"OVIDIUS" UNIVERSITY OF CONSTANȚA
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Thesis for doctoral degree
ABSTRACT

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CARDIOVASCULAR RISK ASSESSMENT IN PATIENTS WITH ANKYLOSING SPONDYLITIS

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Keywords: ankylosing spondylitis, cardiovascular risk, SCORE, relative risk

Motto

"The best way to predict the future is to create it" Abraham Lincoln

INTRODUCTION

Ankylosing spondylitis (AS) or radial axial spondyloarthritis is a chronic, progressive, immune-mediated arthritis that predominantly affects young men. It is characterized by the absence of rheumatoid factor and the presence of inflammation in the axial skeleton (sacroiliac joints and spine), peripheral joints, joints, but also possible extra-articular manifestations: uveitis, cardiovascular, gastrointestinal tract, pulmonary or renal involvement. As a chronic disease, the influence on the patient's quality of life and productivity, and the socio-economic impact, are particularly important and are due to the relatively high prevalence of the disease (1%), but above all to its onset at a young age (under 45), when social mobility is at its peak, the most productive period of life.

This paper comprises two cross-sectional observational studies (prevalence, cross-sectional) of patients with ankylosing spondylitis.

The major research question we addressed in the first study was to determine the prevalence and identify the types of cardiovascular impairment, as well as cardiovascular risk factors, in patients diagnosed with ankylosing spondylitis.

In the second study my aim was to assess whether the Systematic Coronary Risk Evaluation (SCORE) chart underestimates cardiovascular (CV) risk in young (<50 years) patients with (AS) and at the same time I sought to determine whether the use of the Relative Risk (RR) chart score can improve the identification of young AS patients at high and very high risk of cardiovascular disease.

I benefited from the support of the Rheumatology Department of the Medical Section 2 of the "Sfântul Apostol Andrei" Emergency Clinical County Hospital in Constanța and in particular from Professor Maria Șuța, the first scientific supervisor of my PhD thesis, who guided me in the conception and realization of this study and to whom I address my warmest thanks.

The scientific foundation and the elaboration of this PhD thesis would have been impossible without the help, support and guidance of Professor Doctor Irinel Raluca Parepa, who has contributed

decisively to my formation as a doctor, researcher, but especially as a human being, always giving me the courage to go further.

I especially want to thank my family and especially my husband, Paris, for the unconditional love, moral support, financial support, understanding and encouragement they have continuously given me throughout these years.

THE CURRENT STATE OF THE ART

The first part of the PhD thesis, the ***general part***, presents a literature review on current developments in ankylosing spondylitis (AS), notions of epidemiology and etiology, data on the mechanism, treatment and course of the disease, as well as the main comorbidities associated with this condition.

Ankylosing spondylitis (AS) is a chronic inflammatory disease with an incompletely known etiology, usually affecting young men. It progresses to significant disability due to skeletal disorders including: reduced spinal mobility, peripheral joint lesions and extra-articular lesions (including visceral lesions), leading to decreased quality of life and work productivity among these patients (1).

Although the pathogenesis of AS remains incompletely elucidated, the currently accepted hypothesis is that AS develops through complex interactions between immune-mediated mechanisms and genetic conditions, environmental factors, microbial infections and endocrine disorders (2). Various reviews and meta-analyses report that AS is associated with a 1.5- to 2-fold higher mortality rate compared to the general population, mainly related to cardiovascular (CV) complications (3).

There is increasing evidence recently suggesting that atherosclerosis is an inflammatory disease (4). The role of inflammation in the development of heart disease has been recognized relatively recently (84). Rheumatic disease can be viewed as a "natural experiment" in the interaction between chronic inflammation and cardiovascular disease, which may elucidate the fundamental mechanisms by which inflammation accelerates the development of atherosclerosis and the onset of cardiovascular disease (5).

Dysfunction of the arterial endothelium is at the heart of the atherogenesis process, altering the vasomotor response to various neurohumoral stimuli, leading to myocardial ischemia, platelet rupture, thrombosis and subsequently acute myocardial infarction (6). Although the management of inflammatory arthritis does not focus on cardiovascular morbidity and mortality, it appears that the majority of premature deaths are attributed to cardiovascular disease (7).

According to current data, inflammatory arthritis is an independent CV risk factor, so screening and treatment of cardiovascular risk factors is imperative (7).

In conclusion in patients with AS, we can benefit from cardiovascular risk reduction and accelerated atherosclerosis prevention through complex interventions on the inflammatory profile, but also by correcting traditional CV risk factors.

The European Society of Cardiology (ESC) clinical practice guidelines on CV disease prevention recommend the use of the Systematic Coronary Risk Evaluation (SCORE) as a predictive model for estimating the 10-year risk of fatal CV disease (mortality from myocardial infarction, stroke, aortic aneurysm or other), including the following variables: age, gender, total cholesterol, smoking status and systolic blood pressure level - with age having the most significant impact in this model (4). Although risk assessment tools can be useful aids for physicians in establishing a therapeutic plan for patients, "SCORE" has some significant limitations in its ability to identify patients at high CV risk when applied to the population under 50 years of age, thus preventing them from initiating primary prevention (5,6). Pharmacological intervention, particularly statin therapy, used to reduce CV risk is indicated only among those considered at high risk of CV events; thus, a large proportion of younger patients with AS may be neglected (7, 8).

The 2016 ESC guidelines proposed the use of a relative risk (RR) chart rather than the traditional SCORE model in patients younger than 50. Unlike SCORE, RR estimates relative, not absolute, risk, showing the likelihood of developing fatal CV disease in an individual with traditional CV risk factors compared with another with no risk factors (4, 9). This is a central point of concern in AS, a disease associated with early atherosclerosis characterized by oxidative stress and inflammation (10, 11).

PERSONAL CONTRIBUTION

The special part of the thesis comprises two cross-sectional observational studies (prevalence, cross sectional) on patients with ankylosing spondylitis and was conducted in the Rheumatology Department, Medical Ward 2 of the Emergency Clinical County Hospital "St. Apostle Andrew" in Constanta, over a period of 4 years, starting in January 2016. The two studies with the objectives of each, material and method and the results obtained allowed the elaboration of conclusions with practical value.

Study I

The major research question I intend to address in this study is to determine the prevalence and identify the types of cardiovascular impairment, as well as cardiovascular risk factors, in patients diagnosed with ankylosing spondylitis.

This chapter includes a description of the population diagnosed with AS in terms of socio-demographic characteristics, clinical and paraclinical data, characteristic treatment, association of CV risk factors.

Specific objectives

- Characterization of clinical, biological and ultrasonographic features of patients with AS, with detection of cardiovascular risk and prognostic factors;
- Highlighting CV risk using traditional assessment tools (SCORE diagram, calculation of relative risk of death from cardiovascular causes at 10 years);
- Highlighting the role of inflammatory markers (CRP, VSH) in assessing the risk of cardiovascular disease and subclinical atherosclerosis;
- Highlighting the role of carotid artery Doppler ultrasonography and brachial artery flow-mediated vasodilation (FMD) in the evaluation of subclinical atherosclerosis;
- Detecting peculiarities and developing an optimal model for assessing patients at risk.

Materials and methods

Study I includes a group of 100 patients diagnosed with ankylosing spondylitis according to the modified New York 1984 criteria (11) and a control group including 100 patients without chronic inflammatory rheumatic disease (known with arthrosis, osteoporosis), selected consecutively over a period of 4 years starting in 2016, who were admitted to the Rheumatology Department, Medical Ward 2 of the Constanta Emergency Clinical County Hospital.

The similarity of the groups is ensured in terms of age, sex, clinical parameters, body mass index (BMI) characteristics.

We estimated CV risk using SCORE based on total cholesterol, applied to a high-risk population such as Romania.

Study results I

It is well known that males are at higher risk for cardiovascular disease than females; as shown in the Framingham study, for patients aged 35 to 85 years, morbidity and mortality from coronary heart disease is almost double for males (27).

AS is more common in men than in women by a ratio of about 2:1 (3). In my study, of the 100 subjects with AS 73 were male (73%) and 27 were female (27%), identical distribution in the control group, thus correlating with the data in the literature.

The majority of AS patients have an age of disease onset <45 years, and the presence of HLA-B27 antigen and male gender have been associated with early disease onset.

In the present study, a statistically significant difference was demonstrated between the mean age of onset of the disease in the group of patients with AS (31.04 years) and the control group (43.83 years) ($p < 0.001$.). This leads to the conclusion that AS has an onset at an earlier age which may lead to increased cardiovascular risk of multifactorial cause.

Statistical analysis of the study data reveals a significantly higher prevalence of disease duration > 10 years among patients with AS compared to patients in the control group (p=0.031) which would suggest a higher proportion of disease duration > 10 years as a CV risk factor in patients with AS and a 3.341-fold higher risk of having a disease duration > 10 years in patients in the AS group compared to patients in the control group.

The age of onset of AS appears to correlate with whether the patient is HLA-B27 positive or negative, with HLA-B27 positive patients becoming symptomatic earlier, at a mean age of 24.8 years, while HLA-B27 negative patients become symptomatic at a mean age of 32.7 years.

We observed a significantly higher prevalence of CRP>5 (mg/L) among patients with AS compared to patients in the control group (p=0.001) suggesting a risk of having a CRP>5 (mg/L) value 3.205 times higher in patients in the AS group than in patients in the control group.

On the other hand, high blood pressure (>20 mm/h) was associated with an almost threefold increased risk of CV events in our study, consistent with findings in the general population.

CRP values, HRV, BASDAI score and ASDAS score were associated with abnormal IGB (<9 or >1.3), demonstrating a correlation between inflammatory disease activity and arterial stiffness.

Higher BMI was associated with higher disease activity and including poorer response to biological therapy. In the present study it was shown that there was a 1,670-fold higher risk of having hypercholesterolemia among patients in the SA group than in patients in the control group.

Statistical analysis of the presence reveals a significantly higher prevalence of DZ among patients with AS compared to patients in the control group (p=0.031) which would suggest a higher proportion of DZ as a CV risk factor in patients with AS and a 3.341-fold higher risk of having DZ in patients in the AS group compared to patients in the control group.

A statistically significant association was also demonstrated in the group of patients with AS between the presence of diabetes and personal history of cardiovascular disease. We thus hypothesize that diabetes mellitus may be involved in the association between ankylosing spondylitis and myocardial infarction and stroke.

In the present study we showed that there was a significantly higher prevalence of carotid plaques in the AS patients compared to the control patients (p=0.001) which would suggest a higher prevalence of carotid plaques as a CV risk factor in AS patients and a 4.846-fold higher risk of carotid plaques in the AS patients compared to the control patients (Figure 1).

A statistically significant association (p=0.001) was also demonstrated in the group of patients with AS between the presence of carotid plaques and personal history of cardiovascular disease.

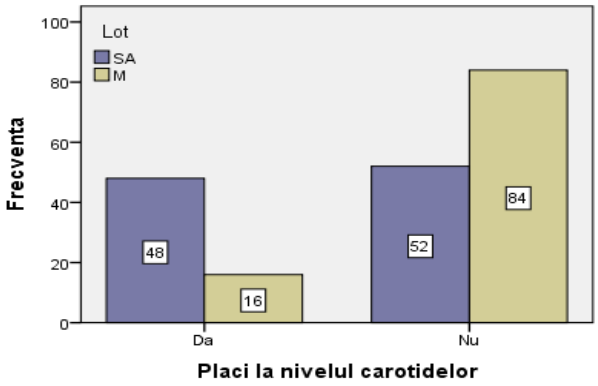

Figure 1. Column plot for variables Batch (SA, M) & Carotid Plates (Yes, No).

Statistical analysis of the study data reveals a significantly higher prevalence of IGB with pathological values (<0.9 or >1.3) among patients with AS compared to patients in the control group ($p<0.001$) which would suggest a higher proportion of IGB as a CV risk factor in patients with AS and a 3,090-fold higher risk of having abnormal IGB in patients in the AS group than in patients in the control group. A statistically significant association was also demonstrated in the AS group between the presence of pathologically significant IGB and personal history of CVD ($p<0.001$).

We found a significantly higher prevalence of high/very high risk of developing a CVD at 10 years according to the SCORE chart among patients with AS compared to patients in the control group ($p = 0.001$) suggesting a 4.462-fold higher high/very high CV risk according to SCORE in patients in the AS group than in patients in the control group (Figure 2).

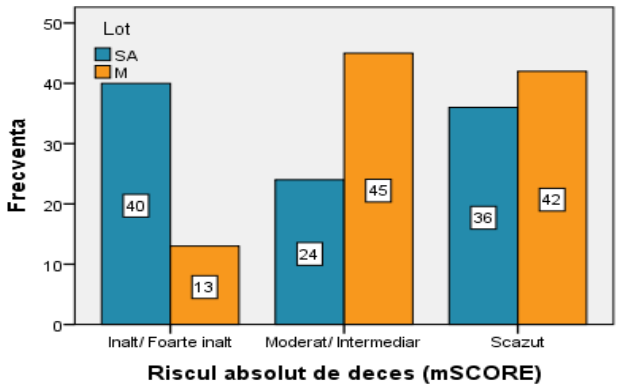

Figure 2. Column plot for the three CV risk categories according to the SCORE diagram, compared in the two study groups

Study I conclusions

- The present research has highlighted that there is still a growing need to improve CV risk prediction models tailored for young patients with chronic inflammatory diseases.
- The effect of additional risk factors, such as C-reactive protein (CRP) and carotid intima-media thickness index (IMT), must be taken into account.

- Given the link between inflammation in AS and worsening CV mortality, the use of anti-inflammatory agents is a topic of considerable interest for both treating AS and mitigating the development of CV sequelae.
- The identification of AS patients at high risk of CV disease is a matter of major relevance for the prompt initiation of early primary prevention treatment and thus reducing the potential risk of fatal CV events in these patients.
- Cardiovascular examination along with imaging tests such as echocardiography and Doppler ultrasound of the carotid arteries of patients with AS should be routinely performed, especially in patients with increased disease duration in whom the HLA-B27 antigen is positive.

Study II

CV risk prediction is an extremely important aspect of CV prevention. Even though there have been significant developments in recent years, risk scores for primary prevention need to be improved, especially in patients under 50, and new prediction models need to be developed and validated.

The aim of the study was to determine whether classical risk charts may underestimate CV risk in young patients with AS and also to promote the need for new risk assessment models and methods to achieve primary prevention of CV disease in this population, thus the present study contributes to a deeper understanding of CV risk in AS, allowing the development of innovative patient-specific CV risk models.

The main objective of Study II

The primary objective of this study was to highlight whether the Systematic Coronary Risk Evaluation (SCORE) chart based on total cholesterol underestimates CV risk in young patients with ankylosing spondylitis (AS) and to promote the need for new models of CV risk assessment in this patient group.

We also sought to determine whether the use of the graphical relative risk (RR) score can improve the identification of young AS patients at high and very high risk of cardiovascular disease.

Materials and methods

Study II includes 70 consecutive patients aged 19 to 50 years, previously diagnosed with AS, with no history of CV disease, no diabetes and no history of chronic kidney disease. We estimated CV risk using SCORE based on total cholesterol, applied to a high-risk population such as that in Romania. CV risk estimation was also performed using the relative risk (RR) graph, taking into account the following variables: smoking, systolic blood pressure and total cholesterol.

Carotid Doppler ultrasound was performed in all patients in the study group and included measurement of carotid intima-media thickness (IMT) at the level of the common carotid artery and detection of focal atheroma plaques in the extra-cranial carotid artery.

Study results II

Regarding the demographic characteristics of patients diagnosed with AS, there is a preponderance of males (54 patients) compared to females (16 patients).

Regarding the presence of the HLA-B27 antigen, according to the literature and in our study out of a total of 70 subjects the majority (79%) are positive, SA being strongly associated with this gene.

Studying the distribution of the group of patients diagnosed with AS according to BASDAI score, we observed that just under half (46%) of the patients had a BASDAI score >4, thus being considered as having active disease.

In our study 52.1% of patients with AS had CRP levels greater than 3mg/L at the time of disease diagnosis before receiving anti-TNFα drugs that can reduce their levels, and this value was significantly associated with the presence of carotid plaques, even after adjusting for confounders (bias).

More than half (61%) of the patients in the study group have an ASDAS-PCR score between 3 and 6, noting increased disease activity.

We find that there is a strong, statistically significant dependence relationship between the variables of hypertension and the existence of atheromatous plaques in the carotid arteries, i.e. a GIM index >9 mm. In other words, the majority of AS patients at very high risk of developing CV disease due to carotid plaques are hypertensive.

Analyzing the entire study group of 70 patients diagnosed with AS, we noticed a large number of patients (no. 30 representing 43% of the total) in whom the Doppler ultrasound of the carotid arteries showed an index of mean intimal thickness > 9 mm or even the presence of carotid plaques, which automatically put patients in a very high cardiovascular risk zone.

The majority of patients (n=59, 84%) had a moderate risk according to the SCORE chart, and 11 (16%) had a low CV risk; no patient had a high or very high CV risk. A total of 4 patients (36.36%) of the 11 considered low risk based on SCORE had carotid plaques, and 26 patients (44%) of the 59 patients with moderate CV risk were found to have carotid plaques (Table I).

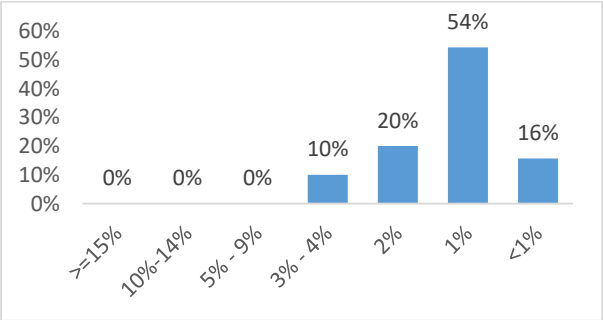

Figure 3. SCORE - absolute risk of cardiovascular death at 10 years

Based on the 2016 ESC guidelines, patients were considered to be at high/very high CV risk if the SCORE system was ≥5 (none of the patients in the study group) or if they had carotid plaques assessed with ultrasound. According to the definition, 30 of all 70 patients were at high/very high CV risk due to the presence of carotid plaques, i.e. GIM index > 9 mm.

We find that the SCORE underestimates the cardiovascular risk of these patients, no patient in the study group is at high/very high risk according to SCORE (Figure 3).

In contrast, when we compared the RR graph score with the presence of carotid plaques, we found that only 2 out of 12 (16%) of patients with RR=1 had carotid plaques, and we note that the frequency was higher in those with RR>1 (28 out of 58 representing 48%) (Table II).

Table II. Relative RR risk correlated with the presence of carotid plaques

	Frequency	**Percent**	**Carotid artery plaques n (%)**
RR = 1	30	17	2 (16%)
RR>1	40	83	28 (48%)
Total	**70**	**100**	**22**

Under these conditions, we can estimate that the relative risk (RR) has the ability to distinguish between the two groups - presence or absence of carotid plaques (sensitivity = 90%, specificity = 85.00%, associated criterion RR>2) (Figure 4).

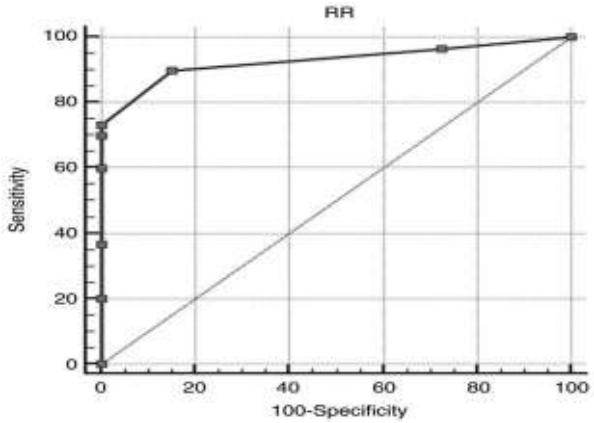

Figure 4. ROC curve of the relationship between relative risk (RR) and the presence of carotid plaques

Under these conditions we can conclude that relative risk has the ability to distinguish between the two groups PCR >3 mg/dl or ≤3 mg/dl (sensitivity = 71.79%, specificity = 83.87%, associated criterion >2) (Figure 5).

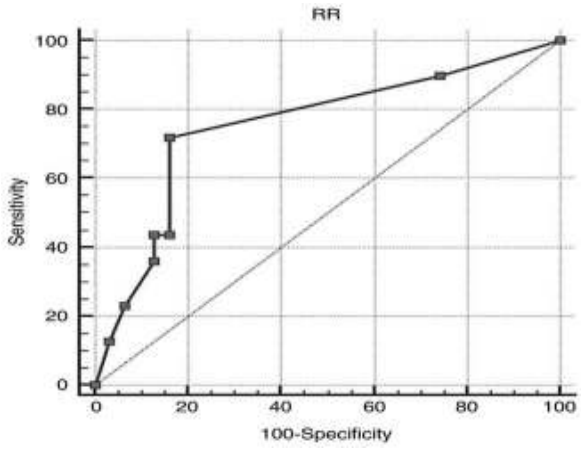

Figure 5. ROC curve of the relationship between relative risk (RR) and C-reactive protein (CRP).

We can appreciate that the RR variable has the ability to distinguish better between the two groups (CRP >3 mg/dl vs. CRP ≤3 mg/dl) than the SCORE variable.

Study II conclusions

In the present study, RR was shown to be superior to the SCORE risk score for identifying patients with AS and age under 50 years at high CV risk, thus allowing them to benefit from intensive preventive treatment. This is crucial given that statins have been shown to decrease mortality by 37% in AS, a decrease double that seen in the general population.

We also confirmed the limitations attributed to the SCORE system when applied to AS patients under 50 years of age. (3). Although significant carotid plaques or a GIM index greater than 9 mm on carotid ultrasound were found in 27% of the patients in the study group, most of them had been classified as "low CV risk" based on the SCORE algorithm.

The use of RR has improved the identification of young AS patients at high CV risk. In this regard, AS patients with RR > 1 were three times more likely to have subclinical atherosclerosis than those with RR = 1 (48% versus 16%).

The combination of an RR > 1 and a CRP level greater than 3mg/L at diagnosis of chronic inflammatory disease increased the probability of revealing severe subclinical CV disease expressed by the presence of carotid plaque by up to 60%.

We concluded that RR was superior to SCORE when attempting to identify young patients at high CV risk; in this regard, SA patients with RR>1 were almost three times more likely to have subclinical atherosclerosis than those with RR=1 (48 vs. 136%), making them high risk.

The effect of additional risk factors, such as CRP and GIM, should be considered. Their contribution to the estimation of absolute CV risk for AS patients is important.

Originality and innovative contributions of the thesis

1. This is the first study to assess the ability of a graphical score such as RR to improve the identification of cases at high risk of developing fatal BCV among young AS patients.

2. The aim of the study was to determine whether classical risk charts may underestimate CV risk in young patients with AS and also to promote the need for new risk assessment models and methods to achieve primary prevention of CV disease in this population, thus the present study contributes to a deeper understanding of CV risk in AS, allowing the development of innovative patient-specific CV risk models.

3. Our study showed that the majority of patients with AS do not have the traditional CV risk factors used by standard scoring tables. However, many of them are at high risk of developing CV disease when considering other parameters such as CRP levels or carotid plaques.

4. The present research has highlighted that there is still a growing need to improve CV risk prediction models tailored for young patients with chronic inflammatory diseases. The effect of additional risk factors, such as CRP and GIM, needs to be considered. To this end, further studies should be carried out by clinical researchers together with statisticians and epidemiologists.

5. The effect of additional risk factors, such as CRP and GIM, must be taken into account. To this end, additional studies should be carried out by clinical researchers together with statisticians and epidemiologists.

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