ISSN 2320-5407 International Journal of Advanced Research (2015), Volume 3, Issue 12, 240 – 246

Journal homepage: http://www.journalijar.com

INTERNATIONAL JOURNAL OF ADVANCED RESEARCH

RESEARCH ARTICLE

Sonographic and Serologic Evaluation of Endothelial Dysfunction in Ankylosing Spondylitis

Doaa S. Atta [(1)], Ahmad A. Elsammak [(2)], Radwa M. Abdullah[(3)] and Rania Ghonaim [(4)]
[(1)]Rheumatology and Rehabilitation, [(2)] Radiology, [(3)] Cardiology, [(4)] Clinical Pathology

Manuscript Info

Manuscript History:

Received: 15 October 2015
Final Accepted: 22 November 2015
Published Online: December 2015

Key words:

Corresponding Author

Doaa S. Atta

Abstract

Objectives: evaluation of subclinical atherosclerosis and endothelial dysfunction sonographically and by serological markers and assess its correlation with disease activity in patients with AS compared with healthy controls. **Methods:** the study population included 42 AS patients and 42 healthy controls were subjected to detailed history and clinical examination. Spinal mobility was assessed using BASMI. Disease activity was evaluated with BASDAI. Functional status was evaluated with BASFI. Laboratory evaluation includes FBG, CRP, total cholesterol, HDL, LDL, triglycerides, CD141, vWF and UT-II. Radiographic assessment included plain X-ray pelvis and ultrasound measurements (CIMT, FMD and ABI). **Results:** AS patients had significantly higher CRP, CD141, VWF and UT-II with mean 21.7, 28.9, 194.5 and 11.4 respectively. With no significant difference between the 2 groups regarding lipid profile and fasting glucose. AS patients had significantly higher CIMT (P = 0.00), FMD (P = 0.03) and ABI (P = 0.00). Significant differences were reported between the 2 groups regarding AS disease activity indices (BASFI, BASDAI and BASMI) (P = 0.00), CRP (P = 0.02) and CIMT (P = 0.02). **Conclusion:** The present study provides evidence of increased association of atherosclerosis in AS subjects compared to controls. This should provide impetus to early intervention strategies to prevent accelerated atherosclerosis which would help in reducing the cardiovascular morbidity and mortality associated with this disease.

Copy Right, IJAR, 2015,. All rights reserved

INTRODUCTION

Ankylosing spondylitis (AS) is a chronic inflammatory rheumatic disease. Its musculoskeletal manifestations include both inflammation and structural damage. Characteristic extra-articular manifestations include aortitis, cardiac conduction defects, pulmonary fibrosis and inflammatory bowel disease indicating that AS is a systemic disease.

As is known primarily for causing a life-time of pain, impaired physical function, work disability and decreased quality of life, rather than for shortening life itself. However, patients with AS also experience premature mortality [(1)].

The standardized mortality rate (SMR) associated with AS are approximately 50% higher than in the general population [(2)]. The four non-inception cohort studies published to date quote SMRs of 1.33 [(3, 4)], 1.5 [(5)] and 1.8 [(6)].

Increased mortality is largely attributable to cardiovascular disease (CV). A large population based study has shown more ischaemic heart disease (prevalence ratio 1.2), peripheral vascular disease (ratio 1.6),

atherosclerosis (ratio 1.5), congestive heart failure (ratio 1.8) and more cardiovascular risk factors (prevalence ratios between 1.3 and 1.7) in AS patients compared to healthy controls. [7]

Ankylosing spondylitis (AS) is a chronic rheumatic disease that comprises mainly the spine and sacroiliac joints. Despite its inflammatory origin like RA and SLE, it is not totally clear if atherosclerosis accounts for higher mortality in these patients. As steroids are not commonly part of clinical treatment of these patients.[8]

High-resolution ultrasonography can be used to measure the intima-media thickness (IMT) as well as vascular elasticity of the carotid artery. An increased carotid IMT reflects the atherosclerotic burden and predicts the development of (clinically apparent) CV disease in the general population [9]. This study was designed to determine whether signs of subclinical atherosclerosis are more prominent in a sample of AS patients compared to controls without the disease but with similar cardiovascular risks. Other studies assessing IMT in AS patients and controls have been published but results were contradictory [10].

AS has been shown to be associated with impaired endothelial function, which is an initial step in the pathogenesis of atherosclerosis may be reflected by changes in various endothelial biomarkers of hemostasis and the release of several cellular adhesion molecules or cytokines. [11]

Even though both serological and sonographic studies have shown endothelial dysfunction in AS its etiopathogenesis is still unclear.[12]

Several biomarkers are currently available to assess endothelial function.[13]

AIM OF THE STUDY

The aim of the study was to evaluate subclinical atherosclerosis and endothelial dysfunction sonographically and by serological markers and assess its correlation with disease activity in patients with AS compared with healthy controls.

PATIENTS AND METHODS

Our study was conducted in out-patient clinic of Rheumatology and Rehabilitation, Radiology, Cardiology and Clinical Pathology Departments, Zagazig University hospitals. In the period from April 2014 to August 2015.

Patients and control

- Forty-two patients were diagnosed according to modified New York criteria [14]. They were above 20 years of age and has disease duration of at least 2 years. We excluded subjects with diabetes mellitus, hypertension, hyperlipidemia, coronary artery disease, peripheral vascular disease, cerebrovascular disease, renal insufficiency hypothyroidism and any tobacco use in the previous 30 days.
- Forty-two healthy controls matched for age, sex and smoking status were recruited. Non of them had a family history of AS, psoriasis, RA, SLE or any inflammatory rheumatic disease.
- Written informed consent was obtained from all subjects.

Clinical assessment:

- A detailed history was recorded from the patients regarding the disease duration, sites involved and the drug used. The joints and entheses involved in the disease process were assessed for swelling and pain. Height and weight of the patient were recorded. Waist circumference was taken midway between lower border of the rib cage and the iliac crest. Chest expansion was measured in centimeters as the difference between maximal inspiration and maximal forced expiration at the level of nipples in males and just below breasts in females. Blood pressure was recorded by sphygmomanometer. Patients who had a systolic blood pressure > 140 mmHg and diastolic blood pressure > 90 mmHg were classified as hypertensive according to the JNC-7 criteria [15] and excluded. All the peripheral pulses were assessed and both the carotid arteries calculated for bruits.
- Spinal mobility was assessed using the Bath Ankylosing Spondylitis Metrology Index (BASMI) [16]. Lateral lumbar flexion, tragus to wall distance, lumbar flexion measured by modified Schober's test, maximal intermalleolar distance and cervical rotation were the criteria employed to assess spinal mobility.
- Disease activity was evaluated with the Bath Ankylosing Spondylitis Disease Activity Index (BASDAI) [17].
- Functional status was evaluated with the Bath Ankylosing Spondylitis Functional Index (BASFI) [18].

- All indices were scored in a scale ranging from 1 to 10.

Laboratory Evaluations :

Following an overnight fast, venous blood samples were collected between 8:00 and 9:00 am for laboratory tests. Fasting blood glucose, standard C-reactive protein (CRP) and serum lipids (total cholesterol, HDL, LDL and triglycerides) were measured using routine techniques.

Blood samples, collected from the patients and controls, were preserved at – 80 °C until assayed for endothelial markers.

Thrombomodulin (CD 141)

- Plasma CD 141 was measured using Human CD 141 ELISA (Gen-Probe, cat. no. 850.720.096) according to the manufacturer's instructions. The minimum detectable dose of human CD141 was less than 0.380 ng/ml. The average concentration of CD141 detected in normal serum was

von Willebrand factor

- Serum von Willebrand factor (vWF) was measured by Human vWF ELISA (Assay Pro, cat no. EV2030-1) according to the manufacturer's instructions. The minimum detectable dose of vWF was <1 mU/ml. The intra- and inter-assay coefficients of variation were 4.9% and 7.6%, respectively.

Urotensin II

- Plasma urotensin (UT-IT) was measured by Human Urotensin ELISA (Phoenix Pharmaceuticals, cat. no. EK-071-05) according to the manufacturer's instructions. The minimum detectable dose of urotensin was 0.06 ng/ml. The infra- and inter-assay coefficients of variation were 5-10% and <15%, respectively.

Radiographic Assessment:

X- Ray

- Plain X-ray pelvis anteroposterior view was used to assess sacroiliitis which was graded according to the Modified New York criterid [14] by a single observer (SG).

Ultrasound Measurements:

- All ultrasound measurements were taken in a quiet, temperature controlled room at the same time of the day (between 8:00 AM and 11:00 AM, to avoid diurnal variation in FMD by a single observer (AE) with the patient fasting for at least 8 hours. The patient was also instructed not to do any night work or exercise on the day before the examination. An ultrasound machine (EUB 5500, Hitachi) with 7.5 MHz linear transducer was used to make the ultrasound measurements.

Carotid intima-medial thickness (CIMT)

- Bilateral assessment of the common carotid artery (CCA) wall thickness was made with the patient supine with the neck extended and the chin turned contralateral to the side being examined. Longitudinal image of the artery was taken.
- Carotid intima-medial thickness (CIMT) was measured 2 cm proximal to the carotid bulb as the distance from the leading edge of the first echogenic line to that of the second echogenic line. The first line represented the lumen-intima interface, and the second line, the collagen-containing upper layer of tunica adventitia. Average of CIMT of right and left common carotid arteries was used.

Flow mediated dilation (FMD):

- The right brachial artery was studied in all subjects. Subjects were in the supine position with the arm outstretched. After at least 10 min of rest, the brachial artery was imaged by a longitudinal section approximately 5 cm above the antecubital fossa.
- After obtaining the baseline image, a blood pressure cuff was placed around the arm above the scanned part of the artery and was inflated to 200 mmHg for 5 min. The blood pressure cuff was then gradually deflated, resulting in reactive hyperemia, which caused an increase in shear stress and dilatation of the brachial artery. The next ultrasound image was obtained 1 min after cuff deflation.
- Measurement of the artery diameter was done between m-lines (media-adventitia interface) of the near and far walls. Endothelium-dependent flow mediated dilatation of brachial artery was expressed as the percentage change in brachial artery diameter from baseline. FMD <4.5% was taken as a measure of endothelial dysfunction. [19]

Ankle-brachial index (ABI):

- For the measurement of ankle-brachial index (ABI), after the subject has been in the supine position in the bed for 15 min, blood pressure cuff was placed over the arm for brachial artery and just above the malleoli for dorsalis pedis and posterior tibial arteries. The Doppler signal of the artery distal to the cuff was visualized. Then the cuff was rapidly inflated to 30 mm Hg above the audible systolic pressure and complete collapse of the artery was ensured by noticing the disappearance of the Doppler signal of the artery. The cuff was then deflated at a rate of 2 mm/s. The systolic pressure was recorded as the pressure at which first doppler signal reappears. The procedure was repeated on the other side also.
- McDermott et al. [20] showed that the association between ABI and lower-extremity function was strongest when ABI was calculated by averaging the values from the dorsalis pedis and posterior tibial arteries, hence we calculated the ankle-brachial index (ABI) for each lower extremity artery by dividing the mean of dorsalis pedis and posterior tibial pressures by the mean of the right and left brachial pressures.
- Final ABI of the patient was taken as the average of the ABIs of each limb. ABI <0.9 was taken as evidence of atherosclerosis [21].

RESULTS

Table (1): Demographic and clinical characteristics of AS patients and controls

	AS patients **N = 42**	Controls N = 42	P value	X^2
Age (years)	29.6 ± 8.2	30.6 ± 8.6	0.59	0.54
Sex (M/F)	36/6	29/13	0.12	2.45
Height (cm)	164.9 ± 4.9	161.9 ± 5.6	0.01*	2.61
Weight (Kg)	56.7 ± 5.2	60.3 ± 5.6	0.00*	3.02
BMI (kg/m^2)	19.1 ± 1.3	21.3 ± 2.0	0.00*	5.97
Waist circumference (cm)	78.1 ± 6.2	82.1 ± 5.4	0.00*	3.15
MAP (mmHg)	86.1 ± 9.1	86 ± 8.7	0.90	

BMI: Body Mass Index

MAP: Mean Arterial Pressure

42 AS patients and 42 healthy controls matched for age, sex and smoking status were studied. The mean age of AS patients was 29.6 years, M/F was 36/6, mean height was 164.9 cm, mean weight was 65.7 kg, mean BMI was 19.1 kg/m^2, mean waist circumference was 78.1 cm and mean MAP was 86.1 mmHg. The mean age of controls was 30.6 years, M/F was 29/13, mean height was 161.9 cm, mean weight was 60.3 kg, mean BMI was 21.3 kg/m^2, mean waist circumference was 82.1 cm and mean MAP was 86 mmHg. Significant differences between AS patients and controls reported in height, weight, BMI and waist circumference.

Table (2): Characteristics of AS patients

Characteristic	AS cases (n = 42)
- Disease duration (years)	11.2 ± 6.05
- NASIDs n (%)	40 (95.2)
- TNF antagonists n (%)	
- Etanercept	5 (11.9)
- Infliximab	-
- Anti-rheumatic agent	
- Sulfasalazine	20 (47.6)
- Methotrexate	2 (4.9)
- Both	2 (4.9)

- BASMI score	2.9 ± 1.9
- BASDAI score	3.9 ± 2.9
- BASFI score	2.9 ± 2.7

NASIDs: Non-Steroidal Anti-Inflammatory Drugs, TNF: Tumor Necrosis Factor, BASMI: Both Ankylosing Spondylitis Metrology Index, BASDAI: Both Ankylosing Spondylitis, Activity Index, BASFI: Both Ankylosing Spondylitis Mfunctional Index

In AS patients mean disease duration was 11.2 years, 95.2% were using NASIDs (Indomethacin was the commonest drug used), 11.9% were on Etanercept, 47.6% on Sulfasalazine, 4.9% on Methotrexate, 4.9% were using both. The mean BASMI, BASDAI and BASFI scores were 2.9, 3.9, 2.9 respectively

Table (3): Laboratory profile in AS patients and controls

	AS patients N = 42	Controls N = 42	P value	t
CRP (mg/L)	21.7 ± 18.4	1.2 ± 1.1	0.00*	
Fasting glucose (mg/dl)	90 ± 7.5	86.6 ± 7.3	0.74	0.32
Total cholesterol (mg/dl)	172 ± 30	185.3 ± 34.8	0.78	0.30
HDL cholesterol (mg/dl)	43.7 ± 9.4	47.6 ± 10.8	0.79	0.27
LDL cholesterol (mg/dl)	107 ± 27	110 ± 25.2	0.94	
Triglycerides (mg/dl)	94 ± 46	101 ± 43	0.92	
CD 141 (mg/dl)	28.9 ± 7.4	2.42 ± 0.1	0.00*	
VWF (mg/dl)	194.5 ± 20.8	78.9 ± 15.9	0.00*	
UT-II (mg/dl)	11.4 ± 2.3	1.21 ± 1.2	0.00*	

CRP: C-Reactive Protein, HDL: High Density Lipoprotein, LDL: Low Density, Lipoprotein, CD 141: Thrombodulin, VWF: Von Willbrand Factor, UT-II: Urotensin II

AS patients had significantly higher CRP, CD141, VWF and UT-II with mean 21.7, 28.9, 194.5 and 11.4 respectively. With no significant difference between the 2 groups regarding lipid profile and fasting glucose.

Table (4): Ultrasound measurements in AS patients and controls

	AS patients (N = 42)	Controls (N = 42)	P value	t
CIMT	1.56 ± 0.1	0.41 ± 0.07	0.00*	43.5
FMD	14.3 ± 9.7	18.9 ± 8.6	0.03*	1.97*
ABI	2.16 ± 0.1	1.0 ± 0.1	0.00*	37.5

CIMT: Carotid Intima- Medial Thickness
FMD: Flow Mediated Dilatation
ABI: Ankle-Brachial Index

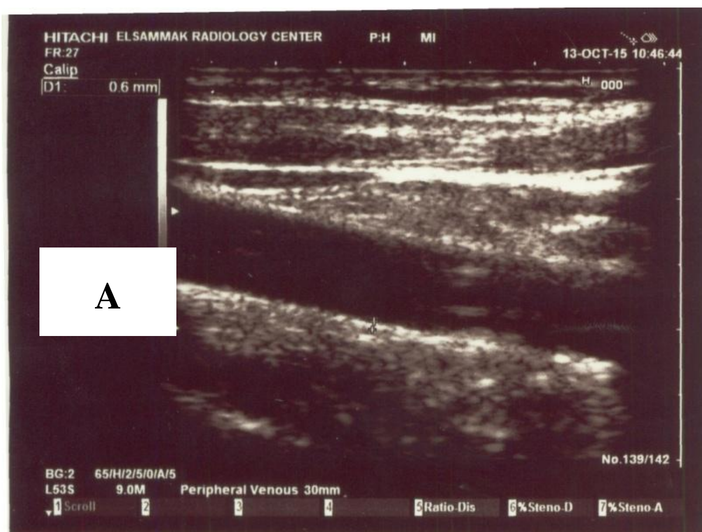
AS patients had significantly higher CIMT (P = 0.00), FMD (P = 0.03) and ABI (P = 0.00).

Table (5): Comparison of AS patients based on disease activity

Characteristic	Active patients (> 4) N = 20	Inactive patients (≤ 4) N = 22	P	t
Disease duration (y)	12 ± 7.2	10.4 ± 4.9	0.23	1.19
BASFI	4.5 ± 1.9	1.2 ± 1.2	0.00*	9.5
BASDAI	5.9 ± 1.2	1.8 ± 0.8	0.00*	18.4

BASMI	2.4 ± 1.8	1.1 ± 2.0	0.00*	4.65
CRP	29.1 ± 27.1	14.4 ± 9.7	0.02*	2.38
VWF	91.6 ± 27.5	102 ± 14.1	0.13	7.65
Thrombodulin (CD 141)	3.6 ± 0.5	4.2 ± 1.8	0.16	1.43
Urotensin II (UT-II)	5 ± 2.9	4.2 ± 2.9	0.38	0.89
CIMT	1.50 ± 0.8	1.22 ± 0.3	0.02*	2.34
FMD	13.4 ± 7.3	15.4 ± 8.4	0.86	0.78
ABI	1.97 ± 0.8	1.46 ± 0.43	0.58	0.57

Significant differences were reported between the 2 groups regarding AS disease activity indices (BASFI, BASDAI and BASMI) (P = 0.00), CRP (P = 0.02) and CIMT (P = 0.02).



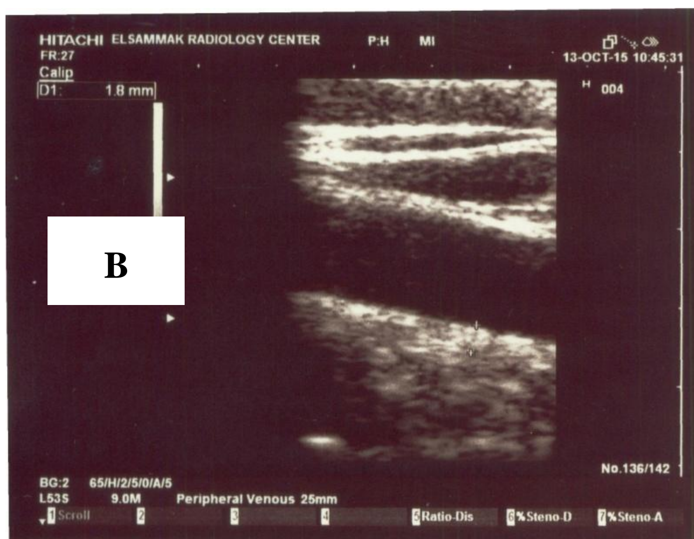


Fig. (1): longitudinal scan of common carotid artery with linear 9 MHz transnducer showing: A) Normal intima-media thickness measuring 0.6 mm. B) Increased intima-media thickness measuring 1.8 mm

DISCUSSION

This study was designed to evaluate subclinical atherosclerosis and endothelial dysfunction sonographically and by serological markers and assess its correlation with disease activity in patients with AS compared with healthy controls.

Endothelial dysfunction is a key early event in atherogenesis, appearing long before the formation of structural atherosclerotic changes[22]. Traditional risk factors, such as higher BMI, smoking and impaired lipid status, are partly responsible for the development of endothelial dysfunction [23]. Current data suggest that inflammation has a role in the development of endothelial impairment[24]. Indeed, chronic inflammation leads to accelerated atherogenesis, particularly in patients with rheumatoid arthritis[25].

AS is a chronic inflammatory rheumatic disease of the spine that affects between 0.2 and 0.9% of the population [26]. There are several characteristic extra-articular manifest-ations involving organs such as the eye, gastrointestinal system, kidneys, lungs and heart [27]. Although mortality from circulatory disease has been found to be increased [28], there is no evidence that it is due to atherosclerosis [29].

In this study 42 AS patients and 42 healthy controls matched for age, sex and smoking status were studied. The mean age of AS patients was 29.6 years, M/F was 36/6, mean height was 164.9 cm, mean weight was 65.7 kg, mean BMI was 19.1 kg/m^2, mean waist circumference was 78.1 cm and mean MAP was 86.1 mmHg. The mean age of controls was 30.6 years, M/F was 29/13, mean height was 161.9 cm, mean weight was 60.3 kg, mean BMI was 21.3 kg/m^2, mean waist circumference was 82.1 cm and mean MAP was 86 mmHg. Significant differences between AS patients and controls reported in height, weight, BMI and waist circumference.

In our study, in AS patients mean disease duration was 11.2 years, 95.2% were using NASIDs (Indomethacin was the commonest drug used), 11.9% were on Etanercept, 47.6% on Sulfasalazine, 4.9% on Methotrexate, 4.9% were using both. The mean BASMI, BASDAI and BASFI scores were 2.9, 3.9, 2.9 respectively.

Our study revealed that AS patients had significantly higher CRP, CD141, VWF and UT-II with mean 21.7, 28.9, 194.5 and 11.4 respectively. With no significant difference between the 2 groups regarding lipid profile and fasting glucose.

vWF is a glycoprotein synthesized by endothelial cells and megakaryocytes and released into the circulation by endothelial cells [30]. Thus, higher levels of this protein may reflect endothelial injury. In a previous study, Divecha et al. showed increased vWF levels in men with AS compared to those in healthy subjects; no correlation was found with CRP, IL-6, BASDAI, and vWF[31].

Thrombomodulin is another glycoprotein that is predominantly expressed on vascular endothelial cells. After proteolytic cleavage from the endothelial surface, sTM can be detected in plasma and has been suggested to be a marker of endothelial function. Thrombo-modulin may reflect the severity of endothelial damage, as clinical studies have demonstrated a significant association between sTM and atherosclerosis [32, 33, 34]. Elevated levels of sTM have also been reported in various inflammatory rheumatic diseases, and some of these studies have shown a relationship with sTM and diseasem activity [35, 36].

Human UT-II is a cyclic peptide with potent vasoconstrictive activity in vascular tissue [37,38,39]. Increased levels of circulating UT-II have been reported as being associated with endothelial injury in conditions such as diabetes mellitus and hypertension [37, 38, 39]. Urotensin II exerts its effect via the urotensin receptor, which is up-regulated by inflammatory stimuli. The activation of this pathway stimulates endothelial and smooth muscle cell proliferation; therefore, UT-II and its receptor may have a role in the initiation and/or progression of atherosclerosis [37, 38, 39]. This is the first study considering the urotensin pathway in inflammatory rheumatic diseases and reporting significantly increased levels of UT-II in AS patients. Notably, both the patients and controls had similar blood pressure levels, glucose and serum lipid concentrations, and age distributions, which might alter the UT-II levels. Therefore, we can speculate that disease-related factorsjriigiitjtiave affected the UT-II concentrations.

Not only plasma levels were similar in AS and control group, but also no correlation was found between minimally modified LDL and CRP levels (data not shown). Whether the same concept can be applied to oxidized LDL levels in patients with AS remains to be exploited. In patients with SLE and RA, not only oxidized LDL, but also their autoantibodies, are associated to atherosclerosis [40, 41].

In the current study, AS patients had significantly higher CIMT ($P = 0.00$), FMD ($P = 0.03$) and ABI ($P = 0.00$).

Peters et al. found a greater IMT in AS patients in comparison with controls [42]. However, the authors also found a high CV risk factor profile in patients with AS, and some of these risk factors (lipids and BMI) were associated with a greater carotid IMT and increased arterial stiffness. No association between large-vessel properties and higher Bath AS indices or CRP values were found. Also, Mathieu et al. [43] found significantly increased IMT in the AS

group compared with healthy controls. However, after adjustment for confounding factors, only an underlying trend towards increased IMT was present. IMT was positively correlated with tobacco use and blood pressure but not correlated with CRP level or mSASS. In the AS group, IMT was correlated with traditional risk factors, such as smoking and systolic blood pressure.

Another study show that there is a higher prevalence of subclinical atherosclerosis in AS patients compared to controls as evidenced by a higher CIMT and a greater prevalence of impaired FMD of brachial artery. CIMT was positively correlated with BASMI indicating that those with more severe disease have a greater probability of having subclinical atherosclerosis. ABI was marginally higher in AS cases as compared tocontrols (1.16 vs 1.1), the reason for which is unclear [44].

Our study revealed significant differences were reported between the 2 groups regarding AS disease activity indices (BASFI, BASDAI and BASMI) (P = 0.00), CRP (P = 0.02) and CIMT (P = 0.02).

Conclusion

The present study provides evidence of increased association of impaired endothelial function in AS subjects compared to controls. This should provide impetus to early intervention strategies to prevent accelerated atherosclerosis which would help in reducing the cardiovascular morbidity and mortality associated with this disease.

REFERENCES

1- Braun J and Pincus T. Mortality, course of disease and prognosis of patients with ankylosing spondylitis. Clin Exp Rheumatol. 2002; 20 (28) : S16-S22.

2- Sokka T, Abelson B, and Pincus T. Mortality in rheumatoid arthritis: 2008 update. Clin Exp Rheumatol. 2008; 26 (51): S356-S61.

3- Kaprover E, Little AH, Graham DC and Rosen Ps. Ankylosing spondylitis: survival in men with and without radiography. Arthritis Rheum. 1980; 23:57-61.

4- Khan MA, Khan MK and Kushner I. Survival among patients with ankylosing spondylitis: a life-table analysis. J Rheumatol. 1981; 8 : 86-90.

5- Lehtinen K. Mortality and cause of death in 398 patients admitted to hospital with ankylosing spondylitis. Ann Rheum. Dis. 1993; 52 : 174-6.

6- Radford EP, Doll R and Smith PG. Mortality among patients with ankylosing spondylitis not given x-ray therapy. N Eng J Med. 1977; 297: 572-6.

7- Han C, Robinson DW Jr, Hackett MV, Paramorre LC, Fraeman KH and Bala MV. Cardiovascular disease and risk factors in patients with rheumatoid arthritis, psoriatic arthritis and ankylosing spondylitis. J Rheumatol. 2006; 33: 2167-72.

8- Azevedo VF and Pecoits-Filho R. Atherosclerosis and endothelial dysfunction in patients with ankylosing spondylitis. J Rheumatol Int. 2010; 30 (11): 1411-6.

9- Bots ML, Dijk JM, Oren A and Grobbee DE. Carotid intima-media thickness, arterial stiffness and risk of ccardiovascular disease: current evidence. J Hypertens. 2002; 20: 2317-25.

10- Copkin E, Kiris A, Karkucak M, Durmus I, Gokmen F, Cansu A et al. Investigation of effects of different treatment modalities on structural and functional vessel wall properties in patients with ankylosing spondylitis. Joint Bone Spine. 2011; 78 (4): 378-82.

11- Sari I, Okan T, Akar S, Cece H, Altay C, Secil M et al. Impaired endothelial function in patients with ankylosing spondylitis. Rheumatology (Oxford) 2006; 45 (3): 283-286.

12- Sari I, Kebapcilar L, Taylan A, Biligr O, Kozaci D, Yildiz Y et al. Fetuin-A and interleukin-18 levels in ankylosing spondylitis. Thi J Rheum Dis. 2010; 13 (1): 75-81.

13- Jaevisalo MJ, Juonala M and Raitakari OT. Assessment of inflammatory markers and endothelial function. Curr Opin Clin Nutr Metab Care. 2006; 9 (5) : 547-552.

14- Van der Linden S, Valkenburg HA and Cats A. Evaluation of diagnostic criteria for ankylosing spondylitis. A proposal for modification of the New York criteria. Arthritis Rheum. 1984 Apr; 27 : 361-3687.

15- Chobanian AV, Bakris GL, Black HR et al. Seventh report of the joint National Committee on prevention, detection, evaluation and treatment of high blood pressure. Hypertension. 2003 Dec; 42: 1206-1252.

16- Jenkinson TR, Mallorie PA, Whitelock HC, Kennedy LG, Garrett SL and Calin A. Defining spinal mobility in ankylosing spondylitis (AS). The bath AS metrology ibdex. J Rheumatol. 1994 Sep; 21: 1694-1698.

17- Garrett S, Jenkinson T, Kennedy LG, Whitelock H, Gaisford P and Calin A. A new approach to defining disease status in ankylosing spondylitis: the bath ankylosing spondylitis disease activity index. J Rheumatol. 1994 Dec; 21: 2286-2291.

18- Calin A, Garrett S, Whitelock H et al. A new approach to defining functional ability in ankylosing spondylitis: the development of the bath ankylosing spondylitis functional index. J Rheumatol. 1994 Dec; 21: 2281-2285.

19- Moens AL, Goovaerts I, Claeys MJ and Vrints CJ. Flow-mediated vasodilatation: a diagnostic instrument, or an experimental tool? Chest. 2005 June; 127: 2254-2263.

20- Mc Dermott MM, Criqui MH, Liuk et al. Lower ankle/brachial index, as calculated by averaging the dorsalis pedis and posterior tibial arterial pressures and association with leg functioning in peripheral arterial disease. J Vasc Surg. 2000 Dec; 32: 1164-1171.

21- Olin JW. The clinical evaluation and office base detection of peripheral arterial disease. In: Hirsch AT, Olin FW ed. An office-based approach to the diagnosis and treatment of peripheral arterial disease. Belle Meade, NJ. Excerpta Medica Inc. 1998; 10-17.

22- Agewall S. Is impaired flow-mediated dilatation of the brachial artery a cardiovascular risk factor? Curr Vase Pharmacol 2003; 1:107-9.

23- Chan NN, Colhoun HM, Vallance P. Cardiovascular risk factors as determinants of endothelium-dependent and endothelium-independent vascular reactivity in the general population. J Am Coll Cardiol 2001 ;38:1814-20.

24- Sattar N. Inflammation and endothelial dysfunction: intimate companions in the pathogenesis of vascular disease? Clin Sci (Lond) 2004; 106:443-5.

25- Sattar N, McCarey DW, Capell H, Mclnnes IB. Explaining how "high-grade" systemic inflamm-ation accelerates vascular risk in rheumatoid arthritis. Circulation 2003; 108:2957-63.

26- Braun J, Bollow M, Remlinger G et al. Prevalence of spondyl-arthropathies in HLA-B27 positive and negative blood donors. Arthritis Rheum 1998: 41:58-67.

27- Lautermann D, Braun J. Ankylosing spondylitis-cardiac manifestations. Clin Exp Rhcumatol 2002; 20 (6) : Sll-5.

28- Lehtinen K. Mortality and causes of death in 398 patients admitted to hospital with ankylosing spondylitis. Ann Rheum Dis 1993;52:J74-6.

29- Divecha H, Sattar N, Rumley A, Cherry L, Lowe GD, Sturrock R. Cardiovascular risk parameters in men with ankylosing spondylitis in comparison to non-inflammatory control subjects: relevance of systemic inflammation. Clin Sci (Lond) 2005; 109:171-6.

30- Vischer UM (2006) von Willebrand factor, endothelial dysfunction, and cardiovascular disease. J Thromb Haemost 4(6): 1186-1193.

31- Divecha H, Sattar N, Rumley A, Cherry L, Lowe GD, Sturrock R (2005) Cardiovascular risk parameters in men with ankylosing spondylitis in comparison with non-inflammatory control subjects: relevance of systemic inflammation. Clin Sci (Lond) 109(2): 1 71-176.

32- Blann AD, McCollum CN (1999) von Willebrand factor and soluble thrombomodulin as predictors of adverse events among subjects with peripheral or coronary atherosclerosis. Blood Coagul Fibrinolysis 10(6):375-380.

33- Blann AD, Seigneur M, Steiner M, Boisseau MR, McCollum CN (1997) Circulating endothelial cell markers in peripheral vascular disease: relationship to the location and extent of atherosclerotic disease. Eur J Clin Invest 27(11):916-921.

34- Li YH, Shi GY, Wu HL (2006) The role of thrombomodulin in atherosclerosis: from bench to bedside. Cardiovasc Hematol Agents Med Chem 4 (2) : 183-187.

35- Vanhoutte PM, Shimokawa H, Tang EH, Feletou M (2009) Endothelial dysfunction and vascular disease. Acta Physiol (Oxf) 196 (2): 193-222.

36- Kotajima L, Aotsuka S, Sato T (1997) Clinical significance of serum thrombomodulin levels in patients with systemic rheumatic diseases. Clin Exp Rheumatol 15(1):59-65.

37- Loirand G, RolJi-Derkinderen M, Pacaud P (2008) Urotensin II and atherosclerosis. Peptides 29 (5):778-782.

38- Kemp W, Roberts S, Krum H (2005) Urotensin IT: a vascular mediator in health and disease. Curr Vase Pharmacol 3 (2): 159-168.

39- Pakala R (2008) Role of urotensin II in atherosclerotic cardiovascular diseases. Cardiovasc Revasc Med 9 (3): 166-178.

40- Hahn BH, McMahon M. Atherosclerosis and systemic lupus erythematosus: the role of altered lipids and of auto-antibodies. Lupus. 2008; 17: 368-70.

41- Peters MJ, van Halm VP, Nurmohamed MT, Damoiseaux J, Tervaert JW, Twisk JW, et al. Relations between auto-antibodies against oxidized low-density lipoprotein, inflamm-ation, subclinical atherosclerosis, and cardiovascular disease in rheumatoid arthritis. J Rheumatol. 2008:35:1495-9.

42-[Peters MJ, van Eijk 1C, Smulders YM, Serne E, Dijkmans ;BA, van der Horst-Bruinsma IE, et al. Signs of accelerated preclinical atherosclerosis in patients with ankylosing spondylitis. J Rheumatol. 2010; 37 (l):161-6.

43- Mathieu S, Joly H, Baron G, Tournadre A, Dubost J, Ristori JM, et al. Trend towards increased arterial stiffness or intima-media thickness in ankylosing spondylitis patients without clinically evident cardiovascular disease. Rheum-atology. 2008, 47 (8): 1203-7.

44- Sharma S.K., Prasad K.T., Handa R. et al. Increased prevalence of subclinical atherosclerosis in ankylosing spondylitis. Indian Journal of Rheumatology., 2015; 10 : 53-57.