

# Allo-HKHN'de Ürün Seçimi Dondurulmuş? Taze?

Hüseyin Saffet Beköz  
Medipol Üniversitesi Tıp Fakültesi  
Erişkin Hematoloji



**14.** ULUSAL AFEREZ  
KONGRESİ  
22-23 KASIM 2019 MARRIOT ASIA HOTEL , İSTANBUL



# Hangi ürünleri donduruyoruz?

## KAYNAK

- KEMİK İLİĞİ
- PERİFERİK KAN
  - Kök hücre nakli için
  - DLI için
- **KORD KANI**

## VERİCİ

- **OTOLOG**
- SİNGENEİK
- ALLOGENEİK
  - TAM UYUMLU AKRABA
  - **AKRABA DIŞI (%2)**
  - HAPLOİDENTİK ?

# Allogeneik ürünü Neden donduruyoruz?

- Allojenik hematopoetik kök hücre nakli için donör kök hücreleri (alloHSCT) genellikle nakilden hemen önce toplanır ve alıcıya "taze" aktarılır.
- Bununla birlikte, **tıbbi nedenlerden dolayı** (ağır enfeksiyon, relaps vb)
- Donöre planlanan günde ulaşamama veya vazgeçme riski olduğunda
- Kök hücre toplanması önceden planlanmış ve **yeniden planlanamadığında**, Hazırlama rejimi ve ardından greft infüzyonunu geciktirmek gerekebilir.  
**Donör kaynaklı olabilecek riskleri azaltmak!**

# Allogeneik ürünü Neden donduruyoruz?

- Ex-vivo T hücre deplesyonu yapılan (**TCR Alpha, Beta and CD19+ Cell Depletion**) Haploidentik nakillerde kök hücre ürünü dondurulması gerekebiliyor.
- **RIC Allo** nakiller sonrası hastaya **DLI** planlaması da yapılıyor ise MUD nakil vericisi olan hastalarda ürün fazla ise bir kısmı DLI için dondurularak saklanabiliyor.
- Donör kilosunun düşük olduğu kemik iliği nakillerinde birden fazla toplama gerekliliğinde
- Bu gibi durumlarda allojenik greftler dondurulabilir ve hastalar çözülmüş ürünler alır.

# Allo-HKHN için dondurarak korunmuş kök hücre ürünlerinin, taze kök hücre ürünlerinin yerine kullanılmasının potansiyel avantajları ve dezavantajları.

## Potansiyel avantajları

- Sağlık sistemi üzerindeki stres azalır
- Donör üzerindeki azalmış stres
- Nakil işleminin 0. Gününden önce fakir mobilize olan donörlerin belirlenmesi
- Donör ölüm, kaza, vazgeçme, red vb ile kullanılamazlığı durumunda donör greftinin mevcudiyeti sağlanır.
- Kriyoprezervasyon sürecinde T hücresi alt gruplarının tercihli imhası nedeniyle GVHD insidansının azalması.

## Yorumlar

- Bağışçı toplama işlemini 0. Gün nakli ile koordine etmek zordur. Kriyoprezervasyon sisteme daha fazla esneklik kazandırır.
- Bağışçının, başka yükümlülüklerin belirlenmesinde toplama planlaması için daha fazla esneklik.
- Pleriksafor'un ümit veren mobilizasyon sonuçları, sağlıklı periferik kan kök hücre bağışçıları arasında mevcut % 2-5 zayıf mobilizasyon oranlarını azaltabilir.
- Nadir olsa da, dondurarak saklama, alıcılar arasında bu sık bildirilen korkuyu giderir.
- Sınırlı klinik ve laboratuvar çalışmalarının çoğu bu hipotezi desteklememektedir. Laboratuvar korelasyonları ile desteklenen klinik çalışmalara ihtiyaç vardır.

# Allo-HKHN için dondurarak korunmuş kök hücre ürünlerinin, taze kök hücre ürünlerinin yerine kullanılmasının potansiyel avantajları ve dezavantajları.

## Potansiyel dezavantajları

- Kriyoprezervasyon sırasında greftin zarar görmesinden dolayı nötrofil ve trombosit engrafmanında gecikme endişesi.
- Kriyoprotektan olarak DMSO varlığına bağlı olarak transfüzyon reaksiyonlarında artış görülmüştür.
- Donma / çözünme işleminde artan kullanım nedeniyle greftin bakteriyel kirlenme oranının artması.
- Asla kullanılmayan greft toplama insidansının artması, bağışçıya gereksiz bir toplama prosedürü uygulanması.

## Yorumlar

- Sınırlı yayınlanmış deneyim, trombosit veya nötrofil yamalanmasına ilişkin zaman içerisinde önemli bir gecikme olmadığını göstermektedir. Laboratuvar korelasyonları ile daha uygun şekilde çalışan daha fazla çalışmaya ihtiyaç vardır.
- Kriyoprezerveli ürünler, daha fazla transfüzyonla ilişkili bulantı, kusma ve ateş ile ilişkilidir, ancak hemodinamik instabilite (hipotansiyon, bradikardi) veya pulmoner riskler gibi daha ciddi olaylar olmaz.
- Bakteriyel kontaminasyon oranlarının yüksek olması, alıcılarda anlamlı derecede yüksek bakteriyemi ve sepsis oranlarına dönüşmez. Periferik kan greftleri daha düşük kontaminasyon insidansı ile koreledir.
- Nakil işleminin 1-3 haftası içerisinde ürün toplandığı zaman, bu kaçınılmaz sonucu en aza indirecektir.

## Use of Non-Cryopreserved Peripheral Blood Stem Cells Is Associated with Adequate Engraftment in Patients with Multiple Myeloma Undergoing an Autologous Transplant.

Kulkarni U<sup>1</sup>, Devasia AJ<sup>1</sup>, Korula A<sup>1</sup>, Fouzia NA<sup>1</sup>, Nisham PN<sup>1</sup>, Samoon YJ<sup>1</sup>, Lakshmi KM<sup>1</sup>, Abraham A<sup>1</sup>, Srivastava A<sup>1</sup>, Mathews V<sup>1</sup>, George B<sup>2</sup>.

### Author information

- 1 Department of Haematology, Christian Medical College, Vellore, India.
- 2 Department of Haematology, Christian Medical College, Vellore, India. Electronic address: bijuhaemat@gmail.com.

### Abstract

Autologous transplantation is the standard of care for transplant-eligible patients with multiple myeloma. Toward making this treatment accessible in developing countries, there are significant challenges like resource constraints and access to cryopreservation facilities. We performed a retrospective analysis of patients with multiple myeloma who underwent autologous transplantation using granulocyte colony-stimulating factor (G-CSF)-mobilized non-cryopreserved grafts at our institution from January 1995 to December 2014. Peripheral blood stem cells (PBSCs) were harvested over 1 to 2 days after G-CSF mobilization. After apheresis, PBSCs were stored at 4°C in a blood bank refrigerator for up to 72 hours. During the study period, 224 patients with multiple myeloma underwent autologous transplantation using G-CSF-mobilized non-cryopreserved grafts. The number of days of stem cell harvest was 1 in 91 patients (40.6%) and 2 in 133 patients (59.4%). The median CD34 cell dose was  $4.87 \times 10^6/\text{kg}$  (range, 1.15 to 23.7). All patients except 1 engrafted. The median time to neutrophil engraftment was 12 days (range, 9 to 22). The median time to platelet engraftment was 17 days (range, 10 to 44). In a resource-limited setting, the use of G-CSF-mobilized non-cryopreserved grafts results in adequate engraftment for most patients with multiple myeloma undergoing autologous stem cell transplantation.



## Outcomes of Fresh Peripheral Stem Cell Transplants (PSCT) compared to Cryopreserved PSCT for the Treatment of Multiple Myeloma

Laine Fiscina,<sup>1,2</sup> Alessandro Moura,<sup>1,2</sup> Suellen Riccio,<sup>2</sup>  
Hugo Carvalho\*,<sup>2</sup> Luciene Oliveira,<sup>2</sup>  
Daniela Dourado,<sup>1,2</sup> Herbert Henrique Santos,<sup>1</sup>  
Bruna Gotardo Salvino,<sup>2</sup> Marcos Chaves,<sup>1</sup>  
Tiago Freitas,<sup>2</sup> Gloria Bomfim Arruda,<sup>1</sup>  
Edvan Crusoé,<sup>1</sup> Marco Aurélio Salvino<sup>1,2</sup>

<sup>1</sup>Universidade Federal da Bahia, Salvador, Bahia, Brazil; <sup>2</sup>Hospital São Rafael, Salvador, Bahia, Brazil

toxicity. **Objective:** To evaluate the time of neutrophil grafting in patients with MM who underwent PSCT with infusion of non-cryopreserved stem cells compared to those who underwent cryopreserved stem cell infusion. **Design:** A historical cohort of MM patients who underwent autologous PSCT, from 2010 and 2017, at a single center (BMT Center of Universidade Federal da Bahia) was carried out. We compared the outcomes of 100 patients submitted to Autologous PSCT using fresh or cryopreserved cells. The fresh Protocol follows Brazilian laws, that allow the infusion of PSC within 48hours after apheresis (D-2 apherese, D-1 Melphalan, D0

infusion). **Setting:** Single center (BMT Center of Universidade Federal da Bahia). **Results:** Of the 100 patients included, 60 received cryopreserved PSC and 40 received fresh (non-cryopreserved cells). Neutrophil grafting was obtained with a median of 11 days in the cryopreserved group and 10 days in the fresh infusion group ( $p < 0.001$ ). There was infusional related acute adverse effect (AE) during the procedure in 5 patients (8.3%) of the cryopreserved group, whereas in the non-cryopreserved group there was only 1 case of intercurrent (2.5%) ( $p < 0.05$ ). There was no difference in the frequency of febrile neutropenia between the two groups (81.7% vs 72.5%), retrospectively for cryopreserved and fresh infusion groups). However, the frequency of mucositis was higher in the first subgroup - 86.6% versus 62.5% ( $p < 0.05$ ). **Conclusions:** In our series autologous fresh PSCT is feasible and safe, with superiority when considering the neutrophil time to recovery. It is a more cost-effective method, especially in resource-limited country. **Keywords:** Multiple Myeloma, Stem Cell Transplants, non-cryopreserved cells





## Is long term storage of cryopreserved stem cells for hematopoietic stem cell transplantation a worthwhile exercise in developing countries?

Santhosh Kumar Devadas<sup>1</sup>, Minal Khairnar<sup>2</sup>, Sumathi S Hiregoudar<sup>2</sup>, Shashank Ojha<sup>2</sup>, Sachin Punatar<sup>1</sup>, Alok gupta<sup>1</sup>, Anant Gokarn<sup>1</sup>, Pallavi Bhole<sup>3</sup>, Sadhana Kannan<sup>3</sup>, Navin Khattri<sup>1</sup>

<sup>1</sup>Department of Medical Oncology and Bone Marrow Transplantation, <sup>2</sup>Department of Transfusion Medicine, <sup>3</sup>Statistics, Tata Memorial Center, Advanced Center for Treatment, Research and Education in Cancer (ACTREC), Mumbai, India

**Gelişmekte olan ülkelerde hematopoetik kök hücre nakli için dondurularak saklanan kök hücrelerin uzun süreli depolanması faydalı bir egzersiz midir?**

Bir üçüncü basamak kanser tedavi merkezinde,  
Kasım 2007 - Ocak 2015 tarihleri arasında  
Otolog kök hücre transplantasyonu (oto-SCT) (N = 239) veya  
Allojenik kök hücre transplantasyonu (allo-SCT) (N = 196) planlayan **435 hastada**  
retrospektif bir SCU kullanımı denetimi yapılmış.  
Haziran 2016'da yapılan analiz sırasında, dondurulmuş kök hücrelerin depolandığı  
**medyan süre 4.1 yıldır (aralık, 1.34–8.4 yıl).**

## Gelişmekte olan ülkelerde hematopoetik kök hücre nakli için dondurularak saklanan kök hücrelerin uzun süreli depolanması faydalı bir egzersiz midir?

**Table 1.** Details of stem cell units stored for autologous stem cell transplantation.

|  | HL         | NHL        | MM         | AML       | NB        | Other     | Total      |
|--|------------|------------|------------|-----------|-----------|-----------|------------|
| N (%)  | 94 (39.3)  | 42 (17.5)  | 75 (31.3)  | 10 (4.1)  | 11 (4.6)  | 7 (2.9)   | 239        |
| Stored SCUs                                    | 669        | 355        | 522        | 64        | 72        | 46        | 1,728      |
| Mean SCU stored/patient                        | 7.1        | 8.4        | 7.0        | 6.4       | 6.5       | 6.5       | 7.2        |
| SCUs reinfused                                 | 427        | 249        | 251        | 22        | 44        | 31        | 1,024      |
| Ratio of infused/stored SCUs                   | 0.6        | 0.7        | 0.5        | 0.3       | 0.6       | 0.7       | 0.6        |
| Patients receiving all SCUs (%)                | 35 (37.2)  | 20 (47.6)  | 4 (5.3)    | 2 (20.0)  | 4 (36.3)  | 3 (42.5)  | 68 (28.5)  |
| Patients partially receiving stored SCUs (%)   | 50 (53.1)  | 16 (38.1)  | 65 (86.6)  | 2 (20.0)  | 6 (54.5)  | 2 (28.5)  | 141 (58.9) |
| Patients who never received any stored SCU (%) | 9 (9.6)    | 6 (14.2)   | 6 (8.0)    | 6 (60.0)  | 1 (9.0)   | 2 (28.5)  | 30 (12.6)  |
| SCUs still stored (%)                          | 242 (36.2) | 106 (29.9) | 271 (51.9) | 42 (65.6) | 28 (38.9) | 15 (32.6) | 704 (40.7) |

Abbreviations: AML, acute myeloid leukemia; HL, Hodgkin's lymphoma; MM, multiple myeloma; NB, neuroblastoma; NHL, Non-Hodgkin's lymphoma; SCU, stem cell unit; Other, other solid tumors.

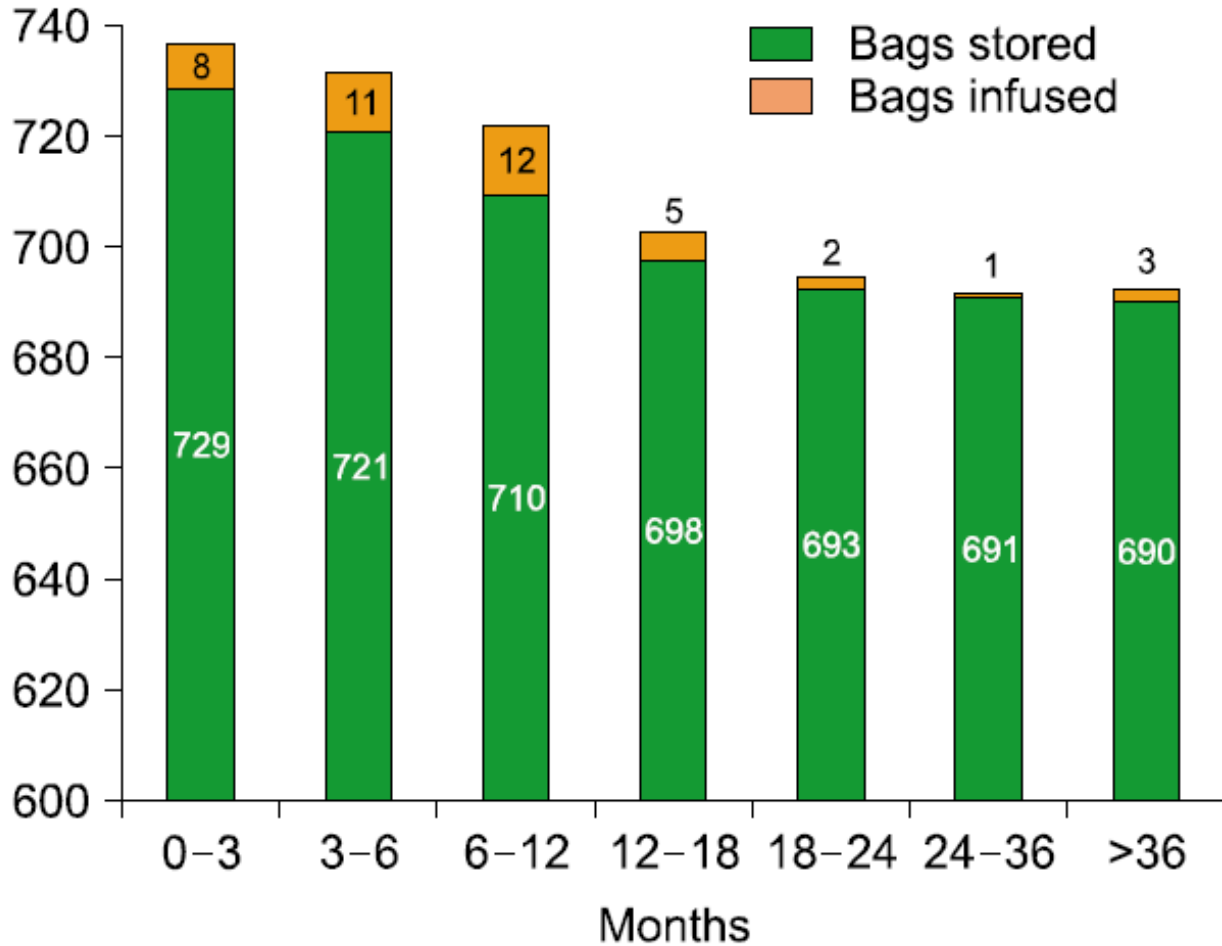
## Gelişmekte olan ülkelerde hematopoetik kök hücre nakli için dondurularak saklanan kök hücrelerin uzun süreli depolanması faydalı bir egzersiz midir?

**Table 2.** Use of SCUs stored for donor lymphocyte infusion.

|                                      | ALL        | AML        | AA        | CML       | MDS       | HL        | Others    | Total      |
|--------------------------------------|------------|------------|-----------|-----------|-----------|-----------|-----------|------------|
| Patients in whom SCUs stored for DLI | 37 (22.4)  | 71 (43.0)  | 16 (9.6)  | 20 (12.1) | 10 (6.1)  | 3 (1.8)   | 8 (4.8)   | 165        |
| Patients whose SCUs were not stored  | 9          | 8          | 7         | 1         | 4         | 2         | 0         | 31         |
| Stored SCUs                          | 169        | 321        | 61        | 94        | 42        | 13        | 29        | 729        |
| Mean stored SCU/patient              | 4.5        | 4.5        | 3.8       | 4.7       | 4.2       | 4.3       | 3.6       | 4.4        |
| SCUs reinfused (%)                   | 9 (5.3)    | 25 (7.7)   | 3 (4.9)   | 0 (0.0)   | 1 (2.3)   | 2 (15.3)  | 2 (6.8)   | 42 (5.7)   |
| SCUs still stored (%)                | 160 (94.6) | 296 (92.3) | 58 (95.0) | 94 (100)  | 41 (97.7) | 11 (84.6) | 27 (93.2) | 687 (94.3) |

Abbreviations: AA, aplastic anemia; ALL, acute lymphoblastic leukemia; AML, acute myeloid leukemia; CML, chronic myeloid leukemia; DLI, donor lymphocyte infusion; HL, Hodgkin's lymphoma; MDS, myelodysplastic syndrome; SCU, stem cell unit.

## Gelişmekte olan ülkelerde hematopoetik kök hücre nakli için dondurularak saklanan kök hücrelerin uzun süreli depolanması faydalı bir egzersiz midir?



### Kullanılmayan SCU'nun saklama maliyeti

- Auto-SCT için kullanılmayan 704 SCU'nun toplam dondurma maliyeti 70.400 dolar,
- DLI için 687 kullanılmamış SCU'nun dondurma maliyeti 43.100 dolar.
- Gerekli elektrik, bakım ve alan maliyeti dahil değil.

# Medipol Üniversitesi Deneyimi

- Pediatrik KİT ünitesinde:
- 23 VAKANIN VERİCİ KÖK HÜCRESİ DONDURULDU
  - 19 vakanın allo donörünün kemik iliği 4 vakanın PKH si dondurulmuş.
    - 19 vakanın yaşı ve kilosu küçük olması
    - 3 ü yabancı uyruku olması nedeni ile zamanlama,
    - 1 tanesi naklin 1 ay ertelenmesi
- Erişkin KİT ünitesinde:
- 13 VAKANIN VERİCİ KÖK HÜCRESİ DONDURULDU
  - 6 vakanın yaşı ve kilosu küçük olması
  - 3 vakada vericinin vazgeçme riskinin yüksek olması
  - 3 vakanın nakil zamanlamasının vericiye uymaması nedeni ile
  - 1 vakanın (aktif hastalık) Hazırlama rejimi esnasında sepsis gelişmesi



# DONDURMA PROTOKOLÜ

- Kriyoprotektan : **%7.5 DMSO + %6 HES** veya **%7.5 DMSO + Otolog plazma**
- **1 ml de 100.000 den fazla WBC olmayacak şekilde ürün + kriyoprotektan**
- Planer kontrollü kademeli dondurucuda
- - 196 °C Sıvı nitrojen buhar fazında
- Dondurma eritme arası 1 ay ile 4 ay ortalama 45 gün
- Eritildikten sonra canlılık 7 AAD kullanarak akım sitometri ile
- **Ortalama %92 ( %82- %98)**

# Taze ve Donmuş allojenik kök hücre ürünleri göz önüne alındığında, iki konu çok önemlidir;

- (1) dondurma / çözme işlemine atfedilebilecek greftlerin hücresel içeriğinde tespit edilebilir bir fark var mı?
- (2) greft içeriğindeki bu farklılıklar
  - GvHD insidansı,
  - nüks oranları,
  - engrafman zamanı ve
  - sağkalım gibi klinik olarak önemli son noktalarla ilişkili midir?

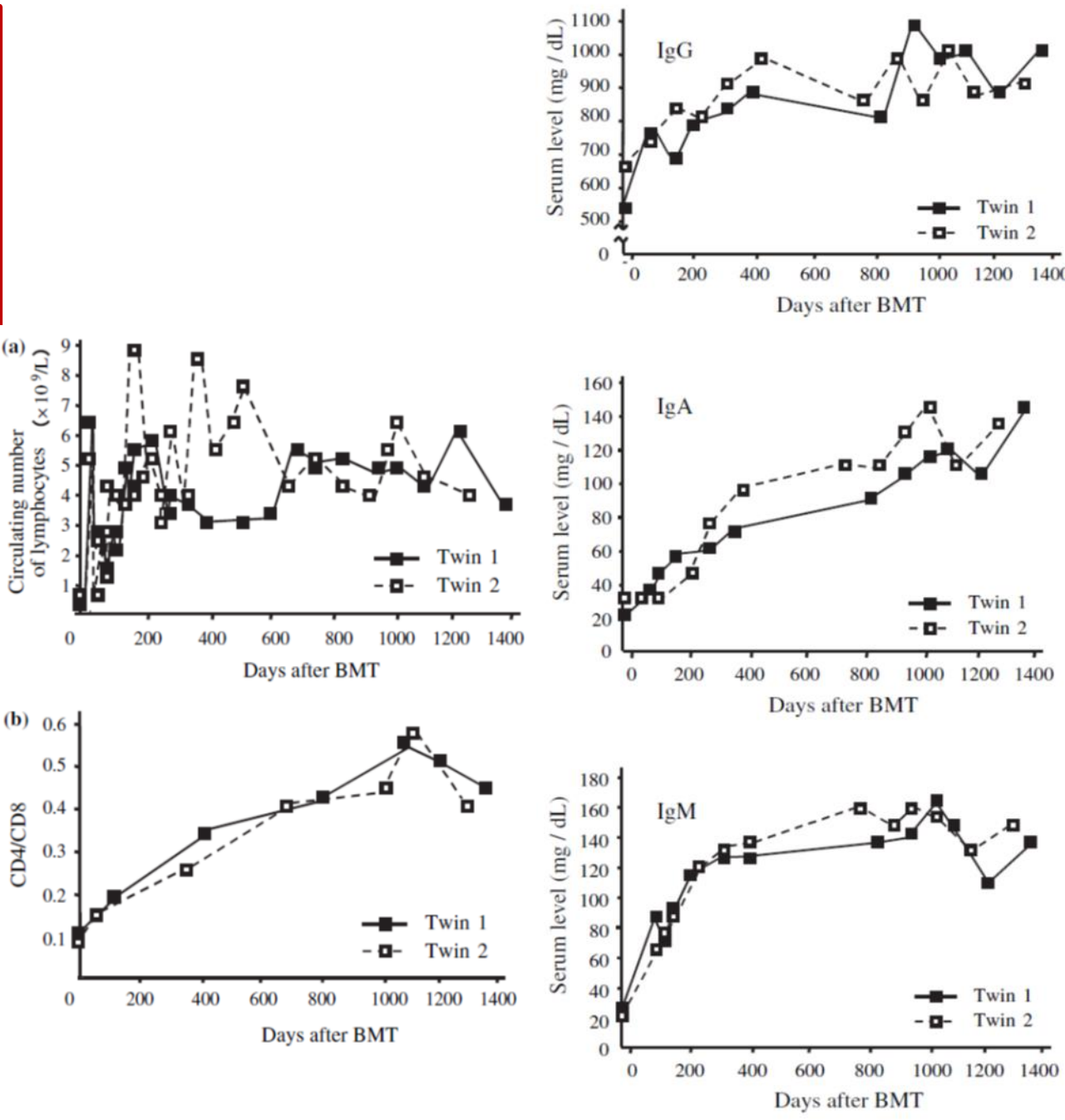
**Case Report**

Identical reconstitution after bone marrow transplantation in twins who received fresh and cryopreserved grafts harvested at the same time from their older brother

İkiz 1: 8 aylık AML B Rh (+)  
İkiz 2: 9 aylık AML B Rh (+)  
Donör 4 yaşında HLA tam uyumlu erkek kardeş. O Rh +  
Kemik iliği toplanıyor. Ürün iki eşit parçaya bölünüyor  
İkiz 1'e taze, İkiz 2 ye dondurulmuş ürün ile nakil.  
**Engrafman günleri benzer.**  
**İkiz 1: grad I aGvHD, cGvHD yok**  
**İkiz 2: grad II a GvHD (hafif Gıs ile Deri), cGvHD yok**  
**Naklin 50 ve 52. aylarında remisyonda**  
**İki hastada lenfositlerin ve plazma Ig rekonstrüksiyonu benzer.**

Table 1. Days to achieve engraftment level of each blood component

|        | Leukocytes<br>( $\times 10^9/L$ ) |           | Platelets<br>( $\times 10^9/L$ ) |     |      | Reticu-<br>locytes<br>(%) |    | Last days of<br>transfusions |          |
|--------|-----------------------------------|-----------|----------------------------------|-----|------|---------------------------|----|------------------------------|----------|
|        | WBC > 1.0                         | ANC > 0.5 | >20                              | >50 | >100 | >1                        | >2 | RBC                          | Platelet |
| Twin 1 | 14                                | 15        | 5                                | 2   | 24   | 14                        | 1  | 6                            | 13       |
| Twin 2 | 12                                | 12        | 5                                | 2   | 28   | 15                        | 1  | 10                           | 10       |



## REVIEW

**Has allogeneic stem cell cryopreservation been given the ‘cold shoulder’?**  
**An analysis of the pros and cons of using frozen versus fresh stem cell products in allogeneic stem cell transplantation**

**Table 1** Summary of engraftment and outcome data for selected reports of allogeneic stem cell transplantation using cryopreserved stem cells

| <i>Reference</i>                         | <i>Storage</i> | <i>No. of patients</i> | <i>Donor</i> | <i>HSC source</i> | <i>Days to ANC &gt; 0.5</i> | <i>Days to Plt engraftment</i> | <i>aGvHD ≥ gr II</i> | <i>Day 100 survival</i> |
|--|----------------|------------------------|--------------|-------------------|-----------------------------|--------------------------------|----------------------|-------------------------|
| Stockschrader <i>et al.</i> <sup>4</sup> | Frozen         | 40                     | Related      | BM                | 17                          | 21(plt > 20)                   | 61%                  | 82%                     |
|  | Fresh          | 40                     | Related      | BM                | 17.5                        | 20                             | 60%                  | 72%                     |
| Stockschrader <i>et al.</i> <sup>6</sup> | Frozen         | 10                     | MUD          | BM                | 22.6                        | NA                             | 75%                  | 55%                     |
|  | Fresh          | NA                     | NA           | NA                | NA                          | NA                             | NA                   | NA                      |
| Eckardt <i>et al.</i> <sup>2</sup>       | Frozen         | 10                     | Related      | BM                | 19                          | 28(plt > 50)                   | 20% <sup>a</sup>     | 70%                     |
|  | Fresh          | 33                     | Related      | BM                | 16                          | 23                             | 57.5%                | 61%                     |
| Lasky <i>et al.</i> <sup>3</sup>         | Frozen         | 6                      | Related      | BM                | 21.3                        | NA                             | NA                   | 50%                     |
|  | Fresh          | NA                     | NA           | NA                | NA                          | NA                             |                      |                         |

Abbreviation: NA = Not available; HSC = hematopoietic stem cell source; ANC = absolute neutrophil count; Plt = platelet; aGVHD = acute graft-versus-host disease; MUD = matched unrelated donor.

<sup>a</sup>Statistically significant difference.

Kesin insidansı bilinmemekle birlikte, NMDP (R King; NMDP, kişisel iletişim) tarafından bildirilen **akraba dışı nakillerin toplam sayısının % 2'sini oluşturur.**

## REVIEW

### Has allogeneic stem cell cryopreservation been given the ‘cold shoulder’? An analysis of the pros and cons of using frozen versus fresh stem cell products in allogeneic stem cell transplantation

**Table 2** Summary of comparative graft contents in selected reports of allogeneic stem cell transplantation using fresh versus cryopreserved cells

| <i>Reference</i>                                      | <i>Storage time<br/>cryopreserved grafts</i>                | <i>Analysis limited to</i>   | <i>Results</i>   |
|---|---|--|--|
| Stockschrader <i>et al.</i> <sup>4</sup>              | Med: 17.5d<br>Range: 3–455d<br>Cryoprotectant:<br>DMSO, 10% | Post-thaw graft content<br>Fresh donor graft content                       | No s.d. between fresh and frozen graft content<br>Post-thaw: CFU-GM (median) = $0.83 \times 10^5/\text{kg}$  |
| <sup>a</sup> Stockschrader <i>et al.</i> <sup>5</sup> | Med: 21d<br>Range: 4–455d<br>Cryoprotectant:<br>DMSO, 10%   | Pre-frozen graft content<br>Post-thaw graft content<br>Fresh graft content | No s.d. between pre-frozen and post-thaw CFU-GM/ $10^5$ MNC<br>No s.d. between infused CFU-GM and MNC for fresh and frozen stem cell recipients<br>Post-thaw MNC (mean) = $0.69 \times 10^8/\text{kg}$<br>Post-thaw CFU-GM (mean) = $0.92 \times 10^5/\text{kg}$ |
| Stockschrader <i>et al.</i> <sup>6</sup>              | Med: 38d<br>Range: 7–139d<br>Cryoprotectant:<br>DMSO, 10%   | Pre-frozen graft content<br>Post-thaw graft content                        | No s.d. between pre-frozen and post-thaw CFU-GM/MNC $\times 10^5$<br>Post-thaw MNC (median) = $1.05 \times 10^8/\text{kg}$<br>Post-thaw CFU-GM (median) = $4.69 \times 10^4/\text{kg}$   |
| Eckardt <i>et al.</i> <sup>2</sup>                    | Med: 10.5d<br>Range: 1–175d<br>Cryoprotectant:<br>DMSO, 10% | Pre-frozen graft content<br>Post-thaw graft content<br>Fresh graft content | No s.d. between pre-frozen and post-thaw CFU cells<br>A small but significant reduction in numbers of frozen MNC versus fresh MNC infused<br>Post-thaw MNC (mean) = $2.67 \times 10^8$   |
| Lasky <i>et al.</i> <sup>3</sup>                      | Med: 21d<br>Range: 6–49d<br>Cryoprotectant:<br>DMSO, 10%    | Pre-frozen graft content<br>Post-thaw graft content<br>Fresh graft content | A small but significant reduction in the CFU% post-thaw<br>Post-thaw MNC (mean) = $3.0 \times 10^8$<br>Post-thaw CFU-GM (mean) = $0.79 \times 10^4$  |



# Long-term follow-up of leukaemia patients after related cryopreserved allogeneic bone marrow transplantation

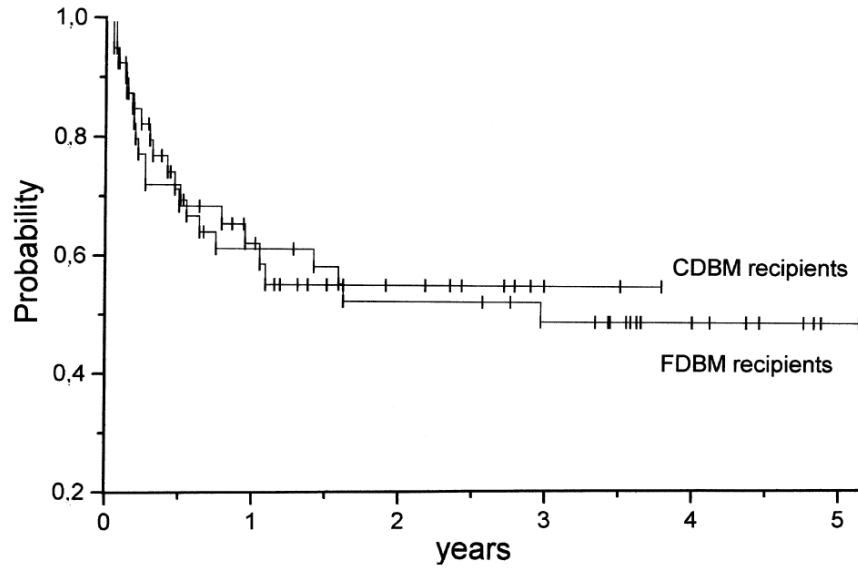
MARCUS STOCKSCHLÄDER,<sup>1</sup> HASSAN T. HASSAN,<sup>1</sup> CORNELIA KROG,<sup>1</sup> WILLIAM KRÜGER,<sup>1</sup> CORNELIUS LÖLIGER,<sup>3</sup> MARTIN HORSTMAN,<sup>2</sup> M. ALTNÖDER,<sup>1</sup> JOHANNES CLAUSEN,<sup>1</sup> JAN GRIMM,<sup>1</sup> HARTMUT KABISCH<sup>2</sup> AND AXEL ZANDER<sup>1</sup>  
<sup>1</sup>Bone Marrow Transplantation, <sup>2</sup>Department of Paediatric Oncology, and <sup>3</sup>Department of Transfusion Medicine, University Hospital Eppendorf, Hamburg, Germany

|   | Cryopreserved<br>BM | Fresh<br>BM | P* |
|---|---------------------|-------------|----|
| BFU-E transplanted ( $\times 10^5/\text{kg}$ )        |                     |             |    |
| Median  | 0.82                | 0.66        | NS |
| Range   | 0.22–6.03           | 0.01–3.20   |    |
| CFU-GM transplanted ( $\times 10^5/\text{kg}$ )       |                     |             |    |
| Median  | 0.83                | 0.72        | NS |
| Range   | 0.14–5.75           | 0.21–3.11   |    |
| ANC $> 0.5 \times 10^9/\text{l}$ (median)             | 17                  | 17.5        | NS |
| Range (d)   | 11–24               | 10–28       |    |
| ANC $> 0.5 \times 10^9/\text{l}$ (before day 15)      | 31%                 | 29%         | NS |
| ANC $> 0.5 \times 10^9/\text{l}$ (before day 21)      | 82%                 | 82%         | NS |
| Platelets $> 20 \times 10^9/\text{l}$ (median)        | 21                  | 22          | NS |
| Range (d)   | 11–85               | 13–69       |    |
| Platelets $> 20 \times 10^9/\text{l}$ (before day 20) | 52%                 | 48%         | NS |
| Platelets $> 20 \times 10^9/\text{l}$ (before day 50) | 87%                 | 94%         | NS |

|                           | Cryopreserved<br>BM (n = 40) | Fresh<br>BM (n = 40) | P  |
|---------------------------|------------------------------|----------------------|----|
| Diagnosis                 |                              |                      |    |
| AML                       | 18                           | 18                   | NS |
| First CR                  | 16                           | 12                   | NS |
| After first CR/refractory | 2                            | 4                    | NS |
| ALL                       | 10                           | 10                   | NS |
| First CR                  | 8                            | 9                    | NS |
| After first CR            | 2                            | 1                    | NS |
| CML                       | 12                           | 12                   | NS |
| Chronic phase             | 10                           | 11                   | NS |
| Accelerated phase         | 2                            | 1                    | NS |
| Age (years)               |                              |                      |    |
| Median                    | 33                           | 28                   | NS |
| Range                     | 4–58                         | 4–53                 |    |
| Conditioning regimens     |                              |                      |    |
| TBI                       | 30%                          | 25%                  | NS |
| Busulphan                 | 70%                          | 75%                  | NS |
| VP16                      | 73%                          | 78%                  | NS |
| GvHD                      |                              |                      |    |
| GvHD                      | 74%                          | 73%                  | NS |
| GvHD $\geq$ grade II      | 61%                          | 60%                  | NS |
| GvHD grade III–IV         | 24%                          | 28%                  | NS |
| Survival                  |                              |                      |    |
| Day 100 survival          | 82%                          | 72%                  | NS |

# Long-term follow-up of leukaemia patients after related cryopreserved allogeneic bone marrow transplantation

MARCUS STOCKSCHLÄDER,<sup>1</sup> HASSAN T. HASSAN,<sup>1</sup> CORNELIA KROG,<sup>1</sup> WILLIAM KRÜGER,<sup>1</sup> CORNELIUS LÖLIGER,<sup>3</sup> MARTIN HORSTMAN,<sup>2</sup> M. ALTNÖDER,<sup>1</sup> JOHANNES CLAUSEN,<sup>1</sup> JAN GRIMM,<sup>1</sup> HARTMUT KABISCH<sup>2</sup> AND AXEL ZANDER<sup>1</sup>  
<sup>1</sup>Bone Marrow Transplantation, <sup>2</sup>Department of Paediatric Oncology, and <sup>3</sup>Department of Transfusion Medicine, University Hospital Eppendorf, Hamburg, Germany



**Dondurulmuş ve taze kemik iliği nakli:  
 Engrafman süresi,  
 aGvHD, cGvHD,  
 Kimerizm,  
 BFU-E, CFU-GM,  
 100 günlük ve uzun dönem yaşam sonuçlarında istatistikî fark yok.**

|                              | Cryopreserved<br>BM (n = 40) | Fresh<br>BM (n = 40) | P  |
|------------------------------|------------------------------|----------------------|----|
| <b>Diagnosis</b>             |                              |                      |    |
| AML                          | 18                           | 18                   | NS |
| First CR                     | 16                           | 12                   | NS |
| After first CR/refractory    | 2                            | 4                    | NS |
| ALL                          | 10                           | 10                   | NS |
| First CR                     | 8                            | 9                    | NS |
| After first CR               | 2                            | 1                    | NS |
| CML                          | 12                           | 12                   | NS |
| Chronic phase                | 10                           | 11                   | NS |
| Accelerated phase            | 2                            | 1                    | NS |
| <b>Age (years)</b>           |                              |                      |    |
| Median                       | 33                           | 28                   | NS |
| Range                        | 4–58                         | 4–53                 |    |
| <b>Conditioning regimens</b> |                              |                      |    |
| TBI                          | 30%                          | 25%                  | NS |
| Busulphan                    | 70%                          | 75%                  | NS |
| VP16                         | 73%                          | 78%                  | NS |
| <b>GvHD</b>                  |                              |                      |    |
| GvHD                         | 74%                          | 73%                  | NS |
| GvHD ≥ grade II              | 61%                          | 60%                  | NS |
| GvHD grade III–IV            | 24%                          | 28%                  | NS |
| <b>Survival</b>              |                              |                      |    |
| Day 100 survival             | 82%                          | 72%                  | NS |

# Similar Outcomes of Cryopreserved Allogeneic Peripheral Stem Cell Transplants (PBSCT) Compared to Fresh Allografts

*Dong Hwan Kim,<sup>1,2</sup> Nazir Jamal,<sup>1</sup> Ronnie Saragosa,<sup>1</sup> David Loach,<sup>1</sup> Janice Wright,<sup>1</sup> Vikas Gupta,<sup>1</sup> John Kuruvilla,<sup>1</sup> Jeffrey H. Lipton,<sup>1</sup> Mark Minden,<sup>1</sup> Hans A. Messner<sup>1</sup>*

**AMAÇ:** Non randomize prospektif Çalışma  
(2003-2005 dondurulmuş – 2001-2003 taze historik kontrol)

- Dondurulmuş ile taze PBSC'nin tarihi kontrol serisi ile **allogreftin güvenlik ve klinik sonuçlarını** karşılaştırmak
- Dondurulmuş **hematopoetik progenitörlerin sürvileri üzerine kriyoprezervasyonun etkisinin** değerlendirmesi
- Transplantasyon sonrası **lenfosit iyileşmesinin yanı sıra yamalanmaya katkılarını** incelemek

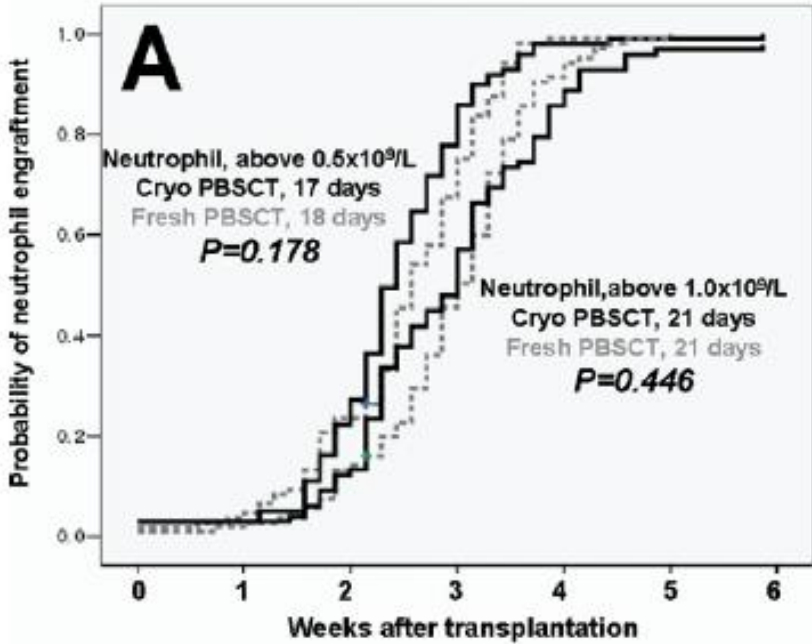
**Table 1.** *Properties of Patients, Donors, Grafts, and Transplant Procedures for Both Cohorts Receiving Either Cryopreserved or Freshly Procured PBSCT*

|   | 2003-2005                     | 2001-2003             | P-Value |
|---|-------------------------------|-----------------------|---------|
|   | Cryopreserved PBSCT (n = 105) | Fresh PBSCT (n = 106) |         |
| <b>Recipients</b>                                 |                               |                       |         |
| Sex (F/M)   | 48/57 (46%/54%)               | 46/69 (43%/57%)       | NS      |
| Age (years)                                       | 51 (17-71)                    | 49 (21-69)            | NS      |
| Weight (kg, mean $\pm$ SE)                        | 73.5 $\pm$ 1.4                | 71.2 $\pm$ 1.9        |         |
| <b>Diseases</b>                                   |                               |                       | NS      |
| AML/ALL   | 42/7 (40%/7%)                 | 44/14 (42%/13%)       |         |
| MDS/MF  | 7/9 (7%/8%)                   | 9/1 (8%/1%)           |         |
| CML/CLL   | 9/15 (8%/14%)                 | 15/7 (14%/7%)         |         |
| NHL/HD  | 13/1 (12%/1%)                 | 12/1 (11%/1%)         |         |
| MM/solid  | 2/0 (2%/0%)                   | 1/2 (1%/2%)           |         |
| <b>Donors</b>                                     |                               |                       |         |
| Sex (F/M)   | 50/55 (48%/52%)               | 65/41 (61%/39%)       | NS      |
| Age (years)                                       | 47 (12-75)                    | 48.5 (13-73)          | NS      |
| Weight (kg, mean $\pm$ SE)                        | 74.5 $\pm$ 1.8                | 73.9 $\pm$ 2.0        | NS      |
| <b>CMV status (R/D, n = 193)</b>                  |                               |                       | NS      |
| (pos/pos)/(pos/neg)                               | 44/11 (44%/11%)               | 41/10 (44%/11%)       |         |
| (neg/pos)/(neg/neg)                               | 20/25 (20%/25%)               | 19/23 (20%/25%)       |         |
| <b>PBSC collection</b>                            |                               |                       |         |
| 1-/2-/ $\geq$ 3 times                             | 90/12/3 (86%/11%/3%)          | 79/24/3 (75%/23%/3%)  | NS      |
| <b>Transplant CD34<sup>+</sup> cell dose</b>      |                               |                       |         |
| CD34 <sup>+</sup> cells ( $\times 10^6$ /kg)      | 4.8 (2.3-14.8)                | 5.2 (1.7-13.4)        |         |
| CD34 <sup>+</sup> cells, $\geq 5 \times 10^6$ /kg | 41 (45%)                      | 43 (54%)              | NS      |

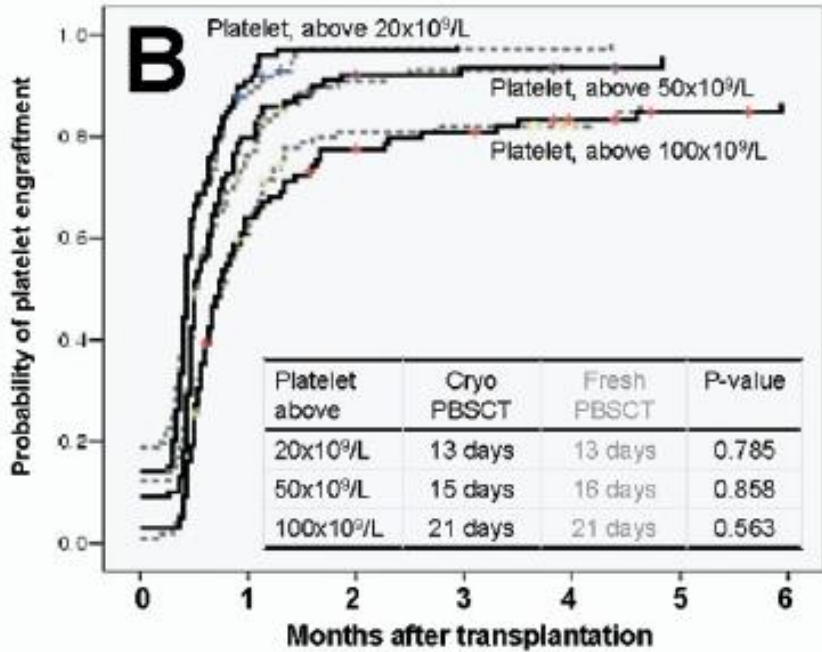
|   | Cryopreserved PBSCT (n = 105) | Fresh PBSCT (n = 106) | P-Value |
|---|-------------------------------|-----------------------|---------|
| <b>CFUs, infused (<math>\times 10^4/\text{kg}</math>)</b> |                               |                       |         |
| <b>CFU-GM</b>   |                               |                       |         |
| (mean $\pm$ SE)   | 134.87 $\pm$ 8.44             | 132.86 $\pm$ 16.12    | NS      |
| (median, range)   | 118.67 (8.95-451.75)          | 108.76 (5.12-1111.81) |         |
| <b>BFU-E</b>  |                               |                       |         |
| (mean $\pm$ SE)   | 141.19 $\pm$ 7.73             | 126.38 $\pm$ 12.53    | NS      |
| (median, range)   | 134.02 (4.41-350.70)          | 112.56 (1.40-942.18)  |         |
| <b>CFU-MEG</b>  |                               |                       |         |
| (mean $\pm$ SE)   | 24.11 $\pm$ 2.27              | 42.28 $\pm$ 6.37      | .009    |
| (median, range)   | 19.68 (0-86.16)               | 32.75 (0.47-475.70)   |         |
| <b>CFU-GEMM</b>   |                               |                       |         |
| (mean $\pm$ SE)   | 8.77 $\pm$ 0.46               | 4.63 $\pm$ 0.35       | <.001   |
| (median, range)   | 8.39 (0-21.04)                | 2.65 (0.47-20.28)     |         |
| <b>Conditioning regimen</b>                               |                               |                       | NS      |
| <b>Myeloablative</b>                                      | 74 (70%)                      | 79 (74%)              |         |
| <b>TBI based/non-TBI based</b>                            | 47/27 (45%/26%)               | 65/14 (61%/13%)       |         |
| <b>Reduced intensity</b>                                  | 31 (30%)                      | 27 (26%)              |         |
| <b>GVHD prophylaxis</b>                                   |                               |                       | NS      |
| <b>CSA/MTX</b>  | 62 (59%)                      | 74 (70%)              |         |
| <b>CSA/MMF</b>  | 31 (29%)                      | 26 (24%)              |         |
| <b>Others*</b>  | 12 (12%)                      | 5 (5%)                |         |
| <b>Type of PBSCT</b>                                      |                               |                       | NS      |
| <b>Syngeneic</b>  | 3 (3%)                        | 1 (1%)                |         |
| <b>Allogeneic sibling</b>                                 | 98 (93%)                      | 94 (89%)              |         |
| <b>Allogeneic family</b>                                  | 4 (4%)                        | 11 (10%)              |         |
| <b>HLA-disparity</b>                                      |                               |                       | NS      |
| <b>HLA-identical</b>                                      | 95 (91%)                      | 93 (88%)              |         |
| <b>I-antigen mismatched</b>                               | 10 (9%)                       | 13 (12%)              |         |



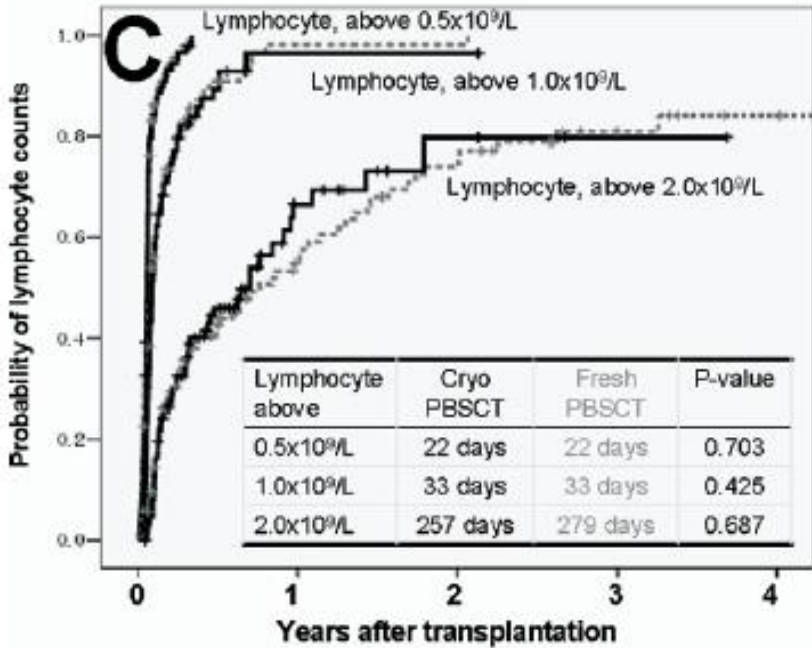
Nötrofil  
engrafmanı



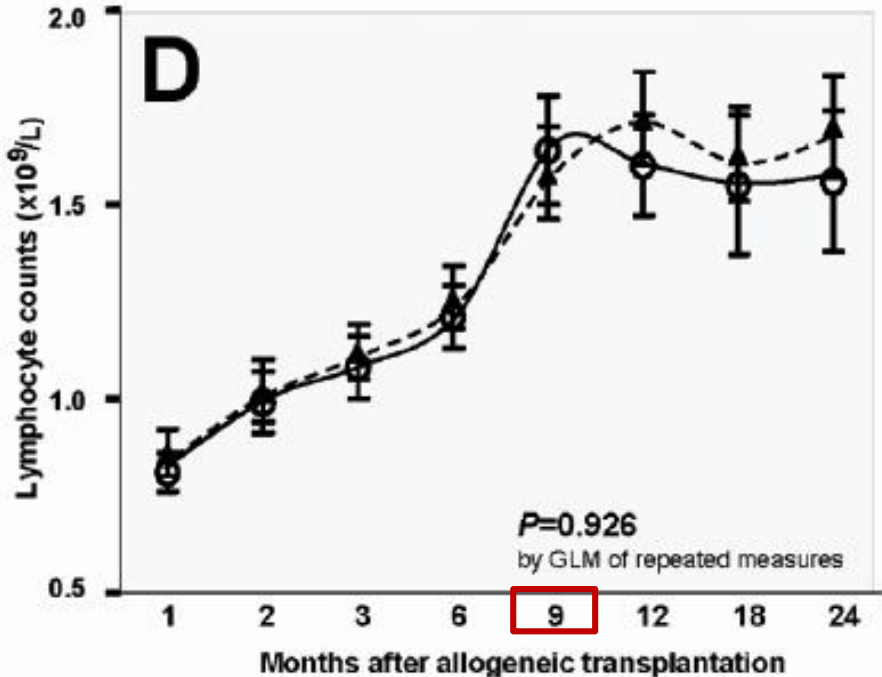
Trombosit  
engrafmanı



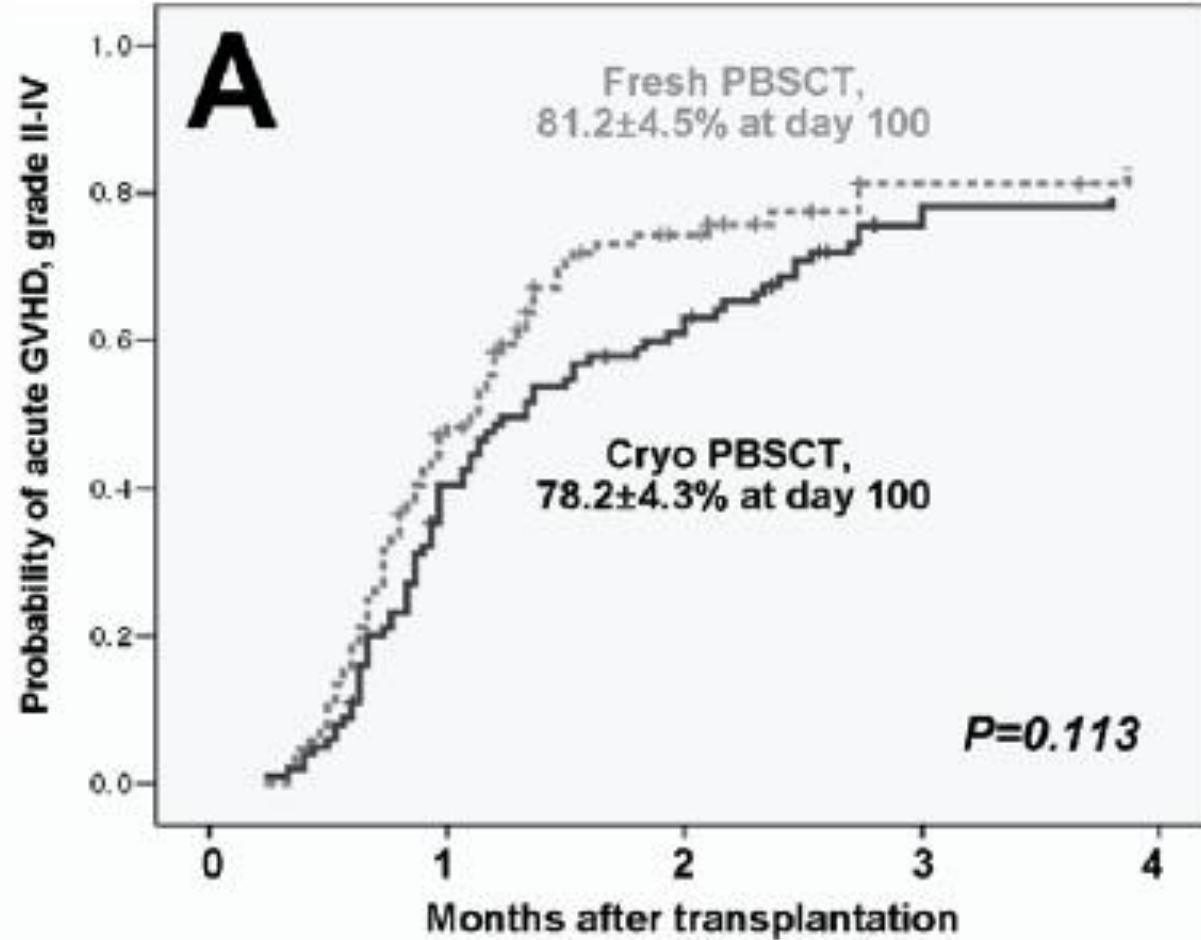
Lenfosit  
engrafmanı



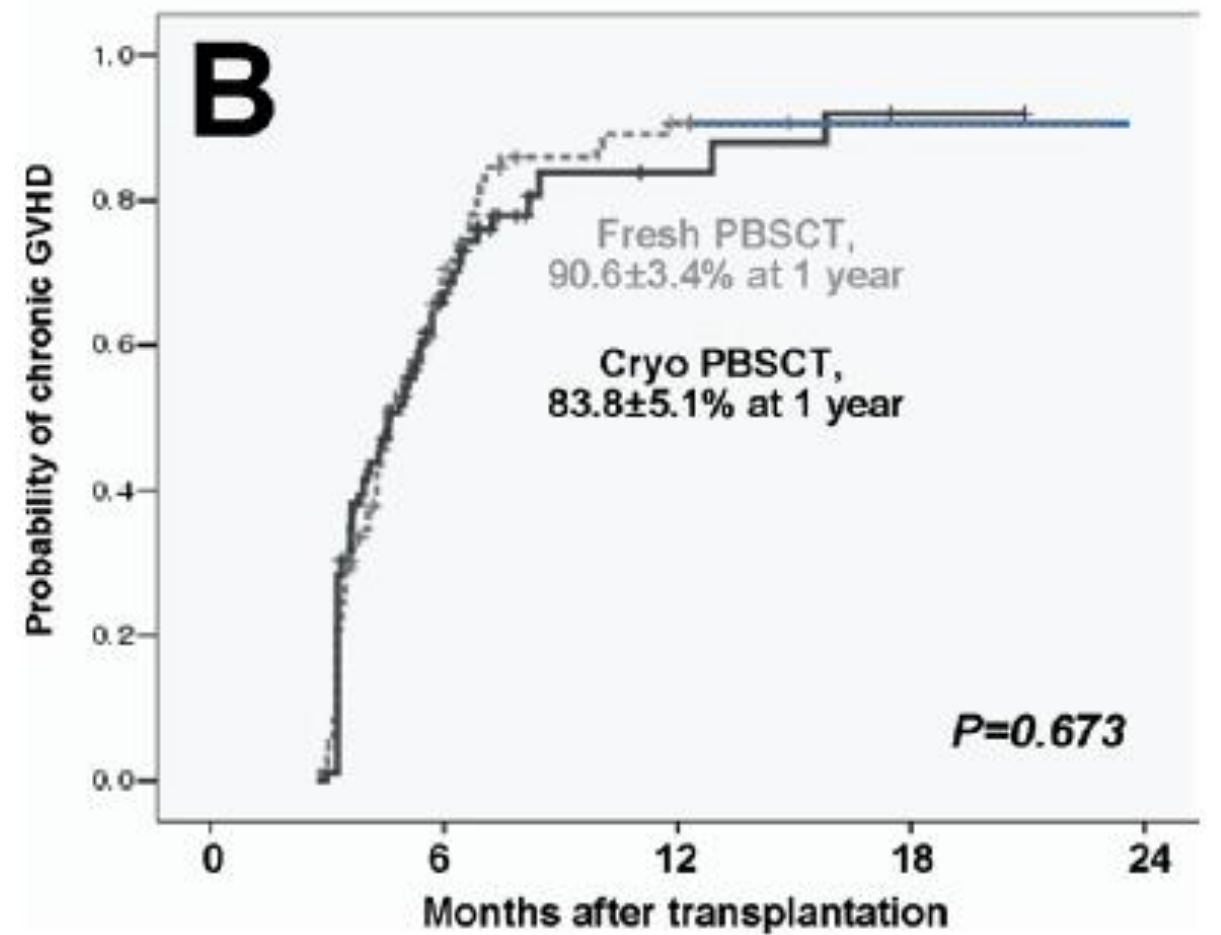
Lenfosit takibi  
1,2,3,6,9,12,  
18,24,36. aylar



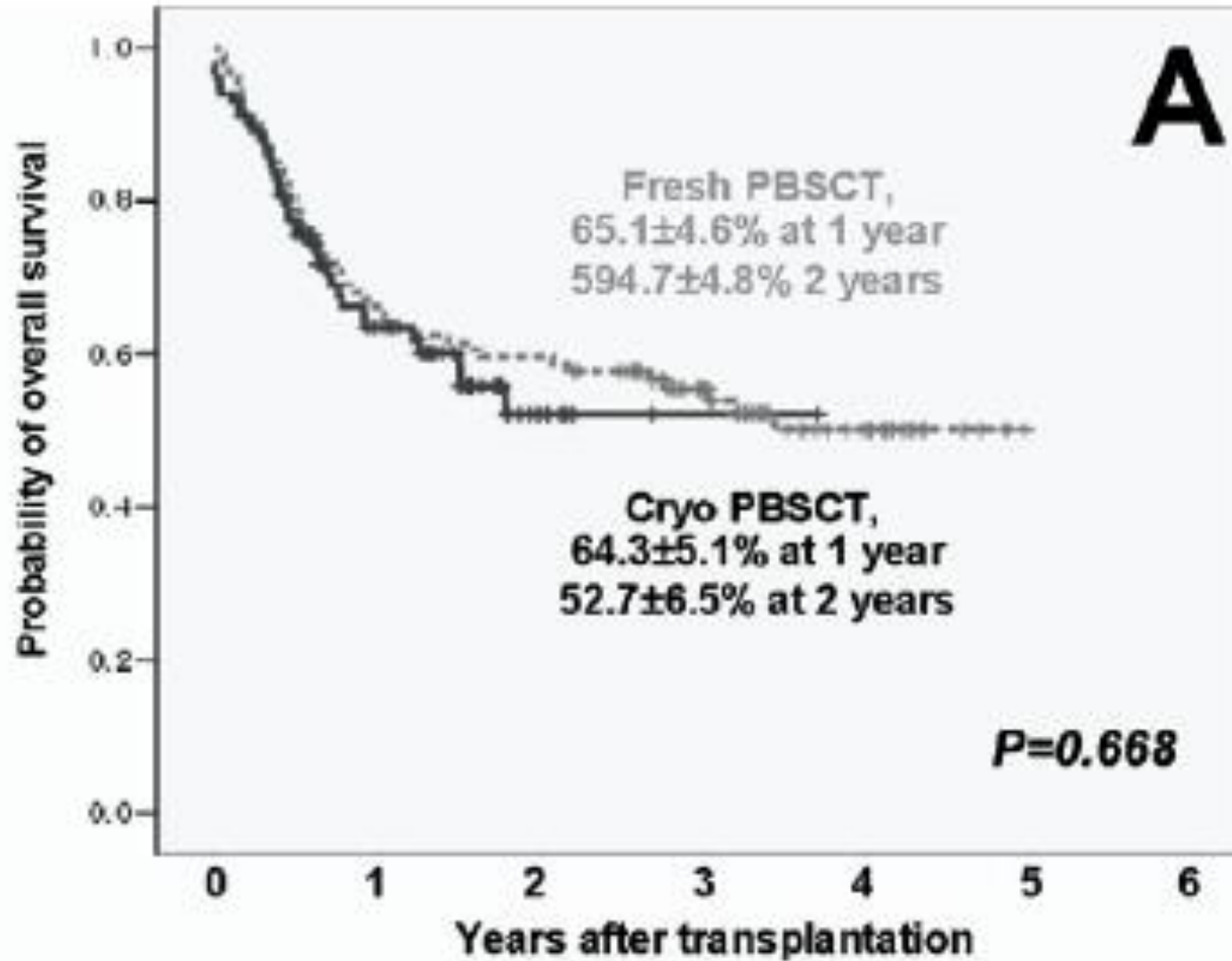
# Dondurulmuş ve taze PBSCT sonrası akut (A) ve cGVHD (B) görülme sıklığı



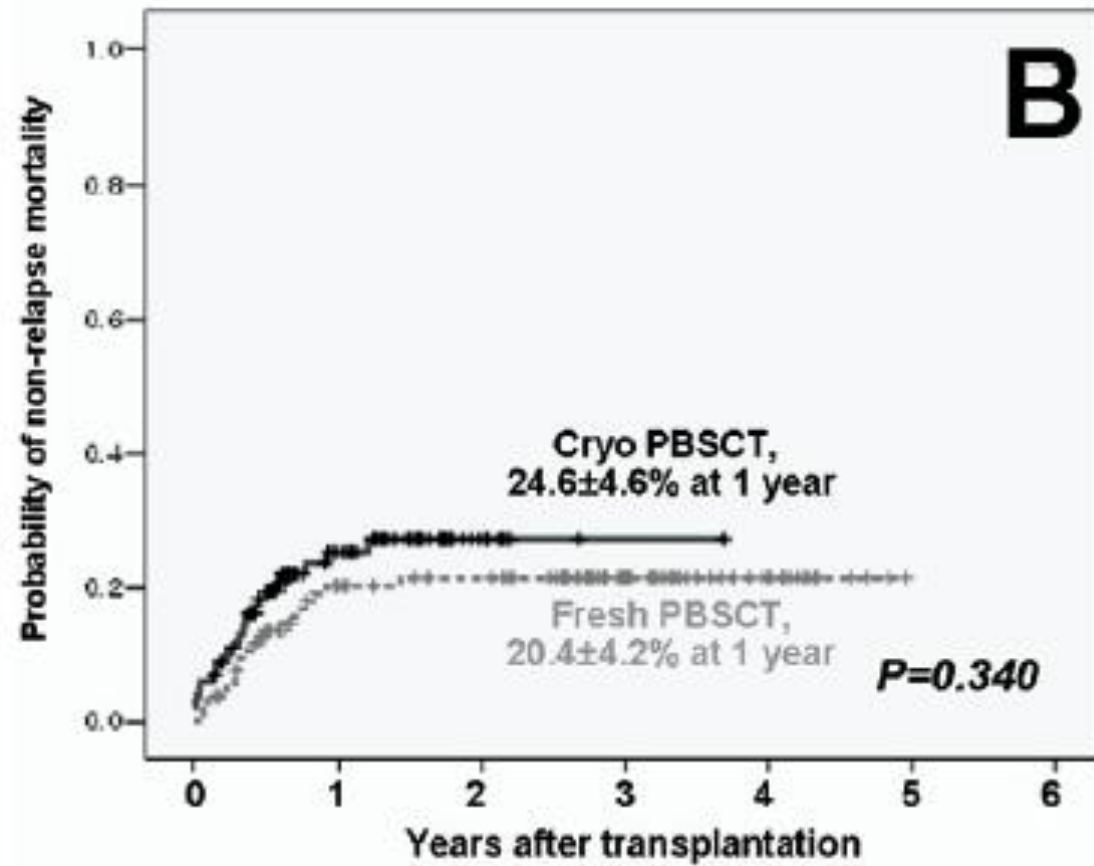
100. Günde aGvHD P:0.113



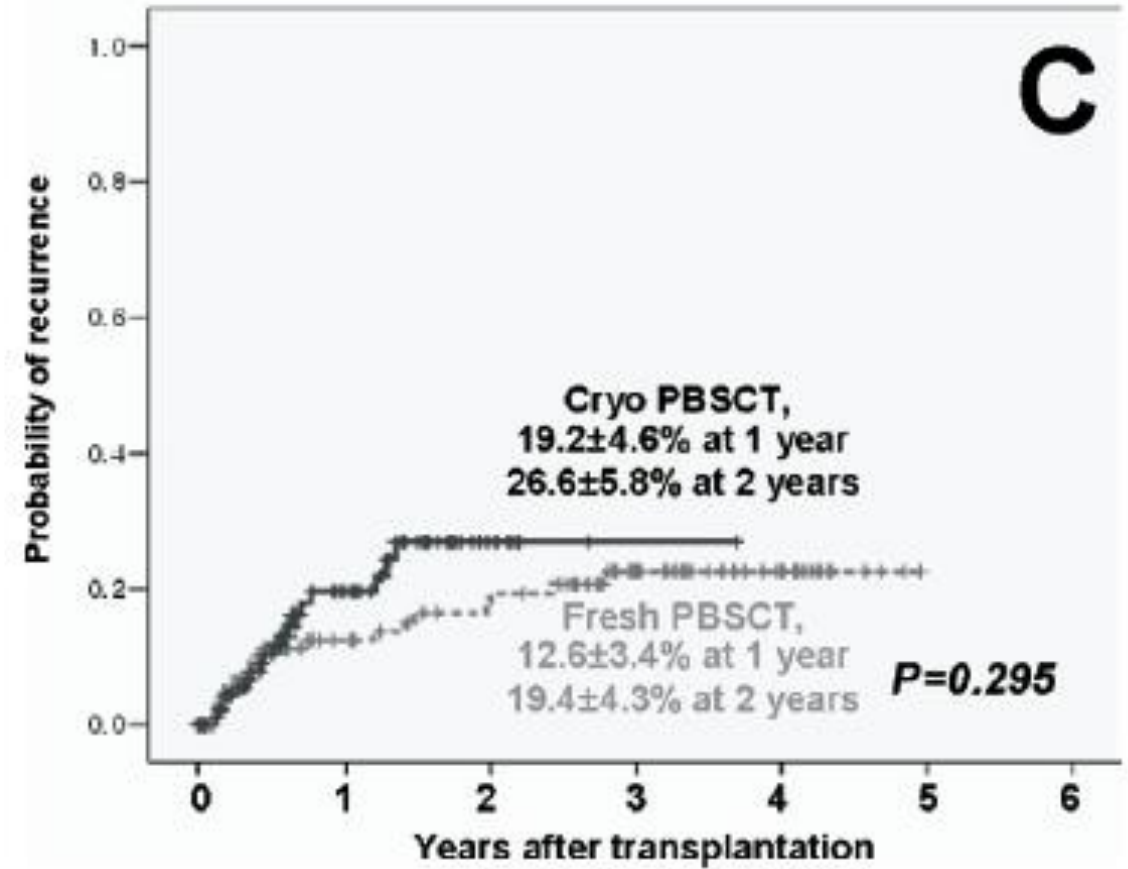
1. Yılda cGvHD P:0.673



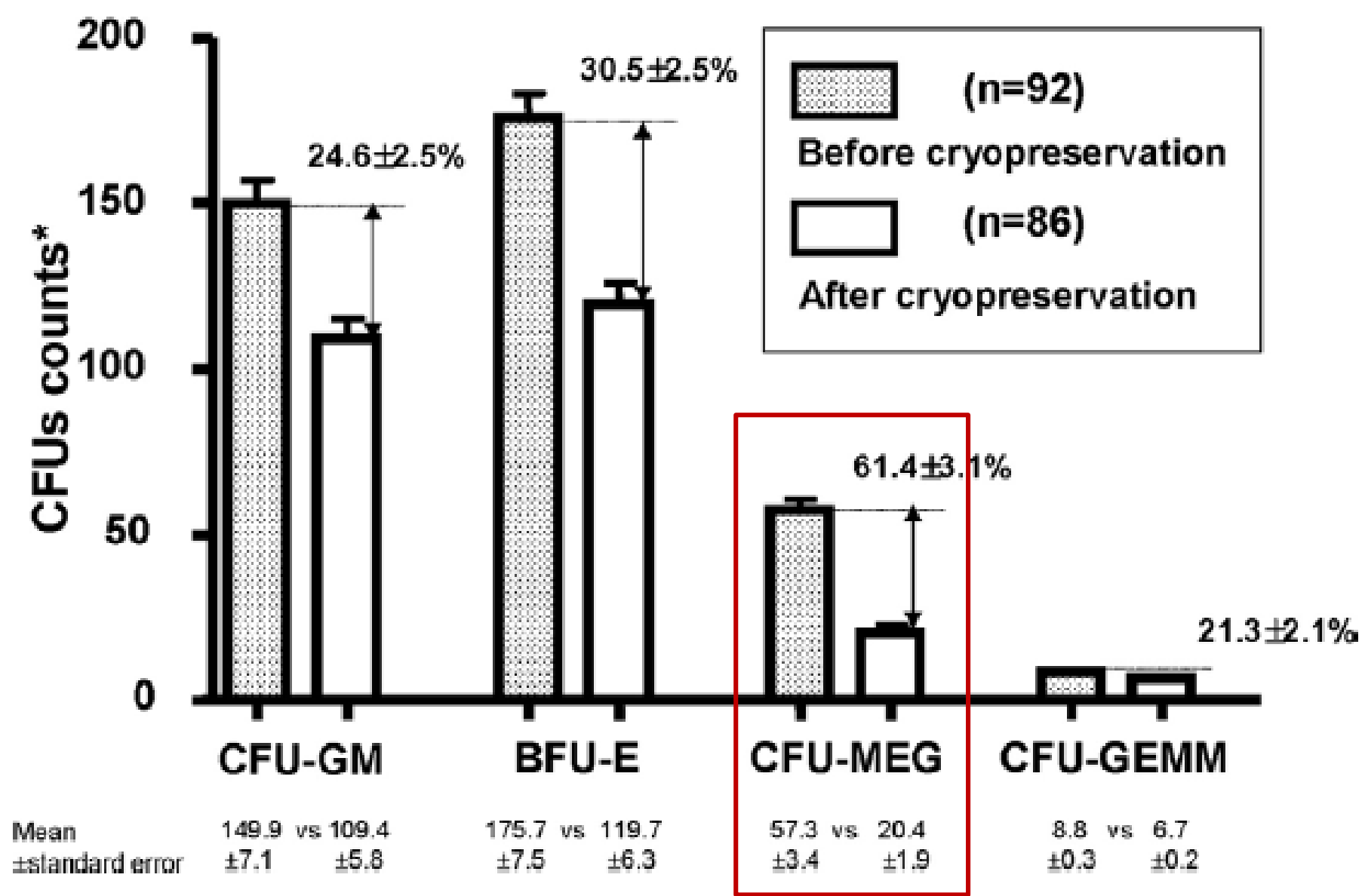
Dondurulmuş ve taze PBSCT almış hasta grubu için **1. ve 2. yıldaki OS oranları (P 0.668)**



1. Yıl NRM: P:0.340



1. Ve 2. yıl relaps oranı: P:0.295



**Tripan Mavisi ile viabilite: median:%71.2 (%53 -98)**

**CFU-MEG çok düşük olan grupta trombosit engrafmanı gecikmekte (med:18 e 29 gün).**



**Table 5.** Uni- and Multivariate Analyses to Evaluate the Contribution of Cryopreservation in the Context of Clinical Risk Factors on OS, NRM, and Disease Recurrence

| Prognostic Factor            | Univariate |                      | Multivariate |                      |
|------------------------------|------------|----------------------|--------------|----------------------|
|                              | P-Value    | HR (95% CI)          | P-Value      | HR (95% CI)          |
| <b>Overall Survival</b>      |            |                      |              |                      |
| Cryopreservation             | .768       | 1.100 (0.601-1.503)  | NS           | —                    |
| Myeloablative conditioning   | .103       | 1.455 (0.926-2.283)  | NS           | —                    |
| High risk disease            | .030       | 2.247 (1.082-4.663)  | .027         | 4.095 (1.170-14.330) |
| Older age ( $\geq 50$ years) | .015       | 1.711 (1.110-2.638)  | .021         | 1.801 (1.094-2.967)  |
| Female donor/male recipient  | .824       | 0.949 (0.597-1.509)  | NS           | —                    |
| Acute GVHD, grade 2-4        | .833       | 0.944 (0.553-1.612)  | NS           | —                    |
| Chronic GVHD                 | <.001      | 0.216 (0.128-0.365)  | <.001        | 0.197 (0.114-0.339)  |
| <b>Non-relapse mortality</b> |            |                      |              |                      |
| Cryopreservation             | .342       | 1.330 (0.739-2.395)  | NS           | —                    |
| Myeloablative conditioning   | .841       | 1.070 (0.551-2.080)  | NS           | —                    |
| High-risk disease            | .439       | 0.457 (0.063-3.321)  | NS           | —                    |
| Older age ( $\geq 50$ years) | .020       | 2.113 (1.124-3.974)  | .041         | 2.299 (1.036-5.102)  |
| Female donor/male recipient  | .745       | 1.110 (0.591-2.087)  | NS           | —                    |
| Acute GVHD, grade 2-4        | .091       | 2.439 (0.866-6.896)  | NS           | —                    |
| Chronic GVHD                 | .021       | 0.372 (0.161-0.858)  | .006         | 0.302 (0.129-0.709)  |
| <b>Recurrence</b>            |            |                      |              |                      |
| Cryopreservation             | .297       | 1.432 (0.729-2.811)  | NS           | —                    |
| Myeloablative conditioning   | .008       | 0.411 (0.214-0.790)  | NS           | —                    |
| High risk disease            | <.001      | 8.431 (3.816-18.625) | <.001        | 4.854 (2.041-11.628) |
| Older age ( $\geq 50$ years) | .111       | 1.719 (0.883-3.348)  | NS           | —                    |
| Female donor/male recipient  | .146       | 0.543 (0.239-1.238)  | NS           | —                    |
| Acute GVHD, grade 2-4        | .094       | 0.888 (0.419-1.883)  | NS           | —                    |
| Chronic GVHD                 | <.001      | 0.121 (0.058-0.249)  | <.001        | 0.135 (0.064-0.285)  |

GVHD indicates graft-versus-host disease; HR, hazard ratio; 95% CI, 95% confidence interval; NS, nonsignificant.



# Cryopreservation of allogeneic PBSC from related and unrelated donors is associated with delayed platelet engraftment but has no impact on survival

P Medd<sup>1,2</sup>, S Nagra<sup>1</sup>, D Hollyman<sup>3</sup>, C Craddock<sup>1</sup> and R Malladi<sup>1</sup>

**Table 1.** Patient, disease and transplant characteristics

| Characteristic  | Cryopreserved    | Fresh             | P                 |
|---|------------------|-------------------|-------------------|
| <i>n</i>  | 76               | 123               |                   |
| Median year of transplant (range)                             | 2008 (2004–2010) | 2009 (2005–2010)  | <b>0.003</b>      |
| Median age (years (range))                                    | 51.5 (20–71)     | 45 (17–71)        | <b>0.048</b>      |
| Recipient sex male, <i>n</i> (%)                              | 44 (57.9%)       | 72 (58.5%)        | 1                 |
| <i>Diagnosis, n (%)</i>                                       |                  |                   |                   |
| Myeloid   | 50 (65.8%)       | 78 (63.4%)        | 0.76              |
| Lymphoid  | 26 (34.2%)       | 45 (36.6%)        |                   |
| <i>Disease status, n (%)</i>                                  |                  |                   |                   |
| CR  | 51 (67.1%)       | 72 (58.5%)        | 0.23              |
| Not in CR   | 25 (32.9%)       | 51 (41.5%)        |                   |
| <i>Donor, n (%)</i>   |                  |                   |                   |
| Related donor   | 57 (75%)         | 63 (51.2%)        | <b>0.001</b>      |
| MUD   | 19 (25%)         | 60 (48.8%)        |                   |
| Female donor/male recipient, <i>n</i> (%)                     | 21 (28%)         | 19 (15.4%)        | <b>0.0046</b>     |
| <i>CMV status recipient/donor, n (%)</i>                      |                  |                   |                   |
| Negative/negative   | 28 (36.8%)       | 59 (48%)          | 0.88 <sup>a</sup> |
| Negative/positive   | 10 (13.2%)       | 9 (7.3%)          |                   |
| Positive/negative   | 15 (19.7%)       | 18 (14.6%)        |                   |
| Positive/positive   | 23 (30.3%)       | 37 (30.1%)        |                   |
| <i>Conditioning, n (%)</i>                                    |                  |                   |                   |
| Myeloablative   | 28 (36.8%)       | 49 (39.8%)        | 0.76 <sup>b</sup> |
| CY/TBI  | 19 (25%)         | 42 (34.1%)        |                   |
| BU/CY   | 7 (9.2%)         | 5 (4.1%)          |                   |
| Other   | 2 (2.6%)         | 2 (1.6%)          |                   |
| Reduced intensity   | 48 (63.2%)       | 74 (60.2%)        |                   |
| Fludarabine/melphalan/alemtuzumab                             | 31 (40.8%)       | 54 (43.9%)        |                   |
| BEAM–alemtuzumab + / – fludarabine                            | 11 (14.5%)       | 13 (10.6%)        |                   |
| FLAMSA  | 5 (6.6%)         | 3 (2.4%)          |                   |
| Fludarabine/BU  | 0                | 2 (1.6%)          |                   |
| Other   | 1 (1.3%)         | 2 (1.6%)          |                   |
| <i>In vivo</i> T-cell depletion, <i>n</i> (%)                 | 55 (72.4%)       | 90 (73.2%)        | 1                 |
| Median CD34 <sup>+</sup> dose (× 10 <sup>6</sup> /kg (range)) | 6.44 (2.4–16.6)  | 6.27 (0.45–14.28) | 0.4               |

Abbreviations: FLAMSA = fludarabine, amsacrine and cytosine; MUD = matched unrelated donor. <sup>a</sup>CMV negative/negative vs any positive; <sup>b</sup>Myeloablative vs reduced intensity. Bold text indicates statistical significance.

**Table 2.** Factors influencing engraftment

| Variable   | Univariate analysis                            |               | Multivariate analysis          |                |
|--|--|---------------|--------------------------------|----------------|
|  | Outcome  | P             | HR (95% CI)                    | P              |
| <i>Neutrophil engraftment (<math>&gt;0.5 \times 10^9/L</math>)</i> | <i>Proportion engrafted at day 14 (95% CI)</i> |               |                                |                |
| Fresh PBSC   | 87.8% (80.5–92.5)                              | Ref           | 1                              | –              |
| Cryopreserved PBSC   | 72.4% (60.7–81.1)                              | 0.067         | 1.44 (1.13–1.84)               | <b>0.0032</b>  |
| Transplant before Dec 2008   | 76.3% (66.2–83.8)                              | Ref           | –                              | –              |
| Transplant Dec 2008 onwards  | 86.8% (78.6–92)                                | <b>0.025</b>  | –                              | NS             |
| CD34 <sup>+</sup> dose < median                                    | 78% (68.4–85)                                  | Ref           | 1                              |                |
| CD34 <sup>+</sup> dose > median                                    | 85.9% (77.1–91.4)                              | <b>0.042</b>  | 0.93 (0.89–0.97) <sup>a</sup>  | <b>0.00094</b> |
| Myeloablative conditioning   | 76.6% (65.3–84.7)                              | Ref           | 1                              | Ref            |
| Reduced intensity conditioning                                     | 85.2% (77.5–90.5)                              | 0.065         | 0.76 (0.59–0.96)               | <b>0.024</b>   |
| <i>Platelet engraftment (<math>&gt;50 \times 10^9/L</math>)</i>    | <i>Proportion engrafted at day 30 (95% CI)</i> |               |                                |                |
| Fresh PBSC   | 90.2% (83.3–94.4)                              | Ref           | 1                              | Ref            |
| Cryopreserved PBSC   | 71.1% (59.3–80)                                | <b>0.0002</b> | 1.85 (1.36–2.51)               | <b>0.00009</b> |
| Transplant before Dec 2008   | 75.3% (65–82.9)                                | Ref           | 1                              | Ref            |
| Transplant Dec 2008 onwards  | 89.6% (81.8–94.2)                              | <b>0.001</b>  | 0.66 (0.49–0.88)               | <b>0.0056</b>  |
| Related donor  | 81.7% (73.4–87.6)                              | Ref           | –                              | –              |
| MUD  | 84.8% (74.5–91.2)                              | <b>0.016</b>  | –                              | NS             |
| Other than female donor/male recipient                             | 83.5% (76.7–88.5)                              | Ref           | –                              | –              |
| Female donor/male recipient  | 80% (63.3–89.7)                                | 0.097         | –                              | NS             |
| CD34 <sup>+</sup> dose < median                                    | 83% (73.9–89.1)                                | Ref           | 1                              | Ref            |
| CD34 <sup>+</sup> dose > median                                    | 82.8% (73.7–89)                                | 0.18          | 0.926 (0.88–0.98) <sup>a</sup> | <b>0.0058</b>  |

Abbreviations: 95% CI = 95% confidence interval; HR = hazard ratio; MUD = matched unrelated donor. <sup>a</sup>Analysed as a continuous variable, HR per unit increase in CD34<sup>+</sup> dose. Bold text indicates statistical significance.

# Cryopreservation of allogeneic PBSC from related and unrelated donors is associated with delayed platelet engraftment but has no impact on survival

P Medd<sup>1,2</sup>, S Nagra<sup>1</sup>, D Hollyman<sup>3</sup>, C Craddock<sup>1</sup> and R Malladi<sup>1</sup>

**Table 3.** Univariate analysis of post-transplant outcomes

| <i>Outcome</i>                             | <i>Cryopreserved (95% CI)</i> | <i>Fresh (95% CI)</i> | <i>P</i> |
|--|-------------------------------|-----------------------|----------|
| OS at 2 years                              | 45.3% (35.7–59.2)             | 60.3% (51.9–70)       | 0.13     |
| RFS at 2 years                             | 41.9% (31.5–55.7)             | 51.2% (42.9–61.2)     | 0.39     |
| Relapse incidence at 2 years               | 35.3% (24.2–46.7)             | 30% (21.8–38.6)       | 0.45     |
| TRM at 1 year                              | 14.6% (7.7–23.5)              | 17.9% (11.7–25.2)     | 0.68     |
| Grade 2–4 acute GVHD incidence at day 100  | 31.7% (21–43)                 | 36.9% (27.1–43.5)     | 0.36     |
| Extensive chronic GVHD incidence at 1 year | 40.3% (28.3–52)               | 28.3% (20–35.8)       | 0.13     |

Abbreviations: 95% CI = 95% confidence interval; RFS = relapse-free survival.

**Table 1** Demographic Data for Total Study Group

| Variable                                 | N = 63 |
|--|--------|
| <b>Original Diagnosis</b>                |        |
| AML/MDS                                  | 32     |
| NHL                                      | 7      |
| HL                                       | 2      |
| ALL                                      | 13     |
| MM                                       | 2      |
| CLL                                      | 2      |
| CML                                      | 4      |
| AML/ALL                                  | 1      |
| <b>Transplantation Conditioning Type</b> |        |
| Full                                     | 40     |
| RIC                                      | 23     |
| <b>Any Chemotherapy Before DLI?</b>      |        |
| Yes                                      | 29     |
| No                                       | 34     |
| <b>Donor Source of Cells for DLI</b>     |        |
| Matched sibling                          | 20     |
| Matched unrelated                        | 11     |
| Mismatched donors                        | 0      |
| <b>Grade III or IV GVHD after DLI?</b>   |        |
| Yes                                      | 30     |
| No                                       | 33     |

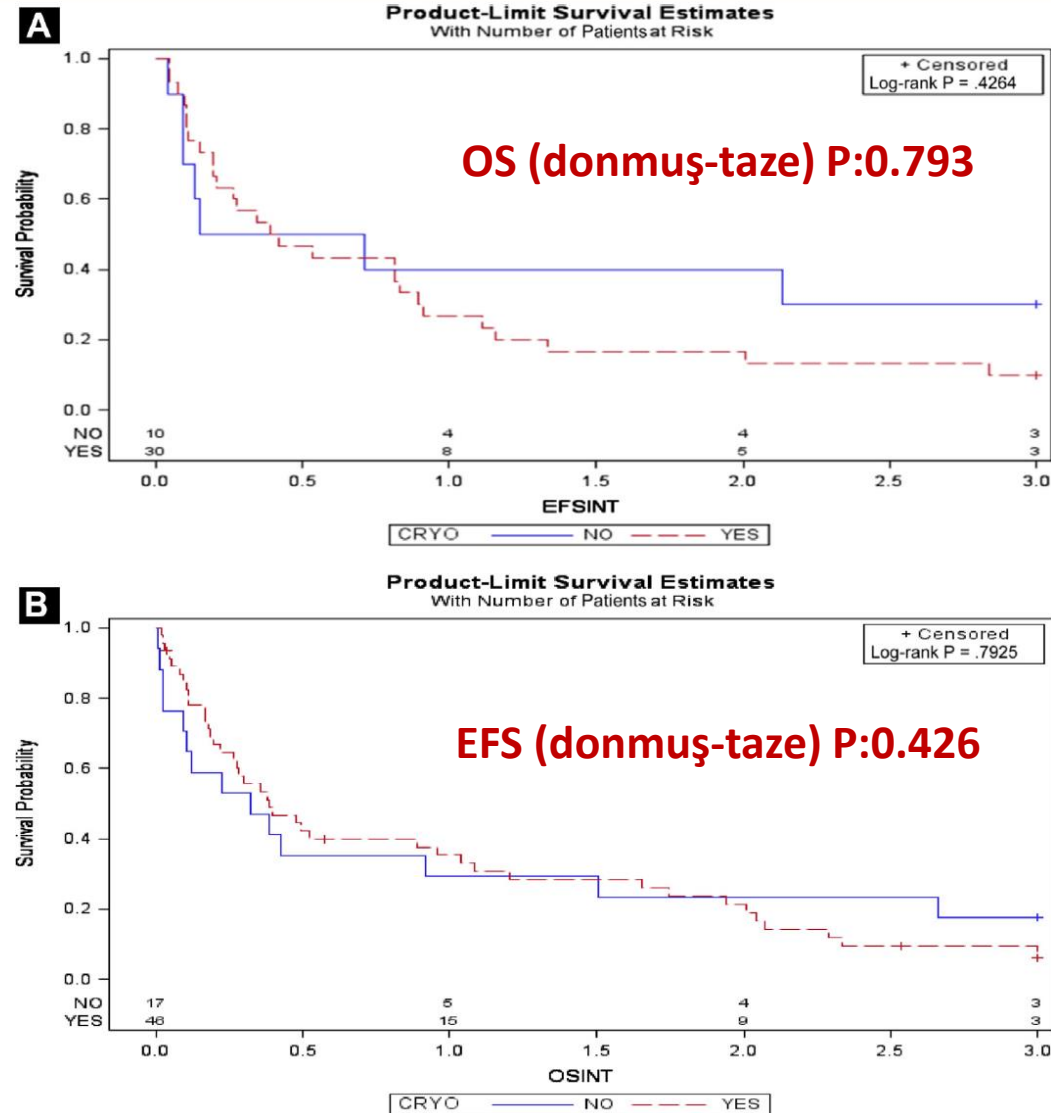
## Donor Lymphocyte Infusion in Hematologic Malignancies—Good to be Fresh?

Nasheed Mohammad Hossain,<sup>1</sup> Thomas Klumpp,<sup>2</sup> John Ulicny,<sup>3</sup> Michael Garner,<sup>3</sup>  
Patricia Lamont Kropf,<sup>3</sup> Kenneth F. Mangan,<sup>3</sup> Stefan Klaus Barta,<sup>4</sup>  
Henry C. Fung,<sup>3</sup> Mary Ellen Martin<sup>3</sup>

**Table 2** AML/MDS Subgroup

|  |    |
|--|----|
| <b>Transplantation Conditioning Type</b>         |    |
| Full   | 21 |
| RIC  | 11 |
| <b>Any Chemotherapy Before DLI?</b>              |    |
| Yes  | 17 |
| No   | 15 |
| <b>Disease Burden at Time of Transplantation</b> |    |
| <10% blasts                                      | 16 |
| ≥10% blasts                                      | 9  |
| Disease burden unavailable                       | 7  |
| <b>Donor Source of Cells for DLI</b>             |    |
| Matched sibling                                  | 19 |
| Matched unrelated                                | 13 |
| Mismatched donors                                | 0  |
| <b>Grade III or IV GVHD?</b>                     |    |
| Yes  | 3  |
| No   | 0  |

Figure 1 (A) Overall Survival (OS) After Donor Lymphocyte Infusion (DLI) for Cryopreserved Cells Versus Fresh Cells. (B) Event-free Survival (EFS) After DLI for Cryopreserved Cells Versus Fresh Cells



## Donor Lymphocyte Infusion in Hematologic Malignancies—Good to be Fresh?

Nasheed Mohammad Hossain,<sup>1</sup> Thomas Klumpp,<sup>2</sup> John Ulicny,<sup>3</sup> Michael Garner,<sup>3</sup> Patricia Lamont Kropf,<sup>3</sup> Kenneth F. Mangan,<sup>3</sup> Stefan Klaus Barta,<sup>4</sup> Henry C. Fung,<sup>3</sup> Mary Ellen Martin<sup>3</sup>

### 63 vaka – 32 AML/MDS

DLI sonrası median takip süresi **5.4 yıl** (dağılım, 0.03-11.88 yıl) idi.

### 40 vaka donmuş 23 vaka taze DLI

Nakil işleminden DLI'ye medyan süre **184 gündü**.

**DLI türünün (taze ya da dondurulmuş), DLI prosedüründen sonra OS ya da EFS üzerinde önemli bir etkisi olmadığını göstermektedir.**



# Pair-matched study of cryopreserved *versus* native graft in adult and pediatric recipients of allogeneic hematopoietic stem cell transplantation

Elena V. Babenko, Ivan S. Moiseev, Mikhail M. Kanunnikov, Alexandr L. Alyanskiy, Dmitrii E. Pevcov, Anastasia V. Frolova, Anna A. Osipova, Tatyana A. Bykova, Olesya V. Paina, Elena I. Darskaya, Ludmila S. Zubarovskaya, Sergey N. Bondarenko, Inna V. Markova, Boris V. Afanasyev

R. Gorbacheva Memorial Research Institute of Children Oncology, Hematology and Transplantation, The First St. Petersburg State I. Pavlov Medical University, St. Petersburg, Russia

- Greft kriyoprezervasyonun allojeneik kök hücre transplantasyonunun sonuçları ve toksisitesi üzerindeki etkisini değerlendirmek için tek merkezli ve eşleşmiş bir retrospektif çalışma
- **2006-2017 döneminde 162 hasta**
- **Dondurma nedenleri:**
  - Koşullandırma başlamadan önce enfeksiyon (% 38,3),
  - Altta yatan hastalığın nüksü (% 32,2),
  - HSCT sırasında akraba donörün bulunmaması (% 17,2),
  - Sonradan dondurularak saklanan greftin mevcudiyeti kısıtlaması nedeniyle ilk bağış (%11.1)
  - Hazırlama rejimi öncesi gebelik (% 1.2).
  - Ortanca takip süresi 25 ay
- İki yıllık takip sonuçları



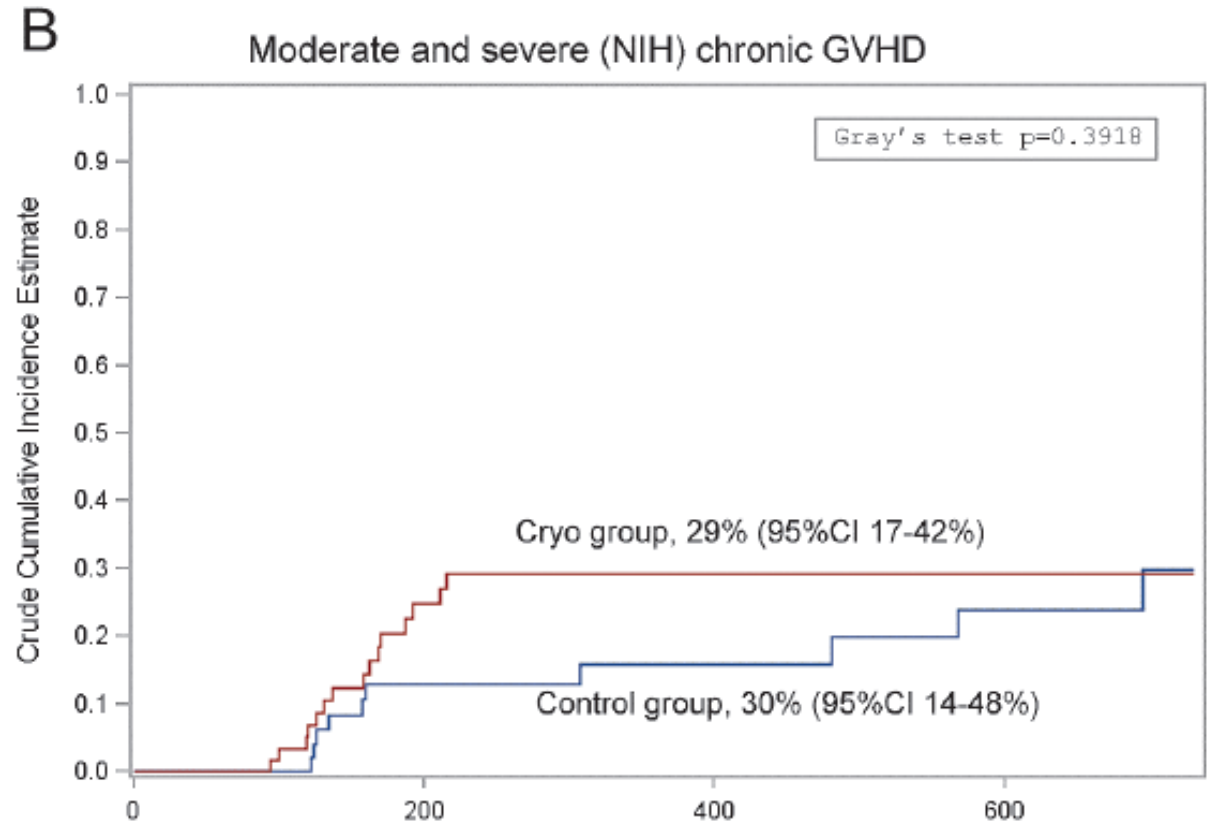
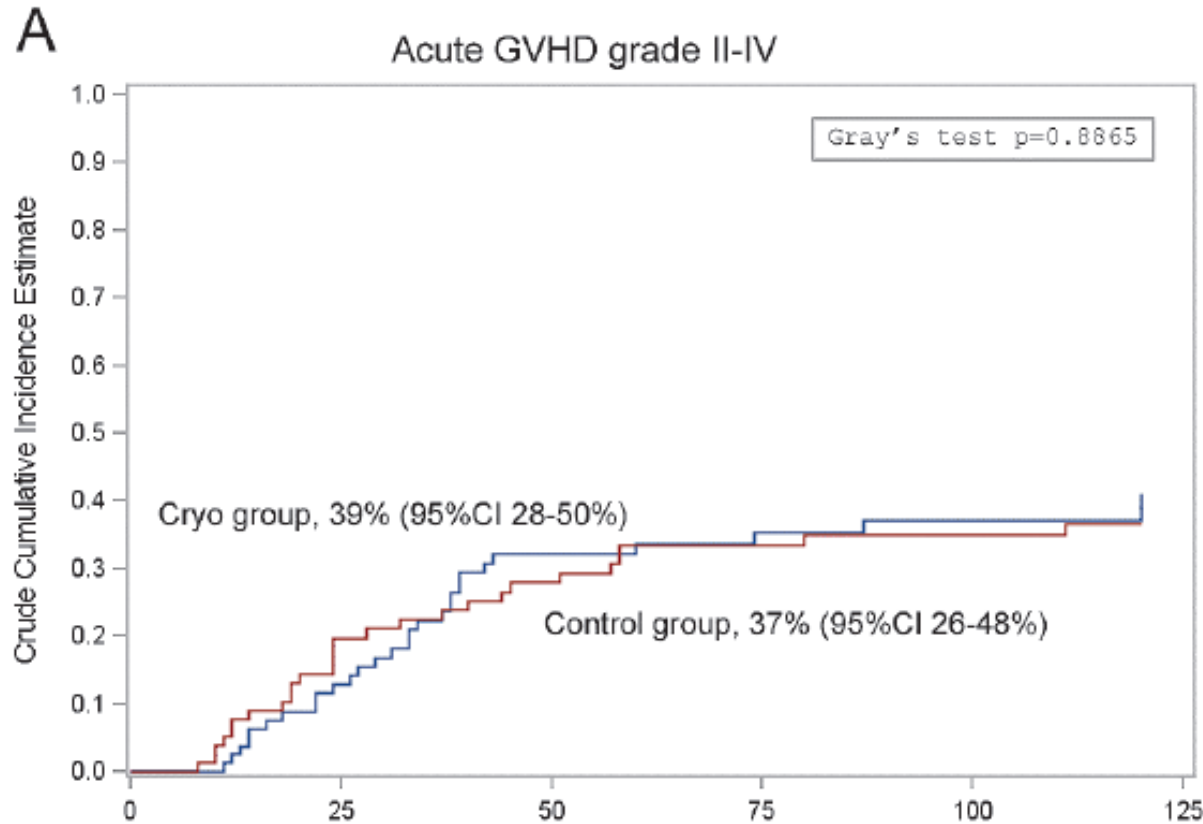
Table 1. Characteristics of patients and transplantations.

| Parameter                                 | Cryopreserved graft (N=81)   | Native graft (N=81)  | p-value          |
|---|--|--|------------------|
| Age                                       | Adult 80.72%<br>Children 19.28%  | Adult 81.01%<br>Children 18.99%  | 0.9626           |
| Male, %                                   | 49.40%   | 50.63%   | 0.8751           |
| Diagnosis                                 | AML 37.97%<br>ALL 31.33%<br>MDS/MPN 10,84%<br>CML 9.64%<br>Lymphoma 6,49%<br>AA 2.53%<br>Solid tumor 1.20% | AML 37.97%<br>ALL 31.33%<br>MDS/MPN 9,64%<br>CML 10,84%<br>Lymphoma 6,49%<br>AA 2.53%<br>Solid tumor 1.20% | 0.9520           |
| Disease risk index                        | 1 – 9.64%<br>2 – 48.19%<br>3 – 33.73%<br>4 – 8.43%   | 1 – 17.95%<br>2 – 44.87%<br>3 – 32.05%<br>4 – 6.17%  | 0.4227           |
| Donor                                     | Related 17.2%<br>Unrelated 82.8%   | Related 17.2%<br>Unrelated 82.8%   | 1.0              |
| Graft source                              | BM 28,4%<br>PBSC 71,6%   | BM 25,9%<br>PBSC 74,1%   | 0.7742           |
| Number of HSCT                            | First – 88,9%<br>Subsequent – 11.1%  | First – 90.2%<br>Subsequent – 9.8%   | 0.3622           |
| Conditioning                              | RIC 75.90%<br>MAC 24.10%   | RIC 78.48%<br>MAC 21.52  | 0.6961           |
| GVHD prophylaxis                          | PTCy-based 32.53%<br>Tacrolimus 73.49%   | PTCy-based 34.18%<br>Tacrolimus 67.09%   | 0.8241<br>0.4740 |
| CD34+ 10x6/kg cells in the graft, mean±SD | 4.8±2.5  | 5.0±2.5  | 0.9266           |

# Pair-matched study of cryopreserved *versus* native graft in adult and pediatric recipients of allogeneic hematopoietic stem cell transplantation

Elena V. Babenko, Ivan S. Moiseev, Mikhail M. Kanunnikov, Alexandr L. Alyanskiy, Dmitrii E. Pevcov, Anastasia V. Frolova, Anna A. Osipova, Tatyana A. Bykova, Olesya V. Paina, Elena I. Darskaya, Ludmila S. Zubarovskaya, Sergey N. Bondarenko, Inna V. Markova, Boris V. Afanasyev

R. Gorbacheva Memorial Research Institute of Children Oncology, Hematology and Transplantation, The First St. Petersburg State I. Pavlov Medical University, St. Petersburg, Russia

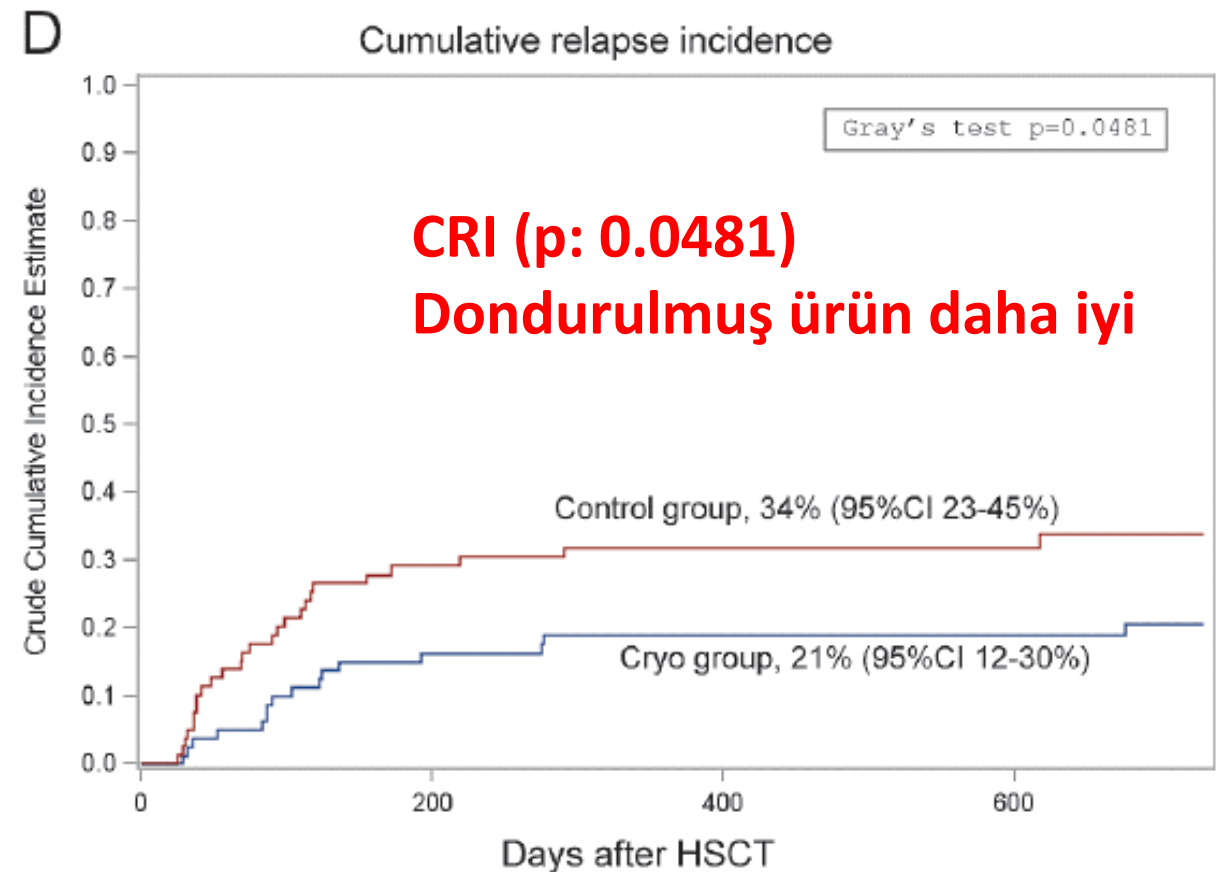
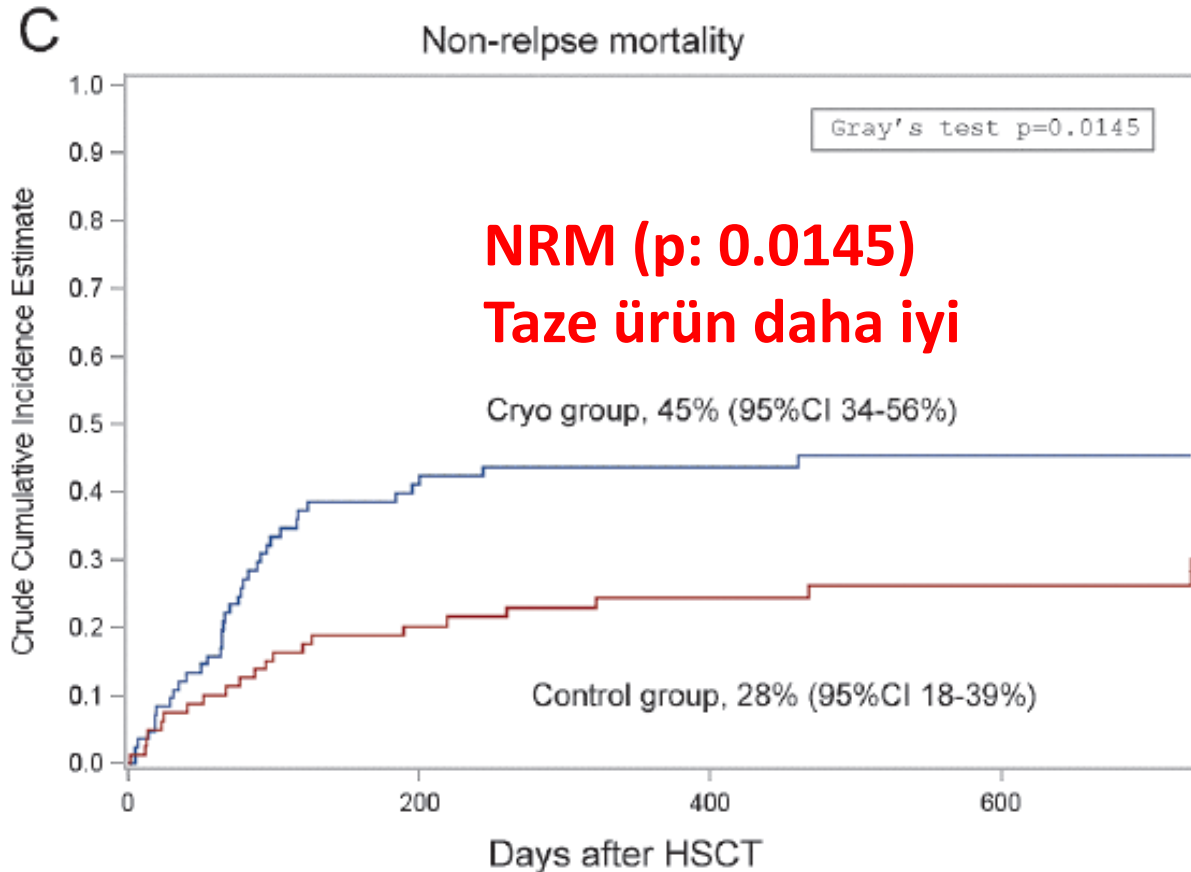


**Akut ve Kronik GvHD açısından iki grup arasında fark yok**

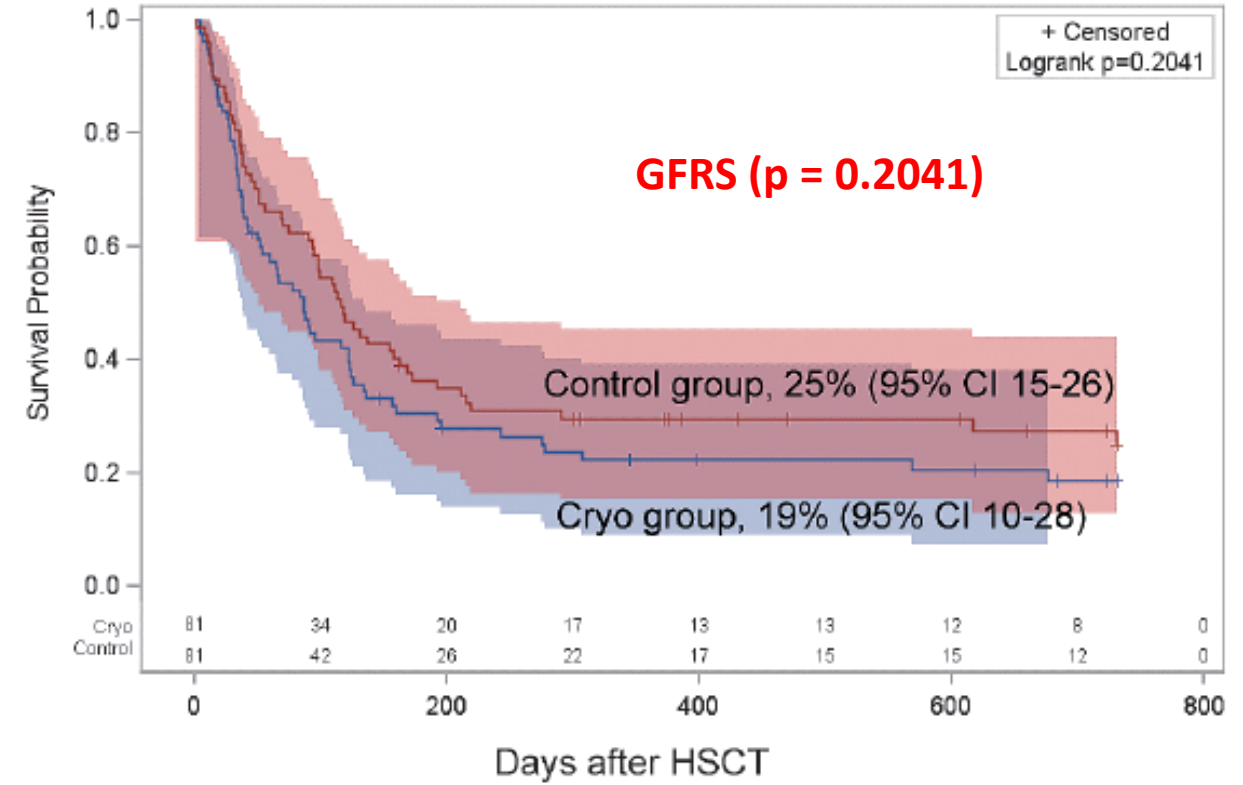
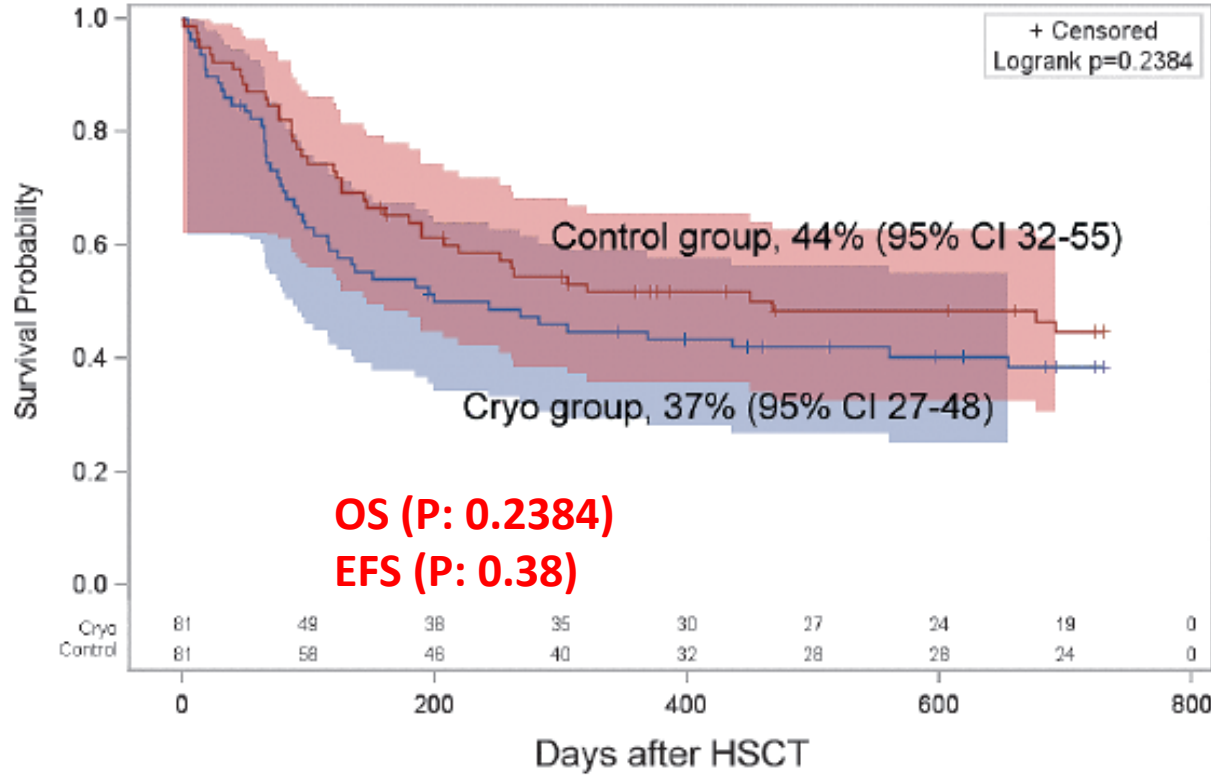
# Pair-matched study of cryopreserved *versus* native graft in adult and pediatric recipients of allogeneic hematopoietic stem cell transplantation

Elena V. Babenko, Ivan S. Moiseev, Mikhail M. Kanunnikov, Alexandr L. Alyanskiy, Dmitrii E. Pevcov, Anastasia V. Frolova, Anna A. Osipova, Tatyana A. Bykova, Olesya V. Paina, Elena I. Darskaya, Ludmila S. Zubarovskaya, Sergey N. Bondarenko, Inna V. Markova, Boris V. Afanasyev

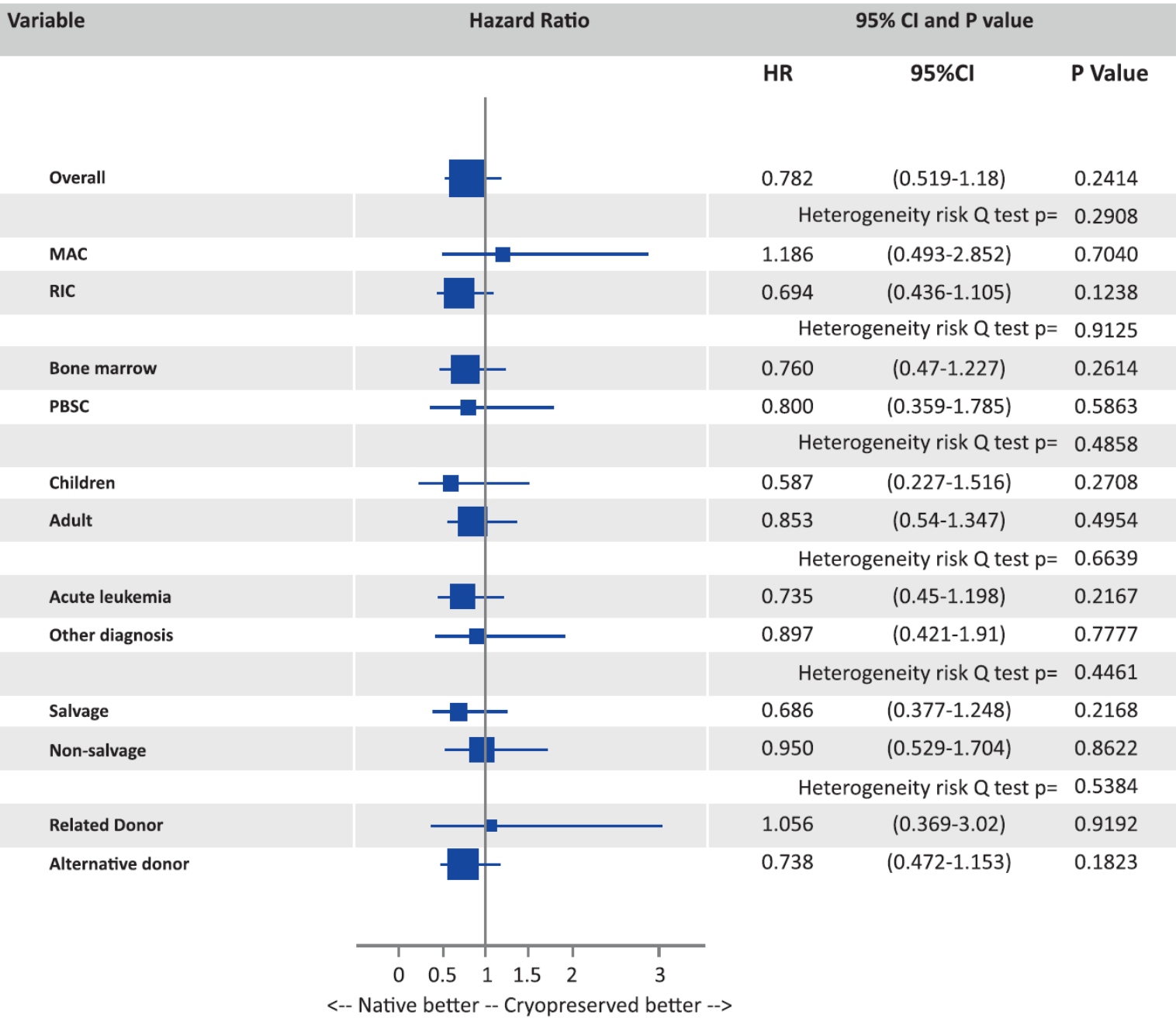
R. Gorbacheva Memorial Research Institute of Children Oncology, Hematology and Transplantation, The First St. Petersburg State I. Pavlov Medical University, St. Petersburg, Russia



# Pair-matched study of cryopreserved *versus* native graft in adult and pediatric recipients of allogeneic hematopoietic stem cell transplantation

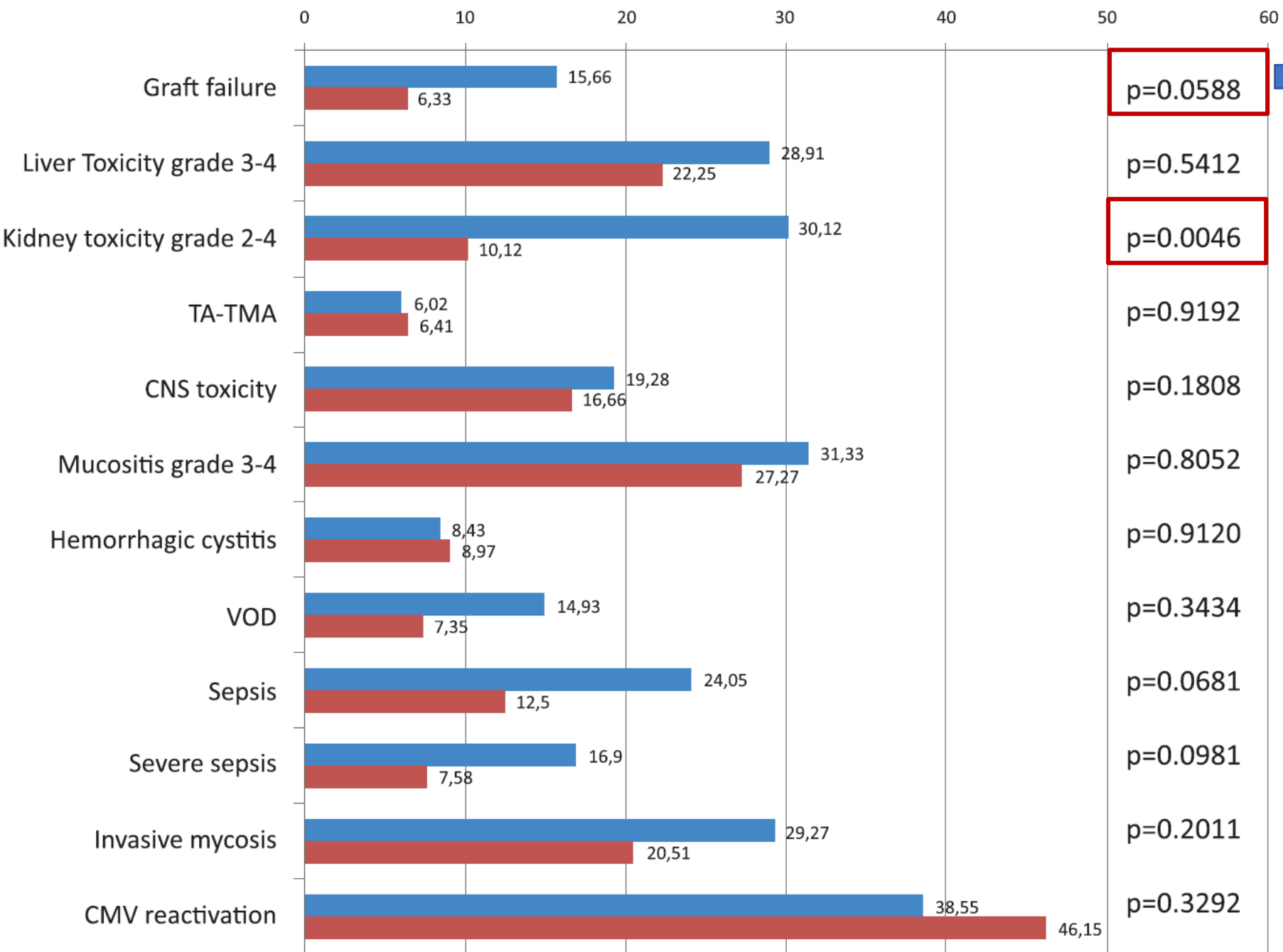


Bu iki yönlü farklılıklar, greft eritmenin OS üzerinde istatistiksel anlamlı bir etkisinin olmamasına neden oldu



**OS sonucunun alt grup analizi,**

- Hazırlama rejimi yoğunluğu,
- Greft kaynağı,
- Hastaların yaşı,
- Altta yatan hastalık,
- Hastalığın durumu ve
- Donör tipine bakılmaksızın kriyoprese edilmiş ve doğal greft arasında bir farklılık göstermedi (**p> 0.2**).



➡ **Graft kaynağı – Graft yetersizliği**  
**Kİ: %26 ya %0 (P:0,025)**  
**PK: %12 ye %9 (P:0,483)**

**Engrafte olan hastalar arasında**  
**Nötrofil engrafmanı**  
 ( ort. 19 - 18 gün, **p = 0.345**,  
 Kriyo ve kontrol gruplarında),

**Trombosit engrafmanı**  
 (17 vs 14 gün, **p = 0.442**).

### Kök hücre nakli komplikasyonları

■ Cryopreserved ■ Native



## Comparison of outcomes post allogeneic hematopoietic cell transplantation using fresh versus cryopreserved peripheral blood stem cell grafts

- Metod: 2003-2017 yılları arasında 951 erişkin allo-periferik kan HCT uygulanan hastanın retrospektif incelemesi (Tek Merkez)
- Trombosit ( $\geq 20 \times 10^9 / L$ )
- Nötrofil ( $\geq 50 \times 10^9 / L$ ) yamalanması
- İlk 100 gün akut greft versus-host hastalığı (aGvHD)
- Genel sağkalım (OS),
- Kümülatif relaps oranı (CIR)
- Non relaps mortalite.

## Comparison of outcomes post allogeneic hematopoietic cell transplantation using fresh versus cryopreserved peripheral blood stem cell grafts

- Median takip 47 ay (4-177)
- 711 (%75) miyeloid, 229 (%24) lenfoid malignite
- 506 (%53) miyeloablatif

|                                  | TAZE        | DONDURULMUŞ           |
|----------------------------------|-------------|-----------------------|
| Vaka Sayısı                      | 525         | 426                   |
| Median Yaş                       | 53          | 54                    |
| <b>Invivo T hücre deplesyonu</b> | <b>%65</b>  | <b>%18</b>            |
| Nötrofil engraftmanı (gün)       | 15 (8-52)   | 15 (10-48)            |
| Trombosit engraftmanı (gün)      | 16 (12-186) | 17 (8-171)            |
| Grad II-IV aGvHD                 | %51         | %54 ( <b>P:0,42</b> ) |
| Grad III-IV aGvHD                | %28         | %28 ( <b>0,96</b> )   |

# Comparison of outcomes post allogeneic hematopoietic cell transplantation using fresh versus cryopreserved peripheral blood stem cell grafts

OS 2 yıl %50 (47-54), OS 5 yıl %40 (36-43)

| Tek değişkenli analiz |      |               |
|-----------------------|------|---------------|
|                       | TAZE | DONDURULMUŞ   |
| OS (2 yıl)            | %47  | %54           |
| OS (5 yıl)            | %39  | %41 (P: 0,21) |
| CIR (2 yıl)           | %16  | %21           |
| CIR (5 yıl)           | %19  | %25 (P: 0,02) |

## Çok değişkenli analizde:

- **OS** taze ve donmuş arasında fark yok. (P:0,25)
- **NRM** için dondurma, artmış risk için bağımsız belirleyici değil (p = 0.06).
- **CIR** için, kriyoprezervasyon tek bağımsız nüks belirleyicisidir (dondurulmuş ürün için **HR 1.43**,% 95 CI 1.07-1.91, p = 0.02)

Sadece MRD transplantlar için çok değişkenli analiz tekrarlandığında (n = 474 vaka 385 dondurulmuş greft, 89 taze)

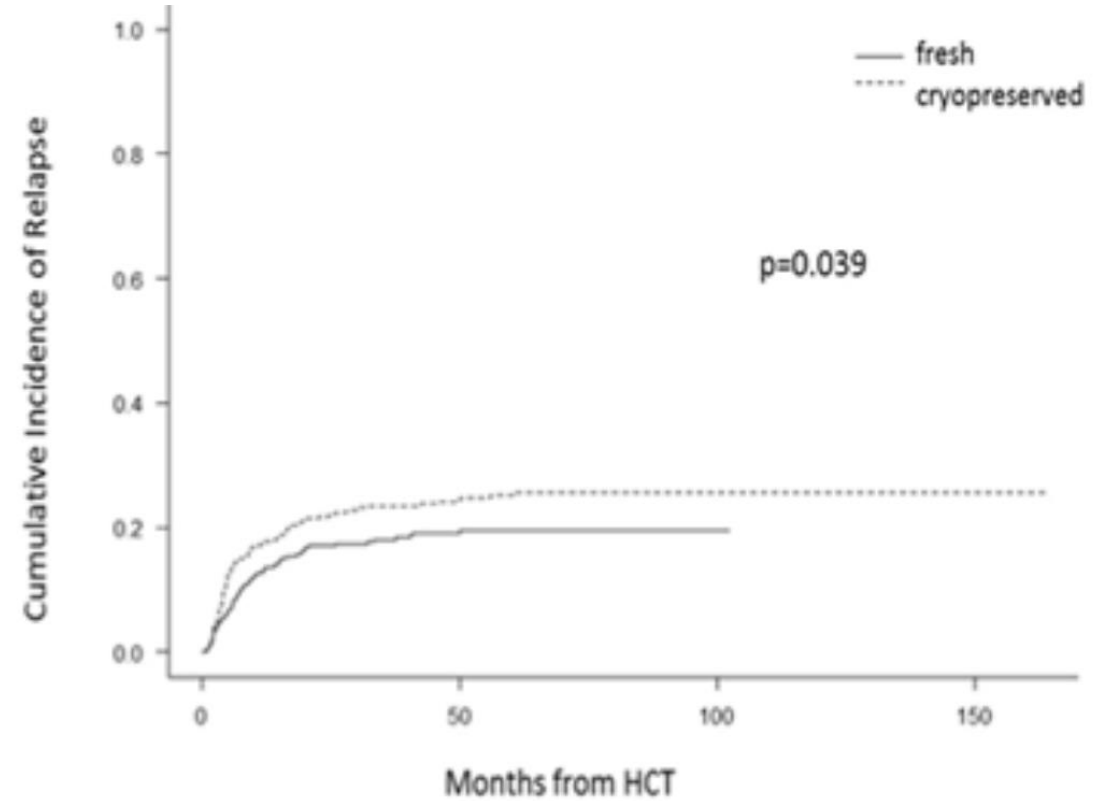
Dondurulmuş greftler için bağımsız nüks riskinin arttığını doğruladı (**HR 2.30**,% 95 CI 1.21-4.38, **p = 0.01**).

# Comparison of outcomes post allogeneic hematopoietic cell transplantation using fresh versus cryopreserved peripheral blood stem cell grafts

## Sonuç:

Tek deęişkenli ve çok deęişkenli analizlerde,

- Taze ve dondurarak korunan periferik kan kök hücre greftleri ile yapılan allojenik nakiller arasında OS'de anlamlı bir fark yok
- **Ancak kriyoprezervasyonla ilişkili nüks riskinde anlamlı bir artış olduęu tespit edilmiş.**



# SONUÇ

- Allojenik hematopoetik kök hücre nakli için donör kök hücreleri (alloHSCT) genellikle nakilden hemen önce toplanır ve alıcıya "taze" aktarılır.
- Tıbbi nedenlerden dolayı alıcı kaynaklı, Kök hücre manuplasyonu yapılacaksa ürün kaynaklı yada Donör kaynaklı riskleri azaltmak için, Vericinin kök hücrelerini ya da donör lenfositlerini dondurmak gerekebilir.
- İnfüzyon ilişkili reaksiyonlar dondurulmuş üründe daha yüksektir.
- Çalışmalarda dondurulmuş üründe CFU-MEG düşük bulunmuştur. Bu da trombosit engrafmanında gecikmeye sebep olabilir.
- DLI türünün (taze ya da dondurulmuş), OS ya da EFS üzerinde önemli bir etkisi olmadığını göstermektedir.
- Genel Kanı engrafman, OS, EFS, GFRS, GvHD parametrelerine etki etmediğidir.
- Kök hücreyi dondururken maliyetlerin de göz önünde bulundurulması gerekmektedir.
- Daha kesin yargı için prospektif yüksek vaka sayılı çalışmalara ihtiyaç vardır



**TEŞEKKÜRLER**