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Analysis of the Relationship Between Soluble Cd40 Ligand and Homeostasis Model Assessment of Insulin Resistance in Obese Non-Diabetic Subjects

Análisis de la relación entre el ligando soluble de Cd40 y la evaluación del modelo de homeostasis de la resistencia a la insulina en sujetos obesos no diabéticos

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SUMMARY

Obesity is a complex multifactorial disease characterized by the accumulation of body fat, leading to adverse effects on one's well-being. Obesity has a strong association with insulin resistance. Insulin resistance can lead to a range of health issues, including type II diabetes mellitus, cardiovascular disease, and other metabolic syndrome diseases. A method that can be employed to evaluate insulin resistance is the measurement of HOMA-IR. sCD40L is the soluble form of the CD40L molecule released on the cell surface through proteolysis. There is evidence that the sCD40L level is a strong predictor of cardiovascular risk, and enhanced levels of CD40L were reported in patients with hypercholesterolemia, diabetes mellitus, and acute coronary syndromes. The objective of the present study is to determine the correlation between sCD40L and HOMA-IR in non-DM subjects. The method was observational analytic with a cross-sectional design involving 70 non-diabetic subjects, 36 obese subjects, and 34 non-obese subjects, comprising 30 males and 40 females. The findings exhibited a significant difference in the value of HOMA-IR between obese and non-obese subjects (p=0.001). No significant difference, however, was observed in sCD40L levels between the two groups (0.117). In addition, no correlation was observed between HOMA-IR and sCD40L levels in non-diabetes mellitus subjects (r= 0.081; p=0.507).

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It was concluded that no significant correlation was found between HOMA-IR values and sCD40L levels in non-diabetes mellitus subjects.

Keywords: *Obesity, HOMA-IR, sCD40L*

RESUMEN

La obesidad es una enfermedad multifactorial compleja caracterizada por la acumulación de grasa corporal, lo que produce efectos adversos sobre el bienestar. La obesidad tiene una fuerte asociación con la resistencia a la insulina. La resistencia a la insulina puede provocar una variedad de problemas de salud que incluyen diabetes mellitus tipo II, enfermedades cardiovasculares y otras enfermedades del síndrome metabólico. Un método que se puede emplear para evaluar la resistencia a la insulina es la medición de HOMA-IR. sCD40L es la forma soluble de la molécula CD40L que se ha liberado en la superficie celular mediante proteólisis. Existe evidencia que el nivel de sCD40L es un fuerte predictor de riesgo cardiovascular, y se ha demostrado niveles elevados de CD40L en pacientes con hipercolesterolemia, diabetes mellitus y síndrome coronario agudo. El objetivo del presente estudio es determinar la correlación entre sCD40L y HOMA-IR en sujetos no DM. El método utilizado fue analítico observacional con un diseño transversal que involucró a 70 sujetos no diabéticos, 36 sujetos obesos y 34 sujetos no obesos, comprendiendo 30 hombres y 40 mujeres. Los hallazgos mostraron una diferencia significativa en el valor de HOMA-IR entre sujetos obesos y no obesos (p=0,001). Sin embargo, no se observaron diferencias significativas en los niveles de sCD40L entre los dos grupos (0,117). Además, no se observó correlación entre los niveles de HOMA-IR y sCD40L en sujetos sin diabetes mellitus (r= 0,081 p=0,507). Se concluyó que no se existe correlación significativa entre los valores de HOMA-IR y los niveles de sCD40L en sujetos sin diabetes mellitus.

Palabras clave: *Obesidad, HOMA-IR, sCD40L*

INTRODUCTION

Indonesia is currently grappling with diverse health challenges, one of which is obesity. Basic Health Research (Riskesdas) data in 2018 revealed that the prevalence of obesity in toddlers was 3.8 %, while individuals 18 years and older experienced a prevalence of 21.8 %. The target obesity rate for 2024 continues to be unchanged at 21.8 % (1). Obesity is a complex multifactorial disease characterized by the accumulation of body fat that negatively impacts health (2).

Obesity is strongly linked to insulin resistance, dyslipidemia, hypertension, and type II diabetes mellitus (3). It is always related to the accumulation of fat or adipose tissue. The accumulation of fat tissue in the central part of the body triggers increased production of excessive free fatty acids, contributing to their heightened transfer to the liver through portal vein drainage. Excessive free fatty acids in the liver trigger visceral fat to release inflammatory cytokines through the portal vein. Consequently, this initiates insulin resistance within the liver and an uncontrollable surge in glucose production. Insulin resistance can lead to various health issues, including type II diabetes mellitus, cardiovascular disease, and other metabolic syndrome diseases (4).

In insulin resistance, low-grade systemic inflammation sets off dysfunction in the hemostatic system through various mechanisms, such as increased activation of platelets. Chronic hyperglycemia and hyperinsulinemia can contribute to an elevated CD40 ligand (CD40L) expression in circulating platelets (5). In cases of insulin resistance, low-grade systemic inflammation also triggers the activation of various other cells such as B cells, T cells, as well as monocytes and endothelial cells, which will then trigger the release of sCD40L into the circulation; this process may exacerbate insulin resistance and contribute to the onset of type II diabetes and other health problems (6).

Studies on the cellular distribution of CD40L indicate that >95 % of the circulating CD40L exists in platelets. Platelets express CD40L on their surface upon stimulation; CD40L is then cleaved and circulates as soluble CD40L (sCD40L). When expressed on the surface of platelets and exposed to CD40-bearing vascular cells, platelet-associated CD40L triggers the expression of various proinflammatory mediators, such as intercellular adhesion molecule-1, vascular cell adhesion molecule-1, IL-1, IL-6, IL-8, IL-12, TNF-α, interferon-γ, and MCP-1. During recent years, the CD40-CD40L system has been implicated in the pathophysiology of chronic inflammatory diseases, including atherosclerosis. There is evidence that the

sCD40L level strongly predicts cardiovascular risk. Enhanced levels of CD40L were reported in patients with hypercholesterolemia, diabetes mellitus, and acute coronary syndromes. Soluble CD40 ligand is a form of the CD40L molecule released on the cell surface through proteolysis. This process involves enzymatic cutting of the CD40L molecule bound to the cell's surface, enabling it to circulate in body fluids in a soluble form (5-8). Insulin resistance is one of the contributing factors to the increased level of plasma sCD40L in obese patients (8).

The homeostasis model assessment (HOMA) is a mathematical model for determining insulin resistance from fasting glucose and insulin concentrations that has been validated against the euglycemic–hyperinsulinemic clamp. Several methods can be employed to evaluate the level of insulin resistance. In individuals diagnosed with pre-diabetes, heightened insulin secretion compensates for the rise in HOMA-IR by pancreatic beta cells (9).

Patients with insulin resistance exhibited a notably higher level of soluble CD40 Ligand (sCD40L) when compared to those without insulin resistance (8). Another study stated that compared to the non-impaired glucose tolerance (IGT)-group, the average levels of sCD40L were significantly higher in the Impaired glucose tolerance (IGT) group (10). The findings of this study differ from those conducted by Linna et al. (2016), which revealed that no correlation was found between sCD40L and body mass index. Furthermore, the current study exhibits a negative correlation with HOMA-IR in female subjects with polycystic ovarian syndrome (PCOS), regardless of their obesity status (11). Meanwhile, a study by Sarray and Almawi also found lower sCD40L values in obese children with insulin resistance compared to the control group (12).

High HOMA-IR values arise from numerous factors linked to metabolic syndrome, such as obesity, hypertension, and dyslipidemia. The insulin resistance commonly observed in obese individuals is connected to heightened HOMA-IR levels likely to correlate positively with sCD40L values (9). Thus, we aimed to investigate and analyze the possible relationship between sCD40L and HOMA-IR in obese non-diabetes mellitus subjects.

MATERIALS AND METHODS

Design and Population of the Research

The current study employed a cross-sectional study design. All adults who willingly volunteered for the research were determined as the research population. A total of 70 non-diabetic (DM) individuals were included, comprising 30 males and 40 females. The inclusion criteria encompassed non-DM males and females aged 18-40 who consented to participate in the study. The exclusion criteria were pregnant women, having a history of DM, having an infection, suffering from malignancy, taking anti-platelet aggregation drugs, taking corticosteroid drugs, icteric, lipemic, or hemolyzed serum samples. The classification of obesity based on body mass index (BMI) according to Asian Pacific criteria includes Underweight (<18.5 kg/m^2), Normal (18.5 - 22.9 kg/m^2), Overweight (≥23.0 kg/m^2), At Risk (23.0 - 24.9 kg/m^2), Obesity I (25.0 - 29.9 kg/m^2), Obesity II (≥30.0 kg/m^2) (13). The study occurred at the Clinical Pathology Laboratory of Hasanuddin University Hospital, Makassar, and Hasanuddin University Medical Research Center (HUM-RC) Laboratory. It was carried out after ethical approval from the Health Research Ethics Commission (KEPK) of Hasanuddin University Hospital (RSPTN UH) with ethical number 858/UN4.6.4.5.31/PP36/2023.

Laboratory Procedure

This study involved documenting the participants' identities, and their height (m) and weight (kg) were assessed. Body Mass Index (BMI) was measured using the weight/(height) formula, with obesity defined as BMI ≥ 25 kg/m^2. Blood samples were collected in the morning after an 8-12 hours fast using a red cap tube without anticoagulant. The samples were left in a vacuum tube for 15-30 minutes to induce clotting. Following this, centrifugation at 3000 rpm for 10-15 minutes was carried out to separate the serum from the blood cell components. The serum obtained was stored at -20°C. Fasting glucose was tested using ABX Pentra 400 with enzymatic method, while insulin levels were measured using Cobas® e411 with Electrochemiluminescence

Immunoassay (ECLIA) method. The analysis of sCD40L was conducted through the Enzyme-linked Immunosorbent Assay (ELISA) method, employing the MyBioSource insert kit. A microplate reader (Type 357, Thermo Fisher Scientific, Shanghai, China) was utilized to assess the absorbance of the ELISA test result.

Data Analysis

The Statistical Package for the Social Sciences (SPSS) version 22 was employed to analyze the data. Meanwhile, the Kolmogorov-Smirnov normality test was utilized to assess the data distribution of sCD40L and HOMA-IR. For non-diabetic subjects, the relationship between sCD40L and HOMA-IR was examined using the Pearson correlation test for normally distributed data and the Spearman correlation test for non-normally distributed data. Results were considered statistically significant if the p-value <0.05.

RESULTS

A total of 70 research subjects were involved, consisting of 30 males and 40 females. The study results show that 36 individuals had obesity non-DM and 34 individuals had non-obesity non-DM, as shown in Table 1.

Table 1. Frequency Distribution of Gender and Group Variables

Characteristics	Category	Nondiabetes Mellitus	
		N	%
Gender	Male	30	42.9
	Female	40	57.1
Group	Obese	36	51.4
	Non-obese	34	48.6

Source: Primary Data

As Table 2 shows, the study participants' ages ranged from 20 to 40, with a mean age of 31.34 years, a mean body weight of 64.33 kg, a mean height of 159.04 cm, a BMI of 25.31 kg/m^2, a mean waist circumference of 88.24 cm, mean HOMA-IR score of 2.58, and mean sCD40L level of 1.83 ng/mL.

Normality Test

Based on the Kolmogorov-Smirnov test conducted on all subjects, the HOMA-IR value in non-DM subjects exhibited a p-value of $0.002 < \alpha$ (0.05), indicating that the data is not normally distributed. Meanwhile, the p-value of sCD40L levels in all non-DM subjects was $0.200 > \alpha$ (0.05), which suggests that the data is normally distributed. For obese non-diabetes mellitus (DM) subjects, the obtained p-value for HOMA-IR levels was $0.124 > \alpha$ (0.05), and the p-value of sCD40L levels was $0.103 > \alpha$ (0.05), indicating normal data distribution. Conversely, in non-obese subjects without DM, the p-value of HOMA-IR value was $0.020 < \alpha$ (0.05), suggesting non-normal data distribution. In contrast, the p-value of the sCD40L level was $0.544 > \alpha$ (0.05), indicating a normal data distribution (Table 3).

Comparison Test

Based on the normality test conducted, the HOMA-IR value for non-obese subjects is the only dataset that does not exhibit a normal distribution. Therefore, to assess differences in

Table 2. Descriptive Analysis of Age Variable, sCD40L Levels, and HOMA-IR Values

Characteristics	n	Mean±SD	Median	Min-Max
Age (Year)	70	31.34±3.80	31.00	20-40
Weight (kg)	70	64.33±14.45	63.30	41.2-130.1
Height (cm)	70	159.04±7.76	157.00	145-178
BMI (kg/m^2)	70	25.31±4.51	25.00	15.3-45
Waist Circumference (cm)	70	88.24±11.18	89.00	61-133
HOMA-IR	70	2.58±1.26	2.39	0.53-5.87
sCD40L (ng/mL)	70	1.83±0.40	1.82	0.38-2.74

Source: Primary Data
Notes : BMT = Body Mass Index, HOMA-IR = Homeostasis Model Assessment Insulin Resistance, sCD40L = Soluble CD40 Ligand

Table 3. Normality Test of sCD40L Level and HOMA-IR Value of Research Subjects

	Normality Test Statistic	N	P	Distribution
HOMA-IR (Overall)	0.132	70	0.002*	Not Normal
sCD40L (Overall)	0.070	70	0.200*	Normal
HOMA-IR (OB)	0.952	36	0.124**	Normal
sCD40L (OB)	0.950	36	0.103**	Normal
HOMA-IR (Non-OB)	0.923	34	0.020**	Not Normal
sCD40L (Non OB)	0.973	34	0.544**	Normal

Source: Primary Data
Notes: *p = Kolmogorov-Smirnov test, **p = Shapiro Wilk test, OB = Obese, Non-OB = Non-obese.

HOMA-IR value between obese and non-obese subjects, the Mann-Whitney test was employed (Table 4). Then, a p-value of 0.001 was obtained from the test conducted. Given that $p<\alpha$ (0.05), the conclusion can be drawn that a significant difference was found in the HOMA-IR values between obese and non-obese subjects within the non-DM group.

Table 4. Comparison Test between HOMA-IR Value and sCD40L Levels in Subjects with Obesity and Non-Obesity

Group	N	Non diabetes melitus			P
		Mean±SD	Median	Min-Max	
HOMA-IR (OB)	36	3.09±1.33	3.11	1.24-5.87	0.001*
HOMA-IR (Non-OB)	34	2.04±0.93	1.74	0.53-3.84	
sCD40L (ng/mL) (OB)	36	1.90±0.47	1.92	0.38-2.74	0.117**
sCD40L (ng/mL) (Non OB)	34	1.75±0.31	1.72	0.93-2.43	

Source: Primary Data. Notes : *p = Mann-Whitney Test, **p= T Independent Test

As seen in Table 4, the statistical tests comparing the sCD40L level between obese and non-obese subjects in the non-DM group using the independent T-test revealed a p-value of 0.117, inferring that no significant difference was observed in the value of sCD40L between individuals with obesity and non-obesity in the non-DM group.

Correlation Test

The Spearman correlation test between HOMA-IR values and sCD40L levels in non-DM subjects obtained a p-value of 0.507. Since 0.507>α (0.05), it was concluded that no significant correlation between HOMA-IR values and sCD40L levels was found in non-DM subjects (Table 5).

Table 5. Correlation Test of HOMA-IR Values and sCD40L Levels in Overall Non-Diabetes Mellitus Subjects

Variable	sCD40L (ng/mL)	
HOMA-IR Value	r =	0.081
	p =	0.507
	n =	70

Source: Primary Data

Source: Primary Data

Data exploration was carried out using scatterplot graphs to determine the linear relationship pattern between HOMA-IR value and sCD40L levels in non-DM subjects.

The scatterplot in Figure 1 revealed that the data distribution does not form a linear relationship pattern between HOMA-IR values and sCD40L levels. This indicates no correlation or relationship between the variables of HOMA-IR values and sCD40L levels in non-DM subjects.

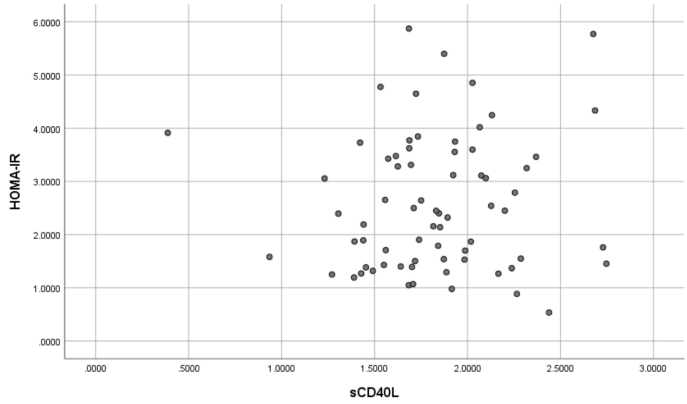

Figure 1. Scatterplot of HOMA-IR value data and sCD40L levels in Overall Non-Diabetes Mellitus Subjects.

DISCUSSION

This study took place between August and November 2023. The results revealed a highly significant difference in the value of HOMA-IR between obese and non-obese subjects in the non-DM group. HOMA-IR is an index used to measure insulin resistance. A high HOMA-IR value indicates the presence of insulin resistance.

This aligns with the research conducted by Abiageli Uzoamaka Agbogu-Ike et al. (14), suggesting a significant increase in HOMA-IR among the obese group compared to the normal control group.

The increase in HOMA-IR values among obese subjects compared to non-obese subjects is attributed to dysfunction in the adipose tissue associated with obesity. Adipocytes can produce several hormones and chemicals called adipocytokines that can affect insulin sensitivity. Increased fat levels in fat cells can cause changes in adipocytokine secretion and exacerbate insulin resistance, increasing HOMA-IR values (15). In addition, decreased mitochondrial function and increased reactive oxygen species (ROS) are also factors that play a part in causing insulin resistance (16).

WHO has set a cut-off of >25 kg/m^2 for the category of obesity in Asian adults. With the increasing prevalence of obesity in Asia, the calculation of body mass index (BMI) has become very important as one of the predictors of obesity. However, there is controversy when applying international criteria for obesity in Asian populations, and there have been attempts to reinterpret BMI cut-offs for Asia-Pacific populations. Furthermore, expert consultants from (WHO) have established Body Mass Index (BMI) cut-offs for obesity and Overweight classifications that are now used worldwide in Asia-Pacific countries; the agreed cut-off for inclusion in the Overweight category is 23.0 kg/m^2. This study uses cut-off obesity based on BMI according to Asian Pacific criteria, namely BMI 25 kg/m^2. This aligns with Weir and Jan's research (2024); Asian and South Asian populations use Overweight criteria, namely BMI between 23 - 24.9 kg/m^2 and Obesity with a BMI of 25 kg/m^2 (17). In addition, Zhao et al. research (2024) on the Overweight and obesity of Asian populations using BMI criteria proposed by WHO for Asian populations: normal weight (BMI < 23 kg/m^2), overweight (BMI: 23-24.9 kg/m^2), and obesity (BMI ≥ 25 kg/m^2) (18). Research by Singh and Chattopadhyay (2024) in the Indian population also used WHO criteria based on BMI for the Asian population, namely (underweight (< 18.5 kg/m^2), normal weight (18.5-22.9 kg/m^2), overweight (23-24.9 kg/m^2), and obesity (≥ 25 kg/m^2) (19).

Our results indicate no statistical differences between the value of sCD40L in obese and non-obese subjects in the non-DM group. This is because most respondents in the obese group are still within a mild obesity condition (average BMI 25.31 kg/m^2), which may not have led to a significant systemic inflammation for high sCD40L in circulation. According to the findings from the study conducted by Guldiken et al. (2016), patients with severe obesity (BMI≥35 kg/m^2) exhibited significantly higher sCD40L values compared to both obese patients (BMI 30-34.9 kg/m^2) and non-obese subjects (BMI<25 kg/m^2) (20). Unek et al. (2010) also found that obese subjects (BMI≥30 kg/m^2) had higher sCD40L values than overweight subjects (BMI 25 to 29.9 kg/m^2) (21).

The correlation between BMI and body fat in Europeans is not appropriate when using the above cut-offs. The prevalence of patients who are metabolically obese but with normal body weight in Asian-Pacific countries is almost twice the Asia-Pacific countries is almost twice that found in the U.S. population due to differences in muscle mass.

Our data indicate no significant correlation between HOMA-IR values and sCD40L levels in non-DM subjects. Various variables, including smoking may influence the results. The release of nicotine from smoking activities can contribute to hemodynamic effects and various cardiovascular occurrences (22). Cardiovascular diseases result from the rupture of atherosclerotic plaques and thrombus formation. Dyslipidemia, endothelial dysfunction, as well as platelet hyperreactivity can lead to atherosclerosis and an increased risk of thrombotic vascular events. Platelet hyperactivation is associated with the release of sCD40L, influencing its level in circulation (23).

Extended periods of obesity will worsen insulin resistance. Persistent low-grade systemic inflammation impedes insulin action within the insulin signaling pathway, disrupts glucose homeostasis, and leads to systemic dysregulation. Generally, sustained obesity and excessive nutrition over an extended period result in insulin resistance and chronic low-grade systemic inflammation through lipotoxicity (24).

Hypertension is also related to the levels of sCD40L. Soluble CD40 ligand (sCD40L)

contributes to the pathogenesis of vascular damage linked to risk factors and is closely associated with inflammation, thrombosis, and angiogenesis. The newly available data point to the vasoactive peptide angiotensin II as a promoter and enhancer of inflammation activation induced by CD40/CD40L ligation in human blood vessel cells (25). Angiotensin II, a vasoactive peptide associated with blood vessel constriction, increases blood pressure as its level rises. Consequently, sCD40L levels will increase in hypertensive patients (26).

The consumption of antihypertensive medications affects blood vessels, reducing blood pressure (exhibiting a pleiotropic effect) and influencing sCD40L levels (26). This research is consistent with the findings reported by Han et al. (2010), stating that either combination therapy or losartan alone significantly decreases the level of plasma sCD40L (27). The consumption of antihypertensive will then affect or even reduce the levels of sCD40L circulating in the body.

The involvement of physical activity, such as exercise, can also impact this study. With or without weight loss, physical activity can reduce the risk of cardiometabolic disorders, partially improve insulin sensitivity, and lower blood pressure (28). This corresponds with the study of Hilberg et al. (2021), which asserts that moderate-intensity exercise inhibits platelets, whereas vigorous exercise promotes the aggregation and activation of platelets (29). Other studies suggest that aerobic exercise in adults with overweight or obesity and cardiometabolic disorders successfully decreases postprandial glucose and insulin levels (30).

The effect of platelet count can also potentially influence the level of sCD40L. Abnormal platelet counts can disrupt the balance of sCD40L in the body. An increase in platelet count may be associated with an increased production or release of sCD40L (31). Calabro et al. (2009) suggests that the adipose hormone resistin produced by adipose tissue in obesity can enhance the expression of sCD40L and tissue factors in human coronary endothelial cells (32). CD40L and platelets have a complex relationship in the immune and inflammatory systems. An increase in platelet count (thrombocytosis) can potentially elevate the concentration of sCD40L in the blood (33).

CONCLUSION

According to the findings in this study, it can be inferred that there is no significant correlation between HOMA-IR and sCD40L levels in non-diabetic subjects.

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Conflict of Interest

All authors affirm that there are no conflicts of interest in this study.

REFERENCES

1. Ministry of Health. Indonesia Health Profile of the Year. Jakarta: Ministry of Health of the Republic of Indonesia. 2019.
2. Lin XL. 2021. Obesity: Epidemiology, Pathophysiology, and Therapeutics. (A. U. Claire Joanne Stocker, Ed.) Department of Endocrinology, Sir Run Run Shaw Hospital, School of Medicine, Zhejiang University, Hangzhou, China. Endocrinol. 2021;12:706978:1-4.
3. Bereket A, Zeynep A. Current status of childhood obesity and its associated morbidities in Turkey. J Clin Res Pediatr Endocrinol. 2012;4(1):1-7.
4. Paleva R. Mekanisme resistensi insulin terkait obesitas. J Ilmiah Kesehatan Sandi Husada. 2019; 10(2):354-358 .
5. Santilli FSP. The role of platelets in diabetes. Department of medicine and aging and haemostasis and thrombosis unit. Amsterdam, Netherlands: Elsevier; 2019:469-503.
6. Henn V, Slupsky JR, Gräfe M, Anagnostopoulos I, Förster R, Müller-Berghaus G, et al. CD40 ligand on activated platelets triggers an inflammatory reaction of endothelial cells. Nature. 1998;391(6667):591-594.
7. Liang Y, Chengrui Z, Sun Y, Zhiliang L, Wang L, Yina L, et al. Persistently higher serum sCD40L levels are associated with outcomes in septic patients. Department of Critical Care Medicine, The First Hospital of China Medical University, Province, China. BMC Anesthesiology. 2021;21:26
8. Gateva A, Yavor A, Tsakova A, Kamenov Z. Soluble CD40L is associated with insulin resistance but not with glucose tolerance in obese nondiabetic patients. Arch Physiol Biochem. 2016;122(3):161-165.

9. Decroli E, Rudy A, Kam A, Puspita U. Correlation between Insulin Resistance with Soluble CD40 Ligand and Plasminogen Activator Inhibitor-1 Plasma in Pre-diabetic Patients. Macedonian J Med Scienc. 2022;10(B):351-356.
10. WHO. Body mass index (BMI) is associated with morbidity and mortality from various diseases & Asian cut-off value. 2012;2:3–4.
11. Linna H, Suija K, Rajala U, Herzig KH, Karhu T, Jokelainen J. The association between impaired glucose tolerance and soluble CD40 ligand: A 15-year prospective cohort study. Aging Clin Exp Res. 2016;28(6):1243-1239.
12. Sarray S, Almawi WY. Levels of CD40L and other inflammatory biomarkers in obese and non-obese women with polycystic ovary syndrome. Am J Reprod Immunol. 2016;76(4):285-291.
13. Luciardi MC, Carizzo T, Diaz EI, Aleman MN, Bazan MC, Abregu A. Proinflammatory state in obese children. Rev Chil Pediatr. 2018;89(3):346-351.
14. Abiageli Uzoamaka Agbogu-Ike, Ogoina D, Geoffrey C. Leptin Concentrations in Non-Obese and Obese Non-Diabetes Nigerian-Africans. Diabetes Metab Syndr Obes. 2021;14:4889-4902.
15. Barazzoni R, Gortan Cappellari G, Ragni M, Nisoli E. Insulin resistance in obesity: an overview of fundamental alterations. Eat Weight Disord. 2018;23(2):149-157.
16. Sangwung P, Petersen KF, Shulman GI, Knowles JW. Mitochondrial dysfunction, insulin resistance, and potential genetic implications. Endocrinology. 2020;161:bqaa017.
17. Weir CB, Jan A. BMI Classification Percentile And Cut Off Points. In: StatPearls Publishing. 2024;26(6):1-2.
18. Zhao R, Yang X, Cui, Wang L, Ye Y. Overweight and obesity determined by body mass index criteria for Asian populations adversely affect assisted reproductive outcomes among Chinese women with polycystic ovary syndrome. Int J Obes. 2024;48:394-402.
19. Singh A, Chattopadhyay A. Age-appropriate BMI cut-offs for malnutrition among older adults in India. Sci Rep. 2024;15072:1-2.
20. Guldiken S, Demir M, Arikan E, Turgut B, Azcan S, Gerenli M, et al. The levels of circulating markers of atherosclerosis and inflammation in subjects with different degrees of body mass index: Soluble CD40 ligand and high-sensitivity C-reactive protein. Thromb Res. 2007;119(1):79-84.
21. Unek ITM, Firat Bayraktar M, Dilek Solmaz M, Hulya Ellidokuz M, Ali Riza Sisman M, Yuksel F, et al. The Levels of Soluble CD40 Ligand and C-Reactive Protein in Normal Weight, Overweight and Obese People. Clin Med Research. 2010;8(2):89-95.
22. Gómez Cerezo JF, López Paz JE, Fernández Pardo J. Update on new forms of tobacco use. Clin Investig Arterioscler. 2022;34(6):330-338.
23. Fuentes E, Palomo I. Mechanism of antiplatelet action of hypolipidemic, antidiabetic and antihypertensive drugs by PPAR activation PPAR agonists: New antiplatelet agents. Vascular Pharmacology. 2014; 62(3):162-166.
24. Bulbul Ahmad RS. Adipose tissue and insulin resistance in obese. Biomed Pharmacoth. 2021;137:111315.
25. Senchenkova EY, Russell J, Vital SA, Yildirim A, Orr AW, Granger DN, et al. A critical role for both CD40 and VLA5 in angiotensin II-mediated thrombosis and inflammation. Faseb J. 2018;32(6):3448-3456.
26. Ferroni P, Fiorella G. Soluble CD40L and its role in essential Hypertension: Diagnostic and Therapeutic implications. Cardiovasc Hematol Disord Drug Targets. 2008;8(3):194-202.
27. Han SH, Koh KK, Quon MJ, Lee Y, Shin EK. The Effects of Simvastatin, Losartan, and Combined Therapy on Soluble CD40 Ligand in Hypercholesterolemic, Hypertensive Patients. Atherosclerosis. 2007;190(1):205-211.
28. Barale C, Melchionda E, Tempesta G, Morotti A, Russo I. Impact of Physical Exercise on Platelets: Focus on Its Effects in Metabolic Chronic Diseases. Antioxidants. 2023;12(8):1609.
29. Hilberg T, Ransmann P, Hagedorn T. Sport and Venous Thromboembolism—Site, Accompanying Features, Symptoms, and Diagnosis. Dtsch Arztebl Int. 2021;118:181-187.
30. Dallak M, Bin-Jaliah I, Sakr HF, Al-Ani B, Haidara MA. Swim exercise inhibits hemostatic abnormalities in a rat model of obesity and insulin resistance. Arch Physiol Biochem. 2019;125:79-84.
31. Antoniades C, Bakogiannis C, Tousoulis D, Antonopoulos AS, Stefanadis C. The CD40/CD40 ligand system: linking inflammation with atherothrombosis. J Am Coll Cardiol. 2009;54(8):669-677.
32. Calabro P, Cirillo P, Limongelli G, Maddalono V, Riegler R, Palmieri R, et al. Tissue Factor is induced by resistin in human coronary artery endothelial cells by the NF-kappaB-dependent Pathway. J Vasc Res. 2009;46(5):423-431.
33. Daub S, Lutgens E, Münzel T, Daiber A. CD40/CD40L and Related Signaling Pathways in Cardiovascular Health and Disease: The Pros and Cons for Cardioprotection. Int J Mol Sci. 2020;21(22):8533.