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Statin Therapy Mitigates Oxidative Stress in Epicardial and Perivascular Adipose Tissue: A Pilot Study in Cardiac Surgery Patients

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Abstract: Epicardial and perivascular adipose tissues have gained significant attention in the past decade due to their involvement in complex metabolic shifts and inflammatory changes in cardiovascular diseases. Statins, the cornerstone of lipid-lowering therapy, have pleiotropic effects, including the potential of alleviating epicardial adipose dysfunction and inflammation, although their impact on local oxidative stress has received less scrutiny. This study was purported to assess the production of reactive oxygen species (ROS) in epicardial and perivascular fat samples harvested from patients undergoing elective cardiac surgery and who were treated or not with statins. Here, we report that patients chronically treated with statins (atorvastatin and rosuvastatin) exhibited significantly lower levels of ROS in their epicardial and perivascular adipose tissues. Additionally, a positive correlation was observed between adipose tissue oxidative stress and the diameter of the right ventricle. Larger studies are required to provide mechanistic insights regarding the sources of epicardial ROS and signal transduction pathways activated by statins.

Keywords: epicardial adipose tissue; perivascular adipose tissue; oxidative stress; reactive oxygen species; statins; cardiac surgery

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Introduction

Epicardial adipose tissue (EAT) represents the true visceral adipose tissue of the heart with which it shares both its embryonic origin as well as blood supply [1]. EAT has unobstructed anatomical contiguity with the myocardium and envelops up to 80% of its surface [2]. Research in the past decade has linked EAT to vast implications in cardiac biology that extend past the mechanical and thermal protection of the heart and coronary arteries. Indeed, epicardial fat provides an energy source for cardiomyocytes, protects them from free fatty acid lipotoxicity and modulates intracellular Ca^{2+} cycling, the electrical and mechanical behavior of heart muscle cells and the myocardial redox state [3]. However, the presence of underlying cardiometabolic conditions, mainly overweight/obesity and diabetes, is a major risk factor leading to EAT expansion and inflammation [4,5]. The subsequent EAT dysfunction is responsible for the shift from a protective phenotype toward a pro-inflammatory, pro-oxidant and pro-fibrotic cardiac micro-environment that contributes to the pathogenesis of cardiometabolic pathologies and their progression toward heart failure with the whole spectrum of the ejection fraction [6]. Nowadays, EAT is regarded as a modifiable risk factor in cardiovascular diseases [7], which can be evaluated with non-invasive imaging techniques [8] and also therapeutically targeted [6].

Statins remain the first-line lipid-lowering treatment in primary and secondary cardiovascular prevention due to their demonstrated long-term benefits in reducing both morbidity and mortality. These drugs primarily decrease low-density lipoprotein cholesterol (LDL-C) levels both by inhibiting 3-hydroxy-3-methylglutarate CoA reductase, a critical component in the cholesterol biosynthesis pathway, and enhancing the expression of the LDL receptor, thereby augmenting LDL-C clearance from the bloodstream [9]. Given the pivotal contribution of LDL-C to the onset of atherosclerosis, the two aforementioned synergistic mechanisms largely account for the observed reductions in major adverse cardiovascular events associated with statin therapy [9]. However, irrespective of the lipid-lowering effect, statins have been unequivocally demonstrated to exert pleiotropic effects, such as improvement in plaque stability, alleviation of endothelial function, reduction in inflammation and thrombosis and, mitigation of oxidative stress [9]. Among these, a highly investigated effect was the diminution of EAT volume, thickness and density, as well as EAT-derived inflammatory mediators [10–12].

Perivascular adipose tissue (PVAT) encompasses the fat depots that are adjacent to the vascular tree and is required to maintain the functional status of vasculature. However, when excessive in various vascular disorders, PVAT becomes dysfunctional and its altered secretome impairs both vascular smooth muscle and endothelial cells [13].

The present study was purported to assess the oxidative stress in samples of epicardial and perivascular adipose tissues harvested from patients undergoing elective cardiac surgery and compare ROS production between patients treated or not with statin therapy. A second objective was to investigate whether correlations could be found between the magnitude of oxidative stress and the echocardiographic parameters.

Materials and Methods

Study Population

This is a pilot study that includes 25 overweight/obese patients with heart failure (HF) with a mildly reduced ejection fraction (HFmrEF, EF = 41% to 49%) and indication of elective cardiac surgery, divided into two groups based on whether they were receiving (n = 17) or not (n = 8) statin therapy.

The study protocol and all related procedures underwent a thorough review and obtained approval by the Commission for Research Ethics of "Victor Babeș" University of Medicine and Pharmacy from Timisoara, Romania (no. 09/22.03.2021 rev. 29.05.2023) and Commision for Ethics in Research and Development of the Institute for Cardiovascular Diseases of Timișoara, respectively. The research adhered to the EU Good Clinical Practice Directives (2005/28/EC) and the principles of the Declaration of Helsinki. To safeguard the confidentiality and privacy of patient information, all identifiable data were removed and the data were anonymized before analysis.

Inclusion and Exclusion Criteria

The inclusion criteria for the study were as follows: (i) consecutive patients referred for cardiac surgery, diagnosed with coronary artery disease (CAD) and valvular heart pathologies, (ii) adults aged 18 years or older and (iii) consent to participate in the study, including agreement to the use of biological samples and medical data for publication. The exclusion criteria for the study were as follows: (i) poor echocardiographic windows that could hinder accurate cardiac assessment, (ii) presence of neoplasia, to avoid potential confounding effects of cancer or its treatments on cardiac and adipose tissue health, (iii) chronic inflammatory or autoimmune diseases, as they could independently impact cardiovascular health and adipose tissue, (iv) active infections or chronic hepatic diseases, considering their systemic effects that could make the results biased and (v) congenital heart disease, to maintain focus on acquired cardiac conditions rather than congenital abnormalities. Patients were further divided into statin- and non-statin-treated groups. The demographic, laboratory and echocardiographic parameters, as well as therapy at admission of the study groups are presented in Table 1.

Table 1. Characteristics of the study groups.

Demographic and Lab Parameters	**(+) Statins (n = 17)**	**(−) Statins (n = 8)**
Age (y)	66.3 ± 8.2	62.6 ± 6.1
Sex (M/F), n(%)	14 (82.4)/3 (17.5)	4 (50)/4 (50)
BMI (kg/m^2)	28.3 ± 4.9	26 ± 4.4
Fasting plasma glucose, mg/dL	128.1 ± 70.1	102.1 ± 22.2
Total cholesterol, mg/dL	181.2 ± 64.1	183.1 ± 50.2
ESR, mm/h	21.8 ± 19.8	27.2 ± 23.1
AST, U/L	22.3 ± 8.7	22.1 ± 5.4
ALT, U/L	30.7 ± 22	20.8 ± 8.7
Creatinine, mg/dL	0.96 ± 0.2	1.04 ± 0.4
Echocardiographic parameters		
LV ejection fraction, %	48.1 ± 0.07	46.8 ± 0.07
LA diameter, cm	4.6 ± 0.7	4.8 ± 0.7
RV diameter, cm	2.8 ± 0.3	3.2 ± 0.5
LV end-diastolic diameter, cm	5 ± 0.6	5.6 ± 1.1
LV end-diastolic volume index, mL/m^2	134.7 ± 39.8	170.3 ± 103.8
Therapy at admission		
Aspirin, n (%)	12 (70.5)	2 (25)
β-blockers, n (%)	12 (70.5)	6 (75)
Nitrates, n (%)	7 (41.1)	0
CCBs, n (%)	6 (35.2)	2 (25)
Diuretics, n (%)	15 (88.2)	8 (100)
Insulin Therapy, n (%)	3 (17.6)	0
OADs, n (%)	5 (29.4)	0
ROS levels		
Epicardial Adipose Tissue	17.9 ± 3.2 *	21.3 ± 2.7
Perivascular Adipose Tissue	18.3 ± 2.8 *	21.8 ± 2.8

CCB = calcium channel blockers; OAD = oral antidiabetics; ROS = reactive oxygen species; EAT = epicardial adipose tissue; PVAT = perivascular adipose tissue; * $p < 0.05$.

Oxidative Stress Assessment in Spectrophotometry

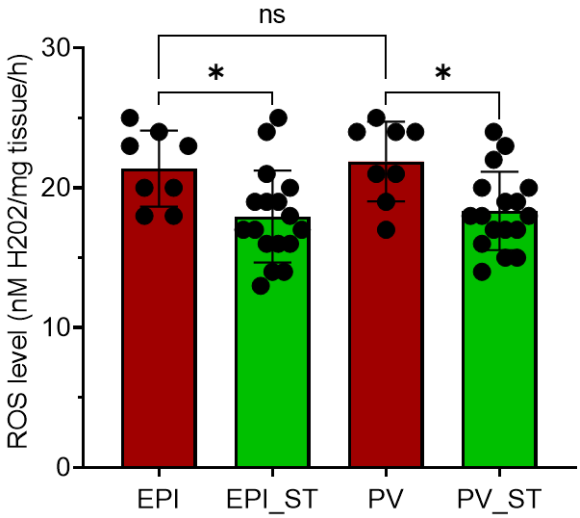
Epicardial adipose tissue samples were collected from the anterior wall of the right ventricle and perivascular adipose tissue from the peri-aortic and peri-pulmonary artery adipose tissue after the initiation of cardiopulmonary bypass. The samples were placed in an ice-cold buffer, transported to the laboratory and utilized for the measurement of the hydrogen peroxide (H_2O_2) level by means of the Ferrous iron xylenol orange oxidation (FOX) assay (PeroxiDetect kit, Merck-Sigma-Aldrich), as previously described [14]. In brief, the principle of the assay is that peroxides oxidize the ferrous (Fe^{2+}) to ferric Fe^{3+} ions at an acidic pH. The Fe^{3+} ion forms a colored adduct with the compound xylenol orange, which can be measured at 560 nm. The results are expressed as nanomoles of H_2O_2 per milligram of tissue per hour (nmol H_2O_2/mg tissue/h).

Echocardiography

The transthoracic echocardiographic exam included conventional, tissue Doppler and speckle tracking imaging parameters, and it was performed one day before the surgical intervention using a modern ultrasonographic system (Vivid E95, General Electric, Milwaukee, WI, USA). Conventional parameters, including the left ventricle (LV) end-diastolic and end-systolic volumes and diameters, right ventricle (RV) diameter and left atrial (LA) maximum volume and area were measured according to the current European guidelines' recommendations [15–17], as previously described [18]. The LA volume and area were measured using the biplane Simpson's disc summation method (apical four-chamber and two-chamber views) at the end-systolic frame in order to achieve the maximum value. The LV volumes and LV ejection fraction (LVEF) were also calculated by using the biplane modified Simpson's method in the apical two- chamber and four-chamber views, carefully delineating the LV endocardium. The image acquisition for trans-mitral flow patterns was performed in an apical four-chamber view, with a horizontal sweep of 100 mm/s and the 3–5 mm pulsed-wave Doppler sample volume placed at the tip of the opened mitral valve leaflets. The early (E wave) and late (A wave) trans-mitral flow were measured on five consecutive cardiac cycles during end-expiratory apnea and the final values were calculated as the average of these measurements.

Results

Patients who were chronically treated with statins (atorvastatin or rosuvastatin) exhibited significantly lower levels of reactive oxygen species (ROS) in their epicardial (EPI) and perivascular (PV) adipose tissue compared to those who were not receiving treatment (Figure 1).



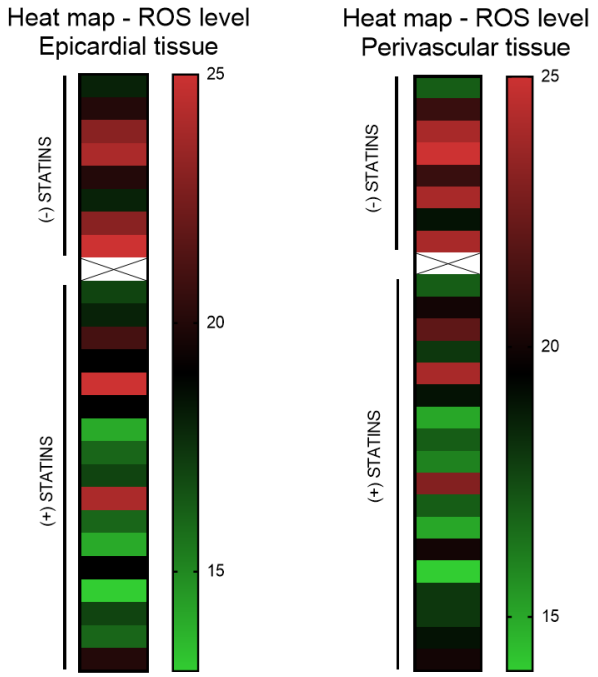


Figure 1. Level of oxidative stress in epicardial (EPI) and perivascular (PV) adipose tissue obtained from patients treated (n = 17) or not (n = 8) with statins (atorvastatin or rosuvastatin); FOX assay for H_2O_2 evaluation (results expressed in nM H_2O_2/mg tissue/h); * $p < 0.05$.

Correlation analysis was performed to evaluate a potential relationship between the echocardiographic parameters (LVEF, LAD, RVD, IVS, LVPWT, LVEDD) and the degree of oxidative stress (H_2O_2 measured by FOX assay) of the analyzed samples. A positive correlation was observed between the level of ROS in epicardial and perivascular adipose tissue and the diameter of the right ventricle (r = 0.47, $p < 0.05$) (Figure 2).

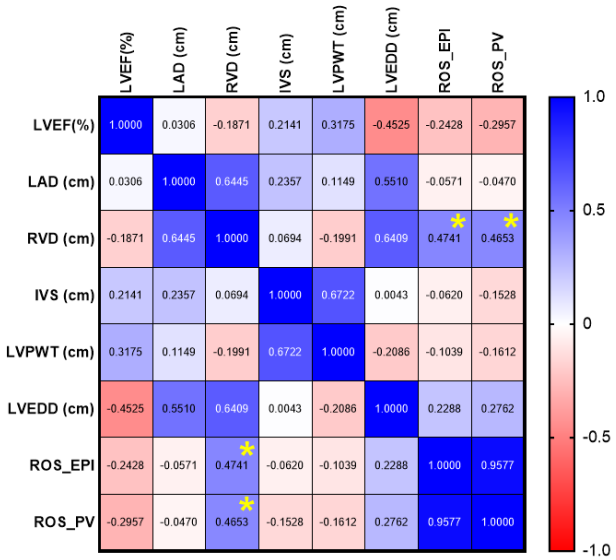


Figure 2. Corelation matrix between the echocardiographic parameters and the degree of oxidative stress (H_2O_2 level measured by FOX assay) in epicardial (EPI) and perivascular adipose tissue (PVAT); * $p < 0.05$. LVEF—left ventricular ejection fraction, LAD—left atrium diameter, RVD—right ventricular diameter, IVS—interventricular septum, LVPWT—left ventricle posterior wall thickness, LVEDD—left ventricular end diastolic diameter.

Discussion

Epicardial and perivascular adipose depots have emerged in the past decades as main modulators of the pathophysiology of both the heart and blood vessels, as well as therapeutic targets [19]. While in the normal heart, EAT exerts protective effects by providing thermogenic and energetic support to the coronary arteries and myocardium and being a source of local anti-inflammatory molecules, in pathological settings, EAT expansion is a major risk factor for cardiovascular diseases, particularly a predictor for coronary artery disease [20,21]. In the presence of multiple risk factors, such as obesity, systemic inflammation, hyperlipidemia and smoking, EAT adipocytes may undergo phenotypic changes and produce a secretome rich in proinflammatory cytokines that promote both atherosclerosis and myocardial fibrosis [5]. EAT plays a distinct pathophysiological role in HF with a reduced ejection fraction (HFrEF), indicating the decreased metabolic reservoir due to the catabolic state, whereas in HF with a preserved ejection fraction (HFpEF), its volume and activity increase, particularly in the presence of metabolic comorbidities [22].

Besides inflammation, oxidative stress is another mechanism contributing to HF pathogenesis and progression. Here, we have assessed the magnitude of oxidative stress in patients with HFmrEF treated or not with statins. In their pioneering study, Salgado-Somoza et al. reported back to 2010 an increased mRNA levels for several proteins related to oxidative stress, e.g., glutathione S-transferase P, protein disulfide isomerase and phosphoglycerate mutase 1 (but not catalase) in EAT compared to the subcutaneous adipose tissue (SAT) harvested from patients undergoing cardiac surgery [23]. In another study that compared EAT with SAT, the p53 mRNA level was up-regulated and increased after ex vivo adrenergic stimulation, mainly in EAT harvested from patients with HF [24]. An elevation of p53 in white AT promotes apoptosis, senescence, chronic inflammation and oxidative stress [25].

Importantly, there is a bidirectional cross-talk between cardiomyocyte and EAT, with adipokines such as adiponectin or leptin affecting the heart's function, and the heart influencing EAT biology through various signaling pathways [26]. For instance, an elevated ROS activates signaling pathways that stimulate epicardial adipocytes to secrete adiponectin with the subsequent inhibition of NADPH oxidase in cardiomyocytes, thereby restoring myocardial redox balance. More recently, Naryzhnaya et al. reported an increased ROS production in EAT adipocytes isolated from explants harvested from prediabetic and diabetic patients undergoing a coronary artery bypass. The ROS level in adipocytes was directly correlated with postprandial glycemia and inversely correlated with serum adiponectin [27]. Sacks et al. reported that at least one of the reasons for EAT producing more ROS than SAT is because of the higher mRNA expression (over three-fold) of $gp91^{phox}$ and $p47^{phox}$, NADPH components [28].

In a recent study, Chen et al. investigated the effects of EAT-derived leptin on myocardium in an experimental model of metabolic syndrome with diastolic dysfunction in rats, and also explored the underlying molecular mechanisms in H9C2 rat cardiomyoblasts exposed to either leptin or an EAT-conditioned medium. The authors reported that the EAT-derived leptin (but not serum leptin) elicited myocardial injury via the activation of the PKC/NADPH oxidase/ROS pathway, which was responsible for two cooperative pathomechanisms: mitochondrial oxidative stress with subsequent apoptosis and inflammation by promoting activator protein 1 nuclear translocation. Interestingly, the increased generation of NADP oxidase-derived ROS was the consequence of leptin-promoted interaction between $p\text{-}p47^{phox}$ and $gp91^{phox}$ in H9C2 cardiomyocytes [29].

Statins serve as the cornerstone therapy in primary and secondary cardiovascular and metabolic disease prevention and treatment due to their pleiotropic effects that extend far beyond the lipid-lowering action [12,30].

In a pioneering retrospective study conducted in patients subjected to a percutaneous coronary intervention, Park et al. showed that the thickness of EAT decreased to a greater extent in patients treated with atorvastatin compared to those treated with the simvastatin–ezetimibe combination [31].

More recently, Parisi et al. demonstrated that atorvastatin therapy was strongly associated between a decreased accumulation of EAT in patients with aortic stenosis subjected to cardiac surgery. Moreover, in vitro incubation with statin elicited a direct anti-inflammatory effect on EAT that was significantly higher compared to its effect on SAT [11]. A possible explanation could be the peculiar EAT structure (adipocytes with a smaller size and an important vascular supply), leading to an improved penetration of lipophilic statin at its level [32]. The efficacy of atorvastatin in diminishing EAT size was reported to be superior to that of either pravastatin or simvastatin [33].

EAT accumulation was associated with the persistence of atrial fibrillation or its recurrence after ablation [34,35]. In an elegant study, Natsui et al. comprehensively assessed the association between the EAT profile and the occurrence of postoperative atrial fibrillation (POAF) in patients who underwent cardiovascular surgery. They reported that the onset of POAF negatively correlated with the EAT adipocyte size and positively with TNF-α expression. Similarly, the mitochondrial respiratory capacity also showed a negative correlation with the size of EAT adipocytes and a positive one with adiponectin secretion [36].

Here, we have reported an increased ROS production of EAT and PVAT samples harvested from patients with HFmrEF with and without diabetes treated with two statins, atorvastatin or rosuvastatin. We have to acknowledge as limitations of this study the fact that we neither assessed the level of inflammatory cytokine (as the anti-inflammatory effect of statins may be responsible for lower oxidative stress) nor provided mechanistic evidence regarding the ROS sources in the adipose tissues harvested from these patients. Also, in the statin-treated group, there was a higher number of males compared to females; thus, the sex differences in ROS production could not be analyzed.

Conclusions

Statin therapy is associated with reduced oxidative stress in both epicardial and perivascular adipose tissue. The generation of reactive oxygen species from cardiac adipose tissues may play a role in the right ventricle enlargement. However, larger studies are required to assess whether the decrease in oxidative stress related to the long-term statin treatment represents a direct effect or indirect consequence of their anti-inflammatory effect.

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Data Availability Statement: Data will be made available by the corresponding author upon reasonable request.

Conflicts of Interest: The authors declare no conflicts of interest.

References

1. Villasante Fricke, A.C.; Iacobellis, G. Epicardial Adipose Tissue: Clinical Biomarker of Cardio-Metabolic Risk. *Int. J. Mol. Sci.* **2019**, *20*, 5989. [CrossRef] [PubMed]
2. Cherian, S.; Lopaschuk, G.D.; Carvalho, E. Cellular cross-talk between epicardial adipose tissue and myocardium in relation to the pathogenesis of cardiovascular disease. *Am. J. Physiol. Endocrinol. Metab.* **2012**, *303*, E937–E949. [CrossRef] [PubMed]
3. Antonopoulos, A.S.; Antoniades, C. The role of epicardial adipose tissue in cardiac biology: Classic concepts and emerging roles. *J. Physiol.* **2017**, *595*, 3907–3917. [CrossRef] [PubMed]
4. Vyas, V.; Blythe, H.; Wood, E.G.; Sandhar, B.; Sarker, S.J.; Balmforth, D.; Ambekar, S.G.; Yap, J.; Edmondson, S.J.; Di Salvo, C.; et al. Obesity and diabetes are major risk factors for epicardial adipose tissue inflammation. *JCI Insight* **2021**, *6*, e145495. [CrossRef] [PubMed]
5. Packer, M. Epicardial Adipose Tissue May Mediate Deleterious Effects of Obesity and Inflammation on the Myocardium. *J. Am. Coll. Cardiol.* **2018**, *71*, 2360–2372. [CrossRef] [PubMed]
6. Rossi, V.A.; Gruebler, M.; Monzo, L.; Galluzzo, A.; Beltrami, M. The Different Pathways of Epicardial Adipose Tissue across the Heart Failure Phenotypes: From Pathophysiology to Therapeutic Target. *Int. J. Mol. Sci.* **2023**, *24*, 6838. [CrossRef] [PubMed]
7. Iacobellis, G. Epicardial adipose tissue in contemporary cardiology. *Nat. Rev. Cardiol.* **2022**, *19*, 593–606. [CrossRef] [PubMed]
8. Braescu, L.; Gaspar, M.; Buriman, D.; Aburel, O.M.; Merce, A.P.; Bratosin, F.; Aleksandrovich, K.S.; Alambaram, S.; Mornos, C. The Role and Implications of Epicardial Fat in Coronary Atherosclerotic Disease. *J. Clin. Med.* **2022**, *11*, 4718. [CrossRef] [PubMed]
9. German, C.A.; Liao, J.K. Understanding the molecular mechanisms of statin pleiotropic effects. *Arch. Toxicol.* **2023**, *97*, 1529–1545. [CrossRef]
10. Alexopoulos, N.; Melek, B.H.; Arepalli, C.D.; Hartlage, G.R.; Chen, Z.; Kim, S.; Stillman, A.E.; Raggi, P. Effect of intensive versus moderate lipid-lowering therapy on epicardial adipose tissue in hyperlipidemic post-menopausal women: A substudy of the BELLES trial (Beyond Endorsed Lipid Lowering with EBT Scanning). *J. Am. Coll. Cardiol.* **2013**, *61*, 1956–1961. [CrossRef]
11. Parisi, V.; Petraglia, L.; D'Esposito, V.; Cabaro, S.; Rengo, G.; Caruso, A.; Grimaldi, M.G.; Baldascino, F.; De Bellis, A.; Vitale, D.; et al. Statin therapy modulates thickness and inflammatory profile of human epicardial adipose tissue. *Int. J. Cardiol.* **2019**, *274*, 326–330. [CrossRef] [PubMed]
12. Toia, P.; La Grutta, L.; Vitabile, S.; Punzo, B.; Cavaliere, C.; Militello, C.; Rundo, L.; Matranga, D.; Filorizzo, C.; Maffei, E.; et al. Epicardial Adipose Tissue Changes during Statin Administration in Relation to the Body Mass Index: A Longitudinal Cardiac CT Study. *Appl. Sci.* **2023**, *13*, 10709. [CrossRef]
13. Chang, L.; Garcia-Barrio, M.T.; Chen, Y.E. Perivascular Adipose Tissue Regulates Vascular Function by Targeting Vascular Smooth Muscle Cells. *Arterioscler. Thromb. Vasc. Biol.* **2020**, *40*, 1094–1109. [CrossRef] [PubMed]
14. Sturza, A.; Duicu, O.M.; Vaduva, A.; Dănilă, M.D.; Noveanu, L.; Varró, A.; Muntean, D.M. Monoamine oxidases are novel sources of cardiovascular oxidative stress in experimental diabetes. *Can. J. Physiol. Pharmacol.* **2015**, *93*, 555–561. [CrossRef]
15. Thomas, L.; Muraru, D.; Popescu, B.A.; Sitges, M.; Rosca, M.; Pedrizzetti, G.; Henein, M.Y.; Donal, E.; Badano, L.P. Evaluation of Left Atrial Size and Function: Relevance for Clinical Practice. *J. Am. Soc. Echocardiogr.* **2020**, *33*, 934–952. [CrossRef]
16. Mitchell, C.; Rahko, P.S.; Blauwet, L.A.; Canaday, B.; Finstuen, J.A.; Foster, M.C.; Horton, K.; Ogunyankin, K.O.; Palma, R.A.; Velazquez, E.J. Guidelines for Performing a Comprehensive Transthoracic Echocardiographic Examination in Adults: Recommendations from the American Society of Echocardiography. *J. Am. Soc. Echocardiogr.* **2019**, *32*, 1–64. [CrossRef] [PubMed]
17. Nagueh, S.F.; Smiseth, O.A.; Appleton, C.P.; Byrd, B.F., 3rd; Dokainish, H.; Edvardsen, T.; Flachskampf, F.A.; Gillebert, T.C.; Klein, A.L.; Lancellotti, P.; et al. Recommendations for the Evaluation of Left Ventricular Diastolic Function by Echocardiography: An Update from the American Society of Echocardiography and the European Association of Cardiovascular Imaging. *Eur. Heart J. Cardiovasc. Imaging* **2016**, *17*, 1321–1360. [CrossRef]
18. Mornos, C.; Cozma, D.; Rusinaru, D.; Ionac, A.; Maximov, D.; Petrescu, L.; Dragulescu, S.I. A novel index combining diastolic and systolic Tissue Doppler parameters for the non-invasive assessment of left ventricular end-diastolic pressure. *Int. J. Cardiol.* **2009**, *136*, 120–129. [CrossRef]
19. Rafeh, R.; Viveiros, A.; Oudit, G.Y.; El-Yazbi, A.F. Targeting perivascular and epicardial adipose tissue inflammation: Therapeutic opportunities for cardiovascular disease. *Clin. Sci.* **2020**, *134*, 827–851. [CrossRef]
20. Karampetsou, N.; Alexopoulos, L.; Minia, A.; Pliaka, V.; Tsolakos, N.; Kontzoglou, K.; Perrea, D.N.; Patapis, P. Epicardial Adipose Tissue as an Independent Cardiometabolic Risk Factor for Coronary Artery Disease. *Cureus* **2022**, *14*, e25578. [CrossRef]
21. Wang, Q.; Chi, J.; Wang, C.; Yang, Y.; Tian, R.; Chen, X. Epicardial Adipose Tissue in Patients with Coronary Artery Disease: A Meta-Analysis. *J. Cardiovasc. Dev. Dis.* **2022**, *9*, 253. [CrossRef] [PubMed]

22. Cho, D.H.; Park, S.M. Epicardial Adipose Tissue and Heart Failure, Friend or Foe? *Diabetes Metab. J.* **2024**, *48*, 373–384. [CrossRef] [PubMed]
23. Salgado-Somoza, A.; Teijeira-Fernández, E.; Fernández, A.L.; González-Juanatey, J.R.; Eiras, S. Proteomic analysis of epicardial and subcutaneous adipose tissue reveals differences in proteins involved in oxidative stress. *Am. J. Physiol. Heart Circ. Physiol.* **2010**, *299*, H202–H209. [CrossRef] [PubMed]
24. Agra, R.M.; Teijeira-Fernández, E.; Pascual-Figal, D.; Sánchez-Más, J.; Fernández-Trasancos, A.; González-Juanatey, J.R.; Eiras, S. Adiponectin and p53 mRNA in epicardial and subcutaneous fat from heart failure patients. *Eur. J. Clin. Investig.* **2014**, *44*, 29–37. [CrossRef] [PubMed]
25. Krstic, J.; Reinisch, I.; Schupp, M.; Schulz, T.J.; Prokesch, A. p53 Functions in Adipose Tissue Metabolism and Homeostasis. *Int. J. Mol. Sci.* **2018**, *19*, 2622. [CrossRef] [PubMed]
26. Lodewijks, F.; McKinsey, T.A.; Robinson, E.L. Fat-to-heart crosstalk in health and disease. *Front. Genet.* **2023**, *14*, 990155. [CrossRef]
27. Naryzhnaya, N.V.; Koshelskaya, O.A.; Kologrivova, I.V.; Suslova, T.E.; Kharitonova, O.A.; Andreev, S.L.; Gorbunov, A.S.; Kurbatov, B.K.; Boshchenko, A.A. Production of Reactive Oxygen Species by Epicardial Adipocytes Is Associated with an Increase in Postprandial Glycemia, Postprandial Insulin, and a Decrease in Serum Adiponectin in Patients with Severe Coronary Atherosclerosis. *Biomedicines* **2022**, *10*, 2054. [CrossRef]
28. Sacks, H.S.; Fain, J.N.; Cheema, P.; Bahouth, S.W.; Garrett, E.; Wolf, R.Y.; Wolford, D.; Samaha, J. Depot-specific overexpression of proinflammatory, redox, endothelial cell, and angiogenic genes in epicardial fat adjacent to severe stable coronary atherosclerosis. *Metab. Syndr. Relat. Disord.* **2011**, *9*, 433–439. [CrossRef]
29. Chen, H.; Liu, L.; Li, M.; Zhu, D.; Tian, G. Epicardial Adipose Tissue-Derived Leptin Promotes Myocardial Injury in Metabolic Syndrome Rats Through PKC/NADPH Oxidase/ROS Pathway. *J. Am. Heart Assoc.* **2023**, *12*, e029415. [CrossRef]
30. Muntean, D.M.; Thompson, P.D.; Catapano, A.L.; Stasiolek, M.; Fabis, J.; Muntner, P.; Serban, M.C.; Banach, M. Statin-associated myopathy and the quest for biomarkers: Can we effectively predict statin-associated muscle symptoms? *Drug Discov. Today* **2017**, *22*, 85–96. [CrossRef]
31. Park, J.H.; Park, Y.S.; Kim, Y.J.; Lee, I.S.; Kim, J.H.; Lee, J.H.; Choi, S.W.; Jeong, J.O.; Seong, I.W. Effects of statins on the epicardial fat thickness in patients with coronary artery stenosis underwent percutaneous coronary intervention: Comparison of atorvastatin with simvastatin/ezetimibe. *J. Cardiovasc. Ultrasound.* **2010**, *18*, 121–126. [CrossRef] [PubMed]
32. Beltowski, J. Epicardial adipose tissue: The new target for statin therapy. *Int. J. Cardiol.* **2019**, *274*, 353–354. [CrossRef] [PubMed]
33. Soucek, F.; Covassin, N.; Singh, P.; Ruzek, L.; Kara, T.; Suleiman, M.; Lerman, A.; Koestler, C.; Friedman, P.A.; Lopez-Jimenez, F.; et al. Effects of Atorvastatin (80 mg) Therapy on Quantity of Epicardial Adipose Tissue in Patients Undergoing Pulmonary Vein Isolation for Atrial Fibrillation. *Am. J. Cardiol.* **2015**, *116*, 1443–1446. [CrossRef]
34. Gaeta, M.; Bandera, F.; Tassinari, F.; Capasso, L.; Cargnelutti, M.; Pelissero, G.; Malavazos, A.E.; Ricci, C. Is epicardial fat depot associated with atrial fibrillation? A systematic review and meta-analysis. *Europace* **2017**, *19*, 747–752. [CrossRef]
35. Zain, S.; Shamshad, T.; Kabir, A.; Khan, A.A. Epicardial Adipose Tissue and Development of Atrial Fibrillation (AFIB) and Heart Failure With Preserved Ejection Fraction (HFpEF). *Cureus* **2023**, *15*, e46153. [CrossRef] [PubMed]
36. Natsui, H.; Watanabe, M.; Yokota, T.; Tsuneta, S.; Fumoto, Y.; Handa, H.; Shouji, M.; Koya, J.; Nishino, K.; Tatsuta, D.; et al. Influence of epicardial adipose tissue inflammation and adipocyte size on postoperative atrial fibrillation in patients after cardiovascular surgery. *Physiol. Rep.* **2024**, *12*, e15957. [CrossRef]