Correlation of echocardiographic findings of pulmonary hypertension with six-minute walk test and plasma pro b-type natriuretic peptide level in systemic lupus erythematous

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Abstract

Introduction: Pulmonary arterial hypertension (PAH) is an increasingly recognized complication of systemic lupus erythematous (SLE), which may remain undiagnosed if asymptomatic.

Objective: This study aimed to determine the correlation between echocardiographic findings of PAH and six-minute walk test (6WMT) and serum pro b-type natriuretic peptide (proBNP) level in patients with SLE.

Methods: This cross-sectional study was performed on 50 SLE patients selected from patients referring to the outpatient's department of the Rheumatology Clinic at Imam Reza Hospital in Mashhad, Iran, from July 2013 through September 2014, using resting transthoracic echocardiography to estimate systolic pulmonary artery pressure (sPAP). Variables were summarized as counts and/or percentages or as mean±SD. Inter-group comparisons were made performing two-tailed Fisher's exact test or Mann–Whitney U test, using SPSS 22.

Results: In general, five out of fifty patients were diagnosed to have PAH with sPAP>30 mmHg (range: 31-40 mmHg) based on echocardiographic findings. Spirometric parameters did not show any differences between the two groups (p>0.05), while the difference in total distance walked during six minutes and serum proBNP level between SLE patients with and without PAH was significant (P<0.05). A high correlation was found between PAP and serum proBNP level, but not between PAP and the distance walked during six-minutes in SLE patients.

Conclusion: The point prevalence of PAH in SLE patients was 10%; the significant correlation between PAP and serum proBNP level suggests that it can be used as a valuable marker for early diagnosis of asymptomatic pulmonary hypertension in patients with SLE.

Keywords: Systemic lupus erythematosus, Pulmonary arterial hypertension, Six-minute walk test, Serum proBNP levels, Transthoracic echocardiography

1. Introduction

Systemic lupus erythematous (SLE) is a well-recognized autoimmune multisystem disorder with frequent respiratory and cardiac manifestations (1) that can lead to high rates of morbidity and mortality (2, 3). To improve prognosis of pulmonary hypertension, new treatment methods have been proposed (4, 5). A multitude of studies were conducted on patients with idiopathic pulmonary hypertension (PAH) and connective tissue disease patients with systemic sclerosis (SSc) (4-6). Approximately 0.5-43% of the cases of PAH are assumed to be associated with SLE (7-10). These results were reported in retrospective studies involving large numbers of SLE patients over a 5

to10-year period or cross-sectional studies on limited groups of patients (11). The wide variability in these prevalence estimates reflect the differences in PAH definitions, population groups, sample sizes, and the applied diagnostic methods (11). The nonspecific nature of symptoms associated with PAH such as dyspnea, fatigue, palpitation, and syncope can result in delayed diagnosis of PAH in SLE patients, indicating the need for appropriate screening methods to identify PAH (11). Although the gold standard test for diagnosis of PAH is right heart catheterization (RHC), its invasiveness and cost make it an unsuitable screening tool (12). Transthoracic Doppler echocardiography, however, is considered a safe, sensitive, and specific tool for screening PAH and assessing its severity in SSc patients (13, 14). In the current study, we aimed to prospectively determine the prevalence of PAH in SLE patients using echocardiography and assess the value of pulmonary function tests, 6MWT, and proBNP level as screening tests for PAH. We also determined the correlation of six-minute walk test (6WMT) and proBNP level with pulmonary arterial pressure as estimated by echocardiography in lupus patients.

2. Material and Methods
2.1. Study design and setting
This cross-sectional study was carried out on fifty SLE patients, selected from patients referring to the outpatient's department of the Rheumatology Clinic at Imam Reza Hospital in Mashhad, Iran, from July 2013 through September 2014. The sampling method was convenient and the cases were among the referrals to our teaching hospital.

2.2. Selection criteria
The inclusion criteria comprised of patients meeting the 1982 revised criteria of the American College of Rheumatology (ACR) for diagnosis of SLE (15), who did not present evidence of other connective tissue diseases. The exclusion criteria included 1) left heart disease, 2) consumption of drugs such as fenfluramine and aminorex, 3) HIV infection, 4) portal hypertension, 5) chronic hemolytic, 6) anemia, 7) pregnancy, 8) significant renal impairment (creatinine≥150 mmol/L), 9) history of unstable angina or a myocardial infarction during the previous month, 10) resting tachycardia (heart rate≥120 beats/min), 11) uncontrolled hypertension≥180/100 mmHg, 12) history of cardiac arrhythmia, and 13) arthritis and other musculoskeletal diseases.

2.3. Data sources and measurements
2.3.1. Questionnaire
The patients completed a questionnaire including the patients' demographic data, medication history, and previous medical documents, and underwent transthoracic echocardiography to identify evidence of PAH. Blood samples were obtained for evaluation of serum proBNP level; then, a pulmonary function test (spirometric test) was run to investigate subclinical pulmonary involvement, and 6 MWT was carried out to assess exercise capacity. A questionnaire was completed based on the patients' demographics and previous medical documents including previous history of Raynaud's phenomenon, previous history of thrombosis, certain autoantibody positivity at any time during the course of the disease, and medication history.
2.3.2. Echocardiography
Two-dimensional, M-mode color Doppler echocardiography was performed for all the patients at rest using a transthoracic echocardiography device by an expert cardiologist, who had no knowledge of the patients' past medical history. Parasternal long and short axis, four-chamber projections, and evidence of pulmonary hypertension (PH) including systolic PAP, tricuspid annular plane systolic excursion (TAPSE), left ventricular ejection fraction (EF), and pericardial effusion were recorded in all the patients. The World Health Organization defines PAH as mean PAP>25 mmHg at rest and >30 mmHg with exercise, in the presence of normal pulmonary capillary wedge pressure as measured by RHC (11). Similar to former studies, the patients with sPAP of higher than 30 mmHg at rest were diagnosed with PHA, using echocardiography as a screening tool for PH (5).
2.3.3. Blood sampling and assay
An aliquot of the serum sample was collected and stored in gel separator tubes on the same day. Samples were transferred to the Chemical Pathology Department in serum gel tubes at room temperature, where they were centrifuged and stored at -20oC for later analysis. Thereafter, the electrochemiluminescence immunoassay was performed for quantitative in-vitro determination of NT-proBNP in human serum. According to the manufacturer of the assay, the analytical range for NT-proBNP assay is 5-35,000 pg/ml and the expected NT-proBNP values (95th percentile) for different age groups and each gender are summarized in Table 1. The aforementioned NT-proBNP values adjusted for age and gender were applied to classify our study patients with and without high proBNP concentrations.

2.3.4. Spirometry

Spirometry was performed according to the standards of the American Thoracic Society at the time of examination, by one technician (16). To ascertain repeatability of the test, spirograms were obtained until three admissible forced vital capacity (FVC) maneuvers were conducted (17). We employed Chest Compression System to gauge FVC, and forced expiratory volume in 1 second (FEV1). The observed values for each individual were compared with those predicted for age, gender, and height. Afterwards, we reported the outcomes as the percentage of predicted values. According to ATS standard, when pulmonary function parameters were below 80% of the predicted value, they were considered abnormal (16). Obstructive pattern was characterized by values below 70% of FEV1/FVC ratio and less than 80% of predicted FEV1; further, the restrictive pattern was characterized by FVC values less than 80% of predicted normal or elevated FEV1/FVC ratio (18). With the subjects in sitting position, measurements were performed without bronchodilator administration.

2.3.5. Six-minute walk test

The 6MWT was performed for all the patients using a standardized protocol in accordance with the American Thoracic Society guidelines to estimate exercise capacity (19). This test has good reproducibility and is easy to administer, well-tolerated by patients with respiratory derangements, and is a better reflection of patients' daily activities compared to the shuttle walk test. To perform the 6 MWT, a 15-m walking course was prepared in a corridor with turnaround points marked with bright color tape at 0 and 15 m. Every 5 m was marked with white tape. In case of fatigue, the patients could sit on chairs provided alongside the corridor. All the participants were asked to wear comfortable clothing, footwear, and walking aids. Before conducting the test, the modified Borg scale for scoring dyspnea and fatigue was used to record the scores at baseline and post-exercise stage, and a pulse oximeter and heart rate monitor were attached to the patients. The subjects were notified that the objective of the study was to walk as far as possible during six minutes and that they were allowed to slowdown, stop and rest (if required), and resume walking. The countdown timer, which was set to six minutes, was started with the patients commencing the test. We informed the participants of the remaining time every minute. We recorded the completed laps and the distance walked in the last partial lap. At the end of the test, Borg dyspnea and fatigue scores were re-recorded, and we made sure that none of the patients had any problems (20). Borg scale is a tool for describing the intensity of perceived dyspnea and fatigue on a scale of zero to ten. At this stage of the study, we recorded the total distance walked (meters) during six minutes, pre- and post-exercise Borg dyspnea and fatigue scores, heart rate (rate/min), and oxygen saturation (%).

2.4. Ethical considerations

The study protocol was approved by the regional Ethics Committee of Mashhad University of Medical Sciences, and written informed consent was obtained from all the subjects for all testing procedures before entering the study.

2.5. Statistical analysis

Variables were summarized as counts and/or percentages or as mean±SD. Inter-group comparisons were made performing two-tailed Fisher's exact test or Mann-Whitney U test, using IBM© SPSS© Statistics version 22 (IBM© Corp., Armonk, NY, USA). In spite of being aware of limited significance due to small sample size, correlations between variables were assessed using Pearson product-moment correlation coefficient. A p-value less than 0.05 was considered statistically significant.

Table 1. NT-proBNP values (95th percentile) in different age groups and gender

Age (year)	95th percentile NT-proBNP value	
	Women	Men
<50	125 pg/ml	64 pg/ml
50-50	186 pg/ml	135 pg/ml
>60	204 pg/ml	194 pg/ml

3. Results

This study was carried out on 50 SLE patients meeting the updated ACR classification criteria for SLE. Patients with SLE were classified into two groups. Group I included five SLE patients with PAH, and group II comprised of 45 SLE patients without PAH. Demographic data, clinical and paraclinical features, the results of echocardiography, spirometry, serum proBNP level, and 6MWT, as well as the comparison between the two groups are summarized in Table 2. The study groups comprised of 50 SLE patients, 48 (94%) of whom were females. The mean age of the participants was 29 years (age range: 21 to 48 years) and mean disease duration was four years (range: 1 to 17 years). All the patients were non-smokers. Comparison of lupus patients with and without PAH showed no

significant differences with respect to gender, age distribution, body mass index (BMI), and disease duration (p>0.05). Past medical history of the patients revealed that none of them had chronic respiratory symptoms, but Raynaud's phenomenon occurred in 30% of the patients and only one patient (2%) had a clinical clotting episode in the past. Twelve patients (24%) had previously undergone immunosuppressive therapy for severe organ involvement other than the lung. The frequency of Reynaud's phenomenon, previous history of thrombosis, and prior therapy with immunosuppressive drugs showed no significant differences between the two groups of SLE patients (p>0.05). Anti-U1-ribonucleoprotein (anti-U1RNP), rheumatic factor, and anti-phospholipid antibody were positive in 17 (34%), 7 (14%), and 11 (22%) patients, respectively. SLE patients with PAH did not show a significantly higher number of cases with positive auto-antibodies such as RF, antiU1RNP, and anti-phospholipid antibodies, compared to SLE patients without PAH (p>0.05). According to echocardiographic findings, all the patients (100%) had normal left ventricular function and none of them had valvular heart disease or pericardial effusion. Five out of fifty (10%) patients had sPAP>30 mmHg (pulmonary hypertension) on echocardiography. The difference between the lupus patients with and without PAH in terms of ejection fraction of left ventricle was not statistically significant (P>0.05), but tricuspid annular plane systolic excursion (TAPSE) and sPAP differed significantly between the two groups of SLE patients (p<0.05). Pulmonary function tests in all the studied patients revealed restrictive lung disease in 10 patients (20%) and normal spirometry in the remaining (80%) cases. There was no significant difference between SLE patients with and without PAH with regard to the presence of restrictive and/or obstructive lung diseases (p>0.05). The 6MWT was successfully completed by all the patients (n=50). In this test, mean six-minute walk distance (6MWD) was 480 m (range: 335–562 m). During this time, oxygen saturation did not occur in any of the patients. At baseline, the dyspnea and fatigue scores were zero, while at the post-exercise stage, mean Borg dyspnea and fatigue scores were 1.37 and 1.49. The difference in total distance walked during six minutes between SLE patients with and without PAH was significant (p=0.04), whereas the pre- and post-exercise Borg dyspnea and fatigue scores were not significantly different between the groups (p=0.05). The mean serum proBNP level measured in this study was 99.64 pg/ml. Comparison of SLE patients with and without PAH showed a significant difference in terms of serum proBNP level (p<0.05).

Table 2. Comparison of gender distribution in SLE patients with and without PAH

Parameter	Total [†]	SLE with PAH[†]	SLE without PAH[†]	P value*
Age (year)	29.10±5.53	31.4±4.9	28.8±5.5	0.267
Gender (F:M)	48:2 (96:4)	5:0 (100:0)	43:2 (95.5:4.5)	0.512
BMI(kg/m^2)	23.46±3.2	23.7±2.8	23.43±3.27	0.788
Disease duration (year)	4.14±2.80	2.9±4.2	1.4±3.2	0.488
Smoking Hx	0	0	0	-----
Immunosuppressive Tx	12 (24)	2 (40.0)	10 (22.2)	0.58
Thrombus	1 (2)	0 (0.0)	1 (2.2)	1.00
Raynaud's	15 (30)	4 (80.0)	11 (24.4)	0.24
Rheumati Factor	7 (14)	3 (60.0)	4 (8.9)	0.16
Anti RNP	17 (34)	4 (80.0)	13 (28.9)	0.40
APL Ab	11 (22)	3 (60.0)	8 (17.8)	0.64
FEV$_1$ (% predict)	80.78±9.3	83.69±8.63	80.45±9.47	0.28
FEV$_1$ (L)	2.76±0.46	2.77±0.47	2.63±0.38	0.48
FVC (% predict)	82.9±10.2	82.54±10.46	86.40±7.90	0.29
FVC (L)	3.13±0.52	3.14±0.54	3.09±0.37	1.00
FEV1/FVC	88.15±4.5	88.48±4.48	85.12±3.88	0.75
LVEF	57.1±3.7	57.11±3.92	57.0±2.73	0.951
TAPSE(mm)	22.1±3.1	18.40±2.88	22.51±2.87	0.007
SPAP(mmHg)	24.3±5.4	37.60±2.51	22.91±3.21	0.000
MWD (m) 6	480.6±49.9	443.20±34.96	484.76±49.95	0.04
Borg dyspnea scale BT.	0	0.00	0.00	---
Borg dyspnea scale AT.	1.37±0.72	2.00±0.70	1.30±0.69	0.05
Borg fatigue scale BT.	0	0.00	0.00	---
Borg fatigue scale AT.	1.49±0.84	2.20±0.83	1.41±0.81	0.05
pro BNP level (pg/ml)	99.6±87.8	347.00±34.56	72.16±27.46	0.000

[†] Values are expressed as mean±standard deviation or counts and percentages; * p value calculated by Mann Whitney U test / Fisher's exact test 2 sided for comparison between groups of SLE patients with and without PAH.

4. Discussion

In this study, we found that the prevalence rate of PAH in SLE patients was 10%, which is within the median range of prevalence rates reported from previous studies worldwide (11). All five of the patients diagnosed with PAH in our study had sPAP<40 mmHg, suggesting that PAH in this study was predominantly mild with no symptoms. The role of left heart disease as a contributing factor in PAH is negligible in the SLE patients participating in our study as none of them had significant left ventricular dysfunction and valvular disease as evaluated by echocardiography. SLE patients with PAH had a shorter mean disease duration (3.2±1.48 years) in comparison with SLE patients without PAH (4.24±2.90 years), but this difference was not statistically significant. Our results are congruent with those of another study by Shereen R. Kamel et al., who compared the demographic and clinical features of SLE patients with and without PAH (21). The frequency of Raynaud's phenomenon in both groups of SLE patients showed no significant differences. In a comprehensive literature review conducted in China on seven former studies, 51.2% of lupus patients with PH had Raynaud's phenomenon contrary to 19.9% of lupus patients (22). Those studies also reported the correlation of Raynaud's phenomenon with PAH (22). In contrast, in a large cohort of SLE patients in the UK, the association of Raynaud's phenomenon with PAH in SLE patients was not considered statistically significant (11). There was no significant difference between the two groups of patients with regard to antiphospholipid antibody positivity, which was also true for rheumatic factor (RF) and antiU1RNP positivity. A recent cohort study on 200 lupus patients from a tertiary care center did not demonstrate any association between PAH prevalence of 17.5% and the level of anti-phospholipid antibodies (23), but in another cohort of 288 SLE patients, with PAH prevalence of 4.2%, a statistically significant association between antiphospholipid antibody and PAH was noted (11). In the present study, most of the patients presented with normal spirometry, where abnormal pattern was only noted in 20% of the patients. All the patients with abnormal spirometry showed restrictive pattern. None of the spirometric parameters were significantly different between the patients with and without PAH and had no value as a screening test for PAH in our study, which is in agreement with results of most previous studies (11). The difference between the total distance walked in the two groups was considered significant; however, no significant correlation was found between the distance walked in 6MWT and sPAP in SLE patients in this study, which is in line with the results of a large cohort study of 283 SLE patients in the UK, in which 6MWT could not differentiate between patients with or without PAH (11). The mean serum proBNP level of the lupus patients with high sPAP was considerably higher at 347 pg/ml in comparison to 72.16 pg/ml in the group with normal sPAP (p<0.05). This is in contrast with the results of another large cohort study conducted by Prabu et al., who found no statistical difference between the proBNP levels in lupus patients with and without PAH (11). Moreover, there was a significant correlation between serum proBNP level and PAP in the current study, and a weak indirect correlation was noted between serum proBNP level and distance walked during six minutes. Nevertheless, Prabu et al. proposed that proBNP level was significantly associated with six minute walk distance, but it was weakly linked with sPAP (11).

5. Limitations of the study

There were some limitations to our study as many eligible patients refused to participate due to various reasons including active disease, distance required to travel to the center, work commitments, inconvenience, and lack of interest in participating in the study, which might have biased our results, and in turn, undermined the point prevalence rate of PAH in our study. Our measurements of PAP were indirect and based on screening criteria of echocardiography and were not confirmed by the gold standard RHC measurement of mean PAP, as we considered it unethical to catheterize patients with only mild disease due to its invasive nature. Limited sample size and lack of a control group due to paucity of time and financial resources were other limitations of our study. The overall mild PAH in the current study suggested lack of association between diverse risk factors and screening tests for PAH related to SLE.

6. Conclusions

It seems there was no association between PAH and history of thrombosis, Raynaud's phenomenon, or autoantibody positivity in lupus patients. The association of smoking, as a pulmonary risk factor, with PAH in SLE patients remains unclear in our study since none of the patients participating in this study had history of smoking. Considering the PFT results, it indicated to us that subclinical pulmonary involvement is not rare in lupus patients, but spirometric parameters did not show any differences between the groups and may not have screening potential for PAH associated with SLE. The 6MWT distance exhibited no significant correlation with PAP, but showed a weak negative correlation with serum proBNP level in SLE patients. Serum proBNP level was significantly correlated with PAP in patients with SLE. To confirm these results, larger cohort studies on SLE-PAH patients with

more severe PAH disease should be conducted. We recommend regular evaluation of patients with SLE, regardless of disease duration, for the development of PAH.

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Conflict of Interest:
There is no conflict of interest to be declared.

Authors' contributions:
All authors contributed to this project and article equally. All authors read and approved the final manuscript.

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