Efficacy and safety of sildenafil in the treatment of severe pulmonary hypertension in patients with hemoglobinopathies

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Background and Objectives. During the last decade new approaches to the treatment of pulmonary arterial hypertension (PH) have increased symptomatic relief and prolonged survival. PH is a common sequel of the hemoglobinopathies, thalassemia and sickle cell anemia, but the use of standard oral treatment options, such as calcium channel blockers, endothelin receptor antagonists, and long-term anticoagulation therapy, is limited because of toxicity and poor effectiveness. Sildenafil citrate is a selective and potent inhibitor of cGMP-specific phosphodiesterase-5 (PDE5) which promotes selective smooth muscle relaxation in lung vasculature and has been utilized successfully in the treatment of PH. The primary objective of this study was to evaluate the efficacy of sildenafil treatment in the control of PH in patients with hemoglobinopathies.

Design and Methods. In this study patients with hemoglobinopathies (thalassemia intermedia n=4; thalassemia major n=2; sickle thalassemia n=1) suffering from severe PH were treated with sildenafil citrate (50 mg b.i.d.) for periods ranging from 4 to 48 months.

Results. A significant decrease in pulmonary pressure and improvement in exercise capacity and functional class were observed in all patients. No significant adverse events were reported.

Interpretation and Conclusions. These data, in a small group of patients, indicate that sildenafil citrate is effective in the treatment of PH in hemoglobinopathies that cannot be treated with alternative oral drugs and is well tolerated long-term at a daily dose of 100 mg, though studies including more patients may uncover toxicities and limitations of efficacy.

Key words: hemoglobinopathies, thalassemia, sickle thalassemia, pulmonary hypertension, sildenafil.

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Pulmonary arterial hypertension (PH) is defined as a sustained elevation of pulmonary arterial pressure, indicated as > 25 mmHg at rest and > 30 mmHg during exercise, with a mean pulmonary-capillary wedge pressure and left ventricular end-diastolic pressure of less than 15 mmHg. Increased pulmonary vascular resistance leading to right ventricular dysfunction, congestive heart failure and ultimately death, is closely associated with PH. Whether PH is defined as idiopathic or associated with a specific condition, the histological pathology of the lung tissue is identical and is characterized by intimal fibrosis, increased medial thickness, pulmonary arteriolar occlusion, and plexiform lesions.[1] A recently revised clinical classification of PH includes the hemoglobinopathies as diseases associated with PH, along with other conditions, such as thyroid disorders, glycogen storage disease, Gaucher's disease, hereditary hemorrhagic telangiectasia, myeloproliferative disorders, and splenectomy.[2] PH in patients with hemoglobinopathies is thought to be due to the state of hypercoagulability in these patients, a reduction in nitric oxide due to the catalytic effect of free hemoglobin, hypoxia, alteration of the elastic fibers of the blood vessels, and the occurrence of subclinical vasocclusive crises in sickle cell anemia.[3] The association between the development of PH in most chronic, hereditary and acquired hemolytic anemias supports the existence of a syndrome of hemolysis-associated pulmonary hypertension.[4] The incidence of pulmonary arterial hypertension in patients with thalassemia intermedia is reported to be 40-50%.[5] In patients with thalassemia major, this incidence rises to 10-75%.[6-8] However, in a recent study of 200 well-controlled thalassemia major patients, no cases of PH were observed.[9] This suggests that the previously described high incidence in patients with thalassemia major may result from the difficulty in differentiating severe thalassemia intermedia from thalassemia major and also that early introduction of regular blood transfusions

and iron chelation therapy may prevent or delay the onset of PH. In sickle cell anemia the incidence of PH is estimated to be 30-60% and a recent study has confirmed that PH determines a mortality risk in these patients.[10] The mean pulmonary artery pressure is inversely related to survival with a 10 mmHg rise being associated with a 1.7-fold increased risk of death.[11]

Current oral treatment options are limited and include calcium channel blockers, prostacyclin analogs, endothelin-1 receptor antagonists, and thromboxane inhibitors.[12] Calcium channel blockers are effective in only 30% of patients[13] and are difficult to manage in hypotensive thalassemic patients. Endothelin-1 receptor antagonists can exacerbate elevated hepatic enzyme levels in thalassemia patents with liver disorders.[14] Prostacyclin analogs improve clinical function in many patients, but the mode of administration (subcutaneous or by continuous infusion) can lead to serious complications as well as being very costly.[15-17] The long-term efficacy of orally or inhaled prostacyclin analogs is yet to be established.[18] Sildenafil citrate, a selective and potent inhibitor of cGMP-specific phosphodiesterase 5 (PDE5), which promotes smooth muscle relaxation in lung vasculature, has been used successfully in the treatment of primary and secondary PH.[19-23] The efficacy of sildenafil has been previously demonstrated in chronic thromboembolic PH.[24] One report of sildenafil utilized in a thalassemia intermedia patient with symptomatic PH documented improved function and significantly reduced PH over time.[25] In this study the efficacy, safety, and tolerability of sildenafil were investigated in hemoglobinopathic patients with severe PH despite previous treatment with anticoagulant drugs, diuretics, cardiac glycosides, vasodilators, and blood transfusion therapy. The objectives of this study were to determine the efficacy of twice-daily dosing of sildenafil in reducing the tricuspid gradient and improving symptoms of PH, and to assess the safety and tolerability of this therapy regimen.

Design and Methods

Patients

Adult hemoglobinopathic patients with severe PH who were symptomatic and unresponsive to conventional treatment were recruited from five Italian Thalassemia Centers. Patients were required to have a mean tricuspid gradient ≥ 45 mmHg at rest as determined by continuous wave echo-Doppler and a modified New York Heart Association (NYHA) functional classification of III or IV, despite therapy.[2] Patients with PH due to left ventricular dysfunction were not considered for the study because of the different physiopathology of the pulmonary vascular involvement and therapeutic approach. Patients did not undergo cardiac catheterization in order to evaluate pulmonary vascular resistance, right atrial pressure and wedge pressure, considering the good relationship between Doppler estimates and invasive measurements of pulmonary arterial pressure described in literature at baseline and after treatment.[26-27] Furthermore, while data relating non-invasive measurements in patients with advanced lung disease indicated that the utility of these measurements is limited,[28] this problem is not present in our patients who did not have chronic obstructive pulmonary disease or an abnormal body surface and had a good acoustic ultrasound thoracic window. The poor compliance of these chronically ill patients to repeated invasive measurements, affecting the possibility of an adequate follow-up, must also be considered. Regular transfusion regimens were maintained in patients with thalassemia intermedia receiving transfusion as first-line treatment in conjunction with chelation therapy. Patients with significant renal or hepatic impairment were excluded. Patients were required to have a stable hematologic status and the study therapy was initiated no more than 6 days after the last transfusion.

Study design and objectives

This was an open-label, multicenter study. Doppler-defined PH, echocardiography measurements, functional class, and exercise capacity were assessed at time 0. Baseline blood pressure and heart rate measurements were also recorded. Two patients were admitted to hospital for two days for the initial treatment phase which consisted of oral sildenafil 25 mg once daily; the other patients started therapy in a day-hospital regime. Dosing was gradually increased to 50 mg b.i.d. over a period ranging from 4 to 15 days. Patients were evaluated on an out-patient basis at weeks 4, 12, 24 and every 6 months thereafter. The primary objective of this study was to evaluate the efficacy of sildenafil treatment in the control of PH. Efficacy was assessed by a change from baseline in tricuspid gradient (as determined using continuous wave Doppler echocardiography),[26] functional class (according to the modified NYHA classification),[12] and exercise capacity (assessed by a six-minute walking test).[27] These efficacy measures were assessed at 4, 12, and 24 weeks and after every 6 months, except exercise capacity, assessed at baseline, 4 weeks, 12 weeks, 12 months and every year. Additional consideration was given to drug safety and tolerability, which was assessed by monitoring patients' discontinuation of treatment and adverse events at each follow-up, and laboratory analyses performed every 4 weeks. All patients underwent a pulmonary perfusion scintigraphy with ^{99}Tc in order to detect perfusion defects in the lungs. At the same time transthoracic echocardiography was also performed.

Imaging protocol

Two-dimensional Doppler ultrasound examinations were performed at the baseline evaluation and at every control visit. All studies were recorded on VHS videocassettes. Patients were imaged in the left lateral decubitus position. Parasternal and apical two-dimensional views were obtained, with the transducer orientation and gain settings adjusted for optimal definition of the endocardial surface of each cardiac chamber. Continuous-wave Doppler recordings of tricuspid regurgitation were obtained from the apical window. The following variables were analyzed:

i) the right ventricular end-diastolic area was measured in the apical four-chamber view by tracing the endocardial edges of the right ventricle and the plane of the tricuspid valve at end diastole. Images were considered technically adequate if no drop-out in the endocardial outlines along the interventricular septum and right ventricular free wall was observed. Right ventricular size was characterized as a planar area rather than calculated volume because derivation of volumes would necessitate geometric assumptions about the right ventricle that may be inaccurate in patients with chronic pulmonary hypertension;
ii) the right ventricular percent change in area was calculated from the areas of the right ventricle in end diastole (EDA) and end systole (ESA) using the formula: right ventricular percent change in area=100×(EDA-ESA)/EDA. This measure of right ventricular function correlates closely with right ventricular ejection fraction as measured by radionuclide angiography;[30]
iii) the tricuspid regurgitant jet area was measured in the apical four-chamber view. Frame-by-frame analysis of each cardiac cycle was used to identify the maximum area of the color flow Doppler jet. The outline of the regurgitant signal was traced and the area determined by computerized planimetry. It has been previously demonstrated that this area is closely correlated with the severity of tricuspid regurgitation measured by a double thermodilution technique;[31]
iv) the maximal tricuspid regurgitant jet velocity, an index of the systolic pressure gradient between the right ventricle and right atrium, was measured by determining the peak regurgitant velocity in the continuous-wave Doppler flow profile obtained from the cardiac apex. Only Doppler signals resulting in a clearly defined envelope of velocities were considered suitable for analysis. Trans-tricuspid gradient was estimated as $4V^2$, where V is maximal tricuspid regurgitant jet velocity. Pulmonary pressure in this particular small cohort of patients derived from different centers is expressed as tricuspid gradient in order to reduce the variability associated with different ways of assessing right atrial pressure.[32] Left heart measurements were performed as previously described.[33]

The study protocol was performed following local Ethics Committee guidelines and all patients provided written informed consent.

Results

Patients' characteristics

A total of seven patients were treated in this study; their baseline characteristics are summarized in Tables 1 and 2. All patients had a stable hematologic status compatible with their disease and had severe PH (functional class III and IV) despite anticoagulant, diuretic, cardiac glycoside or vasodilator treatment, and regular transfusion regimens. All but one patient (patient #5) had been splenectomized. The patient with sickle β thalassemia had β^0 39 C→T mutation.

Following the initial in-patient therapy period, all patients received sildenafil 50 mg b.i.d. Patients received treatment for periods ranging from 4 weeks to 48 months. The mean treatment duration was 12 months. Patient #7 was the last to be enrolled in the study, therefore, only a 12-week period of therapy was available for consideration in the study. No patients discontinued the treatment.

Treatment efficacy

The two patients with the highest levels of PH (patients #1 and 2) showed a rapid decrease in tricuspid gradient at echocardiography following hospital discharge, after up-titration of the drug. At week 12, tricuspid gradient was significantly decreased in each patient and this trend persisted after 12 months of therapy ($p<0.01$; Table 3). A significant improvement in clinical condition was observed in these two patients in functional class IV at baseline: after 6 months they had improved to class II-III. The other patients progressed from NYHA class II-III to class I-II. After 12 weeks of therapy all patients showed a significant fall in PH concomitant with symptomatic improvements (six-minute walking test 301+80 to 445+64 meters; $p<0.002$).

PH increased in patient 2 at 18 months, during an episode of acute pneumonia, but resolved after the acute period. Significant improvements in the tricuspid gradient and the six-minute walking test were observed in patients with the highest and lowest baseline values, respectively.

Improvements were also observed in hemodynamic and functional status (Table 3). Anatomical and functional echocardiographic evaluations were performed at each visit. Ventricular measures were investigated

Table 1. Baseline hematologic characteristics.

Pt #	Sex (age in years)	Diagnosis	Concomitant therapies	Mean Hb-pre (mg/dL)	Mean ferritin (ng/dL)	Units of blood/ year
1	Male (40)	Thalassemia intermedia	Deferoxamine s.c. 50 mg/kg/day (5 days/wk) Hydroxyurea 20 mg/kg/day	8.0	1230	32
2	Male (36)	Thalassemia intermedia	Deferoxamine s.c. 45 mg/kg/day	10.0	2500	32
3	Male (34)	Thalassemia intermedia	Deferoxamine s.c. 50 mg/kg/day	8.5	800	48
4	Female (40)	Thalassemia major	Deferoxamine 40 mg/kg/day	10.4	540	41
5	Male (38)	Sickle thalassemia		11.5	4500	11
6	Female (44)	Thalassemia intermedia	Deferoxamine s.c. 35 mg/kg/day	10.3	440	20
7	Male (41)	Thalassemia major	Deferoxamine s.c. 50 mg/kg/day	8.6	713	30

Mean Hb-pre: mean (in the last year) pre-transfusional hemoglobin level; Mean ferritin: mean ferritin level in the last year.

Table 2. Baseline cardiologic characteristics.

Pt #	Therapy	Cardiac rhythm	Time since diagnosis of PH	Pulmonary scintigraphy
1	Frusemide 25 mg Nifedipine 20 mg S.R. Acetylsalicylic 100 mg	Sinus rhythm	12 months	Isolated perfusion defects, left lung
2	Digoxin 0.25 mg Frusemide 25 mg Warfarin Diltiazem 60 mg b.i.d.	Atrial fibrillation	6 months	Multiple perfusion defects
3	Frusemide 25 mg Warfarin	Atrial fibrillation	12 months	Multiple perfusion defects, right lung
4	Acetylsalicylic 100 mg Diltiazem 120 mg/day Quinapril 5 mg/day	Sinus rhythm	10 years	Multiple perfusion defects, right and left lungs
5	Amlodipine 5 mg/day Digoxin 0.125 mg/day Frusemide 75 mg/day Potassium canrenoate 50 mg/day	Sinus rhythm	5 years	Multiple perfusion defects, right and left lungs
6	Enalapril 5 mg b.i.d.	Sinus rhythm	10 years	Negative
7	Enalapril 2.5 mg/day Amlodipine 5 mg/day Digoxin 0.75 mg Frusemide 50 mg/day	Sinus rhythm	12 months	Negative

Table 3. Hemodynamic and clinical effects of sildenafil.

	Arterial pressure sys/diast (mmHg)	HR (bpm)	Tricuspid gradient sys/mean (mmHg)	NYHA class	Six-minute Walk test (m.)	Hb level (g/dL)
Patient #1						
T0	110/80	95 S.R.	90/55	III–IV	55	12.1
4 wks	120/80	95 S.R.	70/53	II–III	400	12.6
12 wks	110/80	95 S.R.	49/34	I–II	610	13.0
24 wks	120/80	92 S.R.	54/32	I–II	–	12.3
12 mo	115/80	92 S.R.	53/33	I–II	630	12.3
18 mo	115/80	94 S.R.	50/32	I–II	–	11.2
24 mo	110/80	90 S.R.	49/36	I–II	750	12.1
Patient #2						
T0	85/70	140 A.F.	105/70	IV	Not performed	12.1
4 wks	90/70	122 A.F.	95/65	IV	Not performed	12.0
12 wks	95/70	120 A.F.	85/60	III	380	12.3
24 wks	90/75	110 A.F.	75/65	III	–	11.6
12 mo	95/70	105 S.R.	58/37	II	520	11.8
18 mo	85/70	122 A.F.	103/59	III?IV	–	12.0
24 mo	95/70	115 A.F.	85/70	II?III	535	12.2
Patient #3						
T0	105/80	90 A.F.	56/42	III	415	13.0
4 wks	105/85	88 A.F.	50/41	II–III	500	13.1
12 wks	110/75	85 A.F.	50/35	II–III	520	12.8
24 wks	110/70	85 A.F.	42/30	II	–	12.6
12 mo	110/75	85 A.F.	42/25	I–II	600	13.0
18 mo	105/70	82 A.F.	40/25	I–II	–	13.1
24 mo	105/75	80 A.F.	42/28	I–II	620	12.6
30 mo	110/80	80 A.F.	40/25	I–II	–	12.4
36 mo	110/80	83 A.F.	38/25	I–II	615	12.9
42 mo	105/75	80 A.F.	38/26	I–II	–	12.8
48 mo	105/80	88 A.F.	40/25	I–II	630	13.0
Patient #4						
T0	110/80	80 S.R.	158/114	III	115	12.4
4 wks	110/80	82 S.R.	56/48	II	318	12.6
12 wks	120/80	82 S.R.	85/60	I	612	13.0
Patient #5						
T0	100/62	74 S.R.	51/32	III–IV	110	12.9
4 wks	110/65	65 S.R.	40/25	II–III	300	13.0
12 wks	105/65	70 S.R.	40/25	II	550	13.0
24 wks	105/70	75 S.R.	44/19	II	–	12.8
Patient #6						
T0	105/80	85 S.R.	120/87	III	300	12.8
4 wks	105/80	70 S.R.	63/47	II	750	12.7
12 wks	105/80	72 S.R.	45/35	II	760	13.0
24 wks	105/80	85 S.R.	36/29	I	–	12.4
Patient #7						
T0	100/80	80 S.R.	68/47	III	400	13.1
4 wks	100/80	90 S.R.	50/30	II	670	12.9
12 wks	100/85	85 S.R.	48/29	I-II	750	13.2

and several parameters such as right ventricular area and diameter decreased to normal levels in patients with higher values at baseline. Relevant improvements were observed in the tricuspidal regurgitation area in all patients. In patient 5, for example, we registered a decrease from 9.9 to 3.4 cm^2 in 24 weeks (Tables 4, 5).

Table 4. Left heart morpho-functional echocardiographic changes.

Patients	*LVEDD (mm)*	*LVESD (mm)*	*LVEF %*	*LA (mm)*
Patient ##1				
T0	68	56	55	50
4 wks	65	36	65	50
12 wks	65	36	65	50
24 wks	65	36	60	50
12 mo	65	35	60	50
18 mo	65	37	60	50
24 mo	63	37	60	50
Patient #2				
T0	43	27	50	30
4 wks	50	29	50	30
12 wks	50	28	50	30
24 wks	51	33	55	30
12 mo	51	33	55	32
18 mo	51	33	55	32
24 mo	50	33	55	30
Patient #3				
T0	58	40	60	45
4 wks	58	40	60	46
12 wks	58	40	60	46
24 wks	56	38	60	45
12 mo	56	40	60	46
18 mo	56	40	63	45
24 mo	56	40	65	46
30 mo	56	38	65	46
36 mo	56	40	70	46
42 mo	57	38	68	45
48 mo	56	40	70	46
Patient #4				
T0	47	31	62	37
4 wks	48	29	60	38
12 wks	48	29	60	39
Patient #5				
T0	48	31	67	38
4 wks	49	28	66	42
12 wks	48	28	66	40
24 wks	48	28	66	40
Patient #6				
T0	48	34	60	34
4 wks	50	32	60	35
12 wks	50	32	60	33
24 wks	50	38	60	34
Patient #7				
T0	58	35	55	42
4 wks	59	36	60	43
12 wks	57	34	60	43

LVEDD: left ventricular end-diastolic diameter; LVESD: left ventricular end-systolic diameter; LVEF: left ventricular ejection fraction; LA: left atrium.

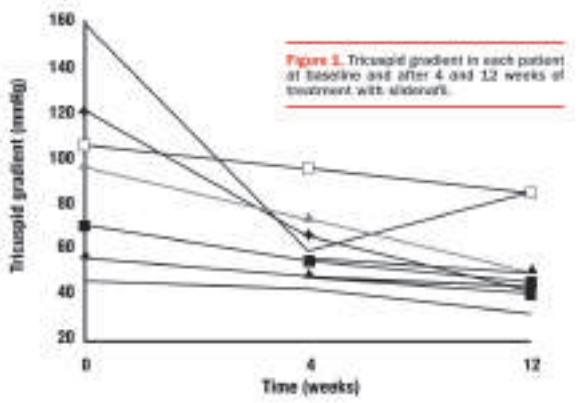

Figure 1. Tricuspid gradient in each patient at baseline and after 4 and 12 weeks of treatment with sildenafil.

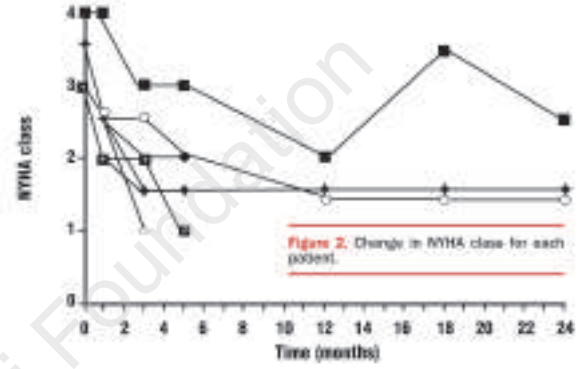

Figure 2. Change in NYHA class for each patient.

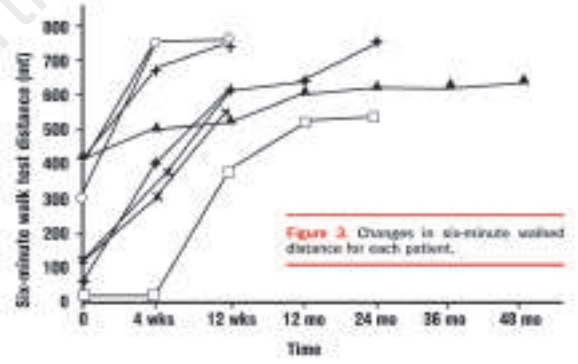

Figure 3. Changes in six-minute walked distance for each patient.

Tolerability

No significant changes in blood pressure or heart rate were observed. In one patient spontaneous recovery of sinus rhythm occurred after 12 months of therapy. One patient experienced a transient episode of nasal mucosal congestion, otherwise no adverse effects such as headache or priapism were observed or reported in the clinical interviews. In particular, priapism was not experienced by any patient during the study and in a previous case report it was suggested that sildenafil can relieve priapism in patients with sickle cell disease.[34] No drug-related withdrawals were reported.

Discussion

In this study of patients with hemoglobinopathies suffering from severe refractory PH, twice-daily sildenafil citrate (100 mg/day, p.o.) was seen to be an effective and well-tolerated therapy for this condition.

Therapeutic efficacy was assessed by measuring tricuspid gradient by continuous wave echocardiography. Sildenafil therapy was associated with a rapid,

sustained decrease in tricuspid gradient in all patients. Long-term drug administration produced progressive reductions in pulmonary pressure, with no reduction in systemic arterial blood pressure. Improved cardiovascular function was also associated with significant symptomatic improvements. These effects were seen in patients with different levels of pulmonary resistance. All patients had a significant improvement in modified NYHA functional class and six-minute walking test and the therapy was well tolerated. No drug-related withdrawals were reported. Spontaneous recovery of sinus rhythm from atrial fibrillation after 12 months of therapy was observed in one patient. No patients experienced adverse collateral effects and there were no significant changes in blood pressure or heart rate.

There is a high incidence of PH among hemoglobinopathic patients. The major determinants of PH in these pathologies are chronic hypoxia, intravascular hemolysis, iron overload and possibly splenectomy and thrombosis. Sildenafil particularly acts in hypoxia-related PH.[3,10,35,36] The quality of life and life expectancy of patients with hemoglobinopathies are affected by PH. In patients with primary PH it has been demonstrated that sildenafil significantly improves exercise tolerance, cardiac index and quality of life.[18] Therefore, if our preliminary data are confirmed by studies on a larger population, with adequate therapy for this severe complication of the disease, patient prognosis will be an open issue.

Four of the patients in this study were taking calcium channel blockers at study enrolment. These agents were not stopped during sildenafil therapy and no adverse effects from combined therapy were noted. Based on the efficacy of sildenafil alone in the other three patients in the study who were not taking calcium channel blockers, combined therapy was not required.

In conclusion, long-term therapy with sildenafil citrate has been shown in this study to produce a significant and persistent reduction in PH in patients with thalassemia and sickle thalassemia. However, further evaluation is required to investigate the full potential of this approach.

GD, GLF: conception and design of the study, carrying out the human assessments, drafting the article, acquisition, analysis and interpretation of data, final approval of the version to be published; MDC, RG, GD'A, PB: acquisition, analysis and interpretation of the data, revising the article for important intellectual content, final approval of the version to be published; CM: critical revision of the article, contributions to conception and design of the study, final approval of the version to be published; ML and FF: acquisition, analysis and interpretation of data, drafting the article, final approval of the version to be published. ML: creation of figures and tables. The authors declare that they have no potential conflict of interest.

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