



Tata Memorial Centre
Advanced Centre for Treatment, Research & Education in
Cancer (ACTREC)
(Grant-in-Aid Institute of DAE, Govt. of India)



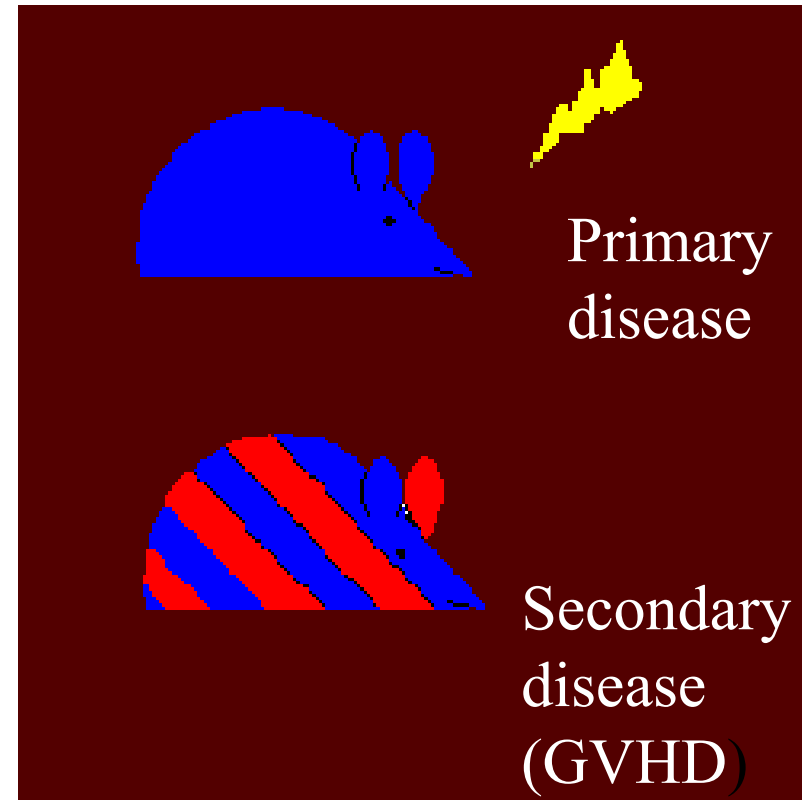
GvHD Prophylaxis: Strategies to mitigate acute GvHD

Dr Navin Khattry



Historical Perspective

- A fatal syndrome of skin abnormalities and diarrhea (or wasting, in newborns) given allogeneic spleen cells after irradiation (or in the newborns, at all).
- Primary disease was the same thing after irradiation itself.
- This was in the 1950's, and in mice.



blood[®] flashback

1965



Georges Mathé was a French pioneer in bone marrow transplantation and cancer immunotherapy contemporaneous with E. D. Thomas and D. W. van Bekkum. He was the first to describe graft-versus-host disease (GVHD) in humans and to understand the potential of the graft-versus-leukemia (GVL) effect to treat human leukemia. Here, he described a 32-year-old man with relapsed refractory acute lymphoblastic leukemia who received a myeloablative transplant using a mixed bone marrow collection from 6 donors (mother, father, and 4 siblings). The patient engrafted and developed short-lived but severe acute GVHD, following which he was shown to have predominance of 1 donor's cells. He recovered but developed recurrent GVHD after donor lymphocyte infusions from the predominant donor given to enhance GVL. Remarkably, his GVHD was controlled and he remained in remission at 12 months.

Successful Allogenic Bone Marrow Transplantation in Man: Chimerism, Induced Specific Tolerance and Possible Anti-Leukemic Effects

By G. MATHÉ, J. L. AMIEL, L. SCHWARZENBERG, A. CATTAN, M. SCH M. J. DE VRIES, M. TUBIANA, C. LALANNE, J. L. BINET, M. PAPIER G. SEMAN, M. MATSUKURA, A. M. MERY, V. SCHWARZMANN AND A. FLAISLER

IT HAS BEEN demonstrated in various animal species that (homologous) bone marrow transplantation is possible after cor the recipient by a lethal dose of total-body irradiation.¹⁰ A success plant is usually complicated by a secondary syndrome,² the mecl which probably involves the reaction of immunologically compe against host antigens.⁴

If the recipient is leukemic, the immune reaction of the graft against leukemic cells^{3,20,25,28,29} and perhaps against the leukemia virus^{19b} may be a powerful therapeutic weapon. Hence, the idea of using allog marrow grafts in the treatment of leukemia. The object in such ca: be to obtain prolonged acceptance of the graft, followed by the syndrome which indicates the graft versus host reaction, and then the undesirable effects of the secondary syndrome.

CASE REPORT

Before Irradiation

B. B., a 26-year-old physician, had been suffering from acute lymphoblast since August, 1961. The first progressive phase was treated with Δ -1-cortison daily from September 10 to November 8, 1961. An apparently complete remission was obtained during the course of which (from November, 1961 to December, 1962) the patient received 6-mercaptopurine in doses of between 50 and 200 mg./day as limited by his gastrointestinal and hematologic tolerance. On December 10, 1962, a differential count of the bone marrow revealed 84 lymphoblasts per 100 nucleated cells. A course of 14 sessions

Total bone marrow graft is associated with specific tolerance towards donor tissues. This is paralleled by the production in the chimaera of immunoglobulins produced by the graft.

The secondary syndrome seems, as in animals, to be related essentially to the graft versus host reaction. It is convenient to distinguish between its various manifestations, on the one hand, those lesions which are readily controlled such as hepatitis or erythrodermia associated with infiltration and proliferation of immunologically competent cells from the graft, and on the other hand immune insufficiency with regard to micro organisms, especially viruses and *Candida albicans*. This latter group, the mechanism of which is complex, still eludes attempts at preventive and curative control.

The use of multiple donors, the administration of cortisone during marrow transfusion and amethopterin and/or cyclophosphamide in the days following transfusions, all seem to have reduced the severity of the secondary syndrome, which however still cannot be satisfactory controlled.

The graft reaction against the leukaemic cells is utilised as an antileukaemic treatment. In the 4 patients who escaped acute secondary syndrome survival has been notably longer, the longest lasted 20 months, when death from zoster encephalitis supervened. There were no clinical signs of leukaemia or histological evidence of this at autopsy.

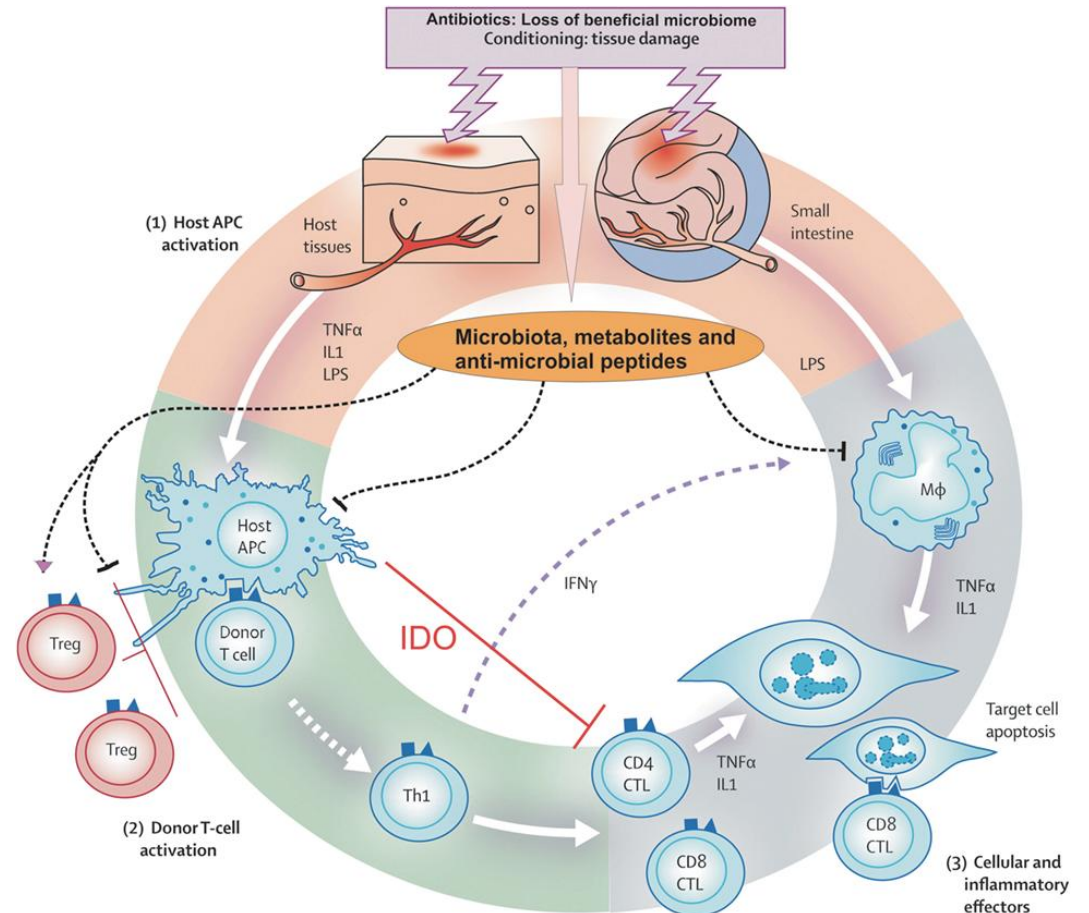
Billingham Postulates for Acute GVHD (1966)

1. The transplanted graft must contain immunologically competent cells – **donor T cells**
2. The recipient must be incapable of rejecting or eliminating transplanted cells – **immune-ablative agents**
3. The recipient must express tissue antigens that are not present in the transplant donor, thus the recipient antigens are recognized as foreign by donor cells – **major and minor HLA-mismatch**

Although aGVHD manifestations occur early after engraftment, late presentations are seen (>100 days – withdrawal of immunosuppression, reduced intensity transplants)

Approaches to blunt the distinct stages of GvHD induction

- Reducing donor & anti-host allogeneic T cells
- Blunting T cell signals
- Inhibiting co-stimulatory signals
- Impairing activating & inflammatory cytokine signals driven GvHD injury
- Regulating histone deacetylase
- Blocking T cell chemokine receptor directed migration into GvHD organs
- Cellular therapy

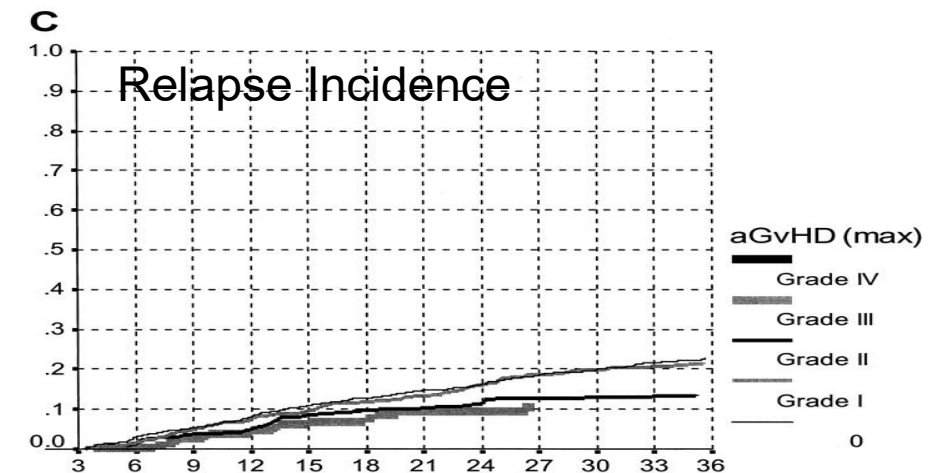
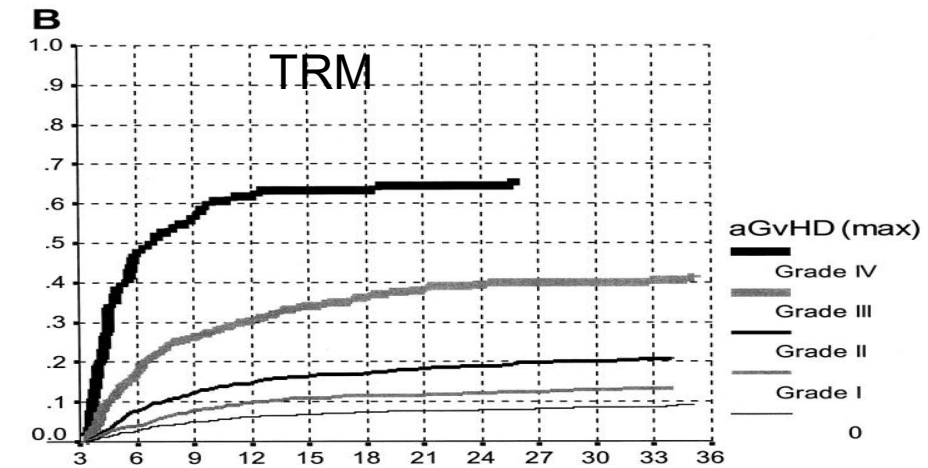
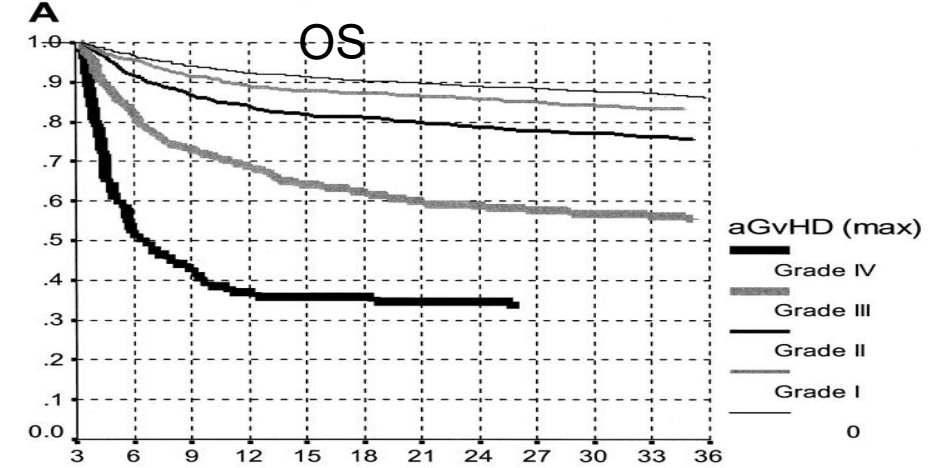


Risk factors for GVHD

Risk factor	Description
HLA disparity	HLAs, encoded by the MHC, present antigens to trigger T-cell activation. These proteins display a high degree of genetic polymorphism, and the degree of HLA mismatch between HSCT donor and recipient is directly related to risk of developing aGvHD
Age	Increased age of either the donor or the recipient is associated with increased aGvHD risk.
Sex mismatch	The incidence of aGvHD increases with female/male donor/recipient pairings.
HSC source	aGvHD is more common in patients receiving peripheral blood stem cell transplants (PBSCT) than bone marrow transplants, likely because PBSCTs contain a greater number of allo-reactive T cells. Umbilical cord blood transplants are associated with a lower risk of aGvHD, likely because more immature cells are contained in the graft.
Intensity of conditioning regimen	Multiple but not all studies show that the risks of aGvHD is lower with reduced-intensity conditioning regimens, compared with myeloablative regimens

A balance of GVH and GVL

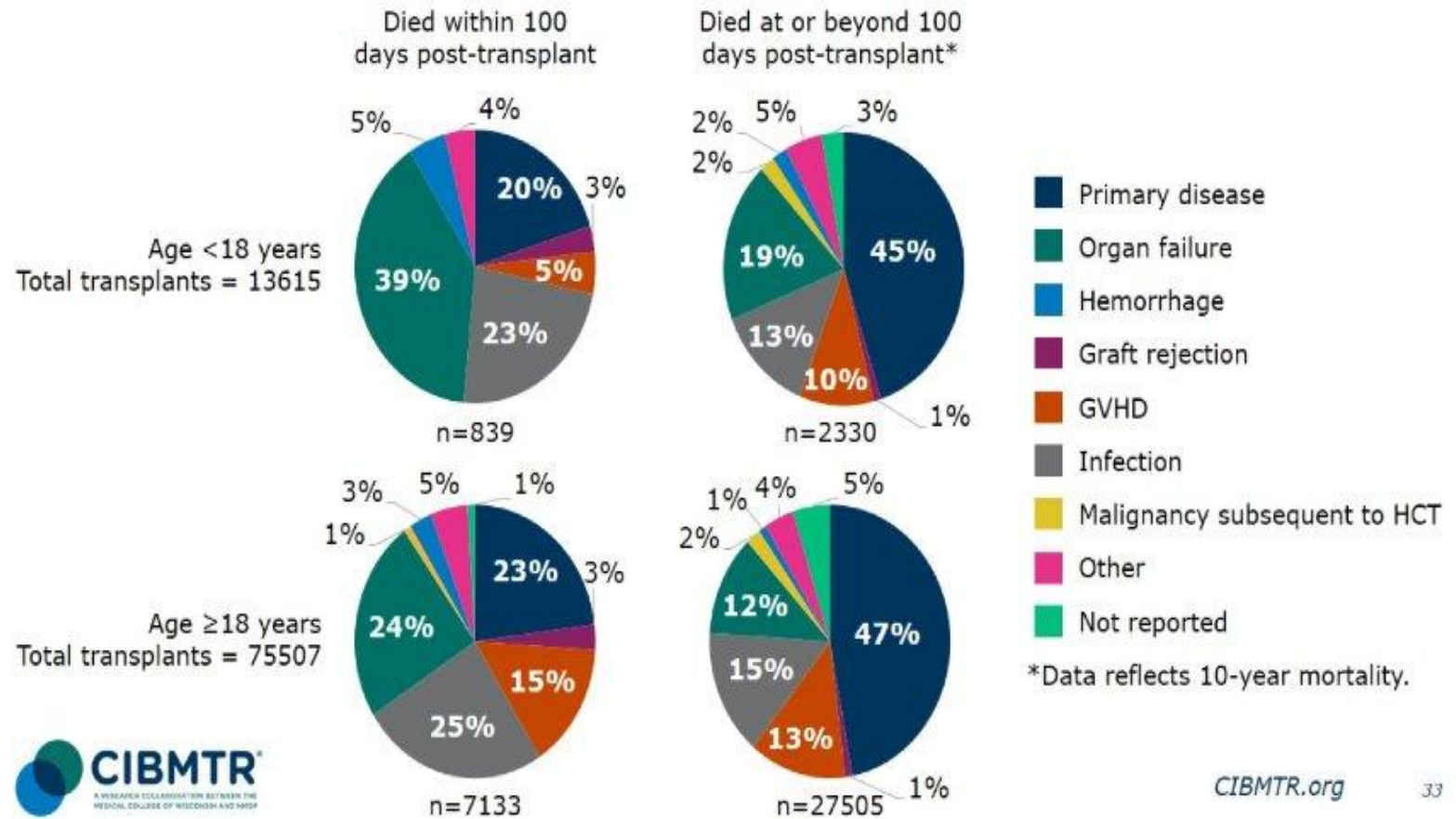
- 4174 HLA-identical sibling transplants for chronic myelogenous leukemia in first chronic phase
- TRM increased with increasing grades of aGVHD
- Increasing degrees of aGVHD reduced the risk of relapse



Problem statement

- Amongst all allogeneic transplants, 30-50% patients develop GVHD
- Severe acute GVHD in 14%

Causes of Death after Allogeneic HCTs in the US, 2012-2022



Strategies to prevent or mitigate aGVHD

- Post-transplant Immunosuppression
 - Antimetabolites
 - Methotrexate – cytotoxic
 - Mycophenolate Mofetil (MMF) – prevention of synthesis/proliferation/function of T/B lymphocytes (↓ purine synthesis)
 - Calcineurin inhibitors
 - Cyclosporine
 - Tacrolimus
 - M-TOR inhibitor - sirolimus
 - High-dose cyclophosphamide (PTCy)
 - Antithymocyte globulin (ATG)
- T cell depletion
 - In vitro and In vivo
 - Pan T cell
 - Selective depletion – alpha/beta
- Novel –
 - use of immune checkpoint inhibitors
 - Inhibition of CTLA-4 pathway: abatacept
 - T cell signaling
 - Enhancing regulatory T cells (T-regs)
 - Invariant NK-T cells
 - Targeting T cell trafficking
 - Targeting cytokine pathways
 - Targeting tissue damage/endothelial pathway
 - T cell modulation

Decreasing the damage to the host

- This can be divided into giving less rigorous conditioning regimens or blocking the cytokines or other products that lead to the activation of APC's.
- Antibiotics have been shown to decrease the frequency of acute GVHD.
- TNF-alpha inhibitors have been tried to decrease GVHD
- Holler et al were able to postpone the development of acute GVHD but not reduce the rate with the use of prophylactic TNF-alpha inhibitors.

Stopping activation and proliferation of donor lymphocytes

- Cyclosporine
- Methotrexate
- Tacrolimus
- Sirolimus
- Mycophenolate mofetil
- Steroids
- Alemtuzamab (Campath)
- ATG
- Abatacept

Methotrexate

- Methotrexate is a folate antimetabolite
- As a single agent significant GVHD develops in 70%
- It has found a significant role in prevention when used in combination with other immune suppressant medications

Cyclosporine/Tacrolimus

- Cyclosporine/Tacrolimus is a calcineurine inhibitor
- Inhibition of calcineurine prevents the transcription of NFAT and many cytokines (IL-2, TNF-alpha, IL-3, IL-4) which decreases the proliferation of lymphocytes

Steroids

Comparison of CSA vs MTX

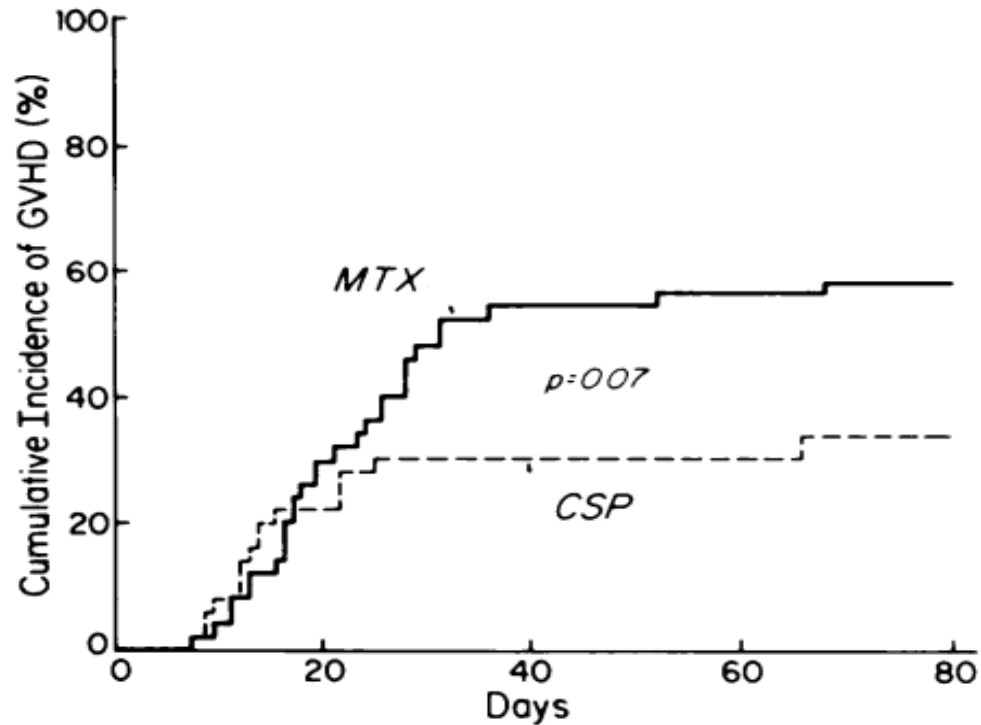


Fig 2. Probability of developing acute GVHD of grades II through IV in 75 patients with ANL in first remission given marrow grafts from HLA-identical siblings and either MTX (n = 39) or CSP (n = 36) after transplantation (Kaplan-Meier product limit estimates).

Fred Hutchinson Cancer Center

CSP reduced the incidence of GVHD as compared to MTX alone however there was no survival advantage

MTX - 15 mg/m² IV on day 1 and 10 mg/m² on days 3, 6, 11, 18, and 25, and then every two weeks until day 95

CSP, 1.5 mg/kg, was given IV (infused over one hour) every 12 hours until the patient had recovered from GI toxicity. CSP was then converted to an oral dose of 6.25 mg/kg every 12 hours

Deeg HJ, Storb R, Thomas ED, et al. Cyclosporine as prophylaxis for graft-versus-host disease: a randomized study in patients undergoing marrow transplantation for acute nonlymphoblastic leukemia. *Blood*. 1985;65(6):1325-1334.

Comparison of CSA+ MTX vs CSA

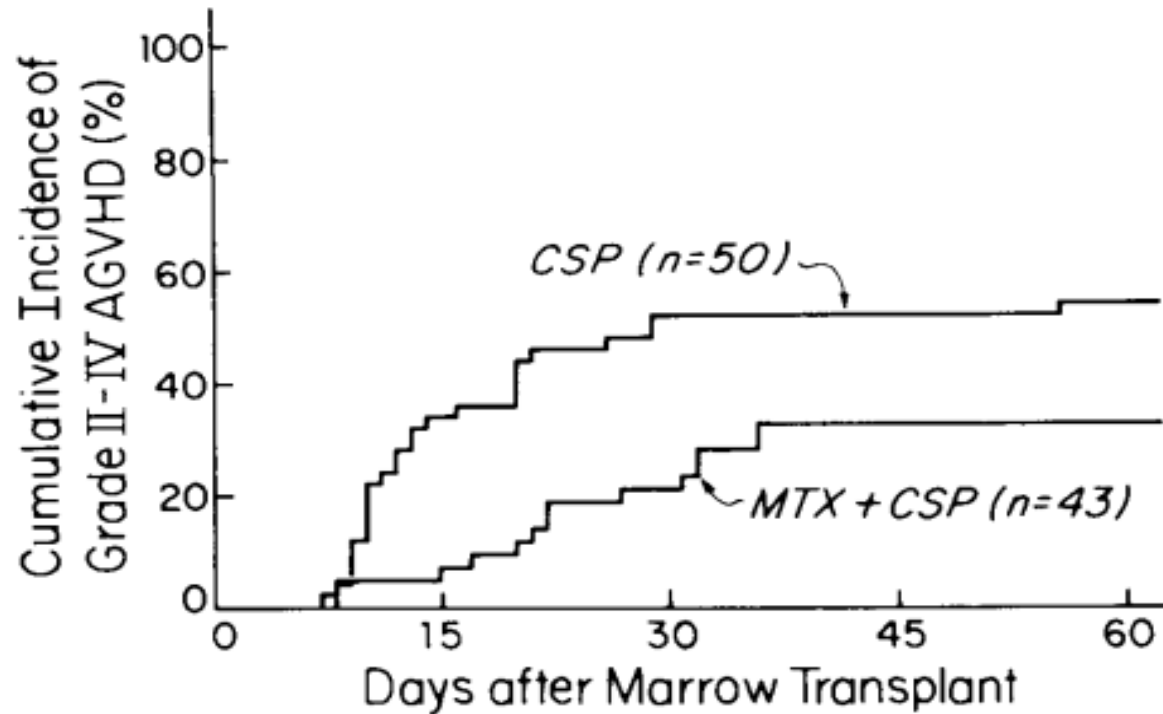


Figure 2. Probability of Grades II to IV Acute Graft versus Host Disease (AGVHD) in 93 Patients with Acute Nonlymphoblastic Leukemia in First Remission or Chronic Myelocytic Leukemia in the Chronic Phase, Given Marrow Grafts from HLA-Identical Siblings and Prophylaxis with Either Methotrexate and Cyclosporine (MTX + CSP) or Cyclosporine Alone (CSP).³²

The incidence of acute GVHD was significantly reduced with **cyclosporine + short course methotrexate – 33% reduction in grade II-IV acute GVHD** as compared to cyclosporine alone

Seven patients who received cyclosporine alone acquired grade IV acute graft versus host disease, as compared with none who received both methotrexate and cyclosporine.

CSA + METHOTREXATE vs TAC + METHOTREXATE in MRD

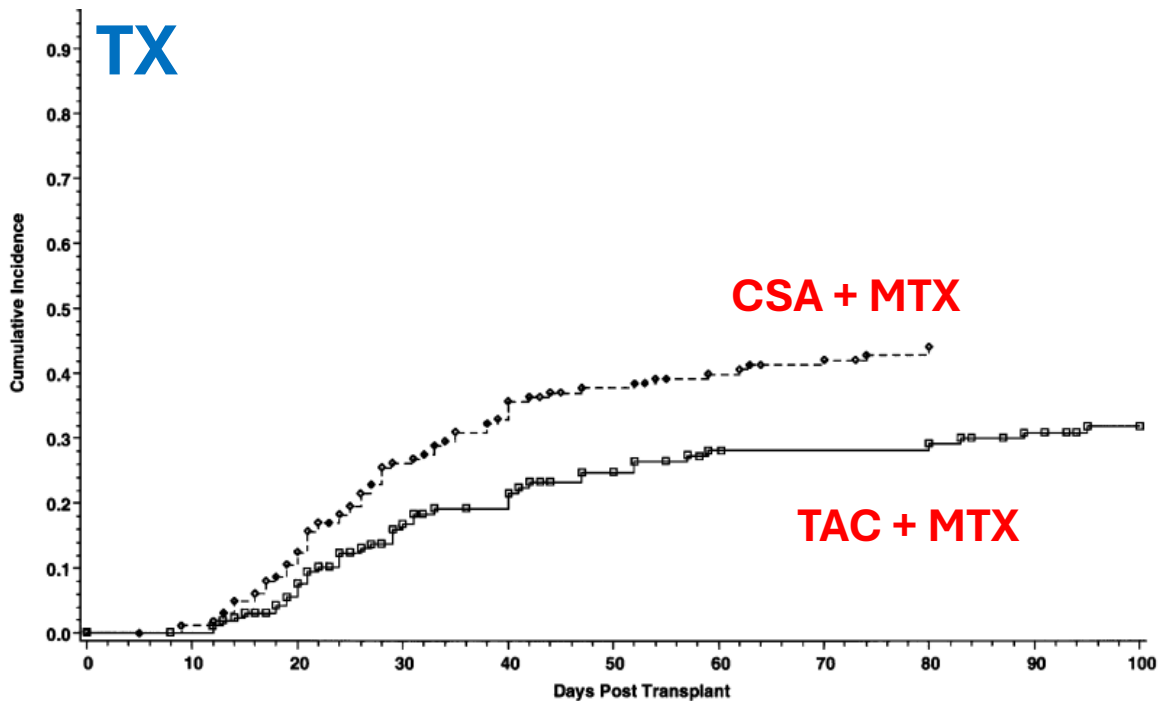


Fig 1. Cumulative incidence of grade II-IV acute GVHD of 165 patients who received cyclosporine/methotrexate, 44.4% (◇) and 164 patients who received tacrolimus/methotrexate, 31.9% (□); absolute difference = 12.5%, 95% CI = -23.9 to -1.2 (P = .01, Wilcoxon).

The incidence of grade III-IV acute GVHD was similar, 17.1% in cyclosporine group and 13.3% in the tacrolimus group.

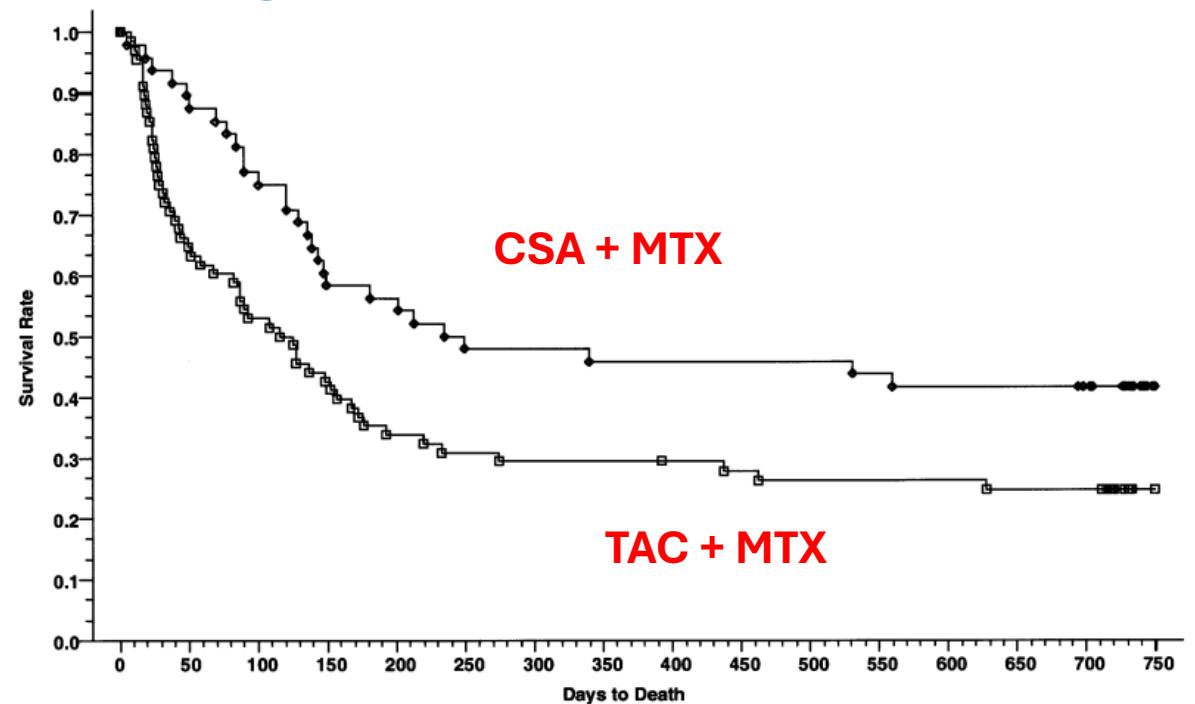


Fig 5. Overall survival at 2 years of patients with advanced disease; 48 patients received cyclosporine/methotrexate, 41.7% (◇) and 68 patients received tacrolimus/methotrexate, 24.8% (□); absolute difference = 16.9%, 95% CI = -34.3 to 0.4 (P = .006, Wilcoxon).

There was a higher frequency of deaths from regimen-related toxicity in patients with advanced disease who received tacrolimus.

Ratanatharathorn V, Nash RA, Przepiorka D, et al. Phase III study comparing methotrexate and tacrolimus (prograf, FK506) with methotrexate and cyclosporine for graft-versus-host disease prophylaxis after HLA-identical sibling bone marrow transplantation. *Blood*. 1998;92(7):2303-2314.

CSA + METHOTREXATE vs TAC + METHOTREXATE in MUD TX

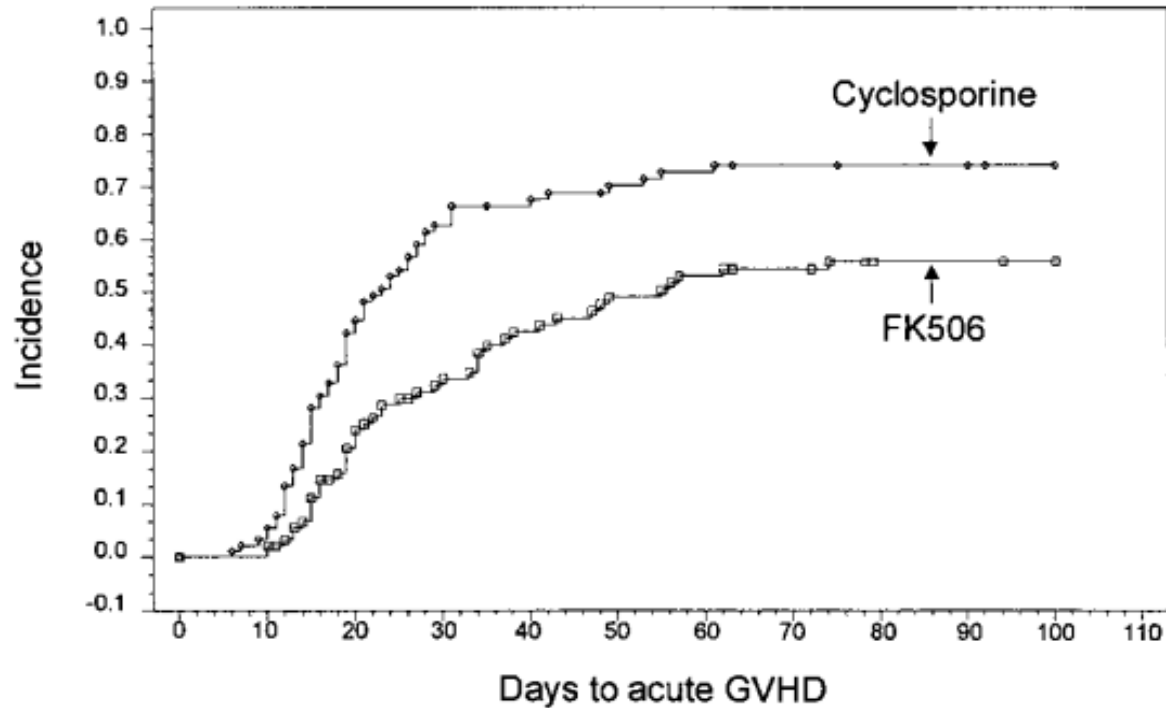


Figure 1. Kaplan-Meier estimate of acute GVHD based on site investigator assessment (tacrolimus, 56%; CSP, 74%; $P = .0002$).

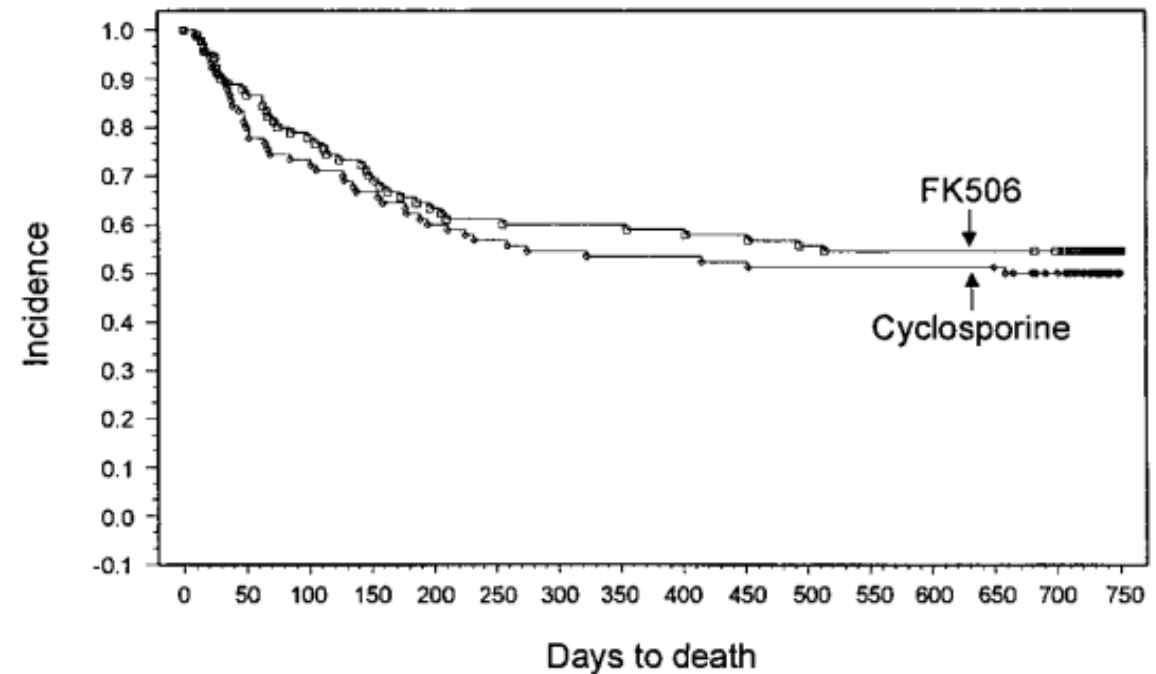
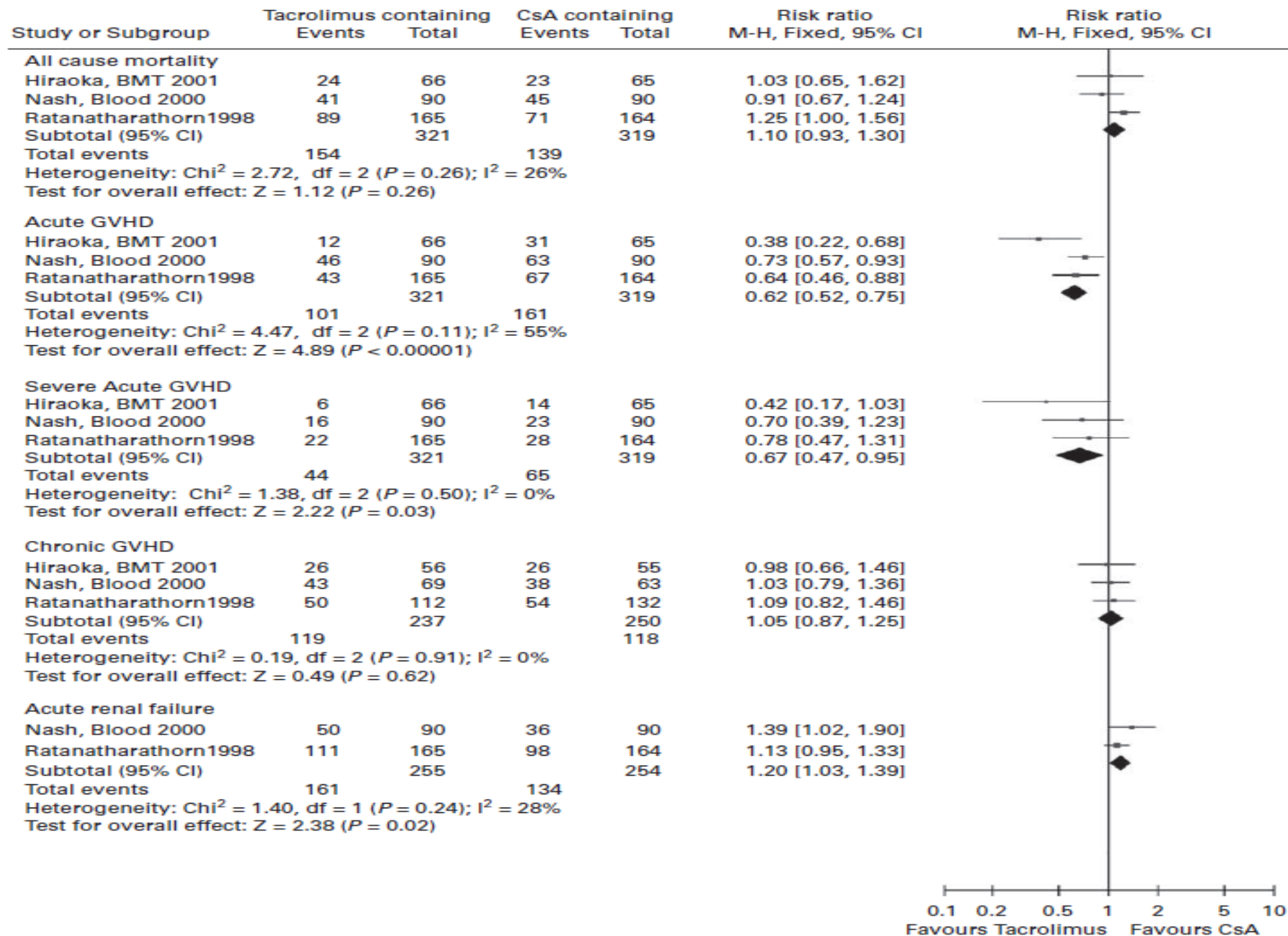


Figure 4. Kaplan-Meier estimate of survival (tacrolimus, 54%; CSP, 50%; $P = .46$).

Nash RA, Antin JH, Karanes C, et al. Phase 3 study comparing methotrexate and tacrolimus with methotrexate and cyclosporine for prophylaxis of acute graft-versus-host disease after marrow transplantation from unrelated donors. *Blood*. 2000;96(6):2062-2068.



Sirolimus

- M-TOR Inhibitor
- Sirolimus is an immune suppressant that appears to work synergistically with Tacrolimus.

TAC + METHOTREXATE Vs TAC + Sirolimus in MRD

TX

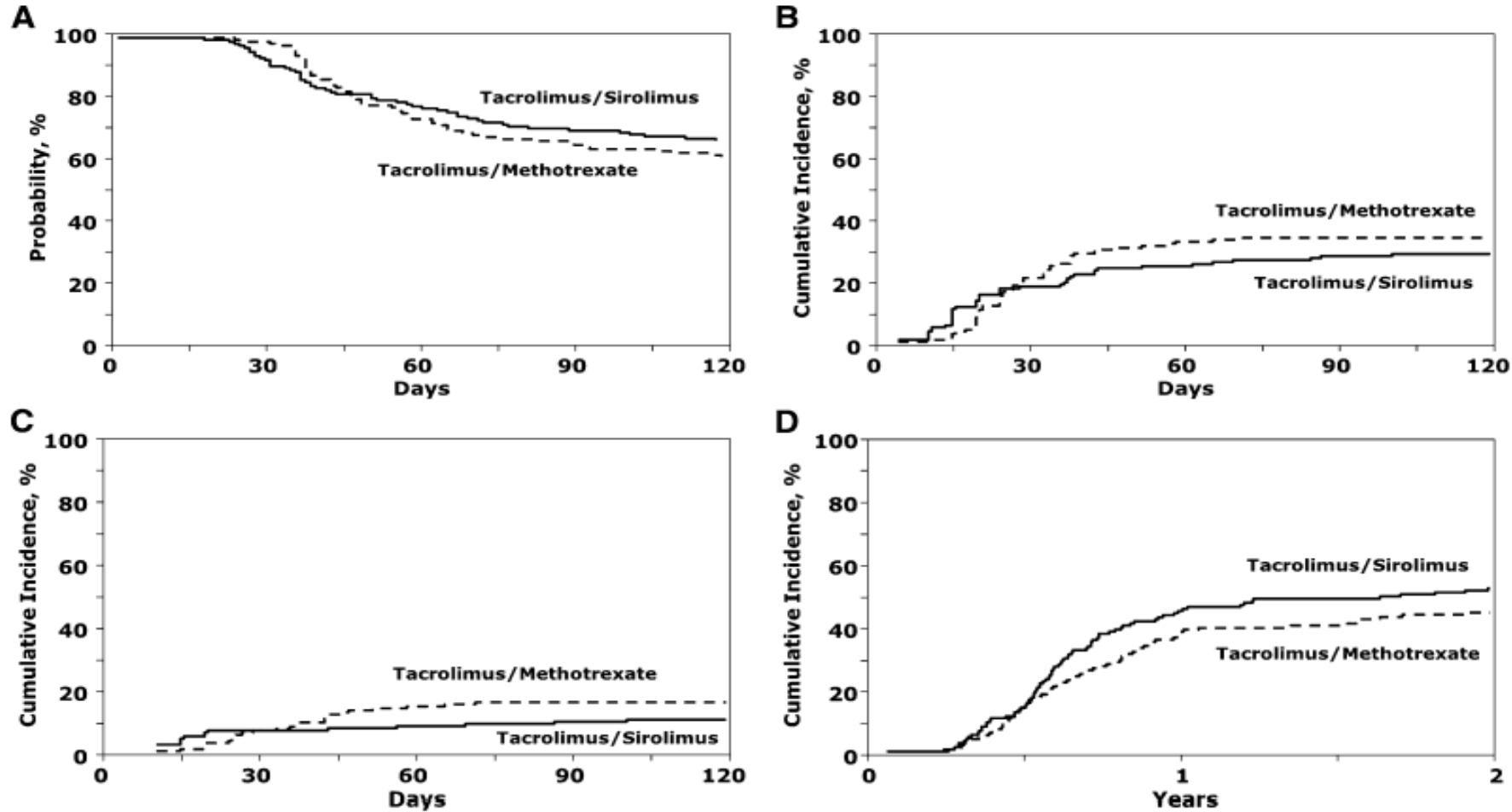


Figure 1. GVHD outcomes. (A) Grades 2-4 acute GVHD-free survival from randomization. (B) Cumulative incidence of grades 2-4 acute GVHD. (C) Cumulative incidence of grades 3-4 acute GVHD. (D) Cumulative incidence of chronic GVHD with death and relapse as competing risks.

Tacrolimus from day -3 at a dose of 0.02 mg/kg/day by continuous IV infusion, target serum concentration of 5 to 10 ng/mL
Sirolimus from day -3 with a 12 mg oral loading dose, followed by daily oral doses of 4 mg, target serum trough concentration of 3 to 12 ng/mL

Based on similar long-term outcomes, **more rapid engraftment, and less oropharyngeal mucositis**, the combination of Tac/Sir is an acceptable alternative to Tac/Mtx after MRD HCT.

Cutler C, Logan B, Nakamura R, et al. Tacrolimus/sirolimus vs tacrolimus/methotrexate as GVHD prophylaxis after matched, related donor allogeneic HCT. *Blood*. 2014;124(8):1372-1377.

Mycophenolate Mofetil

- IMPD inhibitor
- Lymphotoxic
- Doesnot cause mucositis unlike methotrexate containing regimens
- Commonly used regimen in RIT and cord transplants

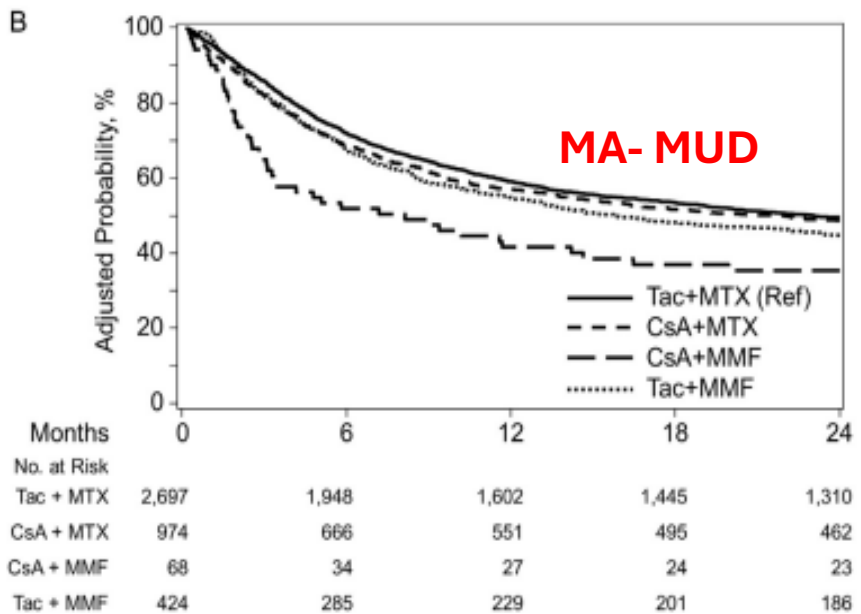
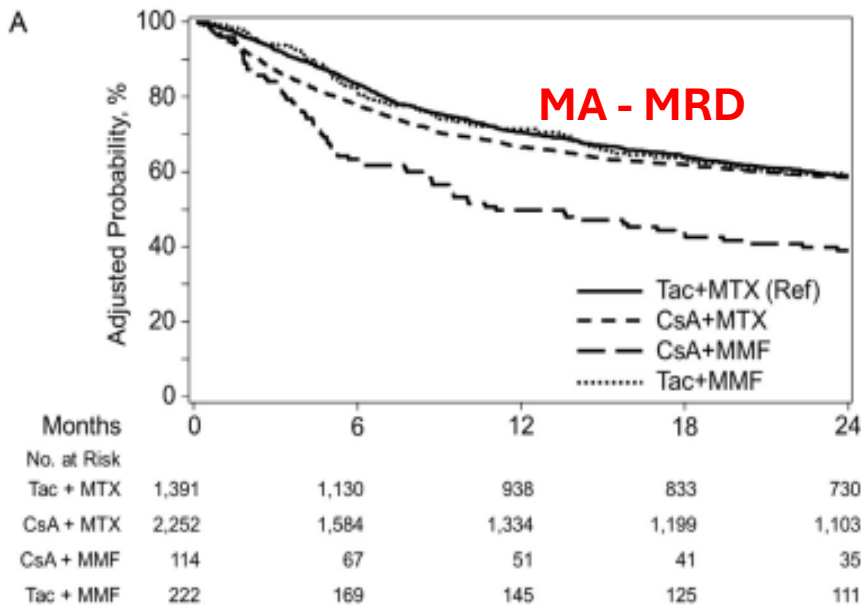


Table 4

Multivariate Outcomes in Myeloablative Related and URD HCT

Outcome	Tac+MTX	CSA+MMF	CSA+MTX	Tac+MMF
MRD, HR (95% CI), P value				
aGVHD grade II-IV		1.65 (1.24-2.20) P < .001	1.17 (1.04-1.33) P = .010	1.05 (.83-1.34) P = .661
aGVHD grade III-IV		1.92 (1.31-2.83) P < .001	1.13 (.92-1.37) P = .283	1.19 (.85-1.66) P = .319
cGVHD* <5 mo		1.34 (.83-2.17) P = .233	1.41 (1.20-1.67) P < .001	1.28 (.94-.74) P = .119
cGVHD* ≥5 mo		.56 (.34-.95) P = .030	.74 (.65-.85) P < .001	.92 (.71-1.19) P = .512
TRM* <5 mo	Ref (1.00)	4.03 (2.61-6.23) P < .001	2.29 (1.78-2.95) P < .001	1.17 (.71-1.94) P = .537
TRM* ≥5 mo		1.38 (.78-2.44) P = .275	.83 (.67-1.03) P = .095	.90 (.59-1.36) P = .609
Relapse		1.43 (1.05-1.93) P = .022	.86 (.74-.98) P = .029	.92 (.73-1.14) P = .444
OS* <5 mo		2.31 (1.73-3.09) P < .001	1.27 (1.10-1.48) P = .002	1.05 (.81-1.37) P = .704
OS* ≥5 mo		.94 (.62-1.43) P = .767	.74 (.64-.87) P < .001	.92 (.70-1.20) P = .530
URD, HR (95% CI), P value				
aGVHD, grade II-IV		1.49 (1.08-2.07) P = .016	1.00 (.90-1.12) P = .961	1.14 (.99-1.32) P = .066
aGVHD, grade III-IV		2.31 (1.57-3.42) P < .001	1.02 (.87-1.20) P = .806	1.26 (1.02-1.56) P = .030
cGVHD* <4 mo		1.54 (.79-2.98) P = .203	1.62 (1.35-1.93) P < .001	1.34 (1.04-1.73) P = .022
cGVHD* ≥4 mo		.92 (.52-1.63) P = .779	.85 (.74-.98) P = .025	1.47 (1.24-1.75) P < .001
TRM* <4 mo	Ref (1.00)	3.09 (2.00-4.77) P < .001	1.24 (1.02-1.51) P = .030	1.02 (.77-1.36) P = .896
TRM* ≥4 mo		.89 (.42-1.90) P = .763	1.00 (.84-1.20) P = .979	1.45 (1.16-1.81) P = .001
Relapse		.81 (.50-1.32) P = .398	.97 (.85-1.12) P = .699	.93 (.78-1.12) P = .449
OS* <4 mo		2.36 (1.67-3.35) P < .001	1.23 (1.08-1.41) P = .002	1.20 (1.01-1.44) P = .044
OS* ≥4 mo		1.10 (.64-1.89) P = .730	.91 (.79-1.05) P = .221	1.34 (1.12-1.61) P = .001

Bolded values represent statistically significant associations.

* Given differential effects over time (nonproportional hazards), models were constructed breaking the post-transplantation time course into 2 periods, using the maximized partial likelihood method to find the most appropriate breakpoint.

Hamilton BK, Liu Y, Hemmer MT, et al. Inferior Outcomes with Cyclosporine and Mycophenolate Mofetil after Myeloablative Allogeneic Hematopoietic Cell Transplantation. *Biol Blood Marrow Transplant*. 2019;25(9):1744-1755.

Figure 3. Adjusted OS by GVHD prophylaxis regimen in myeloablative MRD HCT (A) and myeloablative URD HCT (B).

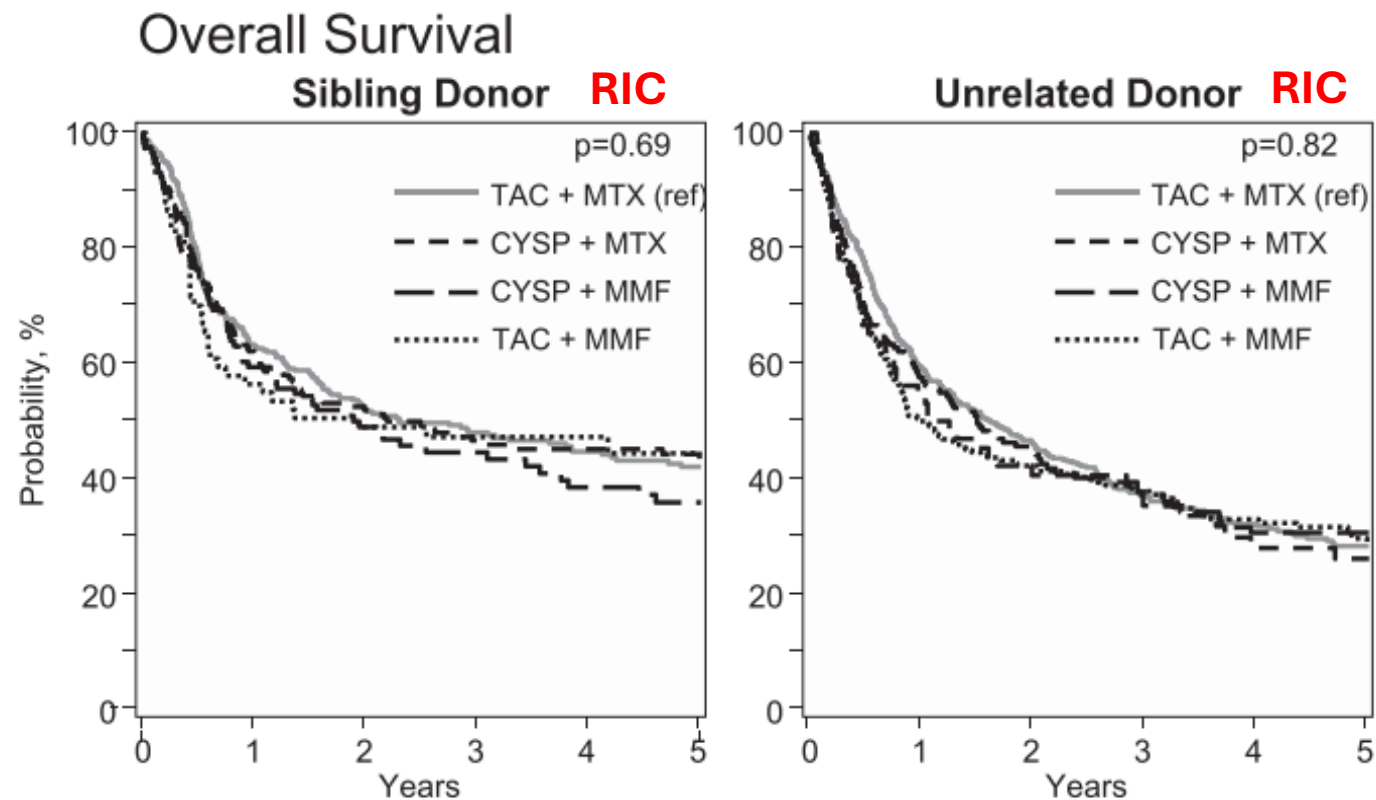


Figure 3. (A). Adjusted curves for OS in MRD RIC alloHCT patients receiving 1 of the 4 GVHD prophylaxis regimens: TAC-MTX, CYSP-MTX, CYSP-MMF, and TAC-MMF. **(B)** Adjusted curves for OS in URD RIC alloHCT patients receiving 1 of the 4 GVHD prophylaxis regimens: TAC-MTX, CYSP-MTX, CYSP-MMF, and TAC-MMF.

In the URD group, MMF-CYSP was associated with increased risk of grade II to IV acute GVHD (relative risk [RR], 1.78; $P < .001$) and grade III to IV acute GVHD (RR, 1.93; $P = .006$) compared with MTX-TAC.

In the URD group, use of MMF-TAC (versus MTX-TAC) lead to higher nonrelapse mortality. (hazard ratio, 1.48; $P = .008$).

Chhabra S, Liu Y, Hemmer MT, et al. Comparative Analysis of Calcineurin Inhibitor-Based Methotrexate and Mycophenolate Mofetil-Containing Regimens for Prevention of Graft-versus-Host Disease after Reduced-Intensity Conditioning Allogeneic Transplantation. *Biol Blood Marrow Transplant.* 2019;25(1):73-85.

CSA + Sirolimus + MMF Vs CSA + MMF

NMA conditioning MUD donors

5.0 mg/kg of cyclosporine orally twice daily from day -3, and tapered from day 96 through to day 150

15 mg/kg of mycophenolate mofetil orally TID from day 0 until day 30, then twice daily until day 150, and tapered off by day 180.

In the triple-drug group, mycophenolate mofetil doses were the same as in the standard group, but the drug was discontinued on day 40.

Sirolimus from day -3, orally at 2 mg once daily and adjusted to maintain trough concentrations between 3–12 ng/mL through to day 150, and (in the absence of GVHD) tapered off by day 180.

Overall survival at 4 years it was 64% in the triple-drug group (54–75) and 46% in the standard group (34–57%; HR 0.62 [0.40–0.97]; p=0.035).

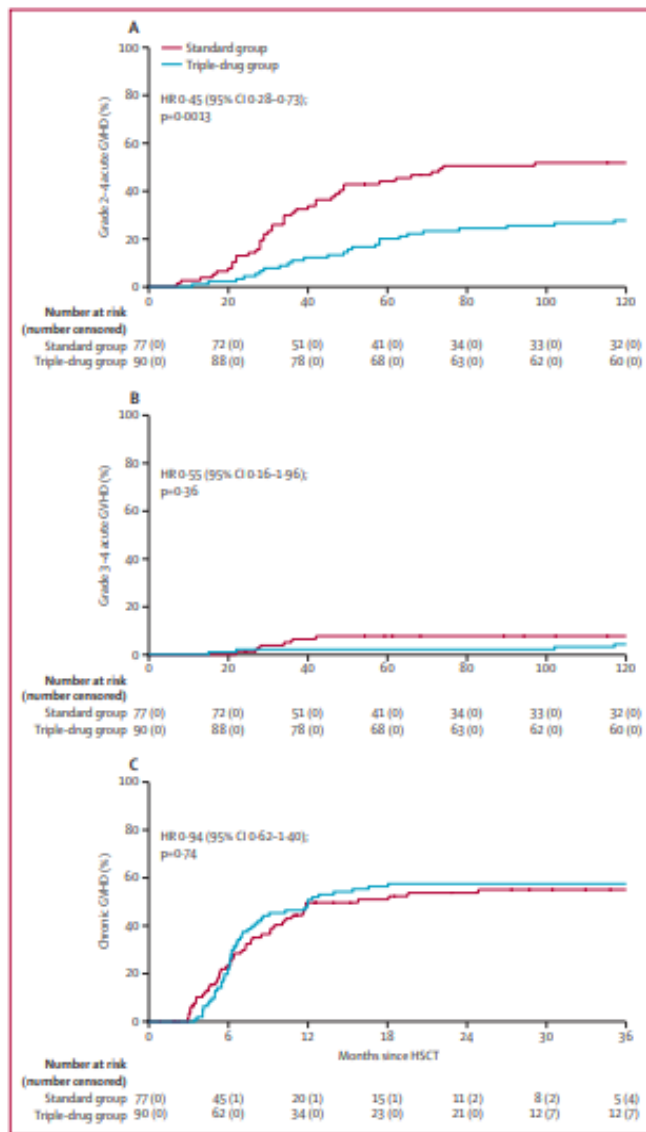
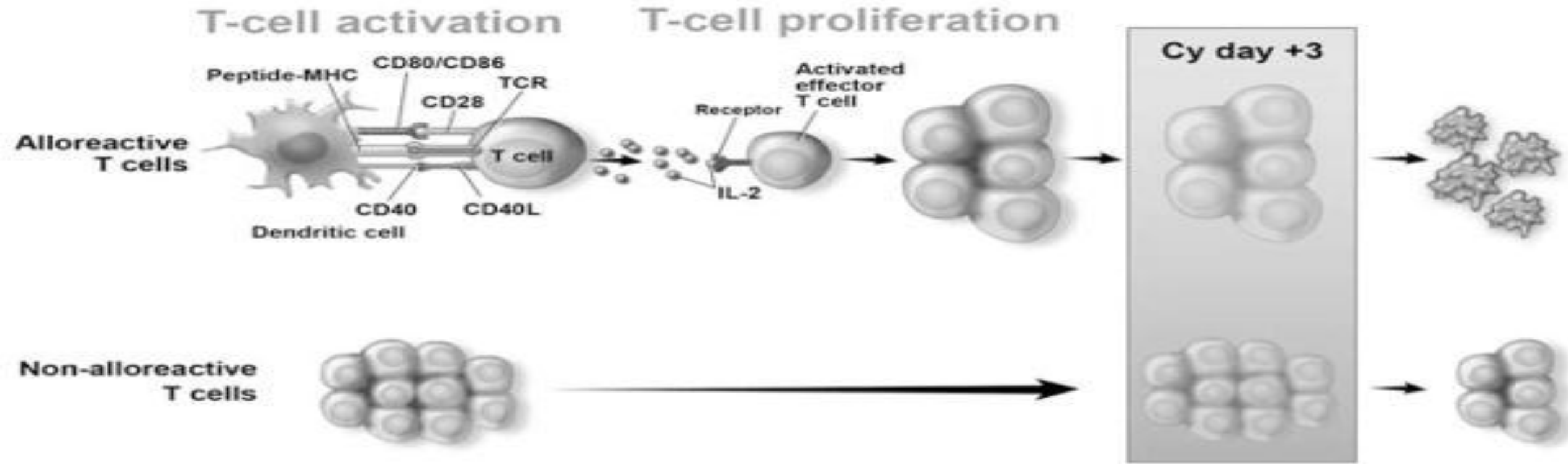


Figure 2: GVHD
Cumulative incidence of (A) grade 2–4 acute GVHD, (B) grade 3–4 acute GVHD, and (C) chronic GVHD by treatment group. GVHD=graft-versus-host disease. HSCT=haemopoietic stem cell transplantation.

Sandmaier BM, Kornblit B, Storer BE, et al. **Addition of sirolimus to standard cyclosporine plus mycophenolate mofetil-based graft-versus-host disease prophylaxis for patients after unrelated non-myeloablative haemopoietic stem cell transplantation: a multicentre, randomised, phase 3 trial.** *Lancet Haematol.* 2019;6(8):e409-e418.

- **Post Transplant Cyclophosphamide as GVHD Prophylaxis**



Based on the concept that

- a) Cyclophosphamide causes selective killing of alloreactive T cells – both anti-host and anti-donor [stem cells and resting memory T cells have high levels of ALDH1 – thus immune to Cy related damage]
- b) Development of peripheral tolerance
- c) Central deletion of donor HSC derived anti-host T cells in the thymus

POST TRANSPLANT CYCLOPHOSPHAMIDE

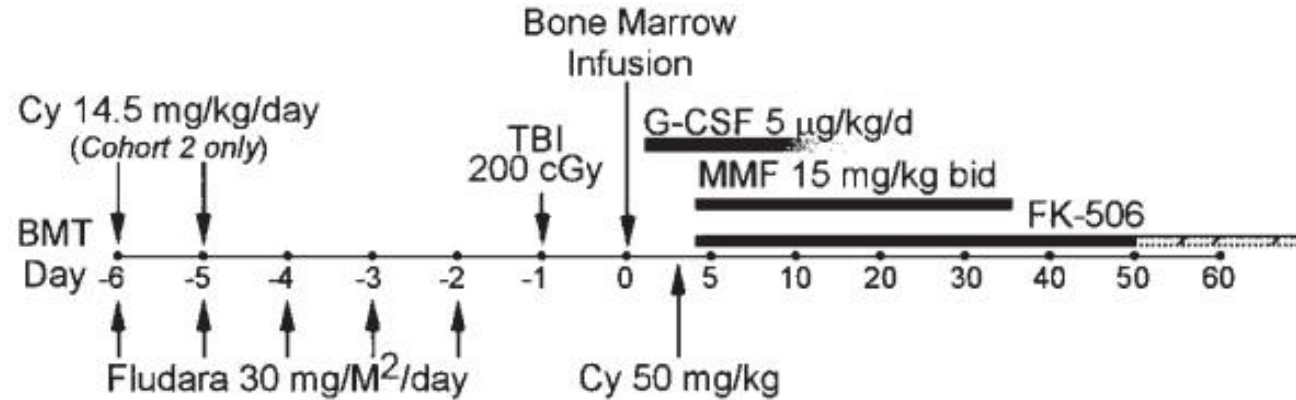


Figure 1. Treatment schema. All patients received fludarabine from days -6 to -2, 200 cGy TBI on day -1, a haploidentical bone marrow infusion on day 0, Cy on day 3, and MMF on days 4 to 35. Granulocyte colony-stimulating factor (G-CSF) treatment was discontinued when the ANC exceeded 1000/ μ L for 3 consecutive days. All patients in cohort 2 received Cy on days -6 and -5; the last 2 patients of this cohort received tacrolimus beyond day 100.

O'Donnell PV, Luznik L, Jones RJ, et al. Nonmyeloablative bone marrow transplantation from partially HLA-mismatched related donors using posttransplantation cyclophosphamide. *Biol Blood Marrow Transplant.* 2002;8(7):377-386.

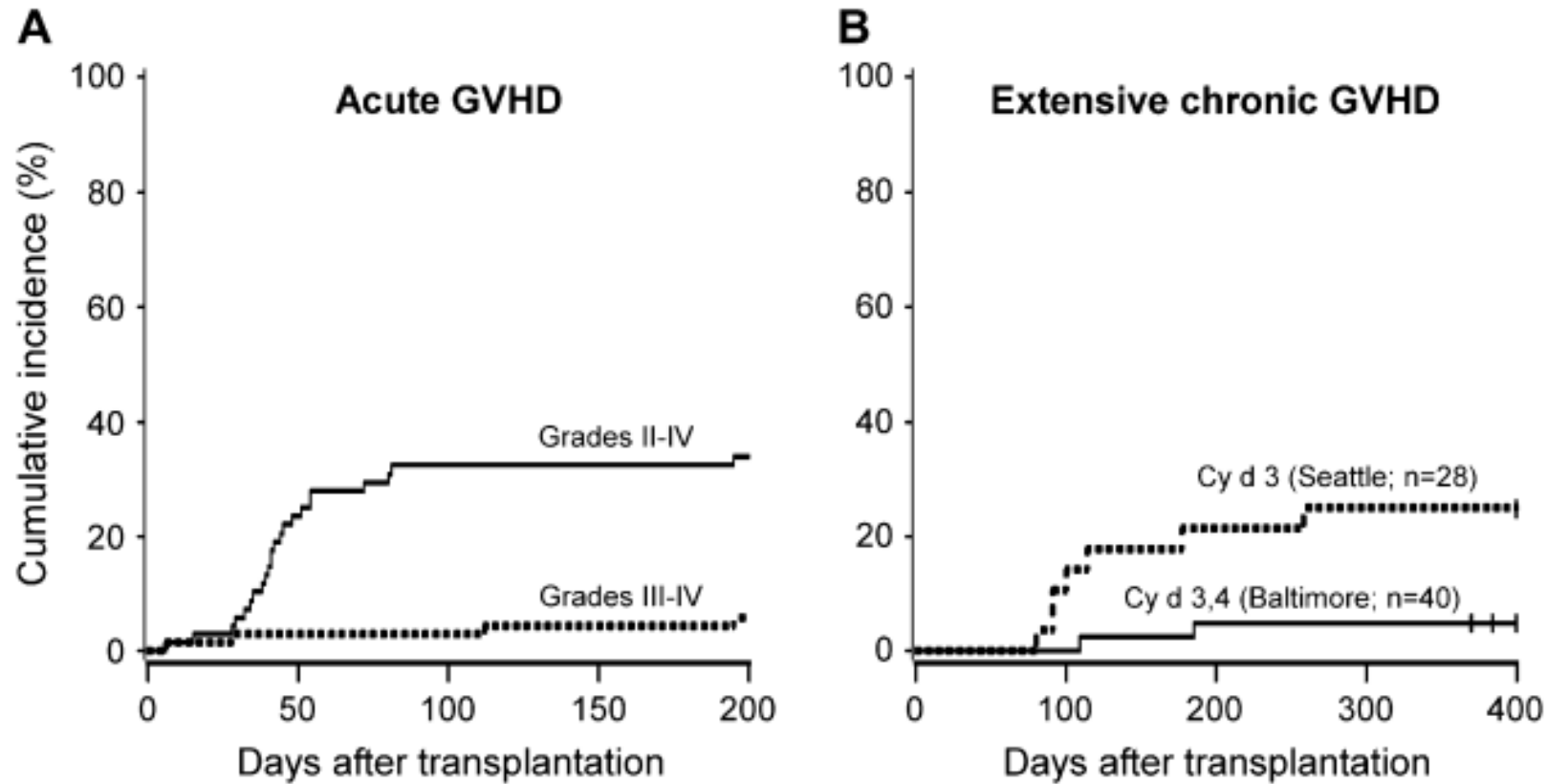


Figure 3. Cumulative incidence of aGVHD and cGVHD. (A) Cumulative incidence of aGVHD grades II-IV and III-IV. (B) Cumulative incidence of extensive cGVHD for patients who received 1 (Seattle) versus 2 (Baltimore) doses of posttransplant Cy.

Luznik L, O'Donnell PV, Symons HJ, et al. HLA-haploidentical bone marrow transplantation for hematologic malignancies using nonmyeloablative conditioning and high-dose, posttransplantation cyclophosphamide. *Biol Blood Marrow Transplant.* 2008;14(6):641-650.

ORIGINAL ARTICLE

Post-Transplantation Cyclophosphamide-Based Graft-versus-Host Disease Prophylaxis

J. Bolaños-Meade, M. Hamadani, J. Wu, M.M. Al Malki, M.J. Martens, L. Runaas, H. Elmariah, A.R. Rezvani, M. Gooptu, K.T. Larkin, B.C. Shaffer, N. El Jurdi, A.W. Loren, M. Solh, A.C. Hall, A.M. Alousi, O.H. Jamy, M.-A. Perales, J.M. Yao, K. Applegate, A.S. Bhatt, L.S. Kean, Y.A. Efebera, R. Reshef, W. Clark, N.L. DiFronzo, E. Leifer, M.M. Horowitz, R.J. Jones, and S.G. Holtan, for the BMT CTN 1703 Investigators*

June 2023

Phase 3 randomised trial

MRD and MUD transplants

Reduced Intensity Conditioning Regimens

Experimental Arm- PTCY followed by Tacrolimus and MMF-214 patients

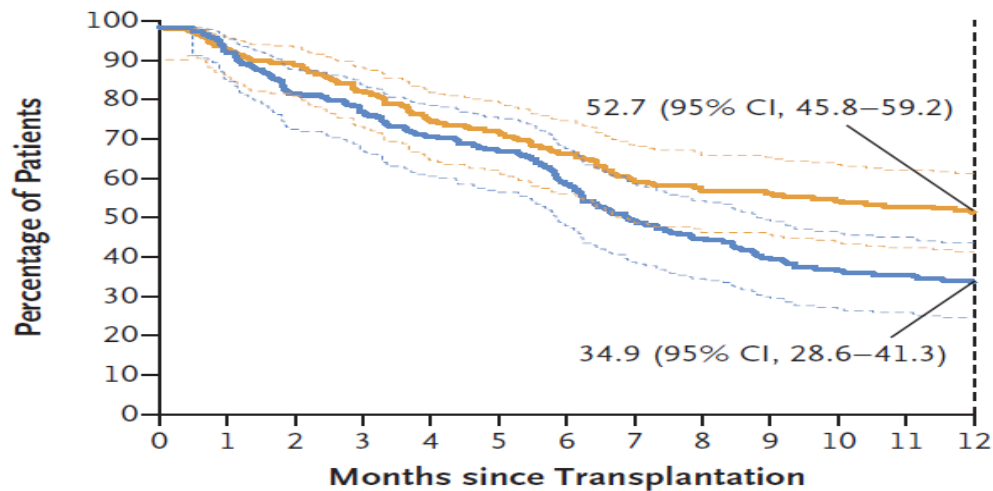
Control Arm- Tacrolimus and Methotrexate-217 patients

Primary End Point- GVHD Free, Relapse Free Survival at 1 year

Events: Grade 3/4 aGVHD, cGVHD requiring systemic immune

suppression, relapse or progression, death

A Adjusted GVHD-free, Relapse-free Survival

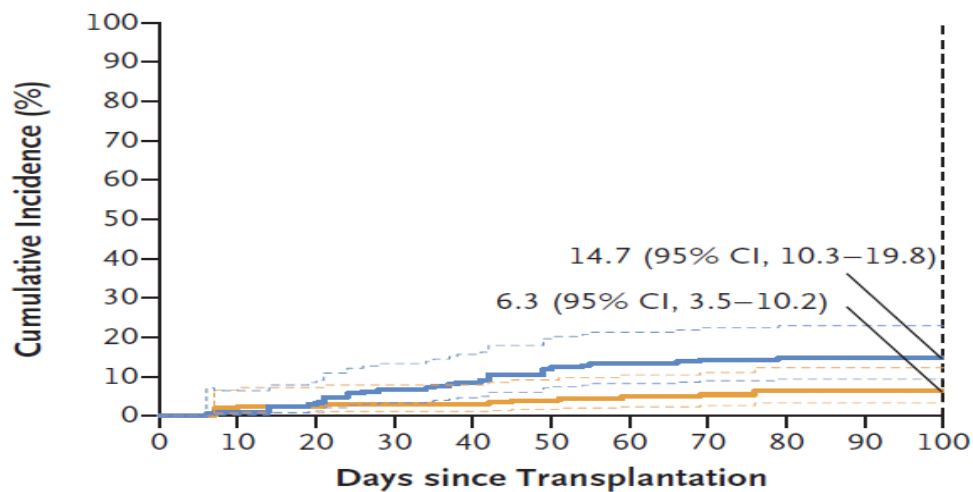


No. at Risk

Experimental prophylaxis	214	197	187	172	155	149	138	123	117	116	112	109	24
Standard prophylaxis	217	199	174	164	150	142	125	106	97	87	80	78	14

— Experimental prophylaxis
— Standard prophylaxis

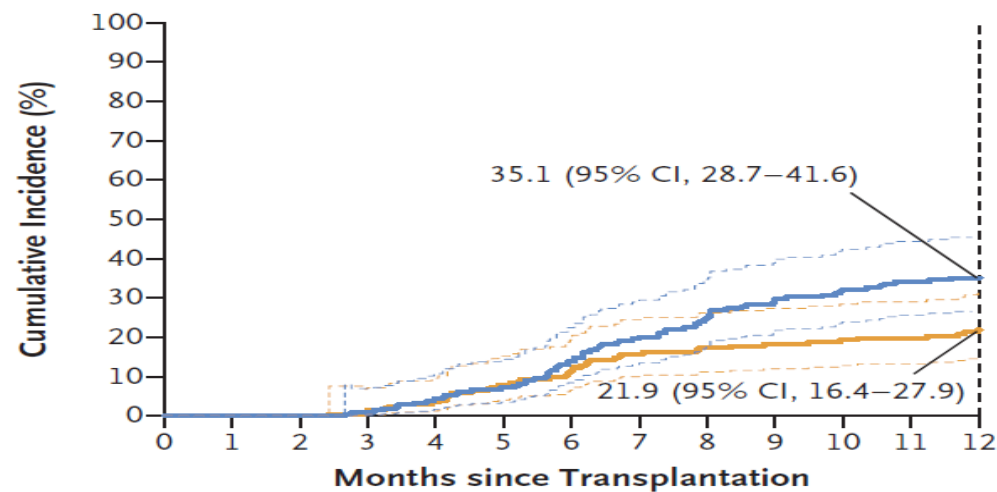
B Acute GVHD, Grade III or IV



No. at Risk

Experimental prophylaxis	208	203	199	195	192	190	186	185	181	179	176
Standard prophylaxis	212	209	204	193	187	178	174	172	170	170	169

C Chronic GVHD



No. at Risk

Experimental prophylaxis	208	200	195	186	175	164	154	139	131	127	123	121	30
Standard prophylaxis	212	207	198	193	179	169	148	129	116	105	94	88	17

ORIGINAL ARTICLE

Graft-versus-Host Disease Prophylaxis with Cyclophosphamide and Cyclosporin

D.J. Curtis,^{1,2} S.S. Patil,¹ J. Reynolds,^{1,2} D. Purtill,³ C. Lewis,⁴ D.S. Ritchie,⁵ D.J. Gottlieb,⁶⁻⁸ D.T. Yeung,⁹ E. Wong,¹⁰ S.-K. Tey,¹¹⁻¹³ T. Perera,¹⁴ J. Moore,¹⁵ R.M. Koldej,¹⁶ R. De Abreu Lourenco,¹⁷ J. Stubbs,¹⁸ C.O. Morrissey,¹⁹ N. Munsef,¹⁸ A. Arenas,¹⁸ and G.R. Hill,^{13,20,21} for the Australasian Leukaemia and Lymphoma Group*

July 2025

Phase 3 Randomised Study

Matched Related Donors

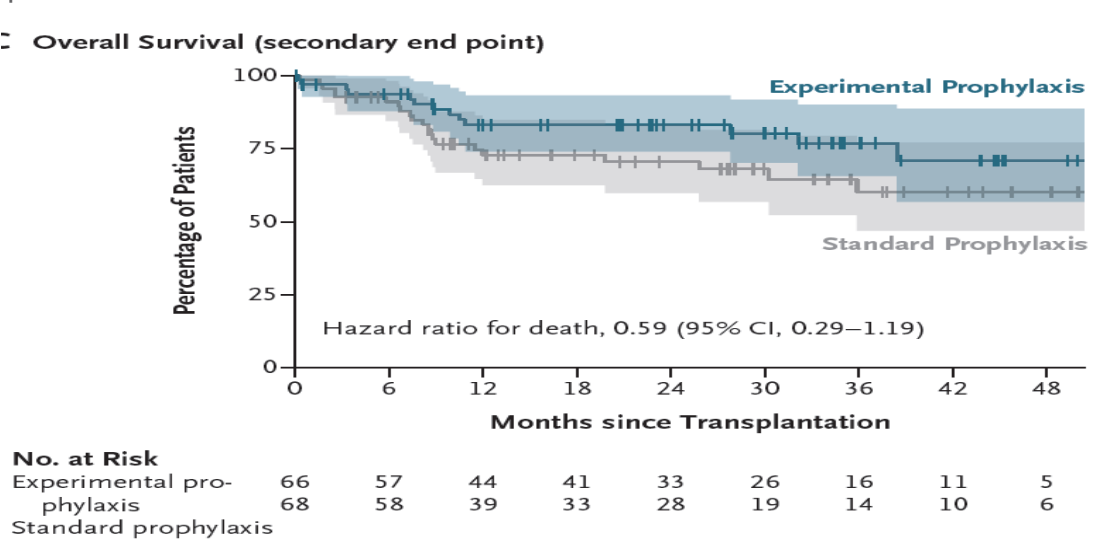
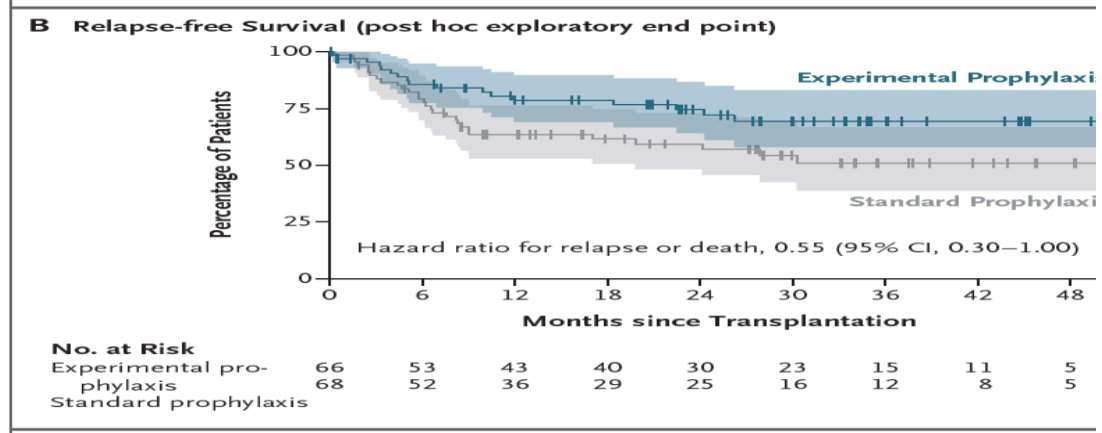
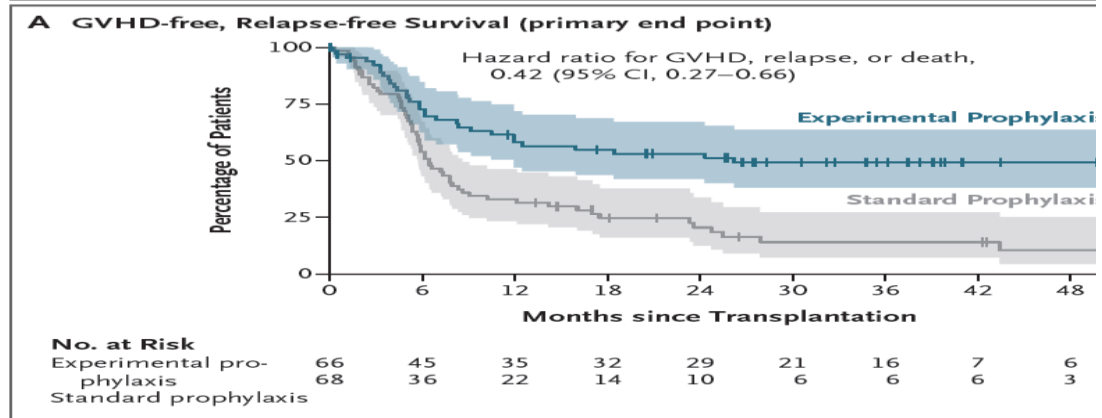
Experimental Arm – PTCY plus Cyclosporin – 66 patients

Control Arm- Cyclosporin and Methotrexate- 68 patients

Conditioning Regimens- Myeloablative and RIC

Primary End Point- GVHD Free Relapse Free Survival

Events: Grade 3/4 aGVHD, moderate to severe cGVHD , relapse or progression or death



	Experimental prophylaxis	Standard prophylaxis
GRFS Percentile	Months (95% Confidence Interval)	
50	26.2 (9.1-not reached)	6.4 (5.6-8.3)
GRFS Months	Percentage (95% Confidence Interval)	
12	58 (45-69)	33 (22-44)
24	53 (40-64)	20 (11-32)
36	49 (36-61)	14 (6-25)

ATG in GVHD Prophylaxis

Antilymphocyte Globulin for Prevention of Chronic Graft-versus-Host Disease

Nicolaus Kröger, M.D., Carlos Solano, M.D., Christine Wolschke, M.D., Giuseppe Bandini, M.D., Francesca Patriarca, M.D., Massimo Pini, M.D., Arnon Nagler, M.D., Carmine Selleri, M.D., Antonio Risitano, M.D., Ph.D., Giuseppe Messina, M.D., Wolfgang Bethge, M.D., Jaime Pérez de Oteiza, M.D., Rafael Duarte, M.D., Angelo Michele Carella, M.D., Michele Cimminiello, M.D., Stefano Guidi, M.D., Jürgen Finke, M.D., Nicola Mordini, M.D., Christelle Ferra, M.D., Jorge Sierra, M.D., Ph.D., Domenico Russo, M.D., Mario Petrini, M.D., Giuseppe Milone, M.D., Fabio Benedetti, M.D., Marion Heinzelmänn, Domenico Pastore, M.D., Manuel Jurado, M.D., Elisabetta Terruzzi, M.D., Franco Narni, M.D., Andreas Völp, Ph.D., Francis Ayuk, M.D., Tapani Ruutu, M.D., and Francesca Bonifazi, M.D.

January 2016

Phase 3 randomized study

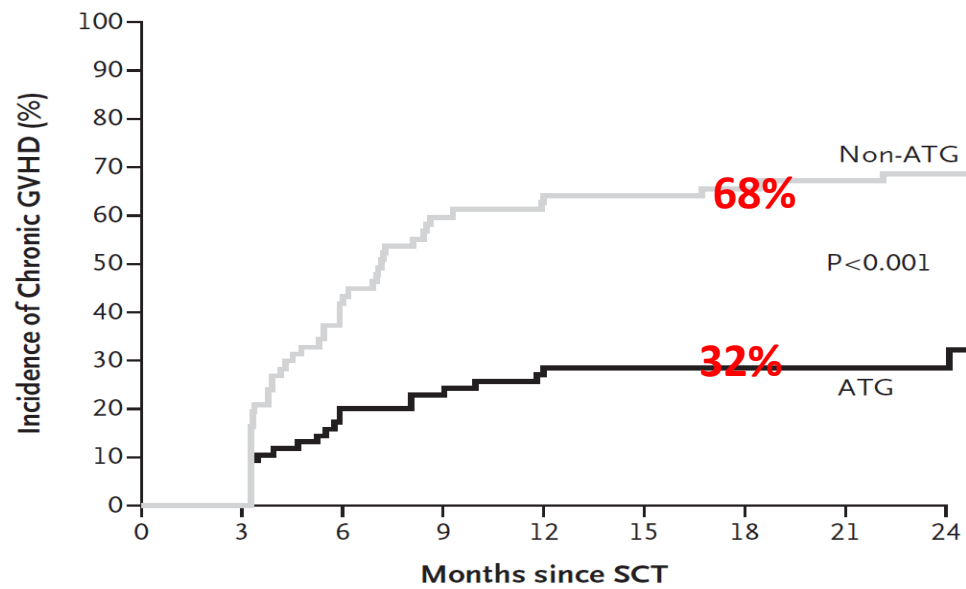
MRD transplants in acute leukemia patients

Myeloablative conditioning regimens

Experimental arm- ATG (Fresenius) -10mg/kg -3 days + CSA+ MTX-83 patients

Control Arm- CSA + MTX- 72 patients

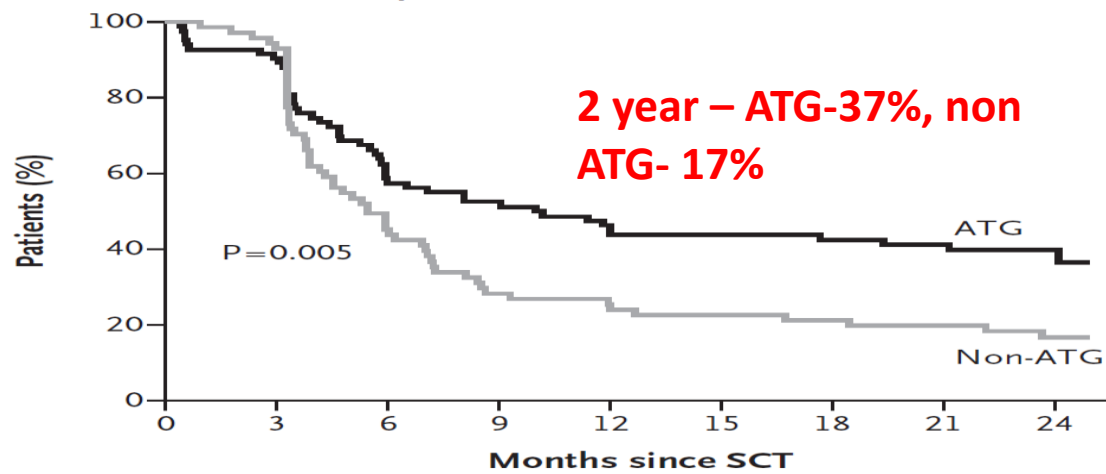
Primary End Point- Cumulative Incidence of cGVHD at 2 years



No. at Risk	0	3	6	9	12	15	18	21	24
ATG	83	78	55	46	42	40	38	38	25
Non-ATG	72	68	34	23	21	20	18	17	9

Figure 1. Cumulative Incidence of Chronic GVHD, According to Treatment Group.

F Chronic GVHD-free+Relapse-free Survival



No. at Risk	0	3	6	9	12	15	18	21	24
ATG	83	76	47	42	37	35	34	34	22
Non-ATG	72	67	32	21	19	17	16	15	8

Variable	ATG Group (N=83)	Non-ATG Group (N=72)	P Value*
Graft failure — no. (%)	0	1 (1.4)	
Days to engraftment — median (range)			
Absolute neutrophil count $\geq 0.5 \times 10^9$ /liter	18 (10–31)	15 (11–34)	<0.001
Platelet count $\geq 20 \times 10^9$ /liter	20 (10–110)	13 (6–29)	<0.001
Infectious complication — no. (%)	48 (57.8)	39 (54.2)	0.65
Cytomegalovirus reactivation — no. (%)	18 (21.7)	18 (25.0)	0.63
Epstein-Barr virus reactivation — no. (%)	3 (3.6)	1 (1.4)	0.38
Post-transplantation lymphoproliferative disorder — no. (%)	0	0	
Pulmonary infection — no. (%)	6 (7.2)	10 (13.9)	0.18
Fungal infection — no. (%)	3 (3.6)	3 (4.2)	0.86
Acute GVHD within 100 days after transplantation — no. (%)	21 (25.3)	25 (34.7)	0.20
Overall grades of acute GVHD — no. (%)			0.15
0	62 (74.7)	46 (65.2)	
1	12 (14.5)	12 (16.7)	
2	7 (8.4)	7 (9.7)	
3	2 (2.4)	4 (5.6)	
4	0	2 (2.8)	
2–4	9 (10.8)	13 (18.1)	0.13
3 or 4	2 (2.4)	6 (8.3)	0.10

Prospective, Randomized, Double-Blind, Phase III Clinical Trial of Anti-T-Lymphocyte Globulin to Assess Impact on Chronic Graft-Versus-Host Disease-Free Survival in Patients Undergoing HLA-Matched Unrelated Myeloablative Hematopoietic Cell Transplantation

Robert J. Soiffer, Haesook T. Kim, Joseph McGuirk, Mitchell E. Horwitz, Laura Johnston, Mrinal M. Patnaik, Witold Rybka, Andrew Artz, David L. Porter, Thomas C. Shea, Michael W. Boyer, Richard T. Maziarz, Paul J. Shaughnessy, Usama Gergis, Hana Safah, Ran Reshef, John F. DiPersio, Patrick J. Stiff, Madhuri Vusirikala, Jeff Szer, Jennifer Holter, James D. Levine, Paul J. Martin, Joseph A. Pidala, Ian D. Lewis, Vincent T. Ho, Edwin P. Alyea, Jerome Ritz, Frank Glavin, Peter Westervelt, Madan H. Jagasia, and Yi-Bin Chen

Phase 3 randomized study

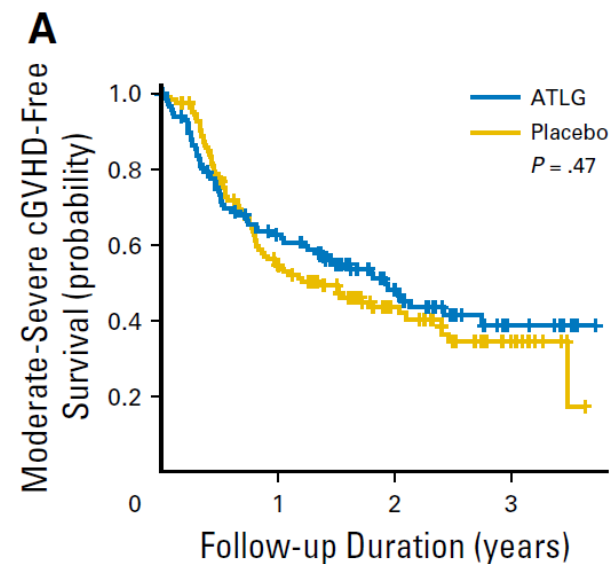
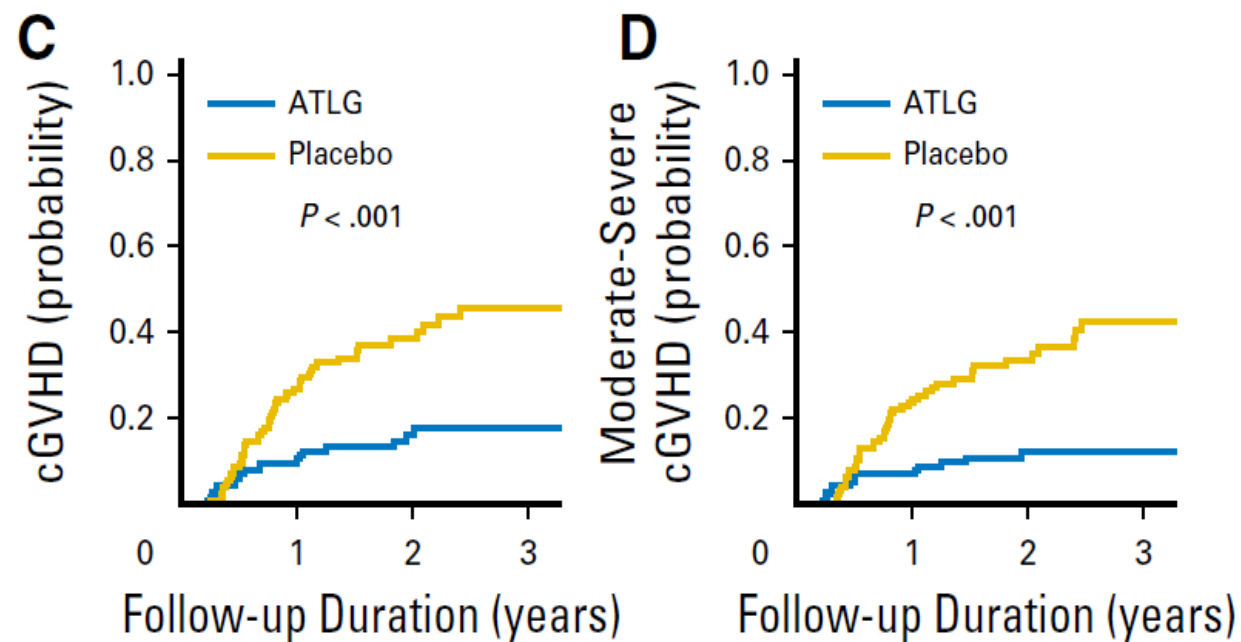
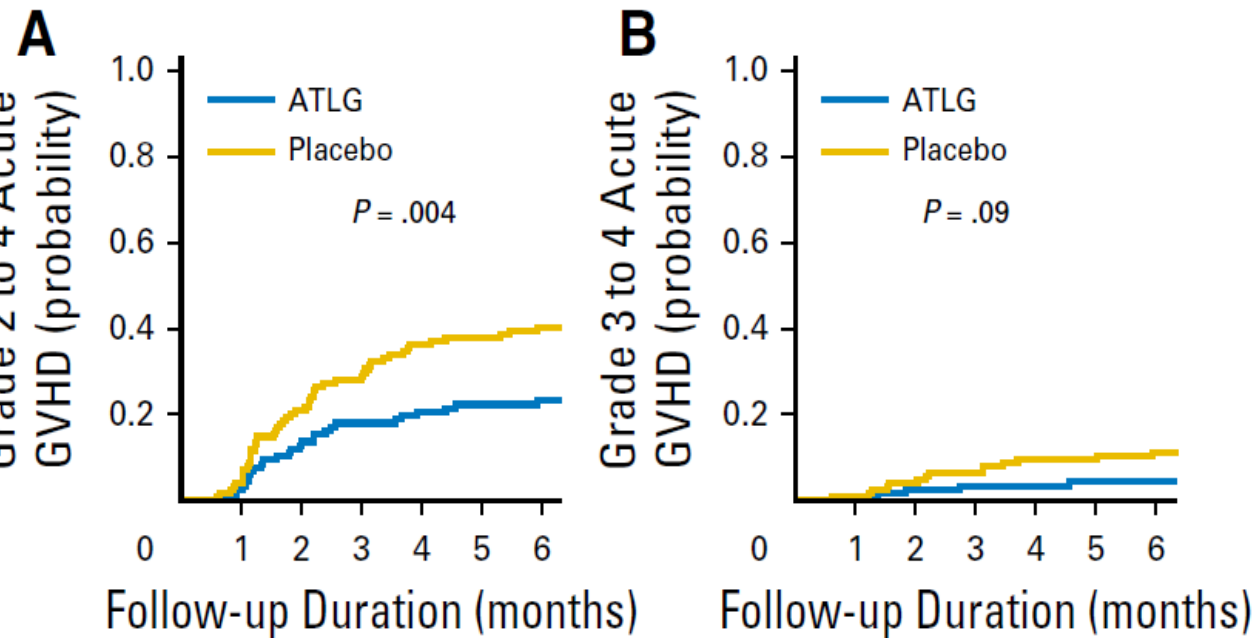
MUD transplants

Myeloablative conditioning regimens for AML/MDS

Experimental arm- ATG (Fresenius) 20mg/kg x 3 days + TAC+ MTX-128 patients

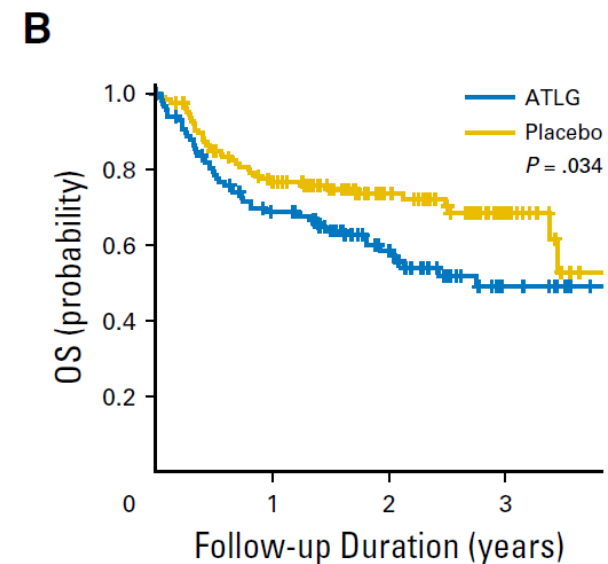
Control Arm- TAC + MTX-132 patients

Primary End Point- moderate-severe cGVHD free survival



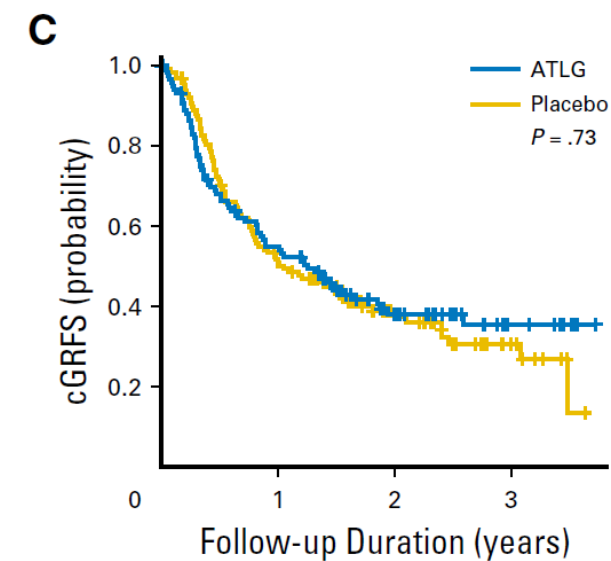
No. at risk:

ATLG	126	67	34	8
Placebo	128	65	27	9



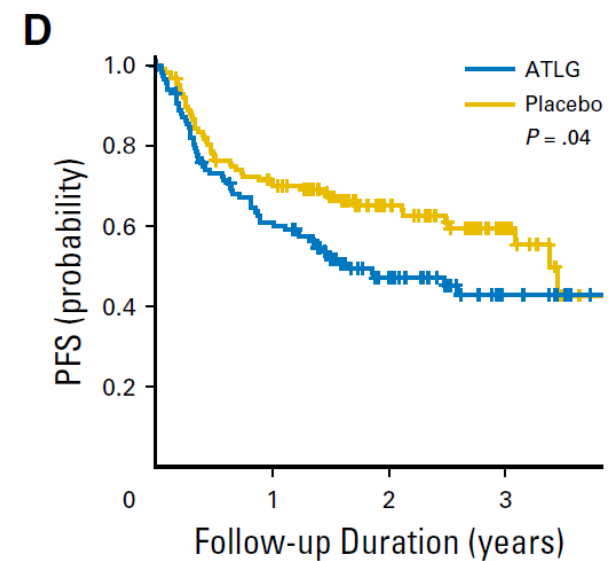
No. at risk:

ATLG	126	74	42	10
Placebo	128	91	51	17



No. at risk:

ATLG	126	62	29	8
Placebo	128	64	27	9



No. at risk:

ATLG	126	69	36	10
Placebo	128	88	50	17

Antithymocyte Globulin for Matched Sibling Donor Transplantation in Patients With Hematologic Malignancies: A Multicenter, Open-Label, Randomized Controlled Study

Ying-Jun Chang, PhD¹; De-Pei Wu, PhD, MD²; Yong-Rong Lai, MD³; Qi-Fa Liu, MD⁴; Yu-Qian Sun, MD¹; Jiong Hu, MD⁵; Yu Hu, MD⁶; Jian-Feng Zhou, MD, PhD⁷; Juan Li, PhD⁸; Shun-Qing Wang, MD⁹; Wei Li, MD¹⁰; Xin Du, MD, PhD¹¹; Dong-Jun Lin, MD¹²; Han-Yun Ren, MD¹³; Fang-Pin Chen, MD¹⁴; Yu-Hua Li, MD¹⁵; Xi Zhang, MD¹⁶; He Huang, MD, PhD¹⁷; Yong-Ping Song, MD, PhD¹⁸; Ming Jiang, MD¹⁹; Jian-Da Hu, MD, PhD²⁰; Ying-Min Liang, PhD, MD²¹; Jing-Bo Wang, PhD²²; Yang Xiao, MD²³; and Xiao-Jun Huang, MD¹

JCO, July 2020

Phase 3 randomized study

MRD transplants

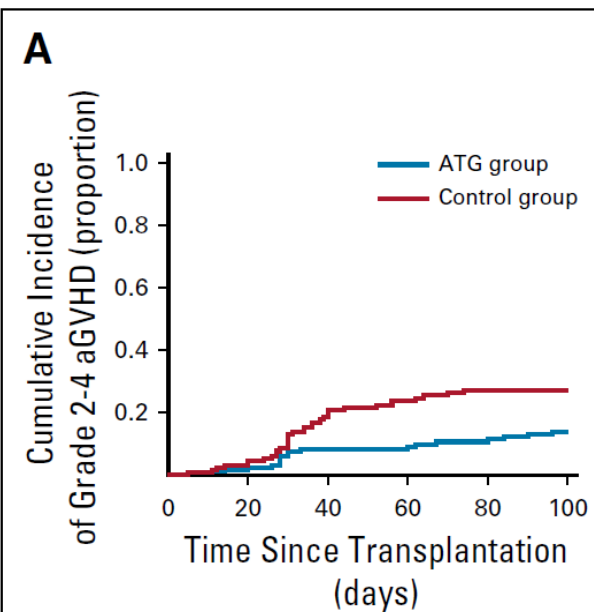
Myeloablative conditioning regimens for acute leukemias

Experimental arm- ATG (Thymoglobulin) - 4.5 mg/kg over 3 days + CSA+

MTX+MMF-132 patients

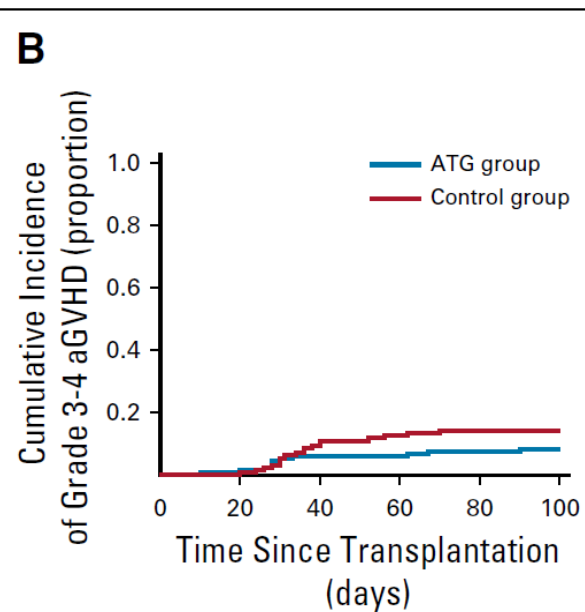
Control Arm- CSA + MTX+ MMF- 131 patients

Primary End Point- Grade 2-4, aGVHD at day 100



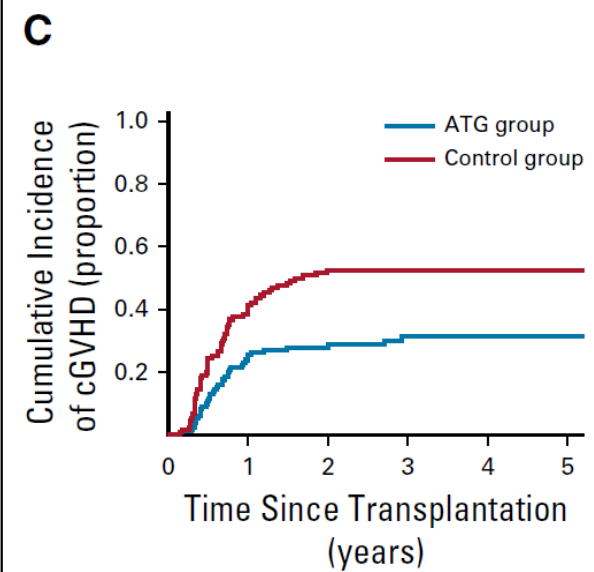
No. at risk:

ATG	132	130	121	120	115	110
Control	131	126	105	96	92	90



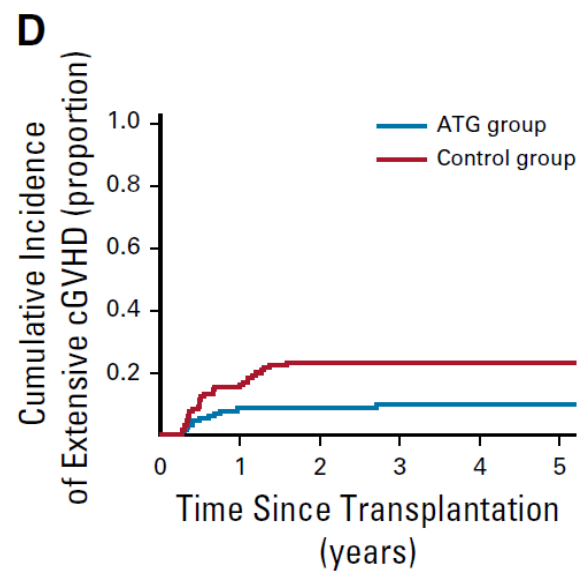
No. at risk:

ATG	132	131	123	122	118	115
Control	131	129	114	107	105	103



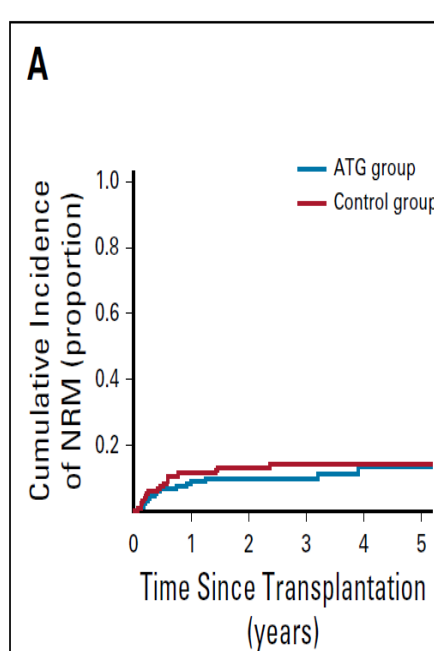
No. at risk:

ATG	132	74	59	34	24	8
Control	131	56	34	15	7	2



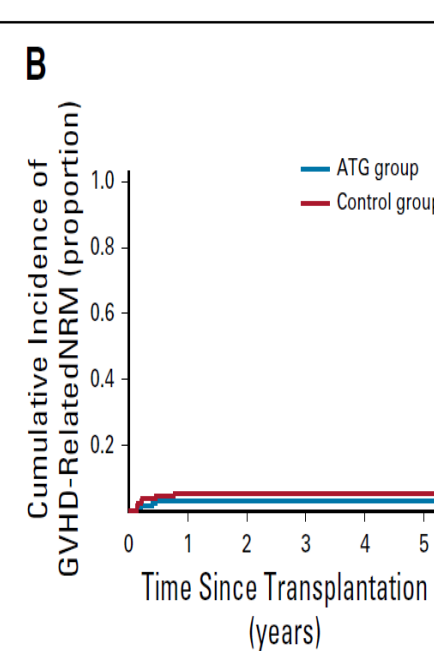
No. at risk:

ATG	132	90	75	42	29	8
Control	131	83	66	36	20	5



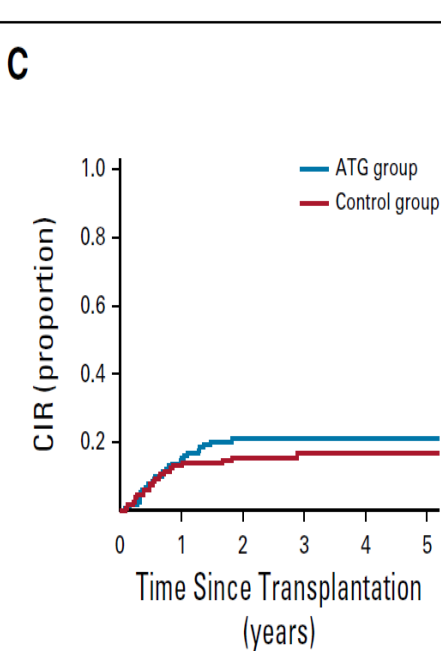
No. at risk:

ATG	132	103	83	49	35	9
Control	131	100	88	50	2	9



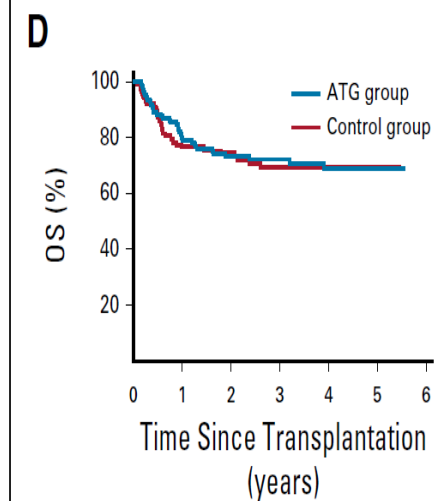
No. at risk:

ATG	132	74	59	34	24	8
Control	131	56	34	15	7	2



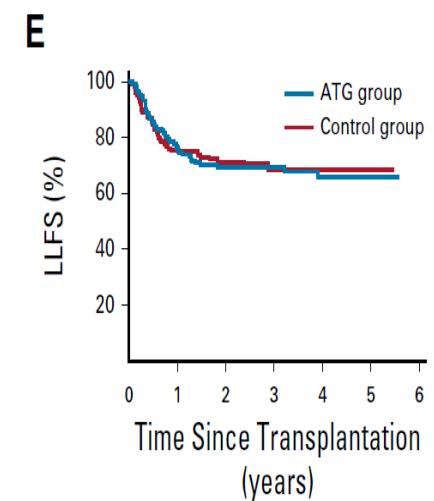
No. at risk:

ATG	132	99	79	48	35	9
Control	131	98	85	49	32	9



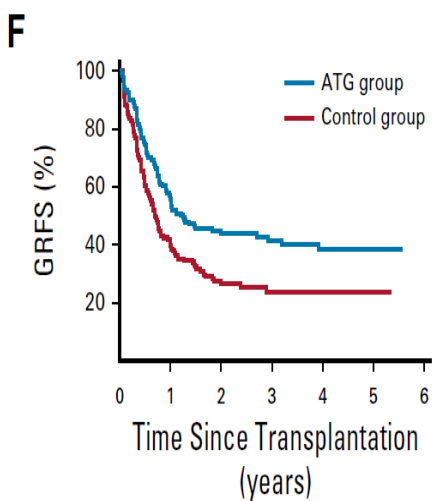
No. at risk:

ATG	132	103	83	49	35	9
Control	131	100	88	50	32	0



No. at risk:

ATG	132	99	79	48	35	9
Control	131	98	86	49	32	9



No. at risk:

ATG	132	72	54	33	23	7
Control	131	54	32	15	8	3

ARTICLE OPEN



Post-transplant cyclophosphamide versus anti-thymocyte globulin after reduced intensity peripheral blood allogeneic cell transplantation in recipients of matched sibling or 10/10 HLA matched unrelated donors: final analysis of a randomized, open-label, multicenter, phase 2 trial

Eolia Brissot ¹✉, Myriam Labopin ^{1,2}, Helene Labussière ³, Gaelle Fossard ³, Patrice Chevallier ⁴, Thierry Guillaume ⁴, Ibrahim Yakoub-Agha ⁵, Micha Srour ⁵, Claude-Eric Bulabois ⁶, Anne Huynh ⁷, Sylvain Chantepie ⁸, Anne-Lise Menard ⁹, Marie-Therese Rubio ¹⁰, Patrice Ceballos ¹¹, Rémy Dulery ¹, Sabine Furst ¹², Florent Malard ¹, Didier Blaise ¹² and Mohamad Mohty ¹

BCJ 2024

Phase 2 randomized study

MRD and MUD transplants

RIC regimen for hematological malignancies- FLU-BU

Experimental arm- PTCY + CSA+ MMF-44 patients

Control Arm- ATG (5 mg/kg over 2 days- thymoglobulin) + CSA + MMF-37 patients

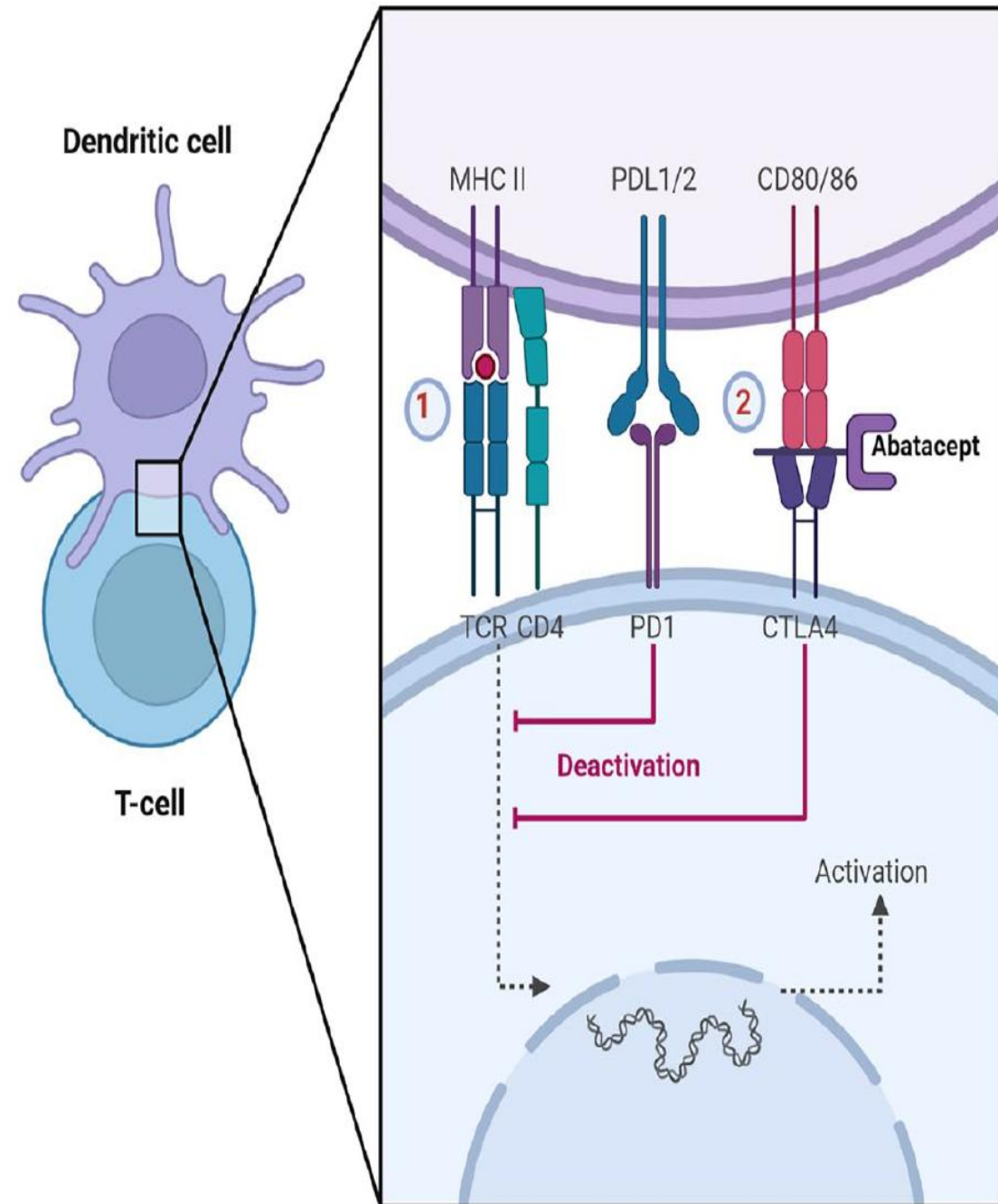
Primary End Point- GRFS at 12 months

Table 2. Outcomes at 1, 3, and 5 years.

		Relapse	NRM	DFS	OS	GRFS	CRFS	chronic GVHD	chronic GVHD requiring systemic treatment
1 year	PTCy (<i>n</i> = 44)	22.7% [11.6–36]	11.4% [4.1–22.8]	65.9% [50–77.8]	79.5% [64.4–88.8]	56.8% [41–69.9]	56.8% [41–69.9]	32.5% [18.5–47.2]	13.6% [5.4–25.6]
	ATG (<i>n</i> = 37)	27% [13.9–42]	8.1% [2–19.8]	64.9% [47.3–77.9]	81.1% [64.4–90.5]	40.5% [24.9–55.7]	40.5% [24.9–55.7]	36.1% [20.7–51.8]	24.3% [11.9–39.2]
	<i>P</i> value (censored at 1 year)	0.61	0.66	0.82	0.91	0.12	0.11	0.72	0.58
3 years	PTCy (<i>n</i> = 44)	27.3% [15–41]	11.4% [4.1–22.8]	61.4% [45.4–73.9]	72.7% [57–83.5]	47.7% [32.5–61.5]	47.7% [32.5–61.5]	32.5% [18.5–47.2]	18.2% [8.4–30.9]
	ATG (<i>n</i> = 37)	32.4% [18–47.7]	10.8% [3.3–23.3]	56.8% [39.4–70.8]	64.9% [47.3–77.9]	37.8% [22.6–53]	37.8% [22.6–53]	41.7% [25.2–57.4]	27% [13.8–42.1]
5 years	PTCy (<i>n</i> = 44)	27.3% [15–41]	18.6% [7.3–33.8]	54.2% [36.8–68.6]	60.3% [42.5–74.2]	43.2% [28.4–57.1]	43.2% [28.4–57.1]	32.5% [18.5–47.2]	22.7% [11.6–36.1]
	ATG (<i>n</i> = 37)	37.6% [20.8–54.4]	10.8% [3.3–23.3]	51.6% [33.2–67.2]	60.5% [42.1–74.7]	37.8% [22.6–53]	37.8% [22.6–53]	41.7% [25.2–57.4]	27% [13.8–42.1]
	<i>p</i> value (entire FU)	0.52	0.57	0.77	0.94	0.39	0.37	0.44	0.58

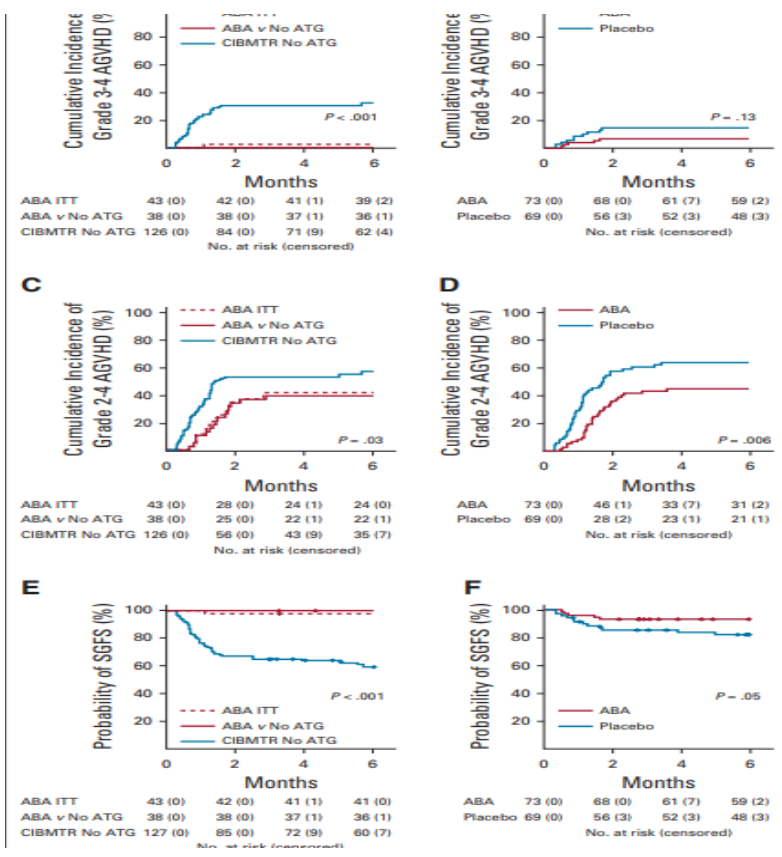
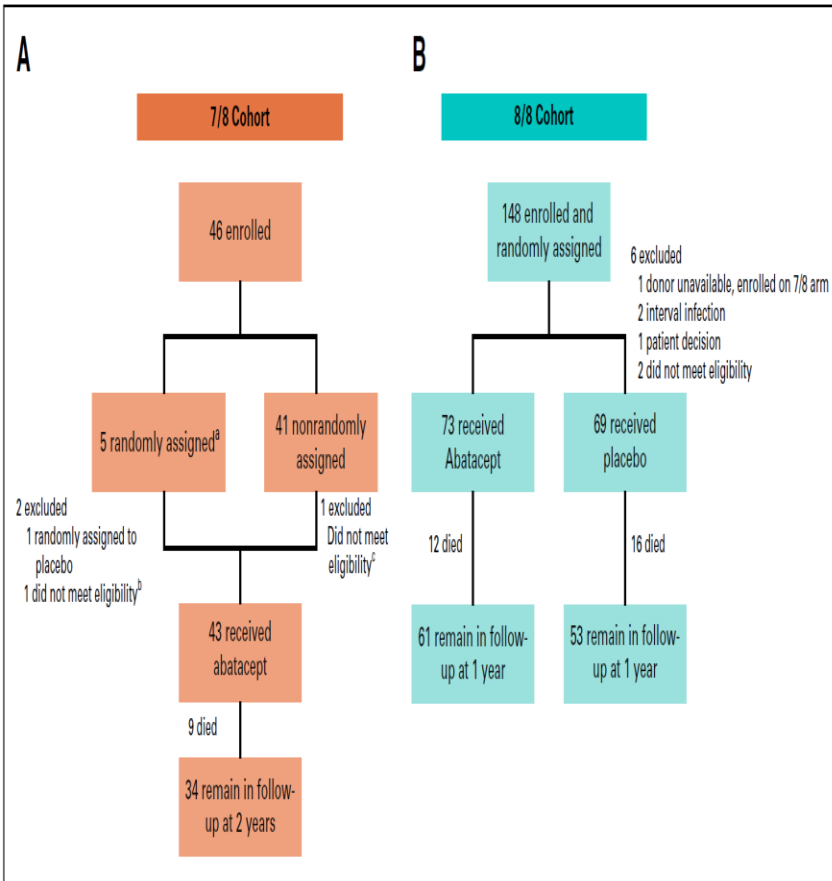
NRM non-relapse mortality, *DFS* disease-free survival, *OS* overall survival, *GRFS* graft-versus-host disease-free, relapse-free survival, *CRFS* chronic graft-versus-host disease-free, relapse-free survival, *GVHD* graft-versus-host disease, *PTCy* post-transplant cyclophosphamide, *ATG* anti-thymocyte globulin, *FU* follow-up.

Abatacept



Phase II Trial of Costimulation Blockade With Abatacept for Prevention of Acute GVHD

Benjamin Watkins, MD¹; Muna Qayed, MD¹; Courtney McCracken, PhD²; Brandi Bratrude, BA³; Kayla Betz, BS³; Yvonne Suessmuth, PhD¹; Alison Yu, PhD³; Shauna Sinclair⁴; Scott Furlan, MD⁵; Steven Bosinger, PhD⁶; Victor Tkachev, PhD³; James Rhodes, PharmD⁷; Audrey Grizzle Tumlin, BS⁷; Alexandria Narayan, BA⁵; Kayla Cribbin, BS⁴; Scott Gillespie, MS²; Ted A. Gooley, PhD⁵; Marcelo C. Pasquini, MD⁸; Kyle Hebert, MS⁸; Urvi Kapoor, MD⁹; Andre Rogatko, PhD¹⁰; Mourad Tighiouart, PhD¹⁰; Sungjin Kim, MS¹⁰; Catherine Bresee, MS¹⁰; Sung W. Choi, MD¹¹; Jeffrey Davis, MD¹²; Christine Duncan, MD³; Roger Giller, MD¹³; Michael Grimley, MD¹⁴; Andrew C. Harris, MD¹⁵; David Jacobsohn, MD¹⁶; Nahal Lalefar, MD¹⁷; Maxim Norkin, MD¹⁸; Nosha Farhadfar, MD¹⁹; Michael A. Pulsipher, MD²⁰; Shalini Shenoy, MD²¹; Aleksandra Petrovic, MD⁴; Kirk R. Schultz, MD¹²; Gregory A. Yanik, MD¹¹; Edmund K. Waller, MD²²; John E. Levine, MD⁹; James L. Ferrara, MD⁹; Bruce R. Blazar, MD²³; Amelia Langston, MD²²; John T. Horan, MD³; and Leslie S. Kean, MD, PhD³



For patients receiving abatacept, four doses were delivered, 10 mg/kg/dose, on days -1, +5, +14, and +28.

With abatacept, rates of AGVHD significantly decreased in HLA-matched and HLA-mismatched URD HCT, with particularly striking results in **HLA-mismatched HCT**.

This decrease in AGVHD was not accompanied by an increase in the risk of relapse or infectious complications

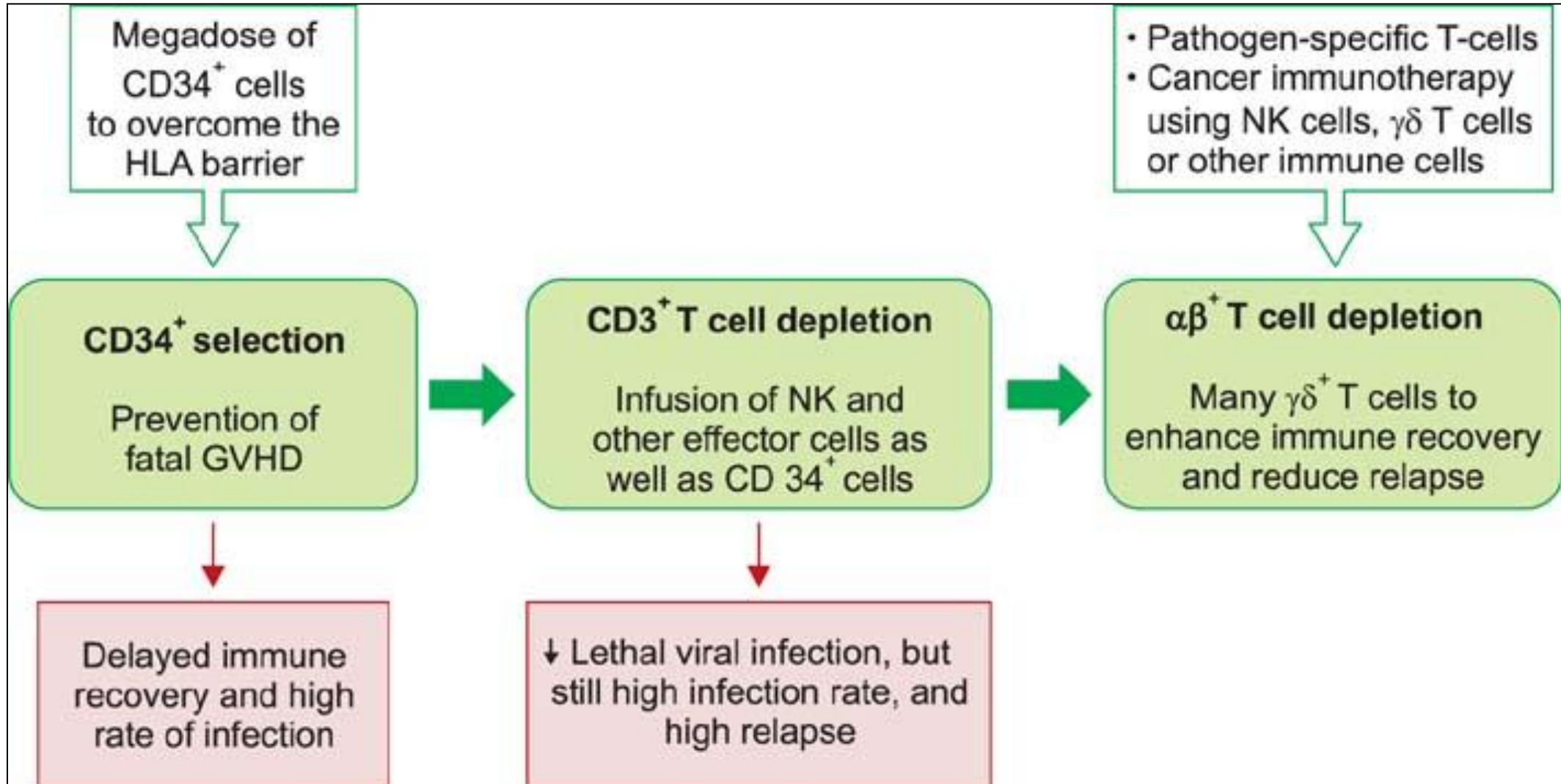
Watkins B, Qayed M, McCracken C, et al. Phase II Trial of Costimulation Blockade With Abatacept for Prevention of Acute GVHD. *J Clin Oncol*. 2021;39(17):1865-1877.

Abatacept and Post-Transplant Cyclophosphamide Along with Short Course Cyclosporine for Matched Family Donor HCT in Acute Leukemia: A Pilot Study.

Transplantation and Cellular Therapy. -Volume 31, Issue 2, Supplement S18-S19 February 2025

Sarita Rani Jaiswal, · Mahak Agarwal, · Kamal Kishor⁴ · Manish Tandon⁵ Suparno Chakrabarti,

Evolution of TCD graft strategies



Naive T-Cell Depletion to Prevent Chronic Graft-Versus-Host Disease

Marie Bleakley, BMBS, PhD, MMSc^{1,2}; Alison Sehgal, MD^{3,4}; Stuart Seropian, MD⁵; Melinda A. Biernacki, MD^{1,6}; Elizabeth F. Krakow, MD, MSc^{1,6}; Ann Dahlberg, MD^{1,2}; Heather Persinger, MA¹; Barbara Hilzinger, BSc¹; Paul J. Martin, MD^{1,6}; Paul A. Carpenter, MBBS^{1,2}; Mary E. Flowers, MD^{1,6}; Jenna Voutsinas, MPH¹; Theodore A. Gooley, PhD^{1,7}; Keith Loeb, MD, PhD^{1,8}; Brent L. Wood, MD, PhD^{1,9}; Shelly Heimfeld, PhD¹; Stanley R. Riddell, MD^{1,6}; and Warren D. Shlomchik, MD^{3,4,10,11}

JCO 2022

Results of 3 phase 2 studies with Naïve T cell depletion

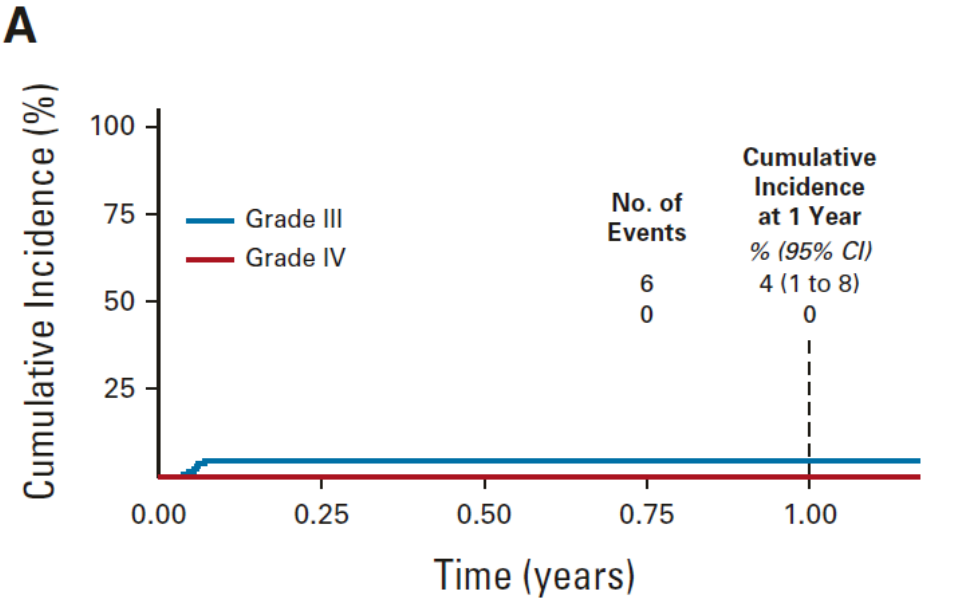
MRD and MUD transplants-

MAC and RIC regimen involving TBI, Fludarabine and Thiotepa for hematological malignancies-

CD 34 selected graft with CD45 RA depleted memory cells infusion

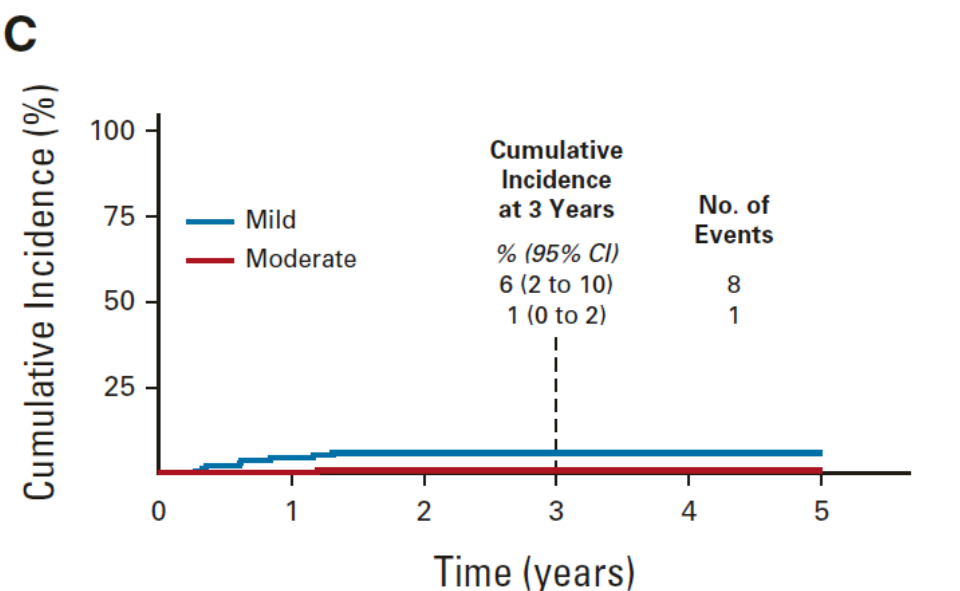
GVHD prophylaxis –Tac +/- MTX or MMF

Primary End Point- incidence of cGHVD and CRFS at 3 years



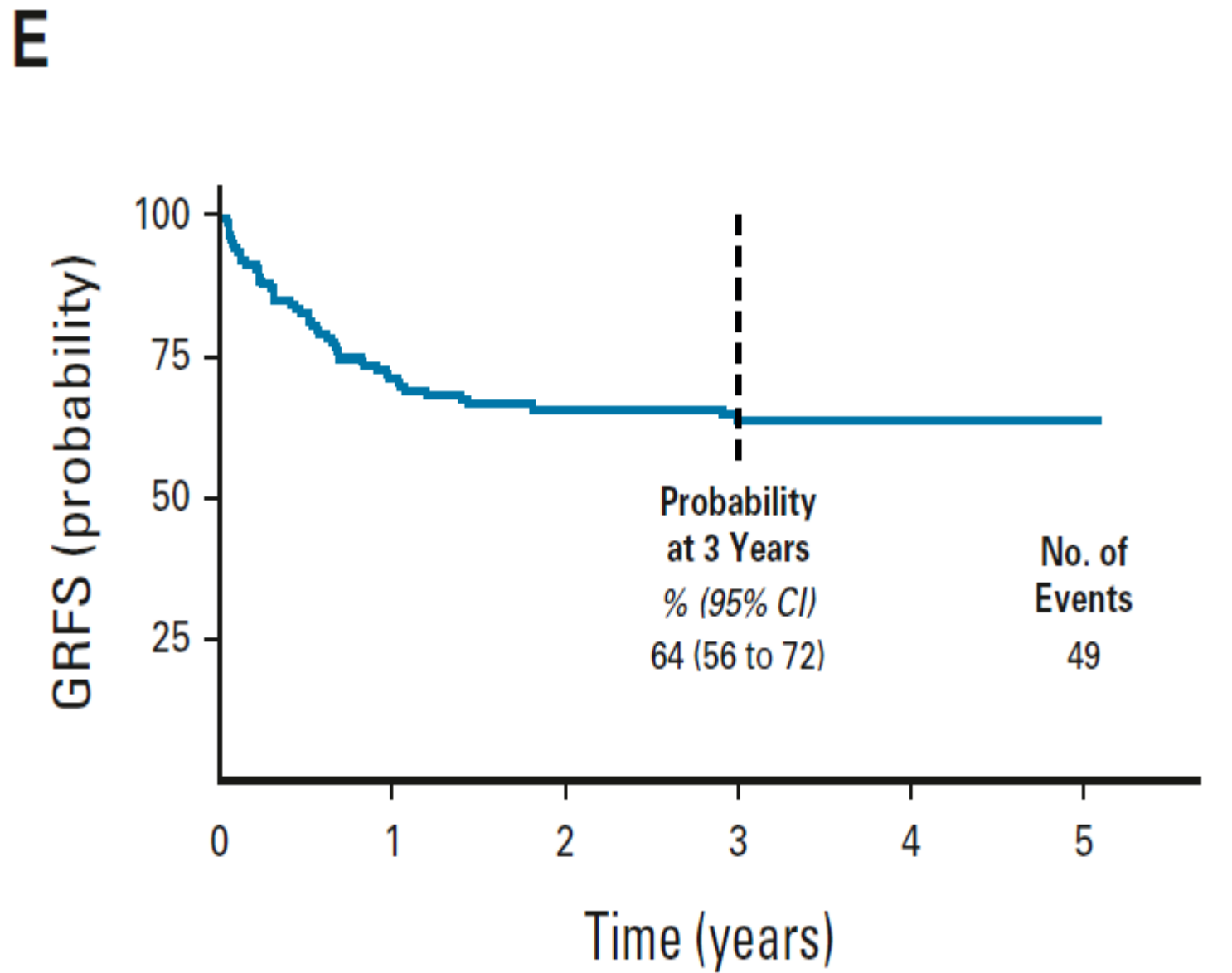
No. at risk:

Time (years)	0	0.25	0.50	0.75	1.00
No. at risk	138	126	123	115	108



No. at risk:

Time (years)	0	1	2	3	4	5
No. at risk	138	98	77	66	48	33



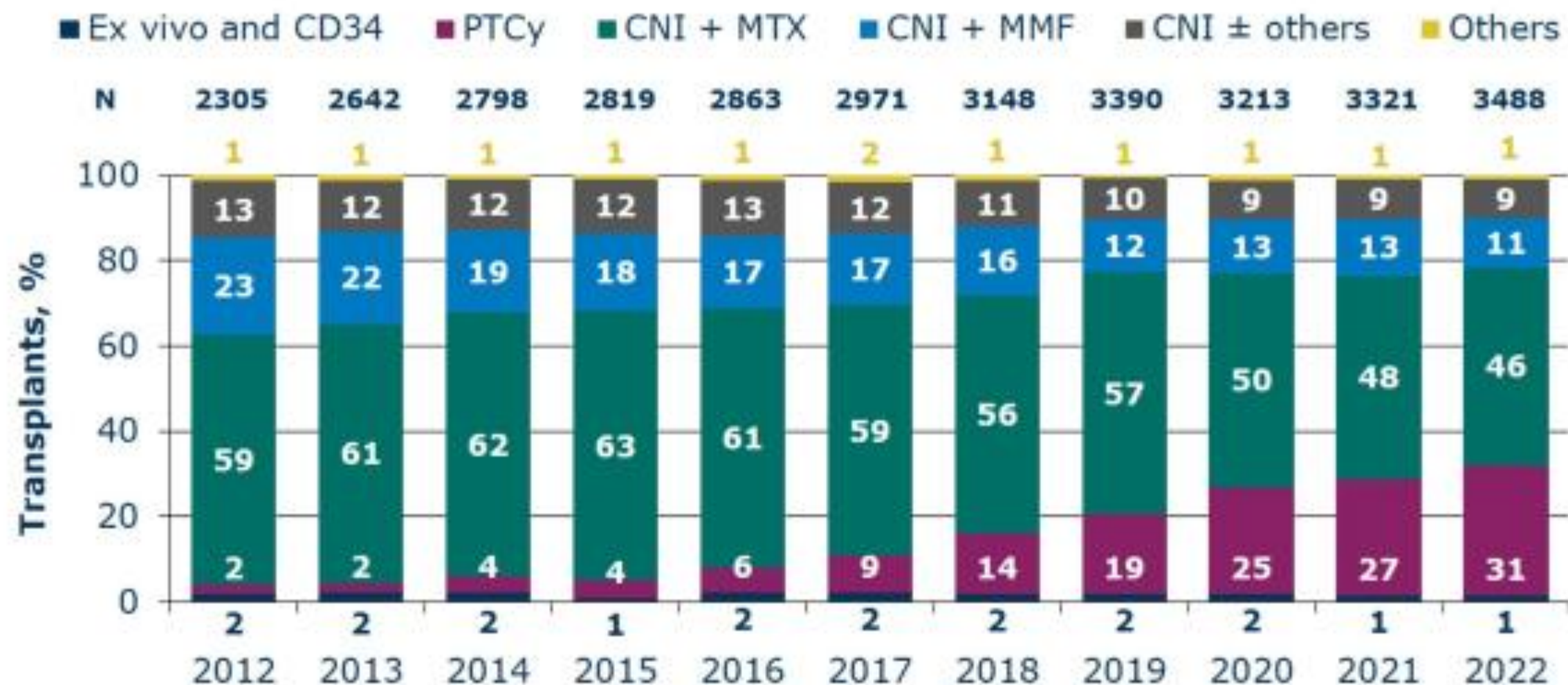
No. at risk:

Time (years)	0	1	2	3	4	5
No. at risk	138	96	77	64	47	31

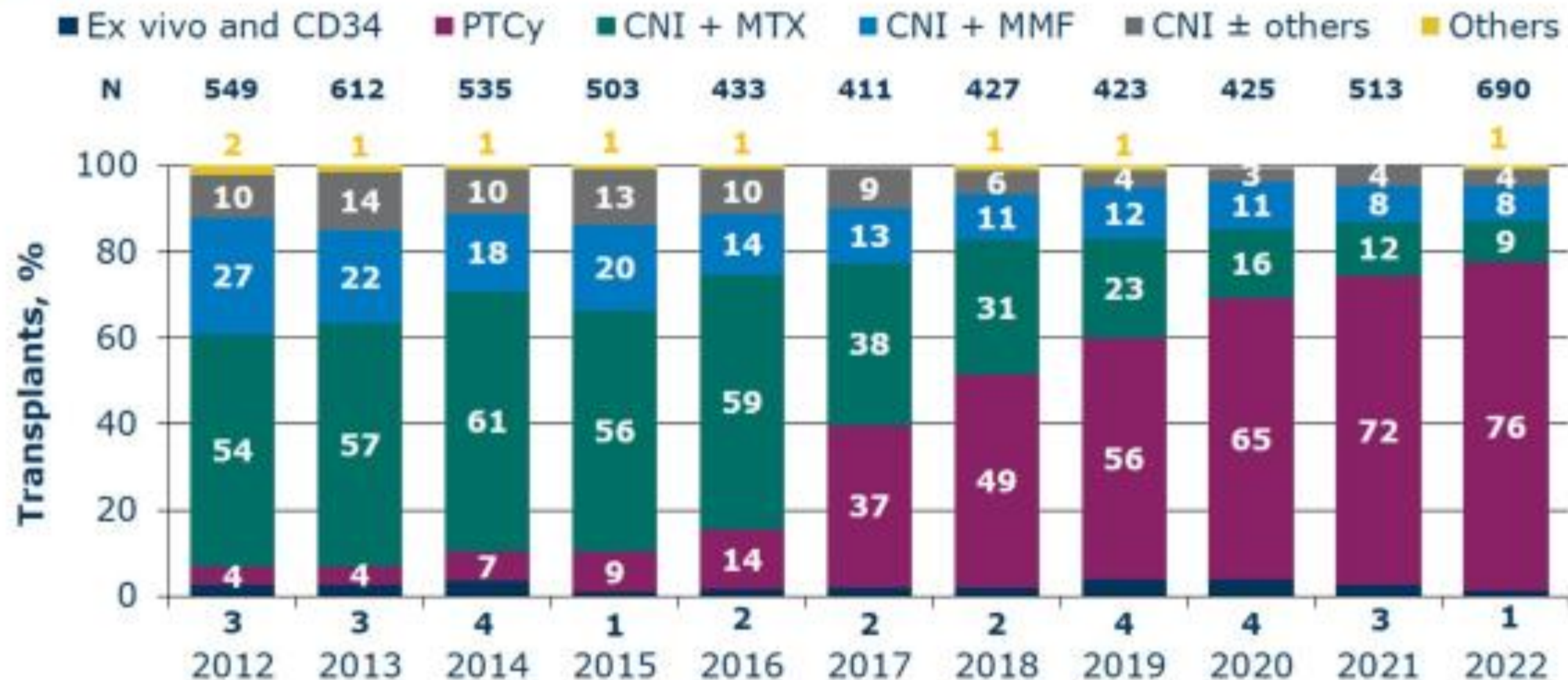
GVHD Prophylaxis of Matched Related Donor HCTs in the US, Adults



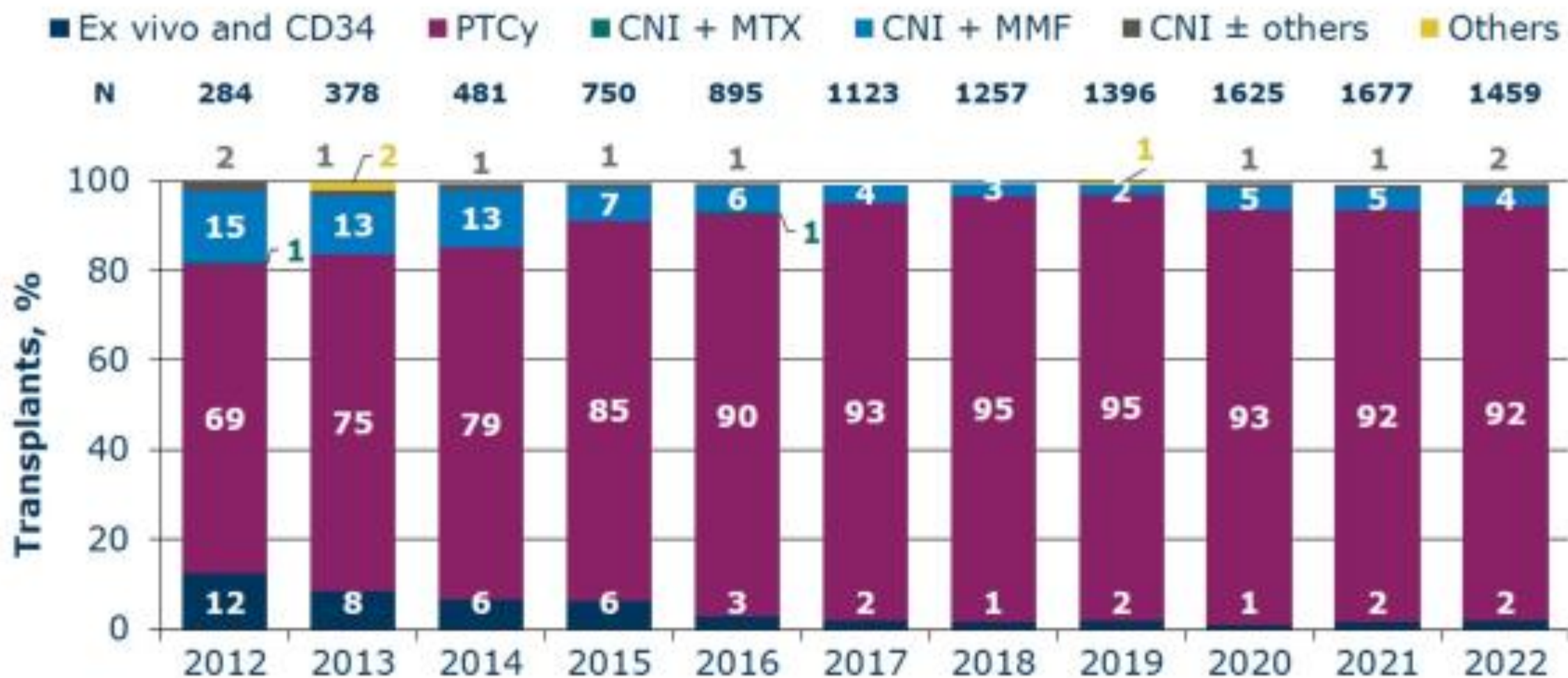
GVHD Prophylaxis of Matched Unrelated Donor HCTs in the US, Adults



GVHD Prophylaxis of Mismatched Unrelated Donor HCTs in the US, Adults



GVHD Prophylaxis of Haplo Donor HCTs in the US, Adults



Summary

- CNI plus methotrexate has been the standard GVHD prophylaxis for last 4 decades for MRD and MUD transplants
- With above regimen- incidence of aGVHD has been atleast 40-50%
- In recent years- use of PTCy or ATG in matched related/unrelated donor transplant has shown to improve GVHD Free Relapse Free Survival
- However, between the 2 strategies, there is not enough data to suggest superiority.
- Addition of abatacept to standard GVHD prophylaxis has also shown decrease incidence of GVHD
- T depleted strategies that depleting naïve T cells (CD 45RA) being co infused with CD 34 + cells or alpha/beta T cell depletion is another strategy that may decrease incidence of aGVHD
- In haplo transplant , both PTCy and T depleted strategies have shown favourable outcomes to decrease incidence of aGVHD