**Streszczenie w języku angielskim.**

Epicardial adipose tissue and the risk of cardiovascular diseases: pathophysiological mechanisms and clinical implications.

Introduction:

Cardiovascular diseases remain one of the leading causes of death globally. Classic and potentially modifiable cardiovascular risk factors include smoking, obesity, low physical activity, lipid disorders, and hypertension. However, increasing attention is also being paid to non-classical cardiovascular risk factors, such as adipose tissue surrounding internal organs.

The adipose tissue surrounding the heart is a subtype of visceral fat, the amount of which correlates with the severity of abdominal obesity and is independently associated with the risk of cardiovascular events. Like abdominal obesity, pericardial adipose tissue is a risk factor for the development of atherosclerotic cardiovascular diseases.

The classification of pericardial adipose tissue varies in the literature. For the purposes of this work, the most important is epicardial adipose tissue (EAT) and pericoronary adipose tissue (PCAT). EAT lies between the visceral layer of the pericardium and the myocardium, while PCAT is defined as fat surrounding the coronary vessels, regardless of its location, considering it as part of epicardial fat.

Pericardial adipose tissue is highly metabolically active, secreting numerous cytokines and chemokines in the vicinity of the heart muscle. Under healthy conditions, EAT and PCAT play a protective role, participating in metabolic, energetic, and anti-inflammatory processes. In pathophysiological conditions, such as in patients with diabetes or metabolic syndrome, these protective properties are replaced with processes promoting inflammation and the development of cardiovascular diseases, including coronary artery disease. Therefore, pericardial adipose tissue may be an interesting target for new cardiovascular therapies in the future.

Aim of the study:

The aim of this doctoral thesis was (i) to identify the relationship between extremely intense physical exertion and the quantity and inflammatory activity of EAT, (ii) to identify potential pathophysiological mechanisms at the level of gene expression in EAT that could be a target for future therapies, and (iii) to summarize the existing data on the role of EAT in the pathophysiology of cardiovascular diseases.

Results:

In the first study, the effect of extremely intense physical activity on the amount of EAT and the correlation between EAT and cardiovascular risk factors were evaluated. The study

included 30 healthy amateur ultramarathon runners and 9 volunteers leading a sedentary lifestyle. EAT surface area was assessed using magnetic resonance imaging at 4 locations: around the 3 main coronary arteries (left anterior descending branch, circumflex branch, right coronary artery) and on the surface of the right ventricle. Additionally, body composition, lipid profile, serum interleukin-6 concentration, and intima-media thickness in the carotid arteries were evaluated. The amount of EAT in the ultramarathon runners was significantly lower at all examined locations compared to the control group ($p < 0.001$). As expected, ultramarathon runners had a lower percentage of visceral fat and a better lipid profile than the control group ($p < 0.001$). However, no differences in intima-media thickness were observed. There was also no statistically significant difference in interleukin-6 levels between the groups, but the frequency of pathologically high interleukin-6 levels (defined as concentration > 1 pg/ml) was three times lower in the ultramarathon runners group than in the control group (17% vs. 56%, $p < 0.05$). Additionally, a positive correlation was found in the ultramarathon runner group between the surface area of EAT surrounding the left anterior descending branch, circumflex branch, and right ventricle and the percentage of total visceral fat, and between the amount of EAT around the circumflex branch and the concentration of LDL and non-HDL cholesterol ($p < 0.05$).

In the second study, gene expression in PCAT was compared between patients with advanced coronary artery disease and a control group. PCAT samples were obtained during aorto-coronary bypass surgery ($n = 21$, study group) or non-coronary cardiac surgery in patients with previously excluded coronary artery disease ($n = 19$, control group). Out of 67,528 transcripts, 1348 were identified as differentially expressed genes (DEGs). Among them, 416 (30.9%) showed overexpression, and 932 (69.1%) were classified as underexpressed compared to the control group. Among the genes showing increased expression were those encoding molecules with pro-inflammatory and pro-atherogenic activity, such as CXCL8, CXCL2, interleukin-6, selectin E, low-density lipoprotein receptor, and ADAMTS metalloproteinases. Genes encoding signalling proteins, enzymes, microRNAs, and various types of collagen were identified among the underexpressed genes.

Additionally, a group of "upstream regulators" associated with differentially expressed genes was distinguished. This term refers to any molecule that can affect the expression, transcription, or phosphorylation of another molecule. In this heterogeneous group, attention was drawn to genes encoding platelet-derived growth factor, high mobility group box 2 protein (HMGB2), and evolutionarily conserved signalling intermediate in toll pathway (ESCIT), which are considered pro-inflammatory and pro-atherosclerotic molecules. Moreover, the

Ingenuity Pathway Analysis (IPA) software used linked differentially expressed genes to entire networks of connections, canonical pathways, and networks. The activated pathways included the "coagulation system pathway" containing molecules known for their pro-atherosclerotic and pro-inflammatory nature (tissue factor pathway inhibitor, plasminogen activator, urokinase receptor, thrombomodulin).

The third work is a review article summarizing the current state of knowledge regarding the involvement of EAT in the pathogenesis of cardiovascular diseases, including coronary artery disease, heart failure, and atrial fibrillation. Attention is also drawn to the possible relationship between EAT and the course of COVID-19. Finally, EAT is presented as a potential therapeutic target in the prevention and treatment of cardiovascular diseases in the future.

Conclusions:

The first study demonstrated that extremely intense physical training may reduce cardiovascular risk by reducing the amount and pro-inflammatory activity of epicardial adipose tissue. However, further research is needed to confirm the relationships identified in this doctoral thesis.

The second study was one of the first to demonstrate altered gene expression not only in individual genes but in entire networks and pathways derived from PCAT, providing further evidence that the tissue under study is an active source of pro-atherosclerotic molecules that may accelerate the development of coronary artery disease.

The third work indicates an extensive interest in EAT in the recent years, suggesting its potential as a new therapeutic target in the prevention and treatment of cardiovascular diseases.

In summary, the series of studies presented demonstrate that pericardial adipose tissue is one of the key elements involved in the pathogenesis of cardiovascular diseases. However, further research at the molecular level is crucial to identify specific genes and proteins encoded by them whose modulation could change the unfavourable phenotype of EAT in patients with cardiovascular diseases. The negative impact of EAT on the circulatory system can be limited through non-pharmacological interventions, and with further development of gene expression studies, there is a chance to identify new therapeutic targets within EAT.