

Marrow Donor Registry and Cord Blood Bank in Taiwan

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Abstract

Unrelated Bone marrow transplant was initiated thirty years ago. Though there are over millions of donors registered with the bone marrow registries worldwide, Asian patients rarely find a match with all these donors. Tzu Chi Marrow Donor Registry was established to meet this need. It has become the largest Asian marrow donor registry in the world. With the introduction of high technology to test the HLA of the donors and recipients, the success rate of bone marrow transplant is greatly improved among Asian countries. 50% of blood disease Asian patients who cannot find a bone marrow matched donor will be complemented by the establishment of cord blood banks in Taiwan.

1. Introduction

The first unrelated donor marrow transplants were initially performed in the early 1970?

The most common disease employing an unrelated donor transplant is chronic myeloid leukemia (CML) as a marrow or hematopoietic transplant is the only known cure for this disease. CML patients transplanted from an HLA identical sibling turned out to be very favorable with probability of survival approaching 80% at 5 years. In accordance to the reports, HLA allele matching and graft rejection in 300 unrelated donor transplants was performed for patients with chronic myeloid leukemia (CML). The overall incidence of graft rejection was 5.6%, but the incidence of rejection was only 2% when patient and donor were fully matched for HLA-A, -B, -C, -DRB1 alleles.

However, in the early 1990, the number of HLA typed volunteers were limited, the chances of matching donors were low by that time. Searching for a donor is costly, time-consuming and the matching rate is rare.

Though there are millions of data registered in the registries worldwide with the development of bone marrow transplant twenty years later, the matching rate for Asian patients is rare. The Tzu Chi Marrow Donor Registry was then established in October 1993 to help Asian patients and, to a less extend, Caucasian patients whenever possible.

2. Methods and Results

The Registry with the positive influences of the Founder of Tzu Chi Foundation - Master Cheng Yen, has recruited over 230,000 volunteer donors up to March 31, 2002. It has become the largest Oriental ethnicity bone marrow donor registry in the world. The average age of the volunteer donors in the Tzu Chi Marrow Donor Registry has been shifted from a range of 40 to 50 to a range of 29 to 30 lately and now over half of the actual marrow donors' age is between 20 and 30 years of age. As of today, 8700 patients from 16 countries across the continents of Europe, America, Africa, South America, Australia and Asia have turned to our registry in hope of allocating compatible donors. The patient groups may be subdivided by ages: 28% of which are from 10 to 19 years of ages; 26% are from 20 to 29; 21% are from 30 to 39; 10% are from 40-49 and 14% are under 10 years old. Among the transplanted patients, 130 of them suffered from CML disease; AML consists of 84 patients, ALL consists of 74 patients, SAA consists of 19 patients; MDS consists of 13 patients; and 57 of them were with other diseases.

It is well recognized that HLA molecules of the MHC function in alloantigen presentation that promote allo-immunization and eventually achieve the inevitable self defense mechanism. Unfortunately, the molecules also

pose an ever increasing obstacle of barriers for transplantation medicine including bone marrow transplants. Serological determination of HLA antigens is unreliable, especially when Caucasoid alloantisera are employed as the typing reagents, let alone crossreactivities and scarcity of certain specificities.

The Tzu Chi Immunogenetics Center of the Tzu Chi Marrow Donor Registry was set up in 1997. Using sophisticated molecular biology high resolution DNA typing methodology (PCR-SSOP technique), it correctly and unambiguously HLA-types all volunteer marrow donor. With all its quality control measures, the typing accuracy reaches virtually up to 99.99%. To ensure a true match for each patient, all patients from Southeast Asia requesting for a compatible marrow donor are required to submit patients' blood samples to the Immunogenetics Center for a confirmatory test. Eventually, we found that we had eliminated a frightening typing error rate of 10-20% of the submitting laboratories. Furthermore, we reduced the incidences of graft versus host disease (GVHD) as well as significantly improved the successful rate of bone marrow transplantation in several areas. For example, the success rate in Mainland China has been upgraded from a poor 0% to 70%. Some hospitals even reach up to 100% success rate. From November 1998 on, the average successful transplantation rate in China is over 70%. Table 1 shows a comparison of survival rate of marrow transplanted patients during the period before and after November 1998 but before December 31, 2001 with or without confirmatory HLA typing by DNA high resolution tests. Search requests for compatible marrow donors from our Registry have been booming ever since and a total change of attitude among physicians and patients in favor of marrow transplantation for the treatment of life-threatening diseases such as leukemia, lymphoma, aplastic anemia, thalassemia and metabolic abnormality has swept the whole nation.

To evaluate the outcomes of unrelated bone marrow transplantation facilitated by TCMDR, we focused on 48 marrow transplants performed in Taiwan during the period of May 1994 to June 1997. Follow-up of these 48 cases was made up to 30 May 1998. The average length of follow-up was 328 days. The actual length of follow-up ranged from 7-1385 days. All the donor/recipient pairs were HLA-A, B, and DR matched and were divided into two risk groups based on diagnosis and stage of disease. There were 24 standard risk and 24 high risk patients. Engraftment occurred in 40 patients (83.3%). Acute and chronic GVHD was observed in 80% of evaluable patients and accounts for 41% of deaths. The second leading cause of death was infection, accounting for 35% of deaths. The overall probability of survival for the 48 patients after BMT over 2 years was 33.3% (95% CI 19.67-46.32%). The probability of survival for standard risk and high risk groups was 54% and 8%, respectively. In multivariate analysis, patients who were younger, in the standard risk group, or with a shorter search interval showed a higher survival probability.

Though the Registry has recruited over 230,000 donors, there are 40% patients from Taiwan, 50% patients from South China, 75% patients from North China and 45% patients from overseas Chinese, overall an average of 50% Asian blood disease patients not be able to find a match. The Tzu Chi Foundation decided to set up Cord Blood Bank in July 1999 to help marrow unmatched patients. We adopted the most advanced Archive System to store the cord blood units. So far, there are five institutes, Koo Foundation Sun Yet-Sen, Chinese Blood Foundation, Babybanks, Tzu Chi Foundation and Geneborn Company, collecting cord blood units in Taiwan. Out of the five Institutes, only the Tzu Chi Foundation and Geneborn Company adopt BioArchive system. Other institutes adopt traditional system to preserve cord blood units.

As a matter of fact, utilization of cord blood has recently created a whole lot of zeal in the medical field as well as in the commercial arena. All this commotion is due to the facts that:

1. It is enriched with stem cells which can be used as a therapy for the treatment of blood disorder such as malignancy, Falconi? anemia, and Wiskott-Aldrich Syndrome.
2. It was found that it is naive in terms of cell differentiation and maturation that the requirement for histocompatibility is less stringent as opposed to unrelated bone marrow donor transplantation.
3. It is derived without the involvement of an invasive surgery. No general anaesthesia and no hazards of post operative infections.

However, the average volume of cord blood is very small compared with bone marrow transplantation. Therefore, it sets limitations for adult patients or patients with large body weight. Freezing of cord blood cells requires advanced technology that guarantees high viability of the frozen stem cells.

To address these two issues, recent developments aiming to enhance the effects of cord blood banking are as follows:

1. Combining infusion of two unrelated cord blood units to accommodate the insufficiency of a single unit. A successive transplantation with two unrelated placental cord blood in a 95 Kg adult was reported by Peking University People? Hospital and Institute of Hematology in a recent scientific conference. According to their report, two units of frozen unrelated cord cells with HLA disparity in -B locus were infused into a 95 Kg, 32 year old male who was diagnosed with acute lymphoblastic leukemia. Complete remission was achieved after chemotherapy with central nervous complications which were resolved after treatment. The first cord blood has a cell count of 18.7×10^8 and the second cord blood 9.6×10^8 . On day 19 his absolute neutrophil count reached $0.5 \times 10^9/L$ and WBC $5.9 \times 10^9/L$ on day 31 since the infusion. His WBC remained between $2.0-5.0 \times 10^9/L$ and did not require platelet transfusion thereafter. There was no

Table 1.

Comparison of the survival rate with or without performing confirmatory HLA tests by DNA high resolution methodology.

Survival Periods	Before November 1998 (Total: 95 Transplants)			Between November 1998 ~ December 31, 2001 (Total: 251 Transplants)		
	Death Tolls	Survivals	Survival Rate	Death Tolls	Survivals	Survival Rate
Within 3 months	30	65	68.09 %	37	214	85.26 %
3~6 months	11	54	57.45 %	32	182	72.51 %
6 months~1 year	19	35	37.23 %	30	152	60.56 %
1~2 year	2	33	35.11 %	12	14	055.78 %
2~3 year	3	30	31.91 %	0	140	55.78 %

obvious sign of GVHD. The bone marrow showed normal cellularity and regeneration of different normal cells, including megakaryocytes. Blood DNA sequential analysis according to Profile Plus system and HLA analysis were in entire agreement with cord blood unit 1. It is evident that one of the two cord blood units with the cell dosage of $1.96 \times 10^7/\text{kg}$ can reconstitute the hemopoietic system promptly together with another cord blood. The engraftment seems to be accelerated with simultaneous transplantation of another cord blood without significant side effect.

- Freezing of cord blood in liquid nitrogen for cryopreservation has been used for some time. Because of the inadvertent warming and vapor condensation process can occur many times in an openable dewar and a removable storage racks, cell viability will be greatly affected. It is vitally important to minimize these problems by freezing a specimen in liquid nitrogen vapor phase compartment above the liquid nitrogen storage dewar system and then removing the specimen to a specific address within the same dewar without opening the lid of the storage tank. To accommodate these needs, the BioArchive system is employed. It is an enclosed automatic system with robotic arm and a periscope. It can perform Bar Code reading and instruct the Canister Hook to store and retrieve cord blood samples in a double-layer, completely insul-

tated stainless steel Dewar. It has a programmed mechanism to preset parameters for selected controlled rate of freezing, for example 2°C per minute and to store blood samples automatically. It greatly improves the old traditional storage method, which is vulnerable for temperature fluctuations and known to be disadvantageous in the preservation of viable cells.

With the above mentioned advancements, however, there is a pivoted issue that must be discussed very seriously--The accuracy of HLA typing.

It is well documented that in transplantation medicine, the time of graft survival is directly proportional to the degree of matching in HLA between a graft donor and the pertaining recipient. The effect is more significant in stem cell transplantation. The adverse effects in stem cell transplantation range from graft rejection to graft versus host disease and the eventual prospect is extremely grey if HLA matching is compromised.

In conclusion, bone marrow transplant and cord blood banking has come a long way since its inception some 10-15 years ago. Inevitably, research endeavors and developmental work on cord blood banking will need to focus on the issue of volume and viability of stem cells. Simultaneously, to improve successful outcomes of such transplantation, uncompromising DNA determination of HLA alleles on cord blood stem cells is imperatively a must.