51.70%)		
Obesity 1	5 (8.60%)		
Obesity 2	19 (32.80%)		
Obesity 3	6 (10.30%)		
Non-obese	28 (48.30%)		
BMI (kg/m²)		27.14±6.79	25.66 (19.00–43.50)
ET-1 urine		83.79±59.75	79.35 (3.26–250.11)
GFR (ml/min/1.73 m²)		137.47±38.00	133.66 (71.9–287.00)
Creatinine (mg/dL)		0.71±0.17	0.70 (0.40–1.20)
Urea (mg/dL)		19.94±5.72	20.50 (8.00–38.00)
Sistolic (mmHg)		112.41±14.64	110.00 (90.00–164.00)
Diastolic (mmHg)		76.60±9.57	78.50 (60.00–111.00)

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Table 2: Differences in urinary ET-1, urea, creatinine, and GFR between obese and non-obese subjects.

Characteristics	Obesity (n=30) Median (Min–Max)	Non-obese (n=26) Median (Min–Max)	p
BMI (kg/m²)	31.86 (25.30–43.50)	21.17 (19.00–23.20)	<0.001*
Systolic (mmHg)	117.00 (100.00–164.00)	110.07 (90.00–120.00)	0.001*
Diastolic (mmHg)	80.00 (63–111.00)	70.00 (60.00–80.00)	0.005*

Note: * – p-value is significant.

Table 3: Differences in urinary ET-1, urea, creatinine, and GFR between obese and non-obese subjects.

Variable	Obesity (n=30) Median (Min–Max)	Non-obese (n=26) Median (Min–Max)	p
ET-1 urine (ng/L)	92.27 (11.31–250.11)	74.41 (3.26–169.04)	0.038*
Urea (mg/dL)	16.50 (8.00–38.00)	22.00 (12.00–28.00)	0.113
Creatinine (mg/dL)	0.70 (0.40–1.00)	0.65 (0.50–1.20)	0.828
GFR (ml/min/1.73 m²)	128.35 (63.00–111.00)	148.97 (71.90–207.70)	0.697

Note: * – p-value is significant.

to enhanced adipocyte activity [9, 10]. ET-1, a potent vasoconstrictor synthesized by endothelial cells and adipocytes, plays a crucial role in maintaining renal hemodynamics [11, 12].

Indicators of kidney function suggest a tendency toward decreased kidney function in young adults with obesity, characterized by high creatinine levels and low GFR. However, kidney function, as measured by urea, creatinine, and GFR, did not show a significant decline in the obese group. The lack of a significant difference in urea, creatinine, and eGFR may reflect the relatively young age of the subjects and the short duration of obesity, which may not yet have led to advanced kidney damage. Age is an unavoidable risk factor for degenerative diseases, as organ function, including kidney function, naturally declines with age. Individuals aged

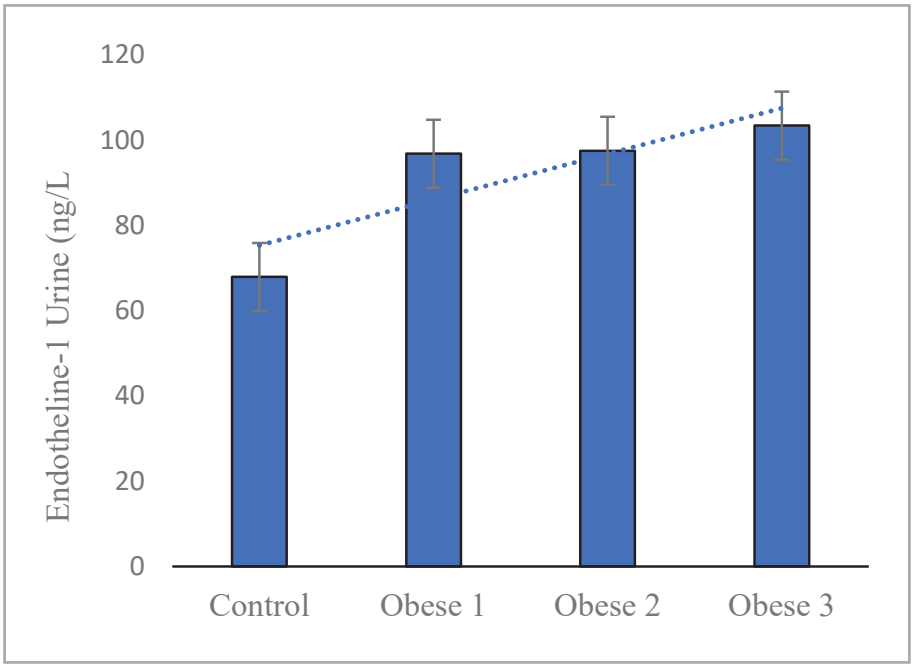

Figure 1: Urinary endothelin-1 levels in obese and non-obese young adults.

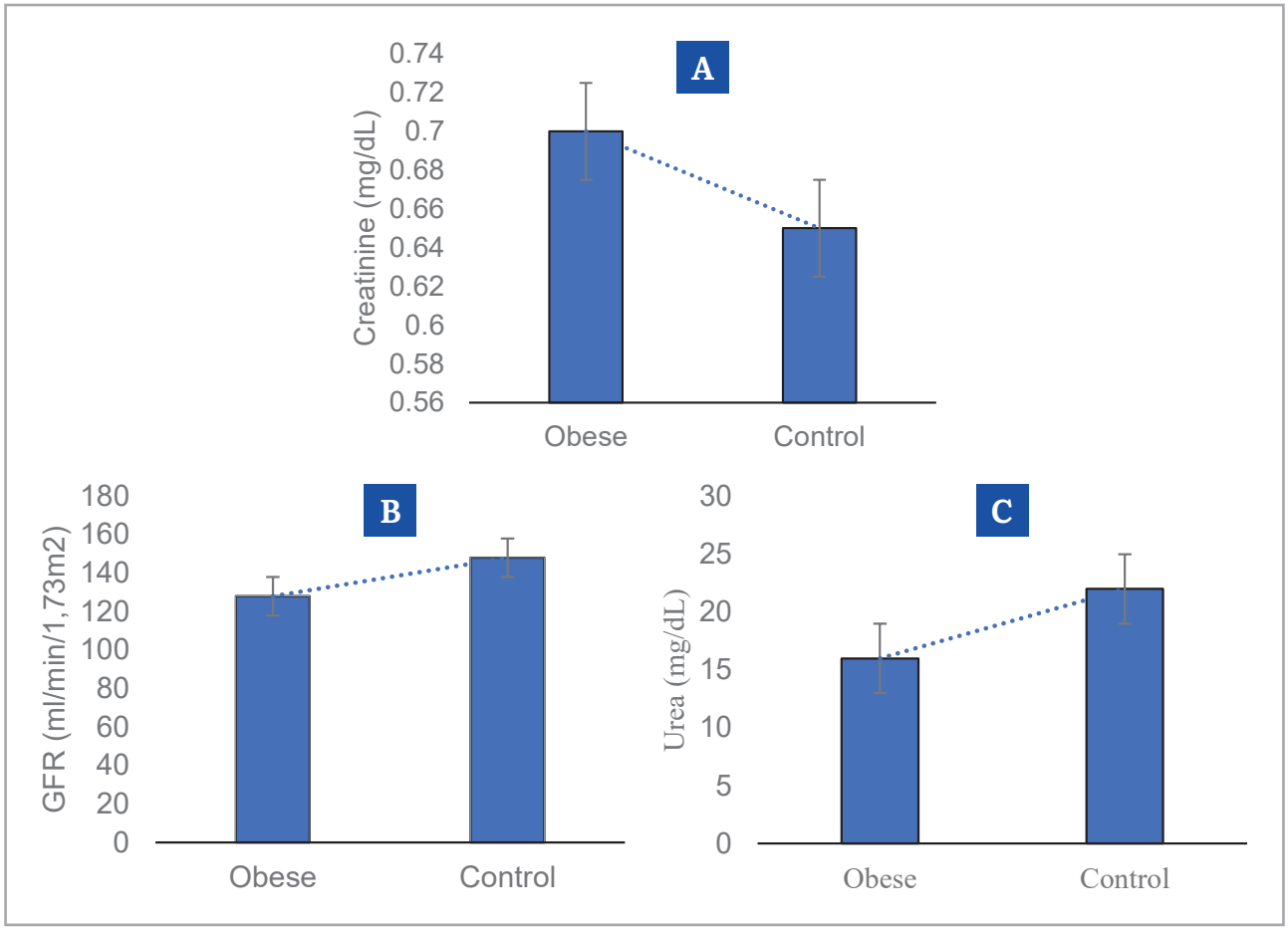

Figure 2: GFR, creatinine and urea levels in obese and non-obese young adults.

61–86 years have a 4.51 times higher risk of developing chronic kidney disease (CKD) compared to those aged 18–30 years [13, 14]. Long-term obesity can lead to kidney disorders such as glomerular hyperfiltration, sodium retention, Bowman's space enlargement, increased glomerular cell proliferation, mesangial matrix expansion, inflammatory cell infiltration, and tubulointerstitial lesions. These early changes can progress to more severe conditions, such as focal segmental glomerulosclerosis, proteinuria, and extensive tubulointerstitial lesions [15]. In a hypertension-independent rat model, ET-1 induction is involved in kidney pathology, leading to renal fibrosis and fatal kidney disease [16]. Although no significant differences were found in kidney function markers, the observed trends suggest that long-term obesity may lead to chronic kidney disease (CKD). The mechanisms involved include glomerular hyperfiltration, mesangial matrix expansion, and tubulointerstitial damage, which can progress to focal segmental glomerulosclerosis and proteinuria.

Spearman's correlation analysis showed no significant association between urinary ET-1 levels and kidney function markers ($p>0.05$). Nonetheless, the weak negative correlations with urea and creatinine, along with the weak positive correlation with eGFR, suggest a more intricate interaction. Studies conducted on patients with Autosomal Dominant Polycystic Kidney Disease (ADPKD) have shown a notable negative correlation between ET-1 and eGFR, highlighting the intricate role of ET-1 in kidney pathology [17].

Future research should investigate the long-term impact of obesity on kidney function in larger and

Table 4: Correlation of urinary ET-1 levels, urea, creatinine, and GFR in obese and non-obese subjects.

Variable (n=58)		ET-1 urine (ng/L)
Urea (mg/dL)	r	-0.200
	p	0.132
Creatinine (mg/dL)	r	-0.167
	p	0.211
GFR (ml/min/1.73 m²)	r	0.168
	p	0.206

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more diverse populations to gain a deeper understanding of ET-1's role in kidney pathophysiology.

Conclusion

The findings of this study indicate that urinary ET-1 concentrations are significantly elevated in young adults (19–25 years) with obesity compared to those without obesity. However, kidney function, as measured by urea, creatinine, and GFR, was not consistently worse in the obese group. No significant relationship was found between ET-1 levels and kidney function.

Conflict of interest

The authors declare no conflict of interest.

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