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Diagnostic values of neutrophil-to-lymphocyte ratio and Tei index for congenital heart disease-associated pulmonary arterial hypertension in children

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About 10% of children with congenital heart disease (CHD) have pulmonary arterial hypertension (PAH). In this study, we explored the diagnostic values of neutrophil-to-lymphocyte ratio (NLR) and Tei index for CHD-PAH in children. In order to complete the study, two hundred CHD children treated in our hospital from January 2015 to January 2017 were enrolled and divided into postoperative-PAH group (n=29) and no postoperative-PAH group (n=171) according to follow-up results. The peripheral blood was drawn after surgery, neutrophils and lymphocytes were measured by routine blood test, and NLR was calculated. Ventricular Tei index was determined by pulse Doppler, and the other basic data were compared. Factors affecting postoperative PAH were explored by multivariate analysis. The correlations of NLR and Tei index with N-terminal pro-B-type natriuretic peptide (NT-proBNP) and cardiac output/pulmonary blood flow (CO/Qp), were analyzed. The structural equation model of PAH was constructed. According to receiver operating characteristic (ROC) curves, the optimal cutoff values of NLR and Tei index for evaluating postoperative PAH were investigated. It has been found that NLR and Tei index are closely related to CHD-PAH in children, which indirectly reflect the severity and have high diagnostic values.

Keywords: Hypertension. Pediatric CHD

Congenital heart disease (CHD) is a type of congenital disease with abnormal anatomical structure triggered by abnormal development of cardiovascular system during embryonic development. CHD, the most typical congenital defect, ranks first among birth defects in China, which is one of the major contributors of neonatal death. CHD accounts for 29% of all deaths from birth defects in newborns[1]. CHD can result in low immunity and malnutrition, easily lead to respiratory infection and other diseases, and even induce congestive heart failure and pulmonary arterial hypertension (PAH), thus affecting the quality of life of the children after birth, reducing their survival rate, shortening their survival time, and bringing burden to their families and the society[2]. About 10% of CHD children have PAH. Among various types of surgical indications and prognosis of CHD, the diagnosis of PAH is the key factor and prerequisite for choosing a reasonable treatment plan for CHD children[3].

In this context, it becomes necessary to assess the indices for diagnosing the severity of CHD-PAH in children, evaluate the correlation between each index and CHD-associated PAH (CHD-PAH) as it would help understanding the situation and development of the children's condition better, formulate reasonable treatment methods, improve the treatment effect, and ameliorate the prognosis of the children. Therefore, in this study, we tried to investigate the value of neutrophil-to-lymphocyte ratio (NLR) and cardiac Tei index and its use in the diagnosis of CHD-PAH in children.

Materials and Methods

Two hundred CHD children treated in our hospital from January 2015 to January 2017 were enrolled as research subjects. Among them, there were 112 males and 88 females, aged 4 months old to 5.4 years old, with a mean of (2.4 ± 0.3) years old, and the body mass index (BMI) was (13.45 ± 1.2) $kg \cdot m^{-2}$. According to the postoperative pulmonary artery pressure (PAP), the children were divided into postoperative-PAH group (n=29) and no postoperative-PAH group (n=171). This study was reviewed and approved by the hospital Ethics Committee, and informed consent was obtained from the family members of all the subjects participated in this study.

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Inclusion criteria

For the study, only children (i) meeting the diagnostic criteria for CHD[4] which could be diagnosed based on the pregnancy history of the child patient's mother, symptoms, development status, X-ray examination, ultrasound, electrocardiogram, cardiac catheterization, angiocardiography, determination of pigment dilution curve and spiral computed tomography (CT); and those with (ii) clinical symptoms such as purple lips, malnutrition, low immunity, upper respiratory tract infection and pulmonary infection; (iii) sinus rhythm; and (iv) those who would not quit halfway and could cooperate during the study were included.

Exclusion criteria

Children with (i) premature contraction, bundle branch block or advanced atrioventricular block; (ii) pulmonary artery stenosis or right ventricular outflow tract stenosis; (iii) endocrine, metabolic, renal or other relevant diseases; or (iv) mania, depression, schizophrenia or other mental disorders were not included in the study.

Methods and Measures

General data such as age, gender and body mass of all patients were collected through the electronic medical record system. According to the relevant criteria in the 2015 ESC/ERS Guidelines for the diagnosis and treatment of PAH, the PAP at rest was estimated by colour Doppler echocardiography in children with CHD, and spiral CT pulmonary angiography (CTPA) was conducted. With systolic PAP (sPAP) ≥40 mmHg as the diagnostic criterion for PAH[5], the CHD children were divided into postoperative-PAH group (n=29) and no postoperative-PAH group (n=171). On the early morning of the next day after surgery, 5 mL of fasting venous blood was drawn from each subjects, injected into an anticoagulant tube, and mixed evenly. Then, the clinical and biochemical indices were detected in the Department of Clinical Laboratory of our hospital within 2 h, and the NLR was compared between the two groups of patients.

The indices of right ventricular function, such as the right atrial pressure (RAP), mean PAP (mPAP), pulmonary artery wedge pressure (PAWP), N-terminal pro-B-type natriuretic peptide (NT-proBNP), cardiac output/pulmonary blood flow (CO/Qp), pulmonary vascular resistance index (PVRI), cardiac index (CI), sPAP, 6-min walk distance (6MWD), transcutaneous oxygen saturation (SPO_2) and BNP, were measured using an EPIQ 7 cardiology 4-dimensional color Doppler ultrasound (Philips).

The children were kept in a quiet state before measurement, and orally administered with 10% chloral hydrate (manufacturer: Qingdao Yulong Algae Co., Ltd., batch number: 20140132, specification: 0.5 mL/kg, total dose <10 mL) for sedation according to the situation. Next, the children were adjusted to the left lateral decubitus position. Parameters of the instrument were tuned to get clear images, the examination angle was adjusted to obtain the best detection signal from each valve orifice, and the sample volume was 0.24 cm^3. Besides, the blood flow signal of each valve artery at the end of breath were recorded at a speed of 50-100 mm/s, and the clear dynamic and static images of the valve orifice section were saved.

The scanning speed of the Doppler ultrasound was set as 50-100 mm/s, the sample volume was placed at the mitral orifice, and the direction of the sound beam was adjusted so that it was parallel to the blood flow direction. Furthermore, the forward blood flow spectrum of the mitral orifice was obtained, peak A and peak E were determined, and the time interval from the end of peak A to the beginning of peak E of the next cardiac cycle was measured, which was the isovolumetric contraction time (ICT). Then, the sample volume was placed at the aortic valve orifice, and the blood flow spectrum of the aortic valve orifice was obtained. The time from the beginning to the end of the forward blood flow of the aortic valve was measured, which was the ejection time and also the isovolumetric relaxation time (IRT). The ICT of the tricuspid orifice and IRT of the pulmonary valve were determined using the same method. Moreover, the spectral waveforms of at least 3 consecutive cardiac cycles were collected, and the average of multiple measurements was obtained. Tei index was calculated as: Tei index = (ICT+IRT)/IRT.

The relevant data of regular follow-up of all subjects were collected by means such as hospitalization, outpatient service and phone call. In this study, the follow-up time was 3 years, and the patients were followed up every 6 months until January 2020, with at least 5 follow-up records for each patient. Pulse Doppler was utilized for echocardiography. sPAP ≥40 mmHg at rest was set as the enrolment criterion for postoperative-PAH group,

and the remaining patients were grouped into no postoperative-PAH group.

SPSS 19.0 software was used for one-way ANOVA, and GraphPad Prism 5.0 was utilized for plotting. The *t*-test and χ^2 test were performed for comparisons between two groups. Pearson's analysis was conducted to analyze the correlations of NLR and Tei index with NT-proBNP and CO/Qp, respectively. The structural equation model of PAH was constructed using the analysis of a moment structures (AMOS) software, and ROC curves were plotted to analyze the values of NLR and Tei index for predicting postoperative PAH. P <0.05 suggested that the difference was statistically significant.

Results

Clinically, there were no statistically significant differences observed in age, BMI, gender and number of Down syndrome patients between the two groups (P >0.05) (Table 1). Similarly, no statistically significant differences were found in parameters such as RAP, mPAP and PAWP between the two groups (P >0.05). Compared with those in no postoperative-PAH group, the NT-proBNP, CO/Qp, NLR and Tei index in postoperative-PAH group were significantly higher, showing statistically significant differences (P <0.05) (Table 2).

The NLR and Tei index had positive correlations with NT-proBNP and CO/Qp, respectively (P <0.05), indicating that NLR and Tei index may rise with the increase of NT-proBNP or CO/Qp (Fig. 1).

Based on the AMOS software, the structural equation model of PAH was constructed using the maximum likelihood estimation method, and the optimal model was identified with the exploration analysis method. Besides, the root mean square error of approximation (RMSEA) and goodness of fit index (GFI) were utilized to evaluate the goodness of fit of the model. The results revealed that the structural equation model of PAH fitted well, and the values of RMSEA and GFI were 0.000 and 1.000, respectively. The path coefficients of NT-proBNP, CO/Qp, NLR and Tei index were 0.106, 0.09, 0.04 and 0.03, respectively (P <0.05), and the coefficient of determination of the model was 0.064 (Table 3 and Fig. 2).

To evaluate the values of NLR and Tei index for predicting postoperative PAH in patients, the optimal (cut off) values of NLR and Tei index for predicting

Table 1 — Baseline clinical data

Group	No postoperative -PAH (n=171)	Postoperative -PAH (n=29)	t/χ^2	P
Age (month)	27.6±0.4	27.7±0.6	1.148	0.253
BMI ($kg \cdot m^{-2}$)	13.4±1.2	13.6±0.8	0.865	0.388
Male (n)	113	18	0.014	0.907
Down syndrome (n)	2	3	0.662	0.416

Table 2 — Clinical and hemodynamics parameters

Group	No post-operative-PAH	Postoperative -PAH	t	*P*
RAP/mmHg	9.2±2.8	9.3±2.5	0.180	0.857
mPAP/mmHg	63.2±18.7	63.4±17.5	0.054	0.957
PAWP/mmHg	8.42±3.98	8.73±4.06	0.387	0.699
NT-proBNP	142±42	279±58	15.291	0.000
$(CO/Qp)/(L \cdot min^{-1})$	1.73±0.43	2.89±0.52	13.014	0.000
PVRI/ $(WU \cdot m^2)$	22±8.3	24.5±8.6	1.492	0.137
CI $(L \cdot min^{-1} \cdot m^{-2})$	3.5±1.2	3.4±1.0	0.424	0.672
ERA	18.5±1.5	19.0±1.3	1.690	0.093
SPAP/mmHg	85.3±10.4	86.1±11	0.380	0.704
6MWD/m	506.4±40.5	508.7±42.1	0.281	0.779
SPO_2/%	92.8±12.8	94.2±13	0.543	0.587
BNP (ng/L)	120.4±11.7	123.6±10.8	1.376	0.170
NLR	4.16±0.4	4.43±0.6	3.098	0.002
Tei index	0.36±0.1	0.43±0.11	3.435	0.001

Table 3 — Parameters of PAH structural equation model

Dependent variable	Independent variable	Standard estimation (path coefficient)	P
PAH	NT-proBNP	0.106	0.000
	CO/Qp	0.09	0.000
	NLR	0.04	0.002
	Tei index	0.03	0.001
	PVRI	–0.08	0.137
	BNP	–0.12	0.170
	CI	0.03	0.672

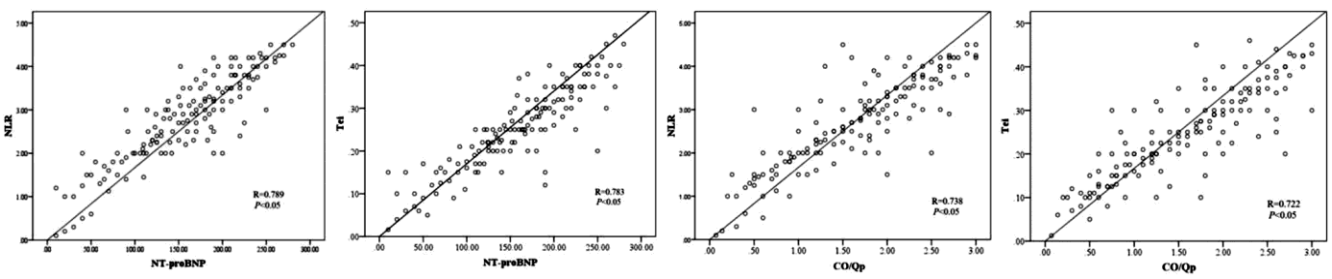

Fig. 1 — Correlations of NLR and Tei index with NT-proBNP and CO/Qp.

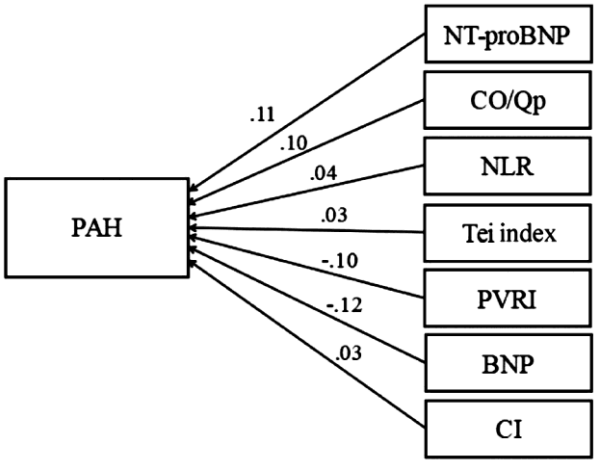

Fig. 2 — PAH structural equation model.

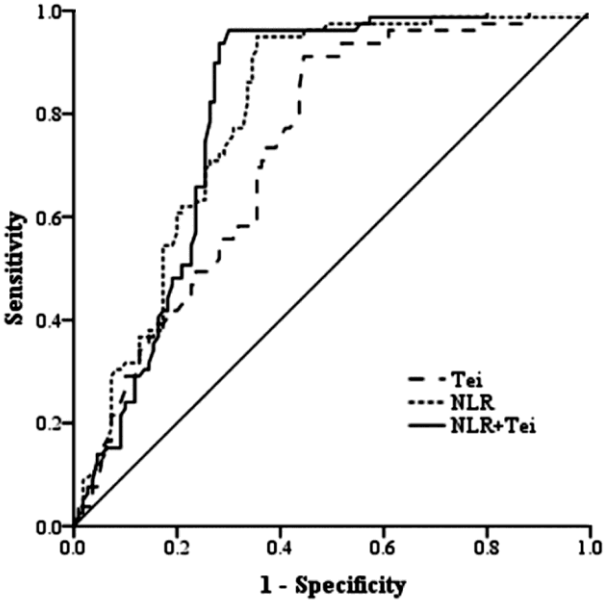

Fig. 3 — ROC curve analysis of values of NLR and Tei index for diagnosing postoperative PAH

Table 4 — Values of NLR and Tei index for predicting postoperative PAH

Diagnostic index	AUC	Optimal cutoff	P	Sensitivity (%)	Specificity (%)	+ve predictive val. (%)	–ve predictive val. (%)
NLR	0.730	6.75	<0.00	82.95	79.48	75.97	85.10
Tei index	0.797	0.68	<0.00	74.83	70.42	77.58	72.94
Combination	0.804	/	<0.001	80.57	73.58	74.26	79.73

postoperative PAH were analyzed and predicted using the ROC curve. The optimal (cut off) values of NLR and Tei index for predicting postoperative PAH were NLR=6.75 and Tei=0.68, respectively. The area under the curve of NLR combined with Tei was 0.804 (95% CI: 0.736-0.849, P <0.001), showing a higher diagnostic value (Table 4 and Fig. 3).

Discussion

Pulmonary arterial hypertension (PAH), one of the most typical complications of CHD in children, can trigger irreversible changes of pulmonary vessels, greatly shortening the life span of patients. Therefore, early diagnosis and early treatment are key for the treatment of CHD-PAH[6]. Due to the pathophysiological features of CHD-PAH and the complexity and diversity of cardiac anatomy, the multiple adaptive mechanisms of CHD-PAH have not been fully understood. As a result, enough attention has not been paid to CHD-PAH. In the past 50 years, surgery and pediatric cardiology have developed extensively, which improves the abilities of early identification and diagnosis of heart defects, and optimizes the treatment plans, thus delaying the progress of PAH in many cases[7].

Tei index, a non-invasive index proposed by Tei *et al.* in 1995[8] to quantitatively assess the ventricular systolic and diastolic function, is associated with cardiac preload. Tei index is able to objectively and truly assess the overall function of the heart, without being affected by heart rate and heart geometry. Meanwhile, its measurement is simple and repeatable[9]. Some scholars have used Tei index to evaluate the cardiac function in adults, children and fetus. The results manifested that Tei index is a sensitive index to assess the changes of cardiac function, possessing a certain clinical application value currently, and having extensive application prospects[10]. Another study have demonstrated that Tei index of the left ventricular in fetus in CHD group is significantly higher than that in normal control group (P <0.05), suggesting that Tei index can serve as an index to assess the left ventricular function in CHD fetus, provide a reliable basis for early evaluation of the prognosis of the fetus, and guide perinatal treatment[11]. It has been reported that Tei index of CHD-PAH patients is dramatically higher than that of normal controls, non-PAH CHD patients, and CHD patients with mild to moderate PAH, suggesting that Tei index can serve as a vital index to diagnose PAH in CHD patients and judge the severity of PAH[12]. The results of this study revealed that Tei index of patients in postoperative-PAH group was prominently higher than that in no postoperative-PAH group, indicating that Tei index may be closely associated with

postoperative PAH in CHD patients. However, further investigation is needed to clarify the association between Tei index and PAH in CHD patients.

The data of white blood cell count (WBC) and its classification are very easy to obtain in clinical practice. NLR refers to the ratio of neutrophils to lymphocytes. In recent years, NLR is considered to be an index that is able to reflect inflammation[13]. NLR is clearly associated with the condition and prognosis of coronary heart disease[14], heart failure[15], type 2 diabetes[16] and acute pancreatitis[17]. NLR is positively correlated with chronic obstructive pulmonary disease (COPD) complicated with PAH (P <0.05), and can be taken as a valuable index to reflect the condition of COPD complicated with PAH[18]. There is a strong correlation between NLR and CHD, and the increase of NLR in mothers during pregnancy will lead to an elevated risk of CHD in their children. Therefore, NLR can be taken as a clinical index to predict CHD in newborns[19]. The association between NLR and PAH has also been explored by scholars, and the results revealed that the number of lymphocytes is decreased markedly, the number of neutrophils is increased dramatically, and NLR was notably higher in PAH patients than that in normal control group, indicating that NLR is associated with the prognosis of PAH[20]. In the present study, the correlation between NLR and postoperative PAH in CHD patients was explored. The results revealed that the NLR of patients in postoperative-PAH group was markedly higher than that in no postoperative-PAH group, implying that NLR may be closely associated with postoperative PAH.

According to a study, the levels of NT-proBNP and CO/Qp are positively correlated with PAP in patients with CHD-PAH (P <0.05), which are able to indirectly reflect the severity of PAH in CHD patients[21]. The results of this study demonstrated that the levels of NT-proBNP and CO/Qp in CHD patients in postoperative-PAH group were dramatically higher than those in no postoperative-PAH group. Therefore, the correlations of NLR and Tei index with NT-proBNP and CO/Qp, respectively, were analyzed. The results indicated that NLR and Tei index had positive correlations with NT-proBNP and CO/Qp, respectively (P <0.05). Based the on AMOS software, the structural equation model of PAH was constructed using the maximum likelihood estimation method. The results indicate the coefficient of determination of the PAH structural equation model to be 0.64 and the fitness degree good, implying that NT-proBNP, CO/Qp, NLR and Tei index are closely related to postoperative PAH in CHD patients. In addition, the value of NLR and Tei index for diagnosing postoperative PAH was analyzed using the ROC curve. The optimal cutoff values of NLR and Tei index for predicting postoperative PAH were NLR=6.75 and Tei=0.68, respectively. However, further studies are needed to clarify the association between NLR and postoperative PAH.

Conclusion

Above observations in the study suggest that NLR and Tei index are closely related to the severity of congenital heart disease-associated pulmonary arterial hypertension (CHD-PAH) in children, indirectly reflect the severity of CHD-PAH in patients and thereby play an important role in the diagnosis of CHD-PAH.

Conflict of interest

Authors declare no competing interests.

References

1 Zhao QM, Liu F, Wu L, Ma XJ, Niu C & Huang GY, Prevalence of congenital heart disease at live birth in China. *J Pediatr*, 204 (2019) 53.

2 Wong P, Denburg A, Dave M, Levin L, Morinis JO, Suleman S, Wong J, Ford-Jones E & Moore AM, Early life environment and social determinants of cardiac health in children with congenital heart disease. *Paediatr Child Health*, 23 (2018) 92.

3 Constantine A, Dimopoulos K, Condliffe R, Clift P, Chaplin G, Muthurangu V, Haworth SG & Moledina S, Pulmonary arterial hypertension associated with congenital heart disease in children: clinical characterisation, outcomes and changes in demographics over time. *Eur Heart J*, 42 (2021) ehab724.

4 Xi SB, Wang SS, Qian MY, Xie YM, Li JJ & Zhang ZW, Predictors of operability in children with severe pulmonary hypertension associated with congenital heart disease. *Chin Med J*, 132 (2019) 811.

5 Vijarnsorn C, Durongpisitkul K, Chungsomprasong P, Bositthipichet D, Ketsara S, Titaram Y, Chanthong P, Kanjanauthai S & Soongswang J, Contemporary survival of patients with pulmonary arterial hypertension and congenital systemic to pulmonary shunts. *PLoS ONE*, 13 (2018) e0195092.

6 Kayali S, Ertugrul I, Yoldas T, Kaya O, Ozgür S, Orün UA & Karademir S, Sensitive Cardiac Troponins: Could They Be New Biomarkers in Pediatric Pulmonary Hypertension Due to Congenital Heart Disease? *Pediatr Cardiol*, 39 (2018) 718.

7 Medjedovic E, Begi Z, Stanojevic M, Begic E, Iglica A, Begic N & Kurjak A, Ultrasound in the service of early diagnosis and treatment of congenital heart defects: bosnian and herzegovinian experience. *Donald School J Ultrasound Obstetr Gynecol*, 2020 (2020) 131.

8 Tei C, New non-invasive index for combined systolic and diastolic ventricular function. *J Cardiol*, 26 (1995) 135.
9 Lee M & Won H. P08.04: Novel technique for measurement of fetal right myocardial performance index using synchronised images of right ventricular inflow and outflow. *Ultrasound Obstetr Gynecol*, 54 (2019) 178.
10 Song ES, Yoon S, Cho JH, Yang EM, Cho HJ, Choi YY, Ma JS, Eom GH & Cho YK, Serial evaluation of myocardial function using the myocardial performance index in Kawasaki disease. *World J Pediatr*, 14 (2018) 259.
11 Goudar SP, Zak V, Atz AM, Altmann K, Colan SD, Falkensammer CB, Friedberg MK, Frommelt M, Hill KD, Hsu DT & Levine JC, Comparison of echocardiographic measurements to invasive measurements of diastolic function in infants with single ventricle physiology: a report from the Pediatric Heart Network Infant Single Ventricle Trial. *Cardiol Young*, 29 (2019) 1248.
12 Carmen Ş, Cristina B, Rodica T & Theodora B, Epidemiological Characteristics and Prospective 6-Months Follow-up of Children with Pulmonary Arterial Hypertension. *Acta Med Marisiensis*, 61 (2015) 75.
13 Fu X, Li T, Dai Y & Li J, Preoperative systemic inflammation score (SIS) is superior to neutrophil to lymphocyte ratio (NLR) as a predicting indicator in patients with esophageal squamous cell carcinoma. *BMC Cancer*, 19 (2019) 721.
14 Hou P, Xue HP, Mao XE, Li YN, Wu LF & Liu YB, Inflammation markers are associated with frailty in elderly patients with coronary heart disease. *Aging* (*Albany NY*), 10 (2018) 2636.
15 Delcea C, Buzea CA & Dan GA, The Neutrophil to Lymphocyte Ratio in Heart Failure: A Comprehensive Review. *Rom J Intern Med*, 57 (2019) 296.
16 Fawwad A, Butt AM, Siddiqui IA, Khalid M, Sabir R & Basit A, Neutrophil-to-lymphocyte ratio and microvascular complications in subjects with type 2 diabetes: Pakistan's perspective. *Turk J Med Sci*, 48 (2018) 157.
17 Borges F, Silva S, Carvalho N & Costa P, Comparison of HAPS, BISAP and NLR as predictors of severity in acute pancreatitis (Atlanta 2012). *HPB*, 22 (S2) (2020) S273.
18 Zuo H, Xie X, Peng J, Wang L & Zhu R, Predictive value of novel inflammation-based biomarkers for pulmonary hypertension in the acute exacerbation of chronic obstructive pulmonary disease. *Anal Cell Pathol*, 2019 (2019) 5189165.
19 Rameh V & Kossaify A, Role of biomarkers in the diagnosis, risk assessment, and management of pulmonary hypertension. *Biomark Insights*, 11 (2016) BMI-S38323.
20 Yıldız A, Kaya H, Ertaş F, Oylumlu M, Bilik MZ, Yüksel M, Polat N, Akıl MA, Atılgan Z & Ülgen MS, Association between neutrophil to lymphocyte ratio and pulmonary arterial hypertension. *Turk Kardiyol Dern Ars*, 41 (2013) 604.
21 Yu X, Zhang Y, Luo Q, Liu Z, Zhao Z, Zhao Q, Gao L, Jin Q & Yan L, Iron deficiency in pulmonary arterial hypertension associated with congenital heart disease. *Scand Cardiovasc J*, 52 (2018) 378.