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New molecular and imaging prognostic markers in young patients with acute ST elevation myocardial infarction

PHD THESIS SUMMARY

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ABBREVIATIONS

ACS – acute coronary syndrome

AMI – acute myocardial infarction

AST – aspartate aminotransferase

AUC – Area Under the Curve

BNP – Natriuretic B peptide

BP – blood pressure CAD – coronary artery disease

CK-MB – creatine kinase MB

CRP – C reactive protein

EA – arterial elastance

EES – end-systolic ventricular elastance

ESP – end-systolic pressure

GCW – global contraction work

GWE – global work efficiency

GWI – global work index

GWW – global wasted work

HF – heart failure

HR – hazard ratio

IL-1 – interleukin 1

IL-1RA – IL-1 receptor antagonist

IL-6 – interleukin 6

LA – left atrium

LAD – left anterior descending artery

LAEF – left atrial emptying fraction

LAEV – left atrial emptying volume

LAS-cd 3D – left atrial conduction strain

LAS-ct 3D – left atrial contraction strain

LAS-r 3D – left atrial reservoir strain

LAVI – left atrial volume index

LAVmax3D – 3D maximal left atrial volume

LAVmin3D – 3D minimal left atrial volume

LAVpreaA3D – 3D pre-atrial contraction LA volume

LCX – circumflex artery

LV – left ventricle

LVEDV – end-diastolic LV volume

LVESV – end-systolic LV volume

LVFE – left ventricular ejection fraction

LVGLS – LV global longitudinal strain

MACE – major adverse cardiac events

miRNA – microARN

PAPs – peak systolic pulmonary artery pressure

PASP – peak systolic pulmonary artery pressure

PCI – percutaneous coronary intervention

RA – right atrium

RCA – right coronary artery

ROC – Receiver Operating Characteristics

RV – right ventricle

STEMI – ST elevation myocardial infarction

SV – stroke volume

TAPSE – tricuspid plane anterior systolic motion

TNFα – α tumoral necrosis factor

VAC – ventriculo-arterial coupling

Introduction

Despite continuous advances in diagnostic and therapeutic methods, acute myocardial infarction (STEMI) remains a leading cause of morbidity and mortality worldwide.

Although myocardial infarction was considered a disease occurring in older adults, in recent years its prevalence in the young population has increased. Patients with an acute coronary syndrome at a young age are at high risk for other future major cardiovascular events. Therefore, close follow-up and preventive strategies are of great importance in this patient group. So far, there have been tested many potential prognostic markers (clinical, echocardiographic, molecular) as predictors of the occurrence of MACE after acute myocardial infarction.

This research aims to identify new molecular (cytokines and miRNAs) and imaging (echocardiographic – myocardial work indices, 3D left ventriculo-arterial coupling and 3D left atrial strain) prognostic markers in young patients with a first acute ST elevation myocardial infarction determined by atherosclerotic plaque rupture.

I. CURRENT KNOWLEDGE

CHAPTER 1. ACUTE ST ELEVATION MYOCARDIAL INFARCTION – CHARACTERISTICS IN YOUNG POPULATION

1.1. General data

Acute ST elevation myocardial infarction is defined as a cardiomyocyte death due to a prolonged ischaemia resulting from an acute imbalance between oxygen supply and demand [1].

In recent years its prevalence in the young population has increased [2]. So far, there has been no standard definition of “young” age in patients with STEMI. Previous studies used different age thresholds, varying from 30 to 55 years [2, 3]. Myocardial infarction has

a significant socioeconomic burden, determined by the long term morbidity and mortality, especially in young patients[4]. Patients with an acute coronary syndrome at a young age are at high risk for other future major cardiovascular events [5].

Previous studies showed certain differences between younger and older patients with STEMI, regarding epidemiology, cardiovascular risk factors, clinical characteristics, angiographic characteristics and prognosis [4].

1.2. Characteristics of young patients with STEMI

Most studies reported a higher incidence of STEMI in young men (between 64.7 and 94.8%), compared with young women [4]. There are two main causes of infarction in young patients: atherosclerotic, and non-atherosclerotic [6]. Atherosclerosis is the most common cause of acute myocardial infarction, 60-65% of the STEMI cases being determined by atherosclerotic plaque rupture [7]. The main cardiovascular risk factors in the young are represented by family history, smoking, dyslipidaemia, and obesity [8]. This research will focus on atherosclerosis.

The diagnosis of acute myocardial infarction is made in accordance with the consensus document ”Fourth Universal Definition of Myocardial Infarction” issued by the European Society of Cardiology in 2018, regardless of age [9]. Many young STEMI patients present with no medical history of chest pain or cardiovascular disease, compared to older patients [10]. Chest pain is either absent or atypical, especially in women [10]. Single coronary artery disease that affects the left anterior descending artery is more prevalent among young patients with acute myocardial infarction [4].

The management of STEMI patients is the same, regardless of age, according to clinical practice guidelines [11]. Besides medical treatment, risk factors management and lifestyle changes are equally important.

Young patients with STEMI represent a subgroup of patients with different risk factor profile, with atypical symptoms at presentation, and different prognosis, compared to older counterparts [10]. They have favourable short-term evolution, however long-term outcome is unfavourable, compared to the general age matched population, especially in those with systolic dysfunction [10].

Considering its increasing prevalence in recent years, especially in the young, close follow up and new preventive strategies for lowering cardiovascular risk are of paramount importance in this patient group [12].

CHAPETER 2. NEW PROGNOSTIC MARKERS IN STEMI

Early identification of patients with potential unfavourable outcome after AMI by measuring various markers could contribute to a more accurate risk stratification [13].

2.1. New echocardiographic prognostic markers in STEMI

Echocardiography has an important role in risk stratification and prognosis assessment after acute myocardial infarction [14].

2.1.1. Myocardial work indices

Left ventricular ejection fraction and global longitudinal strain are the most used echocardiographic methods for the assessment of left ventricular function. They are both load dependent (LVGLS to a lesser extent than LVEF), therefore they are influenced by high preload and afterload [15].

In recent years, an alternative method of assessing ventricular function has emerged: myocardial work. This non-invasive method introduced by Russel et al. is based on a standardised LV pressure curve derived from LV strain analysis, adjusted to arterial pressure [16]. The impairment of myocardial work is the expression of the alteration of the energy metabolism that occurs in the remodelled myocardium [17]. These new markers could detect subclinical myocardial changes, having an incremental diagnostic and prognostic potential over the classical parameters of LV function [17].

2.1.2. Left ventriculo-arterial coupling

Left ventriculo-arterial coupling (VAC) is a key determinant of cardiovascular performance, calculated as the ratio between arterial elastance (EA) and left ventricular end-systolic elastance (EES) [18]. Experimental models showed that maximal mechanical work is achieved when VAC is close to 1, while maximal ventricular efficiency occurs when the ratio approaches 0.5 [18, 19].

In case of left ventricular dysfunction or heart failure, VAC is suboptimal, with EES being decreased secondary to pump dysfunction, while EA is increased [20]. There is even

scientific evidence that VAC alterations occur prior to pump dysfunction [20]. A non-invasive measurement of VAC could provide a comprehensive assessment of ventricular performance and may have incremental prognostic value over left ventricular ejection fraction in patients with CAD [21].

2.1.2. Left atrial function

The importance of assessing left atrial function derives from its contribution to left ventricular filling through its three components: reservoir, conduit and pump. 3D examination has the advantage of an increased accuracy compared to 2D echocardiography, eliminating geometric assumptions. Left atrial structure and function have been less studied in STEMI. Left atrial enlargement is known as an unfavourable prognostic marker both in ischemic heart disease and other cardiovascular pathologies [22], with left atrial volume being an independent predictor for MACE in patients with atrial fibrillation, stroke or AMI [23].

Left atrial strain is considered a more sensitive and earlier marker of left atrial dysfunction, compared with LA volumes, strain impairment occurring before atrial dilatation. 3D speckle tracking examination could detect early functional LA remodelling before the occurrence of anatomical changes, thus bringing additional prognostic information to conventional LA parameters [23].

2.2. New molecular markers in STEMI

Besides echocardiographic parameters, biomarkers like CK-MB, troponin, NTproBNP, CRP, and fibrinogen also have a very important role in risk stratification after AMI [13]. In recent years studies have focused their attention on two new distinct categories of biomarkers: cytokines and miRNAs.

2.2.1. Inflammatory markers – Cytokines

Vascular inflammation plays an important role in the initiation and progression of atherosclerosis, but also in the resolution and healing that occurs after AMI [24]. Among vascular inflammation biomarkers, cytokines and some adipokines were associated with the occurrence of MACE after myocardial infarction [25].

A variety of cytokines are released early after the onset of myocardial ischemia [26]. The main cytokine that mediates the proinflammatory response in STEMI is IL-1. IL-1RA is a competitive inhibitor of IL-1, being released as an acute phase reactant, modulating the

inflammatory response [27]. Ischemia triggers IL-1RA synthesis in cardiomyocytes [28]. IL-1RA correlated positively in various studies with the severity of the inflammatory process [27], proving to be a sensitive diagnostic and prognostic marker in patients with ACS [29].

IL-6, another cytokine released after AMI, proved to have both pro-inflammatory and anti-inflammatory effects [30]. This cytokine is involved in vascular inflammation, the initiation and progression of atherosclerosis, and degradation of fibrous cap contributing to plaque instability [31]. IL-6 propagates inflammation in patients with AMI, and its levels at admission are associated with infarct size and cardiac function, making it a predictor of in-hospital prognosis [32].

Resistin is a pro-inflammatory adipocytokine secreted predominantly by macrophages and adipocytes, with an important role in the pathogenesis and development of atherosclerosis [33]. It upregulates the expression of other pro-inflammatory cytokines, including TNF-α, IL-6, IL-1β, thus promoting the inflammatory process [34]. Resistin levels are high in patients with ACS, its levels increasing early at 3–6 hours after onset making it a potentially useful diagnostic marker [35].

There has been increasing evidence that inflammation following AMI is a complex process, thus assessing multiple cytokines as predictors after STEMI may be more beneficial than focusing on a single marker. [36].

2.2.2. MicroRNAs

Micro RNAs (miRNAs) are endogenously produced small (19-24 nucleotides) RNA sequences that regulate gene expression at the post-transcriptional level, thus affecting a variety of cell processes [37].

miRNAs appear to have a role in the development and progression of ischemic heart disease at multiple levels (angiogenesis, atherogenesis, lipoprotein homeostasis, platelet activation), and also they play a critical role in the STEMI pathophysiological process as well [38]. Many miRNAs proved their prognostic potential after STEMI being involved in atherosclerosis and AMI pathophysiology. MiR-233-3p is almost exclusively of platelet or megakaryocyte origin; its biological activity is related to aggregation and granule secretion [39]. MiR-142-3p plays a role in various inflammatory diseases, such as atherosclerosis[40]. miR-146a-5p is expressed in vascular endothelial cells, smooth muscle cells, and macrophages, and exhibits a protective effect against cardiac ischaemia/hypoxia-induced apoptosis; it is also involved in ischemic heart disease[41]. miR-125a-5p regulates

macrophage activation, lipid metabolism, and the regulation of atherogenesis, essential processes in CAD [42]. miR-486-5p is a muscle-enriched miRNA, found to be upregulated in patients with ACS, with diagnostic potential in STEMI[43]. miR155-5p modulates immune responses via cell differentiation and inflammatory cytokine secretion [44].

miRNAs have emerged as a key epigenetic mechanism in cardiovascular diseases, having both diagnostic and prognostic potential in STEMI [45].

II. PERSONAL CONTRIBUTIONS

CHAPTER 3. WORK HYPOTHESIS AND MAIN OBJECTIVES

The identification of patients at increased risk of developing adverse events after STEMI is important in order to establish more aggressive prevention and treatment strategies, with the aim of improving their prognosis.

Taking this into consideration, this work aims to identify **new prognostic markers (echocardiographic and molecular) for the occurrence of MACE after STEMI in young patients**

The general objectives of the PhD thesis are:

1. Identification of new prognostic echocardiographic markers in young patients with STEMI (myocardial work indices, 3D left ventriculo-arterial coupling and 3D left atrial function);
2. Assessment of the role of cytokines as prognostic markers after STEMI in young patients;
3. Assessment of role of miRNAs as prognostic markers after STEMI in young patients.

CHAPTER 4. RESEARCH METHODOLOGY

4.1. Study population

This prospective study was conducted in the Department of Cardiology of Clinical Emergency Hospital of Bucharest, between 2019 and 2021.

We enrolled 100 young patients (aged between 18-51) with a first STEMI admitted to our hospital and treated by primary PCI (RO STEMI National Programme). 7 patients

were excluded due to poor acoustic window, and 9 were lost during follow-up, leaving a final study group of 84 patients. We also chose 28 age-matched controls (healthy volunteers). Therefore, we analysed 112 subjects in this research.

Patient enrolment, blood harvesting, clinical/echocardiographic assessment, and cytokines quantification were all performed in The Department of Cardiology, Clinical Emergency Hospital of Bucharest. Only miRNA determination was performed at the Institute of Cellular Biology and Pathology "Nicolae Simionescu", Lipidomics Department, as a part of the joint research project "EPITERAMI".

The research was approved by the Medical Ethics Committee of the Bucharest Emergency Clinical Hospital. All patients signed an informed consent at enrolment.

4.2. Inclusion criteria

We enrolled in this study young patients (18-51 years old), with a first STEMI treated by primary PCI. All patients received optimal medical therapy according to current clinical practice guidelines [11].

4.3. Exclusion criteria

Patients with previous myocardial infarction or cardiac surgery, structural cardiac disease of severe pre-existent valvular heart disease, recent surgery or trauma (within 6 months), severe respiratory/hepatic/renal failure, active malignancy or autoimmune diseases, active infections, pregnancy, patients with addictions, poor compliance, or those who refused to sign the informed consent were excluded.

4.4. Study protocol

At enrolment (T0), demographic data, clinical parameters, laboratory analyses, angiographic data, standard and advanced echocardiographic parameters were recorded for each patient.

Blood Sample Collection – whole venous blood samples were harvested, processed and then stored at -80°C for further analysis of the molecular markers.

Echocardiography was performed in all included patients using a GE VIVID E9 ultrasound system. The recordings and measurements were performed in accordance with current echocardiographic guidelines[46, 47].

Echocardiographic parameters measured in this study:

– LV, RV, LA, RA dimensions; LV and LA volumes 2D and 3D;

– LVGLS 2D, left ventricular mechanical dispersion;

– LV diastolic function: E, A waves, E/A ratio, septal e', lateral e', E/e' (classification of diastolic dysfunction was performed according to dedicated guidelines [48]);

– Assessment of valvular heart disease;

– RV function: TASPE, S' wave, PASP;

– Evaluation of mechanical complications/pericardial disease.

New echocardiographic parameters

- Myocardial work indices derived from 2D speckle tracking echocardiography;
- 3D left ventriculo-arterial coupling;
- 3D left atrial function.

Molecular markers

- ❖ Quantification of plasma cytokine levels using ELISA method;
- ❖ miRNA isolation and quantification by RT-PCR.

Patients follow up – T6, T12

Patients were followed up for up to one year after STEMI.

At 6 months (T6), we performed a more detailed follow-up: clinical examination, EKG, standard and advanced echocardiography, blood harvesting for molecular markers.

At 12 months (T12), a telephone follow up was performed due to COVID 19 pandemic restrictions.

During this one year of follow-up, we assessed **the occurrence of MACE**. In this study, MACE was defined as death from cardiovascular causes, heart failure requiring hospital admission, or repeat PCI/CABG due to ischaemia/infarction (in concordance with previous trials) [49].

4.5. **Statistical analysis**

Statistical analysis was performed using SPSS software (IBM SPSS Statistics v.22.0, IBM Corp., Armonk, NY, USA) and GraphPad software (GraphPad Prism 9.0.0, San Diego, CA, USA). We presented the categorical data as percentages, and the continuous variables as means ± standard deviation. A Kolmogorov–Smirnov test, and Mann–Whitney U-test were used for normal and non-normal distribution of data, respectively. Student T test and X2 tests were used to compare continuous and categorical variables. An ROC analysis (receiver operating curve) was used to determine the AUC (area under the curve), representing the predictive power of the tested parameters. We also determined cut-off values for the significant variables using the Youden index. To determine the predictors of MACE, we performed a Cox univariate regression, further incorporating the statistically significant variables in a multivariate analysis. To further test the added values of miRNAs over myocardial work indices, we constructed prediction models and compared their statistical power using the C statistic, and Akaike information criterion (AIC). Kaplan-Meier curves was also performed to represent the differences between patient groups and the occurrence of MACE. P under 0.05 was considered statistically significant.

CHAPTER 5. FIRST STUDY: NEW ECHOCARDIOGRAPHIC PROGNOSTIC MARKERS AFTER STEMI IN YOUNG PATIENTS

5.1. Echocardiographic sub-study nr. 1: Prognostic role of myocardial work indices in young patients with STEMI

5.1.1. Introduction

To determine the predictive value of myocardial work indices on the occurrence of MACE after STEMI in young patients.

Specific objectives

- Determination of myocardial work indices (GWI, GCW, GWW, GWE) in young STEMI patients and in a control group;
- Assessing the prognostic potential of these new parameters;
- Comparison with routinely used echocardiographic parameters.

5.1.2. Patients and methods

– 84 young patients with a first STEMI + control group (28 young healthy subjects)

Echocardiography – All patients underwent standard transthoracic echocardiography.

Myocardial work indices analysis

The quantification of non-invasive myocardial work was performed using a commercially available software package (Echopac V.202, GE). Arterial blood pressure was measured with a brachial cuff sphygmomanometer before image acquisition.

The following parameters were obtained from this analysis:

- Global work index (GWI) – representing the total work performed by the left ventricle in a single cardiac cycle;
- Global constructive work (GCW) – the myocardial work performed during ventricular systole, contributing to the pump function;
- Global wasted work (GWW) – the negative myocardial work performed during the lengthening of a myocardial segment in systole or during shortening in isovolumic relaxation;
- Global work efficiency (GWE) – calculated as the ratio GCW/(GCW + GWW); it is an estimation of mechanical performance and of the energy used by the LV taking into account the filling pressures.

5.1.3. Results

5.1.3.1. Characteristics of the study population

Most of the patients included in this study were male (80%), with a mean age of 44 years old. The most frequent cardiovascular risk factors were smoking (36%), and dislipidemia (40%). Most of the patients had Killip class I at admission (89,3%). Regarding angiographic data – most of the patients had anterior infarction (48,8%); there were no significant differences regarding door to balloon time between groups (P = 0.692).

5.1.3.2. Echocardiographic parameters

The mean ejection fraction in the study group was 42.19%. Patients with MACE at follow-up had lower 2D LVEF, more impaired LVGLS, and higher 2D and 3D left ventricular volumes. LV mechanical dispersion was higher in the MACE group.

5.1.3.3. Myocardial work indices

Compared with control group, GWI, GCW and GWE are lower in STEMI patients, while GWW is higher. Patients in the MACE group had significantly lower values of GWI, GCW, and GCW at admission (GWI: 737.33 ± 199.01 vs. 1169.12 ± 274.17, P < 0.0001, and GCW 1037.83 ± 259.87 vs. 1473.54 ± 287.28, P < 0.0001, GWE: 76.33 ± 5.28 vs. 87.59 ± 5.96, P < 0.0001), and higher GWW values (255.91 ± 103.22 vs. 177.14 ± 93.13, P = 0.009), compared with non MACE group

5.1.3.4. Evolution of echocardiographic parameters in time

GWI, GCW and GWW appear to improve over time (at 6 months follow up), but no significant change is observed in GWW. In MACE group compared to non-MACE group, the improvement of myocardial indices was statistically significant. GWW has a decreasing trend over time, but without statistical significance.

5.1.3.5. Association between clinical, biological, echocardiographic parameters, and MACE at one year follow up

We divided the study cohort in two groups (MACE and non-MACE) considering the presence or absence of MACE at the one-year follow-up. MACE occurred in 18% (13 out of 84) of the studied patients: 2 (15%) died, 7 (53%) developed heart failure requiring hospital admission, and 4 (30%) had a new acute coronary event.

ROC analysis proved that LVGLS has a better predictive power for MACE in young STEMI patients, compared with 2D LVEF, 3D LVEF, and LV mechanical dispersion.

5.1.3.6. Myocardial work indices and MACE

The four myocardial work indices had good prediction potential for MACE, with an AUC greater than 0.7 in the ROC curve analysis (Figure 5.1). Between them GWI and GWE proved to have the best predictive value for MACE. For each of them we also determined cut off values. According to COX univariate regression, all indices were independent predictors for MACE at one year follow-up. In multivariate regression only GWI and GWE remained independent predictors for MACE. Kaplan-Meier analysis proved that patients with lower values of GWI and GWE had higher probability of MACE at follow up.

5.1.4. Discussions and conclusions

In our study we demonstrated that patients with lower values of GWI, GWE and GCW had more important myocardial damage. Patients with myocardial indices below the cut off values had higher CK-MB levels, higher left ventricular volumes, lower left ventricular ejection fraction, and more impaired LVGLS, all being an expression of a more severe cardiac injury. Also, lower values of these markers proved to be associated with worse long term outcome in the study group.

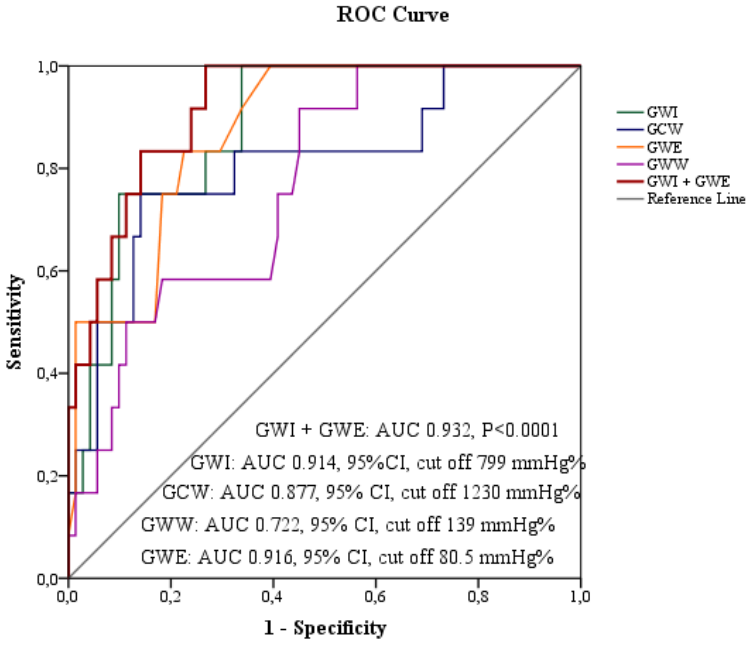

Fig.5.1. ROC curve analysis for baseline values of myocardial work indices

Regarding literature data, 2 other studies showed promising results. Lustosa et al., who tested the long-term prognostic value of GWE in STEMI patients, found that lower GWE values in the acute phase were associated with worse long-term survival [50]. Butcher et al. concluded that lower GWI values were independently associated with increased all-cause mortality at 6 months of follow-up [51]. In our study, cohort of young STEMI patients treated by primary PCI, we found that myocardial work parameters measured noninvasively were independent predictors of MACE at one-year follow-up. Between the 4 tested parameters, GWI and GWE proved to be the best predictors for MACE. Supporting our findings, Steele et al. assessed the prognostic value of myocardial word in 239 patients with STEMI and low ejection fraction; they observed that GWI was associated with all-cause

mortality at 6 months after AMI, proving its incremental prognostic values over LVEF and LVGLS[51].

5.2. Echocardiographic sub-study nr. 2: The prognostic role of ventriculo-arterial coupling in young patients with STEMI

5.2.1. Introduction

Study purpose - To determine the prognostic role of left ventriculo-arterial coupling (determined by 3D echocardiography) in young patients with STEMI.

Specific objectives

- Assessing the prognostic role of EA, EES, VAC in MACE prediction after AMI;
- Comparison of the prognostic value of the new markers with that of the routinely used echocardiographic parameters.

5.2.2. Patients and methods

– 84 young patients with a first STEMI + control group (28 young healthy subjects).

Echocardiography – All patients underwent standard transthoracic echocardiography.

3D left ventriculo-arterial coupling

VAC was calculated as the ratio between arterial elastance (ES) and end-systolic left ventricular elastance (EES). VAC = EA/EES. End-systolic pressure (ESP) is determined as 0,9 x systolic arterial blood pressure. Ventricular volumes were determined by 3D echocardiography. ENd (est) = estimated normalised ventricular elastance at the onset of ejection.

EA = ESP/SV = (0.9 x systolic BP)/3D SV.

EES = [diastolic BP - (ENd(est) x Ps x 0.9)]/(ENd(est) x 3D SV) = (0.9 x systolic BP)/VTSVS 3D.

ENd (est) = 0.0275 - 0.165 x LVEF 3D + 0.3656 x (diastolic BP/(systolic BP x 0.9)) + 0.515 x End (avg).

Simplified formula: 3D VAC = 3D LVEDV/3D SV.

5.2.3. Results

5.2.3.1. EA, EES, and VAC

In the control group compared with study group (STEMI patients), we obtained lower values for VAC (0.8 ± 0.23 control vs. 1.37 ± 0.48 STEMI) and EES (2.3 ± 0,7 control vs.

1.85 ± 0.57 STEMI), but almost similar values for EA (2.1 ± 0.6 control vs. 2.38 ± 0.59 STEMI). VAC lower than 1 in the control group means an optimal coupling between LV and arterial system, as expected, considering the fact that these are healthy subjects.

We divided the study population in 3, according to the degree of LV systolic dysfunction, as follows: preserved EF (> 50%), mildly reduced EF (FE 40-49%), and reduced EF (< 40%) – in accordance with the classification of heart failure from ESC guidelines [52]. We can observe that with the decrease in LV systolic function, GWI, GCW, GWE, and EE decrease progressively while GWW, VAC, and EA increase. The lowest VAC values were encountered in the group with preserved EF, a higher value was in the second group, and the highest values of VAC were observed in the group with decreased EF.

Comparing left ventricular-arterial coupling parameters with LVEF, we observed that patients with severe systolic dysfunction had the lowers EEA, and the highest VAC.

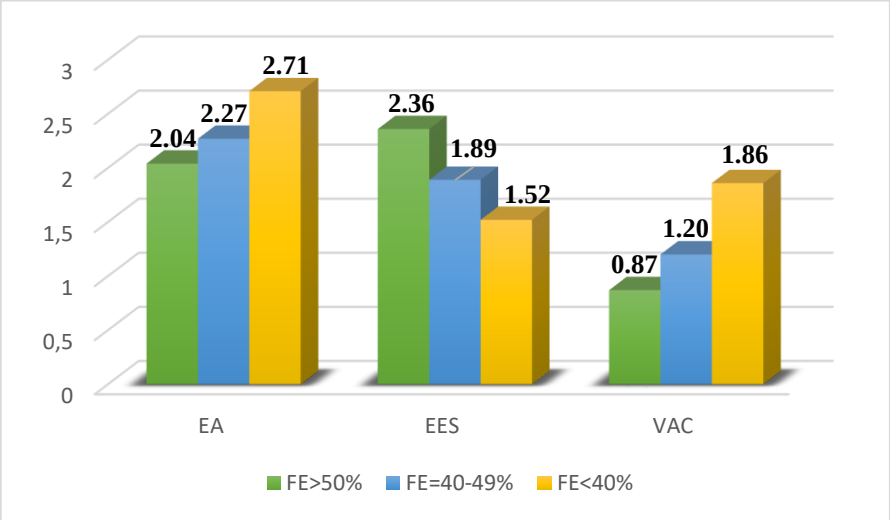

Fig 5.2. VAC, EA and EES according to the degree of LV systolic dysfunction

5.2.3.2. VAC and MACE

There were no significant differences in arterial elastance between the two groups (MACE vs. non-MACE). EA was higher in MACE group, but without statistical significance (2.65 vs. 2.33, P = 0.09). EES was significantly lower in the MACE group (1.25 ± 0.34 vs. 1.91 ± 0.56, P < 0.0001), and VAC was higher (2.2 ± 0.62 vs. 1.24 ± 0.29, P < 0.0001).

Of the new tested parameters, EES and VAC, but not EA were found to be independent predictors of MACE in COX regression analysis. ROC analysis revealed that EA and EES alone had lower predictive value for MACE than their ratio (VAC): AUC for EES 0.847 (P < 0.0001), AUC for EA 0.658 (P = 0.08), AUC for VAC 0.927 (P < 0.0001).

An EA/EES value of more than 1.71 predicts unfavourable outcome with 83.3% sensitivity, and 97.1% specificity.

5.2.4. Discussions and conclusions

This study demonstrated that noninvasively measured left ventricular arterial coupling is a predictor of unfavourable outcome 12 months after a first STEMI in a group of young patients. VAC above 1.71 demonstrated and important prognostic value in predicting adverse events, better compared to LVEF and LVGLS. Patients with higher values of VAC (over > 1.7) had higher ventricular volumes, lower ejection fraction, more impaired LVGLS, and higher values of E/e' ratio, and left atrial volumes in the acute phase. Similar results were obtained in a study on 41 patients with ischemic cardiomyopathy and moderate systolic dysfunction; in this research patients with VAC values under 1.47 had better outcome compared to those with higher EA/EES ratios [19].

By dividing the study group in three according to the degree of systolic dysfunction, we concluded that with the decrease of LV function leading to a decrease in ventricular elastance, a compensatory increase in EA occurs with the aim of maintaining an adequate intravascular volume and optimal left ventriculo-arterial coupling.

VAC proved to be an independent predictor for MACE after STEMI. Furthermore, it had an incremental prognostic value over LVEF and LVGLS in regression models.

5.3. Echocardiographic sub-study nr. 3: The role of left atrial function in predicting MACE in young patients after AMI – a 3D echocardiographic study

5.3.1. Introduction

Purpose of the study - To assess the prognostic role of LA volumes and function determined by 3D echocardiography in young patients with STEMI.

Specific objectives

- Determination of 3D LA volumes and strain derived parameters;
- Assessment of the prognostic role of this parameters in young patients with STEMI.

5.3.2. Patients and methods

– 84 young patients with a first STEMI + control group (28 young healthy subjects).

Echocardiography – All patients underwent standard transthoracic echocardiography. We also analysed myocardial work indices and left ventriculo-arterial coupling.

3D echocardiographic evaluation of the left atrium

3D acquisitions were performed using the same ultrasound system GE Vivid E9, but with a 4D probe (1,5-4 MHz).

We assessed the three functions of the left atrium:

- LA reservoir function – correspond to early atrial diastole, with maximum relaxation of the atrial walls;
- LA conduction function – mid-diastolic LA emptying and passive shortening;
- LA pump function – corresponds to atrial systole, with active myocardial shortening, representing the atrial contribution to ventricular filling.

5.3.3. Results

5.3.3.1 Assessment of LA volumes by 3D echocardiography in patients with STEMI

Volumetric analysis of LA proved that all three phasic volumes were larger in the STEMI group, compared to control group: 25.27 ± 6.93 vs. 21.1 ± 4.6 for LAVmax3D, 11.87 ± 4.91 vs. 9.9 ± 2.8 for LAVmin3D, and 17.98 ± 4.49 vs. 14.1 ± 7.1 for LAVpreS3D. The most affected of the 3 was LAVmin3D.

The emptying fractions were significantly lower in STEMI patients, compared to the control group (54.24 ± 12.03 vs. 61 ± 10.2 LAEF, 29.45 ± 13.92 vs. 30.2 ± 8.5 for passive LAEF, and 35.03 ± 11.54 vs. 49.1 ± 9.12 for active LAEF). Patients in the MACE group had significantly lower volumes and emptying fractions at baseline, compared to those in the non-MACE group.

5.3.3.2. 3D left atrial strain in STEMI patients

Compared to the control group, patients with STEMI had lower values of all LA strain parameters. There were significant differences between MACE and non MACE groups regarding 3D LA strain parameters: LAS-r 3D (15.16 ± 2.58 vs. 22.71 ± 5.25), LAS-cd 3D (-8.62 ± 1.77 vs. -10.97 ± 1.44), and LAS-ct 3D (-9.36 ± 1.51 vs. -12.73 ± 2.53). Patients with MACE at follow up had lower reservoir strain, lower conduction strain, lower pumps strain, but higher maximal and minimal LA volumes, compared to those from the non-MACE group.

5.3.3.3. Prognostic role of 3D LA parameters

The three LA strain parameters proved to have very good predictive abilities for MACE in ROC analysis (AUC = 0.894, 95 %CI, P < 0.0001 for LASr 3D, AUC = 0.854, 95

%CI, P < 0.0001 for LASct 3D, and AUC = 0.883, 95%CI, P < 0.0001 for LAScd 3D). Among them, LASr 3D had the best prognostic value. For each variable we determined a cut off value as follows: 17.5 (Sb 91.7%, Sp 86.7%) for LASr 3D, -10.5 (Sb 75% Sp 86.7%) for LASct 3D, and -9.05 (Sb 75% Sp 91.7%) for LAScd 3D. LASr 3D under 17, and LASct 3D over -10.5 determined an approximately 2-fold increase in adverse events after STEMI.

5.3.3.4. LASr 3D and diastolic dysfunction grades

LASr 3D values showed a proportional decrease with worsening diastolic dysfunction, demonstrating significant differences between diastolic dysfunction grades. We determined LASr 3D cut-off values for discriminating between different degrees of diastolic dysfunction (Fig 5.3.)

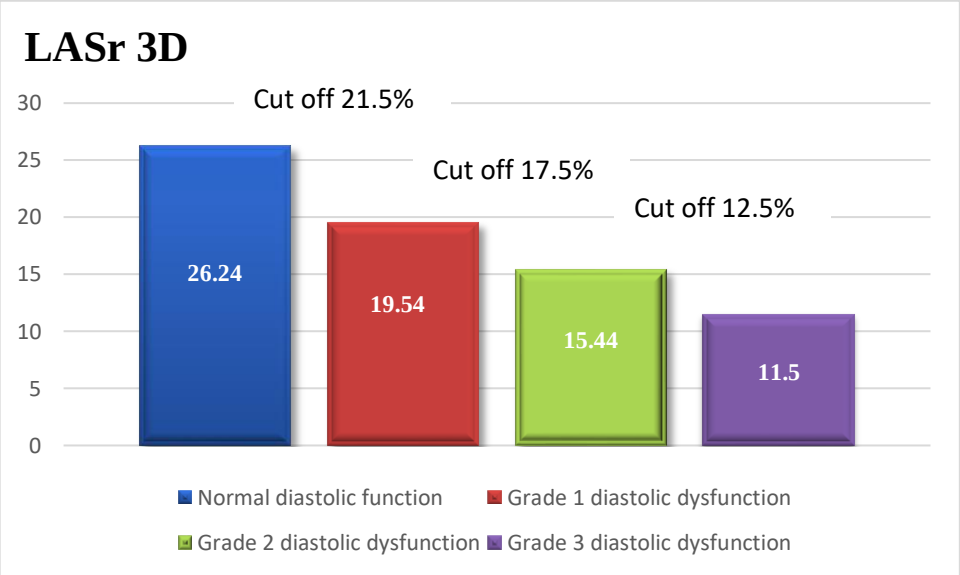

Fig 5.3. LASr 3D value according to the degree of diastolic dysfunction

Univariate COX regression analysis showed that all parameters had a significant effect on MACE, while multivariate analysis identified only reservoir function and contraction as independent predictors for adverse events.

5.3.4. Discussions and conclusions

The main role of the LA is to modulate LV ventricular filling, acting as a reservoir for venous return in ventricular systole, as a conduit in early ventricular diastole, and as a pump in late diastole, supplementing LV filling [53].

In the young STEMI patient group, lower values of reservoir, conduit, and pump strain were associated with worse prognosis after STEMI. A possible explanation for this fact would be that LA function initially compensates for the increase in LV wall stiffness

and filling pressures immediately after AMI, and the loss of this compensatory mechanism through atrial non-compliance determines impaired ventricular filling pressures[53, 54]. LA reservoir and pump strain were found to be independent predictors of MACE in young STEMI patients, independent of LVEF, and also after adjustment with known predictor parameters that proved to be significant in univariate analysis. Among the strain parameters, LASr 3D demonstrated the best prognostic value. Similar results were also obtained by Schuster et al. They highlighted the incremental prognostic role of LASr measured by MRI in a group of patients with ACS [55]. In a study from 2021, Nayyar et al. obtained similar results, also in a cardiac MRI study – LASr and LASct were shown to be predictors of MACE after AMI[56].

Regarding to the LA volumetric analysis, patients with STEMI had higher values of LAV max and min, compared to the control group. Among those with STEMI, those in the MACE group had lower 3D LAEF values, and higher LAVmax3D, and LAVmin3D values, compared to those without MACE in the first year follow up.

In contrast to LA strain, which is relatively independent of left atrial size, LAVmax3D wasn't an independent predictor of MACE in multivariable analysis after adjustment for FEVS and LVGLS. One explanation could be that volumetric indices are limited by a lower sensitivity in the early stages of the disease, compared to LA strain [57]. Reservoir strain reflects atrial compliance, and to a lesser extent contractility and relaxation, modulated by the descent of the LV base plane in systole [53].

CHAPTER 6: SECOND STUDY: NEW INFLAMMATORY MARKERS AS POTENTIAL PREDICTORS FOR MACE AFTER STEMI IN YOUNG PATIENTS

6.1. Introduction

Inflammation plays an important role in the aetiology of myocardial infarction, being involved in the initiation and progression of atherosclerotic plaques [58, 59]. In the current study we aim to assess the prognostic role of three proinflammatory cytokines (IL-1RA, IL6, rezistina) in young patients with STEMI.

Specific objectives

- Quantification of plasma cytokine levels (IL-1RA, IL-6 and resistin);
- Correlation analysis between the proinflammatory status, and biological, and echocardiographic parameters;
- Assessing the prognostic role of cytokines in the occurrence of MACE at one year after STEMI.

6.2. Patients and methods

In this study we included 41 patients with a first STEMI.

Pro-Inflammatory Cytokines Assay – For quantification of cytokine plasma levels at baseline, and at 6 months follow up, we used the enzyme-linked immunosorbent assay (ELISA) method. IL-1RA, IL-6, and resistin levels were determined using R&D Systems kits designated for each cytokine, and following the corresponding protocol**.** All samples were assayed in the Laboratory of Molecular Cardiology, Cardiology Department, Clinical Emergency Hospital Bucharest.

6.3. Results

The levels of the 3 cytokines were significantly correlated with the echocardiographic parameters that reflect left ventricular function (LVFE, LVGLS, mechanical work indices, and VAC), highlighting a possible association between the extent of myocardial damage and their plasma levels.

In the entire cohort, IL-1RA, IL-6, and resistin plasma levels decreased from the baseline at 6 months follow up. In MACE group we observed a more important decrease in cytokine levels compared with non MACE group.

Cytokines and MACE

During the one-year follow-up period, 22% of patients reached the primary endpoint (MACE) as follows: 4.8% cardiac deaths, 27% readmissions for angina, 45% readmissions for heart failure exacerbation.

Cytokine levels were different in the acute phase between the 2 patient groups, patients in the MACE group having significantly higher values (88.91 ± 74.48 vs. 18.90 ± 26.6, P = 0.023 for IL-6, 1643.7 ± 456.22 vs 456.22 ± 371.75, P = 0.05 for IL-1RA, and 9.9 ± 3.7 vs 5.7 ± 1.9, P = 0.01 for resistin). Univariate regression analysis demonstrated an association between the 3 cytokines and MACE.

The three cytokines had good prediction abilities for MACE, with AUC greater than 0.7: AUC 0.876 (95% CI), P = 0.000 for IL-6; AUC 0.827 (95% CI), P = 0.002 for IL-1RA; AUC 0.863 (95% CI), P = 0.001 for resistin. For each variable we determined a cut off value, based on the maximum value of the Youden index as follows: 456.9 pg/mL (Se = 80%, Sp = 77.4%) for IL-1RA, 42.5 pg/dL (Se 70%, Sp 96.8%) for IL-6, and 6.98 ng/dL (Se 80%, Sp 83.3%) for resistin. Out of the three variables, IL-6 proved to have the best predictive value for MACE.

We conducted a further analysis based on the number of cytokines with values above the cut-off, and divided the patients into four groups (with one, two, or three cytokine values above the cut-off, or none above the cut-off). There was a significant difference regarding the number of markers over cut-off between MACE and non-MACE groups (P = 0.000). In the MACE group, 77.8% of patients had three markers with values above the cut-off, 11.1% had two, and 11.1% had only one cytokine over the cut-off. In the non MACE, none had three markers over cut-off, 12.5% had two markers over the cut-off, 18.8% had one, and the majority (68.8%) had no markers over the cut-off value.

The combined analysis of the cytokines with the echocardiographic parameters (Cytokines + GWI AUC 0.948; cytokines + GWE AUC 0.937; cytokines + VAC AUC 0.944, Cytokines + LASr3D AUC 0.920) proved to have better results for MACE prediction than any of the markers analysed individually.

6.4. Discussions and conclusions

The association between circulating levels of IL-6 and the extent of myocardial necrosis has been previously demonstrated [60]. IL-6 plasmatic levels in the acute phase were higher in MACE group, compared to non MACE group. Groot et al. demonstrated that IL-6 levels at 24 h after AMI were independently associated with infarct size, and that higher values correlated with lower LVEF [61]. In the current research, IL-6 > 34.8 pg/dL proved to be an independent predictor for MACE after STEMI.

IL-1RA levels were higher in patients with MACE at follow up (1643.703 ± 1544 pg/mL MACE vs. 456.222 ± 371.75 pg/mL non MACE, P = 0.05). Consistent with our findings, previous studies demonstrated an increase in IL-1RA levels in the acute phase of AMI, and also an association between IL-1RA and myocardial necrosis [27, 32, 62]. IL-1RA proved its predictive values for MACE occurrence in COX regression analysis. Similar results were obtained by Schofer in 2018, who demonstrated that IL-1RA is an independent

predictor of cardiovascular mortality, beyond the prognostic value of CRP and troponin T, in a group of patients with ACS and known CAD (HR = 1.93, 95%, CI = 1.33–2.80, P < 0.001) [63].

Resistin levels were higher in patients who experienced MACE during the follow up period. Similar results were observed by Lubos et al., who proved that resistin might have a role as a diagnostic and prognostic marker, considering its elevated levels in patients presenting with ACS [33]. Other studies also confirmed that the levels of resistin corelate with cardiac fibrosis, and that resistin is an independent predictor of LV remodelling in patients with STEMI and metabolic syndrome at 12 months follow up [64].

This research concluded that the combination of IL-6, IL1-RA and resistin is a better prognostic marker for than each cytokine taken separately, and also compared to some echocardiographic or biological parameter. Moreover, the combined analysis of cytokines with echocardiographic parameters leads to a better prediction of future adverse events after STEMI.

CHAPTER 7: THIRD STUDY: miRNAS AS MACE PREDICTORS IN YOUNG PATIENTS WITH STEMI

7.1. Introduction

In recent years, microRNAs (miRNAs) have appeared as promising diagnostic and prognostic markers, being involved in the pathophysiology of cardiovascular diseases [65].

This study aims to assess the prognostic value of 6 miRNA (miR-233-3p, miR-142-3p, miR-155-5p, miR-486-5p, miR-125a-5p and miR-146a-5p) in young patients with STEMI.

Specific objectives

- miRNA isolation and quantification by RT-PCR;
- Correlations between miRNA and echocardiographic parameters;
- Evaluate the role of miRNAs as MACE predictors after STEMI in young patients.

7.2. Patients and methods

– 50 young patients with a first STEMI + control group 13 healthy subjects for validating miRNA analysis results.

Echocardiography – All patients underwent standard echocardiography. New parameters described in the first study were also determined.

miRNA isolation and quantification

MiRNA isolation and quantification was performed at the Institute of Cellular Biology and Pathology "N. Simionescu", as a part of the joint research project "EPITERAMI". miRNAs were isolated from plasma using the miRNeasy Serum/Plasma kit (Qiagen, Hilden, Germany), following the manufacturer's instructions. The plasmatic levels of the 6 miRNAs were determined by TaqMan technology. Reverse-transcription was performed with a pool of TaqMan miRNA-specific stem-loop primers on a Veriti PCR system. The obtained data were analysed using ViiA7 Software v1.2 with the automatic Cq setting. The expression level of each miRNA was determined relative to that of exogenously added cel-miR-39-2p.

7.3. Results

Patients with STEMI had significantly higher levels of miRNA when compared to the control group. miRNA levels at baseline correlated with the echocardiographic parameters of LV dysfunction (LVEF, VAC, LVGLS).

7.3.1.Patient follow up

Patients were divided in two groups (MACE and non-MACE), considering the presence or absence of MACE at one year follow up. MACE occurred in 18% (9) of the studied patients, 12% readmissions for heart failure, 4% requiring PCI and 2% cardiovascular deaths.

We observed that the expression levels of three miRNAs miR-223-3p, miR-146a-5p, and miR-142-3p were higher in the MACE group, compared with the non-MACE group. There were no significant differences between the levels of the other three miRNAs miR-155-5p, miR-486-5p și miR-125a-5p.

7.3.2.miRNAs as independent predictor for MACE at one year follow up after STEMI

Using a Cox binary univariate regression analysis, we tested the predictive potential of these markers and found that only three of the circulating miRNAs were significantly associated with the primary endpoint (MACE) in young STEMI patients. ROC analysis showed good AUC values for the three variables, greater than 0.7: AUC 0.832, P = 0.002 for miR-223-3p, AUC 0.732, P = 0.031 for miR-142-3p, and AUC 0.848, P = 0.001 for miR-

146a-5p. For each variable we determined a cut-off value, based on the maximum value of the Youden index. miR-146-5p demonstrated the gretest predictive power for MACE in young STEMI patients.

A Kaplan–Meier analysis showed the survival curves for the risk of MACE with respect to miR-223-3p, miR-142-3p and miR-146a-5p expression. Patients with higher miRNA levels at baseline had a higher probability of MACE at one year follow-up

7.3.3. Combined analysis of miRNA and echocardiographic parameters

Each of the three miRNAs and their associations added value to all the proposed predictive models. The association between miRNA and myocardial work indices proved to have a better predictive value compared with the association between miRNAs and LVEF, or miRNAs and LVGLS. The minimal statistical model with the best performance that could predict unfavourable outcome in STEMI patients was the combination of miR-142, miR-146, GWI, and GWE (AUC = 0.997).

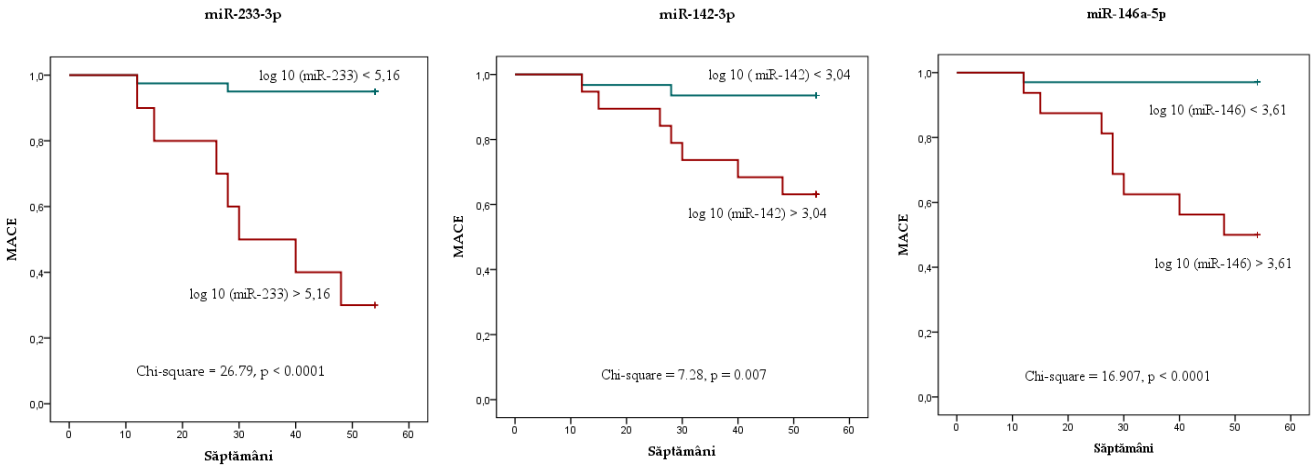

Fig 7.1. Kaplan–Meyer analysis curves showing the risk of MACE stratified by miR-223 (left), miR-142 (middle), and miR-146 (right).

7.4. Discutions and conclusions

Previous studied demonstrated that miR-146a-5p is upregulated in atherosclerotic plaques [66], and also in patients with coronary artery disease [41, 67], confirming its association with the athereosclerotic process. Oerlemans et al. [68] proved that miR-146a-5p expression in acute coronary syndromes is significantly higher, compared to unstable angina. miR-146a-5p demontrated to ba an independent predictor for MACE in patients with CAD and ACS [69], results in agreement with the current research.

miR-223-3p levels at baseline were higher in STEMI patients, compared to controls. This could be explained by the fact that miR-223-3p is strongly upregulated during the early stages of myocardial infarction before the elevation of troponin I and CK-MB [70], with a higher expression in the ischemic, compared to the normal myocardium. Schulte et al. [71] reported that increased circulating miR-223-3p in coronary artery disease could be used to predict cardiovascular death risk in a study on 873 patients.

miR-142-3p, known to be involved in atherosclerosis and ischemic heart disease [40] proved to be an independent prognostic marker of adverse outcome in our study group (P = 0.031). Its prognostic value was tested using ROC curve analysis, where we obtained an AUC of 0.732 (95% CI), p = 0.031, and determined an optimal cut-off value (sensitivity 77.8%, specificity 68.3%) for MACE prediction. In good agreement with these data, a strong predictive potential for subsequent cardiovascular events was proved in a previous study on multiple vascular atherosclerotic patients with peripheral artery disease [72].

Between the three tested miRNAs, miR-146a-5p had the best predictive value for MACE. miR-223-3p, miR-142-3p and miR-146a-5p proved to have incremental prognostic value over myocardial work indices, toegether having a better predictive power for adverse events than each separately.

CHAPTER 8. CONCLUSION AND PERSONAL CONTRIBUTIONS

8.1. Final conclusions

8.1.1. Conclusions from the first study (echocardiographic study)

1. The myocardial work indices GWI, GCW, and GWE had lower values in the MACE group vs. non-MACE, while GWW had higher values.
2. GWI, GWE, GCW, and GWW predict the occurrence of MACE at one year after STEMI in univariate COX regression analysis. In the multivariate analysis only GWI and GWE remain independent predictors for the occurrence of MACE. These parameters had incremental prognostic value over LVEF and LVGLS for the occurrence of MACE in the studied patient group.
3. Patients with GWI < 799 mmHg%, and GWE < 80.5 mmHg% had a higher risk of developing MACE at one year follow up after AMI.

4. Noninvasively measured left ventriculo-arterial coupling is an independent predictor of adverse outcome at 12 months after STEMI.
5. VAC above 1.71 demonstrated a significant prognostic role in the occurrence of adverse events, proving more effective in predicting MACE, when compared to FEVS and LVGLS.
6. Patients in the MACE group had significantly higher values of 3D LA volumes, and lower reservoir, conduit, and pump strain values, compared to those in the non MACE group.
7. 3D LA strain parameters are independent predictors for MACE after STEMI.
8. LASr 3D was significantly correlated with the degree of diastolic dysfunction; its value progressively decreasing with the worsening of the degree of diastolic dysfunction.

8.1.2. Conclusions of the second study (inflammatory markers – cytokines)

9. Higher levels of IL-1RA > 456.9, IL-6 > 42.5 pg/mL, and resistin > 6.98 ng/dl measured in the first 24-48 hours after STEMI were associated with the occurrence of MACE one year after AMI.
10. There was a good correlation between the three cytokines and echocardiographic parameters of left ventricular function, suggesting a possible association between the extent of myocardial damage and the levels of the 3 markers.
11. IL-6, IL-1RA and resistin proved to be independent predictors of MACE. Among them, IL6 had the best predictive value with ROC = 0.852. The combination of the three has better predictive value for MACE than each separately
12. The predictive value for the occurrence of MACE of the combination of cytokines and echocardiographic parameters proved to be superior to each taken separately.

8.1.3. Conclusions of the third study (with miRNAs)

13. Only three of the 6 teste miRNAs proved to have a prognostic role after STEMI (miR-233, miR-142-3p, miR-146a-5p)
14. Circulating levels of the 3 miRNAs measured at 24-48 hours after the onset of STEMI in young patients are independent predictors for MACE one year follow up.
15. The predictive value for the occurrence of MACE of the combination of miRNA and echocardiographic parameters proved to be superior, compared to each used separately.

16. The minimal statistical model with the best performance that could predict unfavourable outcome one year after STEMI in young patients was the combination of miR-142, miR-146, GWI, and GWE – Chi square model 40.04, AUC = 0.997, $P < 0.0001$.

8.2. Limitations of the study

This study included a relatively small number of patients due to the age threshold of the study group and COVID 19 pandemic). Therefore larger further studies are required to validate our findings. Another limitation would be the short follow up period (one year). At one year we only had a telephonic follow-up available due to COVID-19 pandemic restrictions. miRNAs and cytokines quantification was not possible in all patients included in the study due to technical and financial reasons.

Despite these limitations, our study holds great potential, considering the obtained results and their further possible utility in current clinical practice, and with potential utility in post-infarction risk stratification in young patients.

8.3. Personal contributions and future directions

It is worth mentioning that this research is focused on a target population – young patients with STEMI. This is the first study that proposes an association between new echocardiographic parameters and molecular markers as predictors of MACE after STEMI.

Both, the new echocardiographic parameters and the selected molecular markers determined at 24-48 hours after the onset of the acute myocardial infarction proved to be good predictors for the occurrence of MACE one year follow up. Combining new echocardiographic markers with molecular markers (cytokines or miRNA) allows a better prediction of post infarction outcome.

Using this research as a starting point, I want to continue the process of identifying new prognostic markers for young patients after STEMI. I think that it would be interesting to assess the same parameters in a group of older patients, in order to compare the results with the current cohort of young patients. I would extend this research on a larger study group to validate our findings. Also, I propose the elaboration of a score that allows optimal patient risk stratification from the time of hospital admission. I am also considering longer term follow up of the young STEMI patients from the current study.

In conclusion, a multiparametric approach (using echocardiographic markers together with molecular markers – miRNA or cytokines) would lead to a better prediction of the occurrence of adverse cardiovascular events one year after STEMI in young patients, being able to facilitate a better risk stratification, and an improvement of secondary prevention strategies, thus decreasing morbidity and mortality in this group of patients.

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1. MicroRNAs in Acute ST Elevation Myocardial Infarction – A New Tool for Diagnosis and Prognosis: Therapeutic Implications – **Alina Ioana Scărlătescu,** Miruna Mihaela Micheu, Nicoleta-Monica Popa-Fotea, Maria Dorobanțu; International Journal of Molecular Sciences, 30 april 2021, Vol 22, Issue 9, ISI indexed, ***impact factor 4,556***; https://www.mdpi.com/1422-0067/22/9/4799 (chapter 2 of the PhD thesis)

2. IL-6, IL-1RA and Resistin as Predictors of Left Ventricular Remodelling and Major Adverse Cardiac Events in Patients with Acute ST Elevation Myocardial Infarction – **Alina Ioana Scărlătescu**, Miruna Mihaela Micheu, Nicoleta Popa-Fotea, Ana Maria Pascal, Ana Maria Mihail, Ioana Petre, Silvia Deaconu, Aura Vîjîiac, Maria Dorobanțu; Diagnostics, 21 january 2022, vol 12, issue 2, ISI indexed, ***impact factor 3,706;*** https://www.mdpi.com/2075-4418/12/2/266 (Chapter 6 of the PhD thesis)

3. miR-146a-5p, miR-223-3p and miR-142-3p as Potential Predictors of Major Adverse Cardiac Events in Young Patients with Acute ST Elevation Myocardial Infarction – Added Value over Left Ventricular Myocardial Work Indices – **Alina Ioana Scărlătescu**, Teodora Barbălată, Anca Volumnia Sima, Camelia Stancu, Loredan Ștefan Niculescu, Miruna Mihaela Micheu ; Diagnostics, 12 august 2022, vol 12, nr 8, ISI indexed, ***impact factor 3,992***; https://www.mdpi.com/2075-4418/12/8/1946 (Chapter 7 of the PhD thesis)

Papers presented/poster presetations at specialty congresses/conferences – as first author:

1. Correlations between left atrial strain and systolic and diastolic LV function in young patients with STEMI – **Alina Ioana Scărlătescu**, Miruna Mihaela Micheu, Monica Stoian, Maria Dorobanțu – Poster presentation at The 13th congress of the Romanian Medical Association – Romania Academy 18-20 april 2019, Bucharest
2. Early measurement of left ventricular global longitudinal strain and mechanical dispersion predict left ventricular remodeling at 5 year follow up after STEMI: pilot study – **A.I. Scărlătescu**, S. Onciul, D. Zamfir, A. Pascal, M. Dorobanțu – Poster presentation at EuroEcho Congress 4-7 december 2019, Vienna
3. Peak left atrial systolic strain as a marker of left ventricular diastolic dysfunction in patients with ischemic heart failure with depressed ejection fraction after STEMI: pilot study – **A.I. Scărlătescu**, M. Stoian, N.M. Popa-Fotea, G. Nicula, N. Oprescu, C.A. Mihai, V. Bătăilă, L. Câlmâc, D. Zamfir, V. Ploscaru, A. Scafa-Udriște, M.M. Micheu, M. Dorobanțu – Poster presentation at EuroEcho Congress 4-7 december 2019, Vienna
4. Correlations between left atrial strain parameters and left ventricular function in young patients presenting with acute ST elevation myocardial infarction – **A.I. Scărlătescu**, M.M. Micheu, M. Stoian, D. Zamfir, I. Petre, M. Dorobanțu – Poster presentation at EuroEcho Congress, 4-7 december 2019, Vienna
5. Correlations between 3D mitral valve parameters and left ventricular remodeling at 6 months after ST elevation myocardial infarction: pilot study – **A.I. Scărlătescu**, S. Onciul, A. Pascal, D. Zamfir, I. Petre, R. Onuț, S. Iancovici, M. Stoian, C. Guzu, A. Vîjîiac, M. Dorobanțu, Posterat the ESC congress 2020 – The Digital Experience
6. Prediction of left ventricular remodeling following ST elevation myocardial infarction: role of myocardial deformation parameters – **Scărlătescu A.I.**; Onciul S.; Pascal A.; Petre I.; Zamfir D.; Guzu C.; Iancovici S.; Stoian M.; Vîjîiac A.; Onuț R.; Dorobanțu M. - Poster presentation at EACVI Congress – Best of Imaging 2020.