

Autologous hematopoietic stem cell transplant at a tertiary care centre in India: achieving comparable outcomes with adaptations

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Abstract

Autologous stem cell transplantation (ASCT) is the standard treatment for many high-risk solid tumors. Patients undergoing ASCT should be managed in a dedicated hematopoietic stem cell transplantation (HSCT) unit with isolation rooms, high-efficiency particulate air (HEPA) filters, and positive pressure. We report the outcomes of the first 20 pediatric patients who underwent ASCT in isolation rooms with no HEPA filters or positive pressure. Moreover, the isolation rooms were not part of a dedicated HSCT unit.

Data from 20 patients were analyzed. All patients included in the study underwent ASCT after harvest and cryopreservation of the hematopoietic stem cells (HSC). Furthermore, all patients also underwent myeloablative conditioning.

The most common indications for ASCT included high-risk neuroblastoma (HR-NB) (n=9) and refractory/relapsed Hodgkin's lymphoma (HL) (n=6). The median CD-34 positive HSC administered was 4.5 (0.8-21.9) million per kg. The median time to neutrophil and platelet engraftment was 16.5 (10-35) and 19 (10-87) days, respectively. Additionally, only one transplant-related mortality was observed and the mean time to discharge from the hospital was 27.6+8.3 days. The overall survival for all our patients was 75% at a median follow-up of 33.2 months (15 out of 20 patients survived), and the disease-free survival was 60% (median follow-up, 28.4 months). The overall survival for the patients with HL was 85.7% at a median of 45.3 months and for the HR-NB was 66.7% at a median of 34.9 months.

This study provides evidence that ASCT can be safely performed in isolation rooms without HEPA filters and positive pressure if expertise and supportive care are available. In settings with limited resources, such a model could help establish low-cost HSCT units.

Key words autologous SCT, pediatrics, outcomes

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Introduction

Bone marrow toxicity, which leads to myeloablation, is often a limiting factor in the treatment of malignancies by escalation of chemotherapy. The ability to harvest and store a patient's hematopoietic stem cells (HSC) and reinfuse them after myeloablative doses of chemotherapy allow better control of many tumors, making autologous stem cell transplantation (ASCT) an effective modality for the treatment of malignancies.

Moreover, ASCT is a standard treatment for solid tumors such as high-risk neuroblastomas (HR-NB), stage IV Ewing's sarcoma, and relapsed/refractory lymphomas. Recently, ASCT has been used as a clinical option for stage IV-A retinoblastoma (RB), Wilms tumor, brain tumors, and autoimmune diseases^{1,2}.

As infections are a major concern after highly myeloablative chemotherapy, it is recommended that patients undergoing ASCT be cared for in a dedicated hematopoietic stem cell transplant (HSCT) unit. In gen-

eral, HSCT units should have a minimum number of single-bed rooms, depending on the volume of the center. Additionally, it is recommended that the single rooms be positively pressured with differential pressure between the rooms and the hallway along with self-closing doors. Rooms should have more than 12 exchanges of air per hour, continuous air pressure monitoring, and high-efficiency particulate filters (HEPA filters)³.

In low-income and middle-income countries, the aforementioned units should be made accessible. Whether acceptable outcomes can be achieved through the best utilization of existing resources is a question that needs to be addressed. In this article, we report the outcomes of the first 20 pediatric patients who underwent ASCT as part of their treatment at a center that initiated the HSCT program. Moreover, ASCT procedures were performed in rooms without HEPA filters. The rooms were not part of the HSCT unit, and adult and pediatric patients of other medical and surgical specialties were admitted to the unit along with pediatric HSCT patients.

Patients and Methods

The ASCT program for the Division of Pediatric Oncology, Department of Pediatrics at the All India Institute of Medical Sciences, New Delhi, commenced in August 2017. We present the outcomes of the first 20 pediatric patients, with ages equal to or less than 14 years, who underwent ASCT treatment. Informed consent was obtained from all patients. The patients were followed up until April 2023. This retrospective study was approved by the Institutional Review Board of the All India Institute of Medical Sciences, New Delhi, India (IEC-396/17.07.2023).

Description of the rooms where the ASCT was performed

The ASCT procedures were performed in one of the 12 rooms located on the top floor of a five-story hospital wing. The main door to the passage was a manually operated glass door, followed by a sensor-operated automatic door. The rooms in which the ASCT was performed were on either side of the passage with self-closing hand-operated wooden doors. Patients from other specialties and those undergoing HSCT were admitted to the rooms (as the rooms were not a part of the dedicated HSCT unit). The rooms were air-conditioned, and before the admission of a patient, they were fumigated with a fogging machine using a solution of stabilized hydrogen peroxide 11%w/v and 0.01%w/v diluted silver nitrate solution. The machine was operated for 40 min, after which the room was

closed for another 45 min.

Only one attendant (in most cases, the mother) was allowed to be with the child during admission for ASCT treatment. Patients were kept in isolation from admission until discharge.

Stem cell collection and harvest

Peripheral blood HSCs were used in all patients. The patients underwent a femoral line insertion 1 day before harvest. Stem cell mobilization was performed using granulocyte colony-stimulating factor (G-CSF) alone, or G-CSF and plerixafor. Harvesting was performed via a Spectra-Optia apheresis system (Terumo BCT, Lakewood, Colorado, USA) using a continuous mononuclear cell collection program.

Stem cell cryopreservation and transfusion

After collection, the HSCs were cryopreserved at our stem cell facility using established protocols. The harvested stem cells were mixed with a freshly prepared cold cryoprotectant mixture (normal saline, dimethylsulfoxide, albumin, and heparin) in a ratio of 1:1, with a constant swirling of the cryobag. An aliquot of the harvested cryopreserved HSC was sent for sterility testing. The cryobag was properly labeled, sealed, and stored at -80°C for later use. At the time of infusion, the HSC cryobags were retrieved from the -80°C freezer and thawed at 37°C using a water bath at the patient's bedside. An aliquot of the thawed HSC was tested for stem cell viability using the trypan blue dye assay.

Central venous access

Central venous access for patients during autologous HSCT involved Hickman line insertion or the use of a peripherally inserted central catheter line.

Medical and Nursing care

The patients had a dedicated superspecialty trainee doctor assigned to their care. Each patient was examined by consultants twice daily. The nurse-to-patient ratio varied from 1:3 to 1:4. All nurses were experienced in the care of patients undergoing HSCT; however, they also managed other patients. Only healthcare personnel were allowed to enter the rooms after donning a cap and face mask, changing footwear, and thoroughly washing their hands with water or using an antiseptic handrub. The patients were provided cooked food from a common hospital kitchen. Patients were allowed to drink reverse osmosis-filtered water and commercially available mineral water.

Conditioning regimens

The conditioning regimens used for the lymphomas included carmustine, etoposide, cytarabine, and melpha-

lan (BEAM). In most cases, the regimen consisted of carmustine 300 mg/m² for 1 day, etoposide 200 mg/m² for 4 days, Ara-C 200 mg/m² for 4 days, and melphalan 140 mg/m² for 1 day⁴. We also used gemcitabine-based conditioning in Hodgkin lymphoma (HL) if the refractory disease was sensitive to gemcitabine-containing salvage chemotherapy. Furthermore, in one patient, we used carmustine 300 mg/m² for 1 day, gemcitabine 1,000 mg/m² for 2 days, and melphalan 140 mg/m² for 1 day⁵. In another study, busulfan was administered at 80 mg/m² for 4 days, gemcitabine at 2,000 mg/m² for 2 days, and melphalan at 120 mg/m² for 2 days⁶.

The conditioning regimen used in the HR-NB group was a combination of busulfan 4 mg/kg/day for 4 days and melphalan 70 mg/m² for 2 days⁷ or a combination of carboplatin (doses based on Calvert's formula) for 4 days, etoposide 338 mg/m² for 4 days, and melphalan 70 mg/m² for 3 days⁸.

Patients with stage IV-A RB who underwent ASCT received either carboplatin 350 mg/m² for 5 days, etoposide 350 mg/m² for 5 days, and cyclophosphamide 1,600 mg/m² for 4 days⁹ or carboplatin 500 mg/m² for 3 days, thiotepa 300 mg/m² for 3 days, and etoposide 250 mg/m² for 3 days¹⁰.

Supportive care

All patients received fluconazole and acyclovir for antifungal and antiviral prophylaxis, respectively. All patients received ursodeoxycholic acid as prophylaxis against veno-occlusive disease (VOD). The treatments were terminated on Day +90. Co-triamoxazole was stopped before conditioning and restarted on day +30 after engraftment had been achieved.

Blood component and growth factor support

Packed red blood cells (PRBC) were transfused when the hemoglobin level was less than 7 g/dL, and platelets were transfused when they were less than 20,000/cumm. All patients received G-CSF from day +1 until the neutrophils were > 1,000/cumm. All the PRBC and platelets were transfused after irradiation.

Treatment of infections

Any episode of fever after conditioning defined as a single temperature of more than 38.3°C (101°F) or more than 38.0°C (100.4°F) lasting for more than 1 hour was treated with broad-spectrum antibiotics. The initial antibiotics of choice were piperacillin-tazobactam and amikacin. Teicoplanin was added upfront in the presence of mucositis or 48 hours later if the fever persisted. If no response was observed, meropenem was added. Systemic antifungals were initiated if the fever persisted for 5 days or more despite antibiotic treatment or if suspicion of an invasive fungal infection was pre-

sent based on radiological/computed tomography scan findings. If fungal infection was suspected, serum galactomannan levels were evaluated. Cytomegalovirus (CMV) infection was suspected if fever or cytopenia persisted even after the addition of antifungals, and CMV-polymerase chain reaction (PCR) was requested from the blood samples.

Recording of data and statistical analysis

The data of all patients were prospectively recorded after ASCT. For this retrospective study, the data were retrieved and analyzed. Non-normally distributed continuous data were reported as median (range), and normally distributed data were presented as mean ± standard deviation. Qualitative data were expressed as numbers and percentages. Kaplan-Meier survival curves were prepared for individual diseases, as well as for the entire cohort.

Results

Twenty pediatric patients underwent ASCT between August 2017 and the present. The indications for ASCT included HR-NB (n=9), refractory/relapsed HL (n=6), relapsed or refractory non-HL (NHL) (n=3), and stage IV-A RB (n=2). Sixteen patients were males and four were females. The mean age of the patients at the time of transplant was 84.9±39.5 months.

Myeloablative conditioning chemotherapy was used in all cases. Carboplatin, etoposide, and melphalan (n=4) and busulfan and melphalan (BuMel) (n=5) were the conditioning chemotherapies administered to patients with HR-NB. In patients with HL, BEAM (n=5) or gemcitabine-based conditioning (n=2) was used. For NHL, the conditioning chemotherapy was BEAM, and for RB (n=2), the chemotherapy included thiotepa, etoposide, and carboplatin in one patient and carboplatin, etoposide, and cyclophosphamide in the other. (**Table 1**)

The stem cell dose administered was available for 18 patients. The median CD-34 positive HSCs administered were 4.5 (0.82-1.9) million stem cells per kg of recipient body weight. The median time to neutrophil engraftment was 16.5(10-35) days and the median time to platelet engraftment was 19 (10-87) days. One transplant-related mortality (TRM) was observed and the mean time to discharge from the hospital for the remaining patients (n=19) from the day of ASCT was 27.6±8.3 days. (**Table 1**)

The overall survival (OS) for all our patients was 75% at a median follow-up of 33.2 months (15 out of 20 patients survived), and the disease-free survival (DFS) was 60% (at a median follow-up of 28.4 months). The overall survival for the HL patients was

Table 1. Details of all patients in relation to autologous stem cell transplant

SI no	Age (m)	Indication for autologous HSCT	Conditioning	CD-34 (HSC) transfused in million/kg	Neutrophil engraftment (day)	Platelet engraftment (day)	Day of discharge	Status as on 30.08.2023 [Alive/Dead (cause of death)]	Follow up in days since HSCT as on 30.08.2023/ Last contact
1	88	Relapsed Burkitt's Lymphoma	BEAM	0.8	16	19	20	Alive	2,192
2	42	HR-NB	CEM	1.3	31	31	34	Dead (Relapse and progressive disease)	1,076
3	55	Refractory NB	CEM	NA	22	34	37	Dead (Refractory and progressive disease)	1,477
4	120	Relapsed HL	BEAM	10	32	31	36	Alive	1,954
5	120	Relapsed HL	Ge BCNU Mel	NA	17	19	TRM	Dead (HSV encephalitis)	41
6	78	HR-NB	CEM	12.5	15	41	40	Alive	1,602
7	78	HR-NB	Bu Mel	2.6	14	11	27	Dead (Relapse and progressive disease)	357
8	36	HR-NB	CEM	2.9	14	11	20	Alive	1,559
9	120	Relapsed HL	BEAM	2.1	11	13	26	Alive	1,531
10	108	Relapsed HL	BEAM	10	10	15	21	Alive	1,367
11	72	Refractory HL	BuGeMel	5.9	35	87	35	Alive (Relapse and progressive disease)	1,349
12	156	Refractory ALCL	BEAM	3.3	10	10	21	Dead (Relapse and progressive disease)	703
13	36	HR-NB	Bu Mel	9.2	16	25	48	Alive (Relapsed and progressive disease)	363
14	44	HR-NB (n myc positive)	Bu Mel	1	18	24	28	Alive	1,049
15	65	RB stage IV-A	CARBOPEC	5.7	18	18	21	Alive	758
16	36	HR-NB	Bu Mel	12	12	12	21	Alive	661
17	84	Relapsed HL	BEAM	21.9	17	19	17	Alive	405
18	132	RB stage IV-A	TEC	6.2	12	20	31	Alive (Relapsed on salvage treatment)	624
19	168	Refractory DLBCL	BEAM	1.1	18	18	20	Alive	946
20	60	HR-NB	Bu Mel	2.2	19	19	22	Alive	224

SI no, Serial number; HSCT, Hematopoietic Stem Cell transplantation; HR_NB, high risk neuroblastoma; NB, neuroblastoma; HL, hodgkin lymphoma; ALCL, anaplastic large cell lymphoma; RB, retinoblastoma; DLBCL, diffuse large B cell lymphoma; BEAM, carmustine, etoposide, Ara-C and melphalan; CEM, carboplatin, etoposide and melphalan; Ge BCNU Mel, gemcitabine, carmustine and melphalan; BuMel, busulfan and melphalan; BuGeMel, busulfan, gemcitabine and melphalan; CARBOPEC, carboplatin, etoposide and cyclophosphamide; TEC, thiotepa, etoposide and carboplatin; HSC, hematopoietic stem cell; TRM, transplant related mortality; HSV, herpes simplex virus; NA, Not available

85.7% at a median of 45.3 months and the HR-NB was 66.7% at a median of 34.9 months. One TRM was observed in a patient with relapsed HL due to herpes simplex virus (HSV) encephalitis on day +41 post-HSCT. The magnetic resonance imaging (MRI) findings of the patient's brain were suggestive of HSV encephalitis, and HSV PCR of the cerebrospinal fluid (CSF) demonstrated positive results. Additionally, four deaths (three for HR-NB and one for NHL) were observed, all of which were attributed to relapse and progressive disease

in all cases. (**Table 1**) The Kaplan-Meier curves for OS and DFS for all patients and OS for HR-NB and relapsed/refractory HL are displayed in **Figure 1**.

During admission for ASCT, all the patients experienced febrile neutropenia (FN) and required antibiotics. Need for systemic antifungal treatment was required in 40% of cases. Blood cultures were positive in five cases. Moreover, two cases exhibited positivity for serum galactomannan, of which one also demonstrated evidence of IFI on imaging (**Table 2**).

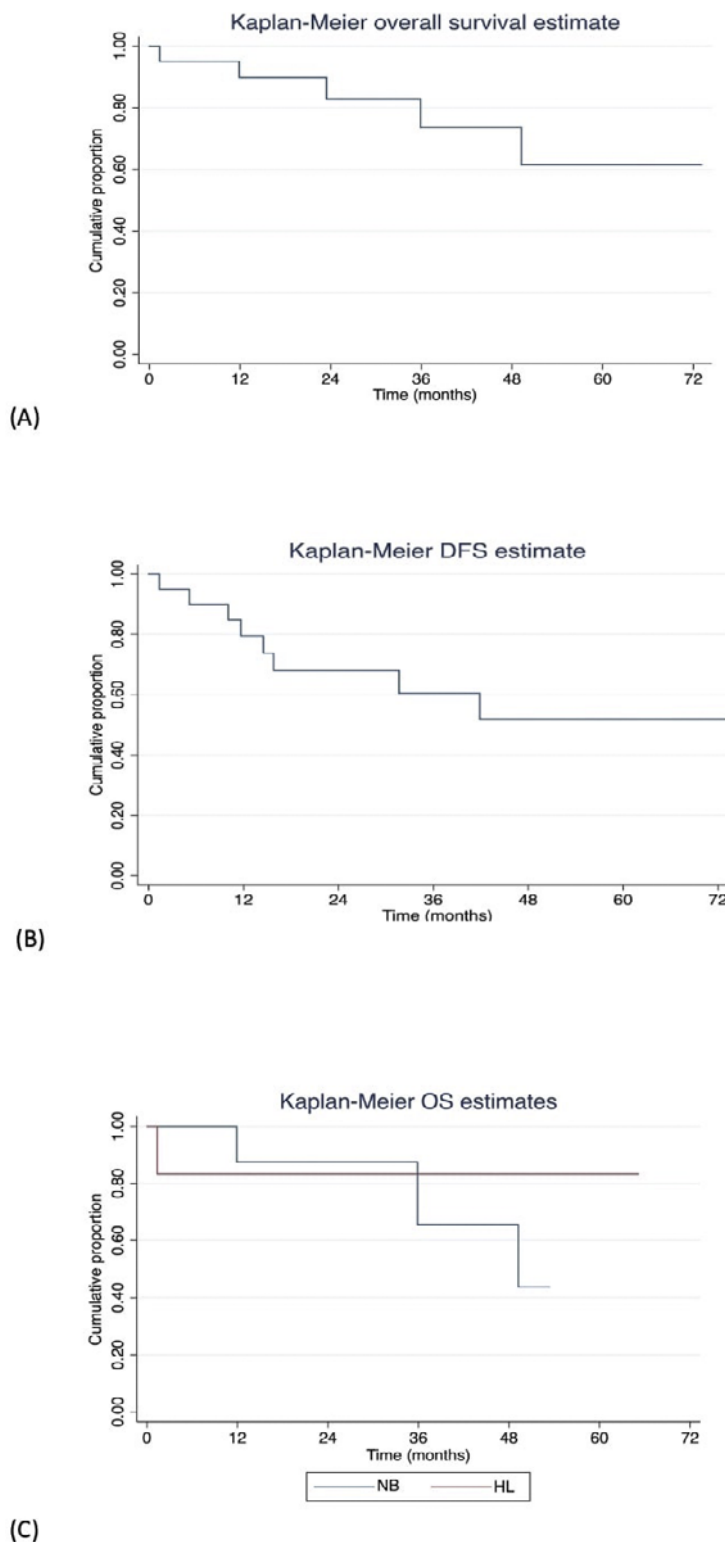


Figure 1. Kaplan-Meier survival curves
 A: Overall survival for all patients; B: Disease free survival for all patients; C: Overall survival of neuroblastoma (blue; n=9) and Hodgkin lymphoma (red; n=6)

Additionally, CMV reactivation was suspected in two cases but was ruled out by quantitative PCR. None of the patients had VOD.

Discussion

Good short- and long-term outcomes were observed in the first 20 pediatric ASCT cases performed at our

Table 2. Infection details of all patients

SI no	Documented infections	FN	Antibiotics	Antifungals	Suspicion of viral infection
1	None	Yes	Yes	No	No
2	Blood: <i>Acinetobacter baumannii</i>	Yes	Yes	No	No
3	Blood: <i>Acinetobacter baumannii</i> Urine: <i>Klebsiella oxytoca</i> Stool: Adenovirus	Yes	Yes	Yes	No
4	None	Yes	Yes	No	No
5	HSV encephalitis	Yes	Yes	No	Yes CMV PCR in blood negative HSV PCR IN CSF positive
6	Blood: <i>Escherichia coli</i> Stool: <i>Giardia sp</i>	Yes	Yes	Yes	No
7	None	Yes	Yes	Yes	No
8	Stool: <i>Entamoeba sp</i>	Yes	Yes	Yes	No
9	Blood: <i>Pseudomonas stutzeri</i>	Yes	Yes	Yes	No
10	None	Yes	Yes	No	No
11	Perianal cellulitis Galactomannan positive	Yes	Yes	Yes	Yes CMV PCR in blood negative
12	None	Yes	Yes	No	No
13	Blood: <i>Klebsiella pneumoniae</i>	Yes	Yes	No	No
14	Galactomannan positive Stool: <i>Clostridium difficile</i>	Yes	Yes	Yes	No
15	None	Yes	Yes	No	No
16	None	Yes	Yes	No	No
17	None	Yes	Yes	No	No
18	Fungal pneumonia Galactomannan positive	Yes	Yes	No	No
19	Furuncles over buttocks	Yes	Yes	Yes	No
20	None	Yes	Yes	No	No

SI no, Serial number; FN, Febrile neutropenia; CMV, Cytomegalovirus; HSV, herpes simplex virus; PCR, polymerase chain reaction; CSF, cerebrospinal fluid; HSC, hematopoietic stem cell; HSV, herpes simplex virus

center. These ASCTs were performed in normal isolation rooms in a common ward; the rooms did not have HEPA filters or positive pressure.

The use of HEPA-filtered rooms is recommended for ASCT recipients to reduce the chances of nosocomial infections as a result of prolonged neutropenia¹¹. On the other hand, improved supportive care and timely engraftment due to the use of peripheral blood HSCs have made ASCT a safe procedure with low mortality¹². The need for HEPA filtration and positive pressure rooms, especially for ASCT, has been debated. Ruiz-Argüelles et al in 2022 published a review in which they recommended that ASCT be performed in an outpatient setting. They considered the need for HEPA-filtered rooms with laminar flow to be optional¹³. A similar experience has been published by Kumar et al., who demonstrated acceptable infection rates and survival in 40 patients who underwent allogeneic HSCT in non-HEPA-filtered rooms without positive pressure¹⁴. Tsai et al. demonstrated that the 100-day mortality in adult patients with multiple myeloma did not differ if the room used was without a HEPA filter¹⁵.

In our experience, we identified an OS of 75% at a median follow-up of 33.2 months. In patients with HR-NB, a disease with a poor prognosis, the OS was 66%. Although our study was limited to 20 patients, these were the first ASCTs performed in our unit.

Sharma et al. demonstrated a 55% OS in a retrospective analysis of 36 patients, (half of them with HR-NB) who underwent ASCT at their center¹⁶. In another study from India, which also included adult patients with multiple myeloma, Yadav et al. demonstrated a 55.6% OS at a median follow-up of 114 days (range: 21-617 days). The median duration of neutrophil engraftment in their study was 12 days (range, 9-30)¹⁷. Relapsed/refractory HL had a 5-year OS of about 68%¹⁸ and in HR NB, an EFS of 50% has been demonstrated by Landenstein et al. in patients undergoing ASCT with BuMel conditioning¹⁹. In our experience, relapsed/refractory HL and HR-NB were the common conditions for which autologous HSCT was performed and the OS observed in our patients was acceptable.

Rates of FN in ASCT up to 93% have been documented, with microbiologically or clinically docu-

mented infections in 50%²⁰. A FN rate of 89% has been documented among 113 ASCT recipients, in adult patients not receiving any antibiotic prophylaxis²¹. Some studies have demonstrated an improvement in FN rates with the use of prophylactic antibiotics, however, we did not use prophylactic antibiotics during the ASCT^{22,23}. All patients had FN and required antibiotics, but all of them recovered completely. Blood culture results were positive in five instances. Our FN and blood culture positivity rates were acceptable in comparison with those reported in other studies. In our study, one case of TRM was observed due to HSV encephalitis.

The neutrophil and platelet engraftment time in patients is comparable to the time documented by others²⁴. Jain et al reported the outcomes of 35 patients with HR-NB who underwent ASCT in regular multi-bedded ward beds without HEPA filtration and cryopreservation of the HSC. They reported a median time to neutrophil engraftment of 11 days, antibiotic usage of 86%, and an OS at 3 years of 41%²⁵.

Our study has some limitations because of the retrospective nature and the inclusion of only 20 patients. However, the study provides evidence that ASCT can be safely performed in non-HEPA filtered and non-positive pressure isolation rooms if expertise and supportive care are available. In resource-limited settings, such a model could help establish low-cost HSCT units.

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Author Contributions

AKG designed the study, analyzed the data, and wrote the manuscript; AKG, JPM, RS, PN, SM and PC were involved in the clinical care of the patients and reviewed the manuscript; All the authors approve the final version of the manuscript.

Conflicts of Interest

The authors declare no conflict of interest. Disclosure forms provided by the authors are available on the website.

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None to declare

Ethical Approval

This retrospective study was approved by the institutional review board of All India Institute of Medical Sciences, New Delhi, India (IEC-396/17.07.2023).

Informed Consent

Informed consent was obtained from all participants or their parents.

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