Original Article

Serum-Soluble ST2 (sST2) and NT-proBNP Levels in Children With Pulmonary Arterial Hypertension

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ABSTRACT

Background: Soluble ST2 (sST2) is a member of the interleukin-1 receptor family and is considered a novel biomarker of inflammation, fibrosis, and cardiac stress. Additionally, sST2 is accepted by guidelines as a measure of risk stratification in patients with heart failure.

Methods: Our study enrolled 53 subjects: 23 patients who were followed up for pulmonary arterial hypertension (PAH) and were prescribed different medications and 30 healthy children admitted to the pediatric cardiology outpatient clinic with chest pain or innocent murmurs as the control group. The plasma concentration of NT-proBNP was analyzed via the electrochemiluminescence method, and the sST2 level was analyzed via the ELISA method.

Results: The mean age was 13.9 years (5.5–18 y) in the case group and 9.6 years (3–17 y) in the control group. The mean NT-proBNP level was significantly higher in the patient group than in the control group (763.73±2432.67 pg/mL vs 51.71± 30.08 pg/mL; $P<0.01$). The mean sST2 level was 1469.26±510.9 pg/mL in the patient group and 1151.30±655.99 pg/mL in the control group ($P>0.05$).

Conclusions: Our results suggest that sST2 could be a significant indicator of right heart failure and cardiovascular mortality in children, as well as a novel biomarker of PAH. However, we found that the serum sST2 level was not as useful as the serum NT-proBNP level in this regard. Further studies with larger patient series are needed to evaluate sST2 as a biomarker in patients with PAH. ***(Iranian Heart Journal 2022; 23(4): 46-51)***

KEYWORDS: Pulmonary hypertension, Biomarker, NT-proBNP, Soluble ST2

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Soluble ST2 (sST2) is a member of the interleukin-1 receptor family and is regarded as a novel biomarker of inflammation, fibrosis, and cardiac stress. Furthermore, sST2 is accepted by guidelines as a measure of risk stratification in patients with heart failure.[1-3] Signaling cardiac fibrosis and restructuring, sST2 has further been suggested as an important indicator of right heart failure and cardiovascular mortality in children while it may also be taken as a biomarker of pulmonary arterial hypertension (PAH).[4-6]

In the present study, we evaluated the value of serum sST2 and N-terminal pro-brain natriuretic peptide (NT-proBNP) levels as biomarkers in the diagnosis and prognosis of PAH in a pediatric population.

METHODS

Having been granted the institutional ethics committee's permission, we conducted the current study between November 2018 and October 2019 at the Department of Pediatric Cardiology, Inonu University. Our study population was comprised of 2 groups: a case group (n=23) and a control group (n=30). The case group patients were followed up for PAH, for which they were prescribed different medications, and the control group consisted of healthy children admitted to the pediatric cardiology outpatient clinic with chest pain or innocent murmurs. The control group shared normal electrocardiography and echocardiography findings, and no issues were recorded at their physical examination. Patients with systemic diseases or infections were excluded from the patient group.

Detailed medical history was taken from the patient group; it included age, sex, weight, height, the body surface area, the diagnosis, the date of diagnosis, hemodynamic data, cardiac catheterization findings, and treatment and follow-up periods. The case group then underwent thorough physical examinations. The functional class of the patients was determined according to the New York Heart Association (NYHA) functional classification criteria.

After the confirmation of PAH diagnosis via left and right heart cardiac catheterization, cardiac catheterization was performed under sedation. For hemodynamic evaluation, right atrial pressure; systolic, diastolic, and mean pulmonary artery pressures; and systemic arterial pressures were measured. Pulmonary flow, systemic flow, flow rates, pulmonary resistance (Rp), systemic resistance (Rs), and the resistance ratio were also calculated during catheterization.[7-10] When required, acute pulmonary vasoreactivity tests were performed after cardiac catheterization. The pulmonary vascular reactivity test results were evaluated in 2 subgroups as positive and negative during catheter angiography. We used the Sitbon and modified Barst criteria to evaluate the pulmonary vascular reactivity test.[8-10]

Written informed consent was obtained from the entire study population. The medical data of the participants in the 2 groups were recorded. Thereafter, venous blood was drawn into EDTA-containing tubes and stored in the form of frozen plasma at −80 °C in aliquots. The plasma concentrations of NT-proBNP were analyzed via the electrochemiluminescence method (BioTek Synergy H1 Biochemistry AutoAnalyzer using the immunoturbidimetric method; NT-proBNP, Siemens IMMULITE 2000 Model Device via chemiluminescence method). Additionally, sST2 levels were analyzed using the enzyme-linked immunosorbent assay (ELISA) method (Human sICAM-1 Elisa Kit RayBiotech Inc USA). The results were recorded in pg/mL.

Statistical Analysis

The SPSS software for Windows, version 17.0, was employed to evaluate the data, and the Mann–Whitney *U* test was utilized to

compare differences between the 2 independent groups for nonparametric data. Additionally, the *t* test was applied to compare independent samples from the groups for data distributions. *P* values smaller than 0.05 were considered significant.

RESULTS

The mean age was 13.9 years (5.5–18 y) in the patient group and 9.6 years (3-17 years) in the control group. In the case group, 3 patients were diagnosed with idiopathic PAH, 6 with postoperative residual PAH (3 of these patients had previously undergone surgery due to ventricular septal defects [VSDs]), 2 with patent ductus arteriosus (PDA), 1 with parachute mitral valve and mitral stenosis, 6 with severe PAH related to congenital heart diseases (2 of these patients had large VSDs, 2 had complete atrioventricular septal defects [AVSDs], and the remaining 2 had PDA), 1 patient with agenesis in the right pulmonary artery, and 7 with the Eisenmenger syndrome. (The condition of 5 of these 7 patients was related to complete AVSDs: 1 patient had single ventricle + double-inlet left ventricle + transposition of the great arteries [TGA], and the Eisenmenger syndrome of another patient was related to single ventricle + TGA).

On the hemodynamic evaluation, the mean pulmonary artery pressure was 62.04±19.7 mm Hg, and the mean aortic pressure was 75.41±12.83 mm Hg. The Qp/Qs ratio was 1.09±(0.2–3.9), Rp was 13.04±9.45 U×m^2, Rs was 21.80±10.9 U×m^2, and Rp/Rs was 0.6 (0.13–1.9). Acute vasoreactivity testing was done for 11 patients. Only 2 of them were considered positive. The mean follow-up period was 3.9 years in the patient group (1–10 y). The clinical and demographic data of the participants are presented in Table 1 and Table 2.

As can be seen in Figure 1 and Figure 2, the mean NT-proBNP level in the patient group was significantly higher than that in the control group (763.73±2432.67 pg/mL vs 51.71±30.08 pg/mL; *P*<0.01). The mean sST2 level was 1469.26±510.9 pg/mL in the patient group and 1151.30±655.99 pg/mL in the control group (*P*>0.05). There was no statistically significant correlation between the mean pulmonary artery pressure and the mean NT-proBNP and sST2 levels (*P*>0.05). The same conclusion was reached concerning the correlation between Rp/Rs and the mean NT-proBNP and sST2 levels (*P*>0.05).

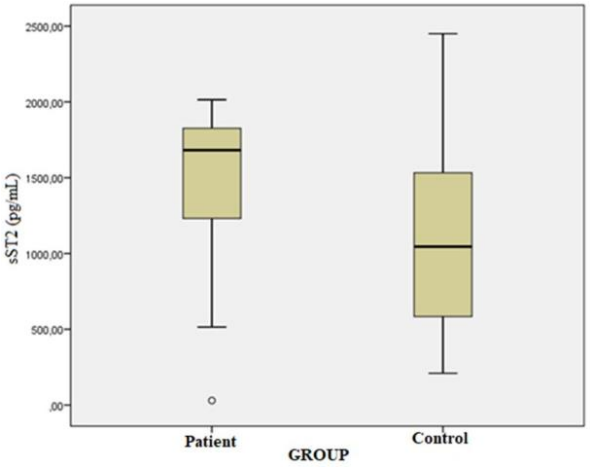

Figure 1: The image presents the mean sST2 level in the case and control groups.
sST2, Serum-soluble ST2

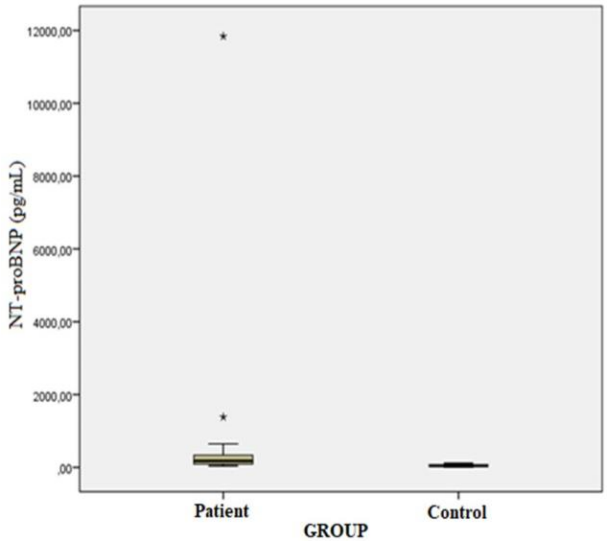

Figure 2: The image illustrates the mean NT-proBNP level in the case and control groups.
NT-proBNP, N-terminal pro-brain natriuretic peptide

Table 1: Diagnosis and hemodynamic data of the case group

Diagnosis	Eisenmenger syndrome	7 patients
	Primary PAH	3 patients
	Postoperative residual PAH	6 patients
	PAH-related congenital heart disease	6 patients
	Agenesis in the right pulmonary artery	1 patient
Hemodynamic Evaluation		
	Qp/Qs	1,09± 0,98 (0,2-3,9)
	Mean pulmonary artery pressure (mm Hg)	62,04± 19,7 (26-99)
	Mean aortic pressure (mm Hg)	75.41 ± 12.83 (43-91)
	Rp (Uxm^2)	13,04 ± 9,45 (1,7-38,8)
	Rs (Uxm^2)	21,80± 10,9 (4,69-45,06)
	Rp/Rs	0,72 ± 0,84 (0.13-1,9)
	Vasoreactivity testing	Negative in 9 patients
		Positive in 2 patients
	Follow-up periods	mean 3.9 years (1-10 years)
	PAH-specific treatment	Bosentan for 14 patients
		Bosentan+Tadalafil for 2 patients
		Bosentan+Tadalafil+Ilioprost for 1 patient

VSD, Ventricular septal defect; PAH, Pulmonary arterial hypertension; AVSD, Atrioventricular septal defect; PDA, Patent ductus arteriosus; DORV, Double-outlet right ventricle; ASD, Atrial septal defect; PAP, Pulmonary artery pressure; Rp, Pulmonary resistance; Rs, Systemic resistance

Table 2: Serum NT-proBNP and sST2 levels in the PAH and control groups

	PAH Group (N=23)	Control Group (N=30)	*P* value
Age (y)	13.9 ± 4.2 (5.5-19)	9.6±4.4 (3-17)	<0.01
Weight (kg)	41.7 ±19.5 (11.4-78)	33.1±15.2 (14-62)	>0.05
NT-proBNP (pg/mL)	763.7±2432.6 (41-11840)	51.7± 30 (4-107)	<0.01
sST2 (pg/mL)	1469.2±510.9 (30-2014)	1151.30±655.9 (210-2450)	>0.05

NT-proBNP, N-terminal pro-brain natriuretic peptide; PAH, Pulmonary arterial hypertension; sST2, Serum-soluble ST2

DISCUSSION

PAH is a life-threatening disease that affects all age groups and is associated with various clinical conditions.[7] PAH is characterized by intimal and medial hypertrophy and the remodeling of the small pulmonary arteries in the pulmonary vascular structures.[11] The diagnostic conclusion for PAH is defined with a mean pulmonary artery pressure exceeding 25 mm Hg and a pulmonary vascular resistance exceeding 3 U×m^2 at resting.[8,9]

Research suggests that the most common types of PAH are congenital heart disease-associated PAH, pediatric lung disease-associated PAH, idiopathic PAH, and postoperative residual PAH. Among the PAH-related congenital heart diseases, acyanotic and cyanotic congenital heart diseases constitute the 2 largest groups, respectively.[11]

Although there have been significant advances in the diagnostic methods and treatment of PAH, late-diagnosed or undiagnosed congenital heart diseases and postoperative PAH are among the important causes of severe PAH or the Eisenmenger syndrome in developing countries. Most of the patients in the present study had the Eisenmenger syndrome or postoperative

residual PAH related to late-diagnosed congenital heart diseases.

There is an increasing number of biomarkers in patients with PAH that can help with the assessment of diagnosis and prognosis. NT-proBNP, secreted from the cardiac myocytes in response to cardiac pressure and volume overload, is the most widely used biomarker for the diagnosis and risk stratification of PAH.[12] NT-proBNP levels reflect the severity of right ventricular dysfunction and right ventricular failure secondary to PAH, the main cause of death related to PAH.[14] NT-proBNP levels in patients with PAH are a significant parameter in determining the risk level at the time of diagnosis and in assessing treatment efficacy during the follow-up. In our study, the mean NT-proBNP level was significantly higher in the patient group than in the control group ($P<0.01$). Most of the patients involved in the present study had the Eisenmenger syndrome and comprised a non-homogenous group. There was no statistically significant correlation between the mean pulmonary artery pressure and NT-proBNP ($P>0.05$) or between Rp/Rs and NT-proBNP ($P>0.05$).

ST2 is a member of the interleukin-1 receptor host defense and inflammation family and encodes sST2 and transmembrane types. ST2 is produced in various cells and tissues, including endothelial cells, smooth muscle cells, and cardiomyocytes.[1,2] The functionally active transmembrane form, the ST2 ligand, is involved in modulating the responses of T helper type 2 cells, thereby improving immunological tolerance. It also plays a prominent role in reducing myocardial fibrosis and hypertrophy by binding to interleukin-33.[3,4] In 2013, Carlomagno et al[13] evaluated sST2 levels in 25 patients with PAH and 10 subjects in a control group with different etiologies and reported that sST2 demonstrated a good correlation with right ventricular structure and function parameters measured by magnetic resonance imaging in patients with PAH. Agoston-Coldea et al[5] reported similar findings insofar as sST2 levels correlated with right ventricular dysfunction in patients with PAH secondary to chronic obstructive pulmonary disease. In a more recent study, Chida et al[15] suggest that high levels of sST2 and NT-proBNP could predict a poor outcome in patients with childhood idiopathic PAH. Zheng et al[6] investigated sST2 levels in 40 patients with idiopathic PAH and concluded that there was a significant relationship between sST2 and clinical deterioration. In our study, we found no statistically significant difference in the mean sST2 level between patients with PAH and the control group.

Recent research suggests that sST2 is an important indicator of right heart failure and cardiovascular mortality in children and could be a novel biomarker of PAH. Based on the findings of our study, however, we were unable to detect any statistically significant correlation between PAH and serum sST2 levels. Since there are only a few investigations on this subject, further studies with more patients are needed to evaluate sST2 as a biomarker in patients with PAH.

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Conflict of Interest: None

REFERENCES

1. Wang TJ, Wollert KC, Larson MG, Coglianese E, et al. Prognostic utility of novel biomarkers of cardiovascular stress: the Framingham Heart Study. Circulation 2012; 25(12):1596-604.

2. Sanada, S., Hakuno D, Higgins LJ, Schreiter ER, McKenzie AN, Lee RT. IL-33 and ST2 comprise a critical biomechanically induced

and cardioprotective signalling system. J Clin Invest, 2007. 117(6): 1538-49.

3. Pascual-Figal, D.A, Januzzi J.L. The biology of ST2: The international ST2 consensus panel. Am J Cardiol 2015; 15(7 Suppl): 3B-7B.

4. Schmitz, J., Owyang A, Oldham E et al., IL-33, an interleukin-1-like cytokine that signals via the IL-1 receptor-related protein ST2 and induces T helper type 2-associated cytokines. Immunity, 2005. 23(5): 479-490.

5. Agoston-Coldea L, Lupu S, Hicea S, et al. Serum levels of the soluble IL-1 receptor family member ST2 and right ventricular dysfunction. Biomark Med. 2014; 8:11–18.

6. Zheng, Y.G.; Yang, T.; He, J.G.; Chen, G.; Liu, Z.H.; Xiong, C.M.; Gu, Q.; Ni, X.H.; Zhao, Z.H. Plasma solubleST levels correlate with disease severity and predict clinical worsening in patients with pulmonary arterial hypertension. Clin. Cardiol. 2014, 37, 365–370.

7. Simonneau G, Montani D, Celermajer DS, et al. Haemodynamic definitions and updated clinical classification of pulmonary hypertension. Eur Respir J. 2018.

8. Hoeper MM, Bogaard HJ, Condliffe R, et al. Definitions and diagnosis of pulmonary hypertension. J Am Coll Cardiol 2013; 62:42-50.

9. Barst RJ. Diagnosis and assessment of pulmonary. Hypertension in infants and children. Prog Pediatr Cardiol 2001; 12:279-88.

10. Haworth SG. The management of children with congenital heart disease. In: Beghetti M, Barst RJ, Naeije R, Rubin LJ (eds). Pulmonary Arterial Hypertension Related to Congenital Heart Disease, 2006, Elsevier, New York, pp 129-41.

11. Duffels MGJ, Engelfriet PM, Berger RMF, van Loon RLE, Hoendermis E, Vriend JWJ, van der Velde ET, Bresser P, Mulder BJM. Pulmonary arterial hypertension in congenital heart disease: an epidemiologic perspective from a Dutch registry. Int. J Cardiol 2007; 120:198 –204.

12. Warwick G, Thomas PS, Yates DH. Biomarkers in pulmonary hypertension. European Respiratory Journal. 2008 32: 503-512.

13. Carlomagno G, Messalli G, Melillo RM, et al. Serum soluble ST2 and interleukin-33 levels in patients with pulmonary arterial hypertension. Int J Cardiol. 2013; 168:1545–1547.

14. McLaughlin VV, Presberg KW, Doyle RL, Abman SH, et al. Prognosis of pulmonary arterial hypertension: ACCP evidence-based clinical practice guidelines. Chest 2004; 126: 78S-92S

15. Chida A, Sato H, Shintani M, et al. Soluble ST2 and N-terminal pro-brain natriuretic peptide combination. Circ J. 2014; 78:436–442.