VICTOR BABEȘ UNIVERSITY OF MEDICINE
AND PHARMACY TIMIȘOARA
FACULTY OF MEDICINE
DEPARTMENT OF INTERNAL MEDICINE

MORARIU VLAD IOAN

PhD THESIS

MULTIDISCIPLINARY APPROACH TO PATIENTS WITH METABOLIC SYNDROME AND SUBCLINICAL CARDIOVASCULAR IMPAIRMENT

Scientific Coordinator
PROF. UNIV. DR. MIRELA CLEOPATRA TOMESCU

Timișoara
2022

Introduction

The Metabolic Syndrome (MS), regardless of its current definition cut-off points, that have generated polarized views regarding incidence of both the disease itself, as well as the complicated consequences it leads to, is characterized by atherogenic dyslipidemia (with high LDL-C levels, as well as elevated triglycerides), abdominal obesity, insulin resistance or Type 2 Diabetes Mellites, as well as arterial hypertension.

Despite the myriad of defining criteria (be it JIS, NCEAP ATPIII or IDF), it is a underdiagnosed nosological entity, studies demonstrating its prevalence varying from 15% to 30% (1), also depending on several factors such as gender, age, ethnicity, inflammation status, genetic background, and sociocultural status of the patient, as well as criteria cut-off values.

All MS components have paramount cardiovascular implications. Elevated LDL-C fractions, and as well as triglycerides can be easily encompassed in the notion defined as atherogenic dyslipidemia, with important vascular impairment and subsequent risk of ischemic manifestations ranging from acute coronary syndromes to stroke and PAD. It has been further noted that the particular dyslipidemia profile found in MS patients is linked to high apolipoprotein B levels, being thus prone to a quicker evolution of atherosclerotic plaques.

Insulin resistance, or diagnosed diabetes is also associated with impaired endothelial function and further cardiovascular risk.

It has long since been demonstrated that systemic arterial hypertension is both a standalone disease as well as a cardiovascular risk factor, being included along with high cholesterol levels and smoking status in cardiovascular risk stratification, as used in the SCORE chart.

Moreover, abdominal obesity is frequently associated with both a low grade chronic pro-inflammatory status, with elevated TNF-alpha, as well as insulin resistance.

Taking into consideration the risk MS poses not only from a cardiovascular standpoint (through severe complications such as acute coronary syndromes, stroke, peripheral artery disease), but also from a endocrinological standpoint (as it is frequently associated with Polycystic Ovary Syndrome) as well as a gastroenterological standpoint (through its frequent association with Non Alcoholic Fatty Liver Disease), it is important to ensure a multidisciplinary approach to the patients who suffer from it, as to provide optimal management both from a lifestyle standpoint and a therapeutical standpoint. Moreover, as its incidence is on the rise, correct and timely management is of the essence in order to greaten life expectancy and quality of life, as well as to decrease general morbidity and mortality, in particular from a cardiological and neurological standpoint.

Hence, it is of a paramount importance to correctly assess MS patients from both a cardiovascular, neurological, imagistic, metabolic as well as gastroenterological perspective as timely as possible.

The current thesis focuses on early, subclinical cardiovascular impairment in MS patients, and the role a relatively new imagistic tool plays in this particular area, namely 2D ultrasound strain imaging, a method that still requires validation as a robust predictor of early deterioration of vascular and cardiovascular structure and functionality.

The General part of the current thesis is divided into three main chapters. The first chapter focuses on the MS definition, criteria used for correct diagnostic and up to date epidemiological data regarding incidence of each particular MS criteria. Then it further reviews several cardiovascular significant MS components, namely atherogenic Dyslipidemia, Systemic Hypertension and Cardiovascular Risk, as well as Endothelial Dysfunction and Inflammation. Finally, it highlight current treatment guidelines for the aforementioned atherogenic dyslipidemia.

The second chapter of the General part reviews the association between metabolic syndrome and subclinical cardiovascular impairment,

including atherosclerotic lesions, asymptomatic Left Ventricle Diastolic Disfunction in the general population, as well as the little know data regarding MS patients present in international studies, from both a epidemiological as well as diagnostic and prognostic standpoint.

The third chapter of the general part focuses on the multidisciplinary assessment of the metabolic syndrome, including its complications, and focuses on the role strain imaging has as a diagnostic and prognostic tool for this particular subgroup of patients.

The Experimental Part, through the contributions it brings, focuses on three main Hypothesis, each developing into a prospective or retrospective studies, as follows :

1.   The existence of a correlation between elastography determined liver stiffness in MS patients and left atrial impairment, as a subclinical cardiovascular marker in predicting a future left ventricular diastolic disfunction, assessed through 2D strain imaging, a novel imagistic approach. A case-control observational study carried out on a umber of 150 MS patients, in the Department of Cardiology and the Department of Neurology of the Victor Babes University of Medicine and Pharmacy from Timisoara.

2.   The confirmation of the prognostic value carotid strain imaging plays in assessing the risk of stroke and acute coronary syndrome in MS patients, over a 3 year interval. A prospective observational study carried out in Timisoara Emergency Municipal Hospital's Cardiology Department over the course of 3 years, encompassing a total of 220 MS patients..

3.   The validation of left atrial 2D strain imaging as a predictor of silent paroxysmal atrial fibrillation in patients with transient ischemic attacks, as MS patients, through the defining components of this particular disease are at great risk of developing vascular complications. A retrospective cohort study encompassing 190 patients belonging to the Timisoara County Clinical Emergency Hospital's Neurology Department

Written informed consent was obtained from all enrolled patients.

It is important to note that CVD risk increases from threefold for only one MS feature to fourteen-fold with four constituting MS features, as portrayed by Nakanishi et al. The presence of diabetes increases CVD incidence fivefold. Compared to the general population, MS patients have a risk of 30% regarding the development of chronic heart disease, preceded by diastolic dysfunction, whereas up to 50% will develop a major coronary event (unstable angina, sudden cardiac death, fatal as well as non-fatal myocardial infarction) over the course of 5 years, when compared to the general population. Moreover, a recent study undertaken by Boer et al, evaluating over 13000 MS patients without constituted CHD or diabetes or stroke history demonstrated that the risk of a first episode of ischemic stroke is directly corelated and increases with the number of MS criteria (HR=1.75, with a 95% CI of between 1.35-2.27)

Experimental Part

1.The Correlation between Left Atrial Impairment and Elevated Liver Stiffness in Metabolic Syndrome Patients

The current study focuses on assessing the correlation between diastolic dysfunction of the LV and variable degrees of liver steatosis and/or fibrosis in MS patients, and, moreover, to establish if LA performance is a predicting factor for LV diastolic dysfunction and whether it can be corroborated with NAFLD (Non Alcoholic Fatty Liver Disease), a nosological entity associated with high cardiovascular mortality.

150 MS subjects were enrolled in the study group and 150 age– and sex– matched subjects without MS were admitted in the control group. The study had the following inclusion criteria for the study group: age greater than 18 years, the presence of diagnosed MS. The following exclusion were used: chronic hepatic illness due to viral infection, excessive alcohol intake, drug addiction; diagnosed systolic heart failure, diagnosed ischemic heart disease; moderate to severe valopathies, history of atrial fibrillation/atrial flutter or their presence upon admission; cardiomyopathies; presence of ICDs; PAD; history

of stroke; severe systemic illness or malignancy; chronic kidney disease; pregnancy or lactation

Subject age varied from 31 to 85 years (mean 62.4±10 years). 164 patients (54%) were of male sex. No significant differences among the two groups were observed regarding heart rate, smoking status, serum transaminase and the low-density lipoprotein (LDL)-cholesterol levels. MS patients had a much higher incidence of hypertension, diabetes and obesity. They displayed higher triglycerides, glycosylated hemoglobin (HbA1c) and FPG values, and lower total cholesterol and of high- density lipoprotein (HDL) cholesterol levels.

MS patients had the following CAP assessed steatosis severity: 21 (14%) –S0, 11 (7%) - S1, 11 (7%)-S2, and 107 (71%) - S3. MS patients had a higher incidence and severity of both liver steatosis and fibrosis (P<0.0001, respectively P=0.04

Heart Ultrasound data revelead no significant differences between the two groups regarding conventional LV structure and systolic function parameters. However, MS patients had a much higher incidence of diastolic dysfunction of the LV. Conventional 2D ultrasound did not reveal any differences between study and control group regarding LA dimensions, volumes and EF.

However, 2D-STI revealed in MS patients subtle LA dysfunction reflected by significantly lower longitudinal strains and strain rates during the ventricular contraction, as well as during early diastolic filling and atrial contraction, MS patients having a significantly higher LA stiffness (P<0.0001).

LA stiffness was a good predictor of subclinical diastolic dysfunction of the LV in MS patients (45% sensitivity and 96% specificity whilst using a cut-off value >0.38.)

Upon comparison of ROC curves of the two independent predictors for LV diastolic dysfunction, the under the curve area (AUC) had a greater area for LA stiffness than for liver stiffness, however the differ-ence was not statistically

significant. A LA stiffness above 0.38 was found in 36 (24%) of the MS patients and in 12 (8%) control group subjects (P<0.0001). The relative risk of MS patients having a LAsf greater than 0.38 was 3.0, upon comparison to the control group (95% CI 1.62 to 5.53, AUC=0.66, P< 0.001).A LAsf greater than 0.38 was proved to be positively correlated with a liver fibrosis stage greater than F2 (r=0.59, 95% CI 0.51- 0.66, P< 0.0001) as well as a hepatic steatosis stage greater than S 2 (r=0.42, 95% CI 0.32 -0.51, P< 0.0001).

The current study demonstrated that, despite the lack of significant difference regarding LA size, volumes, and ejection fractions between control group and MS patients ,2D STI determined LA deformation, assessed by peak LA strain and strain rate values were statistically significant impaired in patients suffering from MetS (P<0.0001).

As up to 16% of NAFLD patients die because of coronary CAD, whereas CAD-related mortality is none NAFLD patients is only 1–3%, there might be a significant association between NAFLD and CVD related mortality in these patients, association that can be explained through several possible theories. The current study has been able to demonstrate that an elevated LAsf is correlated with diastolic dysfunction of the LV. Moreover, a LAsf greater than 0.38 is robustly and proportionally correlated with liver fibrosis with a grade above F2, as well as liver steatosis above grade S 2, all aforementioned parameters having statistical significance (P< 0.0001.

LV diastolic dysfunction, identified by cardiac ultrasonography, is positively, independently and significantly correlated to hepatic fibrosis above grade 2 in patients suffering from MS. Moreover, 2D STI assessed LA stiffness is a robust predictor of diastolic dysfunction of the LV, being independently correlated with both liver steatosis above stage S2,as well as liver fibrosis above grade F2, thus suggesting that CAP, VCTE, as well as 2D-STE need to become standard assessment techniques in all MS patients.

2. Carotid Artery Strain and Strain Rate as a Predictor in Assessing the 3-year Risk of Acute Coronary Syndrome and Stroke in Metabolic Syndrome Patients

Despite advances in 2D strain imaging, the prognostic significance of carotid arterial strain and strain rate in MS patients has yet to be established. In the current study, STE determined CS and CSR has been used to determine CCA stiffness in patients suffering from MS, in order to further establish their predictive significance for MACE over a follow-up period spanning 3 years.

The current study included 220 MS patients. Inclusion criteria for the current study demanded the patients pertain to an age group between 40 and 70 years , and have a certified well documented MS diagnosis as well as the ability to consent. Exclusion criteria comprised refusal to provide their informed consent, congenital vascular anomalies, a history of prior stroke or transient ischemic attacks, established, well diagnosed cardiovascular disease, prior atrial fibrillation and flutter, current life-threatening diseases or cancer, as well as pregnancy or lactation.

The mean age of the studied population was 60.7 ± 7.5 years, among which 102 (47%) were of female gender and 118 (53%) were male. MACE occurred in 14 enrolled patients (7%), as stated bellow: 6 (2%) acute coronary syndromes, 8 (4%) atherothrombotic ischemic strokes, and 2 (1%) hospitalizations for HF decompensation. Henceforth the MS patients were included in two subgroups, depending on whether they had suffered a MACE or not in the three year follow- period.

Conventional ultrasound evaluation of the left and right CCA revealed no statistical differences among MS patients, however significant statistical differences were present among the two studied groups regarding CCA strain and strain rate values, namely: CS (2.12 ± 0.7% vs 3.33 ± 1.14%, $p < 0.0001$) and CSR (0.31 ± 0.11 s^{-1} vs 0.31 ± 0.1 s^{-1}, $p < 0.0001$).

After applying univariate regression algorithm, it prevailed that systemic hypertension, diabetes mellitus, and the circumferential strain and strain rate of

the CCA as well as age were correlated with a much higher risk of MACE in the studied group of MS patients.

Multivariate logistic regression was performed for MACE predictors highlighted during univariate analysis, and identified two independent predicting factors for MACE in MS patients: CCA circumferential strain (%) and circumferential strain rate (1/s), both having statistical significance/

ROC curve for the aforementioned MACE independent predicting factors revealed a good sensitivity and specificity, as follows: CCA circumferential strain rate (AUC = 0.779, with a sensitivity = 82.6%, and a specificity = 72.4%, $p < 0.0001$); CCA circumferential strain (AUC = 0.806, with a sensitivity = 82.6%, and a specificity = 79.2%, $p < 0.0001$). ROC curves cross comparison for these two MACE predictors (CCA CS and CSR) revealed no statistically significant differences between areas under the ROC curves with a $p = 0.67$.

The identified cut-off values for the independent predictors of MACE using ROC curves were ≤2.9% for CS, respectively <0.35 s^{-1} for CSR. Using these particular cut-off values, Kaplan-Meier survival curves were generated, indicating that survival without ischemic stroke, ACS as well as MACE was significantly higher among the MS patients with higher CS and CSR values ($p < 0.0001$),

Based on available data available in the present moment, the current study is among the first to examine the role played by carotid artery strain and strain rate as independent predicting factors for to major cardiovascular events in a population of MS patients. The current study managed to establish the existence of a robust correlation between decreased deformation of the carotid wall, quantified through STI and the atherothrombotic vascular event risk in patients suffering from MS. It is important to note that this particular correlation is independent from the presence of established atherosclerotic plaques determined by IMT, as well as the presence of specific vasodilating drugs that can lead to subsequent decrease in arterial wall deformation.

The present study emphasizes that decreased CCA CS as well as CRS are robust, early predictors of subclinical atherosclerosis, and thus tightly linked to the risk of developing consequent atherothrombotic events.

Carotid artery circumferential strain as well as circumferential strain rate are robust independent predicting factors for major cardiovascular and cerebrovascular events in patients suffering from MS, without any priorly established cardiovascular pathology. Carotid artery deformation measured by 2D STI is a valuable prognostic marker for cardiovascular risk in the aforementioned populational subgroup, despite not being currently used by default in vascular assessment, despite the IMT's lack of predictive capabilities. The current result must be disseminated for better MS patient management.

3. Reduced Left Atrial Strain - a Robust Prognostic Factor for Transient Ischemic Attacks in Patients Suffering from Silent Paroxysmal Atrial Fibrillation

The aim of the present study is to assess the correlation between modified deformation parameters of the LA obtained by 2D STE, as predicting factors, and the risk of cardioembolic TIAs in patients diagnosed with silent PAF.

The current retrospective epidemiological study included patients diagnosed with TIA of more than 50 years of age, discharged Timisoara County Hospital's Neurology Department between November 2017 and November 2021, patients that underwent 2D STI of the LA.

Main exclusion criteria included: previously diagnosed AF, or AF within 24 hours after the onset of TIA or at the moment the cardiologic assessment was performed, moderate/severe valvular regurgitation and/or stenosis, mechanical heart prosthetics, congenital cardiomyopathy, severe left ventricular systolic dysfunction (LEVF under 40%), ventricular akinesia, implantable cardiac devices, such as pacemakers, and, most importantly, a decreased quality of ultrasound images.

The current study inducted 190 patients suffering from TIA, which were then divided into two subgroups, as follows: group I had documented PAF episodes, whereas group II had did not suffer from PAF. 33% of TIA patients suffered from a documented PAF episode. PAF patients had the following characteristics: they had an increased age (mean age of 67.5 vs 60 years in non PAF patients, P<0.0001), were more frequently of female gender (48% vs 32%, with a P=0.04), and more frequently associated a history of neurological vascular events (TIA or stroke - 65% vs 23%, with a P<0.0001).

Furthermore, the current study highlighted significant differences between the two cohorts regarding LA functional parameters. Patients with a history of PAF episodes had a larger LAVI(P<0.001), a decreased total LAEF (P<00001), as well as LA strain patterns. However, LV Diastolic as well as systolic parameters, including LV global strain and strain rate did not vary between the two subgroups.

Univariate analysis of the aforementioned clinical as well as ultrasound parameters revealed that risk of developing PAF was significantly correlated to advanced age, female sex, decreased LAVI, LAEF as well as LA strain and strain rates. Despite its current role, and its statistically significant predominance in group I patients (P<0.001 , an elevated CHA2D2S- VASc score was not significantly correlated with PAF in patients suffering from TIA, as logistical regression revealed. Multivariate logistical regression included the aforementioned variables correlated with the risk of developing PAF and was able to detect the following independent predictors for PAF in patients suffering from TIA: increased age, as well as decreased LAEF and LARVS .

ROC curve revealed a good specificity and sensitivity for these independent predictors of PAF, as follows: age (AUC=0.922 and sensitivity: 72.92, respectively a specificity 90.1, with a P<0.0001), LARVS (AUC of 0.915, with a sensitivity of 100.0, a specificity of 64.8, and P<0.0001), respectively LAEF (AUC=0.717 with a sensitivity of 72.9, a specificity of 90.1 and a P<0.0001).

ROC curve comparison demonstrated a significant difference between areas under ROC curves (AUC) for age as well as LARVS when compared to LAEF (0.198 and P<0.0001). The following cut-off values for PAF correlated predictors were an age over 55 years, a LARVS under -17%, as well as a LAEF under 51%.

A study conducted by Pagola et al demonstrated that 86% of patients suffering from strokes of unknown etiology had episodes of silent AF, correlated with normal LA dimensions, yet a decreased strain.

The current study's results emphasize the importance of routine LA function assessment in patients with TIA of unknown etiology, as a reduced atrial contractility and hence deformation is associated with increased risk of intracavitary thrombus development and subsequent embolization. Moreover, studies have demonstrated the evolution of AF from paroxysmal to persistent consists in a atrial cardiomyopathy, associated with the subsequent risk of stroke. Despite the body of evidence, during stroke risk assessment for patients suffering from AF, TIA is frequently omitted, despite its common pathophysiological mechanism, and the increased risk of progression to stroke that these patients have. In order to reduce risk of stroke, AF predictors such as the ones demonstrated in the current study are paramount, strain imaging being a robust marker of LA structure and function.

The current research demonstrates that LA strain parameters are robust independent predictors of PAF episodes in patients suffering from TIA, thus providing important information regarding the cardioembolic etiology of TIA, and emphasizing the need of proper anticoagulation, LA strain and strain rate having a higher sensitivity and specificity than usual 2D echocardiographic parameters

Despite the important contribution, the limited number of patients in the present study requires further validation by large multicenter research.

The current study is of paramount clinical value, by exploring the role 2D strain imaging of the LA plays in assessing possible PAF risk in TIA

patients, approach not used on a regular basis in the present moment, despite its good reproducibility and ease of use, in a category of patients in which a proper anticoagulant prophylaxis is of dire need, given the risk of recurrent TIA or stroke episodes, in the case of silent PAF.

General Conclusions. Contributions. Limitations. Future Research.

The Metabolic Syndrome, through all its recognized components (obesity, atherogenic dyslipidemia, hypertension, insulin resistance), is by all means a disease of great relevance to the present moment, especially through its rising prevalence (affecting up to 35% of the population, depending on the relatively diverse cut-off points of the defining criteria).

The current study aims to bring a new perspective regarding its most dire of complications, namely the accelerated development of cardiovascular diseases, due to the chronic inflammatory status, endothelial dysfunction, and atherogenic dyslipidemia the metabolic syndrome is characterized by.

The metabolic syndrome's high cardiovascular complication risk (through coronary artery disease, risk of stroke, peripheral artery disease) is impossible to quantify only from one standpoint. A proper management requires the presence of a skilled multidisciplinary team, comprising of at least a cardiologist, a diabetologist, endocrinologist, neurologist, as well as a good imagist, as all the diseases' components have a profound cardiovascular effect, generally mediated by the chronic inflammation-insulin resistance-endothelial dysfunction triad.

Thus, an early detection of cardiovascular impairment in these patients is off the essence, in order to lower their global cardiovascular morbidity and mortality. This can be achieved by using recent advances in ultrasonography, namely strain imaging, better known as speckle tracking, in order to detect timely, subclinical cardiovascular impairment.

The current work, by means of the studies published in high-visibility international journals, has managed to demonstrate the following aspects regarding the multidisciplinary approach to patients with metabolic syndrome and the aforementioned subclinical cardiovascular impairment:

1.It has determined that diastolic dysfunction of the left ventricle, assessed using cardiac ultrasound, is independently and significantly correlated to hepatic fibrosis of grade 2 or above, in patient suffering from metabolic syndrome. Furthermore, the study has established that left atrial stiffness assessed by 2D strain imaging is associated with both diastolic disfunction of the left ventricle, as well as liver fibrosis and steatosis, a left atrial stiffness >0.38 being positively correlated with both liver fibrosis grade F≥2 (r=0.59, with a 95% CI of 0.51 to 0.66,and P< 0.0001) as well as liver steatosis grade S ≥2 (r=0.42, with a 95% CI of 0.32 to 0.51, and P< 0.0001). As NAFLD is associated with higher cardiovascular mortality, liver elastography as well as strain imaging must be routinely used in metabolic syndrome assessment.

2.The study has established that carotid circumferential strain (with a ROC curve determined sensitivity of 82.6%, a specificity of 79.2%, and p < 0.0001) as well as circumferential strain rate (with sensitivity of 82.6%, specificity of 72.4%, and p < 0.0001) measured by 2D speckle tracking are both robust independent predicting factors for major cardiovascular as well as cerebrovascular events in metabolic syndrome patients without any priorly diagnosed cardiovascular pathology, and should be used routinely in examining MS patients.

3.It has demonstrated that altered strain parameters of the left atrium during 2D speckle tracking are robust independent predictors, with good sensitivity and specificity, of possible paroxysmal atrial fibrillation episodes as a thromboembolic etiological cause of transit ischemic attacks. Most associated with atrial fibrillation episodes were left atrial reservoir strain (sensitivity: 100.0, specificity 64.8, and P<0.0001) and left atrial total emptying fraction (sensitivity: 72.9; specificity 90.1; and P<0.0001).

The studies bring novel imagistic prognostic research in a field in which cases are frequently underreported or diagnosed only when severe cardiovascular, neurological or metabolic complications arise. It further validates the hypothesis that 2D strain imaging is useful not only in assessing subclinical disfunction of the left ventricle, but also of the left atrium, with prominent predictive power in detecting probability of paroxysmal atrial fibrillation. Furthermore, it demonstrates the role speckle tracking has in vascular assessment, a field which requires future research.

Despite the important contribution in prognostic and diagnostic multidisciplinary assessment of the metabolic syndrome, the limited number of patients in each of the present studies require further validation by larger, multicenter research. Another possible drawback of the aforementioned studies was the fact that most imagistic assessment was performed by a reduced number of sonographers, although intraobserver reproducibility was promising.

Furthermore, there is a current need for future research regarding myocardial and vascular strain rate as a predicting factors for adverse cardiovascular outcomes in patients with different comorbidities, including amyloidosis and myocarditis, speckle tracking sonography representing a promising and powerful prognostic tool, with high sensitivity and specificity, and ease of use, in the absence of the drawbacks such as angle of insonation and reduced reproducibility.