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Chapter 8

Optimization of Unrelated Donor Cord Blood Transplantation for Thalassemia: Implications for Other Non-Malignant Indications such as HIV Infection or Autoimmune Diseases

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Additional information is available at the end of the chapter

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Abstract

Since the first cord blood transplantation (CBT), many indications have been proven for this stem cell therapy. Besides the standard hematological indications, such as leukemia, lymphomas, and aplastic anemia, CBT has also been a proven curative therapy for non-hematological indications such as Krabbe's disease, and osteopetrosis. As transplant-related mortality (TRM), overall survival (OS) and disease-free survival (DFS) for CBT continue to improve with larger inventories, double CBT, higher cell dose CB products, optimal conditioning, GvHD, HLA matching, and infection prophylaxis and treatment, the utility of this stem cell source will expand to certain indications which in the past, rarely used CBT. For patients and physicians to accept CBT for indications such as thalassemia, autoimmune diseases or HIV, the benefit-risk ratio has to be significantly improved so that patients will take a chance on a risky procedure in order to improve their lifespan or quality of life. We review here some of the efforts to improve clinical outcome of CBT for thalassemia through increasing cell dosage using a combination strategy – (1) Chow's MaxCell second and third generation technologies that maximize CB cell dosage, (2) double CBT, (3) no-wash thaw direct infusion advocated by Chow et al., and (4) optimal product selection.

Keywords: unrelated donor cord blood transplantation, thalassemia cure, HIV cure, autoimmune diseases cure, cord blood banking, cord blood processing, cord blood

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transplant outcome, MaxCell cord blood products, MaxCB cord blood products, homozygous CCR5-Δ32 donor cord blood transplantation

1. Introduction

Like other forms of hematopoietic stem cell transplantation (HSCT), unrelated donor cord blood transplantation (CBT) is a lifesaving therapy capable of curing many diseases, including ~80 standard hematologic and certain nonhematologic indications, such as thalassemia major. In addition, HIV infection and ~80 autoimmune diseases may be curable with unrelated donor CBT as well [1]. Unlike adult donor bone marrow (BM) and peripheral blood (PB) HSCT that require ≥10/12 high-resolution HLA A/B/C/DP/DQ/DR matches, unrelated donor CBT has been performed safely with ≥4/6 HLA A/B/DR matches. One of the reasons for the reduced HLA matching requirement of CBT is the decreased incidence and severity of acute and chronic graft-versus-host disease (GvHD) following transplantation with cord blood (CB), even with mismatched donors [2]. As such, unrelated donor CBT lends itself to minority populations without large BM donor registries and disease indications prevalent in certain populations without many adult donors, such as patients with thalassemia that are prevalent in China, India, Southeast Asia and Middle East.

Though HSCT is currently the only cure for thalassemia, due to the scarcity of suitable related and unrelated HLA-matched adult donors for most of the affected patient population, HSCT for thalassemia has been underutilized. Due to its lowered requirement for HLA matching, CBT has been touted as an ideal donor stem cell source for the cure of thalassemia for these populations. Unfortunately, outcome from previous large series using CBT for thalassemia has been underwhelming [3]. As a result, worldwide use of CBT for thalassemia has been even less than that of adult donors. If patient survival can be improved when using CBT for thalassemia, then utilization rate of this alternative donor source would greatly increase, as we have seen from our experience in Taiwan where many of the pediatric thalassemia patients have been cured in our collaboration with Chang Gung Children's Hospital [4–7]. To improve clinical outcome for CBT for thalassemia in Taiwan, our focus has been to increase the stem cell, progenitor cell and total nucleated cell doses transplanted into patients by (1) raising the average cell dose of CB products in our inventory significantly through the use of Chow's proprietary MaxCell CB processing technologies [8]; (2) using double CBT whenever feasible and necessary [9]; (3) avoiding of the use of post-thaw wash either with direct thaw and infusion or with thaw and reconstitution [8]; and lastly, (4) selecting the best combination of optimally HLA-matched CB units that have high progenitor and total nucleated cell doses [6]. Currently, this strategy has been difficult to fully duplicate for other regions where thalassemia is prevalent as large inventories using the proprietary MaxCell CB processing technologies have been limited mostly to the Taiwanese patients for populations with high prevalence for thalassemia.

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These strategies to improve patient survival and clinical outcome of CBT may have an impact on other chronic nonlethal diseases, such as autoimmune diseases and HIV infections, that can be cured with HSCT, which require high survival-to-mortality ratios to raise utilization rates. Since 2001, Chow and his former colleagues at StemCyte have been working with City of Hope to identify and establish a homozygous CCR5-Δ32 donor CB inventory. These efforts preceded the first patient cured of HIV infection (the "Berlin patient") in 2009 with HSCT using an adult donor homozygous for the CCR5-Δ32 mutation. In 2001, Chow et al. patented the homozygous CCR5-Δ32 donor HSCT technology that was acknowledged as the basis for the HIV infection cure for the Berlin patient by Dr. Gero Hütter, the attending transplant physician for the patient [10]. In the last few years, the StemCyte CB bank founded by Chow has collaborated with other high-quality banks around the world to screen CB inventories from these banks to increase the number of homozygous CCR5-Δ32 CB products available for transplantation of HIV patients. Since the Berlin patient, we and others have been searching for suitable HIV patients to be transplanted using this multi-bank homozygous CCR5-Δ32 donor CB inventory. For HIV infection, the current bottlenecks are (1) the scarcity of HLA-matched CB donor products that are also homozygous for the CCR5-Δ32 mutation that has sufficient cell dose for transplantation, and the (2) lack of HLA typing information on potential HIV patients who are candidates for such transplantation. Nonetheless, HIV-infected patient requiring homozygous CCR5-Δ32 donors remains an ideal indication for unrelated donor CBT.

Lastly, the reduced incidence and severity of acute and chronic GvHD following unrelated donor CBT may also factor into its potential preferential consideration as a hematopoietic cell source for transplantation for autoimmune diseases (AD). This is because if GvHD and TRM can be reduced, then allogeneic HSCT may be used more frequently. Currently, autologous HSCT is most often used in the treatment of severe AD (SAD) due to the high early TRM and severe GvHD associated with allogeneic HSCT; however, the remission rate is not ideal with autologous HSCT.

2. Cure of thalassemia by hematopoietic stem cell transplantation

Thalassemia is a disorder characterized by the formation of abnormal hemoglobin and unequal globin chain synthesis. It is one of the most prevalent genetic disorders in the world. There are a global estimate of 270 million carriers of hemoglobin disorders, 80 million of them carrying β-thalassemia. β-Thalassemia is common in the Southern Asia and Southeast Asian regions (1–40%), especially China and India, Middle East (3%), Mediterranean (1–3%), and in malarial tropical regions due to the selective heterozygote advantage against malaria, thus increasing the frequency of β-thalassemia [11]. Current medical therapy consists of lifelong blood transfusions to maintain hemoglobin levels between 9 and 10 g/dL to suppress the ineffective anemia-causing erythropoiesis. Complications with hemosiderosis or iron overload as a result of frequent transfusions have been curbed with the addition of iron chelation therapy, which has doubled life expectancy [11]. Initial iron chelators were administered as a continuous subcutaneous infusion for 8–12 h daily; however, limitations such as inconvenience, side effects, prohibitive cost, pain and associated reduced compliance of parenteral administration

led to the development of oral iron chelators, which have been demonstrated to be safer and easier to be compliant, though still associated with certain side effects. Despite increasing life expectancy and improving quality of life for children with thalassemia, transfusion and chelation therapies have major pitfalls, stopping short of becoming a cure for thalassemia. Endemic areas where thalassemia is most prevalent struggle with the cost of iron chelation and the risks of hyper-transfusion causing blood-transmitted infections such as hepatitis B and C. Developed countries often encounter patient compliance issues, as effective daily chelation administration is often unpleasant and inconvenient. Even with modern transfusion and chelation therapy, only 68% of patients with β-thalassemia are alive at the age of 35 [12]. Although there are considerable advancements in transfusion and iron chelation, HSCT represents the only curative therapy for patients with β-thalassemia currently.

In 1982, the first successful marrow transplantation for thalassemia was performed on a child by Donnall Thomas and his colleagues [13]. Subsequently, the first of several series of transplants for thalassemia was reported by Lucarelli et al. [14–16]. Today, thousands of patients with thalassemia have been treated using HLA-identical sibling donor bone marrow transplantation (BMT). After decades of optimization by the Italian groups [14–16], over 1000 patients with thalassemia and sickle cell disease have been cured, mostly using HLA-identical sibling donor BM. For low-risk Pesaro class 1 or 2 patients, related BMT could achieve outstanding overall survival of 87–95% and thalassemia-free survival of 64–90%, depending on the disease severity [11]. Even with class 3 (with extensive liver damage from iron overload) patients, with certain new preparatory regimens, patients younger than 17 years can achieve survival rates of 93% with only 8% autologous recovery rate [17].

Unfortunately, <30% of adults have HLA-matched siblings, especially in China, where the one-child policy has hindered widespread use of related donor transplantation. Matched unrelated adult donors also remain unavailable for most thalassemia patients, despite proving to be acceptable alternatives for patients with thalassemia who lack a compatible family donor [18]. The lack of available matched unrelated adult donors is often due to the limited size or lack of bone marrow registries for the endemic regions. Despite there being 14 million potential unrelated adult donors registered in various international registries worldwide, current inventories of HLA-matched donors are especially limited for patients of Asian descent, a region where thalassemia is most prevalent, as the majority of the world's adult donor registries are from Caucasian background. With the expansion of donor registries by tens of millions in regions that are prevalent for thalassemia, the scarcity of donors may be alleviated; however, the endeavor of building BM registries of tens of millions donors is quite cost prohibitive, especially given that most of the countries in the endemic regions are developing economies.

Cord blood offers an alternative for the source of hematopoietic stem cells and is a faster and more economical way to increase the supply of donor stem cells. In fact, unrelated donor CBT may offer the best alternative to adult donor HSCT due to a more lenient requirement for HLA matching, allowing patients to find suitable donors from banks that are several orders of magnitude smaller than BM registries. The relaxed HLA matching requirement is a result of less severe GvHD after CBT compared to adult donor HSCT, which resulted in improved quality of life for patients due to the decreased requirement of GvHD prophylaxis. As such,

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related and unrelated CB may alleviate shortage of matched unrelated donors, since less stringent HLA matching is acceptable. Moreover, due to the lower severity and incidence of GvHD after CBT compared to BMT [19–22], CBT may be preferable to BMT for thalassemia and other nonmalignant diseases, as the decreased GvHD incidence and severity greatly improve quality of life for transplant patients. For thalassemia and other transplant patients with nonmalignant diseases, GvHD offers no advantage of relapse reduction as in the setting as for malignant diseases.

Unrelated CBT for treatment of thalassemia has improved significantly with judicious CB graft selection and consideration of a number of factors, including transplant age, Pesaro class, CB processing, cell dose and post-thaw processing. Working exclusively with CB produced by Chow's proprietary MaxCell technologies, Jaing et al. achieved thalassemia-free survival close to that of related CB transplantations [5, 6]; however, other studies using traditional red cell reduced (RCR) CB (referred to as 1st Gen CB processing elsewhere in this book) produced poor results and the authors advocated cautious use of CB only in clinical trials [3]. Transplant center experience is an important factor in increasing CBT for thalassemia. Although unrelated CBT has the potential for curing thalassemia, thereby drastically improving the quality of life for the patient, it is still not considered optimal even with the industry's best practice due to CB donor scarcity in thalassemia endemic areas, especially for MaxCell CB products, and lack of optimal results for RCR CB. Even so, as blood transfusion and iron chelation therapies are prohibitively expensive and not widely accessible in thalassemia endemic regions, unrelated CBT becomes a viable alternative that is less costly in the long run. In addition, it should be noted that unrelated CBT offers patients significant potential benefits—quality of life and increased life expectancy—thus increasing the need for the optimization of CBT to better treat patients.

Recently, a number of studies have shown significant success using related and unrelated donor CBT. As expected, cell dose is the most critical factor for CBT success, as revealed by almost every major study to date [19–22]. Theoretically cell dose may be less of a problem for thalassemia since CBT is usually performed at an early age when patients have smaller body mass and require less cell dose; however, due to the difficulties of eradicating the endogenous erythron, cell dose has been found to be just as critical for both related and unrelated CBT for thalassemia [6, 7, 23–26]. For unrelated CBT for thalassemia, Jaing *et al*. [6] established institutional guidelines of 2.5×10^7/kg for single unit CBT and $>3.7 \times 10^7$/kg combined nucleated cell dose for double CBT, with at least one unit exceeding 2×10^7/kg. Moreover, at Chang Gung, following the Minnesota recommendations of CD34+ cell dose of 1.7×10^5/kg minimum for single unit CBT [27], with the combined CD34+ cell dose exceeding 3.0×10^5/kg for double CBT.

Due to the proven central importance of cell dose in CBT, different groups have employed various strategies to optimize nucleated and CD34+ cell doses, such as the supplementation of BM stem cells from the same donor to the CB graft [28–30], the use of double CBT when single CB units do not have sufficient cell doses [5, 6, 26, 31–35], the avoidance of post-thaw wash (when indicated), which invariably results in loss of cells [5, 6, 26, 32–50], and usage of MaxCell CB as red cell reduction results in decreased cell recovery [4–7, 26, 31–55].

Various other approaches have been tried to improve the outcome of CBT for thalassemia, ranging from preference for superior HLA matches [4, 6, 26, 31, 34, 37, 51–53], usage of related HLA-identical donors [23–25], directed sibling cord blood bank efforts [30, 56], consideration of non-inherited maternal antigen (NIMA) matches [57], preference for IV busulfan over oral formulations [6, 58], the addition of thiotepa to the conditioning regimen [24, 25], reduced intensity conditioning regimens [3, 59, 60], the avoidance of methotrexate in the prophylaxis regimen [23–25], third-party MSC co-infusion [59], and intrabone direct injection of cord blood products [61, 62].

3. Related donor cord blood transplantation for thalassemia

In 1995, the first CBT for thalassemia was reported, using a HLA-identical sibling donor cord blood for a two-and-half-year old [36]. Busulfan/cyclophosphamide conditioning regimen and cyclosporine/methotrexate GvHD prophylaxis were used. The TNC dose was 3.9×10^7/kg, and the thawed product was not washed and was directly infused on June 12, 1993. Neutrophil and platelet engraftment were achieved by day +23 and day +27, respectively. The patient experienced no GvHD, and the patient was alive and transfusion independent 48 months after transplantation. The various studies using related donor CBT for thalassemia are summarized in **Table 1** [23–25, 28–30, 36, 56, 59, 63–68].

	[36]	[64]	[28]	[65]	[23]	[29]	SDCB [30, 56]	[24]	[25]
# Patients	1	2	3 CB 5 CB + BM	1	33	9; 2 + baby donor's PB	(1) 14 Thal 4/14 + PB (2) 7 Thal	27	66
Age (yrs) median Range	2.5	2.2/3.8	4, 13, 15	3	5 1–20	5.5/ 3.5–10	(1) NA 5.9/ 2–11	6 0.8–18	5.9/ 2–20
HLA A/B/DR 6/6 5/6 ≤4/6 matches	1	2	8	0 0 1	41 3	6 1 2	(1) 14 (2) 7	100% 0 0	100% 0/0 0/0
TNC dose Median Range	3.9	6.2/11.4	CB 2/1.2/2.5 Combined 3.2/2.8/3.7	6.1	5.1/ 1.2–13 PF 4.0/ 1.2–10 PT	6.6/ 3.4–12.7	(1) NA (2) 4.7/ 0.8–7.6	3.3 1.5–6	3.9 1.5–14
Engraftment Myeloid (ANC500) Platelet 20/50K	ANC 1/1; D+23 Plt	2/2	1/3CB 5/5 CB + BM	1/1	ANC 89% D+23 (12–60)	5/9	(1) Thal 12/14 (2) Thal 6/6 with engraftment	100% (27/27) 100% (27/27)	ANC 90 ± 4%; D +23 Plt

	[36]	[64]	[28]	[65]	[23]	[29]	SDCB [30, 56]	[24]	[25]
(Plt 20K or 50K)	1/1; D+37				D +39 (19–92)		data reporting		83 ± 5%; D +38
Graft failure; primary (PGF); secondary (SGF)	None	None	2/3 CB	None	7/33 (21%)	4/9; 2 PGF; 2 SGF	(1) 2 PGF + 1 SGF (2) 1SGF	0 (0%)	10.4%
Acute GvHD	0	2/2 III SR	None	None	11% (4/38) II	3/9 I/II SR 1/9 IV	(1) NA	0 (0%)	11% II–IV
Chronic GvHD	0	None	None	Limited	6% (2/36) limited	1/9	(1) NA	0 (0%)	5 ± 3%; 0 ext
Survival	OS 1/1 DFS 1/1	2/2 Alive & TI	OS 8/8 DFS 7/8	0/1 Died D +300	Thal OS 100% EFS 79%	OS 8/9; 1 death aGvHD IV 4 TI	(1) Thal OS 11/14; DFS 11/14	OS 27 (100%) DFS 27 (100%)	DFS 80 ± 5%
Follow-up (M) median/range	48 (1995b)	11–24	(1) 18/ 16–23	10	24/ 4–76	49/ 38–64	12.4/ 0.5–77	40/ 15–89	CB 70 (12–151)

Thal = thalassemia major; **CB** = cord blood; **CBT** = cord blood transplant; **MaxCell** = non-red blood cell reduced cord blood; **RCR** = red cell reduced cord blood; **SCBT** = single cord blood transplantation; **DCBT** = double cord blood transplantation; **NW** = non-wash post-thaw processing; **N/A** = not available; **LGF** = late graft failure; **AR** = autologous recovery; **TI** = transfusion independent; **TNC** = total nucleated cells in $\times 10^7$/kg patient weight; **CD34+** = total **CD34+** Cells in $\times 10^5$/kg patient weight; **GvHD** = graft-versus-host disease; a**GvHD** = acute graft-versus-host disease; **Ltd** = limited chronic GvHD; **Ext** = extensive chronic GvHD; **TRM** = transplant-related mortality; **M** = months; **3Y** = 3 Year; **1Y** = 1 Year; **D** = days post-transplant; **OS** = overall survival; **DFS** = disease-free survival; **EFS** = time interval from CBT to first event (death or autologous reconstitution or infusion of cryopreserved backup recipient hematopoietic stem cells); **RR** = relative risk; **F/U** = follow-up; **CI** = cumulative incidence; **KM** = Kaplan-Meier estimator survival. **SR** = Resolved with Steroids; **Thal** = Thalassemia.

Table 1. CBT using related donors and sibling-directed donor CB bank (SDCB) for patients with thalassemia.

In 2013, Locatelli et al. published their landmark study for hemoglobinopathies on the comparison of related HLA-identical HSCT with 66 thalassemia patients transplanted with CBT against 259 thalassemia patients transplanted with BMT (**Table 1**) [25]. The CBT cohort was younger (median age 6 versus 8 years; $p = 0.02$), had higher disease severity for the thalassemia patients (Pesaro 2–3 39 versus 44%; $p < 0.01$), and was transplanted more recently (median year 2001 versus 1999; $p < 0.01$), with a significantly higher percentage of BMT patients receiving methotrexate GvHD prophylaxis than CB product recipients. No patients were excluded except for patients who received a combination of CB and BM products. Most thawed CB products were thawed and washed per Rubinstein procedure [20], and no information was provided as to the type of CB processing employed for the units. Compared to BMT recipients, the patients given CBT had slower neutrophil engraftment, less acute GvHD and no extensive chronic GvHD. Graft failure occurred more commonly in CBT patients than recipients of BM

grafts but not significantly (10.4 vs. 7.4%; p = 0.33). Eight of the patients who received CB graft experienced graft failure. Cumulative incidence of primary graft failure was 9 ± 4% and 6 ± 4% after CBT and BMT, respectively. Six patients experienced secondary graft failure after CBT at a median of day +151 (range day +51 to 202). The cumulative incidence of neutrophil engraftment was 90 ± 4% and 92 ± 1% (p = 0.01), and 83 ± 5% and 85 ± 5% for platelet engraftment after CBT and BMT, respectively. For patients who engrafted, the median time to neutrophil recovery was day +23 for CBT and day +19 for BMT, and day +38 and day +25 for CBT and BMT for platelet engraftment, respectively (p = 0.004). The proportion of long-term sustained mixed chimerism was significantly higher after CBT than for BMT (37 versus 22%; p = 0.01). Only 11% of CBT recipients experienced grade II–IV acute GvHD (no grade IV), versus 21% of BMT recipients (2% grade IV acute GvHD), with a cumulative incidence 10 ± 3% and 21 ± 2%, respectively (p = 0.04). Only six of 84 evaluable CB recipients experienced chronic GvHD with no extensive grade versus 42 of 355 (12 extensive) patients of BMT who survived past 100 days, with the cumulative incidence of chronic GvHD at 5 ± 3% and 12 ± 2%, respectively (p = 0.12), and extensive chronic GvHD at 0% and 5 ± 9%, respectively. Twenty-one patients expired from transplant-related causes—three after CBT and 18 after BMT. Most importantly, with a median follow-up of 70 months, the 6-year DFS was 80 and 86% for CB- and BM-transplanted patients, respectively, with no difference in multivariate analysis. This study proved that CBT using related donor for thalassemia is as efficacious and safe as related donor BMT, with potentially better long-term quality of life due to reduced chronic GvHD with minimal extensive grades. The authors point out that the quality of life for BMT patients with extensive chronic GvHD is worse than patients on medical therapy.

The author speculated that due to the higher nucleated cell dosage for most of the CB recipients, nucleated cell dosage was not shown to influence engraftment or disease-free survival. For CBT, methotrexate was shown in multivariate analysis negatively influencing DFS (HR 3.81, CI 1.40–10.87; p = 0.004), with 6-year DFS at 90 ± 4% if methotrexate was avoided versus 60 ± 11% (p < 0.001). Similar to previous studies, thiotepa-containing preparative regimen and Pesaro classification 1 were shown to correlate with better outcome after CBT. These series using related CBT demonstrate the efficacy and high margin of safety of related CB as a source of HSCT for thalassemia. The studies confirmed that even with persistent mixed chimerism, patients are still transfusion independent. Methotrexate GvHD prophylaxis was proven to be detrimental to favorable outcome and the addition of thiotepa to busulfan and cyclophosphamide conditioning regimen favored sustained donor engraftment.

4. Unrelated donor cord blood transplantation for thalassemia

Three single cases employing unrelated CBT for thalassemia were reported early on, with all three patients achieving neutrophil engraftment and transfusion independence [65–67]. Busulfan/cyclophosphamide/ATG-containing preparative regimen was used in all three; however, Fang et al. and Tan et al. used methotrexate-containing GvHD prophylaxis [65, 66]. Nucleated cell dose was high, with the minimum of 6×10^7 nucleated cells/kg. One patient

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received a 6/6 HLA A/B/DR-matched CB and two were transplanted with 4/6 HLA-matched CB. None of the patients experienced chronic GvHD or Grade IV acute GvHD. Vanichsetakul et al. [68] reported on six patients transplanted with three 6/6, one 5/6 and two 4/6 HLA-matched CB from unrelated donors. Patients were ranged from 2 to 15 years old with a median of 5.5 years. Busulfan, cyclophosphamide and fludarabine conditioning regimen was used with cyclosporine and methylprednisolone GvHD prophylaxis. Median TNC dose was 2.8×10^7 nucleated cells /kg with a range of $1.5–5.3 \times 10^7$ nucleated cells. Five of six patients engrafted and survived, while one expired due to infection prior to engraftment. Soni et al. [60] reported on unrelated CBT of two Pesaro class 3 patients with reduced intensity conditioning, with one recipient requiring re-transplantation with CB. After re-transplant, both patients engrafted and were thalassemia-free, with a follow-up of 7 and 8 years. Lastly, Kharbanda et al. [59] reported on the use of unrelated CB supplemented with co-infusion of third-party mesenchymal stromal cells (MSC) in two thalassemia patients, using a reduced intensity condition regimen. Only one patient engrafted, with neither patient survived.

In 2004, Tang-Her Jaing at Chang Gung Medical Center and Robert Chow at StemCyte embarked on a long-term collaborative study using unrelated CBT for thalassemia patients in Taiwan with the hypothesis that if conditions were optimized, HLA-mismatched unrelated CBT may produce results as favorable as unrelated BMT as well as approach that of related BMT and CBT. One strategy was to transplant patients as early as possible when disease stage is least severe. Most importantly, several approaches to optimize stem, progenitor and nucleated cell doses were employed: (1) utilization of CB products that were not reduced in RBC (MaxCell CB) whenever possible. Such non-RBC reduced, plasma depleted/reduced MaxCell ("MaxCell" or "MC") CB products have been shown to have significantly higher recovery for nucleated cell, CD34+ cells and colony-forming units (CFU) following parallel processing comparisons against RCR CB units [5, 8, 35, 39, 47, 48, 50]; (2) avoidance of post-thaw washing unless contraindicated, which has been shown by us and several other groups to be safe, offering enhanced infused cell dose due to zero cell loss from post-thaw washing [5, 6, 26, 32–34, 36–50, 69–71]; and (3) double CBT whenever cell dosage for single CB units was insufficient to meet the study thresholds for nucleated and CD34+ cell doses [9, 31–35]. In practice, the study complied with the first two conditions completely for all Chang Gung patients who have been reported, by sourcing all of its CB products from a single manufacturer of red cell-replete MaxCell CB products (StemCyte). Consequently, the average and median cell dosages achieved in Chang Gung patients were higher than every other large unrelated CBT series for thalassemia. The myeloablative conditioning and GvHD prophylaxis regimens consisted of the standard busulfan, cyclophosphamide and ATG, as well as cyclosporine and methylprednisolone, respectively. IV busulfan accompanied by drug level monitoring and adjustment replaced oral busulfan after a cluster of several autologous recoveries. Initially, pre-freeze nucleated and CD34+ cell dose criteria were set at 2×10^7/kg and 1.7×10^5/kg, respectively, and were later raised to 2.5×10^7/kg (for single CBT) and 2.0×10^5/kg, respectively. Importantly, the outcome data for Chang Gung transplant recipients were audited by CIBMTR appointed auditors on site using actual patient charts as routine for StemCyte supplied CB, which has been verified to be 97.3% accurate, with only minor errors and no errors for survival, mortality, engraftment, GvHD or relapse.

	MC CBT Jaing et al. [26]	**MC CBT Jaing et al. [6]**	**MC CBT Petz et al. [5]**	**RCR CBT Ruggeri et al. [3]**
# Patients	45 total; 32 Thal	35 Thal patients; 5 Re-CBT	120 total; 46 Thal	35 Thal
Type of CB	100% MC CB	100% MC CB	100% MC CB	100% RCR CB
Age (yrs) Median	4.5	5.5	3.5	4
Range	1.2–14	1.2–14	0.1–14	
Pesaro class 1/2/3	21/9/0*	N/A	N/A	9/2/4
N/A	2			20
HLA A/B/DR 6/6	11	8	26	5
5/6	25	16	48	14
≤4/6 matches	27	28	53	16
TNC dose median	7.6	7.8	10.5 pre-freeze	6
Range	2.8–15.0	2.8–14.7	7.7 infused	2–32
CD34+ cell dose	4.0	4.0	3.7	N/A
Median/range	1.3–19.9	1.7–19.9		
% CB not washed	100%	100%	58%	NA
DCBT	13 DCBT	10	15	0
Approaches to maximize cell dose	100% MC CB/ NW/±DCBT/CD34+ Priority	100% MC CB/ NW/±DCBT/CD34+ Priority	100% MC CB/ NW/±DCBT	NA
Engraftment CI				
Myeloid ANC500	ANC500 88%	ANC 88%	ANC 87 ± 6%	ANC500 42.8%
Platelet 20K (Plt20K)	Plt 20K 82%	Plt 20K 78%	Plt 20K 81 ± 6%	
Graft failure	4 Primary	5 Primary 1 Secondary	3 ± 2%	20/35 57.2%
Survival	**OS 5Y 88.1%**	**OS 5Y 88.3 ± 6.7%**	**OS 1Y 79 ± 4%; 3Y 79 ± 4%**	**OS 62 ± 9%**
	DFS 5Y 77.1%	**DFS 5Y 85.7%**	**DFS 1 Y 72 ± 5%; 3Y 70 ± 6%**	**DFS 21 ± 7%**
	2Y TRM 12%	**2Y TRM 11.7 ± 6.7%**	**TRM 100 D 10 ± 3%; 3Y 20 ± 4%**	
F/U (M) median/range	26/3–66	36/6–76	6.5	21/3–138
Acute GvHD	II–IV 76%	6 I; 12 II	0–II 38 ± 5%	23 ± 2%

	MC CBT	**MC CBT**	**MC CBT**	**RCR CBT**
	Jaing et al. [26]	**Jaing et al. [6]**	**Petz et al. [5]**	**Ruggeri et al. [3]**
	III–IV 42%	15 III; 1 IV	III–IV 19 ± 4%	
Chronic GvHD	35%	13/35 limited	36 ± 6% Ltd.	8/35 Ltd.
	1/14 extensive	1/35 extensive	12 ± 4% Ext.	2/35 Ext.
Severe infections	1	2/35		4/35

Thal = thalassemia major; **Other** = other nonmalignant indications; **CB** = cord blood; **CBT** = cord blood transplant; **MC** = non-red blood cell reduced cord blood MaxCell; **RCR** = red cell reduced cord blood; **SCBT** = single cord blood transplantation; **DCBT** = double cord blood transplantation; NW = non-wash post-thaw processing; **N/A** = not available; **GvHD** = graft-versus-host disease; **Ltd**. = limited chronic GvHD; **Ext** = extensive chronic GvHD; **TNC** = total nucleated cells in $\times 10^7$/kg patient weight; **CD34+** = total CD34+ cells in $\times 10^5$/kg patient weight; **OS** = overall survival; **DFS** = disease-free survival; **TRM** = transplant-related mortality; **M** = months; **Y** = year; **D** = days post-transplant; **F/U** = follow-up; **CI** = cumulative incidence; **KM** = Kaplan-Meier estimator survival; * = Modified Pesaro (no liver biopsy), which may underestimate the disease severity; @ = TC data audited by CIBMTR on site.

Table 2. Unrelated CB transplantation for patients with thalassemia—large series—3 MC CBT and 1 RCR CBT.

From 2005 onwards, the Jaing-Chow collaboration reported their experience of a number of studies using unrelated CBT for thalassemia (**Tables 2** and **3**)—both Chang Gung Children Hospital single institution experiences [4, 6, 26, 31–35, 37, 51–53, 58] and multi-institutional studies from the StemCyte cord blood bank outcome database [5, 7, 35, 39, 42, 49, 54, 55]. The first thalassemia patient transplanted by Jaing's group on October 2003 became the first disease-free surviving CBT recipient in Taiwan [37].

A single institution series of unrelated CBT of 45 patients with nonmalignant diseases (32 thalassemia cases) was reported by Jaing's group in 2010 (**Table 2**). Most patients received HLA-mismatched CB grafts with median infused nucleated and CD34+ cell doses at 7.6×10^7/kg and 4.0×10^5/kg, respectively [26]. With cumulative incidence of neutrophil and platelet engraftment at 88 and 82%, four patients experienced primary graft failure. Three patients experienced grade IV acute GvHD and only a single patient suffered extensive chronic GvHD. Five-year OS and DFS were 88.1 and 77.1%, respectively, and TRM was 12% at 2 years.

To study the effect of RBC-replete MaxCell CB in a series of 58 thalassemia patients performed at nine U.S. and five non-U.S. transplant centers, Chow's group [33] compared 48 patients who received MaxCell CB versus 10 patients who received RCR CB (**Table 3**). Though this initial study was not rigorously matched, patients were similar among two groups in age, weight, disease severity, TNC dose, #HLA matches, conditioning regimen, no post-thaw wash, and transplant center experience. There were more double CBTs in the MaxCell group (23 versus 10%). The raw comparison results between the two groups showed no significant differences in cumulative incidence in neutrophil (MaxCell 96 ± 4% vs. RCR 75 ± 15%; RR = 1.31; p = 0.56) or platelet 50K engraftment (MaxCell 95 ± 5% vs. RCR 75 ± 15%; RR = 1.24; p = 0.64). Overall patient survival at 1-year trended higher for MaxCell CBT (MaxCell 89 ± 6% vs. RCR 53 ± 20%; RR = 0.32; p = 0.17); however, importantly, DFS was significantly higher at 89 ± 6% for MaxCell CB compared to 38 ± 17% (RR = 0.17; p = 0.01).

	Jaing et al. 2008 matched pair (MP) MC CB ARM @ [32–34]	**Jaing et al. 2008 matched pair (MP) RCR CB ARM @ [32–34]**	**Chow et al. 2012 MC CB ARM @ [35]**	**Chow et al. 2012 RCR CB ARM [35]**
# Patients Type of CB	58 Thal patients 48 MC patients MP performed on 30 matched pairs 30 MC patients vs. 10 RCR patients	58 Thal patients 10 RCR patients MP performed on 30 matched pairs 30 MC patients vs. 10 RCR patients	91 Thal. 79 MC CBT	91 Thal. 12 RCR CBT
Age (yrs) median	Pre-match 5.0 post-MP 4.0	Pre-match 2.8 post-MP 2.8	MC 5.3	RCR 4.0
Range	Pre-match 0.3–20 post-MP 0.3–12	Pre-match 1–12 post-MP 1–12	0.3–2	0.8–12
Pesaro Class 1	Pre-match 46% MP 57%	Pre-match 50% MP 50%	MC 27	RCR 6
Class 2	Pre	Pre-match 20% MP 20%	MC 17	RCR 2
Class 3	Pre	Pre-match 10% MP 10%	MC 0	RCR 2
N/A			MC 35	RCR 2
Conditioning	Multi-inst. Series	Multi-inst. Series	Multi-inst. Series	Multi-inst. series
Regimens	Mostly BU/CY/ATG	Mostly BU/CY/ATG	Mostly BU/CY/ATG	Mostly BU/CY/ATG
GvHD Prophylaxis	CSA MP	CSA MP	Multi-institution series	Multi-institution series
HLA A/B/DR 6/6	Pre-match median 4.8 MP 4.5	Pre-match median 4.4 MP 4.4	MC 21	RCR 1
5/6	Pre-match range 3–6 MP 3–6	Pre-match range 4–6 MP 4–6	MC 38	RCR 3
≤4/6 matches			MC 45	RCR 8
TNC dose median	Pre-match 9.1 MP 9.1	Pre-match 8.9 MP 8.9	MC 9.8	RCR 8.7
Range	Pre-match 2.5–47 MP 3.4–20	Pre-match 2.3–19 MP 2.3–19	2–23.7	2–18.6
CD34+ cell dose median/range	N/A		MC 3.6 0.4–10.3	RCR1.5 0.4–13.5
% CB not washed	N/A		MC 89%;	RCR 77%
% DCBT	Pre-match 11 (23%) MP 5 (17%)	Pre-match 1 (10%) MP 1 (10%)	MC 20%	RCR 8%
Engraftment Myeloid ANC500 Platelet 20K/50K (Plt20K or 50K)	Pre-Match ANC MC 96 ± 4% vs. RCR 75 ± 15% (RR = 1.31; p = 0.56) Pre-Match Plt50K MC 95 ± 5% vs. RCR 75 ± 15% (RR = 1.24; p = 0.64) MP ANC MC 88 ± 12% vs. RCR80 ± 14% (RR = 1.05; p = 0.90; P = 0.92) MP Plt50K MC 88 ± 12% vs. RCR 70 ± 17% (RR = 1.01; p = 0.98; P = 0.73)		**ANC 83.2% Plt20/50K 79/76%**	
Graft failure	**MP 1Y MC 7 ± 5%; RCR 22 ± 14% (RR = 0.31; p = 0.24; P = 0.04)**		N/A	

	Jaing et al. 2008 matched pair (MP) MC CB ARM @ [32–34]	**Jaing et al. 2008 matched pair (MP) RCR CB ARM @ [32–34]**	**Chow et al. 2012 MC CB ARM @ [35]**	**Chow et al. 2012 RCR CB ARM [35]**
Survival	Pre-matched pair OS MC 89 ± 6% vs. RCR 53 ± 20% (RR = 0.32; p = 0.17) **DFS MC 89 ± 6% vs.** **RCR 38 ± 17%** **(RR = 0.17; p = 0.01)** **Matched pair comparisons (MP)** **3Y OS MC 96 ± 4% vs. RCR 53 ± 18%** **(RR = 0.09; p = 0.03; P = 0.001)** **3Y DFS MC 89 ± 6% vs. RCR 40 ± 15%** **(RR = 0.17; p = 0.01;P = 0.0001)** **3Y TRM MC 4 ± 4% vs. RCR 47 ± 18%** **(RR = 0.09; p = 0.03;** **P = 0.001)**		**MC DFS 61.4%**	**RCR DFS 50.0%**
F/U (M) median/ range	9.5	6	23.7	
Acute GvHD II–IV/ III–IV	N/**A**		MC 75%/25%	RCR 80%/20%
Chronic GvHD Ltd/Ext	N/A		MC 60%/5%	RCR 100%/0%

Thal = thalassemia major; **CB** = cord blood; **CBT** = cord blood transplant; **MC** = non-red blood cell reduced cord blood MaxCell; **RCR** = red cell reduced cord blood; **SCBT** = single cord blood transplantation; **DCBT** = double cord blood transplantation; **NW** = non-wash post-thaw processing; **N/A** = not available; **GvHD** = graft-versus-host disease; **Ltd** = limited chronic GvHD; **Ext** = extensive chronic GvHD; **TNC** = total nucleated cells in $\times 10^7$/kg patient weight; **CD34+** = total CD34+ Cells in $\times 10^5$/kg patient weight; **CI** = cumulative incidence; **KM** = Kaplan-Meier estimator survival; **OS** = overall survival; **DFS** = disease-free survival; **TRM** = transplant-related mortality; **M** = months; **Y** = year; **D** = days post-transplant; RR = relative risk; p = p value of MC versus RCR matched pair comparison; **P** = paired Prentice-Wilcoxon test p value; **F/U** = follow-up; * = modified Pesaro (no liver biopsy), which may underestimate the disease severity; @ = TC data audited by CIBMTR on site; **MP** = MC versus RCR CBT-matched pair analysis used a logistic regression model to find patients with similar characteristics to form 30 pairs, with three MC patients matched to each RCR patient (30 MC patients to 10 RCR patients). Factors matched for were age, weight, #HLA matches, TNC dose, transplant center experience. Univariate comparisons and paired Prentice-Wilcoxon test were performed for the matched pairs.

Table 3. Unrelated CB transplantation for thalassemia patients—comparison of MaxCell (MC) versus red cell-reduced CBT.

To further minimize patient population differences and selection bias, Chow's group performed a rigorous matched pair (MP) comparison analysis using a logistic regression model to find thalassemia patients with similar characteristics to form 30 pairs (**Table 3**), with three MaxCell patients matched to each of the available 10 RCR patients (30 MaxCell CBT patients matched to 10 RCR CBT patients) [32–33]. Factors matched were age, weight, TNC Dose, #HLA matches and transplant center experience. Since all available RCR patients were used in the MP study, and the best-matched MaxCell patients were used in the matched pair, differences in the matched factors were further minimized and selection biases were avoided. After matched pairing, age, disease severity, # HLA matches, TNC dose and usage of double CBT were quite similar between the two groups, with no significant differences. Univariate comparisons and paired Prentice-Wilcoxon test (PPW) were performed for the matched pairs.

The matched pair study results in **Table 3** showed that though engraftment did not improve significantly, autologous recovery rate was significantly lower in the MaxCell group at 7 ± 5% versus 22 ± 14% (RR = 0.31; p = 0.04). Most importantly, 3-year OS (96 ± 4% versus 53 ± 18%; RR = 0.09; p = 0.03 univariate and p = 0.001 PPW), 3-year DFS (89 ± 6% versus 40 ± 15%; RR = 0.17; p = 0.01 univariate and p = 0.0001 PPW), and TRM (4 ± 4% versus 47 ± 18%; RR = 0.09; p = 0.03 univariate and p = 0.001 PPW) were all significantly improved for the MaxCell CBT group. It should be noted again that the RCR and MaxCell OS and DFS rates observed in this study were similar to that reported for previously thalassemia series using unrelated CBT [3, 5–7, 26, 35]. Lastly, the outcome data for MaxCell CBT recipients were rigorously audited by CIBMTR personnel on site at transplant centers using actual patient records and verified to be 97.3% accurate, with no errors for major variables, such as engraftment, survival, mortality, autologous recovery, etc.

In 2012, Chow's group reported a multicenter series of 91 thalassemia patients transplanted with unrelated CB between 1999 and 2011, 79 with MaxCell CB and 12 with RCR CB (**Table 3**), the largest series to date of unrelated CBT for thalassemia [35]. With 45% male recipients, patient median age was 5.6 years (range 0.3–20) and median weight was 18.8 kg (range 4–80). HLA matches were 23 cases at 6/6, 45 cases at 5/6, 54 cases at 4/6 and 3 cases at 3/6 HLA A/B/DR matches. Median pre-freeze TNC dose was 9.4×10^7/kg and median pre-freeze CD34+ dose was at 3.2×10^5/kg—unusually high due to the exclusive usage of MaxCell CB products and the patients' young age in the study. Three-quarters of the patients were Asian and 84% CB was infused directly without post-thaw wash/reconstitution. Seven patients received a second CBT due to graft failure. Acute GvHD grade II–IV and III–IV occurred in 76 and 24% of the patients, respectively, whereas 60% and only 5% of the patients exhibited limited and extensive chronic GvHD, respectively. Overall, cumulative incidence of myeloid, platelet 20 and 50K engraftment of 83, 79 and 76% were achieved, respectively. Median times to myeloid and platelet 20K engraftment were 17 and 47 days, respectively. Most importantly, 180-day OS, 1-year OS and 1-year DFS of 80, 74 and 61% were reported, respectively, with a median follow-up of 711 days. Again, it is important to reemphasize that the outcome data for MaxCell CBT recipients were audited by CIBMTR on site at transplant centers using actual patient records and verified to be error-free for major clinical outcome measurements.

In their 2011 multi-institutional study, combined data of Eurocord, NYBC and CIBMTR, Ruggeri et al. [3] reported on 35 thalassemia patients (**Table 2**). With median age of 4 years, and mostly Pesaro class 1 and 2 patients (11 out of 15), this study used a variety of conditioning regimens (30/35 myeloablative regimens) and mostly calcineurin inhibitor containing GvHD prophylaxis (65% cyclosporine). Myeloid engraftment was only 42.8%, with 57.2% patients suffering primary graft failure. Though 66% were alive, only 23% were alive and thalassemia-free, with OS and DFS at 2-year post-transplant as estimated by Kaplan-Meier method as 62 ± 9% and 21 ± 7%, respectively (**Table 2**). Referring to the Jaing et al. series [26], the authors concluded that "For UCB, only one group has reported 80% DFS at 2 years… DFS was not as good as previously reported by Jaing and colleagues." Besides the single-center versus multi-institutional aspects of the two groups, the most obvious differences were the significantly higher TNC dosage in the Jaing/Chow series [4, 6, 26, 31–33], caused by the exclusive utilization

of RCR CB units in the Ruggeri et al. series [3] versus 100% MaxCell CB products used in the Jaing/Chow series. In a similar multi-institutional setting, using exclusively MaxCell CB products with 38% thalassemia patients, Petz et al. [5] reported 3-year OS at 79 ± 4% and DFS at 70 ± 6% (**Table 3**). As discussed above, Chow et al. [35] showed that in 91 thalassemia patients, employing MaxCell CB in 86.8% of the recipients, higher OS (73%) and DFS (61%) were also achieved compared to patients from the Ruggeri et al. series who received RCR CB units exclusively (**Table 3**).

In 2012, using exclusively unrelated MaxCell CB, Jaing's group transplanted 35 thalassemia patients, with 12 patients receiving double CBT (**Table 2**) [6]. The authors explained that their initial approach of using oral busulfan resulted in five primary and one secondary graft failures due to high pharmacokinetic variability exhibited with oral busulfan, necessitating six re-transplants. In this series, 88% of the transplants (35 first and 5 second transplants combined) achieved neutrophil engraftment. Five-year OS was 88.3 ± 6.7% and 5-year DFS was 85.7% (30 of 35 patients) in this exclusive MaxCell CBT series. The 85.7% 5-year DFS of this experience is in contrast to the 21 ± 7% 2-year DFS obtained by Ruggeri et al. [3] and compares very favorably with the 80 ± 5% 6-year DFS for HLA-identical related CBT reported for Locatelli series in 2013 [25].

5. Cure of nonmalignant diseases by cord blood transplantation

In **Table 2**, to assess whether this observation of the superior efficacy of MaxCell CB can be extended to other nonmalignant diseases, Chow's group conducted a multicenter study of transplantation of 120 patients who received MaxCell CB for nonmalignant diseases—46 for thalassemia, 3 for sickle cell disease and 71 other nonmalignant indications [5]. To maximize cell dose, besides using exclusively MaxCell CB, double CBT was used in 12% of the cases and no-wash thaw procedure was used in 58% of the patients. Median TNC dose was 10.5×10^7/kg at collection and 7.7×10^7/kg on infusion, and median CD34+ cell dose was 3.7×10^5/kg at collection. Twenty-six, forty-eight and fifty-three patients were matched at zero, one and two or more mismatches at HLA A/B/DR loci. Myeloid ANC500 and platelet 20K engraftment occurred at a median of days +21 and +49, respectively. The cumulative incidences to myeloid and platelet 20K engraftment were 87 ± 6% and 81 ± 6%, respectively. Autologous recovery occurred in only 3 ± 2% of the patients in this population made up of 38% thalassemics. Importantly, OS at 3-year was 79 ± 4%, whereas 3-year DFS was 70 ± 6%, respectively, with 100 days and 3-year TRM at 10 ± 3% and 20 ± 4%, respectively. Within the statistical power of this series, univariate analysis showed that ABO match, recipient sex, age, myeloablative conditioning regimen and CMV seropositivity were nonsignificant predictors of particular outcome. Double CBT was associated with a significantly higher incidence of acute GvHD grades II–IV (relative risk 2.23; $p = 0.05$). Higher pre-freeze CD34+ dose improved myeloid (RR = 1.55; $p = 0.05$) and platelet (RR = 2.73; $p = 0.05$) engraftment, OS (RR of death = 0.30; $p = 0.05$) and DFS (RR of death or relapse = 0.27; $p = 0.02$). In this study, TNC was not a significant factor, unlike previous reports [3], probably because the usage of typical TNC dose thresholds of 2.5 or 4.0×10^7/kg for analysis as in the Ruggeri et al. series was not applicable in this series,

due to the low numbers of MaxCell CBT patients with such low TNC doses, making statistical comparisons impossible. This anomaly is due to the significantly higher median and average TNC doses afforded by the usage of MaxCell CB products.

By using these three simple strategies to improve infused cell dose—exclusive MaxCell CB usage, not washing cord blood upon thawing (58%) and double CBT (12%) in this series at 46 U.S. and international centers, with divergent nonmalignant diseases, conditioning and GvHD prophylaxis regimens—results consistently superior to other reported series using unrelated RCR CB were obtained as shown in **Table 2** [3, 68]. In fact, these results approached that of Jaing et al. [6] in 35 thalassemia patients in a controlled environment at a single institution and also transplanted exclusively with unrelated donor MaxCell CB that were 100% directly infused upon thawing, proving that the adoption of this combination approach may be efficacious in diverse nonmalignant settings, such as HIV infection and autoimmune diseases.

6. Cure of HIV infection by cord blood transplantation

The application of the highly active antiretroviral therapy (HAART) has significantly improved the survival of HIV-infected patient and converted HIV infection into a chronic but mostly nonlethal disease for those patients who can afford and tolerate HAART in developed countries. Though significantly improving HIV treatment and patient survival, HAART alone is not sufficient to remove the virus in the long term, with rebound expected without continuous HAART treatment due to the long half-life of latent infected cells [72]. HAART cannot cure patients of HIV infection as clinically undetectable plasma viremia may only be achieved by life-long treatment with serious side effects and risks of viral rebound whenever treatment is interrupted. The reservoir of latent HIV provirus persists in patients' latent infected CD4+ T cells even with continued HAART and remains the major obstacle in achieving functional cure for HIV despite the countless efforts to eliminate it. Compared with HIV-negative people, HIV-infected patients are more prone to hematological malignancies including Hodgkin disease (HD) and non-Hodgkin lymphoma (NHL) [73]. Hence, to optimize the life expectancy and quality of life, and to reduce the economic burden of patients, actual cure of HIV is always preferable. Abbreviated life expectancy, high costs, serious chronic side effects and patient noncompliance of HAART therapy drives the search for a HIV cure. Due to existing techniques in leukemia treatment, HSCT has been investigated as a favorable approach to eliminate the HIV virus reservoir while also curing concomitant malignant diseases in the same patient.

The first clinical attempt to use allogeneic HSCT for cure of HIV infection was performed by Hassett et al. [74]. The patient did not improve clinically, and the immunological status remained stable or worsened. Retrospective analysis of reported cases by Hütter and Zaia [75] showed negligible differences between HIV-positive and HIV-negative patients following allogeneic HSCT. Such results are to be expected as HIV produced by the latently infected host cells that survive the allogeneic HSCT re-infects the newly engrafted donor immune system. HAART administration during and following allogeneic HSCT did not change the clinical course, and HIV in all eight patients in these seven reports who discontinued HAART after

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HSCT rebounded in just a couple of days [76]. Of these, two patients from the Brigham & Women's Hospital were reported initially to be negative for HIV using the most sensitive techniques available following unrelated donor 10/12 HLA-matched HSCT. Unfortunately, after HAART suspension, the virus rebounded in both patients within a few weeks. Without the continued antiretroviral treatment, patients show viral rebound quickly and thus were not functionally cured of HIV. In the study by Henrich et al., two patients received allogeneic HSCT from wild-type chemokine receptor 5 (CCR5) donors and were both reported to be free of detectable virus [77]. However, when HAART treatment was interrupted, viral RNA and proviral DNA became positive again 12 and 32 weeks later [78]. Taken together, these attempts appear to indicate that allogeneic HSCT alone is insufficient to eradicate HIV-1 infection.

As CCR5 is a required co-receptor with CD4 for entry of HIV-1 CCR5-tropic strains, and the mutated form of CCR5 with a 32bp deletion (CCR5-Δ32 mutation) provides resistance to the CCR5-tropic strains of HIV-1 in people homozygous for such mutation [79], Chow reasoned that HSCT with CCR5-Δ32/Δ32 donor will confer such resistance to recipients after developing complete donor chimerism, and if such recipients happen to be HIV-infected and contain only or predominantly CCR5-tropic viruses, such patients may be cured of their HIV infection. In 2001, Chow et al. were the first to propose the use of CCR5-Δ32/Δ32 donor HSCT to cure HIV infection and went on to file a patent application on the concept [80]. In vitro, CD4 cells from people homozygous for CCR5-Δ32 are highly resistant to infection by CCR5-tropic HIV-1, the dominant strains in vivo [81]. Even HIV-infected patients with heterozygous CCR5-Δ32 mutation genotype appear to derive partial benefits in the form of slower progression to acquired immunodeficiency syndrome status [82, 83].

Chow's approach was subsequently validated by Hütter et al. several years later [84]. This HIV-infected patient, now known commonly as the "Berlin patient," became the first and only known case in which a HIV-infected patient was functionally cured of HIV infection and survived [85]. Dr. Hütter later acknowledged that the technology first disclosed by Chow et al. in their 2001 U.S. patent application *#09/998,832* [80] and subsequently in U.S. patent application *2003/0099621 AI* was the basis for the Berlin patient cure [10, 86–88], when he wrote "…in 2001, R. Chow, founder of StemCyte, Inc., applied for a patent to screen allogeneic stem cell donors for a beneficial gene with which to treat HIV infection (U.S. patent *2003/0099621* AI)." [88]. The Berlin patient was diagnosed with acute myeloid leukemia and HIV infection and received an initial HSCT from an adult donor homozygous for the CCR5-Δ32 mutation. The patient's leukemia subsequently relapsed, prompting a re-transplantation with graft from the same donor. To this date, since the first transplantation, the patient has been free of HIV-1 infection by viral RNA load or proviral DNA in peripheral blood, gut, liver and brain tissue samples, even though HAART was stopped during and after the first HSCT. The Berlin patient achieved complete chimerism with homozygous CCR5-Δ32 genotype in his peripheral blood monocytes after initial transplantation, and subsequently, after re-transplantation with the same donor after leukemia relapse. With the absence of antiretroviral treatment for more than 8 years, viral RNA or proviral DNA has been undetectable in various tissues even with the most sensitive techniques and has shown a complete clinical remission of HIV infection [76,

86]. Although the case of the Berlin patient sheds light on one approach of curing HIV infection, this result has not been easy to replicate on a large scale to date.

To expand on the success of the Berlin patient, Petz and Chow reasoned that the odds of finding a 10/12 HLA A/B/C/DR/DP/DQ-matched adult donor who is also homozygous for the CCR5-Δ32 genotype is several logs more difficult than the probability of a 4/12 HLA A/B/C/DR/DP/DQ (matched for HLA A/B/DR loci only)-matched donor CB with the CCR5-Δ32/Δ32 genotype [87, 89, 90]. Starting in 2001, in collaboration with John Zaia, Joseph Rosenthal, John Rossi and Stephen Forman of City of Hope, Chow and Petz started screening StemCyte's CB inventory for the homozygous CCR5-Δ32/Δ32 and heterozygous CCR5-Δ32 genotypes. As Hütter et al. commented in 2011 "…Chows' group built a database with over 10,000 cord blood units, genotyped for the CCR5-delta32 deletion [10]." By 2007, Chow, Petz and City of Hope investigators started to plan for a clinical trial with Yvonne Bryson, Ron Mitsuyasu, Mary Territo, Ted Moore and Tempe Chen from UCLA [90]. By 2009/2010, the CB inventory screening process was expanded to include additional CB banks, and probability of finding matched CB units at various HLA match level and cell doses was calculated with the assistance of the National Marrow Donor Program [87]. By 2013, 180 CCR5-Δ32/Δ32 CB units have been identified [89].

To demonstrate the feasibility of CCR5-Δ32/Δ32 CBT to prevent HIV infection in an in vitro model, Petz et al. reported a case when a HIV-negative adult with acute myelogenous leukemia received CCR5-Δ32/Δ32 CBT [87], who engrafted showing complete donor chimerism. In vitro HIV infectivity tests were performed using the patient's engrafted CCR5-Δ32/Δ32 peripheral blood mononuclear cells (PBMCs) challenged with both CCR5-tropic and CXCR4-tropic HIV laboratory strains (BAL and NL4-3). Both viral strains showed no replication activities when cultured with the patient's engrafted CCR5-Δ32/Δ32 PBMCs, but exhibited robust replication with PBMC from control wild-type (WT) CCR5 or heterozygote CCR5/CCR5-Δ32 individuals, thus demonstrating resistance to HIV-1 of CBT derived donor PBMCs. These in vitro results further support the feasibility of curing HIV by CCR5-Δ32/Δ32 CBT.

Table 4 highlights the eight cases of using homozygous CCR5-Δ32/Δ32 donor HSCT performed up to 2014, with five using adult donors and three using CB plus or minus a second graft. Except for the two patients performed in Berlin (the Berlin patient and one patient with yet unpublished outcome), all have expired [91]. In the three adult donor cases performed outside of Berlin, one died from infection after 4 months and one from pneumonia shortly post-transplantation. In the last case, CXCR4-tropic virus rebounded after transplantation [92] and the patient expired from Non-Hodgkin's Lymphoma relapse after 12 months. Prior to transplantation, the patient appeared to have mixtures of CCR5-tropic and CXCR4-tropic viruses, though HAART therapy prior to transplantation caused a shift to predominantly CCR5-tropic viruses. CXCR4-tropic viruses emerged as the dominant viral type after HSCT probably as a result of the selection in the absence of functional CCR5 receptor. Though rebound occurred with HIV with alternative tropism, this case confirms the effectiveness of the CCR5-Δ32/Δ32 blockade of CCR5-tropic virus cell entry. Moreover, the emergence of viruses using alternative co-receptors does not negate the possibility of a sterilization cure using this approach of

patients infected predominantly with CCR5-tropic HIV, since it was the only case out of seven with known outcome where viral tropism switch occurred.

Transplant center	Adult donor graft cases	Patient age	Concomitant indication	Clinical outcome after transplantation
Berlin, Germany 2009	HLA-matched unrelated	40	AML	Alive after 8 years; no viral rebound without ART [10, 75–76, 84–91]
Munster, Germany	HLA-mismatched unrelated	51	NHL	Died from infection after 4 months [91]
Essen, Germany	HLA-matched unrelated	27	NHL	HAART discontinued after HSCT. CXCR4-tropic HIV–1 rebound, died from NHL relapse after 12 months [91–94]
Santiago, Chile	HLA-matched related	46	NHL	Died from pneumonia shortly post-transplant [91]
Berlin, Germany 2015	NA	NA	NA	NA [76, 91]
	CB donor graft cases			
Utrecht, the Netherlands	CB + Haploidentical graft	53	MDS	HAART continued after HSCT. Died from MDS relapse and pneumonia after 2 months [91, 93, 94]
Minneapolis, USA	CB Alone	12	ALL	No viral rebound. Died from leukemia/GvHD after 3 months [76, 91]
Barcelona, Spain	CB + haploidentical graft	37	NHL	Died from relapse of non-Hodgin's lymphoma after 3 months. No viral rebound prior to death [91, 95–96]

CB = cord blood; **AML** = acute myelogenous leukemia; **MDS** = myelodysplastic syndrome; **NHL** = non-Hodgkin's lymphoma; **ALL** = acute lymphoblastic leukemia; **ART** = anti-retroviral therapy; **GvHD** = graft-versus-host disease.
Note: One more CCR5-Δ32/Δ32 Donor HSCT was performed on a non-HIV-infected patient [87].

Table 4. CCR5-Δ32/Δ32 donor HSCT performed on HIV-infected patients up to 2014 [10, 75–76, 84–96].

In the three cases where CBT was employed with a CCR5-Δ32/Δ32 CB graft, one was a single CBT, with this Minneapolis patient expiring from GvHD after 3 months [91]. In the other two cases (the Barcelona and Dutch patients), both patients died from disease relapse prior to the fourth month, with the Dutch patient also suffering from pneumonia [91, 93, 94].

The Barcelona case was a 37-year-old HIV-1-infected patient with aggressive lymphoma who failed after five rounds of radiochemotherapy and an autologous HSCT. In the absence of a

matched sibling donor, the patient received an allogeneic 4/6 HLA A/B/DR-matched unrelated donor CCR5-Δ32/Δ32 CB product from the StemCyte inventory, supplemented with purified CD34+ cells from a haploidentical sibling [95, 96]. By genotypic and phenotypic analysis, the Barcelona patient's HIV strain was CCR5-tropic, with 2.1 copies of replication competent HIV per 10^7 CD4 T cells and 303 copies per ml enumerated by single copy assay. After HSCT with CCR5-Δ32/Δ32 CB and haploidentical sibling donor purified CD34+ cells, plasma HIV DNA load was confirmed to be undetectable using ultrasensitive analysis. Upon reaching full chimerism with the CB donor, the patient's engrafted CCR5-Δ32/Δ32 CD4 T cells responded to proliferation and activation stimuli in vitro but was resistant to infection by the patient's viral isolate and laboratory strains. Unfortunately, death related to lymphoma progression prevented long-term monitoring of the patient's status; however, this case shows the potential promise and utility of CCR5-Δ32/Δ32 CBT in curing HIV if the patient was infected with CCR5-tropic virus [96].

Despite incredible worldwide attention and many attempts to replicate the success of the Berlin patient, why has there been no other successful cured long-term survivors reported to date? For one, HSCT is a highly risky procedure, with expected worse survival in advanced cases of malignancies. Malignancy is a feature which all of these seven cases share [91]. Four of the six cases expired because of or partially due to relapses from their malignant conditions. The other cases died from infection or pneumonia—all common causes of post-transplant mortalities. Only one case out of eight reported viral rebound, albeit with virus of a different tropism which already existed in the patient prior to transplantation, perhaps indicating that viral rebound through tropism switch is not as easy as one might hypothesize [92].

Currently, the most important barrier to having a large CCR5-Δ32/Δ32 adult donor or CB inventory is the expense and logistical difficulties of screening millions of adult donors or hundreds of thousands of CB archival samples. The 26,000 StemCyte CB units, 10,000 CB units from the M.D. Anderson bank and 8000 adult donors tested in the German Red Cross Donor Registry represent the largest repositories screened for CCR5-Δ32/Δ32 genotypes to date. As such, the current availability of HLA-matched CCR5-Δ32/Δ32 homozygous donors is still extremely limited. The frequency of homozygous CCR5-Δ32 is around 1% in Caucasians, even less in the Middle East and 0% or almost 0 in Africa and in Asian countries like Taiwan, China and Japan [75, 87]. The requirement for rigorous co-selection of both suitable HLA matches and CCR5-Δ32/Δ32 genotypes leads to difficulties in finding appropriate adult donors to treat HIV infection with hematopoietic stem cells from adult donors. Cord blood, however, can tolerate 1 or 2 HLA A/B/DR mismatches and does not necessarily need HLA matches for HLA C/DP/DQ loci; therefore, a CCR5-Δ32/Δ32 CB inventory gives rise to a higher probability of finding a suitable CCR5-Δ32/Δ32 CB unit for individual patients [87, 97]. Instead of needing to find a 10/12 HLA A/B/C/DR/DP/DQ match for homozygous CCR5-Δ32/Δ32 adult donors, the chances of a 4/12 A/B/C/DR/DP/DQ homozygous CCR5-Δ32/Δ32 CB donor unit are several logs easier, despite the cell dose limitations of CB products and the smaller sizes of CB inventories compared to bone marrow registries worldwide. In fact, the difficulties in the logistics and incredible cost barriers of screening tens of millions of adult donors for the CCR5-Δ32/Δ32 genotype cannot be over-emphasized. In contrast, with readily available archived

samples and smaller inventories to screen, screening CB banks' products is an easier and less expensive proposition. Moreover, the fact that CB banks are cryopreserved physical inventories instead of a virtual database allows for easier construction of large banks of genotyped and infectious and genetic disease-screened samples, perpetually available for immediate shipping and transplantation with no possibility of donor refusal.

To explore the potential of CB stem cells as a more accessible source for curing HIV through HSCT, starting in 2001, working with City of Hope, Chow and Petz started screening for CCR5-Δ32/Δ32 genotypes of ~18,000 Caucasian and ~8000 Asian CB units and identified 134 Caucasian and 0 Asian CB units with the CCR5-Δ32/Δ32 genotype from StemCyte International Cord Blood Center [87]. During the last few years, additional CB inventories from other cooperating CB banks are being tested, and the CCR5-Δ32/Δ32 unit number has grown to ~180 by 2013 [89] with the hope of increasing the inventory to at least 300 CCR5-Δ32/Δ32 CB units [87]. Studies have shown that the engraftment and survival of CBT recipient are highly dependent on the number of nucleated and CD34+ cells. The most commonly accepted threshold for TNC dose is 2.5×10^7 nucleated cells/kg recipient body weight, but this number may not be achieved by all CB units collected, especially for adult patients. Delayed engraftment and immune reconstitution are observed with CBT due to the low progenitor cell numbers in CB products. With a theoretical inventory of over 300 CCR5-Δ32/Δ32 cryopreserved units, working with the NMDP bioinformatics team, it has been predicted that the ≥4/6 HLA match rate would be 73.6% for Caucasian pediatric patients (younger than age of 16) at the minimal TNC dose of 2.5×10^7 nucleated cells/kg recipient body weight. Far lower rates at ≥ 4/6 HLA match are found for adult patients (16 years or older) of all race groups at this minimal TNC dose of 2.5×10^7 nucleated cells/kg recipient body weight – 27.9% for Caucasian, 2.7% for Chinese-American, 9.9% for African-American, and 14% for Mexican American. To overcome the problems of insufficient cell dose with CBT, double and sequential CBT methods have been developed to overcome this restraint, expand the access of CBT to patients and improve transplantation outcomes [98–100]. With the option of double CBT, the minimum number of nucleated cells necessary for successful transplant drops to a minimal TNC of 1.0×10^7 nucleated cells/kg for each CB unit and 2.5×10^7 nucleated cells/kg for the combined cell dose, thus allowing more patients, especially adult and heavier pediatric patients, to have the access to suitable CB donors [100]. According to Petz et al., the rate of finding 4/6 HLA-matched units can reach 85.6% for Caucasian pediatric patients and 82.1% for Caucasian adults, when only 1.0×10^7 nucleated cells/kg is applied as the selection criteria [87].

Unfortunately, the possibility of finding two 4/6 HLA-matched CB products for double CBT for a single HIV-infected patient out of a small 300 CCR5-Δ32/Δ32 CB unit inventory is also remote. Since that probability approximates zero, to reach a combined TNC of 2.5×10^7 nucleated cells/kg is essentially the same as just using a single CB unit with TNC of 2.5×10^7 nucleated cells/kg. As you can see from **Table 5**, as such, the chances of a double CBT with both CB units being homozygous for the CCR5-Δ32 mutation and with combined TNC ≥2.5×10^7 are expected to be similar to the probability of finding a single homozygous CCR5-Δ32/Δ32 CB unit with TNC ≥2.5×10^7 projected by Petz et al. [87].

	Double CCR5-Δ32/Δ32 CBT		**CCR5-Δ32/Δ32 CBT + WT CCR5 OR CCR5/CCR5-Δ32 CBT OR Haploidentical Adult Donor Transplant**	
	Adult patients @ combined CCR5-Δ32/Δ32 CB TNC ≥2.5 × 10^7	**Pediatric patients @ combined CCR5-Δ32/Δ32 CB TNC ≥2.5 × 10^7**	**Adult patients @ CCR5-Δ32/Δ32 CB TNC ≥1.0 × 10^7**	**Pediatric patients @ CCR5-Δ32/Δ32 CB TNC ≥1.0 × 10^7**
Caucasians				
6/6 HLA A/B/DR matches	~0.01%	~0.01%	0.09%	1.01%
≥5/6 HLA A/B/DR matches	~4.5%	~10.6%	10.7%	10.8%
≥4/6 HLA A/B/DR matches	~27.9%	~73.6%	82.1%	85.6%
Chinese American				
≥4/6 HLA A/B/DR matches	~2.7%	~12.3%	13.9%	15.7%
African American				
≥4/6 HLA A/B/DR matches	~9.9%	~28.6%	31.6%	34.1%
Mexican American				
≥4/6 HLA A/B/DR matches	~14%	~44.1%	48.9%	52.5%

Adapted from Petz et al. [87].

Table 5. Projected match rate with an inventory of 300 CCR5-Δ32/Δ32 cord blood units using double CCR5-Δ32/Δ32 cord blood transplantation strategy versus the CCR5-Δ32/Δ32 Cord blood + bridging CCR5 or CCR5/CCR5-Δ32 cord blood or haploidentical donor strategy.

Instead of homozygous CCR5-Δ32/Δ32 double CBT, we and others have proposed alternative "bridging" strategies. If a single CCR5-Δ32/Δ32 CB unit does not meet the ideal 2.5 × 10^7 nucleated cells/kg recipient body weight cell dose threshold, a second CB unit with either wild-type (WT) CCR5 or heterozygous CCR5/CCR5-Δ32 donor may be used in a double CB transplant ("the CCR5-Δ32/Δ32 CB + CCR5 CB OR CCR5-Δ32/Δ32 CB + CCR5/CCR5-Δ32 CB double CBT strategy"). Alternatively, it is possible to combine the first homozygous CCR5-Δ32/Δ32 CB product with a bridging haploidentical adult donor with wild-type CCR5 or heterozygote CCR5/CCR5-Δ32 genotype as a second graft to "bridge" the patient until the homozygous CCR5-Δ32/Δ32 CB unit has engrafted ("the CCR5-Δ32/Δ32 CB + haploidentical CCR5 donor OR CCR5-Δ32/Δ32 CB + CCR5/CCR5-Δ32 haploidentical adult donor strategy") [76, 91, 94, 101]. This was the strategy tried for the Barcelona and Dutch patients [93, 94, 96]. The hope is that the survival advantage of CCR5-Δ32/Δ32 CB against HIV infection and lysis of wild-type CCR5 or heterozygote cells would enable the homozygous CCR5-Δ32/Δ32 donor CB to prevail over the wild-type CCR5 or heterozygous CCR5/CCR5-Δ32 CB or haploidentical adult graft eventually. Since wild-type CCR5 or heterozygote CCR5 donors are readily available, this strategy will yield far higher probability of finding a match since only TNC ≥1.0 × 10^7 /kg is required for the first homozygous CCR5-Δ32/Δ32 CB product. As seen in **Table 5**, the effect of just requiring TNC ≥1.0 × 10^7/kg for the first homozygous CCR5-Δ32/Δ32 cord blood product resulted in a much higher match rate for adult patients—27.9 to 82.1% for

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Caucasians, 2.7 to 13.9% for Chinese Americans, 9.9 to 31.6% for African Americans and 14 to 48.9% for Mexican Americans. Moreover, if this strategy is combined with Chow's MaxCell CB processing technologies and no-wash direct infusion strategy, then the probability of finding a CCR5-Δ32/Δ32 CB product with higher cell dose further increases [1, 8].

Instead of having HIV-infected patients search for matched donor CCR5-Δ32/Δ32 CB, the more efficient strategy may be for the limited CCR5-Δ32/Δ32 donor inventory to find matched HIV patients who have concomitant transplant indications [1]. In this scenario, instead of having HIV-infected patients searching for CCR5-Δ32/Δ32 donors, it is possible to reverse the process and have the HLA type CCR5-Δ32/Δ32 CB or adult donors database, look for HIV-infected individuals with transplant indications who have suitable HLA matches among the CCR5-Δ32/Δ32 inventory. The math of a patient looking for a graft and a graft looking for a patient is the same, just in the opposite directions; however, this allows targeted CCR5 genotype screening of HIV patients who are in need of a HSCT [1].

Overall, HSCT has the potential to accomplish functional or sterilization cure of HIV patients and would be especially valuable to HIV patients with hematological malignancy, which may cure both indications with the same HSCT. Cord blood could be a more accessible alternative to HLA-matched adult donor for curing HIV infection through HSCT, even though more clinical trials on HIV patients would be necessary to establish the efficacy in eradicating HIV infection for both adult donor HSCT and CBT. Due to the difficulties of co-selecting for units with ≥4/6 HLA A/B/DR matches, suitable nucleated or CD34+ cell doses and the CCR5-Δ32/Δ32 genotype, innovative strategies such as (1) double grafts combining a homozygous CCR5-Δ32/Δ32 CB with either another wild-type or CCR5/CCR5-Δ32 heterozygote graft, (2) combined with MaxCell CB processing and thawing technologies that yield higher cell doses and (3) the employment of the reverse strategy of looking for matched HIV patients in need of HSCT, can be helpful in expanding the utility of the CCR5/CCR5-Δ32 CB inventory for viable transplantation. Actively screening for more CCR5-Δ32 homozygous CB units, establishment of more comprehensive HIV-infected patient HLA type database and the application of double CBT or bridging strategies would eventually allow more HIV-infected patients find suitable units for transplantation.

7. Cure of autoimmune diseases by cord blood transplantation

Although many patients with autoimmune diseases (AD) can have a relatively normal life expectancy with treatment, some patients with severe, progressive and therapy-refractory autoimmunity require more than medication. Patients with systemic sclerosis (SSc), for example, have shown disappointing results in prospective randomized trials of almost all therapeutic agents reported [102]; therefore, HSCT, both autologous and allogeneic, has been proposed as an effective potential treatment for such patients.

The basis for autologous HSCT for AD is the immune system reset by the generation of fresh self-tolerant lymphocytes after chemotherapy-induced elimination of self or auto-reactive lymphocytes. Elimination of auto-reactive T-effector cells, long-living plasma cells and

antigen-presenting cells as well as increased T-regulatory cells, restoration of thymic function, normalization of T-receptor repertoire, reduced auto-antibodies and long-lasting lymphopenia are the intended effects of autologous HSCT for AD [103]. Autologous HSCT carries the risk that if the basic defect of the AD is in the stem cell, the autoimmunity will probably recur after autologous HSCT; however, if the primary defect is an aberrant immune response to an acquired, for example, viral antigen, or self-antigen, there is a theoretical possibility that tolerance may be acquired in the newly reconstituted autologous immune system—assuming ablation of the offending memory cells.

Autologous HSCT has been applied to treat severe autoimmune diseases (SAD) since 1996 [104], when some of the first autologous transplants specifically for AD were performed by Tyndall and colleagues [105]. Autologous HSCT for AD is based on the elimination of auto-reactive effectors through potent immunosuppressive conditioning followed by subsequent regeneration of self-tolerated lymphocytes capable of "resetting" the immune system. This approach has been recognized to induce remission for some patients with various AD, including SSc, systemic lupus erythematosus (SLE), multiple sclerosis (MS), rheumatoid arthritis (RA), adjuvant arthritis and severe Crohn's disease [104–112]. Multiple sclerosis has been the main indication for autologous HSCT, along with SLE, therapy-refractory Crohn's, vasculitis, autoimmune cytopenia, diabetes mellitus type 1, polyarthritis, adults with rheumatoid arthritis and children with juvenile idiopathic arthritis; however, RA relapse is frequent [103]. According to the data from European Group for Blood and Marrow Transplantation (EBMT) registry from 1996 to 2007, the 5-year OS was 85% among the 900 patients who underwent autologous HSCT for AD; however, the 5-year progression-free survival (PFS) was only 43% [104]. Even lower values, 33% for 5-year PFS (78% for 5-year OS), were reported by the British Society of Blood and Marrow Transplantation (BSBMT) data registry for 1997–2009 [113]. Although these studies proved the relative safety of autologous HSCT, the fact that more than half of the patients suffered from disease relapse is unsatisfactory. This was especially problematic for patients with RA, with a 3-year PFS of only 23% despite its high 3-year OS of 98% [104]. This tendency to relapse was also confirmed in the report from Snowden et al. [111]. With data from both EBMT and Autologous Blood and Marrow Transplant Registry (ABMTR), Snowden suggested that the majority of RA patients experienced a reactivation of the disease eventually and required the re-introduction of immunosuppressive drugs. Though somewhat better than RA, autologous HSCT for other ADs is also hampered by disease progression or relapse, showing unimpressive 3-year PFS ranging from 34 to 63% [104]. Illei and colleagues suggested that the reactivation of lupus is a major contributing factor to the deaths of SLE patients after autologous HSCT treatment [114]. Similar association between autologous HSCT and relapse was also identified with SSc patients [115]. T cell depletion has been proposed to be a relapse-preventing strategy in several clinical trials; however, no significant improvement in either OS or PFS has been observed [104]. The unavoidable high recurrence rate, together with the elevated risks of stem collection procedures on AD patients themselves, lead to increased adverse events or even mortality of patients.

Compared to autologous HSCT, which only produces sustained responses in 30–40% patients, allogeneic HSCT can achieve sustained response in 60–70% patients [113]. Allogeneic HSCT,

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which does not need stem cell collection from the patient and is less prone to disease relapse, is therefore considered higher GvHD and TRM risk, but may produce more favorable long-term results with AD relapse. In fact, evidence for the potential of HSCT to cure or ameliorate AD comes from many cases when patients undergo allogeneic HSCT for another indication with coincident AD, such as RA, psoriasis, psoriatic arthritis and ulcerative colitis, with the AD often in remission post-transplant. Moreover, the converse observation has also been reported, that is, passive transfer of AD from the donor graft through allogeneic HSCT, including myasthenia gravis, Graves' disease and autoimmune diabetes mellitus.

In all but one of the cases reporting the use of allogeneic HSCT for concomitant AD, the donor was an HLA-identical sibling. Therefore, one possible explanation for recurrence of the AD may be the presence of shared genetic factors between the related donors and recipients. Another possible explanation is the persistence of host immune cells resulting in recurrence of disease activity. These case reports, although small in number, were important in establishing our understanding of the potential role of allogeneic HSCT for treatment of patients with severe autoimmune diseases.

The failure of autologous HSCT to cure spontaneous-onset AD and to maintain long-term regression could be due to the incomplete elimination of self-responsive memory T cells and B cells. While the conditioning regimes through chemotherapy or irradiation are unable to eradicate every single memory lymphocyte, complete immune ablation can be achieved by allogeneic HSCT via combined effects of immune system replacement and graft-versus-autoimmunity (GVA) effect [116]. In allogeneic HSCT, the host auto-reactive immune system is replaced by the donors' non-auto-reactive, but potentially alloreactive cells. Therefore, alloreactive donor lymphocytes can undergo a GVA process similar to the graft-versus-leukemia (GVL) effect or GvHD and attack the residual self-reactive host effectors [116, 117]. Thus, the biggest problem with allogeneic adult donor HSCT for AD is GvHD. In fact, several cases of complete donor chimerism after adult donor HSCT cured the patients' AD, but were accompanied by severe acute or chronic GvHD post-transplant [118, 119]. In contrast, cases of mixed chimerism, which sometimes occurs with reduced intensity or non-myeloablative conditioning regimens, often exhibit mild and sometimes no GvHD [120–122]. Despite the differences in the conditioning regimen used by the different groups, many patients treated with allogeneic HSCT have shown sustained remission to AD and amelioration of symptoms, and among patients with severe treatment-refractory ADs, the response rate was higher than 75% [123]. With reduced intensity conditioning, the adverse effects and treatment toxicity observed in myeloablative autologous HSCT could be reduced [107]. However, relatively high transplant-related mortality (22.1% at 2 years and 33.7% at 5 years) and GvHD remain as the biggest challenges to allogeneic HSCT for AD [123]; yet, the 5-year OS for allogeneic HSCT is not significantly different than the 78% 5-year OS for autologous HSCT reported by the BSBMT [113], but appears to be lower than the 85% OS for autologous HSCT reported by the EMBT [104].

To minimize the risks of GvHD with allogeneic HSCT for AD, there are emerging interests in using CB as the alternative source of stem cells to the traditionally used (or simply "traditional") adult stem cell sources of bone marrow (BM) and peripheral blood (PB). While tolerating

far greater degrees of HLA mismatch, CBT is still associated with lower incidences and severity of acute and chronic GvHD than HSCT from adult donors [124]. According to the recent study of 143 patients in 2010, the rate of grade II or higher GvHD is only 9% among HLA-matched (n = 60) and 50% among HLA-mismatched (n = 18) patients [125]. Being less mature than stem cells from adult grafts, CB stem cells show lower alloreactivity and immunogenicity without increasing long-term relapse incidences [126]. Moreover, CB has other advantages including low contamination rate, easy storage and immediate accessibility via cryopreservation [125].

Together with its relatively lenient requirement for HLA matching, CB has been used commonly as an alternative source for transplantation. Indeed, one of the first cases to treat AD with CBT was performed due to lack of HLA-matched, appropriate adult stem cell source [127]. Raetz and colleagues performed myeloablative conditioning HSCT on a 5-year-old boy with severe Evans syndrome, which consists of immune thrombocytopenia and Coombs-positive hemolytic anemia. The graft was a CB from an HLA-matched sibling with 3.85×10^7 nucleated cells/kg patient and 0.96×10^5 CD34+ cells/kg patient infused, leading to complete remission. Acute GvHD prophylaxis regimen was with cyclosporine, with G-CSF initiated on day +1. The patient engrafted with an absolute neutrophil count (ANC) greater than 0.5×10^9/l on day +16. The following day, he developed symptoms of acute GvHD, with temperatures of up to 40°C, skin rash that on biopsy was consistent with GvHD and severe pulmonary insufficiency. He was intubated for 2 days and treated with high-dose steroids, with rapid resolution of symptoms. Platelet engraftment was delayed, with sustained platelets greater than 30×10^9/l by day +170. He was platelet independent from day +240 and RBC independent from day +210. Reevaluation of his RBC antibody status revealed a DAT that was only microscopically positive by day +20 and negative on day +286. Anti-platelet antibodies were negative on day +115 and day +176. He had no evidence of response with cyclosporine pre-transplant and no evidence of disease recurrence 2 months after discontinuation of cyclosporine and 4 months after discontinuation of prednisone. At the time of death, the patient was off all immunosuppression and had complete resolution of his autoimmune disease. However, the patient died unexpectedly from acute hepatic failure 9 months after transplantation [127].

Itamura et al. reported another case for a 48-year-old female patient with RA [128], who was subsequently diagnosed as having autoimmune hemolytic anemia (AIHA). Four months after the diagnosis of AIHA, she suddenly developed hemophagocytic syndrome (HPS) with disseminated intravascular coagulation (DIC) and *Staphylococcus aureus* bacteremia. Her HPS transiently improved after treatment, but relapsed 3 weeks later. Without an immediately available related adult donor, an unrelated donor CB graft was used for HSCT. The patient had HLA antibodies against multiple HLA class I antigens; however, the CB unit did not express HLA antigens reactive to her HLA antibodies and contained sufficient cell dose. HLA type of the CB graft was A*02:06/A*11:01, B*40:06/B*67:01, DRB1*09:01/DRB1*16:02 and that of the recipient was A*11:01/A*26:02, B*40:06/B*67:01, DRB1*09:01/DRB1*16:02. Four months after the onset of HPS, she received a CBT following a conditioning regimen of melphalan (40 mg/m^2/day 2 days), fludarabine (25 mg/m^2/day 5 days) and total-body irradiation. Tacrolimus was used for the prophylaxis of GvHD. Engraftment of neutrophils ANC500 and platelets 20K occurred on days 14 and 32, respectively. Bone marrow examination on day 60 showed

complete donor-type chimerism by short tandem repeat-polymerase chain reaction and no evidence of HPS. EBV-DNA load was under the detectable limit. Post-transplant, the patient was not only cured for HPS, but also showed marked amelioration of preexisting RA and AIHA [128].

Others have performed unrelated donor CBT for AD, including at least two pediatric cases of scleroderma and systemic sclerosis where MaxCell CB products were provided by the StemCyte CB bank founded by the corresponding author. Both cases were performed at the same transplant center; however, the attending transplant physician relocated to a different hospital subsequently and both cases were lost to follow-up [R. Chow, unpublished data]. These CBT cases suggest that allogeneic CBT can potentially be a powerful tool for curing AD without significant GvHD provided that the cell dose and HLA match is adequate.

In summary, by optimizing transplant conditions and maximizing cell dose through the use of products manufactured by the MaxCell CB processing technologies, use of double CBT and direct infusion without post-thaw wash, unrelated CBT has been shown in the largest patient series of its kind to be capable of producing outstanding clinical outcome for thalassemia major, with excellent long-term TRM, OS and DFS that rivals or approaches that of related CBT. Such strategies may be employed to optimize unrelated donor CBT for other nonmalignant conditions, such as HIV infection or AD. For HIV infection, optimization of cell dose may make the difference between a HIV-resistant CB graft being eligible for HSCT, especially if in combination with other grafts. Similarly, cell dose maximization will minimize graft failure and TRM for CBT of AD. After the establishment of large racially and ethnically diverse CB banks with MaxCell CB products, further CBT studies of more unrelated patients with thalassemia, HIV infection and AD will be needed to see whether these hypotheses can be validated and the strategies can be widely applicable.

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References

[1] Chow M, et al. Curative cord blood hematopoietic stem cell transplantation (CBT) for HIV infection and thalassemia major (Thal). *J Sci Appl Biomed.* 4: 15–16 (2016).

[2] Rocha V., Sanz G., Gluckman E. Umbilical cord blood transplantation. *Curr Opin Hematol.* 11: 375–385 (2004).

[3] Ruggeri A, et al. Umbilical cord blood transplantation for children with thalassemia and sickle cell disease. *Biol Blood Marrow Transplant.* 17: 1375–1382. (2011).

[4] Jaing TH, et al. Rapid and complete donor chimerism after unrelated mismatched cord blood transplantation in five children with beta-thalassemia major. *Biol Blood Marrow Transplant.* 11: 349–353 (2005).

[5] Petz L, et al. Analysis of 120 pediatric patients with non-malignant disorders transplanted using unrelated plasma depleted/reduced cord blood. *Transfusion* 52: 1311–1320 (2012).

[6] Jaing TH, et al. Unrelated cord blood transplantation for thalassemia: a single-institution experience of 35 patients. *Bone Marrow Transplant.* 47: 33–39 (2012).

[7] Petz L, et al. Cure of thalassemia major using related and unrelated cord blood transplantation. In *Thalassemia: causes, treatment options and long-term health outcomes.* Ed. Greene, Makenzie, Chapter 1, pp. 1–36; 263 pages. Hauppauge: Nova Science Publishers, Inc., Hauppauge, New York, (2014).

[8] Chow R, et al. Cell recovery comparison between plasma depletion/reduction and red cell reduction processing of umbilical cord blood. *Cytotherapy* 13: 1105–1119 (2011).

[9] Barker JN, et al. Transplantation of two partially HLA-matched umbilical cord blood units to enhance engraftment in adults with hematologic malignancy. *Blood* 105: 1343–1347 (2005).

[10] Hütter G, Ganepola S. Eradication of HIV by transplantation of CCR5-deficient hematopoietic stem cells. *Scientific World J.* 11: 1068–1076 (2011).

[11] Lucarelli G, Gaziev J. Hematopoietic cell transplantation for thalassemia. In *Thomas' Hematopoietic Cell Transplantation, 5th Edition*, Ed. Forman, S.J., Negrin, R.S., Antin, J.H., Appelbaum, F.R. pp. 842–854. New York: John Wiley & Sons (2016).

[12] Cao A. Quality of life and survival of patients with beta-thalassemia major. *Haematologica* 89: 1157–1159 (2004).

[13] Thomas ED, et al. Marrow transplantation for thalassaemia. *Lancet* 2: 227–229 (1982).

[14] Lucarelli G, et al. Bone marrow transplantation in thalassemia. *J Exp Clin Cancer Res.* 3: 313–315 (1983).

[15] Lucarelli G, et al. Marrow transplantation in patients with advanced thalassemia. *N Engl J Med.* 316: 1050–1055 (1987).

[16] Lucarelli G, et al. Bone marrow transplantation in patients with thalassemia. *N Engl J Med.* 322: 417–421 (1990).

[17] Sodani P, et al. New approach for bone marrow transplantation in patients with class 3 thalassemia aged younger than 17 years old. *Blood* 104: 1201–1203 (2004).

[18] La Nasa G, et al. Unrelated donor bone marrow transplantation for thalassemia: the effect of extended haplotypes. *Blood* 99: 4350–4356 (2002).

[19] Gluckman E, et al. Outcome of cord-blood transplantation from related and unrelated donors. Eurocord Transplant Group and the European Blood and Marrow Transplantation Group. *N Engl J Med.* 337: 373–381 (1997).

[20] Rubinstein P, et al. Outcomes among 562 recipients of placental-blood transplants from unrelated donors. *N Engl J Med.* 339: 1565–1577 (1998).

[21] Rocha V, et al. Transplants of umbilical-cord blood or bone marrow from unrelated donors in adults with acute leukemia. *N Engl J Med*. 351: 2276–2285 (2004).

[22] Laughlin MJ, et al. Outcomes after transplantation of cord blood or bone marrow from unrelated donors in adults with leukemia. *N Engl J Med*. 351: 2265–2275 (2004).

[23] Locatelli F, et al. Related umbilical cord blood transplantation in patients with thalassemia and sickle cell disease. *Blood*. 101: 2137–2143 (2003).

[24] Lisini D, et al. Donor/recipient mixed chimerism does not predict graft failure in children with β-thalassemia given an allogeneic cord blood transplant from an HLA-identical sibling. *Haematologica* 93: 1859–1867 (2008).

[25] Locatelli F, et al. Outcome of patients with hemoglobinopathies given either cord blood or bone marrow transplantation from an HLA-identical sibling. *Blood* 122: 1072–1078 (2013).

[26] Jaing TH, et al. Transplantation of unrelated donor umbilical cord blood for nonmalignant diseases: a single institution's experience with 45 patients. *Biol Blood Marrow Transplant.* 16: 102–107 (2010).

[27] Wagner JE, et al. Transplantation of unrelated donor umbilical cord blood in 102 patients with malignant and nonmalignant diseases: influence of CD34 cell dose and HLA disparity on treatment- related mortality and survival. *Blood* 100: 1611–1618 (2002).

[28] Goussetis E, et al. Combined umbilical cord blood and bone marrow transplantation in the treatment of beta-thalassemia major. *Pediatric Hematol Oncol* 17: 307–314 (2000).

[29] Fang J, et al. Umbilical cord blood transplantation in Chinese children with beta-thalassemia. *J Pediatric Hematol Oncol*. 26: 185–189 (2004).

[30] Walters MC, et al. Sibling donor cord blood transplantation for thalassemia major: experience of the sibling donor cord blood program. *Ann NY Acad Sci.* 1054: 206–213 (2005).

[31] Jaing TH, Yang CP, Hung IJ, Chen SH, Sun CF, Chow R. Transplantation of unrelated donor umbilical cord blood utilizing double-unit grafts for five teenagers with transfusion-dependent thalassemia. *Bone Marrow Transplant.* 40: 307–311 (2007).

[32] Jaing TH, et al. Unrelated cord blood transplantation (UCBT) for transfusion-dependent thalassemia—a CIBMTR audited retrospective analysis of 51 consecutive patients. *Biol Blood Marrow Transplant.* 14(2S): 6 (2008).

[33] Jaing J, et al. Matched pair comparisons of unrelated cord blood transplantation (CBT) using plasma depleted cord blood products versus red cell reduced (RCR) CB in 30 pair of patients with thalassemia. *Cytotherapy* 10(S1): 186 (2008).

[34] Jaing T, Wang B, Gjertson D, Law P, Petz L, and Chow R. Unrelated cord blood transplantation (UCBT) for transfusion-dependent thalassemia—a CIBMTR audited retrospective analysis of 30 consecutive patients from a single center. *Blood* 112: 11 (2008).

[35] Chow R, et al. Unrelated cord blood transplantation (CBT) of 101 hemoglobinopathy (HGB) patients. *Biol Blood Marrow Transplant.* 18: 2–2; S268 (2012).

[36] Issaragrisil S, et al. Brief report: transplantation of cord blood stem cells into a patient with severe thalassemia. *N Engl J Med.* 332: 367–369 (1995).

[37] Jaing T, et al. Successful unrelated cord blood transplantation in a child with beta-thalassemia major. *J Trop Ped.* 51: 122–124 (2005).

[38] Chow R, et al. Hematopoietic stem cell transplantation (HSCT) using plasma depleted umbilical cord blood units (UCB) and the effect of post-thaw washing. In Wagner J, Champlin R, Petz L (Eds). Proceedings of the 4th Annual International Cord Blood Transplantation Symposium. *Biol Blood Marrow Transplant.* 12: 1206–1217 (2006).

[39] Chow R, et al. Analysis of hematopoietic cell transplants using plasma-depleted cord blood products that are not red blood cell reduced. *Biol Blood Marrow Transplant.* 13: 1346–1357 (2007).

[40] Chow R, et al. Selection of Post-thaw manipulations prior to transplantation of plasma depleted umbilical cord blood products. *Transfusion* 47(3S): 28A (2007).

[41] Chow R, et al. Avoidance of post-thaw wash prior to transplantation of plasma depleted cord blood is associated with improved engraftment & decreased severity of chronic GvHD (cGvHD) without increased relapse. *Blood* 110: 494a (2007).

[42] Chow R, et al. Matched pair comparisons of unrelated cord blood transplantation (CBT) using plasma depleted cord blood products versus red cell reduced (RCR) CB in 92 pair of patients with pediatric malignancies. *Cytotherapy* 10(S1): 178 (2008).

[43] Chow R, et al. A novel method to reduce rates of extensive chronic GvHD (cGvHD) without increased relapse for cord blood transplant. *Biol Blood Marrow Transplant.* 14(2S):11 (2008).

[44] Chow R, et al. Avoidance of post-thaw washing prior to transplantation of plasma depleted umbilical cord blood improves outcome in a matched pair audited analysis of 258 patients. *Transfusion* 48:(2S:17A) S43–020F (2008).

[45] Chow R, et al. Negative impact of post-thaw washing on the overall survival (OS) and disease free survival (DFS) of patients receiving plasma depleted cord blood (CB) transplantation. *Biol Blood Marrow Transplant.* 15: 2–2 (2009).

[46] Chow R, Cord blood unit selection: engraftment, GvHD and overall survival using cord blood units processed using plasma depletion. In: Wagner J, Laughlin M, Petz L (Eds) Symposium summary of the 7th annual international cord blood transplantation symposium. *Biol Blood Marrow Transplant.* 16: 12–27 (2010).

[47] Chow R, et al. Optimal recovery of SSEA-4+/Oct-4+/CD133+/CXCR4+/Lin-/CD45- very small embryonic-like (VSEL) stem cells from umbilical cord blood (CB) using plasma depletion/reduction compared to red cell reduction (RCR). *Biol Blood Marrow Transplant.* 17: 2–2 (2011).

[48] Chow R, et al. Cell recovery comparison between plasma depletion/reduction & red cell reduction processing of umbilical cord blood. *Cytotherapy* 13: 1105–1119 (2011).

[49] Rosenthal J, et al. Improved outcome for transplantation of pediatric patients with non-malignant disorders with unwashed plasma depleted cord blood. *Biol Blood Marrow Transplant.* 14(2S): 28 (2008).

[50] Petz L, Chow R. Correspondence: selection of unrelated cord blood grafts. *Blood* 118: 478–479 (2011).

[51] Jaing T, Tan P, Rosenthal J, Chow R. Rapid and durable engraftment after unrelated cord blood transplantation (CBT) for children with transfusion-dependent thalassemia. In Wagner, J, Champlin, R, Petz, L. eds. Proceedings of the 4th Annual International Cord Blood Transplantation Symposium. *Biol Blood Marrow Transplant* 12: 1206–1217 (2006).

[52] Jaing T, et al. Unrelated cord blood transplantation (UCBT) for transfusion-dependent thalassemia—a retrospective audited analysis of 41 consecutive patients. *Biol Blood Marrow Transplant.* 13(S2): 62 (2007).

[53] Jaing TH, Wang B, Gjertson D, Petz L, Chow R. Unrelated cord blood transplantation (UCBT) of 30 consecutive patients with transfusion-dependent β-thalassemia from a single center. *Biol Blood Marrow Transplant.* 15: 2–2 (2009).

[54] Rosenthal J, et al. Unrelated cord blood transplantation (UCBT) for 59 pediatric patients with benign indications using plasma depleted cord blood, an audited retrospective analysis. *Biol Blood Marrow Transplant.* 13(S2): 7 (2007).

[55] Graham M, et al. A retrospective study and matched pair analysis of 240 pediatric patients with malignancies transplanted with plasma depleted or red

cell reduced (RCR) cord blood (CB) products. *Biol Blood Marrow Transplant.* 14(2S): 133 (2008).

[56] Smythe J, et al. Directed sibling cord blood banking for transplantation: the ten year experience in the National Blood Service in England. *Stem Cells* 25: 2087–2093 (2007).

[57] Rocha V, et al. Effect of HLA-matching recipients to donor non-inherited maternal antigens on outcomes after mismatched umbilical cord blood transplantation for hematologic malignancy. *Biol Blood Marrow Transplant.* 18(12):1890–1896 (2012).

[58] Jaing TH, et al. Second transplant with two unrelated cord blood units for early graft failure after cord blood transplantation for thalassemia. *Ped Transplant.* 13:766–768. (2009).

[59] Kharbanda S, et al. Unrelated donor allogeneic hematopoietic stem cell transplantation for patients with hemoglobinopathies using a reduced-intensity conditioning regimen and third-party mesenchymal stromal cells. *Biol Blood Marrow Transplant.* 20: 581–586 (2014).

[60] Soni S., Breslin N, Cheerva A. Successful unrelated umbilical cord blood transplantation for class 3 β-thalassemia major using a reduced-toxicity regimen. *Ped. Transplant.* 18: E41–E43 (2014).

[61] Brunstein CG, et al. Intra-BM injection to enhance engraftment after myeloablative umbilical cord blood transplantation with two partially HLA-matched units. *Bone Marrow Transplant.* 43(12): 935–940 (2009).

[62] Rocha V, et al. Unrelated cord blood transplantation: outcomes after single-unit intrabone injection compared with double-unit intravenous injection in patients with hematological malignancies. *Transplantation* 95: 1284–1291 (2013).

[63] Lau YL, et al. Sibling HLA-matched cord blood transplant for beta-thalassemia: report of two cases, expression of fetal hemoglobin, and review of the literature. *J Pediatr Hematol Oncol* 20: 477–481 (1998).

[64] Hongeng S, et al. Full chimerism in nonmyeloablative stem cell transplantation in a beta-thalassemia major patient (class 3 Lucarelli). *Bone Marrow Transplant.* 30: 409–410 (2002).

[65] Fang J, Huang S, Chen C, Zhou D. Umbilical cord blood transplantation for beta-thalassemia. *J Trop Pediatrics* 49: 71–73 (2003).

[66] Tan PH, et al. Unrelated peripheral blood and cord blood hematopoietic stem cell transplants for thalassemia major. *Am J Hematol.* 75: 209–212 (2004).

[67] Hall JG, Martin PL, Wood S, Kurtzberg J. Unrelated umbilical cord blood transplantation for an infant with beta-thalassemia major. *J Pediatr Hematol Oncol.* 26: 382–385 (2004).

[68] Vanichsetakul P, et al. Umbilical cord blood transplantation in children with beta-thalassemia diseases. *J Med Assoc Thailand* 87(S2): S62–67 (2004).

[69] Nagamura-Inoue T, et al. Wash-out of DMSO does not improve the speed of engraftment of cord blood transplantation: follow-up of 46 adult patients with units shipped from a single cord blood bank. *Transfusion* 43: 1285–1295 (2003).

[70] Laroche V, et al. Cell loss and recovery in umbilical cord blood processing: a comparison of postthaw and postwash samples. *Transfusion* 45: 1909–1916 (2005).

[71] Stiff P, Shane M, and Rodriguez T. Successful umbilical cord blood transplants in adults who received a nucleated cell dose = 1×10(7) cells/kg processed by a post-thaw non-wash procedure. *Blood* 106: 580a (2005).

[72] Richman DD, et al. The challenge of finding a cure for HIV infection. *Science* 323: 1304–1307 (2009).

[73] Krishnan A, et al. Durable remissions with autologous stem cell transplantation for high-risk HIV-associated lymphomas. *Blood* 105: 874–878 (2005).

[74] Hassett JM, Zaroulis CG, Greenberg ML, Siegal FP. Bone marrow transplantation in AIDS. *N Engl J Med.* 309: 665 (1983).

[75] Hütter G, Zaia JA. Allogeneic haematopoietic stem cell transplantation in patients with human immunodeficiency virus: the experiences of more than 25 years. *Clin Exp Immunol.* 163: 284–295 (2011).

[76] Hütter G. Transplantation of CCR5-Δ32/Δ32 stem cells may cure HIV infection. *Stem Cells Cancer Stem Cells* 13:35–41 (2015).

[77] Henrich TJ, et al. Long-term reduction in peripheral blood HIV type 1 reservoirs following reduced-intensity conditioning allogeneic stem cell transplantation. *J Infect Dis.* 207: 1694–1702 (2013).

[78] Henrich TJ, et al. Antiretroviral-free HIV–1 remission and viral rebound after allogeneic stem cell transplantation: report of 2 cases. *Ann Intern Med.* 161: 319–327 (2014).

[79] Chow R, et al. U.S. patent application #09/998,832 (2001).

[80] Liu R, et al. Homozygous defect in HIV-1 coreceptor accounts for resistance of some multiply-exposed individuals to HIV-1 infection. *Cell* 86: 367–377 (1996).

[81] Samson M, et al. Resistance to HIV-1 infection in Caucasian individuals bearing mutant alleles of the CCR-5 chemokine receptor gene. *Nature* 382: 722–725 (1996).

[82] Eugen-Olsen J, et al. Heterozygosity for a deletion in the CCR-5 gene leads to prolonged AIDS-free survival and slower CD4 T-cell decline in a cohort of HIV-seropositive individuals. *AIDS* 11: 305–310 (1997).

[83] Cohen OJ, et al. Heterozygosity for a defective gene for CC chemokine receptor 5 is not the sole determinant for immunologic and virologic phenotype of HIV-infected long-term nonprogressors. *J Clin Invest.* 100: 1581–1589 (1997).

[84] Hütter G, et al. Long-term control of HIV by CCR5 Delta32/Delta32 stem-cell transplantation. *N Engl J Med.* 360: 692–698 (2009).

[85] Allers K, et al. Evidence for the cure of HIV infection by CCR5Δ32/Δ32 stem cell transplantation. *Blood* 117: 2791–2799 (2011).

[86] Yukl SA, et al. Challenges in detecting HIV persistence during potentially curative interventions: a study of the Berlin patient. *PLoS Pathogens* 9: e1003347 (2013).

[87] Petz LD, et al. Hematopoietic cell transplantation with cord blood for cure of HIV infections. *Biol Blood Marrow Transplant.* 19(3): 393–397 (2013).

[88] Chow et al. U.S. patent 2003/0099621 AI (2003).

[89] Petz L. Cord blood transplantation for cure of HIV infections. *Stem Cells Transl Med.* 2(9): 635–637 (2013).

[90] Chen TK, et al. The feasibility of using CCR532Δ/32Δ hematopoietic stem cell transplants for immune reconstitution in HIV-infected children. *Biol Blood Marrow Transplant.* 14(2S): 119 (2008).

[91] Hütter G. More on shift of HIV Tropism in stem-cell transplantation with CCR5 delta32/delta32 mutation. *N Engl J Med.* 371: 2437–2438 (2014).

[92] Kordelas L, Verheyen J, Esser S. Shift of HIV tropism in stem-cell transplantation with CCR5 delta32/delta32 mutation. *N Engl J Med.* 371: 880–882 (2014).

[93] Kwon M, et al. Single cord blood combined with HLA-mismatched third party donor cells: comparable results to matched unrelated donor transplantation in high-risk patients with hematologic disorders. *Biol Blood Marrow Transplant.* 19: 143–149 (2012).

[94] Kwon M, et al. A single cord blood transplantation combined with a HLA mismatched third party donor for a patient with Burkitt Lymphoma and HIV infection. In*: Proceedings of the 10th International Cord Blood Symposium*, San Francisco, June 7–9, 2012 abstract book (2012).

[95] Henrich TJ. HIV eradication: is cord blood the answer? *Lancet HIV* 2(6): e219–20 (2015).

[96] Duarte RF, et al. CCR5 Δ32 homozygous cord blood allogeneic transplantation in a patient with HIV: a case report. *Lancet HIV* 2(6): e236–42 (2015).

[97] Smith AR, Wagner JE. Alternative haematopoietic stem cell sources for transplantation: place of umbilical cord blood. *Br J Haematol.* 147: 246–261 (2009).

[98] Majhail N, Brunstein C, Wagner J. Double umbilical cord blood transplantation. *Curr Opin Immunol.* 18: 571–575 (2006).

[99] Brunstein C, et al. Umbilical cord blood transplantation after nonmyeloablative conditioning: impact on transplantation outcomes in 110 adults with hematologic disease. *Blood* 110: 3064–3070 (2007).

[100] Brunstein CG, et al. Allogeneic hematopoietic cell transplantation for hematologic malignancy: relative risks and benefits of double umbilical cord blood. *Blood* 116(22): 4693–4699 (2010).

[101] Liu H, et al. Reduced-intensity conditioning with combined haploidentical and cord blood transplantation results in rapid engraftment, low GvHD, and durable remission. *Blood* 118: 6438–6445 (2011).

[102] Viganego F, Nash R, Furst DE. Bone marrow transplantation in the treatment of systemic sclerosis. *Curr Rheumatol Rep.* 2: 492–500 (2000).

[103] Hügle T, Daikeler T. Stem cell transplantation for autoimmune diseases. *Haematologica* 95: 185–188 (2010).

[104] Farge D, et al. Autologous hematopoietic stem cell transplantation for autoimmune diseases: an observational study on 12 years' experience from the European Group for Blood and Marrow Transplantation Working Party on Autoimmune Diseases. *Haematologica* 95: 284–292 (2010).

[105] Tyndall A, Gratwohl A. Haemopoietic stem and progenitor cells in the treatment of severe autoimmune diseases. *Ann Rheumatic Dis.* 55: 149–151 (1996).

[106] Burt R, et al. Association of nonmyeloablative hematopoietic stem cell transplantation with neurological disability in patients with relapsing-remitting multiple sclerosis. *JAMA* 313: 275–28 (2015).

[107] Nash RA, et al. High-dose immunosuppressive therapy and autologous hematopoietic cell transplantation for relapsing-remitting multiple sclerosis (HALT-MS): a 3-year Interim report. *JAMA Neurol.* 72: 159–169 (2015).

[108] Burt R, et al. Autologous nonmyeloablative hematopoietic stem cell transplantation in patients with severe anti-TNF refractory Crohn's disease: long-term follow-up. *Blood* 116: 6123–6132 (2010).

[109] Burt R, et al. Hematopoietic stem cell transplantation for progressive multiple sclerosis: failure of a total body irradiation–based conditioning regimen to prevent disease progression in patients with high disability scores. *Blood* 102: 2373–2378 (2003).

[110] Burt R, et al. Autologous hematopoietic stem cell transplantation in refractory rheumatoid arthritis. *Arthritis Rheum.* 42: 2281–2285 (1999).

[111] Snowden JA, Autologous hemopoietic stem cell transplantation in severe rheumatoid arthritis: a report from the EBMT and ABMTR. *J Rheumatol.* 31: 482–8 (2004).

[112] Verburg RJ, et al. High-dose chemotherapy and autologous hematopoietic stem cell transplantation in patients with rheumatoid arthritis. *Arthritis Rheum.* 44: 754–760 (2001).

[113] Snowden JA, et al. Haematopoietic stem cell transplantation (HSCT) in severe autoimmune diseases: analysis of UK outcomes from the British society of blood and marrow transplantation (BSBMT) data registry 1997–2009. *Br J Haematol.* 157: 742–746 (2012).

[114] Illei GG, et al. Current state and future directions of autologous hematopoietic stem cell transplantation in systemic lupus erythematosus. *Ann Rheum Dis.* 70: 2071–2074 (2011).

[115] Bink M, et al. Phase I/II trial of autologous stem cell transplantation in systemic sclerosis: procedure related mortality and impact on skin disease. *Ann Rheum Dis.* 60: 577–584 (2001).

[116] Sykes M, Nikolic B. Treatment of severe autoimmune disease by stem-cell transplantation. *Nature* 435: 620–627 (2005).

[117] Slavin S, et al. Graft vs autoimmunity following allogeneic non-myeloablative blood stem cell transplantation in a patient with chronic myelogenous leukemia and severe systemic psoriasis and psoriatic polyarthritis *Exp Hematol.* 28: 853–857 (2000).

[118] Oyama Y et al. Allogeneic stem cell transplantation for Evans syndrome. *Bone Marrow Transplant.* 28: 903–905 (2001).

[119] Marmont AM, Gualandi F, Van Lint MT, Bacigalupo A. Refractory Evans' syndrome treated with allogeneic SCT followed by DLI. Demonstration of a graft-versus-autoimmunity effect. *Bone Marrow Transplant.* 31: 399–402 (2003).

[120] Burt RK et al. Induction of remission of severe and refractory rheumatoid arthritis by allogeneic mixed chimerism. *Arthritis Rheum.* 50: 2466–2470 (2004).

[121] Loh Y, et al. Non-myeloablative allogeneic hematopoietic stem cell transplantation for severe systemic sclerosis: graft-versus-autoimmunity without graft-versus-host disease? *Bone Marrow Transplant.* 39: 435–437 (2007).

[122] Khorshid O, et al. Nonmyeloablative stem cell transplant in a patient with advanced systemic sclerosis and systemic lupus erythematosus. *J Rheumatol.* 12: 2513–2516 (2004).

[123] Daikeler T, et al. Allogeneic hematopoietic SCT for patients with autoimmune diseases. *Bone Marrow Transplant.* 44: 27–33 (2009).

[124] Eapen M, et al. Outcomes of transplantation of unrelated donor umbilical cord blood and bone marrow in children with acute leukaemia: a comparison study. *Lancet* 369: 1947–1954 (2007).

[125] Francese R, Fiorina P. Immunological and regenerative properties of cord blood stem cells. *Clin Immunol.* 136: 309–322 (2010).

[126] Brunstein CG, Weisdorf DJ. Future of cord blood for oncology uses. *Bone Marrow Transplant.* 44: 699–707 (2009).

[127] Raetz E, Beatty PG, Adams RH. Treatment of severe Evans syndrome with an allogeneic cord blood transplant. *Bone Marrow Transplant.* 20: 427–429 (1997).

[128] Itamura H, et al. Successful reduced-intensity umbilical cord blood transplant for fulminant hemophagocytic syndrome in an adult with pre-existing rheumatoid arthritis and autoimmune hemolytic anemia. *Leukemia Lymphoma* 53: 2307–2309 (2012).

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