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Pediatric males receiving hematopoietic stem cell transplant lose their male disadvantage in disease risk after the procedure: A retrospective observational study

Laura De Nardi[1] | Roberto Simeone[2] | Lucio Torelli[1] | Alessandra Maestro[3] | Davide Zanon[3] | Egidio Barbi[1,4] | Natalia Maximova[4]

[1]Department of Medicine, Surgery and Health Sciences, University of Trieste, Trieste, Italy

[2]Department of Transfusion Medicine, ASUGI, Trieste, Italy

[3]Pharmacy and Clinical Pharmacology Department, Institute for Maternal and Child Health—IRCCS "Burlo Garofolo", Trieste, Italy

[4]Department of Pediatrics, Institute for Maternal and Child Health—IRCCS "Burlo Garofolo", Trieste, Italy

Correspondence
Maximova Natalia, Bone Marrow Transplant Unit, Institute for Maternal and Child Health—IRCCS "Burlo Garofolo", 34137 Trieste, Italy.
Email: natalia.maximova@burlo.trieste.it

Abstract

Sex differences play a relevant role in cancer susceptibility, incidence and survival. Exploring such differences is difficult because of the close interplay of genetic, epigenetic and hormonal factors. However, a better understanding of the role of such disparities in cancer mechanisms could improve its prevention and therapy. Our study explores how sex differences in pediatric outcomes vary after undergoing first and advanced-line therapy for hematological malignancies. The primary goal was to evaluate if sex differences in pediatric outcomes after first-line therapy persist after allogeneic hematopoietic stem cell transplantation (HSCT). The secondary goal was to analyze sex differences in disease risk at onset and pediatric outcomes after first-line therapy to compare our results with the literature's reported results. Among a total of 485 patients (280 males, 205 females) admitted for hematological malignancies, disease risk at the onset was significantly higher in males ($P < .05$). One hundred and seventy-four patients (111 males and 63 females) had a high-risk disease requiring HSCT. Before HSCT, all patients underwent myeloablative conditioning, which substantially impaired gonadal function. Although the number of boys undergoing HSCT was almost double that of girls, there were no sex-related differences in overall survival, cancer relapse and complications after HSCT exposure ($P > .05$). These findings suggest that the existing sex differences in cancer risk ab initio can be somehow

Abbreviations: ALL, acute lymphoblastic leukemia; AR, androgen receptor; ASBMT, American Society for Blood and Marrow Transplantation; CRI, cumulative incidence of relapse; DFS, disease-free survival; DRM, disease-related mortality; EFS, event-free survival; GvHD, graft-vs-host disease; HRG, hypothalamic-pituitary-gonadal; HSCT, hematopoietic stem cell transplantation; MDS, myelodysplastic syndrome; miRNA, microRNA; OS, overall survival; SD, standard deviation; SOS, sinusoidal obstruction syndrome; TRM, transplant-related mortality; VEGFR2, vascular endothelial growth factor receptor-2.

Previous presentation: Some of the data were previously presented during the EHA 2021 Meeting.

flattened by a conditioning regimen, shedding new light on the role of hormonal factors in cancer mechanism and management.

KEYWORDS
cancer risk, gonadal impairment, male disadvantage, sex disparity, transplant-related outcomes

What's new?

Sex differences resulting in a male disadvantage have been observed in cancer susceptibility, incidence and survival. A better understanding of the role of sex differences in cancer mechanisms could improve prevention and therapy. This pediatric oncological population-based study suggests that the existing sex differences in cancer risk ab initio can be somehow flattened by a myeloablative conditioning regimen before hematopoietic stem cell transplantation, which substantially impairs gonadal function. The findings shed new light on the role of hormonal factors in cancer mechanisms and management, suggesting a major role of androgens in cancer relapse for pediatric hematological malignancies.

1 | INTRODUCTION

Sex differences in cancer incidence and mortality throughout the world are a recognized fact. Several epidemiological studies in the literature agree that in a wide range of cancer types unrelated to reproductive function,[1] females have a lesser risk of cancer and better prognosis than males, independent of the population's ethnicity.[2] The analysis of age- and sex-specific cancer incidence data from Cancer Incidence in Five Continents provided by the International Agency for Research on Cancer documented the universal nature of the sex disparity in cancer.[3] Males not only develop cancer more often but also are more likely to die from their disease,[4] and this "male disadvantage" in terms of malignancies risk applies to every age.[5] Males are also at higher risk than females concerning childhood cancer, with a male:female ratio for all incident cancers being around 1.2, and with substantial sex differences in particular for T cell acute lymphoblastic leukemia (T-ALL) and hepatoblastoma.[6] Prognosis is also worse in boys than in girls,[7] as confirmed by data showing that significantly more males than females affected by acute lymphoblastic leukemia (ALL) need to undergo hematopoietic stem cell transplantation (HSCT).[8]

Differences in sex chromosomes, immune systems and environmental exposures may contribute to the sexual disparity in cancer susceptibility and progression.

Assessing the actual weight of each one of these factors is challenging. To date, immune surveillance has been recognized as one of the main physiologic mechanisms protecting against cancer progression.[9-11] Generally, females are reported to mount higher innate and adaptive immune responses than males, which could result, on the one hand, in faster clearance of pathogens and cancer cells, and on the other, contribute to increased susceptibility to inflammatory and autoimmune diseases. Sex hormones, including estrogens, progesterone and androgens, and sex-chromosome-related genes, are considered the main factors are guiding such differences in the immune response.[5] The expression of X-linked genes and microRNA (miRNA), as well as sex steroid hormone signaling through hormone receptors in immune cells, can affect responses to immunological stimuli, possibly resulting in male-female differential in incidence rates not only of cancer but also of infectious diseases, inflammatory disorders, autoimmune diseases and vaccine responses.[12] Such sexual dimorphism in immune surveillance is the major contributor to the gender effect on cancer mechanisms.[1,6]

Notably, myeloablative conditioning regimens in HSCT are associated with a significant risk of infertility due to gonadal impairment, with hormonal levels remaining low for years after treatment and sometimes throughout life.[13,14]

Our study aims to explore sex differences in first and advanced-line therapy outcomes in pediatric oncology. In particular, we investigated whether the known sex differences occurring after first-line therapy, and leading to a higher number of male patients requiring HSCT, persist after transplant. Our study setting may represent an experimental human model that reveals the possible effects of the impairment of gonadal hormones on cancer progression, which is poorly explored in the literature.

2 | MATERIALS AND METHODS

2.1 | Study design and outcomes

This is a single-center retrospective cohort observational study conducted at the pediatric Transplant Center of the Institute for Maternal and Child Health of Trieste, Italy.

The study's primary goal was to evaluate if sex differences in pediatric outcomes occurring after first-line therapy persist after allogeneic HSCT. Secondary goals were to analyze sex differences in disease risk at onset and after first-line therapy to compare our results with the literature reported. In particular, the main examined outcomes were overall survival (OS) and cumulative incidence

of relapse (CRI) among patients who underwent advanced-line therapy. OS is the time from HSCT until death from any cause, and CRI is the time from HSCT until relapse of the primary disease that required a transplant. Thus, CRI includes both disease-related mortality (DRM) and disease relapse after HSCT. After advanced-line therapy, the analysis of outcomes was limited to patients undergoing allogeneic HSCT for reasons of statistical convenience. Tertiary goals assessed sex differences in HSCT-related complications and transplant-related mortality (TRM) at 12 months. HSCT-related complications included conditioning-related organ toxicity such as mucositis and radiation-induced lung injury, sinusoidal obstruction syndrome (SOS) and other forms of vascular endothelial syndrome and graft vs host disease (GvHD). TRM is the time from HSCT until death from any cause, with relapse as a competing event.

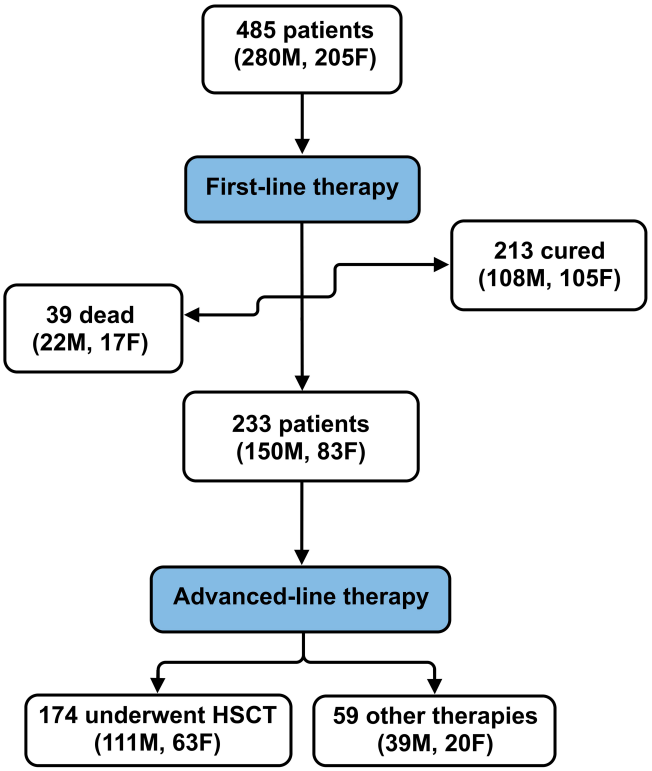

FIGURE 1 Flow diagram showing the breakdown of patients

2.2 | Patient population, analysis and treatment

Potentially eligible participants were pediatric patients between 0 and 18 years of age who received a diagnosis of malignant diseases at our Institute between 2000 and 2018. Diagnoses of malignant diseases included acute or chronic leukemia and myelodysplastic syndromes (MDSs). Leukemia risk at the onset was defined according to a previously published classification.[15] Transplant disease risk index was defined according to a previously published classification considering both the risk upon the first presentation of each patient and the risk after first-line therapy.[16] Patients who had malignant diseases previously diagnosed or treated at other hospitals were excluded from our study. After first-line therapy, patients' outcomes were collected and examined; those who died or recovered from their malignancy were excluded from further analysis. Patients who underwent advanced-line therapy of any kind were followed, and their outcomes were analyzed. Patients who underwent any therapy other than HSCT were excluded from the final analysis due to the small sample sizes. All patients enrolled in the transplant program were analyzed individually. All data were collected from clinical records, analyzed anonymously. All patients' parents had provided informed consent for research purposes, and each patient was allocated through an identification code.

Indications for HSCT conformed to the American Society for Blood and Marrow Transplantation (ASBMT) task force

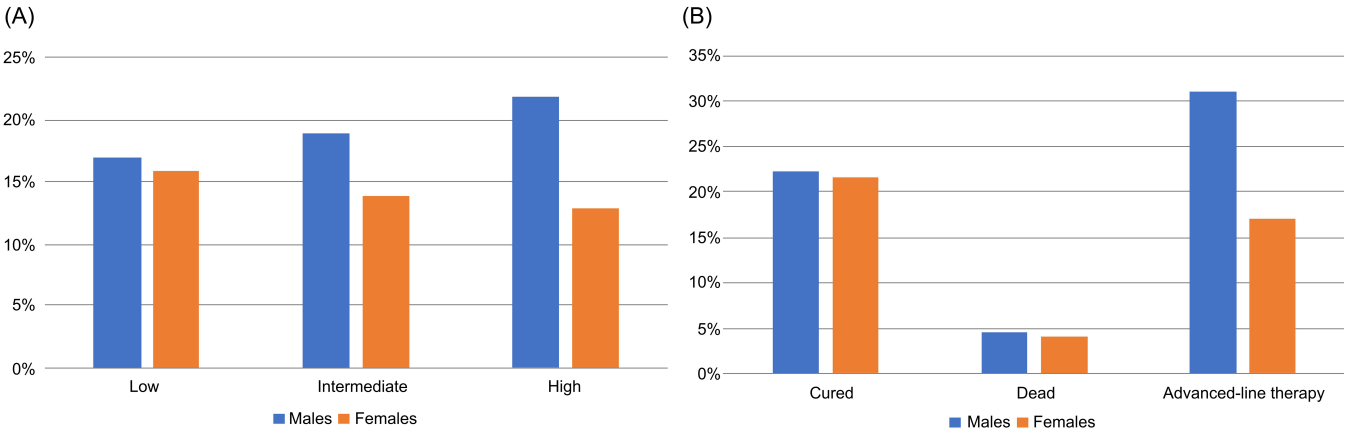

FIGURE 2 (A) Histogram showing disease risk at diagnosis in males and females. Males show a statistically significant higher risk than females ($P = .043$). (B) Histogram showing disease outcomes after first-line therapy: cured, dead and requiring advanced-line therapy. Significantly more males than females require advanced-line therapy ($P = .006$)

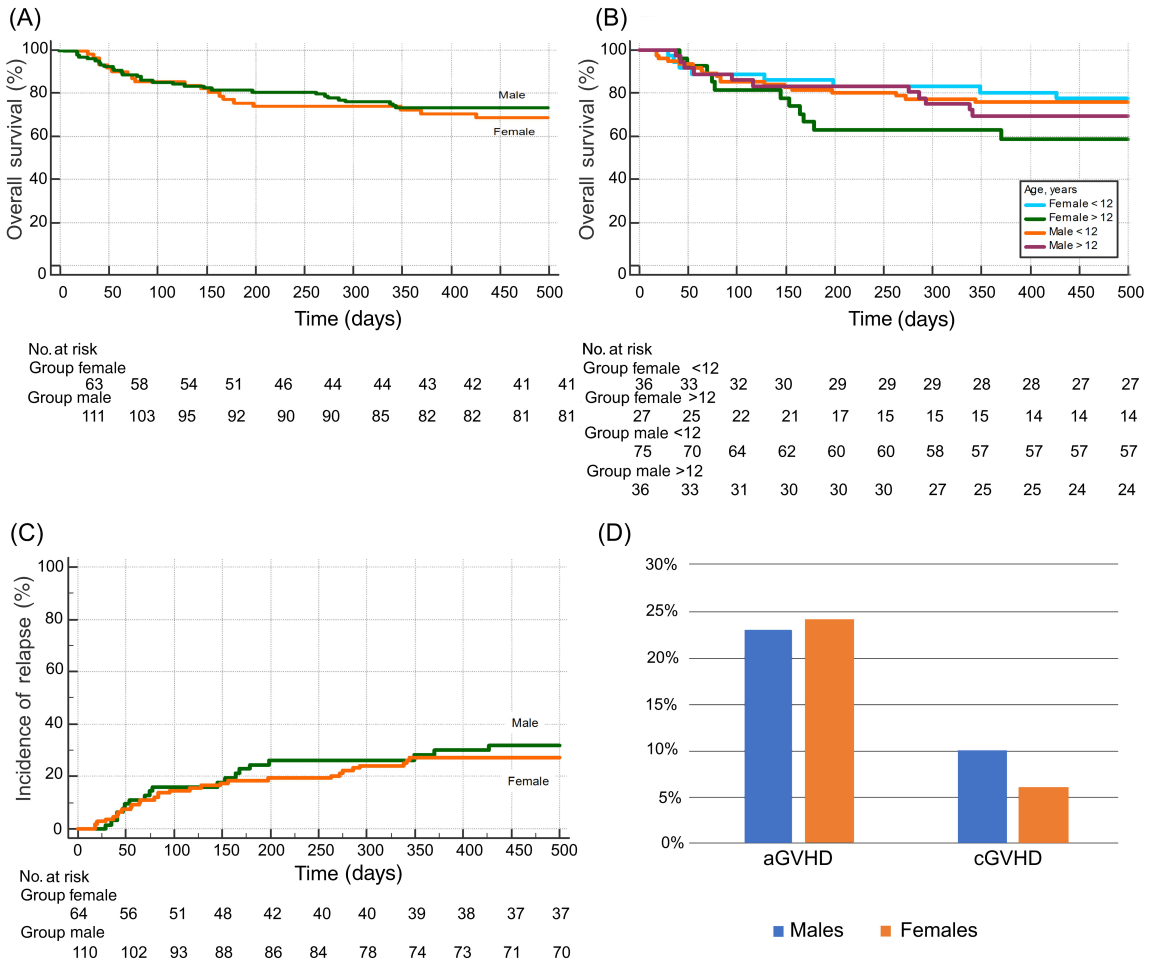

FIGURE 3 (A) Kaplan-Meier curves showing sex differences in overall survival (OS). There are no statistically significant differences ($P = .655$). (B) Kaplan-Meier curves showing sex differences in OS divided for age groups. There are no statistically significant differences ($P = .324$). (C) Kaplan-Meier curves showing sex differences in cumulative relapse incidence (CRI). There are no statistically significant differences ($P = .538$). (D) Histogram showing posttransplant acute and chronic graft-vs-host disease (GvHD) in males and females. There are no statistically significant differences were found ($P = .397$)

recommendations for pediatric patients.[17] All patients who underwent allogeneic HSCT were treated according to standard myeloablative protocols, as previously described in the literature.[18] High-dose posttransplantation cyclophosphamide-based platforms were administered to all haploidentical recipients so that composite endpoints across conditioning intensity, donor type and HLA match were comparable.[19]

Transplant-related toxicity was graded for each sex according to The National Cancer Institute Common Terminology Criteria for Adverse Events (CTCAE v. 5.0), a standardized list of adverse effects terms commonly found in oncology,[20] while acute and chronic GvHD was defined according to standardized criteria.[21,22] For each patient, the following variables were collected: sex, age when undergoing HSCT, underlying disease, disease risk, type of conditioning regimen, stem-cell source and donor type, time of engraftment,[23] rates of engraftment failure and recovery time from chemotherapy-induced toxicity.

2.3 | Statistical analysis

Sample size: Setting $\alpha = .05$ and $\beta = .20$, and assuming an effect size of 0.4, a minimum total sample size of 170 patients was required (presuming the same number of patients for each gender group) to assess the "average gender effect" on transplant outcomes and transplant-related toxicity at 12 months. Categorical variables are presented as frequency and percentage, and χ^2 test or Fisher's exact test are used as appropriate. Continuous data are presented as mean and SD. To evaluate sex differences over time in OS and incidence of relapses, Kaplan-Meier analyses are used and Kaplan-Meier plots are compared using the log rank test. For secondary outcomes, differences between the two groups for continuous variables are assessed using the non-parametric Mann-Whitney *U* test, while Fisher's exact test is adopted for categorical variables. $P < .05$ is considered statistically significant. Statistical analysis has been performed using GraphPad Prism (RRID: SCR_002798) software.

TABLE 1 Comparison of the differences in HSCT-related complications and transplant

Variables	Male (n = 111)	Female (n = 63)	*P*-value
Age at transplant, years, mean (±SD)	8.3 (6)	9.8 (5)	.095
Underlying disease, number (%)			
Acute lymphoblastic leukemia	61 (55)	38 (60)	.273
Acute myeloid leukemia/MDS	50 (45)	25 (40)	.453
Transplant-related toxicity, number (%)			
Mucosal I-II grade	78 (70)	39 (62)	.185
Mucosal III-IV grade	28 (25)	21 (33)	.301
Gastrointestinal I-II grade	82 (74)	49 (77)	.856
Gastrointestinal III-IV grade	28 (25)	13 (21)	.466
Hepatic I-II grade	40 (36)	27 (43)	.519
Hepatic III-IV grade	19 (17)	11 (18)	>.999
Pulmonary	8 (7)	3 (5)	.328
Renal	13 (12)	9 (14)	.646
Engraftment, days, mean (±SD)			
White blood cells	17.6 (7)	16.9 (6)	.733
Neutrophils	17.6 (7)	16.4 (6)	.332
Platelets	26.2 (26)	25.9 (24)	.794
Infection events, number (%)			
Sepsis	15 (13)	9 (14)	>.999
Fungal	22 (20)	17 (27)	.349
Viral	83 (75)	55 (87)	.122
Virus coinfection, virus number, mean (±SD)	1.8 (1)	2.5 (2)	.003
Transplant outcomes[a], number (%)			
Overall survival	82 (74)	44 (69)	.482
Event-free survival	57 (52)	40 (63)	.206
Early death[b]	15 (13)	9 (14)	>.999
Transplant-related mortality	14 (13)	13 (21)	.198
Disease-related mortality	15 (13)	6 (9)	>.999
Successive HSCT, number (%)	28 (25)	6 (9)	.01
Event-free survival	10 (36)	4 (67)	.577

Abbreviations: HSCT, hematopoietic stem cell transplantation; MDS, myelodysplastic syndrome; SD, standard deviation.
[a]Transplant outcomes at last follow-up.
[b]Death within the first 3 months from HSCT.

3 | RESULTS

Four hundred and eighty-five patients (280 males, 205 females) were admitted to the Institute for malignant diseases between 2000 and 2018 (Figure 1). Disease risk at the onset was significantly higher in male patients ($P < .05$) (Figure 2A). All children underwent first-line chemotherapy, after which 213 (44%) were cured, and 39 (8%) died. No differences by sex were observed between these two groups (Figure 2B).

Out of the remaining 233 patients (48%), who needed advanced-line therapy for their primary disease, 83 were females (36%) and 150 were male (64%). Among these patients, 174 underwent HSCT (75%), while 59 required other types of treatment (25%). These other therapies were heterogeneous and included one or more bouts of chemotherapy (38 patients, 23 of them male), surgery/radiotherapy (15 patients, 10 of them male), immunotherapy (3 patients, 2 of them male) or cell-based therapy (3 males). The HSCT cohort included 111 males (64%) and 63 females (36%). The mean (±SD) age at HSCT was 8.3 (5.6) years for boys and 9.8 (4.9) years for girls.

3.1 | OS, relapse and TRM

Among the 174 patients undergoing HSCT, 15 males (9%) and 6 females (3%) died from primary disease relapse, providing a DRM of 12%. Fourteen males (8%) and 13 females (7%) died from transplant-related

TABLE 2 Comparison of the differences in HSCT outcomes and transplant-related complications when donor-recipient pairs are matched and mismatched for sex

Variables	D/R matched M/M (n = 63)	D/R matched F/F (n = 25)	*P*-value	D/R mismatched M/F (n = 38)	D/R mismatched F/M (n = 48)	*P*-value
Transplant-related toxicity, number (%)						
Mucosal I-II grade	47 (73.4)	17 (68)	.599	22 (58)	30 (64)	.825
Mucosal III-IV grade	13 (20.6)	5 (20)	>.999	15 (39)	16 (33)	.653
Gastrointestinal I-II grade	50 (79.4)	19 (76)	.777	29 (76)	32 (67)	.351
Gastrointestinal III-IV grade	13 (20.3)	4 (16)	.769	9 (24)	15 (32)	.477
Hepatic I-II grade	23 (36.5)	9 (36)	>.999	18 (47)	17 (35)	.279
Hepatic III-IV grade	10 (15.6)	3 (12)	.751	8 (21)	9 (19)	.793
Pulmonary	4 (6.3)	0	.574	3 (8)	4 (8)	>.999
Renal	10 (15.6)	2 (8)	.496	7 (18)	4 (8)	.203
Transplant outcomes[a], number (%)						
Overall survival	45 (71)	20 (80)	.591	24 (63)	37 (77)	.232
Event-free survival	31 (50)	18 (72)	.061	21 (55)	27 (55)	>.999
Early death[b]	10 (16)	4 (16)	>.999	9 (24)	5 (11)	.142
Transplant-related mortality	7 (19)	3 (12)	>.999	6 (16)	7 (15)	>.999
Disease-related mortality	11 (17)	1 (4)	.167	5 (13)	4 (8)	.5
Successive HSCT, number (%)	14 (22)	3 (24)	.375	3 (8)	14 (30)	.016
Event-free survival	5 (36)	3 (100)	.683	1 (33)	5 (36)	.222

Abbreviations: D/R, donor/recipient; F/F, female/female; F/M, female/male; HSCT, hematopoietic stem cell transplantation; M/F, male/female; M/M, male/male.

[a]Transplant outcomes at last follow-up.

[b]Death within the first 3 months from HSCT.

causes, providing a TRM of 15%. We found no statistically significant differences between males and females in DRM and TRM ($P > .05$). The OS was 72%, including 82 males (47%) and 44 females (25%). One hundred and eleven patients (64%) showed a disease-free survival (DFS), including 72 males (41%) and 39 females (22%). We found no statistically significant differences in OS between males and females after HSCT (Figure 3A), not even when patients were divided into two age groups that might reflect hormonal maturation (<12 years, >12 years; Figure 3B). The CRI was 21%, with 25 males (14%) and 11 females (6%) having or dying from primary disease relapse (Figure 3C).

Consistent with what we expected, the χ^2 test showed that patients who had the major risk at the outset also had a major number of relapses ($P < .00001$). Additional data about event-free survival (EFS), early posttransplant death, the need for a further HSCT and its outcomes are displayed in Table 1. In addition, we analyzed differences in OS, EFS, early death, TRM, DRM and the need for a further HSCT by comparing donor-recipient pairs matched and mismatched for sex (Table 2) and by comparing two age groups (<12 years, >12 years; Table 3).

3.2 | HSCT-related complications

Data related to mucositis, gastrointestinal, hepatic, lung and kidney toxicity and GvHD were collected, along with the incidence of infection events and the engraftment of neutrophils and platelets. There are no statistically significant differences ($P > .05$) in the incidence of acute and chronic GvHD in males and females (Figure 3D). Posttransplant complications found in both sexes and assessed for different degrees of severity, together with engraftment data, are displayed in Table 1. We found no statistically significant differences between the sexes ($P > .05$). In addition, we found no statistically significant differences for HSCT-related complications between donor-recipient pairs matched and mismatched for sex (Table 2). Similarly, no statistically significant differences in HSCT complications were found between males and females when comparing two age groups (<12 years, >12 years) (Table 3).

4 | DISCUSSION

Our study shows no statistically significant sex differences in OS, CRI, HSCT-related complications and TRM at 12 months. The higher disease risk in male patients and their increased need for advanced-line therapy are consistent with the "male disadvantage" reported in the literature.[8] Considering the major male incidence and severity of malignancy, which more often require a transplant,[7] we would expect this sex difference to be maintained, ending in a higher number of disease relapses among males. In reality, it is not what happens, suggesting that something acting during the HSCT process may flatten such male disadvantage at entry.

TABLE 3 Differences in HSCT outcomes and transplant-related complications between males and females divided in two age groups

Variables	Male 0-12 years (n = 74)	Female 0-12 years (n = 37)	*P*-value	Male 12-18 years (n = 37)	Female 12-18 years (n = 26)	*P*-value
Transplant-related toxicity, number (%)						
Mucosal I-II grade	53 (71)	23 (62)	.390	25 (68)	16 (62)	.435
Mucosal III-IV grade	17 (23)	10 (27)	.643	11 (30)	10 (38)	.435
Gastrointestinal I-II grade	56 (76)	26 (70)	.647	25 (68)	22 (85)	.232
Gastrointestinal III-IV grade	18 (24)	9 (24)	>.999	10 (27)	4 (15)	.359
Hepatic I-II grade	24 (32)	14 (38)	.669	16 (43)	13 (50)	.803
Hepatic III-IV grade	12 (16)	7 (19)	.787	7 (19)	4 (15)	.745
Pulmonary	6 (8)	2 (5)	.725	2 (5)	1 (4)	>.999
Renal	10 (13)	4 (11)	.771	3 (8)	5 (19)	.272
Transplant outcomes[a], number (%)						
Overall survival	56 (76)	29 (78)	.828	25 (68)	16 (62)	.435
Event-free survival	36 (49)	27 (73)	.021	21 (57)	13 (50)	.454
Early death[b]	11 (15)	4 (11)	.765	4 (11)	5 (19)	.480
Transplant-related mortality	8 (11)	7 (19)	.249	6 (16)	6 (23)	.748
Relapse-related mortality	10 (13)	1 (3)	.103	5 (14)	5 (19)	.733
Successive HSCT, number (%)	22 (29)	3 (8)	.009	6 (16)	3 (12)	.721
Event-free survival	7 (32)	2 (67)	.714	3 (50)	2 (67)	>.999

Abbreviation: HSCT, hematopoietic stem cell transplantation.
[a]Transplant outcomes at last follow-up.
[b]Death within the first 3 months from HSCT.

Since all patients undergoing HSCT received a myeloablative conditioning regimen, known to provide a gonadal impairment,[13,14] we can hypothesize that the reduction of circulating levels of gonadal hormones somehow influences the severity and prognosis of hematological malignancies. There is evidence in the literature of the effects of sex hormones on cancer susceptibility, development and progression.[1,3,4,6,12] Sex hormones exert their action through three major sex steroid intracellular receptors—estrogen receptors (ERα and ERβ) and an androgen receptor (AR)—which not only can shuttle to the nucleus and affect gene expression but also act at an epigenetic level through DNA methylation and chromatin conformation. To some extent, sex differences in cancer mechanisms could be attributed to a simplified dichotomy, which includes sex chromosomes and sex hormones (Graphical abstract). These factors act through intrinsic and extrinsic mechanisms on cancer-initiating cell populations, affecting their self-renewal, the tumor microenvironment and the overall metabolic balance of the organism.[1]

ERα significantly reduces macrophages' proinflammatory activity and tumor-promoting properties, such as the secretion of interleukin-6, a constituent of the tumor microenvironment. In endothelial cells, AR can directly associate with and stimulate vascular endothelial growth factor receptor-2 (VEGFR2) activity and promote proliferation and angiogenesis of the stromal compartment of tumors. In addition, several critical immune-related genes, such as FOXP3 and CD40L, are located on the X-chromosome,[4] along with toll-like receptor 7 (TLR7), a central actor of innate immunity expressed at a higher level in females than males.[6] Also, numerous genes expressed in T cells carry the estrogen response element in their promoters, leading to more robust inflammatory and cytotoxic T cell responses in females.[4] On the other hand, testosterone has been shown to promote neutrophil production and restrain T cell proliferation while increasing the number of regulatory T cells.[1,6] This is relevant for cancer, as a higher neutrophil/lymphocyte ratio is considered a poor prognosis biomarker in several tumors.[1] In the same way, the disruption of ERβ in mice promotes the hyperplastic expansion of leukocytes resembling chronic myeloid leukemia.[1] The evidence for the protective role of estrogens against the formation of tumors in nonreproductive tissues and sex differences in immune response should be carefully considered when using cancer immunotherapy.[24,25] Epigenetic alterations mediated by sex chromosome genes is another possible mechanism for gender difference in autosomal gene expression, as shown, for example, by the gender-specific locus of the interferon regulatory factor-4 (IRF4), which has been associated with childhood ALL in males only.[6] Estrogens have an inhibitory effect on the transcription factor NF-kB, which regulates IRF4 transcription, and males lack this effect.[6] Last but not least, several X-linked miRNAs may contribute to the sex-specific regulation of immune-related genes. For example, some X-linked miRNAs overexpressed in T cells of female patients are located within 5000 base pairs of an estrogen response element whose presumed target is the E3-ubiquitin ligase CBL. This myelomonocytic leukemia proto-oncogene is a negative regulator of T cell receptor activity.[1] Circulating gonadal hormone levels are influenced by patients' age, particularly depending on patients'

pubertal development. The hypothalamic-pituitary-gonadal (HPG) axis starts acting in the midgestational fetus, is silenced towards term by the negative feedback of placental hormones and then reactivates at birth when this restraint is removed. Gonadotropin levels then remain high during the first 3 months of life in both sexes. In boys, testosterone levels rise to a peak at 1 to 3 months and then decline along with falling luteinizing hormone (LH) levels towards 6 months. In girls, follicle-stimulating hormone (FSH) levels remain elevated until 3 to 4 years of age and result in the maturation of ovarian follicles and increase of estradiol levels. The HPG axis then remains silenced until puberty.[26] Because sex hormone levels are very low during childhood, and not much difference between males and females, this suggests that prenatal programming plays a role in the sex hormone effect on autosomal gene expression. Exposure to gonadal hormones already begins in utero during a critical window of fetal development, determining differences in sex-specific methylation patterns at epigenetic levels. Females show higher methylation of the promoter of the ERα gene. Testosterone is a regulator of DNA methylation, influencing histone acetylation levels of ERα and aromatase promoters.[4] Gonadal hormone-dependent and -independent effects on sexually dimorphic immune responses have been investigated in murine models,[6,27] but they have never been studied in vivo. Palaszynski et al showed that the XY-genotype is relatively immunostimulatory compared to the XX genotype, confirming that it is the male hormone phenotype that is immunoinhibitory.[27]

Our study is the first report exploring sex differences in pediatric outcomes after HSCT. Thus, our study population can be considered an experimental human model in which the hormonal factor is suppressed by the conditioning regimen, enabling the chromosomal factor to express its immunostimulatory action and thus protect against tumor relapse. Although the HSCT process does not reverse the earlier effects of the gonadal hormones, reducing circulating hormonal levels it provides seems sufficient to flatten the sex-based disparity in cancer outcomes. Notably, this difference has otherwise been shown to persist in males notwithstanding overall improvements in standard chemotherapy, including extending treatment duration and supplemental intrathecal therapy.[28]

Our study has some limitations. First, it is a single-center retrospective study with a relatively small sample size; also for the retrospective nature of the study, the selection bias cannot be ignored. Second, while hormonal levels vary depending on age, information concerning baseline and pretransplant sex hormones levels was unavailable for all patients. Furthermore, the impact of cytostatic drugs on fertility depends on the drug type and dosage and patients' age at the time of treatment.[14,29] The lack of data on patients' hormonal profiles for all stages of the therapeutic program may limit the strength of our conclusions. However, we analyzed outcomes tailored for prepubertal and pubertal age groups and did not find statistically significant differences. Finally, the relatively short follow-up period may have led to misconceptions, and the chosen outcome measures may be debatable. However, we believe that the cumulative relapse incidence can be considered a reasonably robust outcome measure to quantify the residual cancer risk burden after transplant.

Our study shows that the sex discrepancy in cancer severity observed at diagnosis and after first-line therapy is canceled after exposure to HSCT. Since the pretransplant myeloablative conditioning regimen induces gonadal impairment and consequent reduction in the circulating levels of sex hormones, we can speculate that androgens play a major role in cancer relapse, rather than a deficiency in the protective role of estrogens. Further studies involving a larger sample size are warranted to understand better the impact of sex hormone modulation on the outcomes of hematological malignancies.

CONFLICT OF INTEREST

The authors declare no conflicts of interest.

AUTHOR CONTRIBUTIONS

Laura De Nardi: writing—original draft preparation (lead); writing—review and editing (equal). Roberto Simeone: formal analysis (lead); data curation (equal). Lucio Torelli: formal analysis (supporting); writing—review and editing (supporting). Alessandra Maestro: methodology (lead). Davide Zanon: methodology (supporting); formal analysis (supporting); data curation (equal); visualization (supporting). Egidio Barbi: writing—review and editing (equal); supervision (equal). Natalia Maximova: Conceptualization (lead); writing—review and editing (equal); data curation (equal); visualization (lead); supervision (equal). The work reported in the article has been performed by the authors, unless clearly specified in the text.

DATA AVAILABILITY STATEMENT

The data that support the findings of our study are available from the corresponding author upon request.

ETHICS STATEMENT

The study was approved by the Institutional Review Board (IRB, RC 25/19) and has been retrospectively registered on ClinicalTrials.gov Identifier: NCT04580576. All parents of the patients gave written consent for the collection and use of personal data for research purposes.

ORCID

Laura De Nardi https://orcid.org/0000-0002-8141-3389
Egidio Barbi https://orcid.org/0000-0002-6343-846X
Natalia Maximova https://orcid.org/0000-0003-0934-1875

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