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Sinusoidal Obstruction Syndrome/Veno-Occlusive Disease after Autologous or Allogeneic Hematopoietic Stem Cell Transplantation in Children: a retrospective study of the Italian Hematology-Oncology Association–Hematopoietic Stem Cell Transplantation Group

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A B S T R A C T

Sinusoidal obstruction syndrome (SOS), also known as veno-occlusive disease (VOD), is a potentially life-threatening complication that may develop after hematopoietic stem cell transplantation (HSCT). The aims of this retrospective multicenter study were to evaluate the incidence of SOS/VOD in a large cohort of children transplanted in centers across Italy by applying the new European Society for Blood and Marrow Transplantation (EBMT) criteria and to analyze the risk factors underlying this complication. We retrospectively reviewed data of pediatric HSCTs performed in 13 AIEOP (Associazione Italiana di Ematologia e Oncologia Pediatrica)-affiliated centers between January 2000 and April 2016. The new pediatric EBMT criteria were retrospectively applied for diagnoses of SOS/VOD and severity grading. Among 5072 transplants considered at risk for SOS/VOD during the study period, 103 children (2%) developed SOS/VOD, and the grade was severe or very severe in all patients. The median time of SOS/VOD occurrence was 17 days after HSCT (range, 1 to 104). Sixty-nine patients (67%) were treated with defibrotide for a median time of 16 days (range, 4 to 104). In multivariable analysis age < 2 years, use of busulfan during the conditioning regimen, female gender, and hemophagocytic lymphohistiocytosis were risk factors statistically associated with the development of SOS/VOD. The overall mortality directly related to SOS/VOD was 15.5%. Overall survival at 1 year was worse in patients with SOS/VOD ($P = .0033$), and this difference disappeared 5 years after HSCT. Nonrelapse mortality was significantly higher 1 and 5 years after transplantation in patients who developed SOS/VOD ($P < .001$). Based on the application of new EBMT criteria, the overall incidence of SOS/VOD recorded in this large Italian pediatric retrospective study was 2%. Nonrelapse mortality was significantly higher in patients who developed SOS/VOD. Identifying the risk factors associated with SOS/VOD can lead to more effective early treatment strategies of this potentially fatal HSCT complication in childhood.

INTRODUCTION

Sinusoidal obstruction syndrome (SOS), also known as veno-occlusive disease (VOD), is a potentially life-threatening complication that can develop after hematopoietic stem cell transplantation (HSCT). Known risk factors for SOS/VOD

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include patient-associated risk factors, such as age, underlying disease, hepatic dysfunction, ferritin levels, genetic factors (*GSTM1*-null genotype), previous exposure to gentuzumab ozogamicin or inotuzumab; and transplant-associated risk factors, such as type of HSCT (autologous or allogeneic), conditioning regimen (myeloablative with busulfan [Bu] or total body irradiation), and graft-versus-host disease (GVHD) prophylaxis [1].

The use of different diagnostic criteria (like the Seattle or Baltimore criteria) [2-4] and the analysis of cohorts of patients with different characteristics explain the variable incidence of SOS/VOD reported so far, although, overall, the incidence in children is higher than in adults (20% to 30% versus 9% to 14%, respectively) (1-5). The recent revision of the diagnostic criteria for SOS/VOD, proposed by the European Society for Blood and Marrow Transplantation (EBMT) [5], focuses on new criteria for the diagnosis of SOS/VOD in children and adds to the criteria proposed for adults [6]: (1) no limitation for the time of onset of SOS/VOD, (2) unexplained platelet consumption and refractoriness to platelet transfusions, (3) weight gain on 3 consecutive days, and (4) increase of bilirubin from baseline value over 3 consecutive days. Furthermore, the EBMT panel of experts proposed a scale to grade the severity of SOS/VOD in children, providing a useful tool that, once properly validated, could be used to predict outcome and evaluate the effectiveness of different treatment approaches. Although mild or moderate SOS/VOD may resolve within a few weeks in most patients, the severe form is often associated with multiorgan dysfunction and high mortality rate (>80%) [3].

To evaluate the incidence of SOS/VOD in an Italian pediatric population receiving either autologous or allogeneic HSCT, we conducted a retrospective, multicenter study among the transplant centers affiliated with the Italian Association for Pediatric Hematology and Oncology (Associazione Italiana di Ematologia e Oncologia Pediatrica [AIEOP]). We describe the characteristics of patients who developed SOS/VOD, analyzing risk factors and transplant outcome.

METHODS

We retrospectively reviewed all HSCTs performed in 13 of 26 AIEOP centers (50%) between January 2000 and April 2016. All patients were reported to the AIEOP-HSCT registry, and those who developed SOS/VOD after either allogeneic or autologous HSCT were considered eligible for this study. Data were retrieved from the AIEOP-HSCT registry, which collects information on demographics, diagnosis, date and type of transplant, conditioning regimen, occurrence and grade of acute and chronic GVHD, toxicity, and date and status at last follow-up. For the purposes of this study, the new EBMT criteria for diagnosis and grading for severity of pediatric SOS/VOD were retrospectively applied [5]. To evaluate the occurrence of multiorgan dysfunction, respiratory failure was defined as the need for oxygen supplementation, whereas renal failure was defined as an elevation of serum creatinine greater than twice the value observed at start of the conditioning regimen.

For this study each center was required to report all eligible patients in whom SOS/VOD was diagnosed. In case of SOS/VOD occurrence, further details were collected through ad hoc requests on the date of diagnosis and clinical characteristics at diagnosis, including transfusion-refractory thrombocytopenia (defined as 1 weight-adjusted platelet substitution/day), weight gain, hepatomegaly, ascites, level of bilirubin, thickness of gallbladder walls, and abnormalities of plasma coagulation factors (prothrombin time, activated partial thromboplastin time, antithrombin III, fibrinogen, D-dimer concentration). Additional data were also requested to evaluate the outcome of patients with SOS/VOD, such as transaminase and creatinine levels, occurrence of respiratory failure with or without pleural effusion and with or without the need for respiratory assistance, presence of renal failure with or without the need for dialysis, encephalopathy, admission to the intensive care unit, possible hepatic biopsy, and evolution to multiorgan failure (MOF). Type, dosage, and timing of drugs used for prophylaxis (defibrotide [DF], ursodeoxycholic acid [UCD]) and treatment (DF, UCD, steroids, recombinant tissue plasminogen activator, N-acetylcysteine) of SOS/VOD were also obtained. Before 2014 DF was administered only to patients enrolled in the pediatric prospective EBMT study [7] or as compassionate use; after 2014 the drug was approved by the European Medicine Agency for the treatment of severe SOS/VOD in many European countries.

The study was approved by the AIEOP-HSCT working group. All patients or their legal guardians had previously signed a consent allowing use of clinical data for research purposes.

Statistical Analysis

Analysis used January 1, 2017 as the reference date. Quantitative variables were reported as median value and range, whereas categorical variables were expressed as absolute value and percentage. Demographic and clinical characteristics of patients were compared using the chi-square test or Fisher's exact test for categorical variables, whereas the Mann-Whitney rank sum test or the Student's *t*-test were used for continuous variables, as appropriate. Overall survival (OS) and event-free survival were calculated according to the Kaplan-Meier method [8]. The risk of death for causes unrelated to malignant progression, defined as nonrelapse mortality (NRM), and that of developing SOS/VOD were calculated as cumulative incidences to adjust the analysis for competing risks [9]. Comparisons between different OS and event-free survival probabilities were performed using the log-rank test [10], whereas Gray's test was used to assess, in univariable analyses, differences between cumulative incidences [11]. Multivariable analysis was performed using logistic regression or the Cox proportional hazard regression model, as appropriate [10].

All results were expressed as 5-year probabilities or 5-year cumulative incidences (%) and 95% confidence interval (95% CI). $P < .05$ was considered to be statistically significant. Statistical analysis was performed using NCSS 10 statistical software (NCSS, LLC, Kaysville, UT) and Stata MP/15 (StataCorp LP, College Station, TX).

RESULTS

Between January 2000 and April 2016, 5072 transplants were performed in 4021 patients. Eight hundred seventy-one patients received additional transplants, either because of disease recurrence or as part of a specific treatment protocol. The characteristics of the 5072 transplants are shown in Table 1. During the study period 103 children developed SOS/VOD, with a cumulative incidence of 2% (95% CI, 1.7 to 2.5) (Figure 1). The SOS/VOD cumulative incidence was lower in 2000 to 2004 (.8%; 95% CI. .4 to 1.4), increased between 2005 and 2009 (2.52%; 95% CI, 1.8 to 35), and remained stable between 2010 and 2015 (2.54%; 95% CI, 2.0 to 3.3). Among these patients the 77.6% above 2 years of age developed SOS/VOD, and the underlying diseases were malignant in 84 children (81.5%) and nonmalignant in the remaining 19 (18.5%). Forty-four children underwent autologous HSCT (42.6%), whereas 59 were given an allogeneic stem cell transplant from a matched unrelated donor in 30 cases (29%), a matched family donor in 19 (18.4%), and an haploidentical donor in 10 (10%). As a stem cell source, we used bone marrow in 46 patients (44%), peripheral blood in 55 (54%), and cord blood in 2 children (2%) (Table 1).

Table 2 shows the symptoms of these 103 patients. The median time of SOS/VOD occurrence was 17 days after HSCT (range, 1 to 104; interquartile range, 11 days; 90th percentile, 29 days) (Figure 2). Fifty-three patients (62%) received i.v. Bu, whereas 32 (38%) were given oral Bu. Applying the new EBMT criteria for grading the severity of SOS/VOD [4], all 103 children showed signs and symptoms of severe SOS/VOD, and 36 (35%) of them had a very severe form. In particular, respiratory failure requiring invasive pulmonary ventilation occurred in 23 of 103 patients (22%), and it was associated with renal insufficiency in 17 and with encephalopathy in 7. Very severe renal failure requiring dialysis occurred in 12 patients (12%), associated with encephalopathy in 1 case; isolated encephalopathy occurred in 1 child (1%). By applying the classic criteria (Seattle or Baltimore criteria [2-4]) to our patients with SOS/VOD, 17% of patients showed severe SOS/VOD, 28% had moderate forms, and 55% had mild SOS/VOD.

The results of univariable analysis of the risk factors for SOS/VOD are shown in Table 3. The univariable analysis

Table 1
Patient Characteristics

	No. of Cases or Median	Percent or Range
Number of analyzed transplants	5072	100
Number of HSCTs		
First	4021	79.3
Subsequent	1051	20.7
Patient gender		
Male	3054	60.2
Female	2018	39.8
Patient age at transplantation		
Median age, yr	8	.2-20
<2 yr	573	11.3
≥2 yr	4499	88.7
Diagnosis		
Acute lymphoblastic leukemia	961	18.9
Acute myeloid leukemia	584	11.5
Chronic myeloid leukemia	47	.9
Lymphoma	350	6.9
Myelodysplastic syndrome	175	3.4
Neuroblastoma	625	12.4
Other solid tumors	1650	32.6
Nonmalignant	680	13.4
Year of transplantation		
2000-2004	1421	28.0
2005-2009	1431	28.2
2010-2016	2220	43.8
Type of transplant		
Autologous	2464	48.5
Matched family donor	799	15.8
Matched unrelated donor	1336	26.4
Haploidentical family donor	473	9.3
Stem cell source		
Bone marrow	1772	34.9
Peripheral blood	2980	58.7
Cord blood	346	6.8
Conditioning regimen		
TBI-based	934	18.4
Bu-based	1521	29.9
Treosulfan-based	362	7.2
Other chemotherapy*	2039	40.3
Missing	216	4.2
GVHD prophylaxis		
CsA or tacrolimus	453	17.5
CsA or tacrolimus + steroids	39	1.5
CsA or tacrolimus + ATG	304	11.8
CsA or tacrolimus + MTX	1040	39.5
CsA or tacrolimus + MTX + ATG	299	11.5
T cell depletion	329	12.7
Other/unknown	144	5.5

CsA indicates cyclosporine; ATG, antithymocyte globulin; MTX, methotrexate.
* Thiotepa = 545; thiotepa + cyclophosphamide = 131; thiotepa + melphalan = 282; thiotepa + etoposide + cyclophosphamide = 84; thiotepa + carboplatin = 55; thiotepa + fludarabine ± cyclophosphamide or melphalan = 92; thiotepa + fludarabine + alemtuzumab = 3; etoposide + carboplatin ± cyclophosphamide or ifosfamide or melphalan = 279; fludarabine + cyclophosphamide = 155; melphalan = 145; carmustine++cytarabine+ melphalan (beam) = 108; cyclophosphamide ± atg = 55; cyclophosphamide ± fludarabine or melphalan = 43.

showed that female gender, age at HSCT < 2 years; diagnosis of hemophagocytic lymphohistiocytosis (HLH), neuroblastoma, or thalassemia; and use of Bu or melphalan as part of the conditioning regimen and methotrexate as part of the GVHD prophylaxis were associated with a higher incidence of SOS/VOD. Univariable analysis also showed that use of cord blood stem cells was associated with a lower risk of SOS/VOD. No statistically significant difference in the cumulative incidences of SOS/VOD was observed between allogeneic (2.27%; 95% CI, 1.77 to 2.92) and autologous (1.79%; 95% CI, 1.34 to 2.40) HSCT (Grays test, $P = .222$).

In the multivariable logistic regression analysis the following 4 variables remained independently associated with an increased risk of SOS/VOD: female gender (hazard ratio [HR], 1.62; 95% CI, 1.09 to 2.41; $P = .018$), age at HSCT < 2 years (HR, 2.09; 95% CI, 1.40 to 3.11; $P < .001$), diagnosis of HLH (HR, 2.81; 95% CI, 1.06 to 7.44; $P = .038$), and use of Bu during the preparative regimen (HR, 5.20; 95% CI, 2.97 to 9.09; $P < .001$) (Table 4). The use of use of cord blood cells was confirmed to be associated with a lower risk of developing this complication also in multivariable analysis (Table 4).

An analysis of the data on the prophylaxis and treatment of SOS/VOD demonstrated that the prophylactic approach included DF (at dosage of 25 mg/kg/day) and UCD (at median dosage of 10 mg/kg/day), which were administered to 11 (11%) and 44 (43%) patients who developed severe or very severe SOD/VOD, respectively. The therapeutic agents given to patients with SOS/VOD included DF in 69 children (67%; dose 25 mg/kg/day for a median time of 16 days; range, 4 to 104); UCD in 43 patients (42%; 19 as prophylaxis and 24 as therapy), corticosteroids in 39 (38%; methylprednisolone at a dosage of 1 mg/kg/day for a median time of 10 days; range, 4 to 93), N-acetylcysteine in 28 children (27%), and recombinant tissue plasminogen activator in 8 children (8%; median dosage, .2 mg/kg/day). DF was started a median of 1 day (range, 0 to 14) after SOS/VOD diagnosis in 69 children (67%) and was administered alone in 15 patients (22%), in association with steroids in 13 (19%), with UCD in 12 (17%), with N-acetylcysteine in 12 (17%), and with recombinant tissue plasminogen activator in 3 (4%). In the remaining 14 children (20%) DF was administered in combination with 2 or more of the previously reported agents.

Eighty-seven patients (84%) had complete resolution of SOS/VOD after a median of 15 days from the diagnosis (range, 3 to 81). Resolution occurred in 80% of patients with a maximum bilirubin value ≥ 2 mg/dL and in 96% of those with a maximum bilirubin value < 2 mg/dL (anicteric SOS/VOD).

Sixteen of 103 patients with SOS/VOD (16%), all of whom had very severe disease, died a median of 20.5 days (range, 3 to 75) after SOS/VOD diagnosis for MOF. The survival probability (OS) at 1 year was 61% (95% CI, 51% to 71%) for patients with SOS/VOD versus 77% (95% CI, 76% to 78%) for patients who did not develop SOS/VOD ($P = .0033$), whereas the 5-year OS was 55% in both groups (Figure 3). NRM was significantly higher (HR, 2.12; 95% CI, 1.45 to 3.08; $P < .001$) in patients with SOS/VOD as compared with those without SOS/VOD as the incidence of NRM was 22% versus 6% at 100 days, 30% versus 12% at 1 year, and 35% versus 23% at 5 years after HSCT ($P < .001$) (Figure 4).

DISCUSSION

To the best of our knowledge, this is the largest retrospective study describing the incidence, risk factors, clinical characteristics, prophylaxis, treatment, and outcome of SOS/VOD in children given either autologous or allogeneic HSCT [12-14]. Among the 5072 transplants performed between 2000 and 2016 in 13 pediatric Italian HSCT centers, SOS/VOD, defined according to the new EBMT criteria, was diagnosed in 2% of cases (N = 103). No difference in the cumulative incidence of SOS/VOD was observed among the different transplant centers (data not shown). Several studies have reported that the incidence of SOS/VOD is higher in children than in adults (the relative risk ranging from 5.2 to 9.5) [1], most likely because of the immature hepatic metabolism in infancy and childhood [5]. The retrospective use of EBMT criteria in our population may represent a limit of our study because it is possible that patients with mild or moderate forms were not included in the collection of SOS/VOD. Moreover, with the improvement of

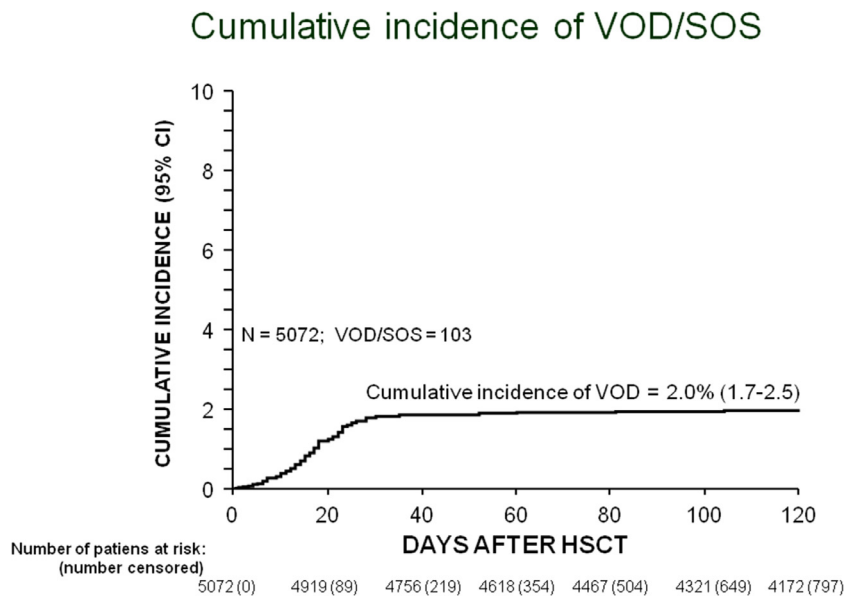

Figure 1. Cumulative incidence of VOD/SOS.

supportive therapies, the changes in the conditioning regimens used during the most recent years, including the use of drugs with lower hepatic toxicity (treosulfan and fludarabine in 7% and 13% of children, respectively) and of DF as prophylaxis in patients at risk (these data are not available), may explain the low incidence of SOS/VOD in our population.

Table 2
Characteristics of SOS/VOD According to EBMT Criteria

	No. of Cases (%)
EBMT criteria at diagnosis	
Transfusion-refractory thrombocytopenia	93 (90.2)
Weight gain on 3 consecutive days despite use of diuretic	93 (90.3)
Weight gain > 5% above baseline value	
Hepatomegaly above baseline value	71 (69)
Ascites	95 (92.2)
Rising bilirubin from a baseline value on 3 consecutive days or bilirubin ≥ 2 mg/dL within 72 hr	75 (72.8)
Median time of SOS/VOD after HSCT, days (range)	17 (1-104)
Additional criteria	
Thickness of gallbladder	70 (68)
Abnormalities of plasma coagulation	87 (84.4)
PT/PTT abnormalities	73 (84)
Decrease in fibrinogen	12 (13.7)
Increase in D-dimer	41 (47.1)
Deficiency of ATIII	54 (62)
Liver function test (alanine and aspartate amino-transferase values)	
≤2 x normal	18 (17.4)
>2 and ≤5 x normal	21 (20.3)
>5	64 (62.1)
Liver biopsy	2 (1.9)
Organ failure	
Respiratory failure	35 (34)
Pleural effusion	38 (36.8)
Need for respiratory assistance	23 (22.3)
Renal failure (creatinine value)	
Within normal range	70 (68)
<2 x normal	17 (16.5)
>2	16 (15.5)
Need for dialysis	8 (7.7)
Encephalopathy	11 (10.6)
MOF	16 (15.5)
Patients admitted to the pediatric intensive unit care	20 (19.4)

PT/PTT indicates prothrombin time/partial thromboplastin time; ATIII, antithrombin III.

For the purpose of this study, we retrospectively applied the new EBMT pediatric criteria [5] to evaluate the best tool for the diagnosis and grading of the SOS/VOD. Based on the data reported to the AIEOP registry and after interrogating the centers participating into the study, all patients meeting the criteria for SOS/VOD had the new pediatric EBMT criteria applied, and all were shown to have severe or very severe disease. Remarkably, the incidence of severe and very severe SOS/VOD reported in this study is lower than the incidence of overall SOS/VOD reported in other pediatric studies (13.2% to 30%) [15,16]. It has been reported that the incidence of severe SOS/VOD was 20% in children and 48% in adults [17,18].

By applying the new pediatric EBMT criteria, all patients with SOS/VOD were matched with severe or very severe forms of this complication, whereas according to the Seattle and Baltimore criteria [2-4] only 17% of these children presented severe SOS/VOD disease. The less accurate diagnosis using the classic criteria of severe SOS/VOD and the better details of the new EBMT criteria of these forms, including many groups of symptoms/signs and laboratory abnormalities, could explain the difference in the incidence of severe forms observed in our experience with these 2 different classifications. We suggest that the classic criteria could overestimate the mild or moderate SOS/VOD diagnosis, whereas the new EBMT criteria are more detailed in the identification of severe SOS/VOD that required a treatment.

Concerning the risk factors for SOS/VOD, some diseases have been reported to be associated with a higher risk of developing SOS/VOD, namely osteopetrosis (60%) [19], HLH (30%) [20], thalassemia (30% to 40%) [1], and neuroblastoma (15% to 30%) [21]. In our study, which enrolled a heterogeneous cohort of patients with both malignant (82%) and nonmalignant diseases (18%), univariable analysis confirmed that patients affected by HLH, thalassemia, and neuroblastoma have an increased risk of SOS/VOD (HR, 2.81 for HLH, 1.86 for thalassemia, and 1.13 for neuroblastoma). Notably, despite the use of a Bu-based conditioning regimen, none of the 13 children affected by osteopetrosis developed severe SOS/VOD.

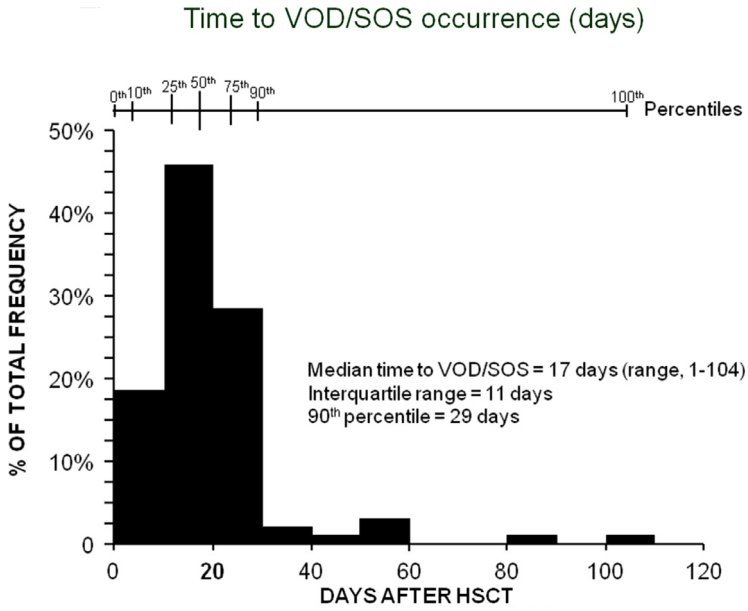

Figure 2. Time to VOD/SOS occurrence (days).

Table 3
Univariable Analysis of Variables Associated with Severe SOS/VOD Development

	No. of Patients	SOS/VOD	Cumulative Incidence (95% CI)(%)	*P*	HR* (95% CI)	*P*
All transplants	5072	103	2.0 (1.7-2.5)			
Gender						
Male	3054	51	1.7 (1.3-2.2)			
Female	2018	52	2.6% (2.0-3.4)	.025	1.55 (1.05-2.28)	.026
Age at HSCT						
≥2 yr	4499	80	1.8 (1.4-2.2)			
<2 yr	573	23	4.0 (2.7-6.0)	<.001	2.29 (1.44-3.64)	<.001
Diagnosis						
Malignant	4392	84	1.9 (1.6-2.4)			
Nonmalignant	690	19	2.8 (1.8-4.3)	.124	1.48 (.90-2.43)	.126
Osteopetrosis	13	0	0	.603	0	
HLH	66	5	7.6 (3.3-17.6)	.001	3.97 (1.62-9.76)	.003
JMML	65	1	1.6 (.2-1.9)	.783	.76 (.11-5.42)	.783
Neuroblastoma	625	24	3.9 (2.6-5.7)	.001	2.17 (1.38-3.43)	.001
Thalassemia	150	9	6.0 (3.2-11.3)	<.001	3.23 (1.62-6.42)	.001
Sickle cell disease	48	0	.0	.316		
No. of transplants						
First	4021	92	2.3 (1.9-2.8)			
Subsequent	1051	11	1.1 (.6-1.9)	.012	.46 (.24-.85)	.014
Type of transplant						
Autologous	2464	44	1.8 (1.3-2.4)			
MFD	799	19	2.4 (1.5-3.7)		1.34 (.78-2.29)	.288
MUD	1336	30	2.3 (1.6-3.2)		1.26 (.80-2.01)	.322
Haploidentical	473	10	2.1 (1.2-3.9)	.662	1.19 (.60-2.37)	.612
Disease status at transplant						
Malignant in remission	2444	51	2.1 (1.6-2.8)			
Malignant not in remission	1938	33	1.7 (1.2-2.4)		.81 (.53-1.26)	.354
Nonmalignant	690	19	2.8 (1.8-4.3)	.396	1.33 (.79-2.26)	.288
Stem cell source						
Bone marrow	1773	46	2.6 (2.0-3.5)			
Peripheral blood	2982	55	1.9 (1.4-2.4)		.71 (.48-1.05)	.084
Cord blood	317	2	.6 (.2-2.5)	.090	.24 (.06-1.00)	.049
Conditioning regimen						
Containing TBI	934	12	1.3 (.7-2.3)	.053	.55 (.30-1.02)	.055
Containing Bu	1521	77	5.1 (4.1-6.3)	<.001	6.61 (4.23-10.3)	<.001
Containing treosulfan	362	3	.8 (.3-2.6)	.008	.37 (.12-1.17)	.092
Containing TT	1851	35	1.9 (1.4-2.6)	.600	.48 (.32-1.03)	.081
Containing fludarabine	649	20	3.1 (2.0-4.8)	.058	1.11 (.68-1.81)	.671
Containing Cy	1242	34	2.7 (2.0-3.8)	.043	.94 (.62-1.41)	.751
Containing LPAM	1424	56	3.0 (2.4-3.9)	<.001	1.95 (1.32-2.87)	.001
Containing VP16	422	7	1.7 (.8-3.5)	.570	.55 (.25-1.18)	.123
Containing ATG	564	16	2.9 (1.8-4.6)	.145	.99 (.58-1.7)	.992

JMML indicates juvenile myelomonocytic leukemia; MFD, matched family donor; MUD, matched unrelated donor; TBI, total body irradiation; TT, thiotepa; Cy, cyclophosphamide; LPAM, melphalan; VP16, etoposide; Tac, tacrolimus.

* The first row and value of each variable was considered a reference value to estimate the HR.

Table 4
Multivariable Analysis of the Risk of Developing SOS/VOD

	HR (95% CI)	*P*
Gender		
Female vs. male	1.62 (1.09-2.41)	.018
Age at HSCT		
<2 yr vs. ≥2 yr	1.97 (1.32-2.94)	.001
Diagnosis		
HLH vs. other	2.71 (1.03-7.08)	.043
Neuroblastoma vs. other	1.39 (.80-2.52)	.240
Thalassemia vs. other	1.53(.66-3.52)	.322
No. of transplants		
Subsequent vs. first	1.03 (.66-1.72)	.929
Stem cell source		
Cord blood vs. other	.21 (.05-.86)	.030
Conditioning regimen		
Bu vs. no Bu	5.37 (3.16-9.10)	< .001
Melphalan vs. no melphalan	1.07 (.65-1.77)	.796
GVHD prophylaxis		
Methotrexate vs. no methotrexate	1.47 (.93-2.31)	.095

However, considering the very low number of patients affected by osteopetrosis in this cohort, no conclusions can be drawn.

Conversely, our data confirmed that females, children younger than 2 years of age, and a Bu-based conditioning regimen represent risk factors for SOS/VOD development, as already reported in other studies [1,12,14-16,22]. No information about the Bu route of administration are collected in the AIEOP registry, so no information on the role of either oral or i.v. Bu can be obtained from our analysis. However, after contacting the participating centers, we were able to obtain data relative to children who developed SOS/VOD: 35 patients (34%) received oral Bu and 55 (66%) received i.v. Bu. Five of 35 patients (14.2%) treated with oral Bu developed MOF, whereas 10 of 55 patients treated with i.v. Bu (18.1%) had MOF. Notably, plasma Bu pharmacokinetic was evaluated in all patients.

Interestingly, the use of cod blood as a stem cell source was associated with a lower risk of SOS/VOD both in univariable and multivariable analyses. Considering the low number of patients who received cord blood cells (n = 317) in our cohort as compared with those who received other stem cell sources (peripheral blood stem cells + bone marrow, 4755), no definitive conclusion in the benefit of using cord blood cells can be drawn.

Applying the new pediatric EBMT criteria for the diagnosis of SOS/VOD [5] to our cohort of patients, we found that the median day of diagnosis of SOS/VOD was 17 days but also that about 10% of cases occurred after day +30. Nonetheless, prospective studies are need to better understand whether the EBMT criteria may be helpful in making an early diagnosis.

Moreover, in our cohort 27% of patients with SOS/VOD had no jaundice, confirming the higher frequency of anicteric SOS/VOD forms in children than in adults [23,24]. In addition, in our cohort the development of thrombocytopenia refractory to platelet transfusions and of abnormalities of coagulation represented important and frequent findings in children with SOS/VOD (90.2% and 84.4% respectively; Table 2).

The overall mortality of patients with SOS/VOD was 39.8%, whereas that due to MOF in SOS/VOD patients was 15.5%. Both results are remarkably lower than the mortality (>80%) due to severe SOS/VOD described in the literature. A better knowledge of the disease, a more timely start of treatment, and, in the second period of the study, the availability of DF, a drug shown to rescue patients with SOS/VOD [25-29], may have contributed to the lower mortality rate observed in our study population.

The difference in both the early and late cumulative incidences of NRM between patients who either did or did not develop SOS/VOD was statistically significant ($P < .0001$), suggesting that patients experiencing SOS/VOD are at greater risk of transplant-related fatalities, also because of the contribution of other types of post-transplant complications (such as GVHD and thrombotic microangiopathy) as a possible result of their fragility of endothelial cells. On the contrary, the apparently lower risk of relapse observed in the group of patients who experienced SOS/VOD could probably be explained by the very low number of patients still at risk at more than 2 years after HSCT (only 32 in the SOS/VOD group versus 2203 in the non-SOS/VOD group). The impact on 5-year OS of SOS/VOD was attenuated by other causes of treatment failure, mainly

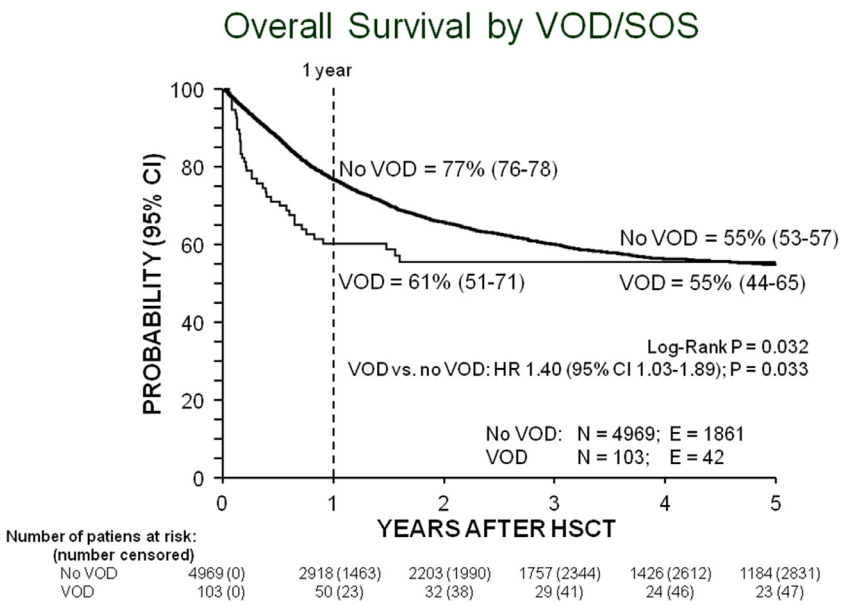

Figure 3. OS of patients who either did or did not experience SOS/VOD.

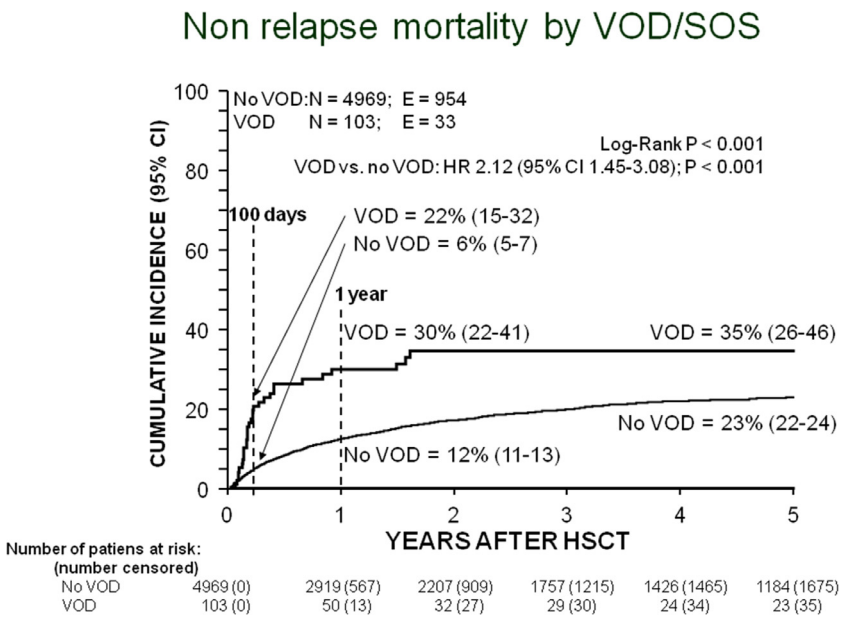

Figure 4. Cumulative incidence of NRM in patients who either did or did not experience SOS/VOD.

recurrence of original disease in children with malignancies or other transplant-related early and late fatal toxicities. In this Italian multicenter pediatric study, DF was the most frequently used therapeutic agent (55%), followed by UCD (36%) and steroids (29%) [30]. Because of the retrospective design of the study and the low number of patients who received DF as SOS/VOD prophylaxis, it is impossible to make any comparison on the efficacy of the different treatment modalities.

In conclusion, the results of this large pediatric, multicenter, retrospective study show that using the new EBMT pediatric criteria for the diagnosis and grading of SOS/VOD, the incidence of SOS/VOD observed in our study is lower (2%) than what was reported in previous studies. Second, female gender, age < 2 years, a diagnosis of HLH, and the use of Bu in the preparative regimen represent, in multivariable analysis, independent risk factors for the development of SOS/VOD. Third, the cumulative incidence of NRM of patients who developed SOS/VOD was higher than that in patients who did not develop this complication. Finally, the mortality rate directly attributable to SOS/VOD was 15.5%. Our findings may be used in the future to conduct prospective studies on this complication and its treatment.

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REFERENCES

1. Dalle JH, Giralt SA. Hepatic veno-occlusive disease after hematopoietic stem cell transplantation: risk factors and stratification, prophylaxis, and treatment. *Biol Blood Marrow Transplant*. 2016;22:400–409.
2. McDonald GB, Sharma P, Mattews DE, Shulman HM, Thomas ED. Venocclusive disease of the liver after bone marrow transplantation: diagnosis, incidence, and predisposing factors. *Hepatology*. 1984;4:116–122.
3. Coppel JA, Richardson PG, Soiffer R, et al. Hepatic veno-occlusive disease following stem cell transplantation: incidence, clinical course, and outcome. *Biol Blood Marrow Transplant*. 2010;16:157–168.
4. Dignan FL, Wynn RF, Hadzic N, et al. BCSH/BSBMT guideline: diagnosis and management of veno-occlusive disease (sinusoidal obstruction syndrome) following haematopoietic stem cell transplantation. *Br J Haematol*. 2013;163:444–457.
5. Corbacioglu S, Carreras E, Ansari M, et al. Diagnosis and severity criteria for sinusoidal obstruction syndrome/veno-occlusive disease in pediatric patients: a new classification from the European Society for Blood and Marrow Transplantation. *Bone Marrow Transplant*. 2017;53(2):138–145.
6. Mohty M, Malard F, Abecassis M, et al. Revised diagnosis and severity criteria for sinusoidal obstruction syndrome/veno-occlusive disease in adult patients: a new classification from the European Society for Blood and Marrow Transplantation. *Bone Marrow Transplant*. 2016;51:906–912.
7. Corbacioglu S, Cesaro S, Faraci M, et al. Defibrotide for prophylaxis of hepatic veno-occlusive disease in paediatric haemopoietic stem-cell transplantation: an open-label, phase 3, randomised controlled trial. *Lancet*. 2012;379:1301–1309.
8. Kaplan EL, Meier P. Non parametric estimation from incomplete observations. *J Am Stat Assoc*. 1958;53:457–481.
9. Gooley TA, Leisenring W, Crowley J, Storer BE. Estimation of failure probabilities in the presence of competing risks: new representations of old estimators. *Stat Med*. 1999;18:695–706.
10. Klein JP, Rizzo JD, Zhang MJ, Keiding N. Statistical methods for the analysis and presentation of the results of bone marrow transplants. Part 2: regression modeling. *Bone Marrow Transplant*. 2001;28:1001–1011.
11. Gray RJ. A class of K-sample tests for comparing the cumulative incidence of a competing risk. *Ann Stat*. 1998;16:1141–1154.
12. Reiss U, Cowan M, McMillan A, Horn B. Hepatic veno-occlusive disease in blood and bone marrow transplantation in children and young adults: incidence, risk factors, and outcome in a cohort of 241 patients. *J Pediatr Hematol Oncol*. 2002;24:746–750.
13. Maximova N, Ferrara G, Minute M, et al. Experience from a single paediatric transplant centre with identification of some protective and risk factors concerning the development of hepatic veno-occlusive disease in children after allogeneic hematopoietic stem cell transplant. *Int J Hematol*. 2014;99:766–772.
14. Cesaro S, Pillon M, Talenti E, et al. A prospective survey on incidence, risk factors and therapy of hepatic veno-occlusive disease in children after hematopoietic stem cell transplantation. *Haematologica*. 2005;90:1396–1404.
15. Cheuk DK, Wang P, Lee TL, et al. Risk factors and mortality predictors of hepatic veno-occlusive disease after pediatric hematopoietic stem cell transplantation. *Bone Marrow Transplant*. 2007;40:935–944.
16. Cacchione A, LeMaitre A, Couanet DV, et al. Risk factors for hepatic veno-occlusive disease: a retrospective unicentric study in 116 children autografted after a high-dose BU-thiotepa regimen. *Bone Marrow Transplant*. 2008;42:449–454.
17. Yakushijin K, Atsuta Y, Doki N, et al. Sinusoidal obstruction syndrome after allogeneic hematopoietic stem cell transplantation: Incidence, risk factors and outcomes. *Bone Marrow Transplant*. 2016;51:403–409.
18. Carreras E, Díaz-Beyá M, Rosiñol L, Martínez C, Fernández-Avilés F, Rovira M. The incidence of veno-occlusive disease following allogeneic hematopoietic stem cell transplantation has diminished and the outcome improved over the last decade. *Biol Blood Marrow Transplant*. 2011;17:1713–1720.
19. Corbacioglu S, Hönig M, Lahr G, et al. Stem cell transplantation in children with infantile osteopetrosis is associated with a high incidence of VOD, which could be prevented with defibrotide. *Bone Marrow Transplant*. 2006;38:547–553.

20. Ouachée-Chardin M, Elie C, de Saint Basile G, et al. Hematopoietic stem cell transplantation in hemophagocytic lymphohistiocytosis: a single-center report of 48 patients. *Pediatrics*. 2006;117:e743–e750.
21. Lee SH, Son MH, Sung KW, et al. Toxicity of tandem high-dose chemotherapy and autologous stem cell transplantation using carboplatin-thiotepa-etoposide and cyclophosphamide-melphalan regimens for malignant brain tumors in children and young adults. *J Neurooncol*. 2014;120:507–513.
22. Kami M, Mori S, Tanikawa S, et al. Risk factors for hepatic veno-occlusive disease after bone marrow transplantation: retrospective analysis of 137 cases at a single institution. *Bone Marrow Transplant*. 1997;20:397–402.
23. Naples JC, Skeens MA, Auletta J, et al. Anicteric veno-occlusive disease after hematopoietic stem cell transplantation in children. *Bone Marrow-Transplant*. 2016;51:135–137.
24. Myers KC, Dandoy C, El-Bietar J, Davies SM, Jodele S. Veno-occlusive disease of the liver in the absence of elevation in bilirubin in pediatric patients after hematopoietic stem cell transplantation. *Biol Blood Marrow Transplant*. 2015;21:379–381.
25. Carreras E. How I manage sinusoidal obstruction syndrome after haematopoietic cell transplantation. *Br J Haematol*. 2015;168:481–491.
26. Corbacioglu S, Richardson PG. Defibrotide for children and adults with hepatic veno-occlusive disease post hematopoietic cell transplantation. *Expert Rev Gastroenterol Hepatol*. 2017;11:885–898.
27. Richardson PG, Smith AR, Triplett BM, et al. Earlier defibrotide initiation post-diagnosis of veno-occlusive disease/sinusoidal obstruction syndrome improves day +100 survival following haematopoietic stem cell transplantation. *Br J Haematol*. 2017;178:112–118.
28. Corbacioglu S, Carreras E, Mohty M, et al. Defibrotide for the treatment of hepatic veno-occlusive disease: final results from the International Compassionate-Use Program. *Biol Blood Marrow Transplant*. 2016;22:1874–1882.
29. Strouse C, Richardson P, Prentice G, et al. Defibrotide for treatment of severe veno-occlusive disease in pediatrics and adults: an exploratory analysis using data from the Center for International Blood and Marrow Transplant Research. *Biol Blood Marrow Transplant*. 2016;22:1306–1312.
30. Gloude NJ, Jodele S, Teusink-Cross A, et al. Combination of high-dose methylprednisolone and defibrotide for veno-occlusive disease in pediatric hematopoietic stem cell transplant recipients. *Biol Blood MarrowTransplant*. 2017;53(2):138–145.