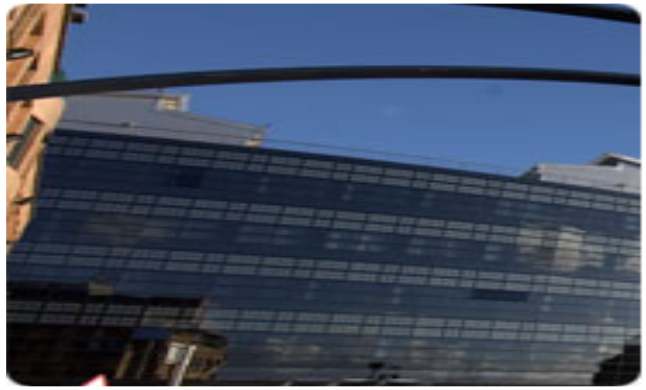
The FCC platform and T-cell replete haplo-ID HSCT in Aplastic Anaemia

ANTONIO PAGLIUCA
KING'S COLLEGE HOSPITAL & KING'S COLLEGE LONDON

GITMO MEETING, NAPLES
24-25th January, 2017



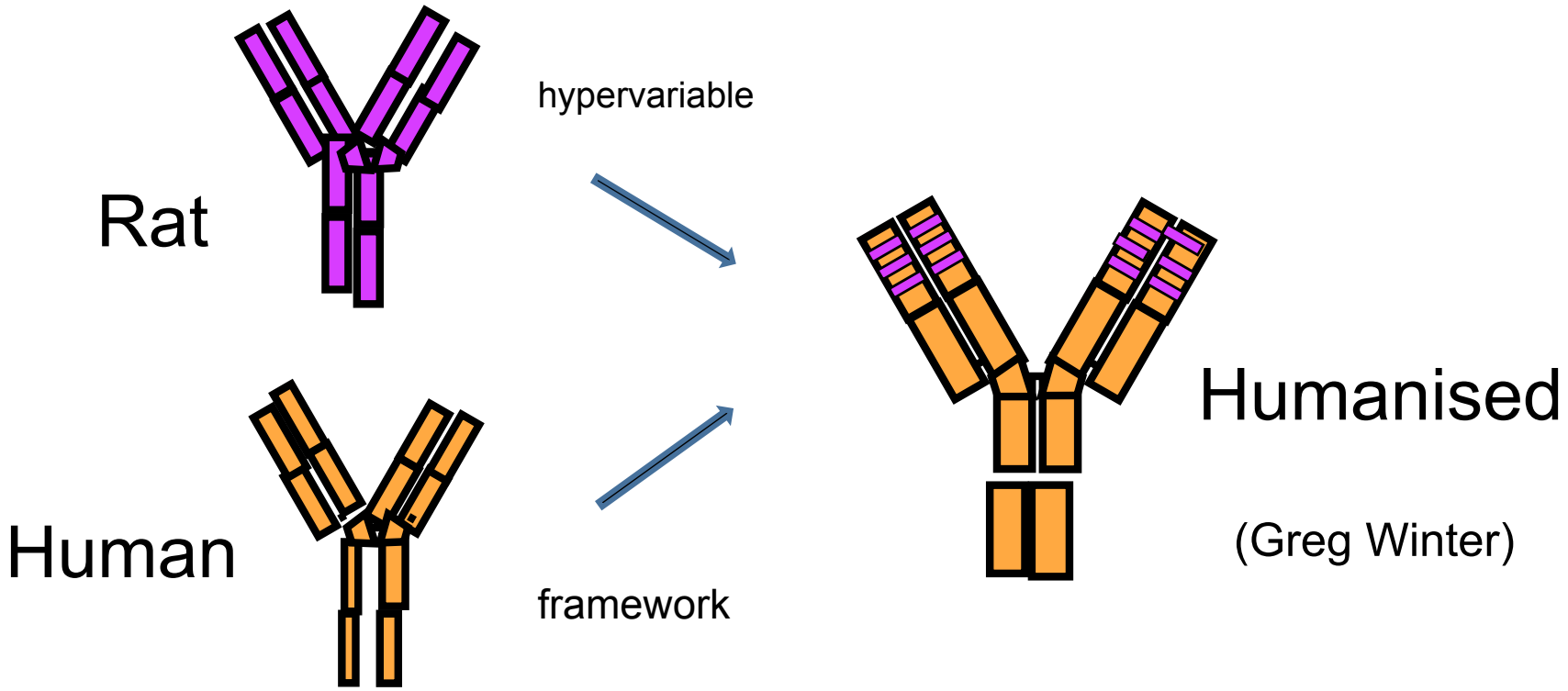
TALK OUTLINE

- Introduction to Campath
- Current guidelines and outcomes
- FCC platform and data
- Immunological reconstitution of FCC and tolerance
- Haplo data and case study
- Conclusion

Development of Campath

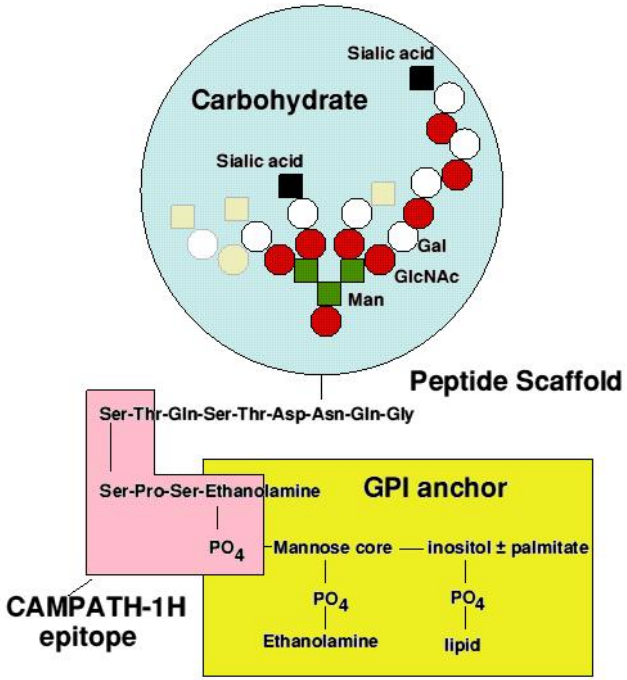
1980 CAMPATH-1M (rat)
1982 First bone marrow transplant
1986 CAMPATH-1G (rat IgG2b)
1987 CAMPATH-1H (IgG1)
Leukemia, transplantation, autoimmune disease

Herman Waldmann and Geoff Hale
Cambridge **Path**ology Laboratories



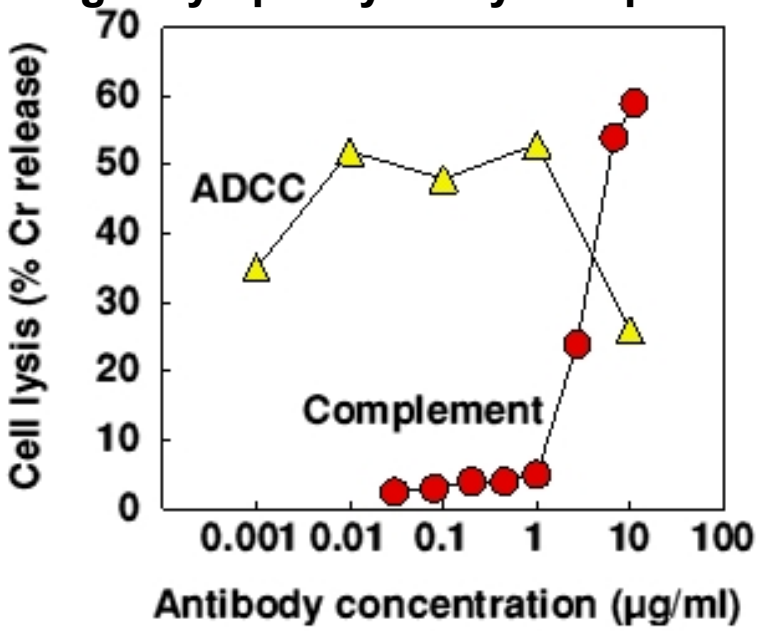
CAMPATH-1 monoclonal antibodies

- Recognise CD52, GPI-linked protein
- CD52 expressed on T, B, dendritic cells, monocytes, eosinophils, (neutrophils), but not on CD34+ HPC
- Campath-1M and 1G lytic with complement
- Campath-1G and 1H also act by ADCC



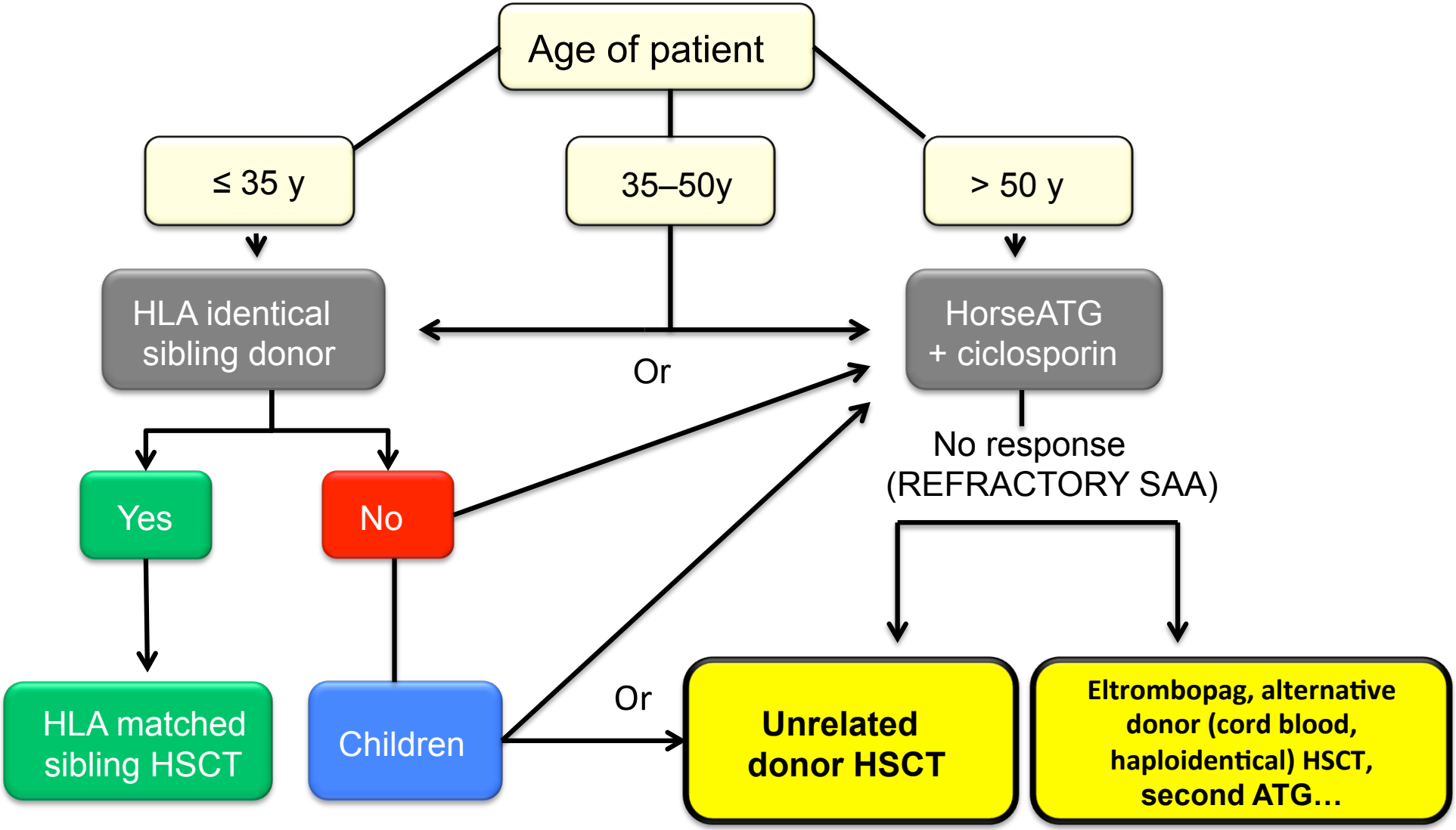
Structure of CD52 antigen

Killing of lymphocytes by Campath-1H



Riechmann. Nature 1988;322:323

Treatment of acquired severe aplastic anaemia



BCSH guidelines: Killick et al. Br J Haematol. 2016 Jan;172:187-207; EBMT guidelines: Sureda et al. BMT 2015;50:1037

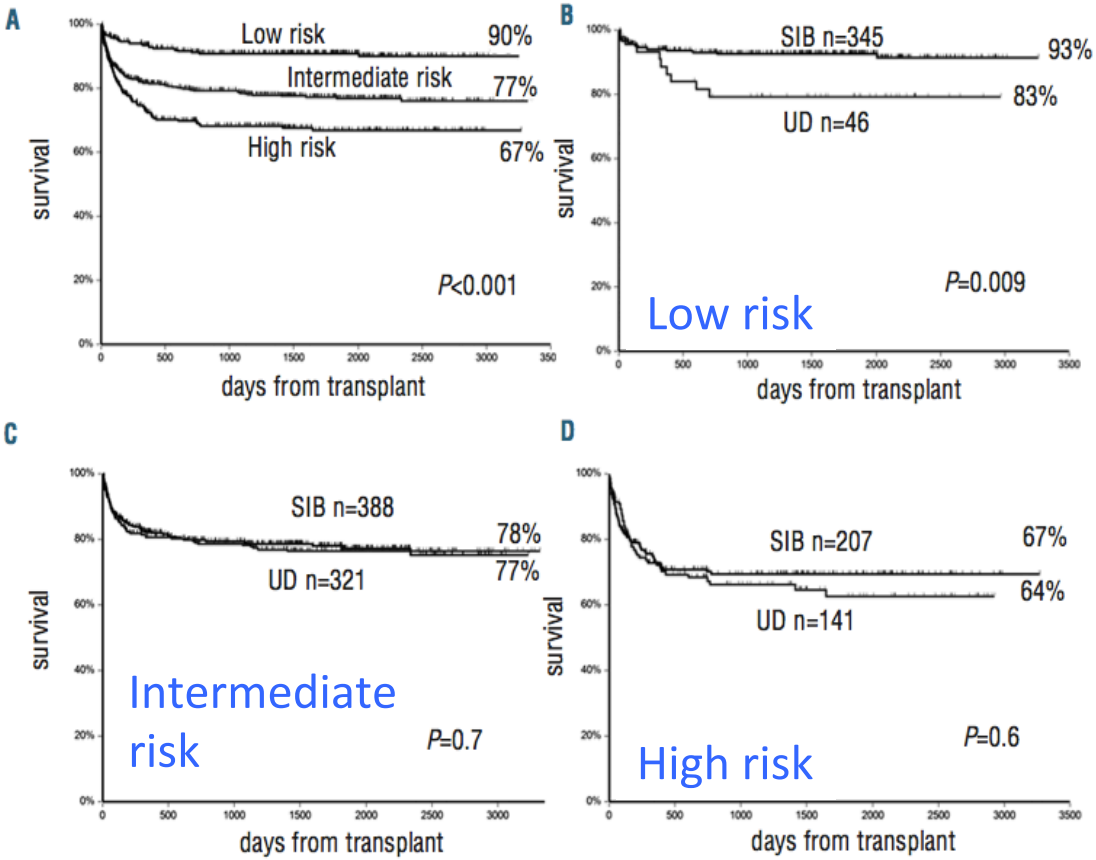
What are the goals of stem cell transplantation in severe aplastic anaemia?

Minimal toxicity from conditioning regimen
Sustained haematological engraftment: • *Full donor myeloid chimerism* • *Stable mixed T-cell chimerism ?*
Absence of any chronic GVHD
Minimal long term complications

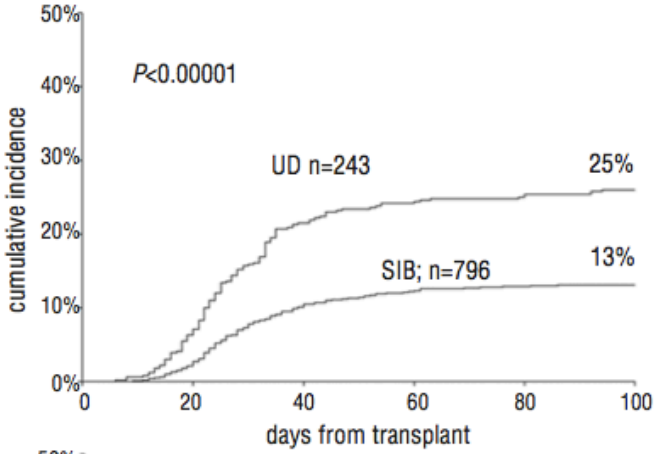
Matched sibling versus unrelated donor HSCT for SAA

EBMT, n = 1448, 2005-2009

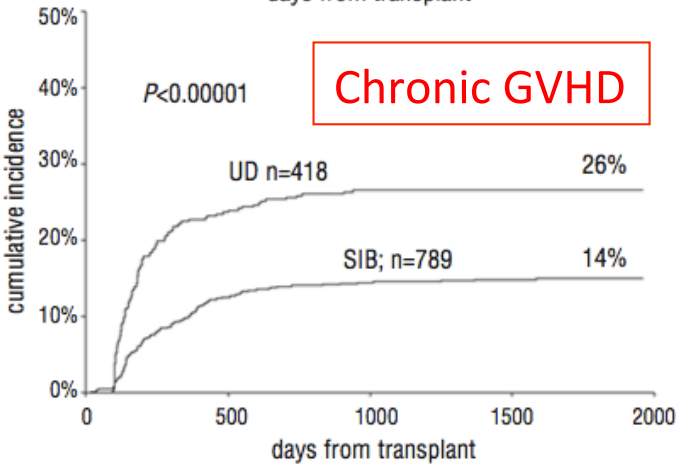
Overall survival



Acute II-IV GVHD



Chronic GVHD



Risk score

Based on: Age > 20yr, time Dx-Tx > 6mo, CMV -/-, ATG in conditioning, BM stem cells

Bacigalupo et al. Haematologica 2015;100:696

blood

2011 118: 2351-2357
Prepublished online April 25, 2011;
doi:10.1182/blood-2010-12-327536

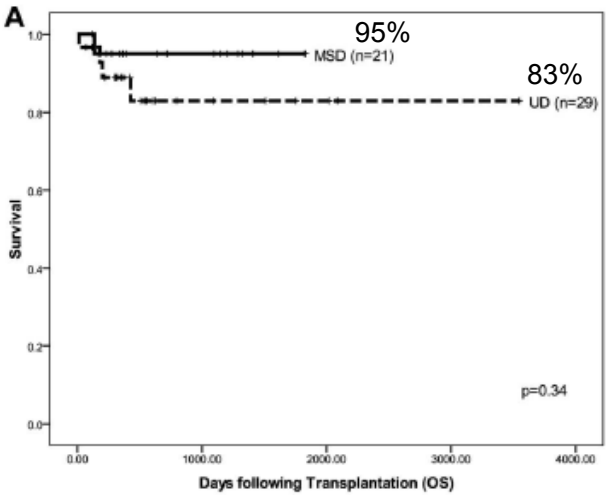
Alemtuzumab with fludarabine and cyclophosphamide reduces chronic graft-versus-host disease after allogeneic stem cell transplantation for acquired aplastic anemia

Judith C. Marsh, Vikas Gupta, ZiYi Lim, Aloysius Y. Ho, Robin M. Ireland, Janet Hayden, Victoria Potter, Mickey B. Koh, M. Serajul Islam, Nigel Russell, David I. Marks, Ghulam J. Mufti and Antonio Pagliuca

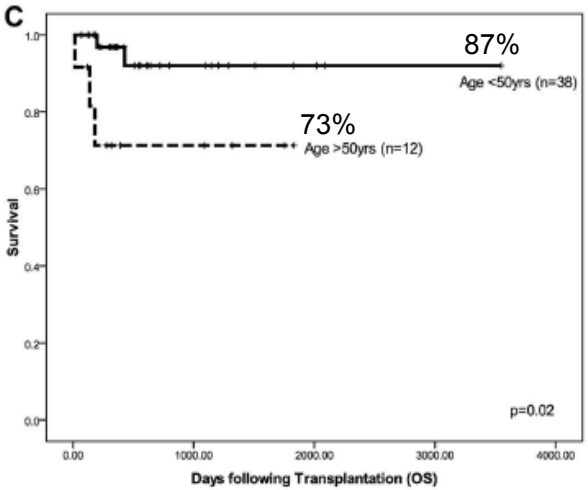
King's FCC conditioning

Fludarabine 30mg/m^2 x 4
CY 30mg/kg x 4
Alemtuzumab 0.2mg/kg x 5
Ciclosporin alone; no methotrexate
Irradiation-free regimen

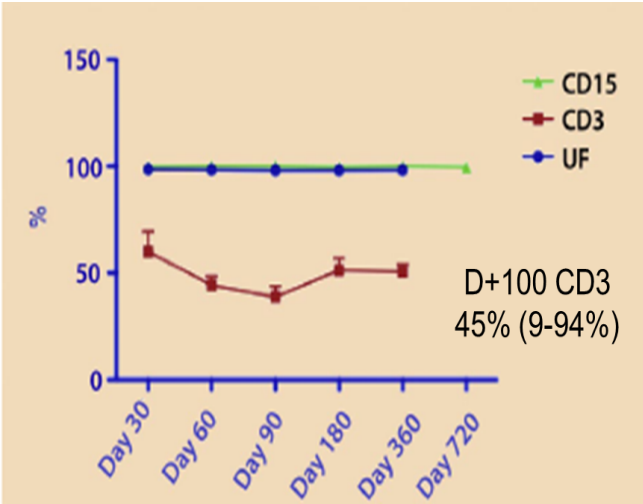
MSD vs MUD



Age < 50 > yr



Co-morbidity score



Acute GVHD
13.5% (16.5% CI at 1yr)

Chronic GVHD
4% (7% CI at 1yr)

Graft failure
6 (12%), 3 primary, 3 secondary
9.5% MSD, 14.5% UD

TRANSPLANTATION

Comment on Marsh et al, page 2351

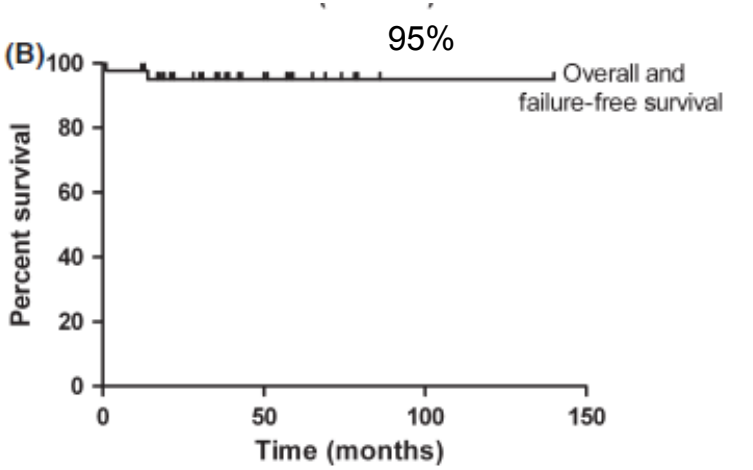
GVHD-free with Campath?

H. Joachim Deeg FRED HUTCHINSON CANCER RESEARCH CENTER

Excellent outcome of matched unrelated donor transplantation in paediatric aplastic anaemia following failure with immunosuppressive therapy: a United Kingdom multicentre retrospective experience

FCC regimen
Flud 30mg/m2 x5
CY: 200mg/kg (n=11)
120mg/kg (n=33)
Alemtuzumab: 0.2mg/kg x5 (n=30)
0.3mg/kg x3 (n=14)

Acute GVHD I-II	31.8%	**5y CI**
III-IV	2.3% (n=1)	38.5%
Chronic GVHD	6.8% (n=3) Ltd (2), ext (1)	11.5%



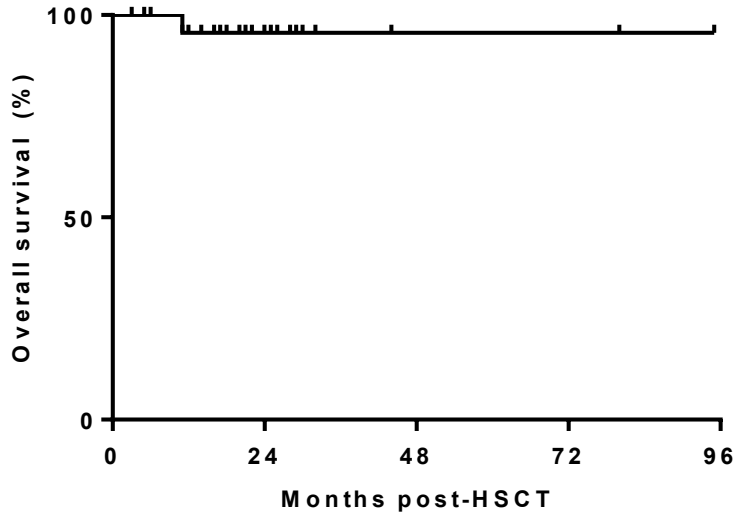
Upfront matched and mismatched unrelated donor transplantation in paediatric idiopathic severe aplastic anaemia:

A United Kingdom multicentre retrospective experience

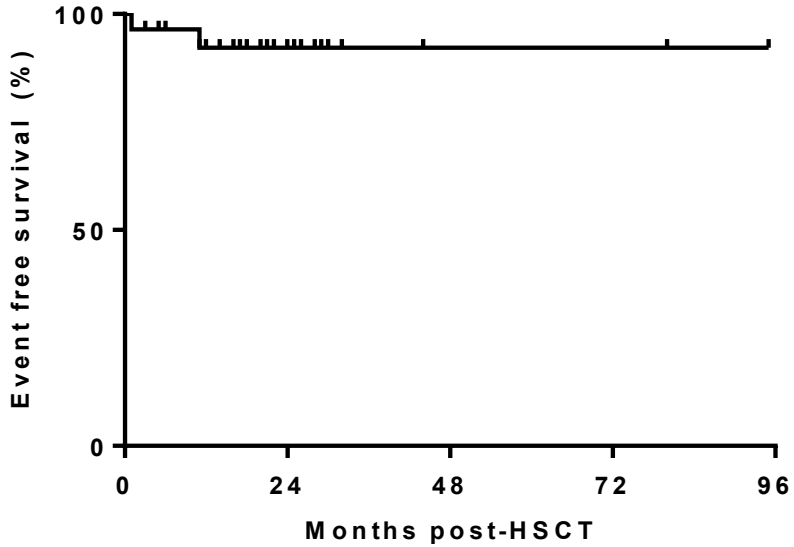
Neha Bhatnagar, Rob Wynn, Mark Velangi, Ajay Vora, Denise Bonney, Brenda Gibson, Rod Skinner, Anna-Maria Ewins, Persis Amrolia, Rachael Hough, Josu De La Fuente, Abigail Shaw, Colin Steward, Paul Veys, Sujith Samarasinghe

Excellent OS and EFS following upfront MUD/MMUD with FCC conditioning regimen

2.5-year OS 95.7%



2.5-year EFS 92.2%



1 death due to idiopathic pneumonia syndrome at 11 months post-HSCT
& 1 primary graft failure following a single antigen-A MMUD HSCT
Acute GVHD III/IV: 7%; chronic 21%, all limited, skin only

National, retrospective, multi-center comparison of Alemtuzumab- versus ATG-based conditioning regimens in HSCT for severe aplastic anemia (SAA): a study from the British Society for Blood and Marrow Transplantation (BSBMT) (CTCR 09-03)

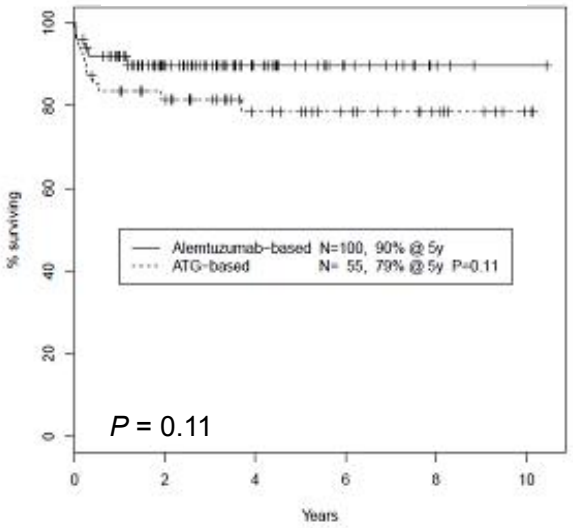
Marsh JC, Pearce RM, Ko M, Tang D, Lim Z, Pagliuca A, Mufti GJ, Snowden J, Vora A, Gibson B, Gilleece M, Lee J, Kirkland K, Cook G, on behalf of the BSBMT CTC Committee.

- Transplants performed 1999-2009
- 22 centres in the UK
- First allograft for acquired SAA
- N = 155
- Median follow up: 38 mo (3-125)
- Median age: 20yr (1.5-67.5)

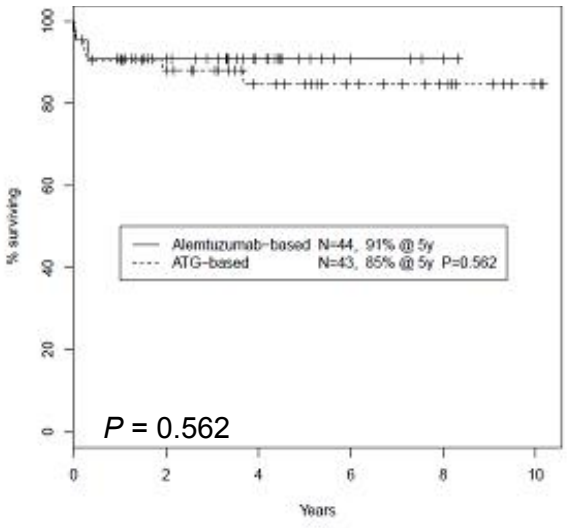
BMT 2014; 49:42

National, retrospective, multi-center comparison of alemtuzumab- versus ATG-based conditioning regimens in HSCT for severe aplastic anaemia

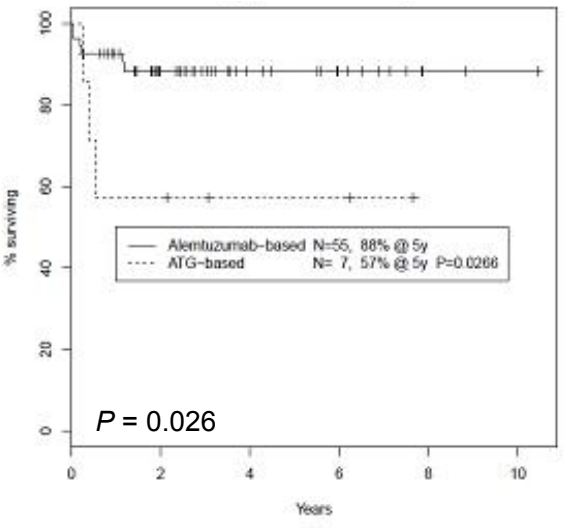
All patients



Matched siblings



Unrelated donors



Marsh et al. BMT 2014;49:42

Conclusions

- Confirms excellent outcomes after HSCT for SAA
- 5 YR OS of 88% for UD HSCT using alemtuzumab, without using irradiation
- No difference in OS for MUD and MSD HSCT in children
- Lower risk of chronic GVHD, and less grade III/IV acute GVHD with alemtuzumab
- Better OS, EFS, less GVHD using BM compared to PB

bjh research paper

Similar outcome of upfront-unrelated and matched sibling stem cell transplantation in idiopathic paediatric aplastic anaemia. A study on behalf of the UK Paediatric BMT Working Party, Paediatric Diseases Working Party and Severe Aplastic Anaemia Working Party of EBMT

Upfront UD HSCT	N=29
Upfront MSD HSCT	N=87
Upfront IST	N=58
UDHSCT after failed IST	N=24

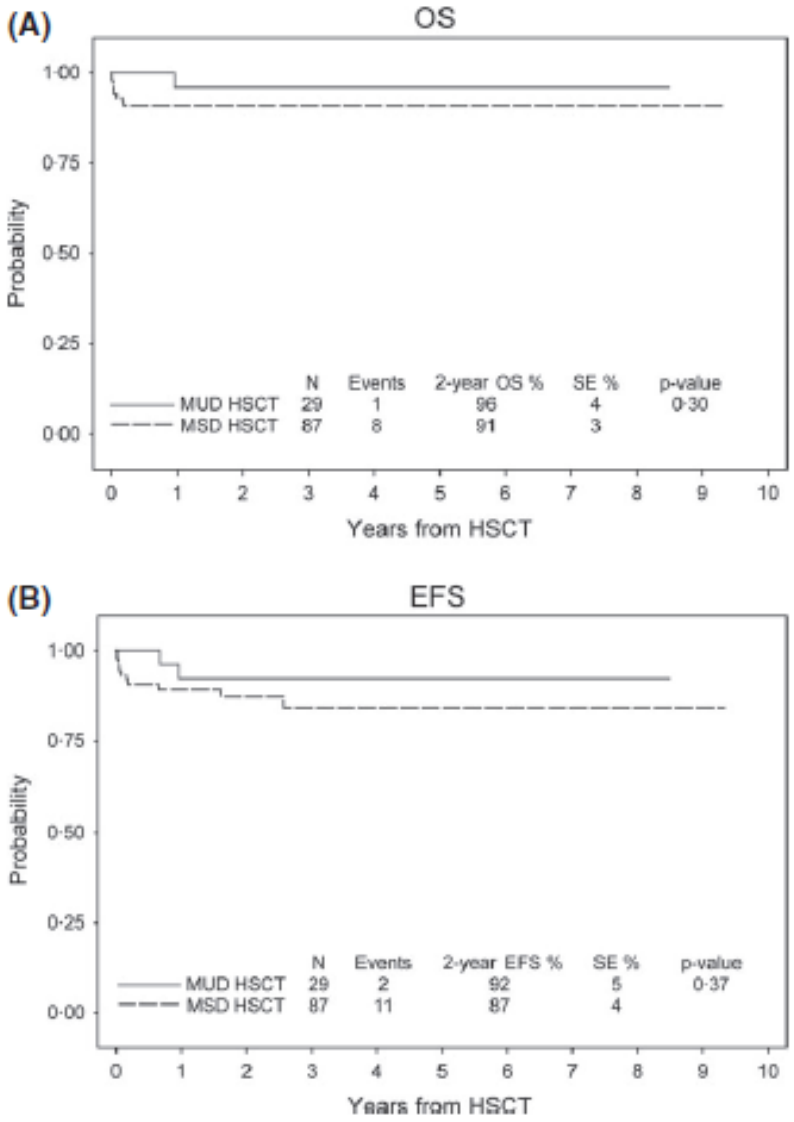
FCC regimen
- Fludarabine 150mg/m2
- Cyclophosphamide 120mg/kg
- Alemtuzumab 0.9-1mg/kg

	Upfront MUD/MMUD HSCT	MSD HSCT Controls	
	n	n	p-value
Total	29	87	
Gender			
Male	12	36	1·0
Female	17	51	
Source of stem cell			
Bone marrow	21	63	1·0
Peripheral blood	8	24	
Mean age at HSCT (years ± SE)	8·9 ± 0·9	8·9 ± 0·5	0·95
Mean interval from diagnosis to HSCT (years ± SE)	0·41 ± 0·05	0·38 ± 0·04	0·69

HSCT, haematopoietic stem cell transplantation; MUD, matched unrelated donor MMUD, mismatched unrelated donor; MSD, matched sibling/family donor.

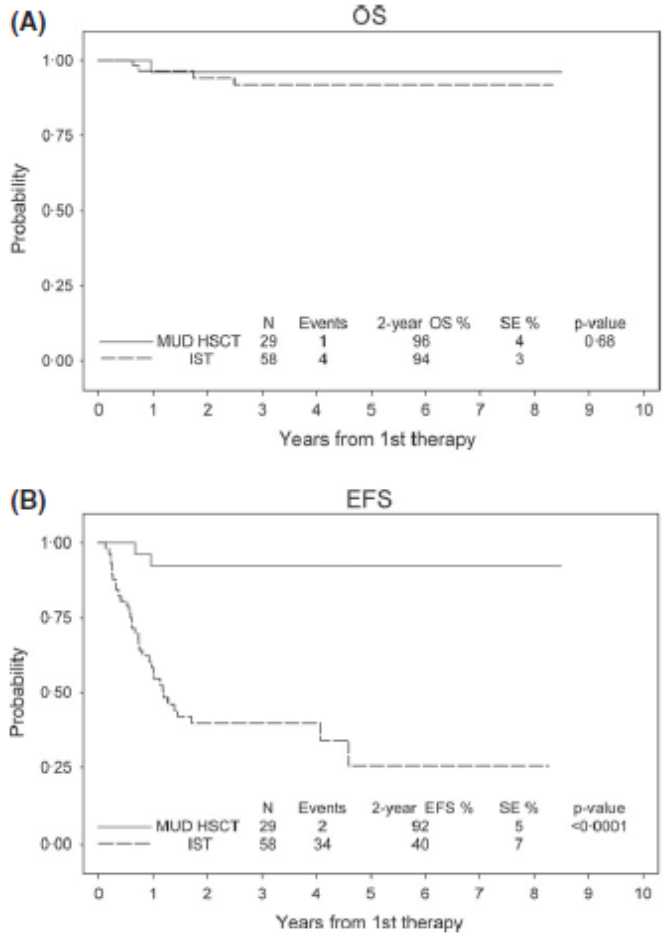
Dufour et al. Brit. J. Haematol. 2015; 171: 585

Upfront UD HSCT vs MSD HSCT

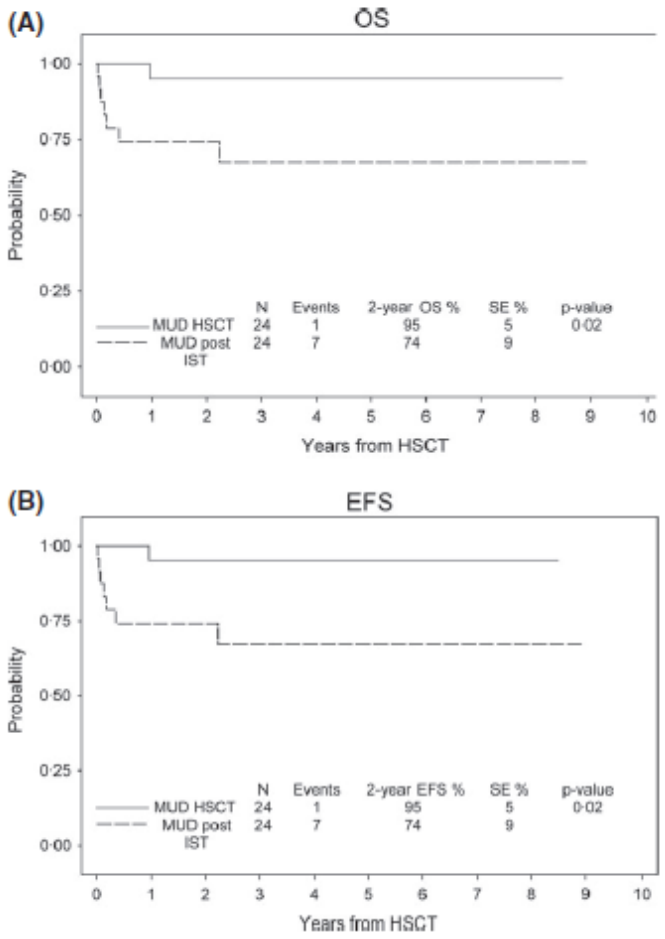


Dufour et al. Brit. J. Haematol. 2015; 171: 585

Upfront UD HSCT vs upfront IST



Upfront vs 2nd line UD HSCT



Dufour et al. Brit. J. Haematol. 2015; 171: 585

Relapse and Clonal Evolution After IST

Studies	Period	N	Age (median)	Resp	Relapse	Clonal Evolution	Survival
Germany	1986-1989	84	32	65%	19%	8%	58% à 11 ans
NIH	1991-1998	122	35	61%	35%	11%	55% à 7 ans
EGMBT	1991-1998	100	16	77%	12%	11%	87% à 5 ans
Japan	1992-1997	119	9	68%	22%	6%	88% à 3 ans
Germany/Australia	1993-1997	114	9	77%	12%	6%	87% à 4 ans
Japan	1996-2000	101	54	74%	42%	8%	88% à 4 ans
NIH	1999-2003	104	30	62%	37%	9%	80% à 4 ans
NIH	2003-2005	77	26	57%	26%	10%	93% à 3 ans

Biology of Blood and Marrow Transplantation

journal homepage: www.bbmt.org

Mixed T Cell Chimerism After Allogeneic Hematopoietic Stem Cell Transplantation for Severe Aplastic Anemia Using an Alemtuzumab-Containing Regimen Is Shaped by Persistence of Recipient CD8 T Cells

Francesco Grimaldi [1,2], Victoria Potter [1], Pilar Perez-Abellan [3], John P. Veluchamy [3], Muhammad Atif [3], Rosemary Grain [3], Monica Sen [3], Steven Best [1], Nicholas Lea [1], Carmel Rice [1], Antonio Pagliuca [1], Ghulam J. Mufti [1,3], Judith C. W. Marsh [1,3,*], Linda D. Barber [3,†]

[1] *Department of Haematology, King's College Hospital NHS Foundation Trust, London, United Kingdom*
[2] *Department of Clinical Medicine, Haematology Division, AOU Federico II, Naples, Italy*
[3] *Division of Cancer Studies, King's College London, London, United Kingdom*

Grimaldi et al, Biol Blood Marrow Transplant. 2016 Nov 2. doi: 10.1016/j.bbmt.2016.11.003. [Epub ahead of print]

Patient pre-transplant characteristics

N	**45**
Median age (range) **Number of patients aged > 50yr**	32 (15 – 63) 14 (31.1%)
M : F	28 : 17
Type of donor - **Matched sibling (MSD)** - **Unrelated donor (UD)** - **9/10 UD***	 12 (26.6%) 33 (73.4%) 8 of 33 (24.2% of UD)
Previous immunosuppressive therapy - **MSD** - **UD**	 6 of 12 (50%) 27 of 33 (81.8%), p = 0.055
Median time to transplant - **MSD - 5.9 months (Range 2.7 – 34.7)** - **UD - 8.4 months (Range 2.1 – 178.9)**	 MSD UD, p = 0.095
Number of patients HLA alloimmunised	11 (24.4%)
Stem cell source - **BM** - **PB**	 7 (15.5%) 38 (84.5%)
Number of patients with PNH clone **Median PNH clone size:** - **Granulocytes** - **Red cells**	21 (46.6%) 2% (range 0.02 – 40) Not available
Median alemtuzumab dose	70 mg (Range 45 – 100)
Median CD34+ stem cell dose	6.55 x 10^6 CD34+/Kg (Range 1.97 – 12.40)
Median follow up	31.4 months (Range 3 – 93)

**FCC + 2Gy TBI for 9/10 UD transplants*

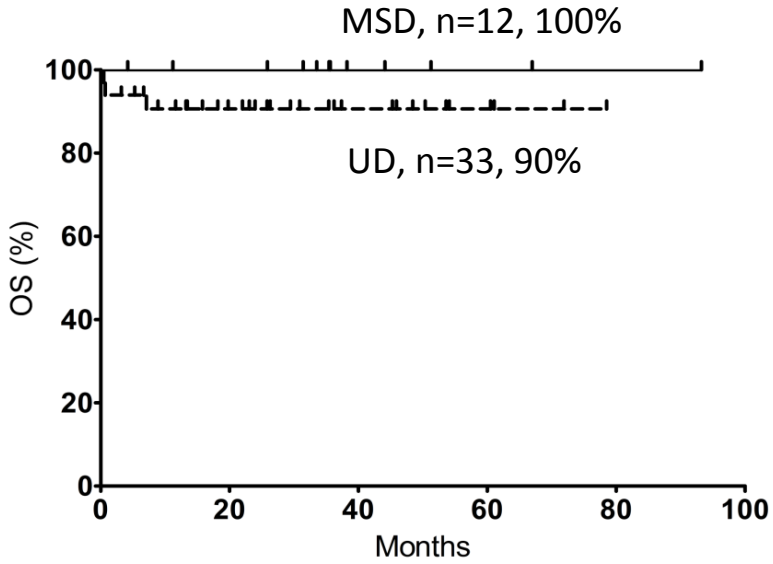
Patient outcomes after FCC HSCT

Median time to neuts > 0.5 x 10^9/l	12 days (10 – 22)
Median time to platelets > 20 x 10^9/l	12 days (9 – 61)
Primary graft failure	1 (2.2%)
Acute GVHD: ▪ Grade I/II ▪ Grade III/IV	6 (13%) ▪ 6 of 6 (100%) ▪ 0 of 6
Chronic GVHD: ▪ Mild ▪ Moderate ▪ Severe	6 (13%) ▪ 4 of 6 (66%) ▪ 1 of 6 (17%) ▪ 1 of 6 (17%)
1-year TRM	3 (6.6%)
5-year OS	93%
5-year EFS	90.7%
5-year EFS MSD (n=12) vs UD (n=33)	100% vs 87% (P=0.219)
5-year EFS Age ≤ 50 years (n=31) vs >50 years (n=14)	93% vs 86% (P=0.356)

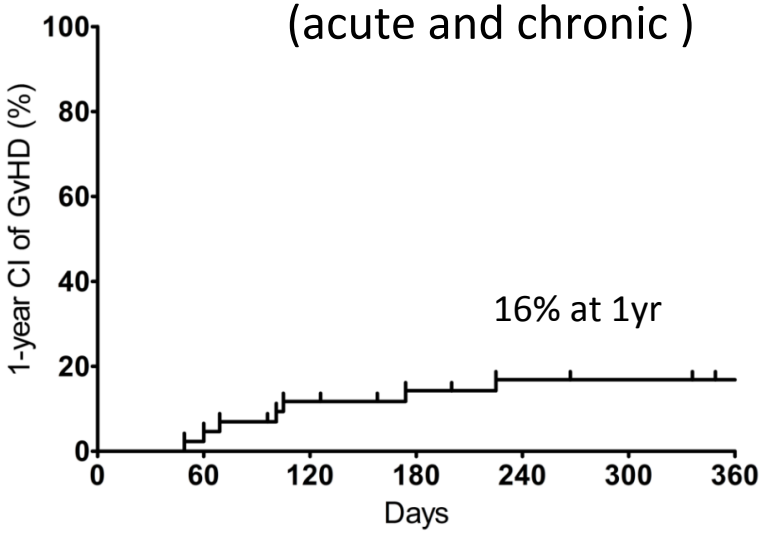
King's experience of FCC HSCT for acquired SAA 2007-2015

Acute GVHD	6 (13.6%) - 5/6 grade I/II, skin only
Chronic GVHD Mild/Moderate/Severe	6 (13.3%) 4/1/1*

* 9/10 UD SCT



Cumulative incidence of GVHD (acute and chronic)



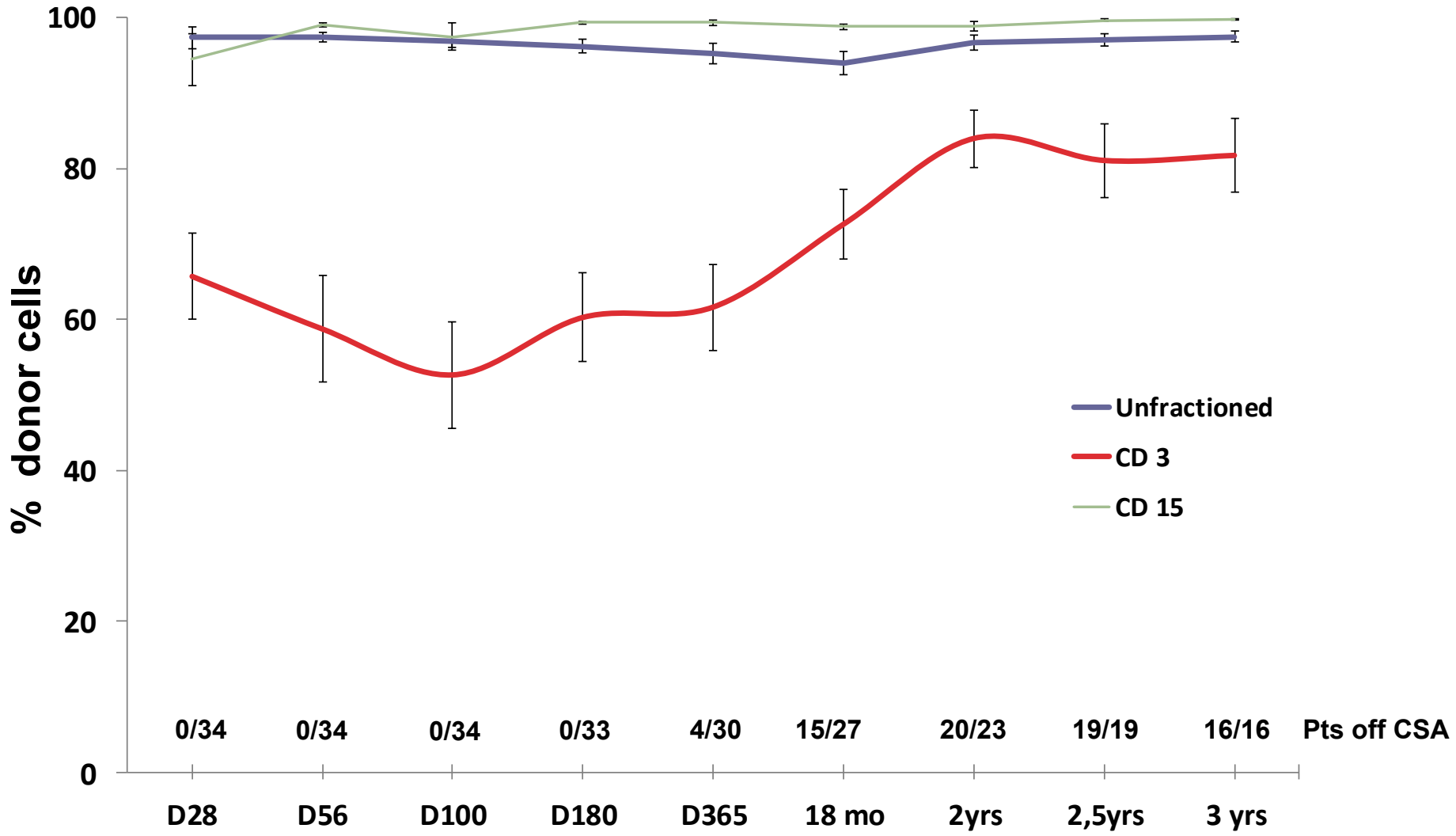
Grimaldi et al, Biol Blood Marrow Transplant. 2016 Nov 2. doi: 10.1016/j.bbmt.2016.11.003. [Epub ahead of print]

Autoimmune disorders post FCC HSCT

N = 9 (20%)

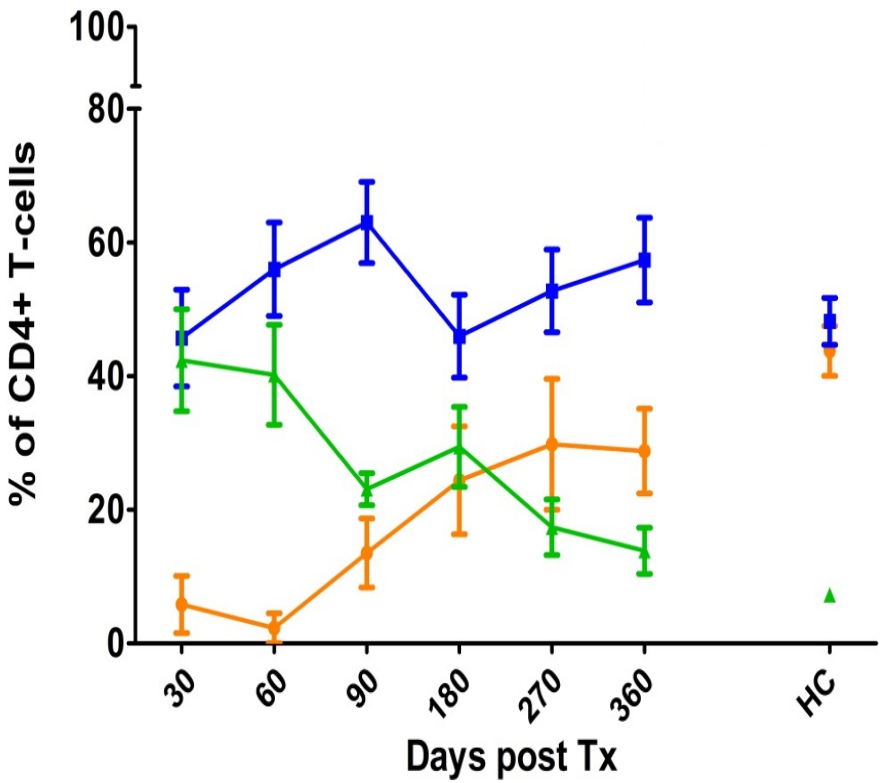
- Autoimmune haemolytic anaemia (4)
  - Warm type
  - All responded to prednisolone; 3 relapsed of whom 2 responded to Rituximab and one refractory fulminant haemolysis died (9/10 UD)
- PRCA (4)
  - 3 major ABO mismatch
  - 3 of 4 recovered, one refractory had successful 2nd MUD (ABO matched)
  - One also had thryoiditis, one ITP
- Probable autoimmune neutropenia (1)

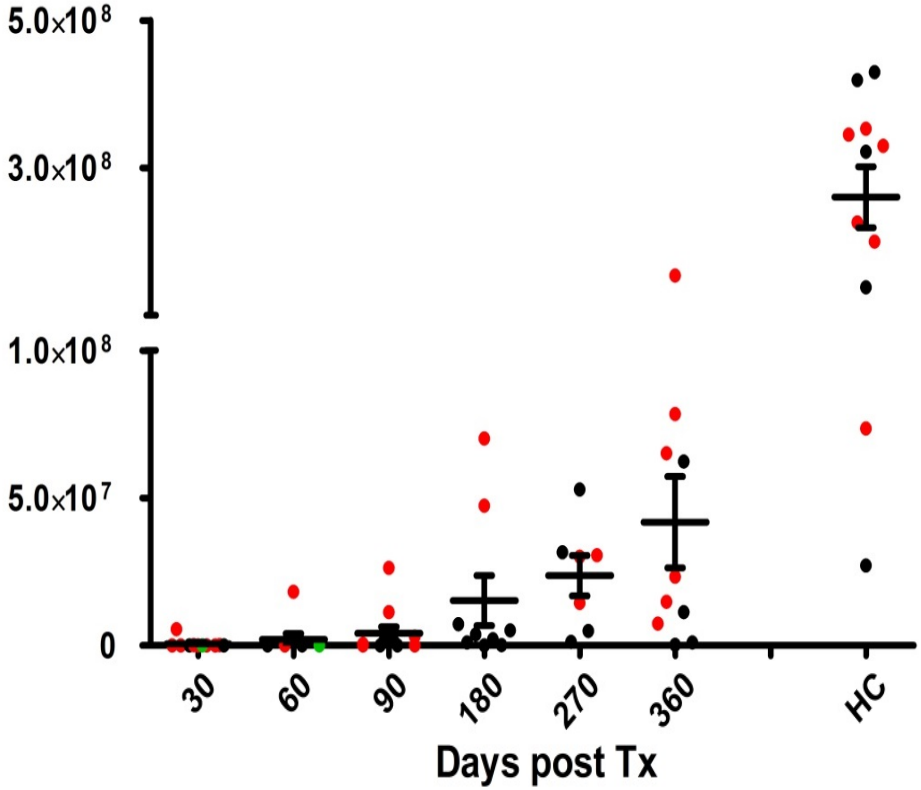
Persistent mixed T-cell chimerism despite ciclosporin discontinuation following FCC HSCT for SAA



Grimaldi *et al*, 2016 BBMT, in press

CD4+ T-cell composition of naïve, memory and effector subsets near normal at 1 year and expression of CD31 by naïve CD4 T cells indicates renewed thymopoiesis





CD4+ Naive: CD45RA+, CD27+ CD62L+

CD4+ Memory: CD45RA-, CD27+

CD4+ Effectors: CD45RA+/-, CD27- CD62L-

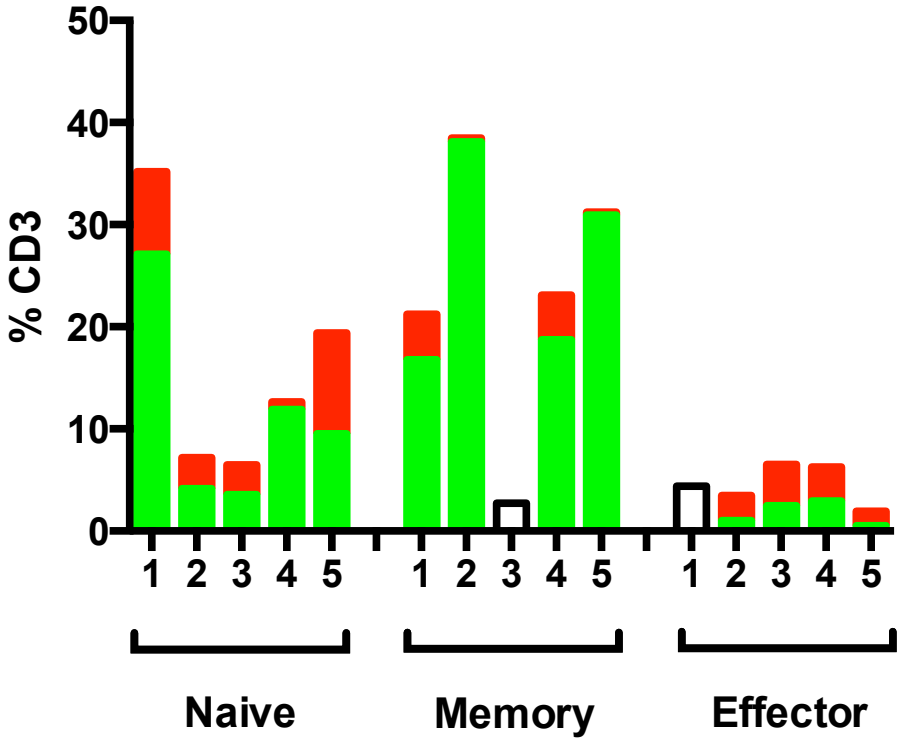
CD4+ Recent Thymic Emigrants:

CD4+ CD45RA+ CD27+ CD62L+ CD31+

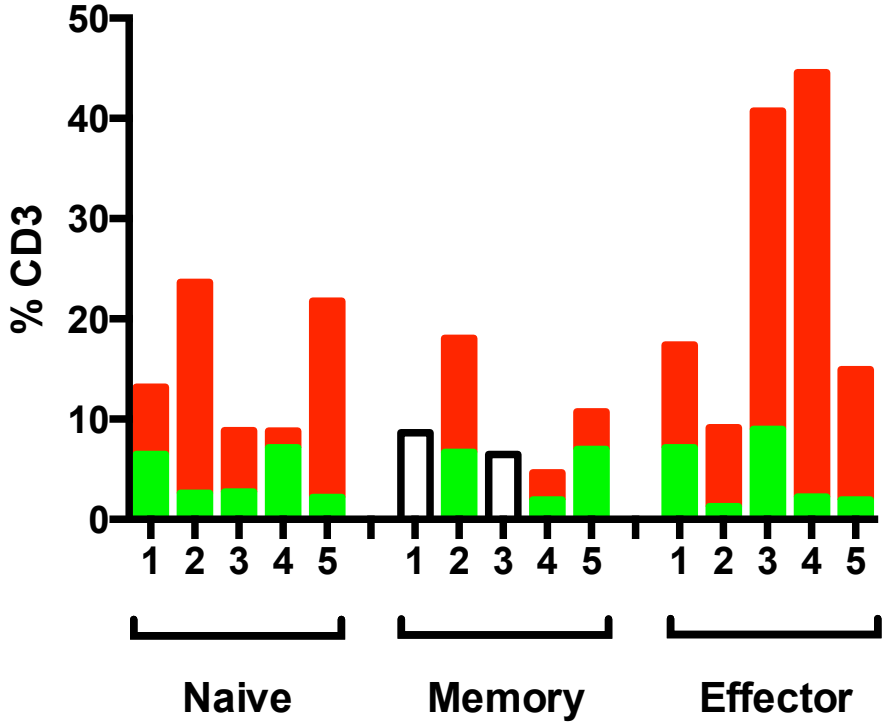
● Age <50 yrs ● Age ≥ 50 yrs

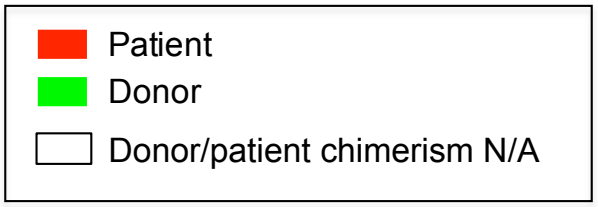
Mixed T-cell chimerism at 1 year principally due to persistence of patient CD8+ T-cells with notable contribution of effector subset

CD4 T cell subset chimerism at 1 year



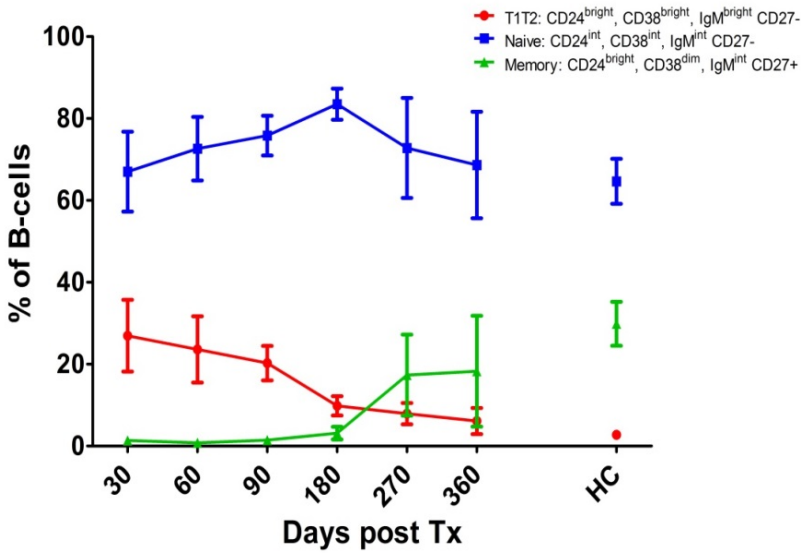
CD8 T cell subset chimerism at 1 year

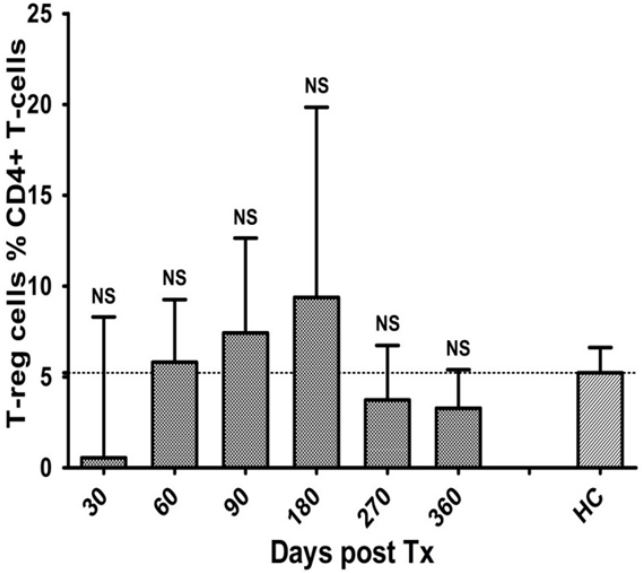




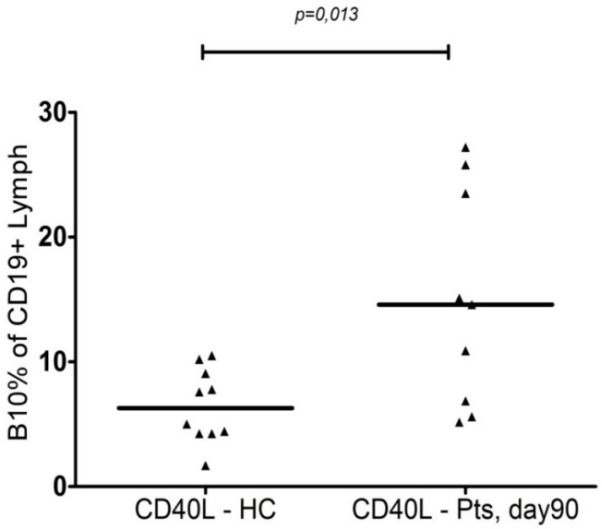
Presence of B and T cells with regulatory properties may facilitate co-existence of donor and patient T cells and low rates of GVHD

Increased proportion of B cells with an immature transitional phenotype present early after HSCT, (with regulatory properties)





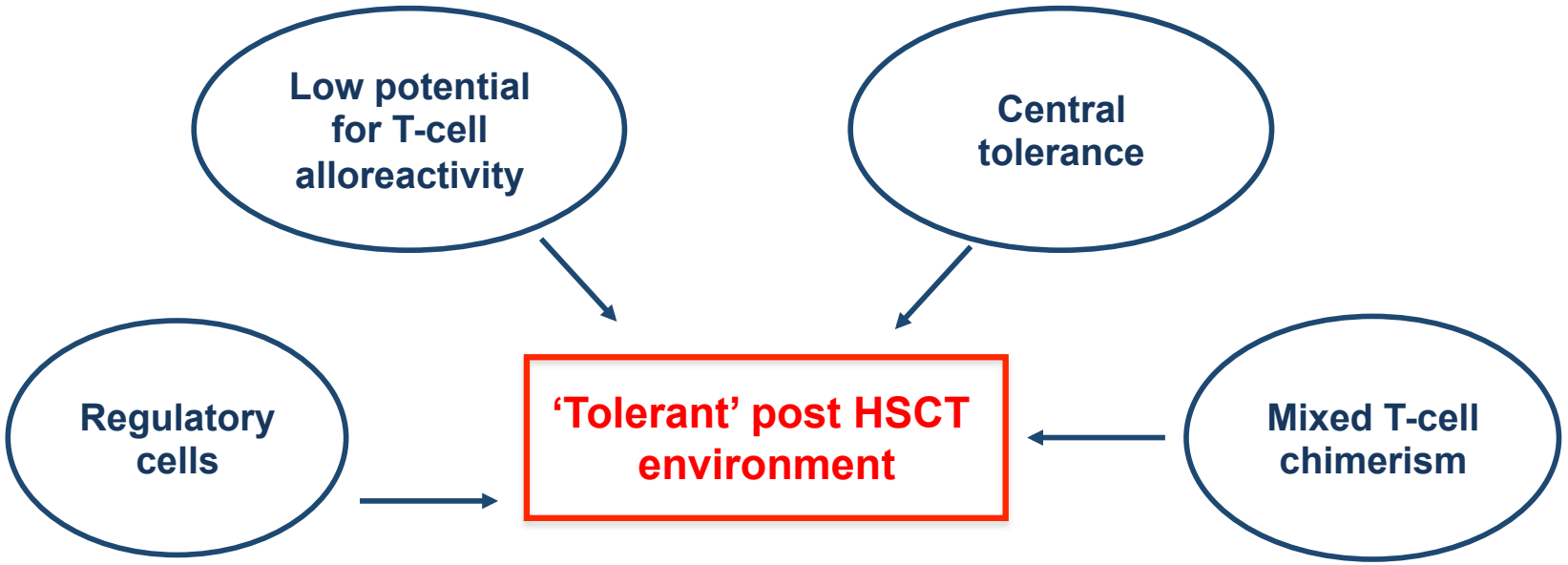
More IL-10 producing B cells present early (day 90) after HSCT



Normal proportions of regulatory T cells ($CD25^{high}$ $CD27^{+}$ $FoxP3^{+}$) within the CD4+ T-cell population

Basis for low incidence of GvHD and prolonged mixed T-cell chimerism following FCC HSCT appears to be multifactorial:

- Sustained low T cells numbers
- CD4+ T-cell recovery donor-derived with thymic education
- Recipient-derived effector CD8+ T-cells shape mixed CD3+ chimerism
- CD4+ T cells with regulatory phenotype present
- High number IL10 producing B cells



FCC conditioning for SAA HSCT

Advantages

- Sustained myeloid engraftment
- Mixed T-cell chimerism
- Low risk of GVHD
- Low toxicity; no need for MTX, or
- Irradiation in MUD HSCT

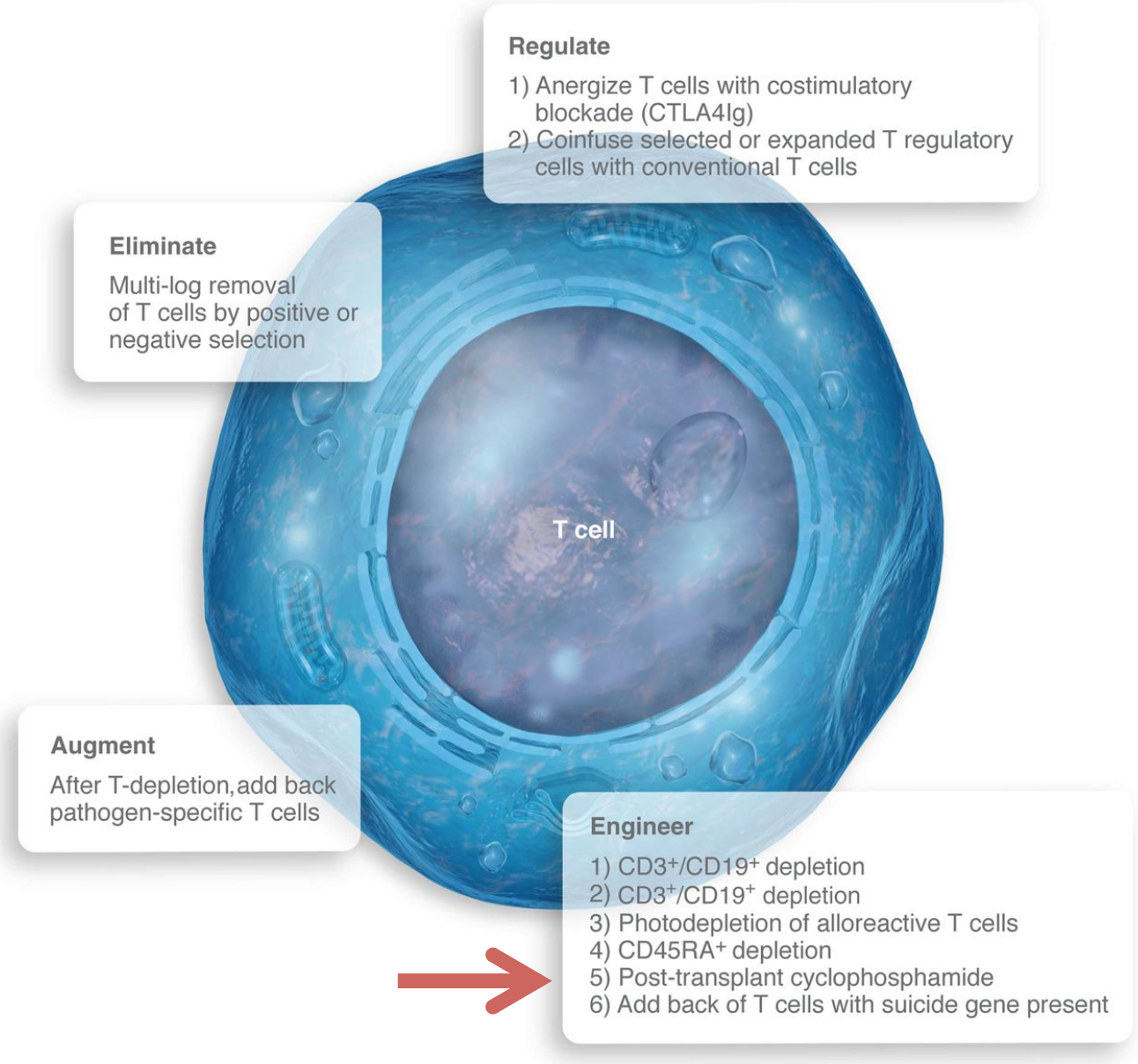
Disadvantages

- Risk of viral infections?
- Secondary autoimmune disorders
- Drug availability

Especially attractive for older patients

General categories and specific approaches that have been taken to facilitate and improve outcomes after haploidentical allogeneic hematopoietic cell transplantation.

Pulsipher M A, "Haplo is the new black" Blood 2014;124:675-676



Haploidentical HSCT for refractory SAA

Attractions

- Graft is available for most patients
- Cost is low compared to cord blood unit(s)
- Time to procure graft is short

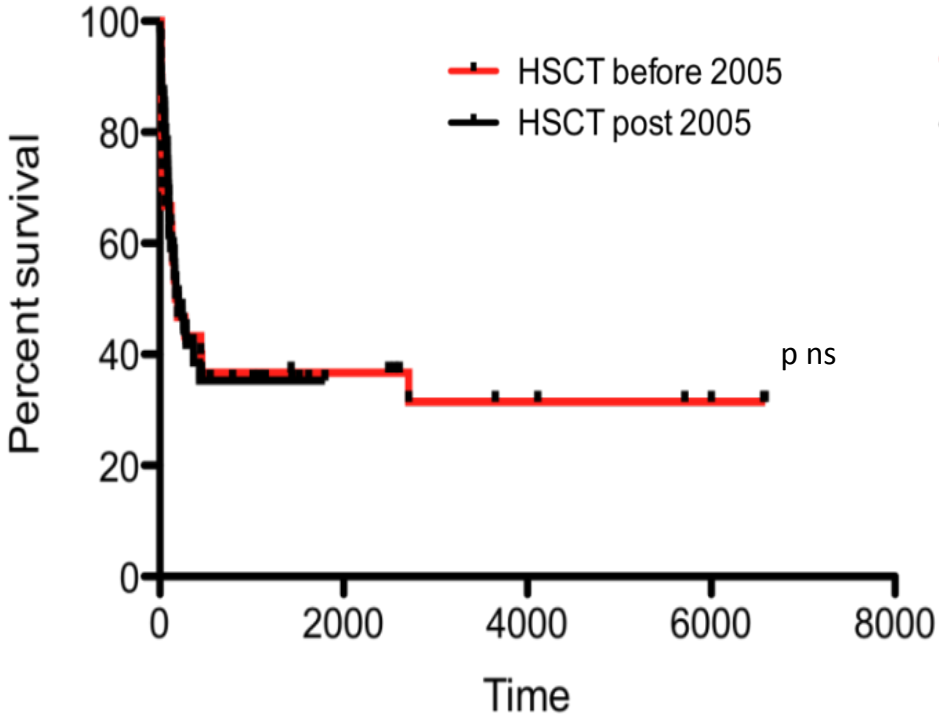
But...

- Recipient HLA antibody(ies) directed against donor precludes use of that donor
- Poor outcomes: High risk GVHD and graft failure, immune deficiency
- Published data are limited, restricted mostly to younger patients

EBMT survey of haploidentical HSCT in refractory SAA

Retrospective study, 1976-2011
82 transplants in 73 patients
Myeloablative regimen (42%)
Ex vivo TCD (52%);
Med age 12 y (2-69)

Results
Engraftment 58%
Acute GVHD (II-IV): 35%
Chronic GVHD 25%

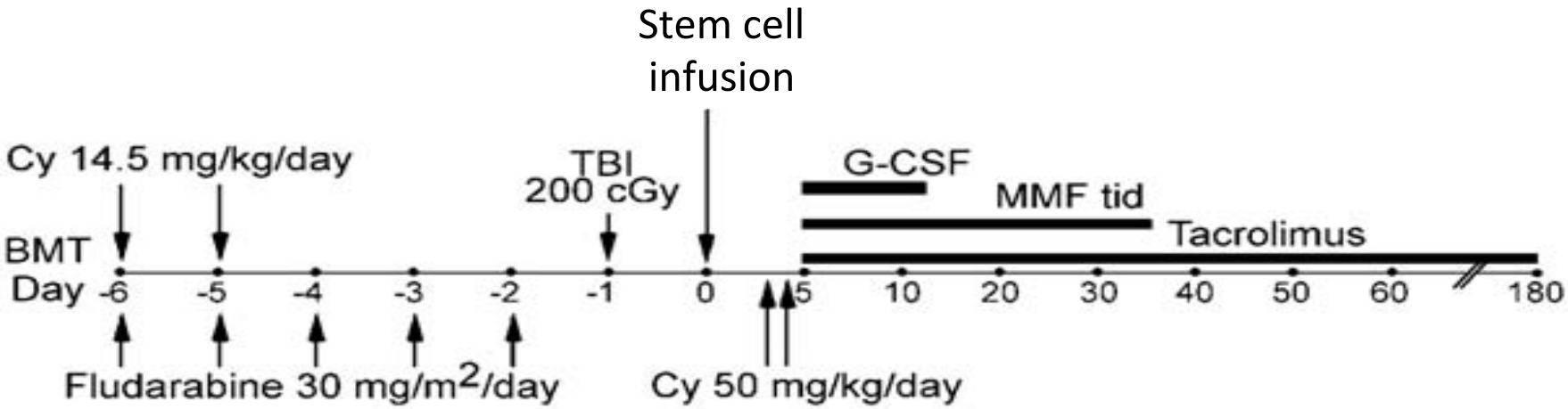


Hence a new approach is needed .....

Ciceri et al . Bone Marrow Transplant. 2012; 48:183

Non-myeloablative peripheral blood HSCT for refractory SAA using post transplant CY

Johns Hopkins protocol



Luznik. BBMT 2008; 14: 641

Results of haploidentical HSCT for refractory SAA

Age/ donor	Disease	Previous therapy	Previous HSCT	CD34 dose ($x 10^6$/kg)	Engraftment (neut, platelet)	GVHD	Status
19y, mother	SAA-MDS	ATG +CSA x 3	Cord-GF	6.7	D+18, D+ 21	None	Alive
51y, sister	2^0 SAA	ATG+CSA	MUD-GF	4.5	D+18 ↓ plt engraftment	None	Dead 22mo GBS,sepsis
23y, sister	SAA/PNH	ATG+CSA	None	5.8	D+16, D+26	Gd II acute	Alive Healthy baby
57y, brother	SAA/PNH	ATG+CSA, danazol, eltrombopag	MUD-GF MUD-GF	4.9	D+20, D+27	None	Alive
22y, brother	VSAA	ATG+CSA	None	6.9	D+23, D+27	None	Alive
20y, father	VSAA	CSA, ATG,MMF androgens	None	6.7	Non-engraftment HLA Ab +	N/A	Dead: GF
50y, son	2^0 SAA	None	MUD-GF MUD-GF	8.3	D+19, D+25	None	Alive
41y, mother	VSAA/PNH	ATG+CSA x2	None	1.8	Non-engraftment HLA Ab +	N/A	Dead: GF
44y,brother	VSAA	CSA. Recurrent E.coli sepsis, liver, rectal abscesses	None (MUD withdrew)	11.1	D+ 17, no plt engr.	None	Dead: E coli, restrictive pericarditis

Clay et al. Biol. Blood Marrow Transplant 2014

Recent results of HAPLO transplants in adult SAA

Study	N	Conditioning	GVHD prophylaxis	Engraftment	aGVHD II-IV	Chronic GVHD	Alive at 1 yr
Esteves, 2015	16	RIC	PT-CY#	100%	2 (12%)	3 (20%)	67%
Jaiswal, 2015	10	RIC: Flu/Mel/ sirolimus (5)	PT-CY	100%	1 (11%)	2 (22%)	60%
Clay, 2014	8	RIC	PT-CY	75%*	1 (12%)	0%	63%
Li, 2014	17	RIC	ATG,CSA, MTX,MSC	94%	23%	14%	76%
Gao, 2014	26	RIC	ATG, CSA	92%	12%	40%	84%

PT-CY = post transplant high dose cyclophosphamide, with CSA and MMF

- 2 graft rejection, both had donor directed HLA antibodies
- Miao et al, Blood 2015, 3227 n=39, haplo + cord , OS 83,2%

Modified from Bacigalupo and Sica. Semin. Hematol 2016;53:115

Miss KCB

- 20 yr old female of Jamacian origin referred with pancytopenia
- Initial Investigations consistent with Aplastic Anaemia
- Commenced on ATG +CsA (EBMT Rabbit ATG prospective study) 2009
- Partial response with development of PNH clone
- Admitted with Gram negative septicaemia → ITU requiring ventilation and dialysis
- Steroid induced diabetes
- AVN of hips and shoulder → bilateral decompression of femoral heads 2010
- Right humeral and Left femoral Klebsiella osteomyelitis 2011
- Vulval abscess (E.coli, P. aeruginosa)
- Recurrent admissions to local hospital with sepsis

Aiming for transplant

- Sibling typing → no full match but haploidentical sibling 16 years of age
- Unrelated donor search → no potential donors
- Repeat bone marrow: consistent with AA, no evidence of evolution to MDS
- Feb 2012 → decision to proceed to RIC-Haplo
- Patient concerned about fertility - Egg harvest, Soliris commenced in view of concerns re thrombosis
- 12th July 2012 admitted with neutropenic sepsis → proceed with transplant when stable.
- Pre-transplant investigations
  - FEV1 77%, Predicted TLCO 58%
  - GFR 61
  - EF >55%
  - No donor directed HLA antibodies

Transplant

- PBSC harvest from younger sister → 1/8/2012
- Admitted to ITU with sepsis at D16 → intubated for five days, requiring inotropes, Candida grown from blood cultures
- Engraftment D17
- Discharged at D40
- Now 54 months post HSCT
  - No GVHD
  - Delivered male child at 18 months (no assisted conception required)

Conclusions

- FCC protocol achieves excellent results in both adults and children with matched and unrelated donors
- Mixed donor chimerism provides the platform for very low rates of acute and chronic GVHD
- PBSC may be an acceptable option with Campath
- OS and EFS is better than IST
- Consideration, with caveats, to upfront unrelated donor transplant in both children and adults (age discussion)
- Haplo using post Cy (Baltimore protocol) is an effective protocol when no other donor is available/emergency situation
- Both protocols retain fertility

THANKYOU

Judith Marsh, Ghulam Mufti, Victoria Potter, Austin Kulasekararaj, Kavita Raj, Hugues de Lavallade, Donal McLornan

Rest of the Laboratory, Data Managers and Nursing teams

UD selection algorithm
King's

- HLA compatibility
  - 10/10, 9/10 (DQB1, A, any, B last) - UD 1
- **Availability/Urgency** Trumps
- CMV status 2
- Age 3
- Gender 4
- Parity 5
- ABO Trumps
- Route of donation Trumps