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Combination of Mangosteen (*Garcinia mangostana* L.) Pericarp Extract and Physical Exercise Decreases Atherosclerotic Lesions in Atherogenic Diet-Fed Rats

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Abstract

The mangosteen pericarp (MP) has xanthone compounds, particularly alpha-mangostin (α-MG), which have been shown to exhibit potent antioxidant and anti-inflammatory activities. Physical exercise (PE) has also been shown to have atheroprotective effects. To date, their combined effect on atherosclerosis has not been studied histologically. This study aimed to investigate the effect of MP extract combined with PE on atherosclerotic changes in rats fed an atherogenic diet. Twenty-five male Wistar rats (Rattus norvegicus) aged 6-8 weeks were randomly divided into five groups of five rats each: C (control, normal diet), CH (atherogenic diet), T1 (atherogenic diet plus MP extract 800 mg/kg/day in three divided doses with PE), T2 (atherogenic diet plus MP extract-loaded nanoemulsion 50 mg/kg/day with PE), and T3 (atherogenic diet plus atorvastatin 1.44 mg/day with PE). The PE protocol involved daily treadmill running for 60 min at 12 m/min, 5 days/week. All rats were treated for eight weeks. Following eight weeks of treatment, the animals were sacrificed and the aortic tissues were taken for histological study. The histological features in groups CH, T1, T2, and T3 were consistent with type Vc atherosclerotic lesion (fibrotic lesion) with intimal thicknesses of 11.99 ± 0.88 μm, 3.61 ± 0.53 μm, 3.48 ± 0.47 μm, and 2.96 ± 0.20 μm, respectively. A comparative analysis revealed a significant reduction in intimal thickness in groups T1, T2, and T3 compared to the CH group ($p < 0.001$). Group T2 exhibited a non-significant decrease compared to group T1 ($p = 0.691$). Moreover, neither the T1 nor T2 groups significantly differed from the T3 group ($p = 0.052$ and 0.128, respectively). In conclusion, the combination of MP extract (and its nanoemulsion) and PE decreases atherosclerotic lesions and provides comparable results to the atorvastatin-treated group, indicating their potential as alternative options for preventing atherosclerosis.

Keywords: Atherosclerosis; *Garcinia mangostana* L.; Nanoemulsion; Statin; Physical exercise

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Introduction

Despite accelerating advances in its medical and surgical management, coronary artery disease (CAD) remains a significant health and economic burden worldwide. According to the Global Burden of Disease (GBD) 2016 study, CAD was responsible for an estimated 2.2% of the overall global burden of disease and 32.7% of cardiovascular diseases (CVD). In the United States, CAD is estimated to cost the healthcare system over 200 billion dollars annually [1,2]. Consequently, the main focus of research on CAD has shifted from curative to preventive efforts. CAD refers to a condition that typically involves the formation of atherosclerotic lesions in the coronary arteries, leading to a reduction in blood flow and thus oxygen delivery to the myocardium [3]. Atherosclerosis is a slowly progressive disease characterized by chronic inflammation, oxidative stress, and the buildup of lipids, fibrous elements, and calcification within the intima of large and medium-sized arteries. This pathology is initiated by endothelial dysfunction in which dyslipidemia, diabetes, hypertension, cigarette smoking, and obesity play a pivotal pathogenic role [4]. CAD can be prevented by addressing atherosclerosis risk factors through lifestyle changes and medications such as statins. However, the use of statins, although therapeutically effective, is not without potential adverse effects. Rhabdomyolysis, elevation of liver enzymes, new-onset diabetes mellitus, and intracranial hemorrhage have been reported [5]. Moreover, statin therapy is only cost-effective for those at high risk of CAD, but is not cost-effective for other population subsets [6]. Therefore, the search for effective, safe, and inexpensive alternatives to statins is of great interest.

Mangosteen (*Garcinia mangostana* L.) is a tropical evergreen tree belonging to the Guttiferae family that grows indigenously in Southeast Asia and is cultivated mainly for its edible fruit. The pericarp of the fruit has a long history of use as a traditional medicine to treat a wide variety of ailments [7]. Phytochemically, the mangosteen pericarp (MP) contains bioactive secondary metabolites like xanthones, mostly alpha-mangostin (α-MG), which have been reported to possess antioxidant, anti-inflammatory, antidyslipidemic, antidiabetic, anti-obesity, and cardioprotective properties [8-13]. Despite its immense therapeutic potential, α-MG still has limited clinical application. This is attributed to the low water solubility of α-MG, which limits its bioavailability. To address the pharmacological limitations of α-MG, the application of nanoparticle-based drug formulation has emerged as a promising solution [14]. In summary, MP shows promise as an anti-atherosclerotic agent and warrants further evaluation.

Physical exercise (PE) has been shown to have beneficial anti-atherogenic effects, including decreased oxidative stress and inflammation [15]. The combined treatment of PE with dietary antioxidants, such as vitamins C and E, significantly decreased the progression of atherosclerotic lesions and prolonged survival in hypercholesterolemic mice [16,17]. However, little is known regarding the effect on atherosclerosis of combining PE and MP extract. Thus, the present study aimed to investigate the effect of MP extract (and its nanoemulsion) and PE on atherosclerotic changes in rats fed an atherogenic high-cholesterol diet.

Methods

This experimental laboratory study was conducted in the Biomolecular Laboratory of the Universitas Islam Sultan Agung (Semarang City, Central Java, Indonesia). The experimental protocols were approved by the Health Research Ethics Committee of Universitas Diponegoro, Faculty of Medicine (approval number: 51/EC/H/FK-UNDIP/VI/2022) and followed the national guidelines for the care and use of laboratory animals.

Animals and experimental design

Twenty-five male Wistar rats (*Rattus norvegicus*) aged 6-8 weeks with an average body weight of 309±41 g were obtained from the Experimental Animal Research Laboratory, Universitas Negeri Semarang. The animals were caged individually at room temperature and humidity with a 12 h light/dark cycle and were provided with a standard pellet diet and water *ad libitum* for one week before starting the experiment. After one week of acclimatization, the rats were randomly divided into five groups of five: C (control, normal diet), CH (atherogenic diet), T1 (atherogenic diet plus MP extract 800 mg/kg/day in three divided doses with PE), T2 (atherogenic diet plus MP extract-loaded nanoemulsion 50 mg/kg/day with PE), and T3 (atherogenic diet plus atorvastatin 1.44 mg/day with PE). The dosage of MP extract was selected according to an earlier report, which identified 800 mg/kg/day as the most efficacious dose [18]. The preparation and dosage of nanoemulsion were adapted based on a previous study that demonstrated improved oral bioavailability of α-MG with a dosage of 50 mg/kg/day [19]. To conform to the human daily dose of atorvastatin (80 mg/day), the rat dose was adjusted based on the ratio of human to rat body surface area [20]. The PE protocol consisted of daily exercise of 60 min of running at 12 m/min, 5 days/week on a customized rodent treadmill set at 0° incline. A modified version of a previously evaluated PE protocol was used [21]. The experiment

was continued for eight weeks, after which the animals were sacrificed by chloroform inhalation and the aortic tissues were collected for histological analysis.

Atherogenic diet

Rats in groups CH and T1-3 were fed with an atherogenic diet containing high levels of fat (20-23% by weight; 40-45% kcal from fat), saturated fatty acids (>60% of total fatty acids), butterfat, sucrose (34% by weight), and cholesterol (0.2% total) for eight weeks to induce hypercholesterolemia and atherosclerosis.

Preparation of MP Extract and Nanoemulsion

The MP extract employed in this study was the standardized herbal product Mastin® (Borobudur Natural Herbal Industry, Semarang, Indonesia). It was prepared from dried fruit pericarp using a 10:1 ratio of raw material to final product, employing a percolator system extraction method with 70% ethanol as the solvent. Thin-layer chromatography was used to confirm that the final extract contained a minimum of 25% α-MG. The extract was then dried using a vacuum belt dryer. The required MP extract was weighed and dissolved in distilled water prior to feeding. The bioavailability of bioactive compounds in the MP extract can be improved using nanoemulsion formulation. Briefly, high-energy ultrasonication technique was utilized to prepare the MP extract-loaded nanoemulsion system [19]. The oil phase was prepared by mixing oleic acid and isopropyl myristate using a magnetic stirrer at 2000 rpm and 40 °C for 30 min. Cremophor® EL (polyoxyl 35 castor oil) was added and the mixture was stirred at 2000 rpm and 40 °C for 30 min. Then, ethanol and MP extract were added to the oil mixture and stirring was continued at 2000 rpm and 40 °C for 120 min. The aqueous phase (distilled water) was then gradually poured over 60 min into the lipid phase at a stirring speed of 2000 rpm at room temperature. Finally, the generated nanoemulsion was ultrasonicated at a power of 1 kW for 10 min.

Histological analysis

After sacrificing the rats, the aorta was removed and fixed in 10% neutral buffered formalin. Transverse sections from aortic specimens were obtained from the aortic arch and aortic sinus. These samples were embedded in paraffin blocks, sectioned, and stained with hematoxylin-eosin (HE) for microscopic evaluation by two anatomical pathologists who were blinded to group assignment. The histopathologic atherosclerotic lesions of the aorta were classified according to the American Heart Association (AHA) classification [22]. Several histological features were evaluated, including intimal thickness and the presence of foam cells, lipid-laden smooth muscle cells, extracellular lipid pools and cores, calcium deposits, fibrous tissue, fissures, hematoma, and thrombus. For each section, the evaluation was performed on four microscopic fields (12, 3, 6, and 9 o'clock positions) at 400× magnification. Photomicrographs were obtained using a Nikon Eclipse E200 microscope equipped with a Sony IMX265 CMOS camera and were analyzed using IndomicroView software.

Statistical analysis

Statistical analysis was performed using SPSS Statistics software (version 26.0, SPSS Inc., Chicago, IL, USA). Data were presented as mean ± standard deviation (SD) or median as appropriate. The Shapiro-Wilk test and Levene's test were used to determine data normality and homogeneity of variances, respectively. One-way analysis of variance (ANOVA) followed by LSD post-hoc test for multiple comparisons among the groups was performed. The chi-square and Kruskal-Wallis tests were used to compare categorical variables. Statistical significance was defined at $p < 0.05$.

Results

The atherosclerotic morphology changes in the aortic wall of the five groups are shown in figure 1. The control group showed healthy aorta without any lesions in the intima or media, with intimal thickness of 2.92 ± 0.28 μm. In contrast, histopathological findings from the CH group revealed the presence of thickened tunica intima (11.99 ± 0.88 μm), layers of foam cells and lipid-laden smooth muscle cells, extracellular lipid pools, and fibrous connective tissue. Samples from groups T1, T2, and T3 displayed similar but less pronounced features (Table 1), with intimal thicknesses of 3.61 ± 0.53 μm, 3.48 ± 0.47 μm, and 2.96 ± 0.20 μm, respectively. These features are consistent with type Vc atherosclerotic lesion (fibrotic lesion) as classified by the AHA.

Table 2 provides a comparative analysis of intimal thickness across the different treatment groups. The intimal thickness was significantly increased in the CH group compared to the control group ($p < 0.001$). Statistical analysis showed a significant reduction ($p < 0.001$) in the thickness of the tunica intima of T1, T2, and T3 groups compared to the CH group. While there was a reduction in intimal thickness in the T2 group compared to the T1 group, this difference was not statistically significant ($p = 0.691$). Additionally, both the T1 and T2 groups did not show statistically significant differences compared to the positive control (T3) group ($p = 0.052$ and 0.128, respectively).

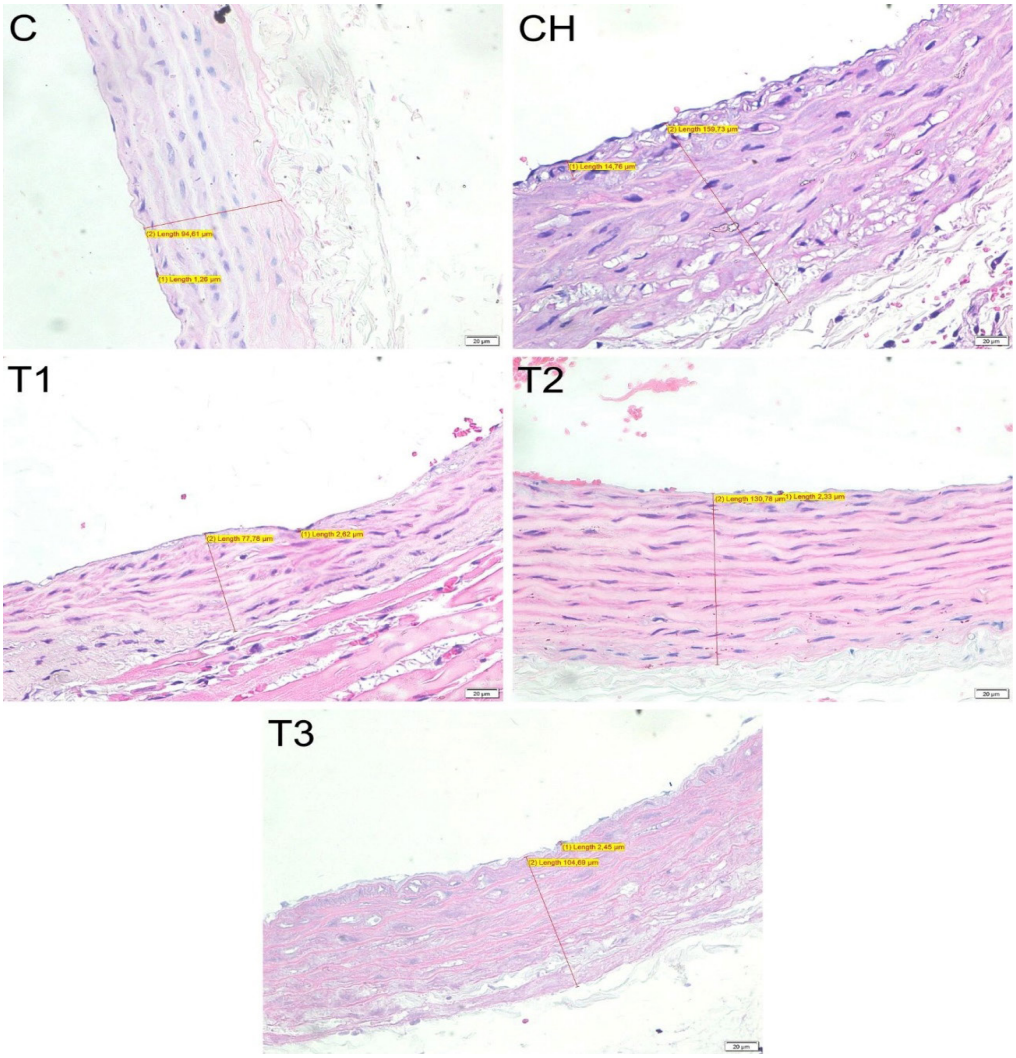

Figure 1. Representative photomicrographs of rat aorta samples from the five groups. C = control, normal diet; CH = atherogenic diet; T1 = atherogenic diet plus MP extract with PE; T2 = atherogenic diet plus MP extract-loaded nanoemulsion with PE; T3 = atherogenic diet plus atorvastatin with PE (HE stain, original magnification 400×).

Discussion

Hypercholesterolemia is a major risk factor for CAD, and the Framingham study has demonstrated a direct relationship between serum cholesterol levels and the incidence of CAD [23]. The present study showed that a high-cholesterol dietary treatment for eight weeks led to the development of atherosclerotic lesions in the aortic wall of rats. This finding is in accordance with earlier studies, which have reported that feeding rodents a high-cholesterol diet for 6-10 weeks resulted in hypercholesterolemia and, subsequently, atherosclerosis [24,25]. Atorvastatin, a potent cholesterol-lowering agent, was used as a positive control due to its ability to inhibit HMG-CoA reductase, a key enzyme in the cholesterol biosynthesis pathway [5]. Oxidative stress, characterized by the overproduction of reactive oxygen species, represents one of the main pathogenetic mechanisms underlying atherosclerosis and is closely related to endothelial dysfunction, oxidative modification of low-density lipoprotein, and inflammatory response within the injured vascular wall. The close interplay between oxidative stress and inflammation creates a vicious cycle that promotes the initiation and progression of atherosclerosis [4]. Therefore, targeting both mechanisms could provide a promising strategy to prevent atherosclerosis. Previous studies have shown clear beneficial effects of antioxidant supplementation (vitamins C and E) and PE

Table 1. Quantitative assessment of histological fea tures

Histological Feature	Group			
	CH (n = 5)	T1 (n = 5)	T2 (n = 5)	T3 (n = 5)
Layers of foam cells and lipid-laden smooth muscle cells[†]				
1 layer	1	2	2	4
>1 layer	4	3	3	1
Presence of extracellular lipid pools[†]				
Absent	0	0	0	0
Present	5	5	5	5
Extent of fibrous connective tissue[†]				
0–5%	0	2	3	3
5–25%	5	3	2	2

CH = atherogenic diet; T1 = atherogenic diet plus MP extract with PE; T2 = atherogenic diet plus MP extract-loaded nanoemulsion with PE; T3 = atherogenic diet plus atorvastatin with PE. [†]No significant overall difference among the groups

Table 2. Comparison of aortic intimal thickness in the five groups.

Group	Mean ± SD (*μm)*	Median (min–max)
C (n = 5)	2.92 ± 0.28	2.86 (2.56–3.31)
CH (n = 5)	11.99 ± 0.88[a]	12.04 (11.01–13.29)
T1 (n = 5)	3.61 ± 0.53[b]	3.71 (2.62–4.11)
T2 (n = 5)	3.48 ± 0.47[b]	3.70 (2.74–3.87)
T3 (n = 5)	2.96 ± 0.20[b]	2.90 (2.73–3.21)
Overall Sig	<0.001	

C = control, normal diet; CH = atherogenic diet; T1 = atherogenic diet plus MP extract with PE; T2 = atherogenic diet plus MP extract-loaded nanoemulsion with PE; T3 = atherogenic diet plus atorvastatin with PE; [a] p <0.001 by LSD post-hoc test compared to C group; [b] p <0.001 by LSD post-hoc test compared to CH group; SD = standard deviation; Overall Sig = overall significance.

on atherosclerotic lesion formation [16,17]. Hence, it is plausible that combined treatment with MP extract (as a source of natural antioxidants) and PE can also produce similar effects.

Current evidence shows that most of the biological effects of mangosteen are significantly correlated with the concentration of α-MG [26]. α-MG, a major xanthone derivative found abundantly in MP, has been shown to exhibit potent antioxidant and anti-inflammatory activities [8,9]. A study by Wihastuti et al. found that administering MP extract to hypercholesterolemic rats significantly suppressed oxidative stress and inflammatory markers and reduced aortic intima-media thickness. These findings provide evidence that targeting these specific mechanisms can attenuate atherosclerosis progression [18]. PE is well known for its cost-effective benefits in preventing and managing CVD, especially CAD [15]. Rentz et al. reported that the anti-atherogenic role of PE is associated with the attenuation of oxidative damage and inflammatory state of atherosclerotic lesions via inhibition of macrophage infiltration and activation. Moreover, PE increased the levels of high-density lipoprotein cholesterol, which has been shown to exert atheroprotective effects, mainly by promoting macrophage cholesterol efflux [27]. The results of this study extend previous findings, which found that MP extract and PE each confer atheroprotective effects [18,27]. Our findings emphasize that their combined action also provides protective effects against atherosclerosis by reducing intimal thickness and improving several histological features. These outcomes may be attributed to their synergistic antioxidant and anti-inflammatory activities. Moreover, to the best of our knowledge, this is the first *in vivo* study on MP extract that utilizes the histopathological parameters recommended by the AHA for atherosclerosis research.

As a drug delivery system, nanoemulsions are promising as carriers for enhancing the aqueous solubility of poorly water-soluble drugs. Increased solubility, facilitated by a larger surface area and faster dissolution rate, improves drug bioavailability and therapeutic efficacy [14]. It is noteworthy that the observed effects were not limited solely to the MP extract-treated group (T1), but also extended to its nanoemulsion form (T2). Albeit not statistically significant, group

T2 specimens showed less extensive lesions than those of group T1. This finding is in good agreement with a previous study reporting that a nano-sized delivery system efficiently improved the pharmacokinetic performance of α-MG and modified its tissue distribution features, thereby enhancing its clinical efficacy [19]. In addition, neither group T1 nor T2 showed a significant difference in aortic intimal thickness compared to the atorvastatin-treated group (T3), suggesting that the MP extract and nanoemulsion groups offer a comparable protective effect against atherosclerosis.

One limitation of this study was the use of rats as an experimental model. Larger animal models like rabbits are preferred for atherosclerosis research since their lipoprotein profile and metabolism are more similar to those of humans. In order to gain a deeper understanding of the impact of each intervention on the progression of atherosclerotic lesions, it is recommended that future studies compare the atheroprotective effects between MP extract, PE, and their combination.

Conclusion

In conclusion, histological analysis demonstrated that the combination of mangosteen pericarp (MP) extract and physical exercise (PE) (T1) significantly reduced atherosclerotic lesion formation in the aorta of atherogenic diet-fed rats. Furthermore, treatment with MP extract-loaded nanoemulsion and PE (T2) resulted in a greater reduction in aortic intimal thickness compared to group T1, suggesting an enhancement in the solubility and bioavailability of α-MG. Additionally, either the T1 or T2 group was equally effective in decreasing aortic intimal thickness when each was compared to the atorvastatin-treated group. These results indicate that MP extract, particularly its nanoemulsion, combined with PE may serve as a potential alternative for preventing atherosclerosis. This study provides a basis for further research into the clinical application of MP combined with PE in the prevention of CAD.

Conflict of Interests

None.

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None.

References

[1] Bauersachs R, Zeymer U, Brière JB, Marre C, Bowrin K, et al. Burden of coronary artery disease and peripheral artery disease: a literature review. Cardiovasc Ther 2019;2019.

[2] Brown JC, Gerhardt TE, Kwon E. Risk Factors for Coronary Artery Disease. Treasure Island (FL). StatPearls Publishing 2024.

[3] Prasad K. Current status of primary, secondary, and tertiary prevention of coronary artery disease. Int J Angiol 2021;30:177-186.

[4] George J. Pathophysiology of coronary artery disease. In: Interventional Cardiology Imaging: An Essential Guide. Abbas A. London: Springer. London 2015; pp 29-46.

[5] Ruscica M, Ferri N, Banach M, Sirtori CR, Corsini A. Side effects of statins: from pathophysiology and epidemiology to diagnostic and therapeutic implications. Cardiovasc Res 2023;118:3288-3304.

[6] Franco OH, Peeters A, Looman CWN, Bonneux L. Cost effectiveness of statins in coronary heart disease. J Epidemiol Community Health 2005;59:927-933.

[7] Ovalle-Magallanes B, Eugenio-Pérez D, Pedraza-Chaverri J. Medicinal properties of mangosteen (Garcinia mangostana L.): A comprehensive update. Food Chem Toxicol 2017;109:102-122.

[8] Vien LC, Chinnappan S, Mogana R. Antioxidant activity of Garcinia mangostana L and alpha mangostin: A review. Res J Pharm Technol 2021;14:4466-4470.

[9] Setiawan AA, Budiman J, Prasetyo A. Anti-inflammatory potency of mangosteen (Garcinia mangostana L.): A systematic review. Open Access Maced J Med Sci 2023;11:58-66.

[10] Warditiani NK, Astuti KW, Sari PMNA, Wirasuta I. Antidyslipidemic activity of methanol, ethanol and ethyl acetate mangosteen rind (Garcinia mangostana L). Res J Pharm Technol 2020;13:261-264.

[11] Kim HM, Kim YM, Huh JH, Lee ES, Kwon MH, et al. α-Mangostin ameliorates hepatic steatosis and insulin resistance by inhibition C-C chemokine receptor 2. PLoS One 2017;12:e0179204..

[12] Choi YH, Bae JK, Chae HS, Kim YM, Sreymom Y, et al. α-Mangostin regulates hepatic steatosis and obesity through SirT1-AMPK and PPARγ pathways in high-fat diet-induced obese mice. J Agric Food Chem 2015;63:8399-8406.

[13] Sampath PD, Vijayaragavan K. Ameliorative prospective of alpha-mangostin, a xanthone derivative from Garcinia mangostana against β-adrenergic cathecolamine-induced myocardial toxicity and anomalous cardiac TNF-α and COX-2 expressions in rats. Exp Toxicol Pathol 2008;60:357-364.

[14] Alam M, Rashid S, Fatima K, Adnan M, Shafie A, et al. Biochemical features and therapeutic potential of α-Mangostin: Mechanism of action, medicinal values, and health benefits. Biomed Pharmacother. 2023;163:114710.

[15] Chacon D, Fiani B. A review of mechanisms on the beneficial effect of exercise on atherosclerosis. Cureus 2020;12:e11641.

[16] Napoli C, Williams-Ignarro S, De Nigris F, Lerman LO, Rossi L, et al. Long-term combined beneficial effects of physical training and metabolic treatment on atherosclerosis in hypercholesterolemic mice. Proc Natl Acad Sci U S A 2004;101:8797-8802.

[17] Napoli C, Williams-Ignarro S, De Nigris F, Lerman LO, D'Armiento FP, et al. Physical training and metabolic supplementation reduce spontaneous atherosclerotic plaque rupture and prolong survival in hypercholesterolemic mice. Proc Natl Acad Sci U S A 2006;103:10479-10484.

[18] Wihastuti TA, Aini FN, Tjahjono CT, Heriansyah T. Dietary ethanolic extract of mangosteen pericarp reduces vcam-1, perivascular adipose tissue and aortic intimal medial thickness in

hypercholesterolemic rat model. Open Access Maced J Med Sci 2019;7:3158-3163.
[19] Xu WK, Jiang H, Yang K, Wang YQ, Zhang Q, et al. Development and in vivo evaluation of self-microemulsion as delivery system for α-mangostin. Kaohsiung J Med Sci 2017;33:116-123.
[20] Laurence DR, Bacharach AL. Evaluation of drug activities: pharmacometrics. Elsevier. Amsterdam (The Netherlands) 1964.
[21] Kadoglou NPE, Kostomitsopoulos N, Kapelouzou A, Moustardas P, Katsimpoulas M, et al. Effects of exercise training on the severity and composition of atherosclerotic plaque in apoE-deficient mice. J Vasc Res 2011;48:347-356.
[22] Stary HC, Chandler AB, Dinsmore RE, Fuster V, Glagov S, et al. A definition of advanced types of atherosclerotic lesions and a histological classification of atherosclerosis. A report from the Committee on Vascular Lesions of the Council on Arteriosclerosis, American Heart Association. Circulation. 1995;92:1355-1374.
[23] Kannel WB, Dawber TR, Kagan A, Revotskie N, Stokes J. Factors of risk in the development of coronary heart disease–six year follow-up experience. The Framingham Study. Ann Intern Med 1961;55:33-50.
[24] Nakashima Y, Plump AS, Raines EW, Breslow JL, Ross R. ApoE-deficient mice develop lesions of all phases of atherosclerosis throughout the arterial tree. Arterioscler Thromb 1994;14:133-140.
[25] Plump AS, Smith JD, Hayek T, Aalto-Setälä K, Walsh A, et al. Severe hypercholesterolemia and atherosclerosis in apolipoprotein E-deficient mice created by homologous recombination in ES cells. Cell 1992;71:343-353.
[26] Gutierrez-Orozco F, Failla ML. Biological activities and bioavailability of mangosteen xanthones: A critical review of the current evidence. Nutrients 2013;5:3163-3183.
[27] Rentz T, Wanschel ACBA, de Carvalho Moi L, Lorza-Gil E, de Souza JC, et al. The anti-atherogenic role of exercise is associated with the attenuation of bone marrow-derived macrophage activation and migration in hypercholesterolemic mice. Front Physiol 2020;11:599379.