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Pulmonary hypertension diagnosed by right heart catheterisation in sickle cell disease

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ABSTRACT: Recent studies have recognised the importance of pulmonary hypertension (PH) in sickle cell disease (SCD). The aim of this study was to determine the prevalence and prognostic impact of PH and its features in patients with SCD.

80 patients with SCD underwent baseline clinical evaluation, laboratory testing, 6-min walk tests (6MWTs) and echocardiography. Patients with a peak tricuspid regurgitant jet velocity (TRV) of $\geqslant$2.5 m·s^{-1} were further evaluated through right heart catheterisation (RHC) to assure the diagnosis of PH.

Our study evidenced a 40% prevalence of patients with elevated TRV at echocardiography. RHC (performed in 25 out of 32 patients) confirmed PH in 10% (95% CI 3.4–16.5%) of all patients, with a prevalence of post-capillary PH of 6.25% (95% CI 0.95–11.55%) and pre-capillary PH of 3.75% (95% CI -0.4–7.9%). Patients with PH were older, had worse performance in 6MWTs, and more pronounced anaemia, haemolysis and renal dysfunction. Survival was shorter in patients with PH.

Our study reinforced the use of echocardiography as a screening tool for PH in SCD and the mandatory role of RHC for proper diagnosis. Our findings confirmed the prognostic significance of PH in SCD as its association to pronounced haemolytic profile.

KEYWORDS: Haemodynamics, prevalence, pulmonary hypertension, right heart catheterisation, sickle cell disease, survival

Sickle cell disease (SCD) encompasses a group of hereditary haemoglobinopathies resulting from inheritance of at least one mutant version of the β-globin gene (βA) on chromosome 11, designated haemoglobin S (HbS) [1]. In HbS, valine replaces glutamic acid in position 6 of the β-chain. This substitution leads to polymerisation of haemoglobin when it is deoxygenated [2].

In homozygous sickle cell anaemia (SCA), two βS-globin alleles are inherited. In addition to SCA, other major sickle genotypes resulting from co-inheritance of other βA-globin gene mutations or β-thalassaemia mutations are recognised [1]. Individuals who have SCA are clinically indistinguishable from those who have Sβ0 thalassaemia and both are more severe than other sickle genotypes [1]. SCD is characterised clinically by the presence of chronic haemolytic anaemia and acute and chronic vaso-occlusion phenomena with cumulative and widespread organ damage. The World Health Organization (WHO) estimates there are ~275,000 births per year worldwide with SCD [3].

Pulmonary hypertension (PH) associated with SCD was first described in 1936 by YATER and HANSMANN [4], and corresponds to a frequent and serious complication associated with increased morbidity and mortality. Traditionally, PH in SCD was considered to be a result of vaso-occlusive events, such as acute chest syndrome (ACS) [5, 6]. In the last few years, however, PH has been attributed to intravascular haemolysis and consequent deregulated nitric oxide metabolism [7, 8].

The early symptoms of PH are nonspecific and do not differ from those experienced by SCD patients without PH [9]. Previous studies suggested that the prevalence of PH in this disease is between 0 and 35% [10–14]. However, these studies were limited by their retrospective data collection, patient selection, and lack of right heart catheterisation (RHC) to confirm the presence of PH and differentiate pre- from post-capillary PH.

The aim of this study was to determine the prevalence and prognostic significance of PH in SCD patients.

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METHODS

Population

In this cohort, we prospectively evaluated 80 consecutive adult patients (>18 yrs of age) with SCD (subtypes SCA and Sβ0 thalassaemia) followed at our university hospital (Hospital das Clínicas, University of São Paulo Medical School, São Paulo, Brazil). The diagnosis of SCD was based on haemoglobin electrophoresis and A2 haemoglobin level determination by chromatography. To be included, patients had to be clinically stable, without history of ACS or blood transfusion in the previous 2 weeks. All patients provided written informed consent.

Clinical and laboratorial evaluation

All patients underwent baseline clinical evaluation and haematological testing, including complete blood count, reticulocyte count, haemoglobin electrophoresis, fetal haemoglobin level determination and biochemical testing to evaluate their haemolytic profile. History of ACS was assessed from medical records. The serum levels of inflammatory markers, and liver and renal function were also determined. On the same day, all patients performed a 6-min walk test (6MWT), as previously described [15].

Echocardiographic studies

All Doppler echocardiography was performed by the same cardiologist, who was blinded to all other patient data except the diagnosis of SCD. The two-dimensional echocardiographic images were obtained using a Sequoia 512 ultrasound system (Acuson, Mountain View, CA, USA). Cardiac measurements were performed according to the guidelines of the American Society of Echocardiography [16]. Tricuspid regurgitation was assessed in the parasternal right ventricular inflow, parasternal short-axis and apical four-chamber views. Continuous-wave Doppler sampling of the peak tricuspid regurgitant jet velocity (TRV) was used to estimate the right ventricular-to-right atrial systolic pressure gradient using the modified Bernouille equation ($4 \times TRV^2$). The presence of valvular heart disease or left ventricular systolic dysfunction was an exclusion criterion.

Invasive haemodynamic evaluation

RHC was performed in patients with a TRV of $\geqslant 2.5\ m \cdot s^{-1}$. A 7 French flow-directed pulmonary artery catheter (Baxter Healthcare Corporation, Irvine, CA, USA) was used in all patients. Cardiac output (CO) was measured by the standard thermodilution technique. We defined PH as the presence of a

TABLE 1 Sickle cell disease patient characteristics, according to tricuspid regurgitant jet velocity (TRV)

	Baseline data	TRV <2.5 m·s^{-1}	TRV ⩾2.5 m·s^{-1}	p-value
Subjects n	80	48	32	
Age yrs	33±10	30±8	38±11	0.001
Females %	61.2	68.7	50	0.07
BMI kg·m^{-2}	20.6±2.6	21.1±2.8	20±2.3	0.6
Left ventricle ejection fraction %	64±7	64±7	64±6	0.67
6MWT m	499±141	515±148	475±131	0.004
O_2 saturation %	92±13	93±15	90±6	0.38
Clinical history %				
Hydroxyurea use	13.7	18.7	6.2	0.102
Priapism	70.9	62.5	80	0.18
Hepatitis C	27.5	16.6	34.3	0.04
Leg ulcer	30	12.5	50	0.001
Proteinuria	36.2	25	40.6	0.06
DVT	10	4.1	18.7	0.07
Ischaemic stroke	13.7	14.6	12.5	0.53
ACS	13.7	18.8	6.3	0.18
Haematological findings				
Hb g·L^{-1}	82±12.9	86.7±16.1	75.5±12.1	<0.001
Reticulocytes %	11.2±5.43	10.9±5.33	11.8±5.65	0.51
Leukocytes $\times 10^9$ cells·L^{-1}	11.4±3.4	11±3.5	12.1±3.2	0.11
Platelets $\times 10^9$ cells·L^{-1}	407±124	427±121	378±126	0.082
Fetal Hb %	7.4±5.41	8.29±5.78	6.22±4.64	0.15
LDH U·L^{-1}	1116±553	909±445	1427±561	<0.001
Uric acid mmol·L^{-1}	332.5±125	290±100	390±112	0.001
AST U·L^{-1}	48.8±20.7	42.6±17.6	54.9±22.6	0.007
GGT U·L^{-1}	89.7±62.3	71±64.5	120±45.6	<0.001
Unconjugated bilirubin μmol·L^{-1}	53.1±50.44	53.35±56.4	53.7±41.9	0.211
C-reactive protein ng·mL^{-1}	6.4±7.7	5.3±5.5	7.98±9.92	0.36
BUN mmol·L^{-1}	4.21±2.9	3.5±1.7	5.2±3.7	0.006
Creatinine μmol·L^{-1}	61.9±29.2	56.4±22.8	71.4±36.2	0.027

Data are presented as mean±SD, unless otherwise stated. BMI: body mass index; 6MWT: 6-min walk test; DVT: deep venous thrombosis; ACS: acute chest syndrome; Hb: haemoglobin; LDH: lactate dehydrogenase; AST: aspartate transaminase; GGT: γ-glutamyltransferase; BUN: blood urea nitrogen.

mean pulmonary artery pressure of ⩾25 mmHg. Further classification into pre- or post-capillary PH was based on pulmonary artery occlusion pressure (P_{pao}). PH patients with a P_{pao} of ⩽15 mmHg were classified as having pre-capillary PH. All patients presenting with PH at RHC were evaluated for the presence of chronic thromboembolic disease by means of pulmonary ventilation/perfusion (V'/Q') scan.

Statistical analysis

Continuous data are presented as mean±SD. Qualitative data are presented as proportions with 95% confidence intervals (CIs) and were compared using Fisher's exact test. All continuous variables were compared using unpaired t-tests. Laboratory measurements were log-transformed in order to reduce the influence of data distribution. A p-value of <0.05 was considered significant. Survival time was estimated using the Kaplan–Meier method. The log-rank test was used for curve comparison.

RESULTS

80 patients were included in the study (61.2% females) with mean age of 33.3±10.1 yrs. Median follow-up time was 31.9 months (range 3.3–34.6 months). An interpretable TRV was obtained in all patients and ranged between 0.79 and 4.17 m·s^{-1}. In 32 (40%, 95% CI 29.2–54.7%) patients, TRV was >2.5 m·s^{-1}, suggesting the presence of elevated pulmonary arterial systolic pressure (P_{pas}). Clinical, haematological, biochemical and echocardiographic characteristics of all patients are presented in table 1. Patients with a TRV of ⩾2.5 m·s^{-1} presented more pronounced anaemia, more haemolysis (reflected by increased levels of lactate dehydrogenase (LDH) and aspartate transaminase), and higher levels of uric acid, γ-glutamyltransferase, blood urea nitrogen (BUN) and creatinine.

Among 32 patients referred for RHC, 26 (81.2%) underwent the procedure. Four patients did not perform the exam for social or clinical reasons. Two patients refused the procedure. Haemodynamic data are showed in table 2.

The P_{pas} estimated by echocardiography and measured by RHC were well correlated (r=0.77, p<0.001); however, invasive measurement confirmed the presence of elevated pulmonary artery pressure in only eight out of the 26 patients submitted to the procedure, corresponding to 10% of the total sample (95% CI 3.4–16.5%). Of these, five (6.2%, 95% CI 0.9–11.5%) patients presented with a P_{pao} of >15 mmHg, thus being classified as post-capillary hypertension; the remaining three (3.7%, 95% CI 0.4–7.9%) patients were classified as having pre-capillary (arterial) pulmonary hypertension. Patient distribution according to echocardiographic and invasive haemodynamic characteristics is presented in figure 1.

The comparison between patients with true PH (n=8) and the remaining patients from our cohort (n=66) revealed a significant difference in age, distance walked in the 6MWT, presence of proteinuria, platelets count, and BUN, LDH, uric acid, γ-glutamyltransferase and haemoglobin levels (table 3). Although the number of patients in each subgroup precluded further evaluation, no significant difference was found in the comparison of clinical and haemodynamic variables between patients with pre- and post-capillary PH, except for P_{pao} (data not shown). It is noteworthy that all confirmed PAH patients presented pulmonary vascular resistances (PVRs) >2.5 Wood

TABLE 2 Invasive haemodynamic data, according to the presence of pulmonary hypertension (PH)

	Without PH	PH	p-value
Subjects n	18	8	
Age yrs	35.3±11.6	45.6±10.7	0.04
PVR Wood units	0.8±0.6	2.24±1.5	0.002
Cardiac index L·min^{-1}·m^{-2}	5±1.36	4.9±1.7	0.85
P_{pao} mmHg	13.3±2	16±5.7	0.07
P_{pas} mmHg	28.5 ±4.5	48.1±13.3	<0.001
$\bar{P}_{pa}$ mmHg	18.7±2.8	33.1±8.9	<0.001
6MWT m	511±78.9	460±152	0.25

Data are presented as mean±SD, unless otherwise stated. PVR: pulmonary vascular resistance; P_{pao}: pulmonary artery occlusion pressure; P_{pas}: pulmonary artery systolic pressure; $\bar{P}_{pa}$: mean pulmonary artery pressure; 6MWT: 6-min walk test.

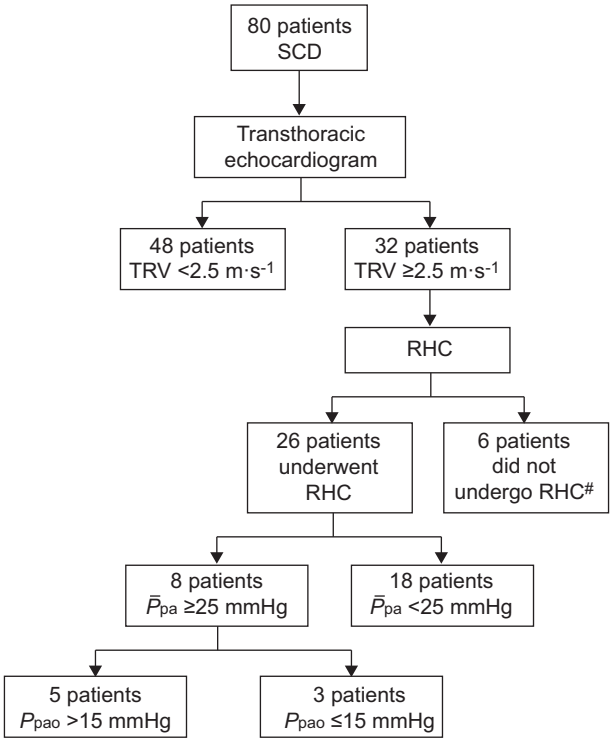

FIGURE 1. Patient distribution according to echocardiographic and invasive haemodynamic characteristics. SCD: sickle cell disease; TRV: peak tricuspid regurgitant jet velocity; RHC: right heart catheterisation; $\bar{P}_{pa}$: mean pulmonary artery pressure; P_{pao}: pulmonary artery occlusion pressure. #: reasons for failure to perform RHC were either clinical (hyperhaemolytic reaction in a 28-yr-old female with a TRV of 3.05 m·s^{-1}, alloimmunisation and severe anaemia in steady state in a 36-yr-old female with a TRV of 3.9 m·s^{-1}, and infection in a 21-yr-old male with a TRV of 2.61 m·s^{-1}), patient refusal (a 37-yr-old female with a TRV of 2.65 m·s^{-1} and a 40-yr-old male with a TRV of 2.7 m·s^{-1}) or social problems (a 45-yr-old male with a TRV of 3.37 m·s^{-1}).

TABLE 3 Sickle cell disease patient characteristics, according to the right heart catheterisation

	TRV <2.5 m·s^{-1} plus TRV ⩾2.5 m·s^{-1} and $\bar{P}$pa <25 mmHg	$\bar{P}$pa ⩾25 mmHg	p-value
Subjects n	66	8	
Age yrs	31.3±11.6	45.6±11	0.04
Females %	64.6	37.5	0.3
BMI kg·m^{-2}	20.8±2.65	20.1±2.8	0.5
Left ventricle ejection fraction %	64.2±7.1	62.5±6	0.53
6MWT m	53±71	460±152	0.02
O_2 saturation %	92.1±6.12	87.5±7.41	0.77
Clinical history			
Hydroxyurea use	15.4	12.5	0.4
Priapism	62.5	80	0.47
Hepatitis C	24.6	25	0.33
Leg ulcer	23.1	37.5	0.41
Proteinuria	30.7	87.5	0.02
DVT	7.7	37.5	0.04
Ischaemic stroke	15.4	0	0.22
ACS	16.6	12.5	0.62
Haematological findings			
Hb g·L^{-1}	84.4±12	73±10.2	0,01
Leukocytes ×10^9 cells·L^{-1}	11±3.4	12.1±4.27	0.58
Reticulocytes %	11±5.3	13.5±6.5	0.23
Platelets ×10^9 cells·L^{-1}	420±122	308±137	0.02
Fetal Hb %	7.8±5.7	6.21±3.44	0.9
LDH U·L^{-1}	1041±535	1596±424	0.006
Uric acid mmol·L^{-1}	282.5±109	516±81.5	<0.001
AST U·L^{-1}	46.3±18.8	61.2±25.5	0.04
GGT U·L^{-1}	82.2±61.7	135±64.3	0.34
Unconjugated bilirubin μmol·L^{-1}	55.7±28	72.84±65	0.22
C-reactive protein ng·mL^{-1}	5.28±5.5	6.36±4	0.61
BUN mmol·L^{-1}	7.74±3.57	18.6±11.5	<0.001
Creatinine μmol·L^{-1}	56.6±21.1	99±45.9	<0.001

Data are presented as mean±SD, unless otherwise stated. TRV: tricuspid regurgitant jet velocity; $\bar{P}$pa: mean pulmonary artery pressure; BMI: body mass index; 6MWT: 6-min walk test; DVT: deep venous thrombosis; ACS: acute chest syndrome; Hb: haemoglobin; LDH: lactate dehydrogenase; AST: aspartate transaminase; GGT: γ-glutamyltransferase; BUN: blood urea nitrogen.

units. Chronic thromboembolic disease was excluded in all patients by means of pulmonary V'/Q' scanning.

Seven patients died during the follow-up period (table 4): two in the group with a TRV of <2.5 m·s^{-1} and five in the group with a TRV of ⩾2.5 m·s^{-1}. In the latter group, three patients had been submitted for RHC: two presenting as post-capillary PH and the other one presenting as pre-capillary PH. Although there was a trend toward a survival difference between the two groups classified according to the presence of a TRV of ⩾2.5 m·s^{-1}, it did not quite reach statistical significance (p=0.07) (fig. 2). Patients with PH confirmed by RHC had worse survival compared with the remaining patients, regardless of their measured TRV (p=0.0005) (fig. 3).

DISCUSSION

Our study was the first prospective study to estimate the prevalence and prognostic significance of pulmonary hypertension in SCD patients based on RHC. Our results indicated that SCD patients with PH presented worse clinical, laboratory and functional profiles, associated with a worse prognosis.

Clinical and echocardiographic characteristics of our population are similar to others previously described [10, 11, 13–15]. Approximately 40% of our patients presented with a TRV of ⩾2.5 m·s^{-1}, and this subgroup presented more anaemia, haemolysis and renal dysfunction compared with the group with a normal TRV. Similarly to the literature, there is no difference between the two groups by fetal haemoglobin level, vaso-occlusive episodes and ACS.

This specific subgroup of patients has previously been described as presenting a higher mortality. These results have been ascribed to increased prevalence of PH in this population [7, 8, 13]. Nevertheless, the presence of PH was solely addressed by echocardiography in these previous studies. Our findings lead us to speculate that the higher mortality associated with a TRV of ⩾2.5 m·s^{-1} may not totally be related to the presence of

TABLE 4 Cause of death and patient characteristics

Patient	Sex	Age yrs	TRV m·s^{-1}	$\bar{P}_{pa}$ mmHg	P_{pao} mmHg	Cause of death
1	F	23	1.99	NA#	NA#	Cranial traumatism
2	M	37	1.16	NA#	NA#	Spontaneous intracranial haemorrhage
3	F	65	3.32	28	12	Chronic renal failure
4	F	36	3.9	NA#	NA#	Acute anaemia, pneumonia
5	M	40	2.7	NA#	NA#	Hepatic failure
6	M	51	3.37	40	18	Right ventricular dysfunction, pneumonia
7	M	58	4.17	43	8	Right ventricular dysfunction

TRV: tricuspid regurgitant jet velocity; $\bar{P}_{pa}$: mean pulmonary artery pressure; P_{pao}: pulmonary artery occlusion pressure; F: female; M: male; NA: not applicable. #: right heart catheterisation was not performed; in this case, patients were included in the survival analysis based on the results of the echocardiogram.

PH but may also be associated with other pathophysiological phenomena of SCD.

Furthermore, invasive haemodynamics revealed a higher prevalence of a post-capillary component (P_{pao} >15 mmHg) in the subgroup of patients with true PH, indicating a possible underlying left ventricular dysfunction.

In 2003, CASTRO *et al*. [17] also reported a large proportion of patients (50%) with high P_{pao} (>15 mmHg), despite the normal ejection fraction in echocardiography. In 2007, ANTHI *et al*. [18] found a prevalence of 46.2% of venous (post-capillary) hypertension among patients with SCD and PH. In a recent study, using only echocardiography, SACHDEV *et al*. [19] reported a left heart disease prevalence of 10–11% in SCD patients with PH. Although the increased P_{pao} was once attributed to an ''inversed'' Bernheim effect (left ventricular filling inhibition by paradoxical ventricular septal motion) [17], the most probable explanation is an underlying diastolic dysfunction. Diastolic dysfunction is well described in SCD and is imputed to microvascular occlusion, iron overload and chronic high output [19].

Consequently, our findings reinforced the multifactorial mechanisms that may be related to the development of PH in SCD patients. Among those multiple mechanisms, haemolysis, asplenic status, thrombosis and left heart disease (with a diastolic component) may share a role in vascular damage. The implication of our findings are particularly important if a specific therapeutic approach for the management of PH is considered, as treatments for pre- and post-capillary PH may be considerably different. The clinical results of controlled trials of endothelin receptor antagonists use in heart failure have been disappointing so far, despite their proven efficacy in pulmonary arterial hypertension [20, 21].

Moreover, several considerations of the pathophysiology of PH in SCD have emphasised only the role of haemolysis and pulmonary vasoconstriction in the genesis of this complication. In fact, our study found a very similar pattern to those observed in other studies. Although it has been questioned [22], there is evidence of a role of haemolysis in the pathogenesis of PH in SCD. Our study also confirms the impact of PH on the mortality of SCD patients.

Nonetheless, it is relevant to consider other factors. The haemodynamic profile has some patterns already described in the literature [17, 23]. PVR is markedly low, if compared with other populations of PH patients [24]. The mean PVR in this group was 2.3 Wood units and the highest PVR was 5.1 Wood units. In the

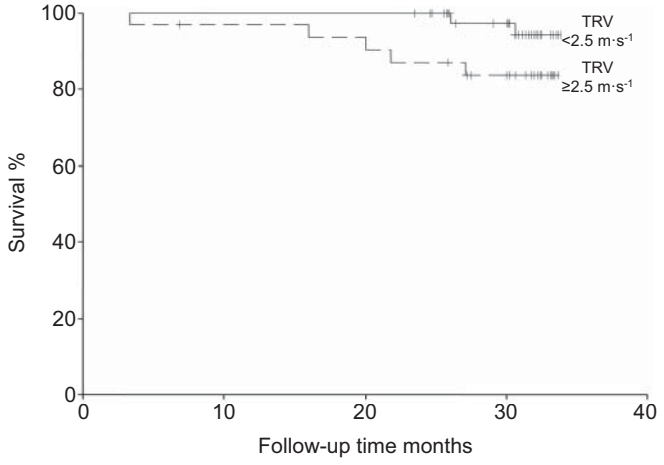

FIGURE 2. Probability of survival according to tricuspid regurgitant jet velocity (TRV) group. Kaplan–Meier estimates for the probability of survival at 36 months are shown. Vertical ticks represent censored patients. p=0.07.

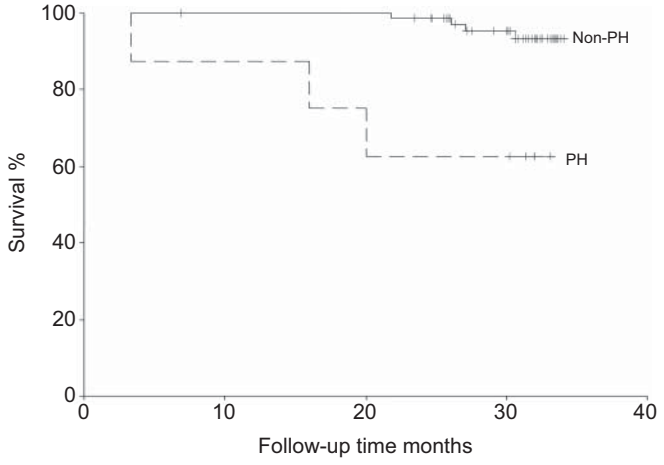

FIGURE 3. Probability of survival according to confirmed diagnosis of pulmonary hypertension (PH). Kaplan–Meier estimates for the probability of survival at 36 months. Vertical ticks represent censored patients. p=0.0005.

group without PH, the mean PVR was 0.78 Wood units, which is much lower than the usual reference value of 1.7 Wood units [25]. These findings are attributable to the lower viscosity and the decreased vascular resistance in response to high CO secondary to the treatment regimen of chronic anaemia [17, 26].

When individuals with PH are compared with the remaining patients, differences in clinical, haematological, biochemical and functional characteristics become more evident. It is noteworthy that uric acid is elevated (512 ± 78 *versus* 306 ± 99 mmol·L^{-1}; $p<0.0001$) and platelet count is decreased (284 ± 129 *versus* $422\pm122\times10^9$ cells·L^{-1}; $p=0.0015$) in the PH group.

High levels of uric acid have been observed in patients with other forms of PH and were assigned to the depletion of adenosine triphosphate in the tissues and worsening of oxidative metabolism [27]. Hyperuricaemia also occurs in haemolytic disease as a consequence of the increased production of uric acid after the recycling of purines. Associated renal dysfunction may also be related to increased uric acid level [28].

The decrease in platelet count was not expected in a context of an inflammatory disease and the presence of hyposplenism. It is not possible, however, to exclude that lower platelet counts could be associated with the presence of hepatitis C or older age, as already described in SCD [29, 30]. Although our study evidenced older age in this subgroup of patients, no association with hepatitis C could be noted.

Survival was significantly lower in the group with PH confirmed by RHC, regardless of the haemodynamic pattern (pre- or post-capillary). This finding is particularly relevant and raises the importance of the appropriate diagnosis of PH in this particular subgroup of patients.

Our study has some limitations that need to be accounted for. The monocentric design and small sample size should be taken into consideration when extrapolating our results. To allow us to better scrutinise our results, 95% CIs were included. Moreover, not all patients selected for completion of catheterisation could be subjected to the procedure. Even under these limitations, to our knowledge, our study presented the largest proportion of individuals evaluated by invasive haemodynamic assessment.

We conclude that PH is a significant complication of SCD with direct prognostic implications. Our study reinforces the use of echocardiography as a screening tool for PH in SCD patients and the mandatory role of RHC for the proper diagnosis. The higher prevalence of post-capillary PH associated with SCD strengthens not only the evidence for multifactorial pathways that may be involved with this particular clinical condition but also the need for appropriate haemodynamic characterisation before any attempt at a specific therapeutic approach targeting PH management.

STATEMENT OF INTEREST

None declared.

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