ORIGINAL ARTICLE

Aromatase Activity and Its Association with Coronary Artery Disease in Males

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ABSTRACT

Objective: To determine the association among aromatase activity, testosterone-to-estradiol (T/E2) ratio, body mass index (BMI), and coronary artery disease in males.

Study Design: Comparative cross- sectional study.

Place and Duration of Study: Department of Chemical Pathology, from Feb2023 –Jan2024.

Materials and Methods: This cross-sectional study used the T/E2 ratio as a marker for aromatase activity and assessed the levels of plasma testosterone, estradiol, and T/E2 in 300 males. In order to evaluate the evolution of CAD, T/E2 was compared across serum quartiles and cardiac calcium score groups using ANOVA. It also showed correlations with atherosclerotic plaque, CRP, cholesterol, and BMI.

Results T/E2 ratio and plaque calcification score were found to be negatively correlated in atherosclerotic plaques. These effects were observed to be greater in men with elevated body mass indexes (BMI). BMI, CRP, and Calcium Score show statistically significant differences across the three T/E2 ratio groups

Conclusion: Males with low T/E2 ratio had higher levels of calcified plaque, systemic inflammation and evident atherosclerosis. These effects were strongest in men with higher BMI, which increased risk of future major acute coronary event.

Key Words: *BMI; Cardiac Calcium Score; Coronary Artery Disease; CT Angiography; Testosterone to Estradiol Ratio; Myocardial Infarction.*

Introduction

The development of atherosclerotic plaques in the arterial lumen is a typical sign of coronary artery disease. This results in decreased blood flow, Consequently, this hinders the myocardium's capacity to take in oxygen.[1] Superimposed atherothrombosis and subsequent artery blockage can result from plaque erosion or rupture, this may result in cardiovascular (CV) events such as myocardial infarction (MI), stroke, limb ischemia, and death from CV.[2] Age is the most significant risk element for coronary heart disease development and mortality after coronary atherosclerosis appears.[3] CAD affects the lives of almost four million people annually in the 49 nations that make up Europe and Northern Asia. An estimated 1.5 million Americans suffer a heart attack or stroke every year.[4] In 2019, CADs accounted for up to 32% of all deaths worldwide, with myocardial infarction and stroke accounting for 85% of these deaths.[5]

There is still uncertainty regarding the connection between testosterone and cardiovascular health. While some research indicates testosterone has a preventive impact, other studies imply it raises the chance of cardiovascular incidents.[6] The enzyme aromatase, which changes testosterone into estrogen, has been discovered to play a major role in the development of CAD.[7] Heart disease (CAD) and poorer cardiovascular outcomes are linked to a low serum testosterone/estradiol (T/E2) ratio, which indicates aromatase enzyme activity in males.[8] Systemic inflammation, bigger plaque size, and an increased risk of cardiovascular events are associated with low testosterone/estradiol (T/E2) ratios, which are regulated by testosterone being converted into estradiol by white fat cells.[9] There are a number of known direct and indirect effects on artery health of steroid hormones related to testosterone and estrogen, including 17β estradiol. An elevated BMI is associated with coronary artery disease because estradiol depends on both aromatase and circulating free testosterone.

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Received: July 23, 2024; Revised: June 26, 2025
Accepted: June 30, 2025

https://doi.org/10.57234/jiimc.june25.2176

Because of the increased white adipose tissue, an elevated BMI raises aromatase activity, which increases the production of estrogen and upsets the hormonal balance.[10]

Although there has been progress in identifying the hormonal factors involved in coronary artery disease (CAD), the aromatase activity, as measured by the testosterone-to-estradiol (T/E2) ratio, is poorly comprehended in men. Earlier reports have shown mixed results on its relationship with systemic inflammation, coronary calcification, and obesity-related risks and there is an urgent need to fill this knowledge gap. The study was conducted to fill this gap and examine the relationship between aromatase activity, which is assessed by T/E2 ratio, and CAD and its associations with clinical markers including BMI and coronary calcification. The results are to elucidate the influence of hormonal imbalances on the cardiovascular pathology and guide the development of better risk assessment and specific treatment.

Materials and Methods

In this comparative cross-sectional study, conducted at Department of Chemical Pathology, Armed Forces Institute of Pathology, Rawalpindi, in collaboration with the Armed Forces Institute of Cardiology, Rawalpindi, from Feb2023 –Jan2024. Blood samples and data was collected after taking informed consent from every patient and the institutional review board's ethical clearance of participating centres - IRB letter no.2773 dated 27.7.24. Following a comprehensive review of the literature, we used the WHO calculator to determine a sample size of 300, maintaining a 5% margin of error, a 95% confidence level, and an 80% test power.[11]

All male patients (40-60 years) presenting to Institute of Cardiology for coronary CT angiography were incorporated into the research. Patients on hormone replacement therapy, diagnosed with testosterone producing tumors, diabetics and patients on insulin therapy, hypertensive patients, patients on lipid lowering drugs, patients with history of angioplasty or previous myocardial infarction, acute and chronic inflammatory conditions were taken out of the study.

Blood samples were drawn at the time of cannulation of patient for CT angiography. Using an automated analyzer and the CHOD-PAP enzymatic colorimetric technique, serum total cholesterol was measured. Serum testosterone and estradiol levels were analyzed by chemiluminescence dedicated reagent method on Advia centaur XPT. CRP was performed on Roche Cobas 600 through Turbidimetric inhibitory immunoassay (TINIA).

SPSS version 26 was used to do the statistical analysis for this investigation. The Shapiro-Wilk test was used to assess the normality of the data prior to analysis. The median and interquartile ranges (IQR) were given for data that was not regularly distributed, whereas the mean and standard deviation (SD) were computed for variables that were normally distributed. The testosterone to estradiol (T/E2) ratio was initially treated as a continuous variable and expressed as median (IQR) due to non-normal distribution. For additional analysis, the T/E2 ratio was also categorized into three groups to assess its relationship with BMI and calcium score. The groups were defined using tertiles as follows: Low T/E2 ratio (<1.41), Normal T/E2 ratio (1.41–1.89), and High T/E2 ratio (>1.89). To compare the testosterone to estradiol (T/E2) ratio, CRP levels, and total cholesterol between two groups based on BMI (High BMI >25 kg/m², and Normal BMI <25 kg/m²), the Mann Whitney U test was applied. The Kruskal Wallis Test was used for comparison of BMI and Calcium Score across T/E2 Ratio Groups. For pairwise comparisons among the three T/E2 ratio groups (Low, Normal, and High), Dunn's Post Hoc Test was employed because of the data's non-parametric character. Spearman The association between the T/E2 ratio and BMI was investigated using correlation analysis. Significant p-Values were those that were less than 0.05.

Results

This study covered 300 patients in total. The patients' median age was 46.00 years, with an 11-year IQR.

The table I presents a comparison of various biochemical and clinical variables between two BMI groups: Normal BMI (<25 kg/m²) and High BMI (>25 kg/m²). The significant differences was found between BMI groups for the T/E2 ratio, CRP levels, and calcium score, with p-values of <0.001 for each, demonstrating substantial statistical significance. In contrast, the p-value for total cholesterol is 0.626 mmol/L showing no significant difference between the two groups. These findings suggest that

individuals with higher BMI have increased aromatase activity, inflammation, and coronary artery calcification.

TABLE I: Comparison of T/E2 Ratio, CRP, Total Cholesterol, and Calcium Score across BMI Groups (n=300)

Variables	BMI Groups		p-Value
	Normal (<25 kg/m²) Median, IQR (n=98)	High (>25(<25 kg/m²) Median, IQR (n=202)	
T/E2 ratio	1.52 (1.98-0.96)	0.54 (0.79-0.34)	<0.001
CRP (mg/L)	9.50 (15.00-5.00)	29.00 (61.00-7.00)	<0.001
T. CHOL (mmol/L)	4.00 (5.00-2.18)	4.20 (6.40-1.90)	0.626
Calcium score	50.00 (100.00-10.00)	110.00 (412.00-60.00)	<0.001

The table-II compares BMI and calcium score across three T/E2 ratio groups: Low (<1.41), Normal (1.41-1.89), and High (>1.89). For BMI, the group with a low T/E2 ratio has a significantly higher median compared to the Normal and High groups, having a p-value of <0.001, suggesting a strong connection between T/E2 ratio and BMI. Similarly, the calcium score is highest in the Low T/E2 ratio group compared to the Normal and High groups, with a p-value of <0.001, showing vital differences in coronary artery calcification among the groups with T/E2 ratios.

Table II: Comparison of BMI and Calcium Score across T/E2 Ratio Groups (n=300)

Variables	T/E2 Ratio Groups			p-Value
	Low (<1.41) (n=236)	Normal (1.41-1.89) (n=25)	High (>1.89) (n=39)	
BMI (kg/m²)	29.00 (32.00-26.00)	23.00 (25.10-21.50)	21.00 (23.00-21.00)	<0.001
Calcium Score	100.00 (410.00-50.00)	50.00 (100.00-10.00)	50.00 (90.0-0.00)	<0.001

T/E2 ratio groups were defined using tertiles: Low (<1.41), Normal (1.41–1.89), High (>1.89).

The Table-III presents a pairwise comparison of BMI and calcium score among the T/E2 ratio groups (n=300). For BMI, no significant difference was found between the High and Normal T/E2 ratio groups (p=0.721). However, significant differences were observed between the High and Low groups (p<0.001) and the Normal and Low groups (p<0.001), indicating a relationship between lower T/E2 ratios and higher BMI. For the calcium score, there was no significant difference between the High and Normal T/E2 ratio groups (p=1.00), but significant differences were found between the High and Low groups (p<0.001) and the Normal and Low groups (p=0.002), suggesting a greater coronary artery calcification in the Low T/E2 ratio group.

TABLE III: Pairwise Comparison of BMI and Calcium Score among T/E2 Ratio Groups (n=300)

Variables	T/E2 Ratio Groups		p-Value
	(I) Ratio group	(J) Ratio group	
BMI	High	Normal	0.721
	High	Low	<0.001
	Normal	Low	<0.001
Calcium Score	High	Normal	1.00
	High	Low	<0.001
	Normal	Low	0.002

The table-IV shows the correlation between T/E2 ratio and BMI, with a correlation coefficient of r = -0.641, indicating a strong negative correlation between these two variables. The p-value of <0.001 suggests that this correlation is statistically significant. This means that as the T/E2 ratio increases, BMI tends to decrease.

Table IV: Correlation of T/E2 Ratio with BMI

	T/E2 Ratio	
	r	p-Value
BMI	-0.641	<0.001

Discussion

The importance of the testosterone-to-estradiol (T/E2) ratio as a possible indicator of coronary artery disease (CAD) risk is highlighted in the current study, especially in people with high body mass indexes (BMIs). Prior studies have demonstrated a substantial correlation between systemic inflammation and vascular calcification and hormonal abnormalities, which are manifested by low T/E2 ratios. A major cause of morbidity and death globally, coronary artery disease (CAD) is influenced by intricate interactions between hormonal, metabolic, and inflammatory pathways. New research identifies the testosterone-to-estradiol (T/E2) ratio as a crucial indicator of hormonal imbalance and a possible predictor of CAD; low T/E2 ratios have been linked to increased vascular calcification and systemic inflammation.[12] Due to the greater buildup of white adipose tissue, elevated BMI raises aromatase activity, which in turn converts testosterone to estrogen. Obesity-related metabolic and cardiovascular risks are associated with this hormonal imbalance, which is characterized by higher levels of estrogen than testosterone.[13]

The results of this research serve as evidence of the complexity of the connection between the

https://doi.org/10.57234/jiimc.june25.2176

testosterone-to-estradiol (T/E2) ratio, BMI, and the coronary artery calcification. Decreased T/E2 ratio was significantly related to higher BMI, more systemic inflammation as shown by CRP levels, and more coronary artery calcification. This implies that the hormonal imbalance that is caused by the high activity of aromatase in those with higher BMI can be a contributing factor to the pathogenesis of coronary artery disease (CAD). Elevated C-reactive protein (CRP) levels indicate systemic inflammation and calcified plaques, which are linked to low T/E2 ratios in males. Higher BMI individuals experienced these effects more strongly, indicating that white adipose tissue's elevated aromatase activity is a key factor in the development of atherosclerosis. Prior research has similarly shown that increased systemic and plaque inflammation is associated with lower T/E2 ratios, highlighting the part aromatase plays in cardiovascular risk (van Koeverden et al., 2019). These results align with the research of Naftolin et al., 2016 states that low T/E2 ratios exacerbate vascular inflammation and atherosclerosis through aromatase-mediated conversion of testosterone to estradiol, and the observed differences in calcium scores and BMI across T/E2 quartiles further support the role of hormonal imbalance in CAD progression. The importance of the T/E2 ratio as a possible biomarker for determining CAD risk is emphasized in the study.

The rate of coronary artery calcification was much more in the low T/E2 ratio group than in the normal and high ratio group, which shows that the severity of vascular calcification is strongly associated with hormonal imbalances. The absence of a strong association between the cholesterol levels and BMI groups supports the hypothesis that the classical markers of lipids might not be sufficient in reflecting the metabolic and inflammatory pathways involved in the development of CAD.[16] The T/E2 ratio shown a substantial connection with CAD markers, in contrast to conventional markers like cholesterol levels, which did not demonstrate any significant changes between BMI groups ($p = 0.626$). This implies that it might be useful as a diagnostic instrument. These findings align with the study of Huang et al., 2019 showing that hormonal abnormalities, rather than traditional lipid profiles, are more important in the development of CAD.

According to the study's findings, a higher BMI is linked to higher T/E2 ratio quartiles, which probably explains the connection between raised T/E2 and higher CRP levels as well as enhanced arterial wall inflammation. Given its association with atherosclerosis and cardiovascular control, this implies that the T/E2 ratio, which represents aromatase activity, may be a valuable indicator for determining a person's personal cardiovascular risk. The results emphasize how important it is to keep researching the metabolic and hormonal processes. More details about the relative contributions of this ratio in men would be beneficial to better understand the possible application of T: E2 ratio as a clinical biomarker (Wang et al., 2019).

Our study compared the T/E2 ratio in CAD patients grouped on basis of cardiac calcium score on CT angiography while a study by Van et al and Huang et al had a significant difference in groups based on T/E2 ratio when compared to endarterectomy patients results of plaque's histological appearance, type of plaque and their 3 year survival rate and future occurrence of MACE Van Koeverden D. et al. (2019) and HTERT expression in patients with worse cardiovascular status respectively (Huang et al., 2019).

Recent studies of Kusters et al., 2024 also confirm the mechanistic connection between increased expression of aromatase in adipose tissue and systemic inflammatory markers in obese men. In their observation, they found a correlation between production of adipose tissue-derived estrogens and elevated levels of C-reactive protein (CRP) and interleukin-6 (IL-6), which are major mediators of vascular inflammation. This is similar to our observation that people with lower T/E2 ratio have higher levels of CRP, highlighting the inflammatory process in case of hormonal imbalance. Inflammation plays a crucial role in CAD, with the T/E2 ratio emerging as a potential measure for aromatase activity, particularly in obese patients. Increased systemic inflammation and vascular calcification are associated with low T/E2 ratios. Our findings add relevance to the idea that a key connection between obesity and CAD may be disrupted sex hormone balance, which is triggered by elevated aromatase activity in the setting of higher BMI. This is in line with the data of the van

Koeverden et al., 2019, which showed that lower T/E 2 ratios correlate with higher systemic and plaque inflammation, higher calcification and twofold risk of major adverse cardiovascular events in overweight men.

Thirumalai et al., 2022 elucidate the simultaneous functions of estrogen and testosterone in cardiovascular health. Although men have historically been at higher risk for CAD due to testosterone, new research indicates that aromatase may be able to reduce some of these risks by converting testosterone to estrogen.

Conclusion

This study emphasize the prospect of the testosterone to estradiol (T/E2) ratio as a measure of coronary artery disease (CAD) risk, especially among males with high body mass index (BMI). The fact that low T/E2 ratio is strongly linked to systemic inflammation, vascular calcification, hormonal imbalance highlights the significance of dealing with metabolic and hormonal aspects of CAD management. This highlights the wider aspect of the hormonal pathways aim of reducing cardiovascular risk. The research needs to be conducted in the future to examine the potential role of T/E2 modulation in therapeutic approaches and its relevance in various populations to improve cardiovascular outcomes.

Future recommendations: Long-term, randomized, double-blind, placebo-controlled studies to assess the effects of aromatase blockers on cardiovascular disease, cardiovascular death, and all-cause death in men with low testosterone estradiol levels could be extremely useful in advancing our understanding of the atherosclerotic process.

Funding sources: None

Acknowledgement: We would like to acknowledge all those who participated directly or indirectly in the study.

REFERENCES

1. Komilovich EB. Coronary Artery Disease. EUROPEAN JOURNAL OF MODERN MEDICINE AND PRACTICE. 2023 Dec 10;3(12):81-7.
2. Korkmaz UT. Cardiovascular diseases and prevention. Surgical Medical Sciences Diagnosis and Treatment. 2021 Jan 20;31.
3. Frąk W, Wojtasińska A, Lisińska W, Młynarska E, Franczyk B, Rysz J. Pathophysiology of cardiovascular diseases: new insights into molecular mechanisms of atherosclerosis, arterial hypertension, and coronary artery disease. Biomedicines. 2022 Aug 10;10(8):1938. https://doi.org/10.3390/biomedicines10081938
4. Martin SS, Aday AW, Allen NB, Almarzooq ZI, Anderson CA, Arora P, Avery CL, Baker-Smith CM, Bansal N, Beaton AZ, Commodore-Mensah Y. 2025 Heart Disease and Stroke Statistics: A Report of US and Global Data From the American Heart Association. Circulation. 2025. https://doi.org/10.1161/CIR.0000000000001303
5. Młynarska E, Czarnik W, Fularski P, Hajdys J, Majchrowicz G, Stabrawa M, Rysz J, Franczyk B. From atherosclerotic plaque to myocardial infarction—The leading cause of coronary artery occlusion. International Journal of Molecular Sciences. 2024 Jul 2;25(13):7295. https://doi.org/10.3390/ijms25137295
6. Kaur H, Werstuck GH. The effect of testosterone on cardiovascular disease and cardiovascular risk factors in men: a review of clinical and preclinical data. CJC open. 2021 Oct 1;3(10):1238-48. https://doi.org/10.1016/j.cjco.2021.05.007
7. Obukohwo OM, Benneth BA, Simon OI, Oghenetega OB, Victor E, Faith FY, Okwute PG, Rume RA, Godswill OO, Kingsley NE. Testosterone: The Male Sex Hormone. InTestosterone-Functions, Uses, Deficiencies, and Substitution 2023 May 11. IntechOpen.
8. Liu H, Dai W, Cui Y, Lyu Y, Li Y. Potential associations of circulating growth differentiation factor-15 with sex hormones in male patients with coronary artery disease. Biomedicine & Pharmacotherapy. 2019 Jun 1;114:108792. https://doi.org/10.1016/j.biopha.2019.108792
9. Kusters CD, Paul KC, Lu AT, Ferruci L, Ritz BR, Binder AM, Horvath S. Higher testosterone and testosterone/estradiol ratio in men are associated with decreased Pheno-/GrimAge and DNA-methylation based PAI1. GeroScience. 2024 Feb;46(1):1053-69. https://doi.org/10.1007/s11357-023-00832-3
10. Kuryłowicz A. Estrogens in adipose tissue physiology and obesity-related dysfunction. Biomedicines. 2023 Feb 24;11(3):690. https://doi.org/10.3390/biomedicines11030690
11. Benjamin EJ, Blaha MJ, Chiuve SE, Cushman M, Das SR, Deo R, De Ferranti SD, Floyd J, Fornage M, Gillespie C, Isasi CR. Heart disease and stroke statistics—2017 update: a report from the American Heart Association. circulation. 2017 Mar 7;135(10):e146-603. https://doi.org/10.1161/CIR.0000000000000485
12. Dai W, Ming W, Li Y, Zheng HY, Wei CD, Rui Z, Yan C. Synergistic effect of a physiological ratio of estradiol and testosterone in the treatment of early-stage atherosclerosis. Archives of medical research. 2015 Nov 1;46(8):619-29. https://doi.org/10.1016/j.arcmed.2015.11.003
13. Blakemore J, Naftolin F. Aromatase: contributions to physiology and disease in women and men. Physiology. 2016 Jun 1. https://doi.org/10.1152/physiol.00054.2015
14. van Koeverden ID, de Bakker M, Haitjema S, van der Laan SW, de Vries JP, Hoefer IE, de Borst GJ, Pasterkamp G, den Ruijter HM. Testosterone to oestradiol ratio reflects

systemic and plaque inflammation and predicts future cardiovascular events in men with severe atherosclerosis. Cardiovascular Research. 2019 Feb 1;115(2):453-62. https://doi.org/10.1093/cvr/cvy188
15. Naftolin F, Mehr H, Fadiel A. Sex steroids block the initiation of atherosclerosis. Reproductive sciences. 2016 Dec;23(12):1620-5. https://doi.org/10.1177/1933719116674078
16. Wu L, Bao X, Xu J, Ma L, Kang L, Zhang R. The triglyceride-glucose index positively associates with the prevalence and severity of coronary heart disease in patients among hypertension. Scientific Reports. 2025 Jun 4;15(1):1-9.
17. Huang Y, Dai W, Li Y. Potential associations of testosterone/estradiol ratio, leukocyte hTERT expression and PBMC telomerase activity with aging and the presence of coronary artery disease in men. Experimental gerontology. 2019 Mar 1;117:38-44. https://doi.org/10.1016/j.exger.2018.08.008
18. Wang F, Wei XL, Wang FH, Xu N, Shen L, Dai GH, Yuan XL, Chen Y, Yang SJ, Shi JH, Hu XC. Safety, efficacy and tumor mutational burden as a biomarker of overall survival benefit in chemo-refractory gastric cancer treated with toripalimab, a PD-1 antibody in phase Ib/II clinical trial NCT02915432. Annals of Oncology. 2019 Sep 1;30(9):1479-86. https://doi.org/10.1093/annonc/mdz197
19. Kusters CD, Paul KC, Lu AT, Ferruci L, Ritz BR, Binder AM, Horvath S. Higher testosterone and testosterone/estradiol ratio in men are associated with decreased Pheno-/GrimAge and DNA-methylation based PAI1. GeroScience. 2024 Feb;46(1):1053-69.
20. Thirumalai A, Anawalt BD. Relationships between endogenous and exogenous testosterone and cardiovascular disease in men. Reviews in Endocrine and Metabolic Disorders. 2022 Dec;23(6):1305-22.

CONFLICT OF INTEREST

Authors declared no conflicts of Interest.

GRANT SUPPORT AND FINANCIAL DISCLOSURE

Authors have declared no specific grant for this research from any funding agency in public, commercial or nonprofit sector.

DATA SHARING STATEMENT

The data that support the findings of this study are available from the corresponding author upon request.