Cord blood Services

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Cord Blood Programmes
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ANTHONY NOLAN
BE A MATCH, SAVE A LIFE

BANC DE SANG I TEIXITS

Services

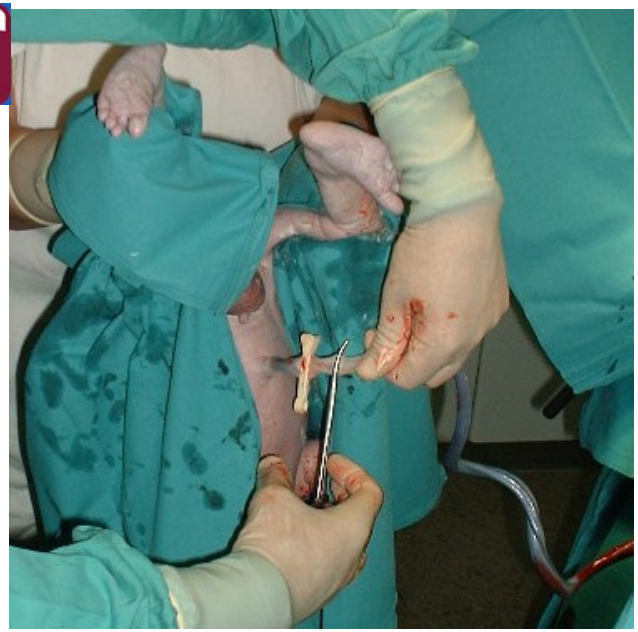
- CBB
  - Unrelated
  - Related
  - Autologous
- CB Infusion
- Biobanking
- CordPharm

Advantages of CB

Assuming equivalent outcomes to adult sources:

- Equitable / Universal
- Timely (“off-the-shelf”)
- Practical (multiple options)
- Donor care: attrition, safety
- Sustainable
- Room for improvement

Cord blood is a naturally discarded tissue…

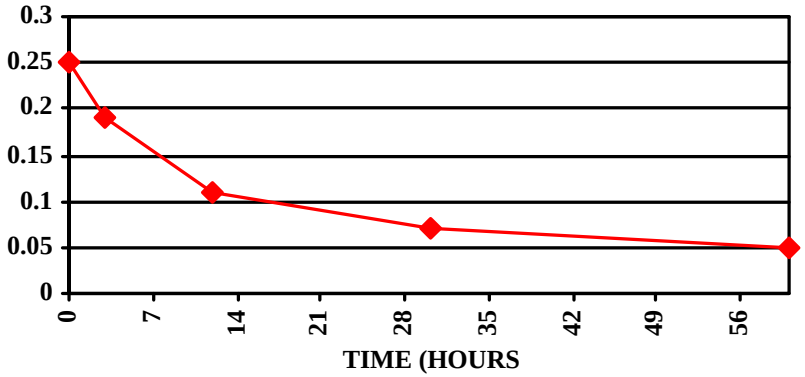


That contains a high proportion of circulating haematopoitic stem cells…

Foetal and neonatal peripheral blood contains circulating stem cell filling a constant space

Edad	Hgb	Hematies	Hcto (%)	Eritro (%)	Leucos	PMN (%)	Plaquetas	CD34
10ª semana	ND	ND	ND	90	1,1	0	ND	56
18-21ª semana	11,69	2,85	37	45	2,57	6	234	50
22-25ª semana	12,2	3,09	39	21	3,73	6,5	247	55
26-29ª semana	12,91	3,46	41	21	4,08	8,5	242	41
30-parto	13,64	3,82	44	17	6,4	23	232	40
SCU	**15,3**	**4,3**	**49**	**4**	**15,7**	**50**	**290**	**47**
Dia 1 postnatal	18,4	4,8	58	3	18,8	70	192	20
Dia 3 postnatal	17,8	4,6	55	1	9,5	50	213	5

Hgb: g/dL; Htes: 10e9/L, Leucos: 10e3/u; Plaquetas:10e6/mL; CD34: cel/uL

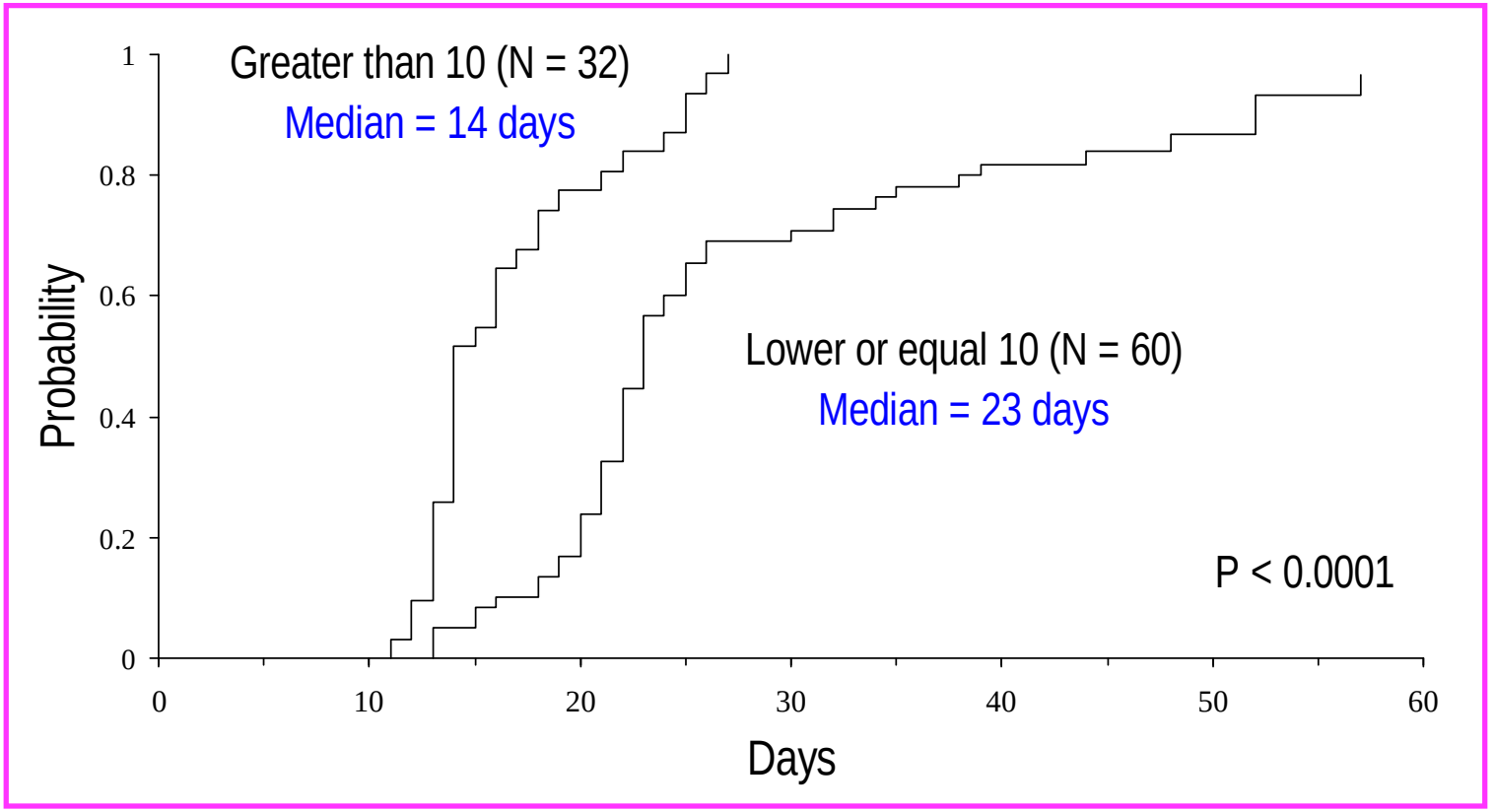


Gonzalez S et al. Cytotherapy, 2008

Able to reconstitute the haematopoietic tissues…

Myeloid Engraftment

Absolute number of CD34+ cells infused ($x10^6$)

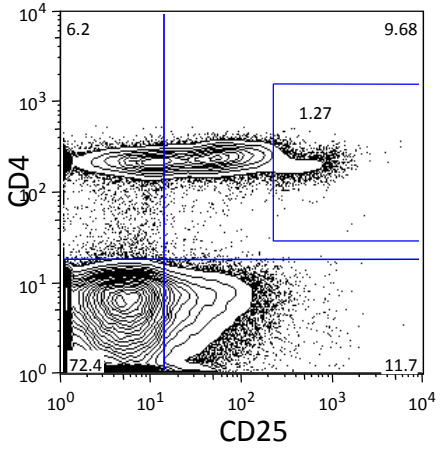


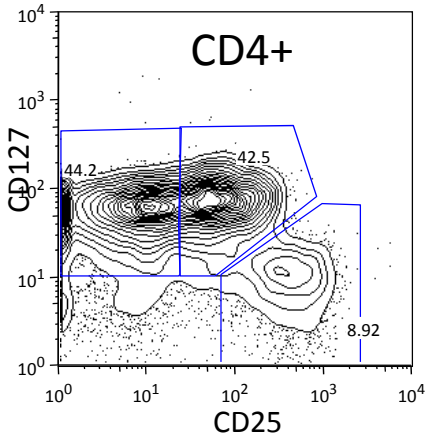
Hospital Universitario La Fe, Valencia, Spain

But also carries a unique profile of immune cells…

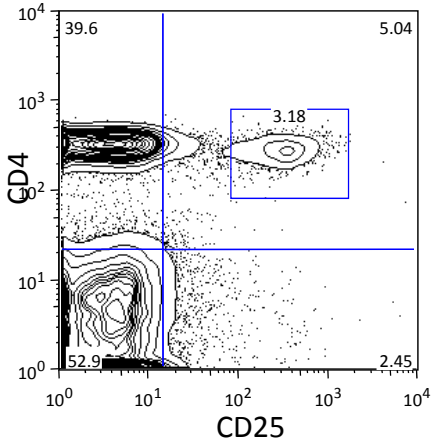
Cord Tregs are a much more defined population than adult PBMC

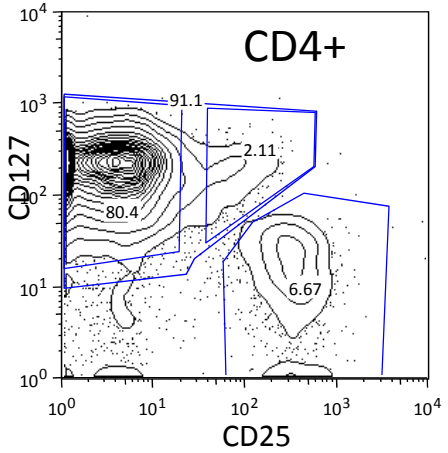
Adult



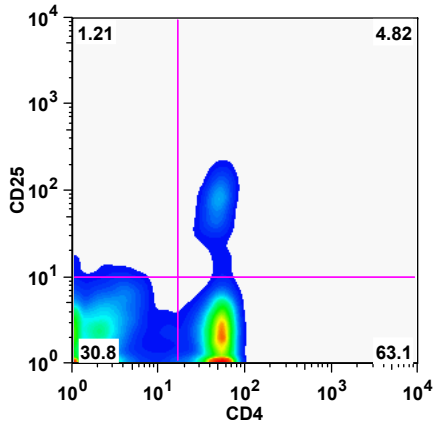


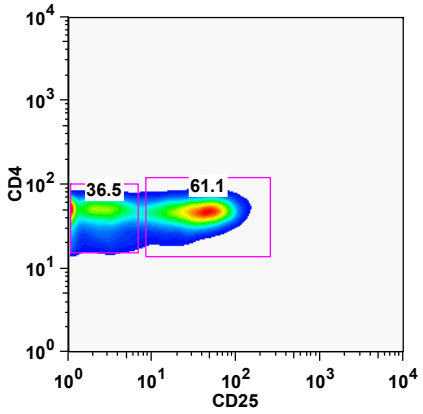
Cord

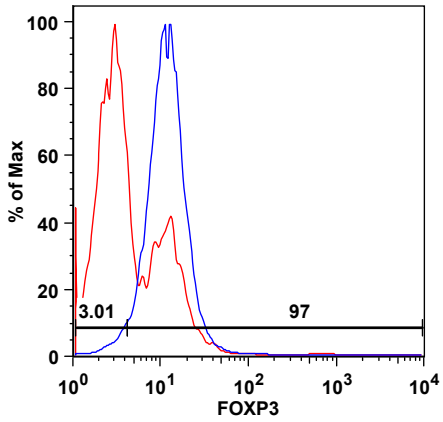


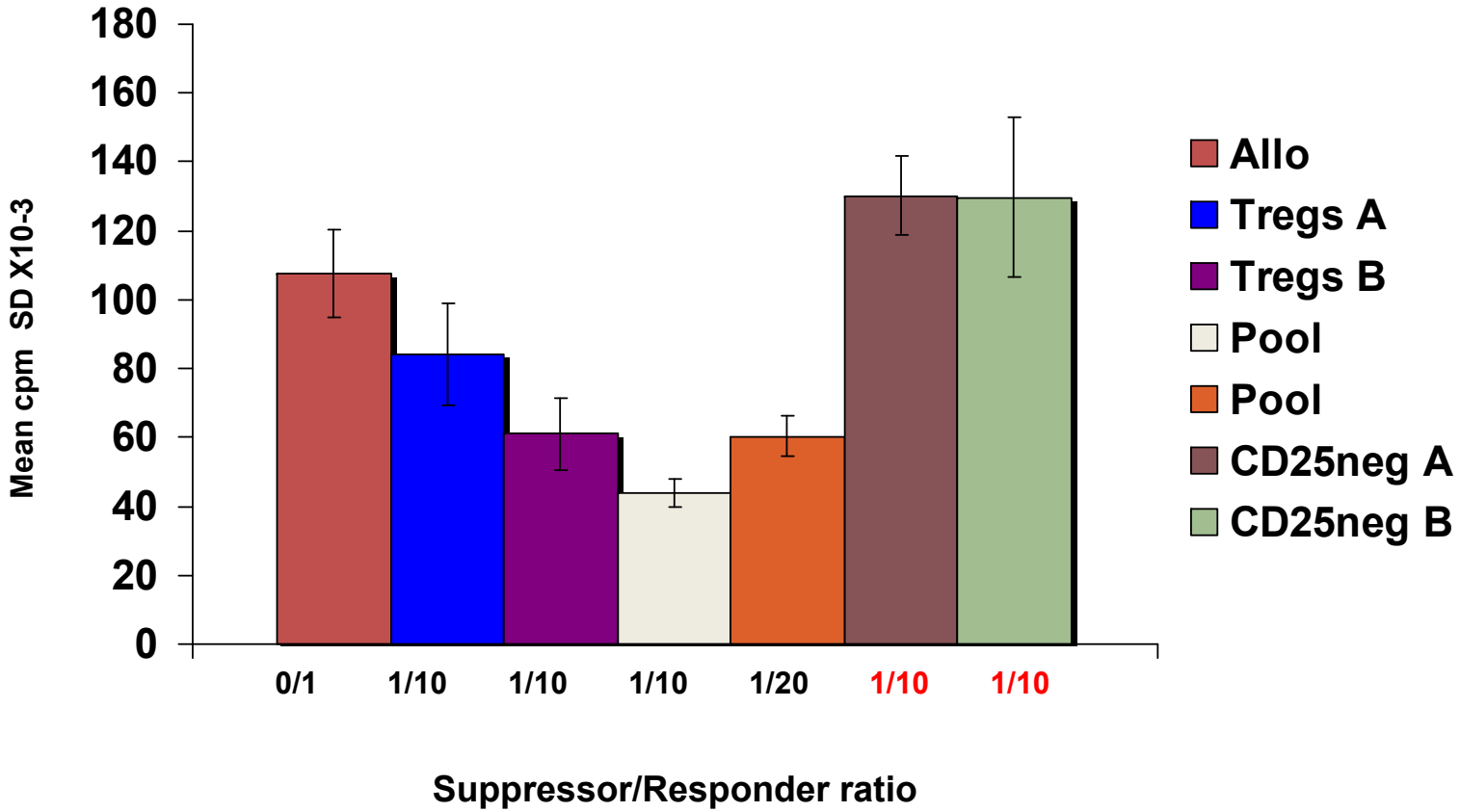


CB Tregs suppress third party alloreactions

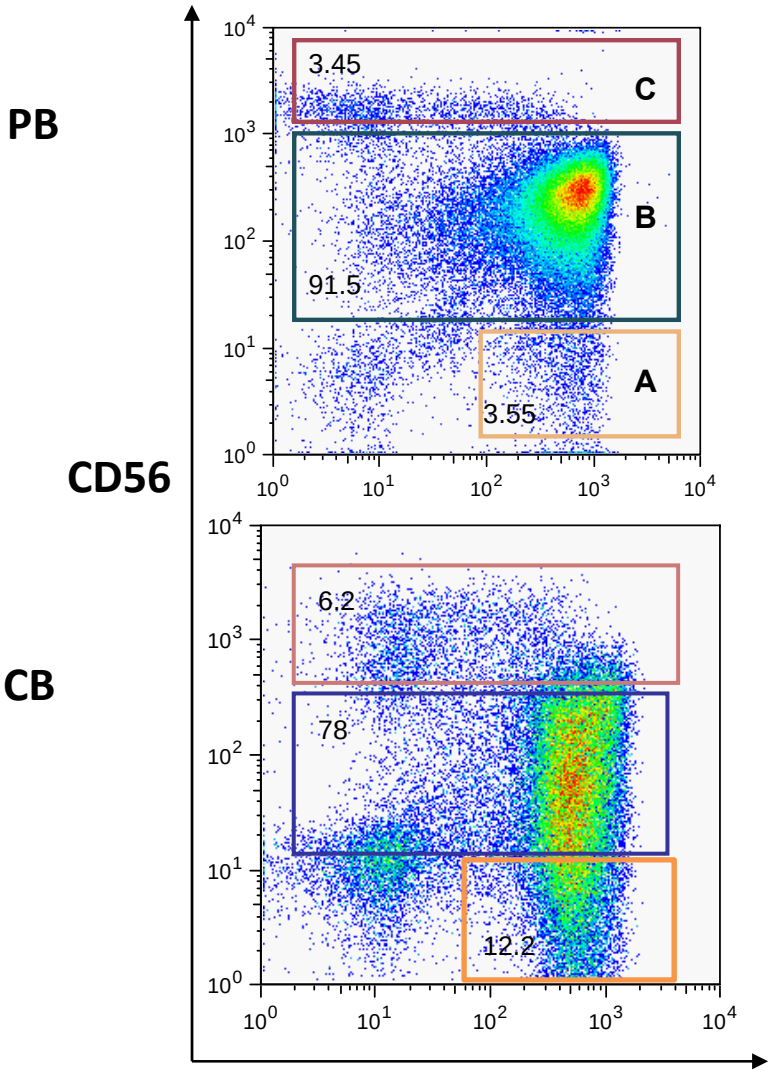








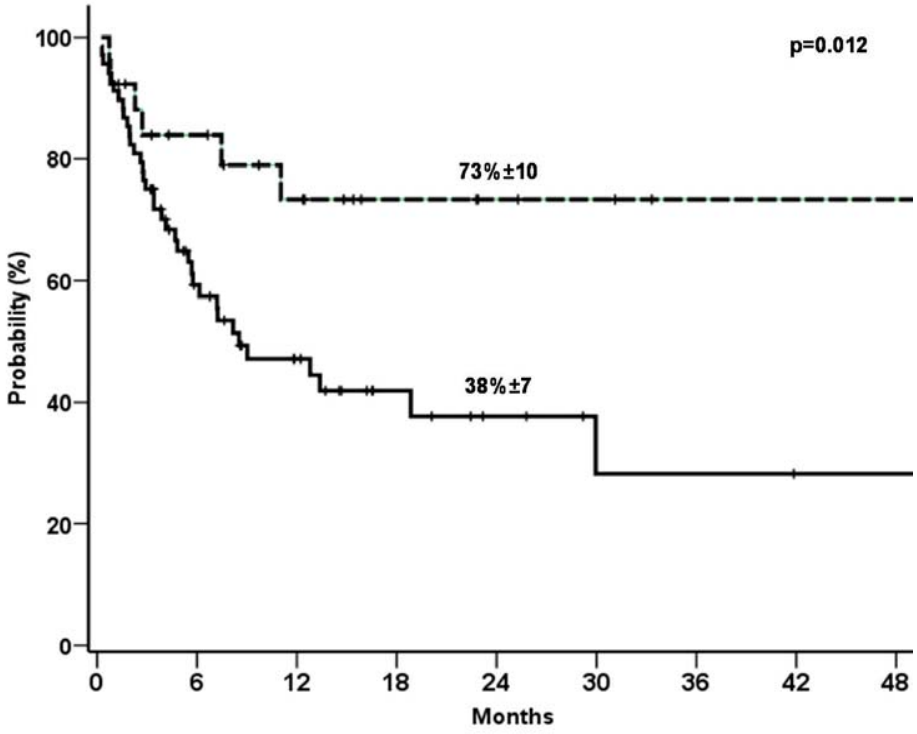
Heterogeneous expression of CD56 on CB NK cells

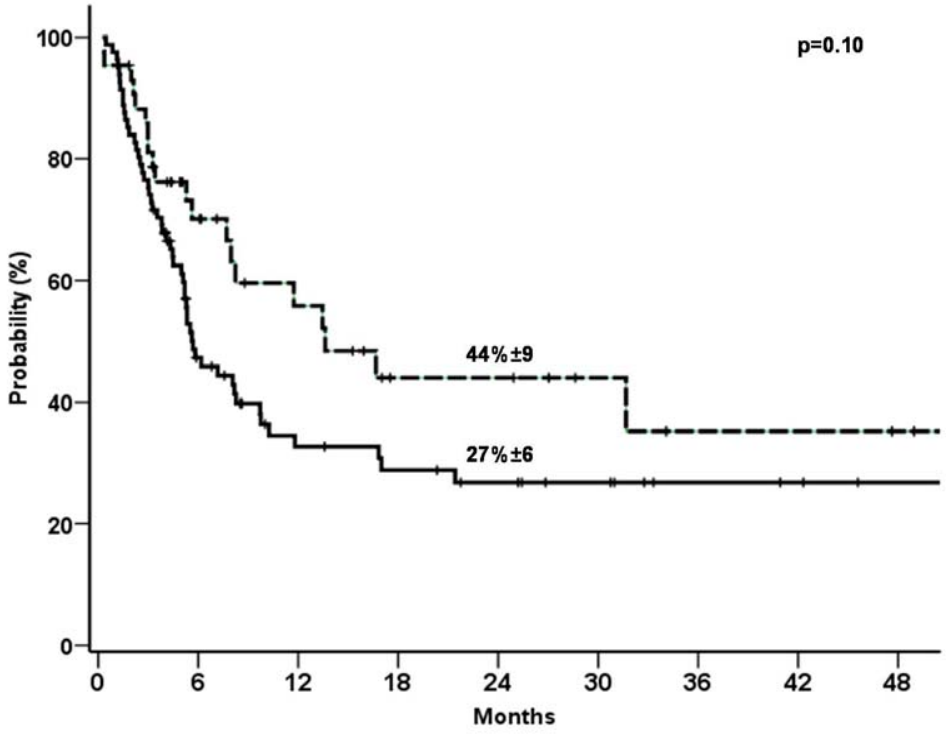


CD56 subpopulations in CB and PB

Effect of KIR Ligand Mismatching on GVH Direction after UCBT: LFS in AML and ALL

Willemze et al. Leukemia 2008





Allowing HLA mismatch transplantation…

High Resolution HLA Match

10 of 10					• 79%	14%
9 of 10				• 26%	16%	
8 of 10			• 8%	• 49%		64%
7 of 10		• 4%	• 49%	• 19%	• 5%	
6 of 10	• 7%	• 29%	• 33%	• 6%		
5 of 10	• 7%	• 35%	• 11%			22%
4 of 10	• 43%	• 27%				
3 of 10	• 36%	• 4%				
2 of 10	• 7%					
	2 of 6 (N=14)	3 of 6 (N=52)	4 of 6 (N=113)	5 of 6 (N=72)	6 of 6 (N=19)	N=216

Source: COBLT study

A typical CBT is 7/10

Providing cure to haematological malignancies disorders (GVL effect) with an attenuated risk of severe graft versus host disease…

Defective Th1 responses of UCB T cells may contribute to a reduce GVHD

- ✓ UCB **T cells are largely naïve** due to low exposure to environmental pathogens that decrease their activation capacity
- ✓ **Impaired** expression and **activation of Th1-driven transcription factors** (i.e. T-bet, STAT4, NFAT pathway) leading to reduce capacity to produce IFN-g.
- ✓ **IL10 production** during TCR stimulation which drives Th2 responses

Kim, Broxmeyer. Crit Rev Oncol/Hematol 2010

Impaired activity of UCB-derived DCs

- ✓ CB-CD14+ derived DC produce **lower amount of TNFa** and defect in **IL12** production
- ✓ **Lower expression of HLA-DR, CD40, CD86, CD83** after LPS stimulation, with normal LPS receptors (CD14 and TLRs) that can be restored after prolonging stimuli.
- ✓ **Higher ratio of pDCs** (CD123+) over mDCs (CD11c+)
- ✓ Inability of a full maturation generate antigen tolerance that maybe related to **plasma factors** as M-CSF and IL4

Kim, Broxmeyer. Crit Rev Oncol/Hematol 2010

EFF/REG BALANCE OF HLA-SPECIFIC NAIVE T CELLS

Total infused T cells

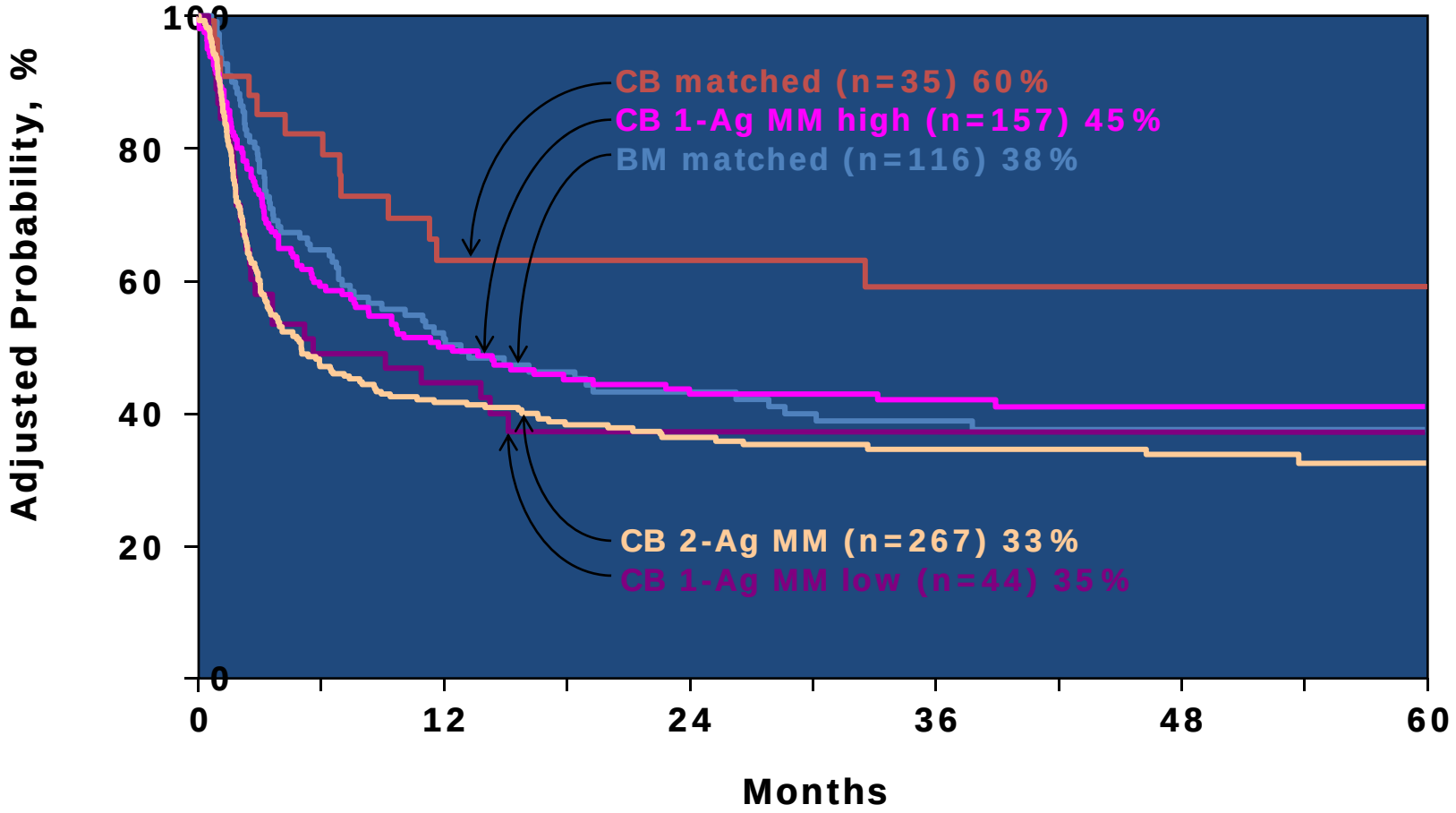
Subpopulation x10(8)	Cord Blood		Mobilized PB
CD4+CD45RA+CD25-	3.1		73
CD4+CD45RA+CD25+	0.2		0.4
Effector/Suppressor	x16		x199

Balance Eff/Reg on HLA specific T clones according HLA match

MATCH DEGREE	9/10	5/10
CB	2/1	16/1
BM/PB	22/1	199/1

Providing promising clinical results…

Leukemia-free Survival
Children



Eapen et al. Lancet 2007

And having room for improvement …

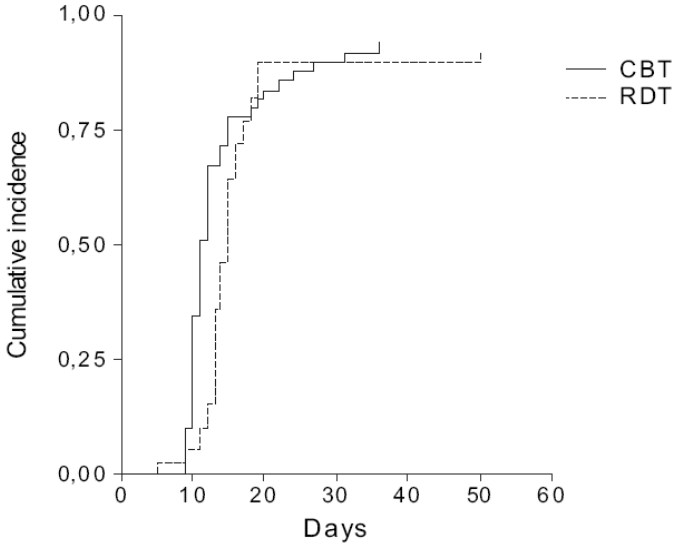
Biology of double CBT engraftment

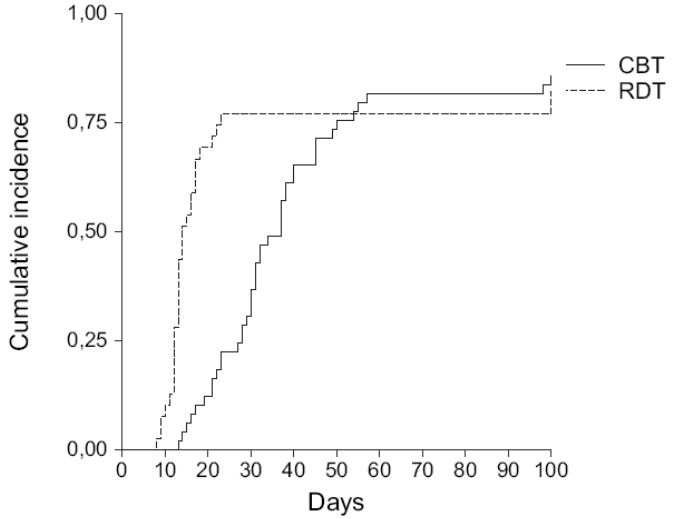
(summary from Eldjerou et al. Blood 2010)

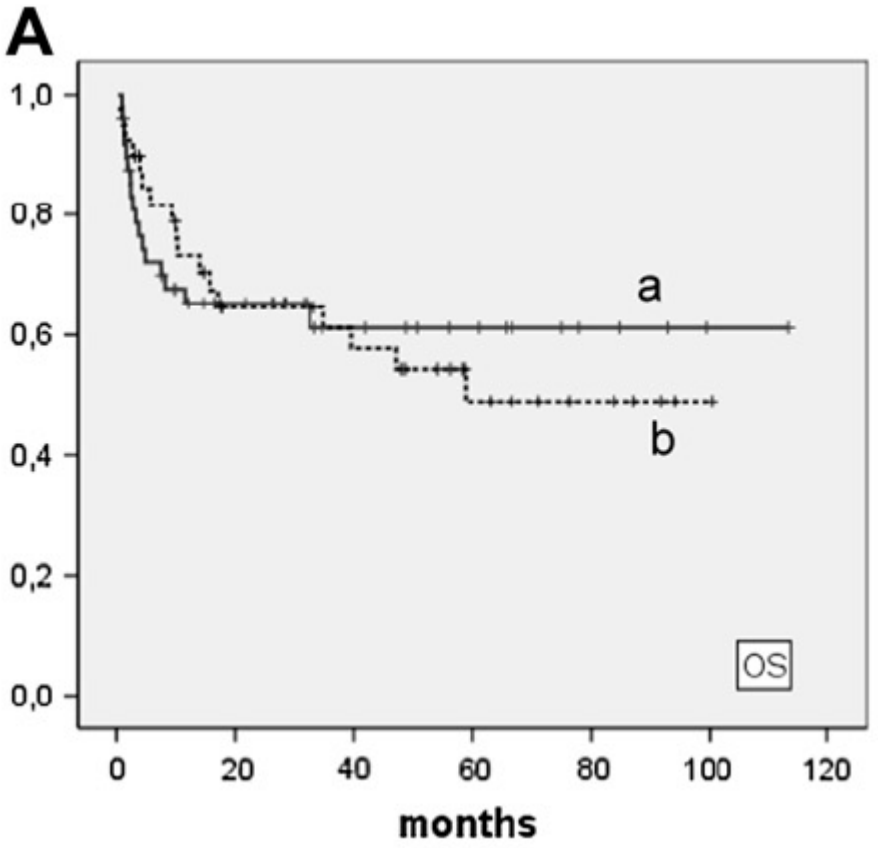
- ✓ When 2 CB units with similar potency are concomitantly infused to a conditioned patient, **immunecompetition between them select the winner** (i.e. **CD3 contain**, **T cell alloreactivity** (Gutman et al, Blood 2010)
- ✓ In a mouse model (NSG) which predicts the clinical winner unit, co-infusion of CD34+ selected cells promotes a mixed chimera. Adding-back **CD34-fractions** of any of them ,**restored single donor dominance** that coincides with their donor origin.
- ✓ It has been shown that when there is a significative difference on cell potency between the 2 CB units, **CD34 viability can predict the winner** (Scaradavou et al, BBMT 2009).

Dual CB transplants (CB plus CD34+TPD) provide comparable results to siblings in AL/MDS

(Sebrango et al, Best Practice & Res Clin Hematol 2010)

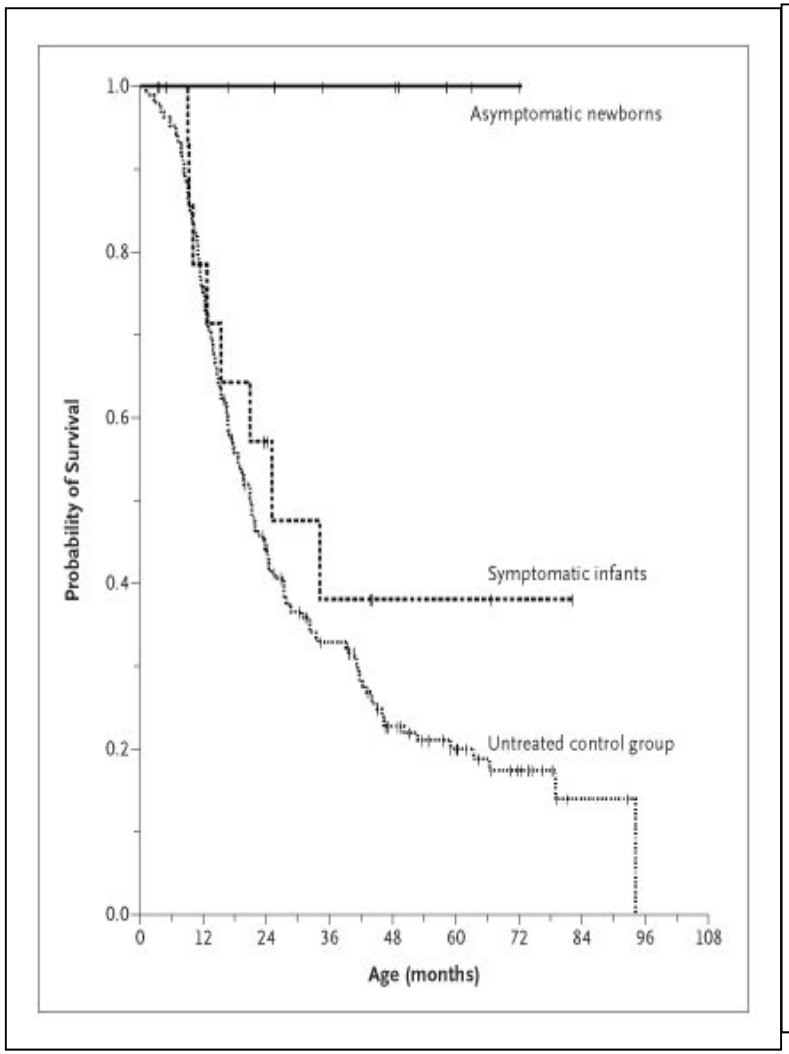


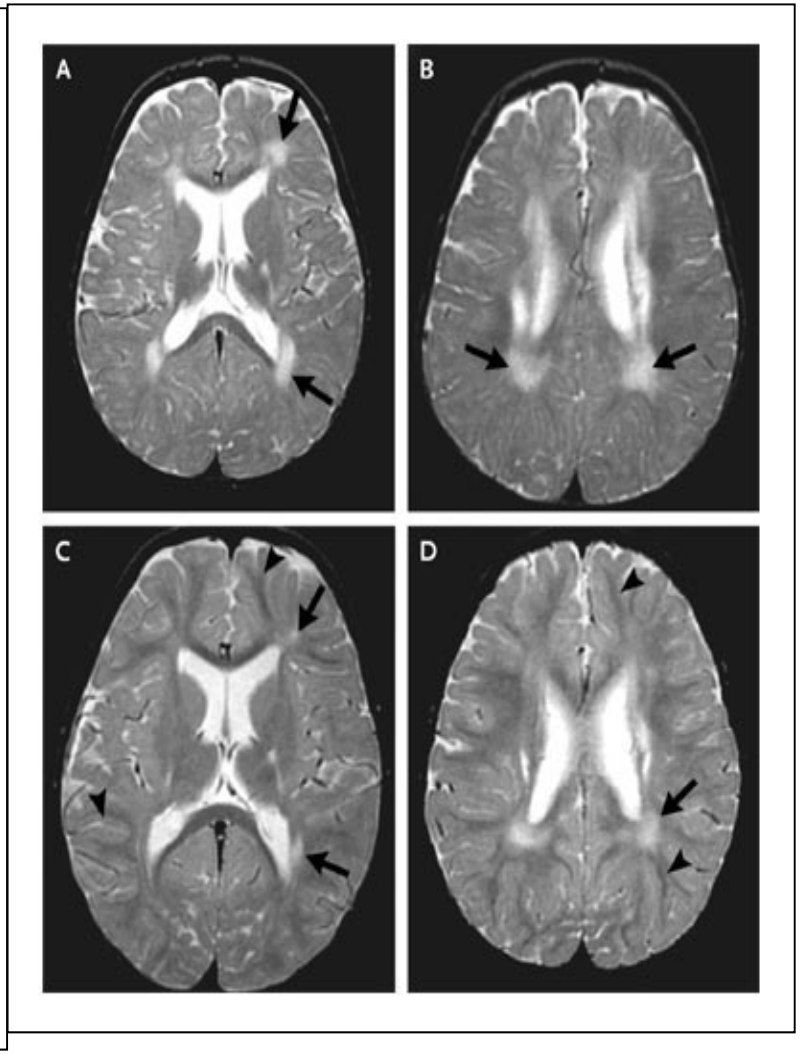




Additionally, it might contain certain multipotent stem cells

Cord blood and CNS regeneration





Krabbe's disease

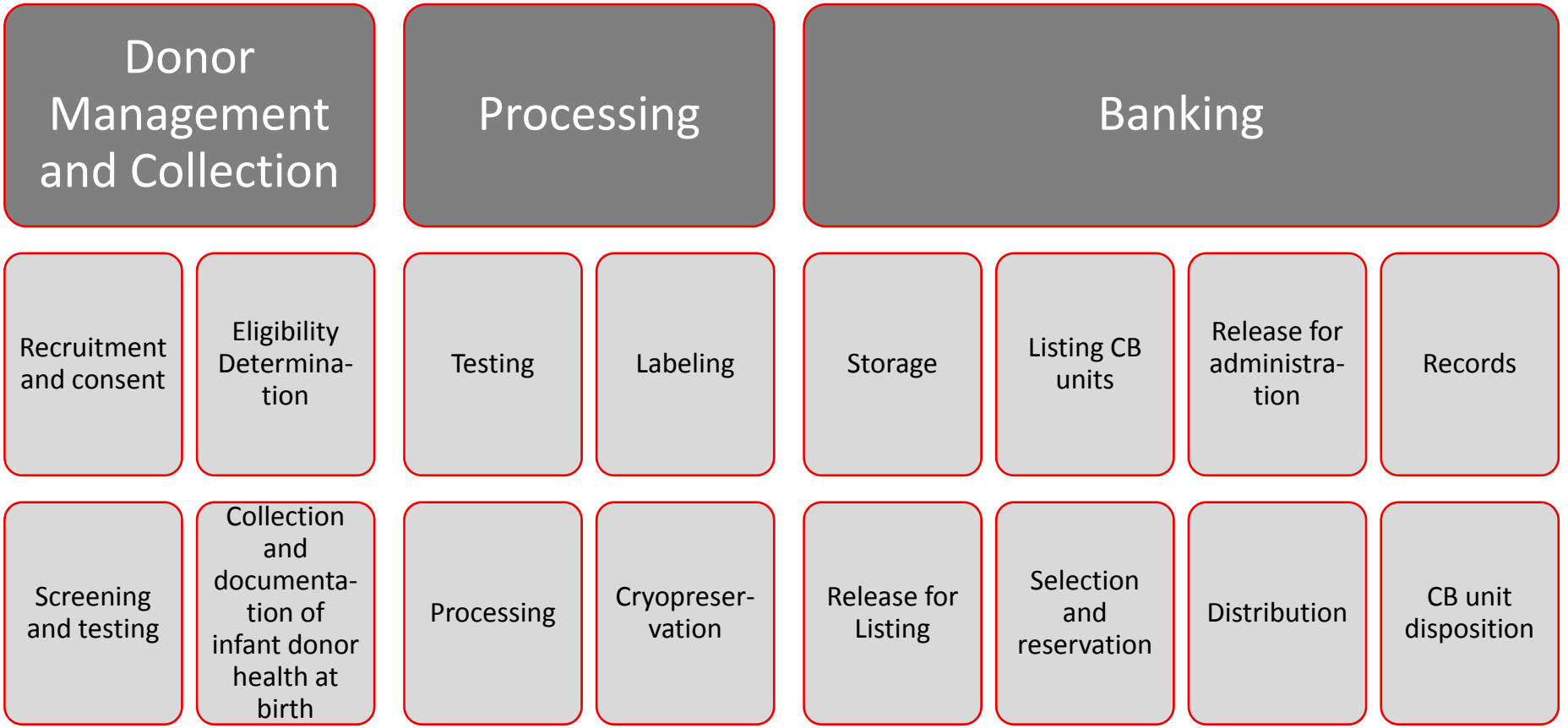
Escolar ML et al. New Engl J Med 2005, May 19

Multicompatiple donors

Haplotipo HLA	Frecuencia	Número homozigotos
A*29, B*44, DRB1*07	7,1%	16
A*30, B*18, DRB1*03	5,0%	11
A*01, B*08, DRB1*03	5.2%	7
A*03, B*07, DRB1*15	3.6%	2
A*23, B*44, DRB1*07	2.3%	2
A*02, B*44, DRB1*01	2.7%	2
A*02, B*44, DRB1*07	5.9%	2

This properties suggested the development of relatively large inventories of ready-to-use, fit-for-purpose tissue products for bone marrow transplantation: CORD BLOOD BANKS

Responsibilities of CBB

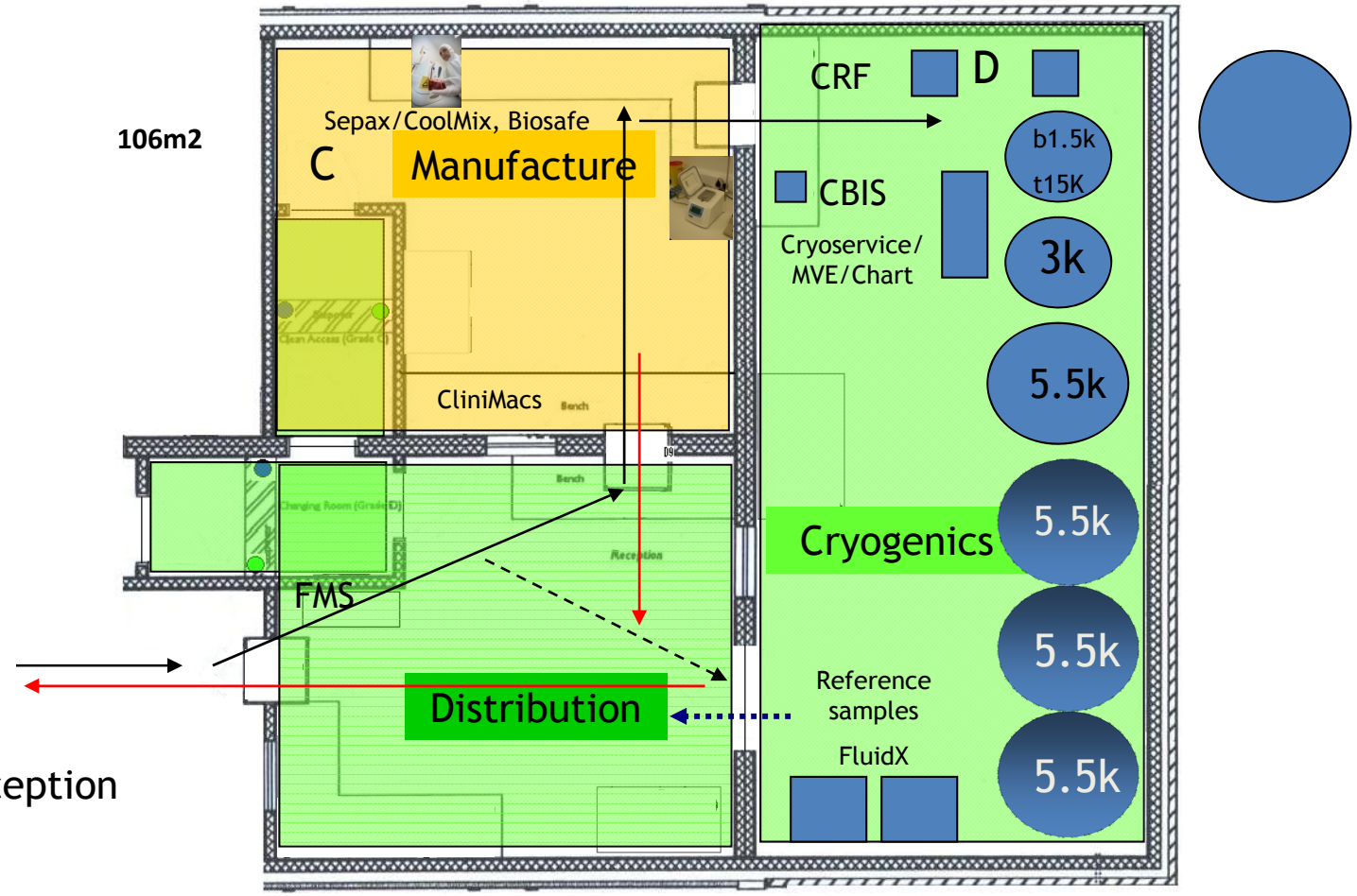


FACILITY: CLEAN ROOM

Ground floor:

106m2

Reception



Size: ~25,000

Reception

Distribution

ANTHONY NOLAN

BE A MATCH, SAVE A LIFE

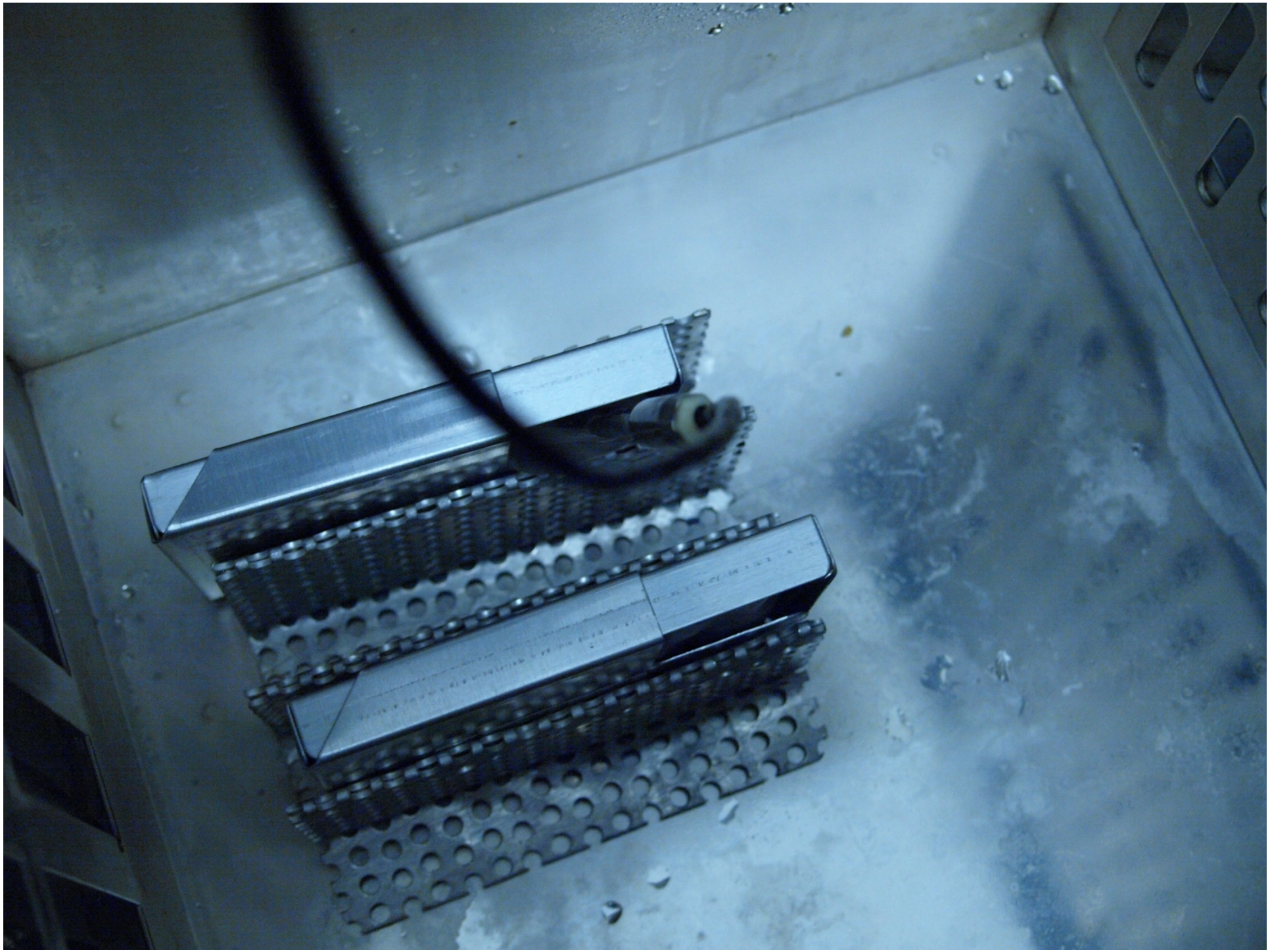
Manufacturing

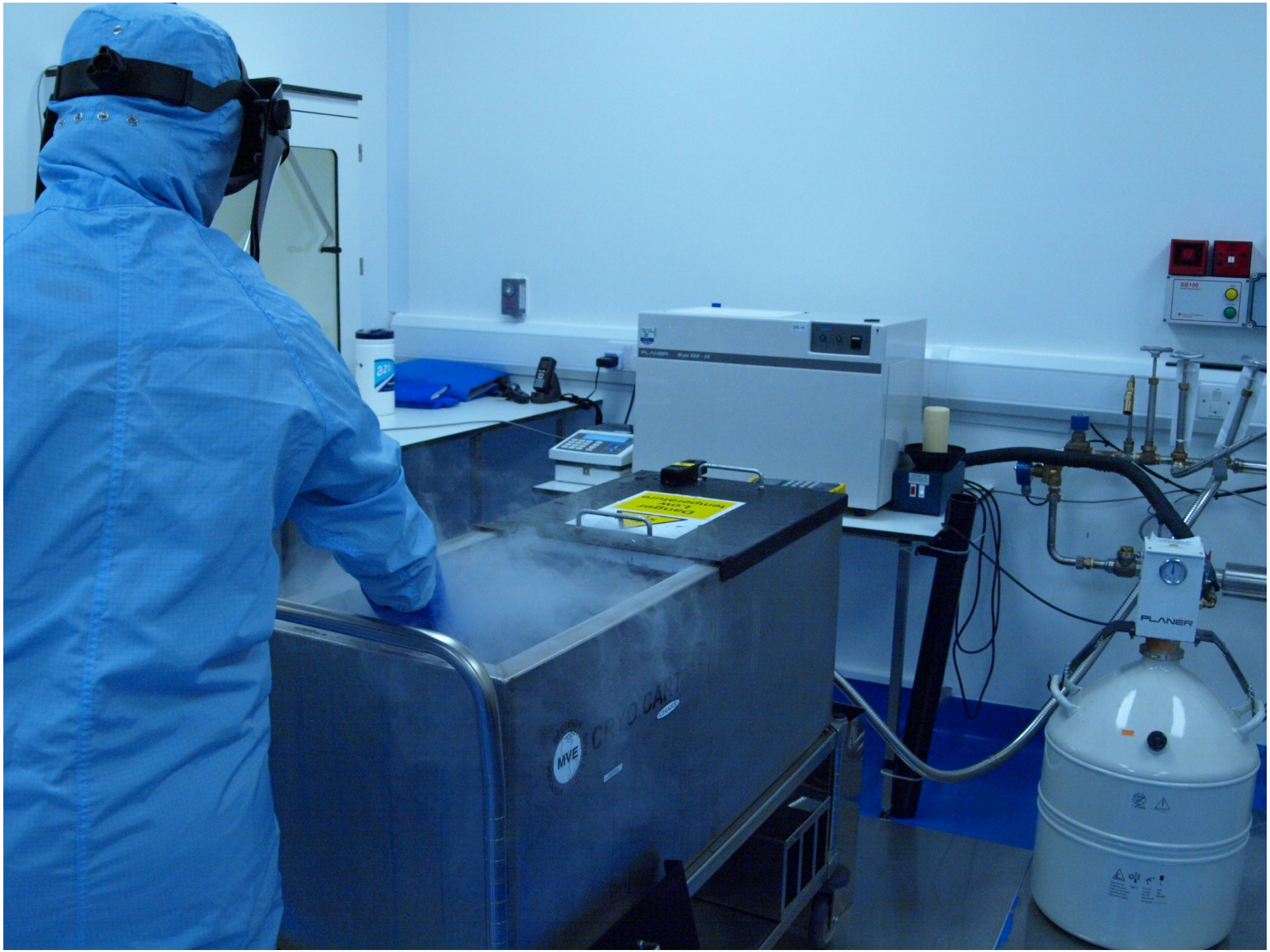
Cryopreservation

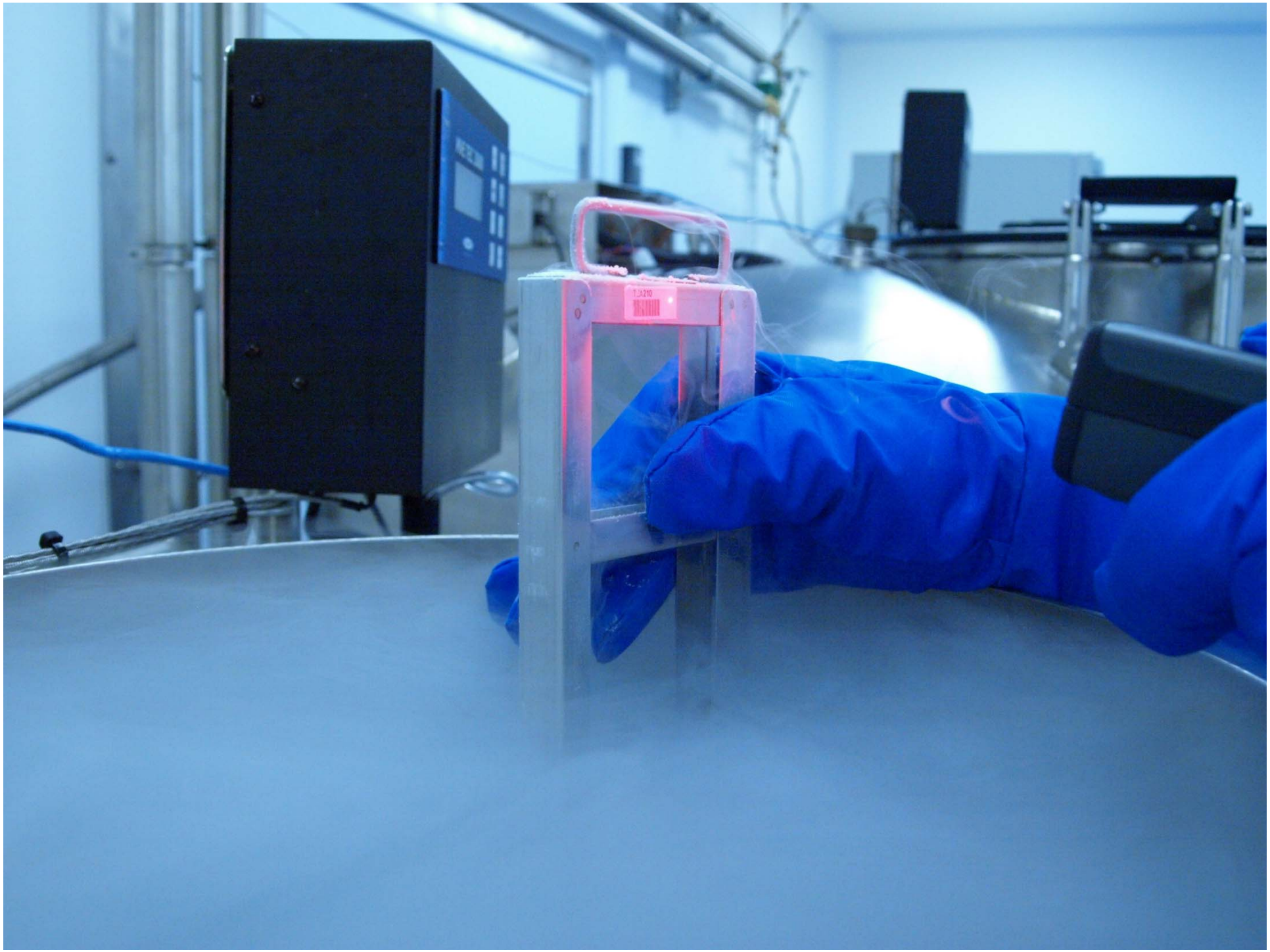
Long-term storing

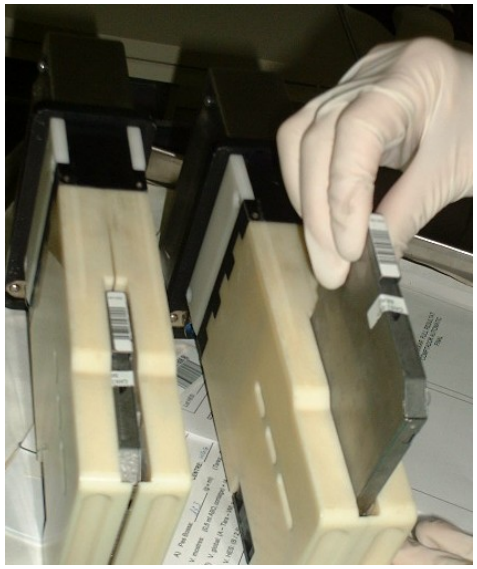

BIOHAZARD
+03.8°C
COOLMIX
CRM-05
CRM-07
PASS


G2212 09 100869
ANTHONY NOLAN CORD
BLOOD BANK, UK
Cryopreserved HPC, CORD BLOOD
Date Of Collection: 26 Oct 2009 16:20
Store < -150 C
Expiry Date: None
Applies
For Use Intended By Recipient Only
Big Compartment
Approx 20ml in 10%
DMSO, 1% Dextran




MVE
PLANER





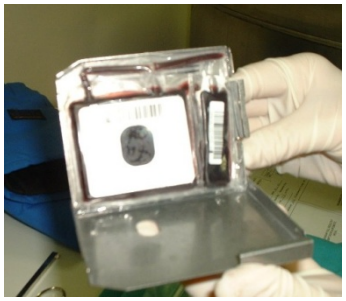
Programa
de Sang de Cordó
BANC DE SANG I TEIXITS

CORD BLOOD PROGRAMME CONCORDIA-BST

BARCELONA CBB

FINAL PRODUCT= volume 25 ML

Last 12 months	Mean (SD)	Median(Range)	
Volume of cryopreserved units (gr, including cryoprotectant)	25.9 (0.8)	25.9 (21.6-29.3)	n=478
NC/mL (x10e6)	46.5 (16)	44 (16-159)	
NC after processing (x10e6)	1206 (414)	1133 (404-3799)	
RBC/mL (x10e9)	2.2 (1.4)	2 (0.1-3.5)	
Platelet/mL (x10e6)	683 (184)	679 (61-1399)	
Hgb gr/dL	7.6 (4.1)	7.3 (0.3-24)	
Hto (%)	23 (12)	22 (1-75)	
Lymph (%)	41 (8)	40 (24-72)	
Mono (%)	7 (5)	6 (0.3-57)	
Granulo (%)	52 (10)	54 (8-72)	
CD34 (%)	0.34 (0.21)	0.31 (0.02-2.24)	CV 62%
CD34 cryopreserved (x10e6)	4.35 (3.68)	3.41 (0.3-33.4)	
CD34 viability (% 7AAD+)	99 (1)	100 (92-100)	
NC yield (%)	83 (10)	85 (34-112)	
RBC depletion (%)	88 (9)	90 (27-99)	
Platelet depletion (%)	50 (13)	49 (8-99)	



CBU VALIDATION

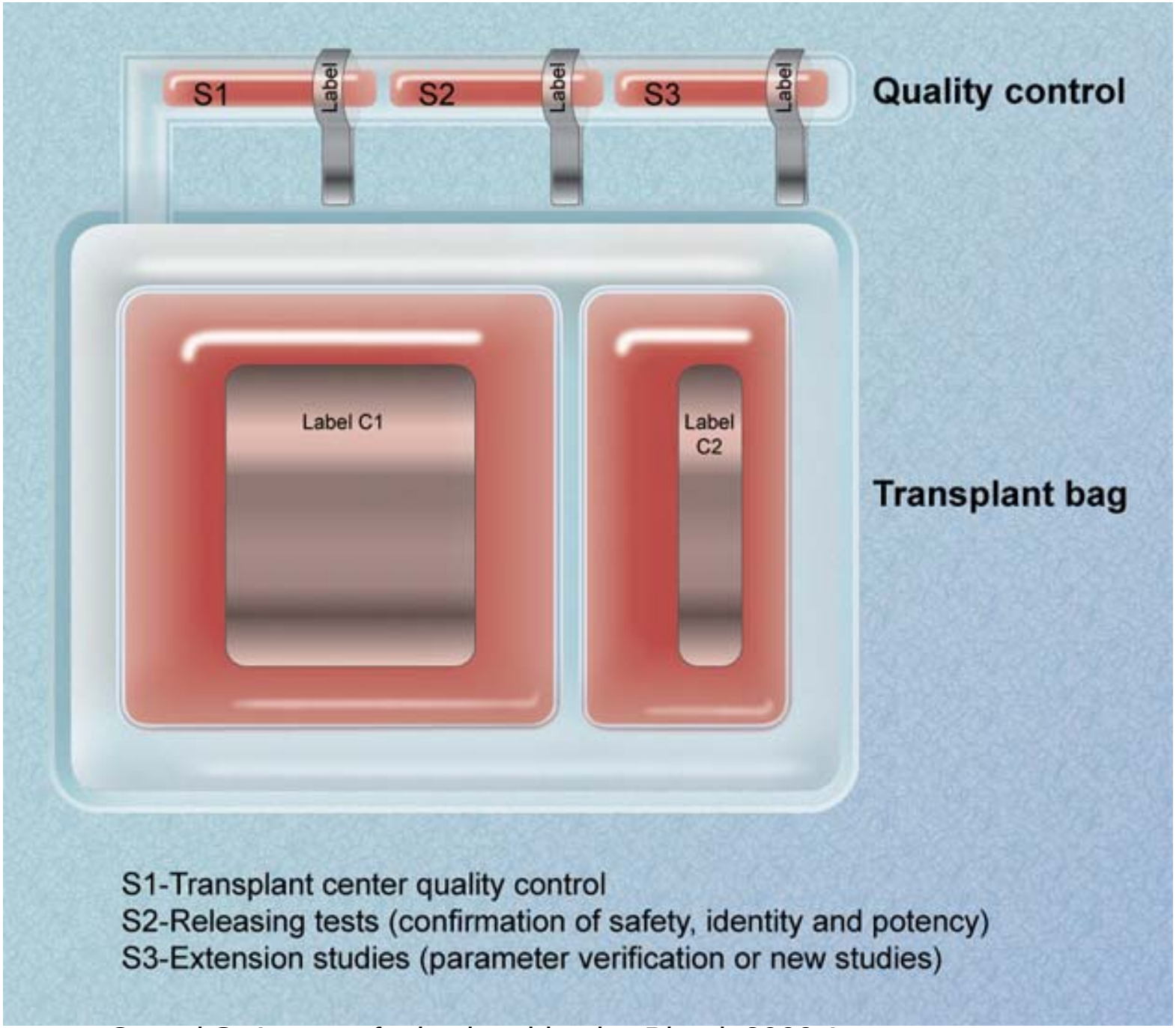
ELIGIBILITY	CONFORMITY	COMPATIBILITY	BATCH RELEASE	FOLLOWUP
INFORMED CONSENT				
TRANSMISIBILITY				
	SAFETY			
	PURITYPOTENCY			
	IDENTITY			
	COMPATIBILITY			
		MATCH		
		DOSE		
			HLA	
			GENDER	
			GROUP	
			MATERNAL HLA	
				ENGRAFTMENT
				GVHD
				RELAPSE
				E-DFS/OS

Qualified CBU

Table 2. Required and recommended tests and tests results according US FDA for cord blood and HPC-C (final cord blood product) (reference 45)

Product Characteristics	Testing	Sample (Type and Timing)	Results of Product Testing
Safety	Infectious diseases – Testing Required (21 CFR 1271.45 through 1271.90)	Maternal peripheral blood obtained within 7 days of cord blood collection – Type and Timing Required. (21 CFR 1271.80(a) and (b))	All tests negative except non-treponemal test for syphilis when confirmatory test is negative. (Cytomegalovirus (CMV) results are recorded). CMV-report
	Sterility - Bacterial and fungal cultures – Testing Required. (21 CFR 211.165(b), and 21 CFR 610.12)	HPC-C *(pre-cryopreservation)	No growth
	Hemoglobin	Cord blood ** or appropriate donor sample obtained at time of cord blood recovery	No homozygous Hemoglobinopathy
Purity and Potency	Total nucleated cells (TNC)	HPC-C (pre-cryopreservation)	≥ 5.0×10^8 TNC ***/ unit HPC-C
	Viable nucleated cells	HPC-C (pre-cryopreservation)	≥ 85% viable nucleated cells
	Viable CD34+ cells (flow cytometry)	HPC-C (pre-cryopreservation)	≥ 1.25×10^6 viable CD34+ cells ****/ unit HPC-C
Identity	Human leukocyte antigen (HLA) Typing	Cord blood	Report
	Confirmatory HLA typing	Attached segment of HPC-C	Confirms initial typing
	Blood Group and Rh Type	Cord blood	Report

*Sample may be obtained before or after addition of the cryoprotectant.
**Cord blood = cord blood before undergoing volume reduction.
***Based on 20 kg recipient, a target dose of ≥ 2.5×10^7 nucleated cells/kg and 70% post-thaw recovery = 1.7×10^7 nucleated cells/kg.
****Based on CD34+ cells ≥ 0.25% of TNC prior to freezing.

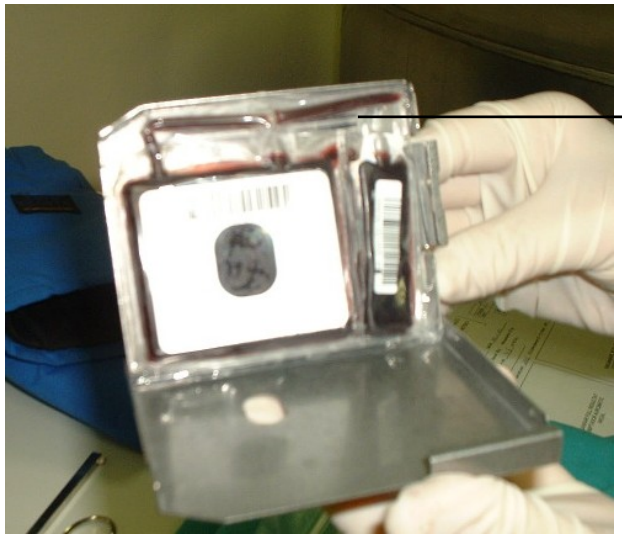


Querol S. A case of mistaken identity. Blood. 2009 Aug

Releasing tests

QUALITY:

- Safety
- Identity
- Potency



What to do:

- Built quality in front
- Verify: releasing tests

Safety:

-*Serology on cord blood*

Identity:

-HLA

-Blood group and gender

- *Maternal haplotype*

Potency:

-CFU/CD34/CLONE

-Viability

-NC recovery

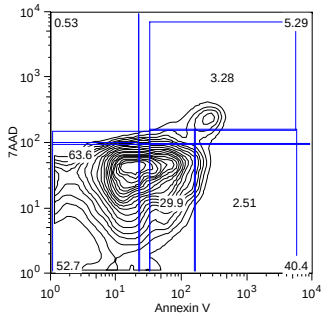
-Volume

TESTING: Functional flow cytometry

Frozen cords can have a high degree of variability

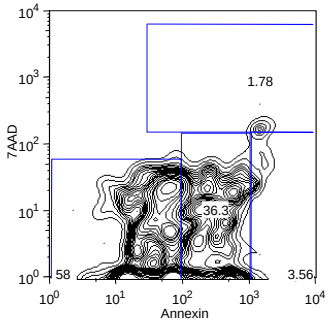
Transplanted cord

successful engraftment



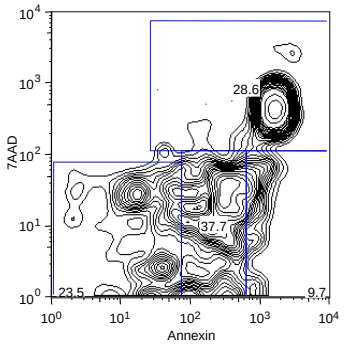
An 100012

mid viability frozen cord



An 100147

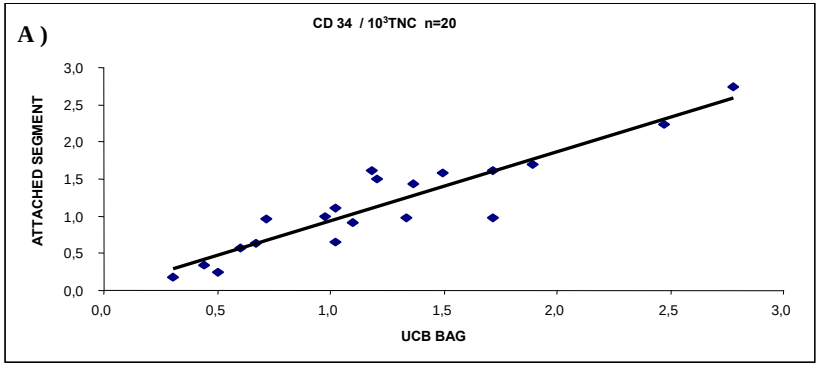
Low viability frozen cord



Assessment with Annexin V and 7AAD of the ISHAGE gated stem cells reveals that there can be a significant number of apoptotic cells present post thaw of cord units.

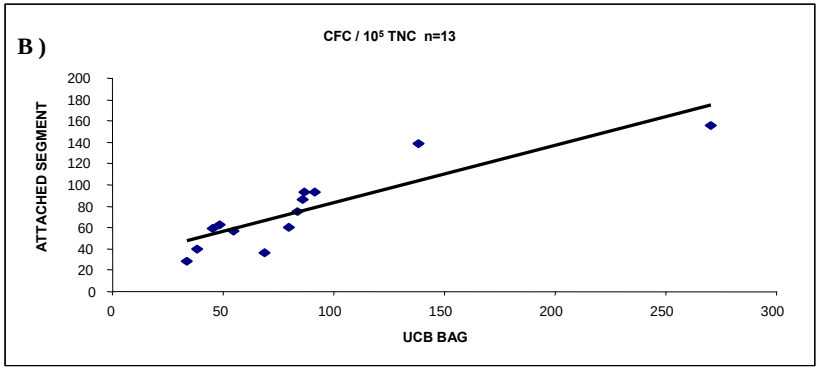
PREDICTIVE UTILITY OF THE ATTACHED SEGMENT IN THE QUALITY CONTROL OF A CORD BLOOD GRAFT.

CD34



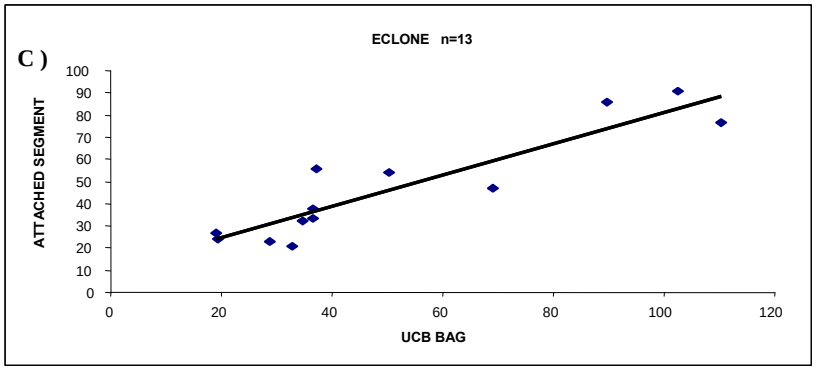
Y = 0,930 x + 0,008. R^2=0,85

CFU



Y = 0,540 x + 29,130. R^2=0,78

CLONE

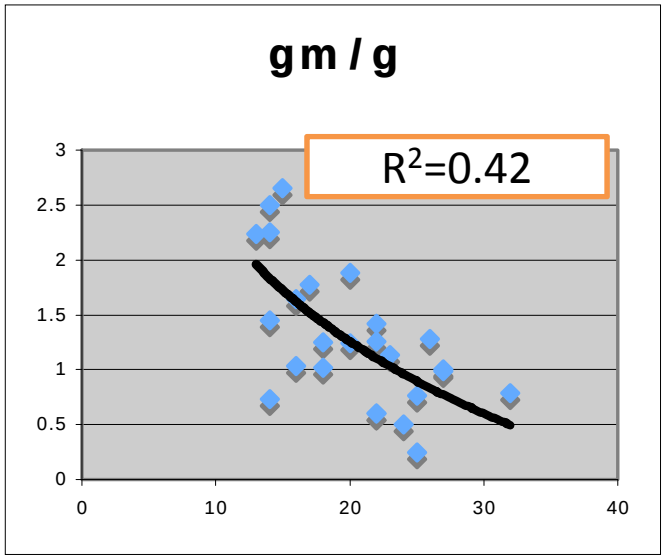


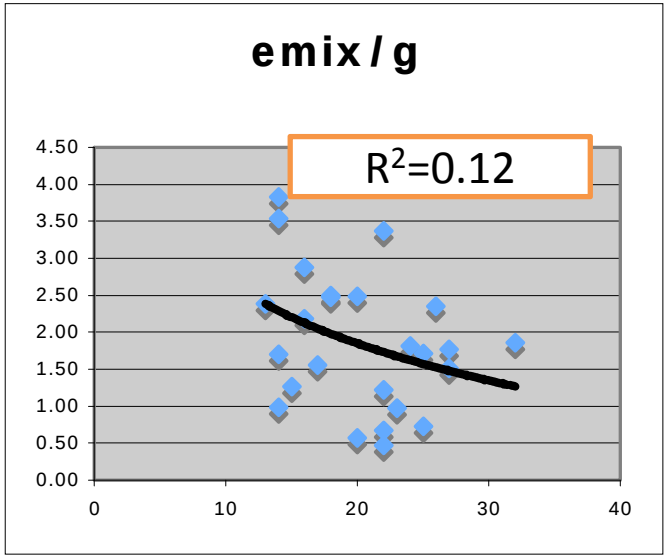
Y = 0,711x + 10,31. R^2=0,83

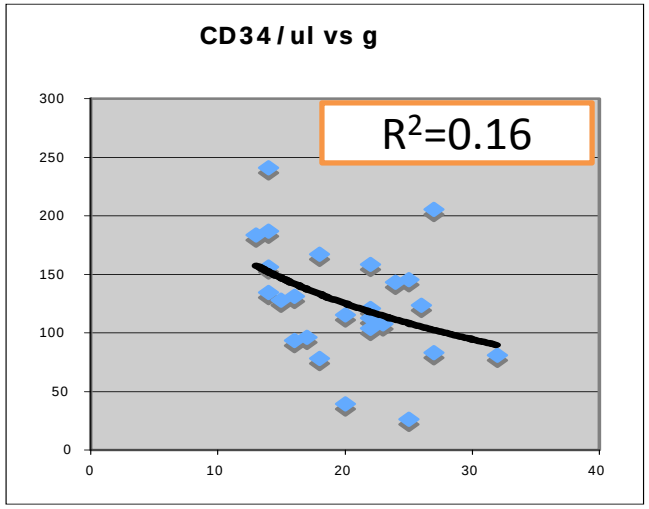
Rodriguez L, Garcia J and Querol S. Biol Biol Blood Marrow Transpl 2005

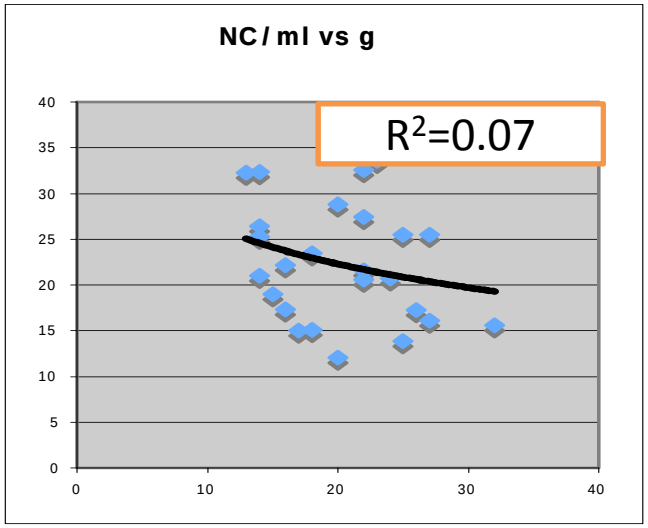
Analysis of 37 pairs Barcelona CBB units-H.La Fe-Valencia TC

CFUGM is the most predictive factor for granulocyte recovery









QMS: continous improvement:
i.e. Bag issues

i.e. Transport issues

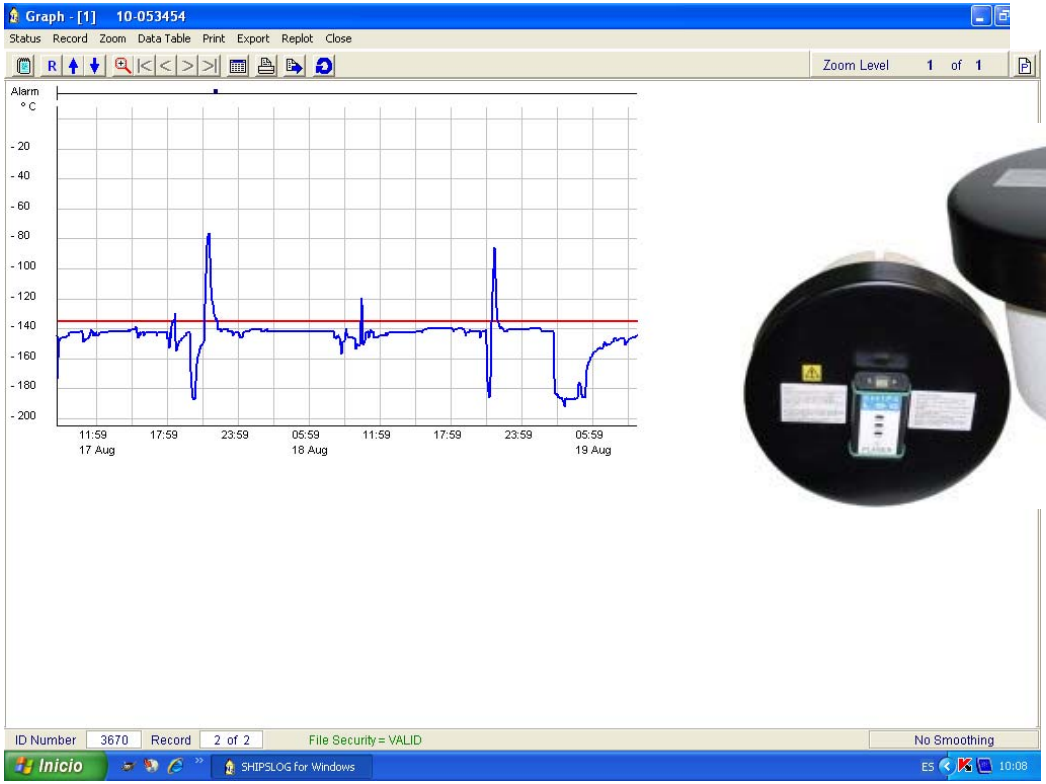
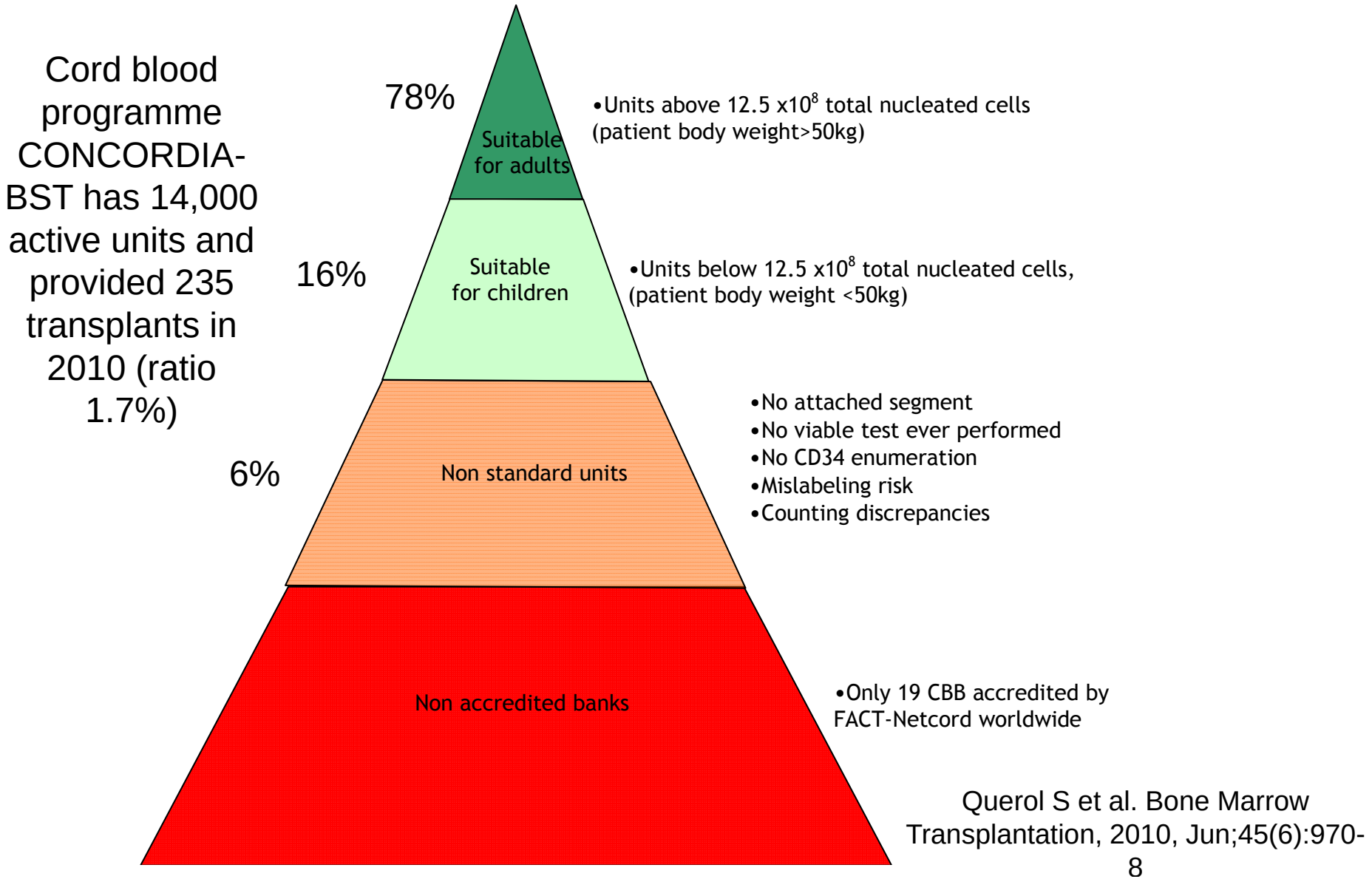


Figure 1. Figurative representation of the quality of the international cord blood inventory (400,000 units from 107 cord blood banks according www.bmdw.org) and suitability for adult and paediatric transplantation based on accreditation, administrative and quantitative factors – The Iceberg effect

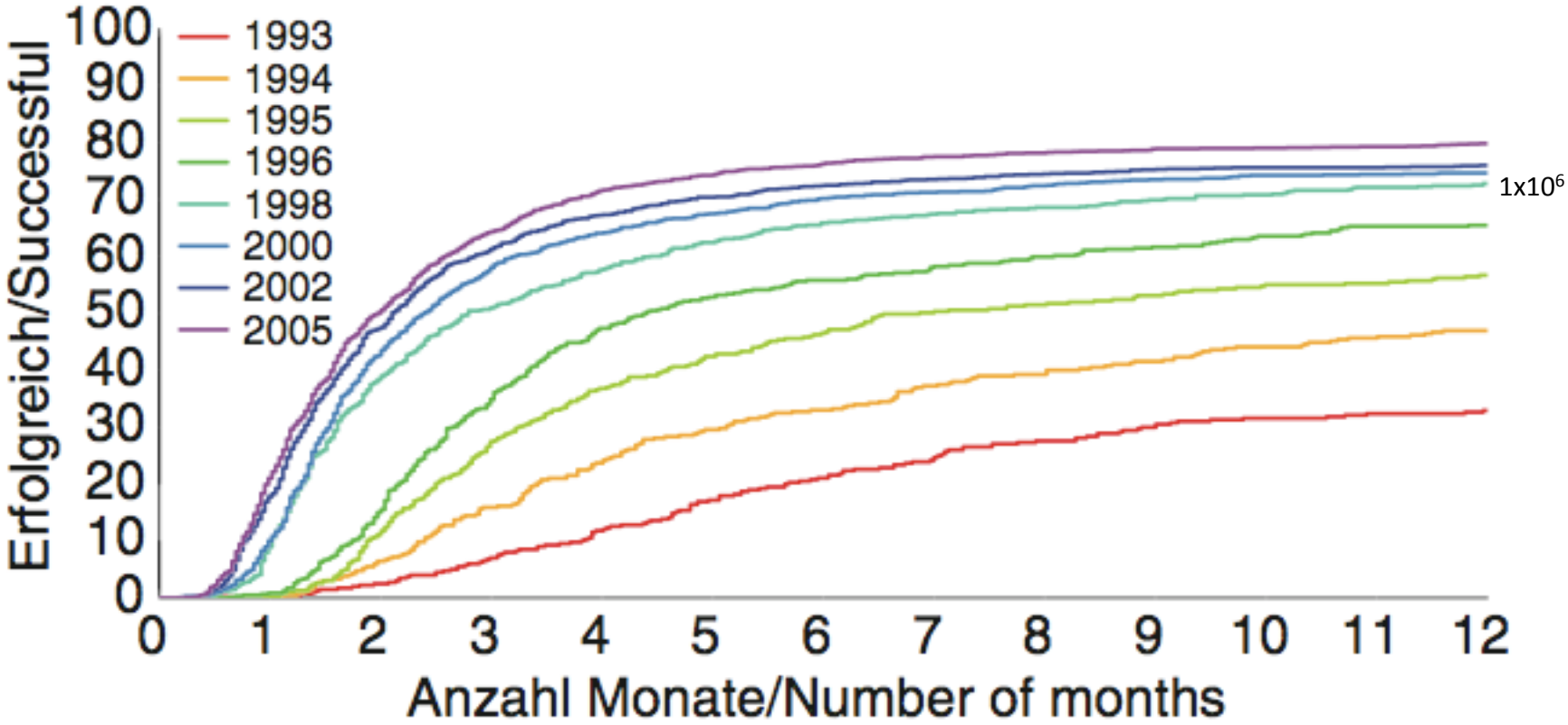


Querol S et al. Bone Marrow Transplantation, 2010, Jun;45(6):970-8

Why national programmes?

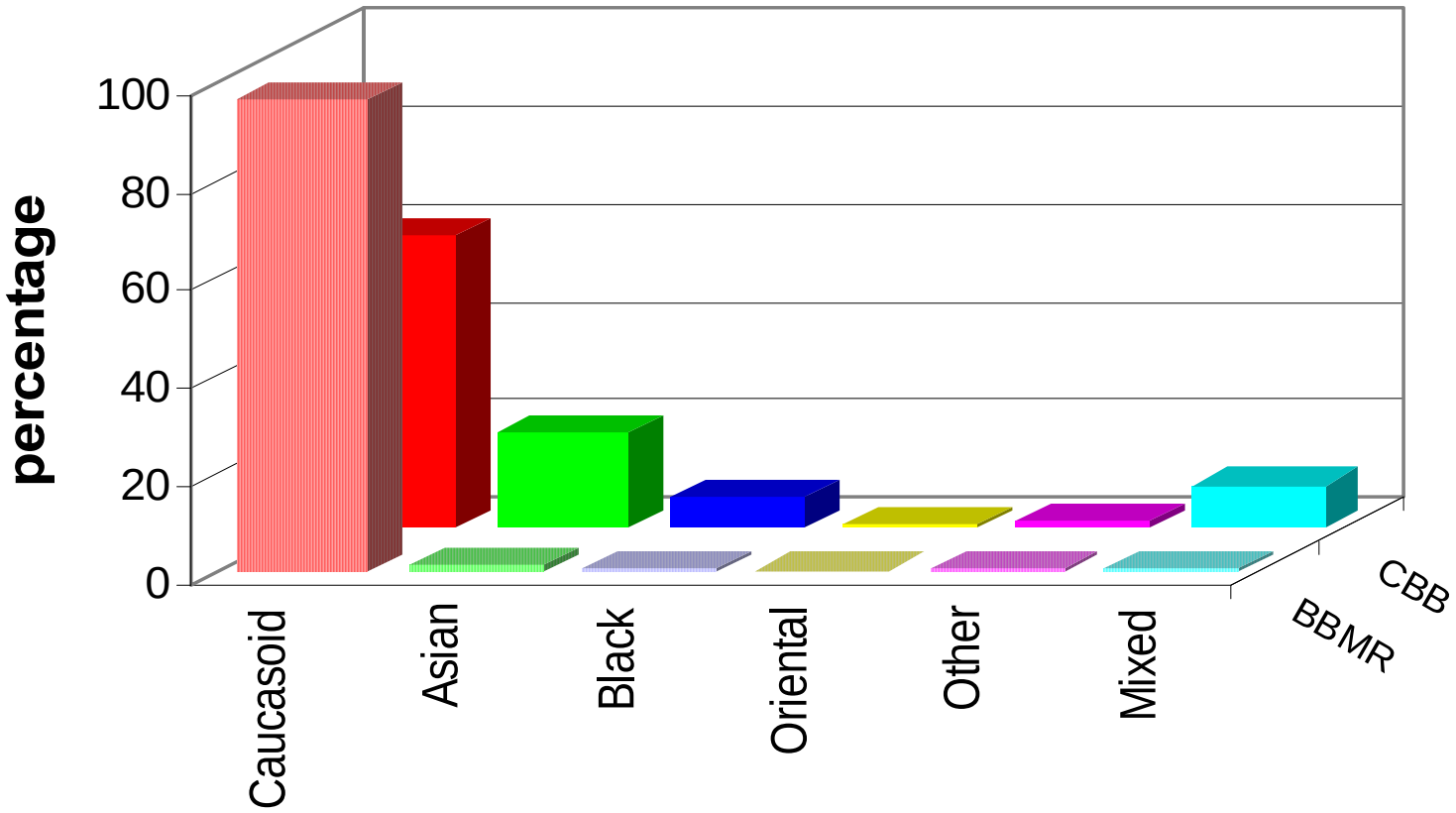
- Meeting National Regulations
- Pre-defined Quality
- Easy Logistics
- Better Feedback
- R&D using CB surplus
- Economic control (auto-sufficiency)

LIMITATIONS OF THE ADULT BONE MARROW REGISTERS: ACCESS AND TIME



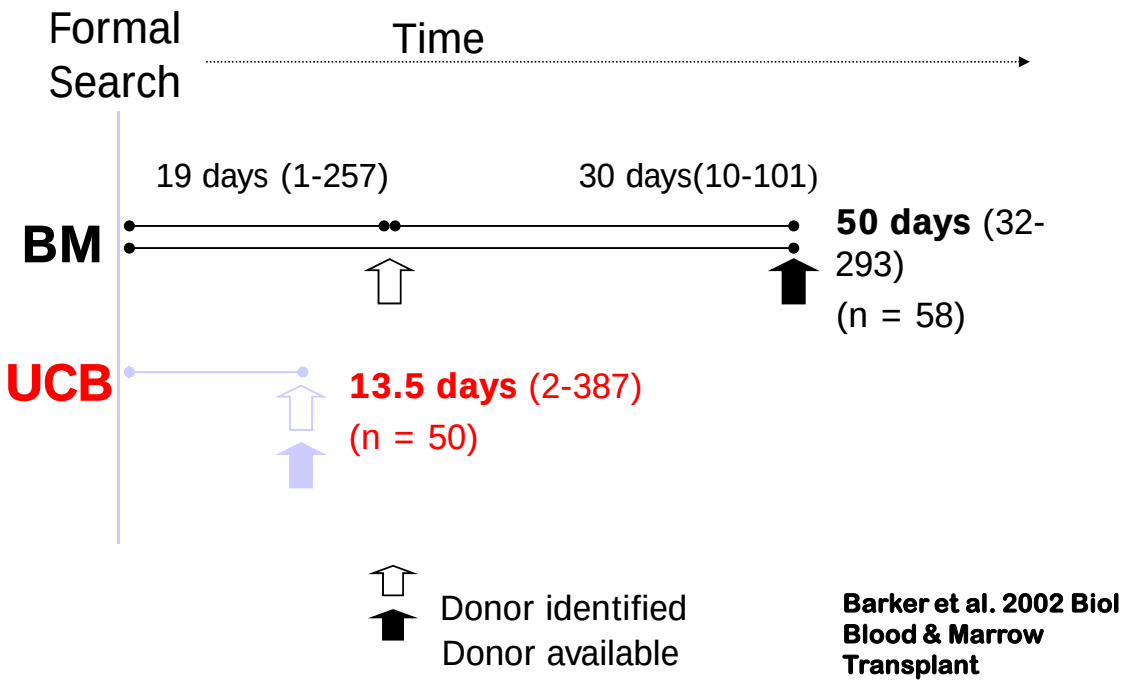
Querol S, Rubinstein P, Marsh SG, Goldman J, Madrigal JA. Cord blood banking:'providing cord blood banking for a nation'. Br J Haematol 2009;147(2):227-35

Cord blood diversity compared to an adult registry: the NHSBT experience



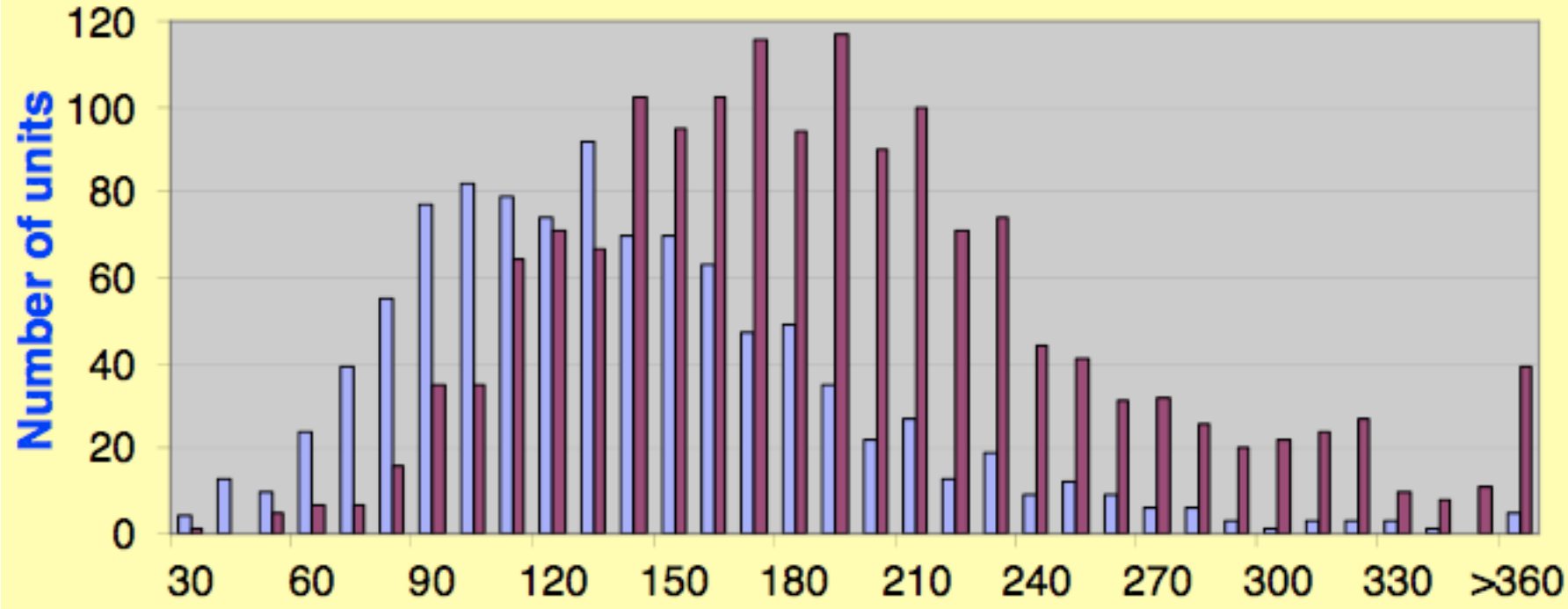
Courtesy Prof Navarrete, NHSBT

Rapid Availability



Barker et al. 2002 Biol Blood & Marrow Transplant

Figure 14: Total Nucleated Cell (TNC) counts of the cord blood units provided for transplantation



TNC median unit children: 136x10E7
TNC average unit children: 143x10E7
TNC median unit adult: 196x10E7
TNC average unit adult: 196x10E7

Table 5 Cell dose of units selected for transplantation over time within the Barcelona Cord Blood Programme

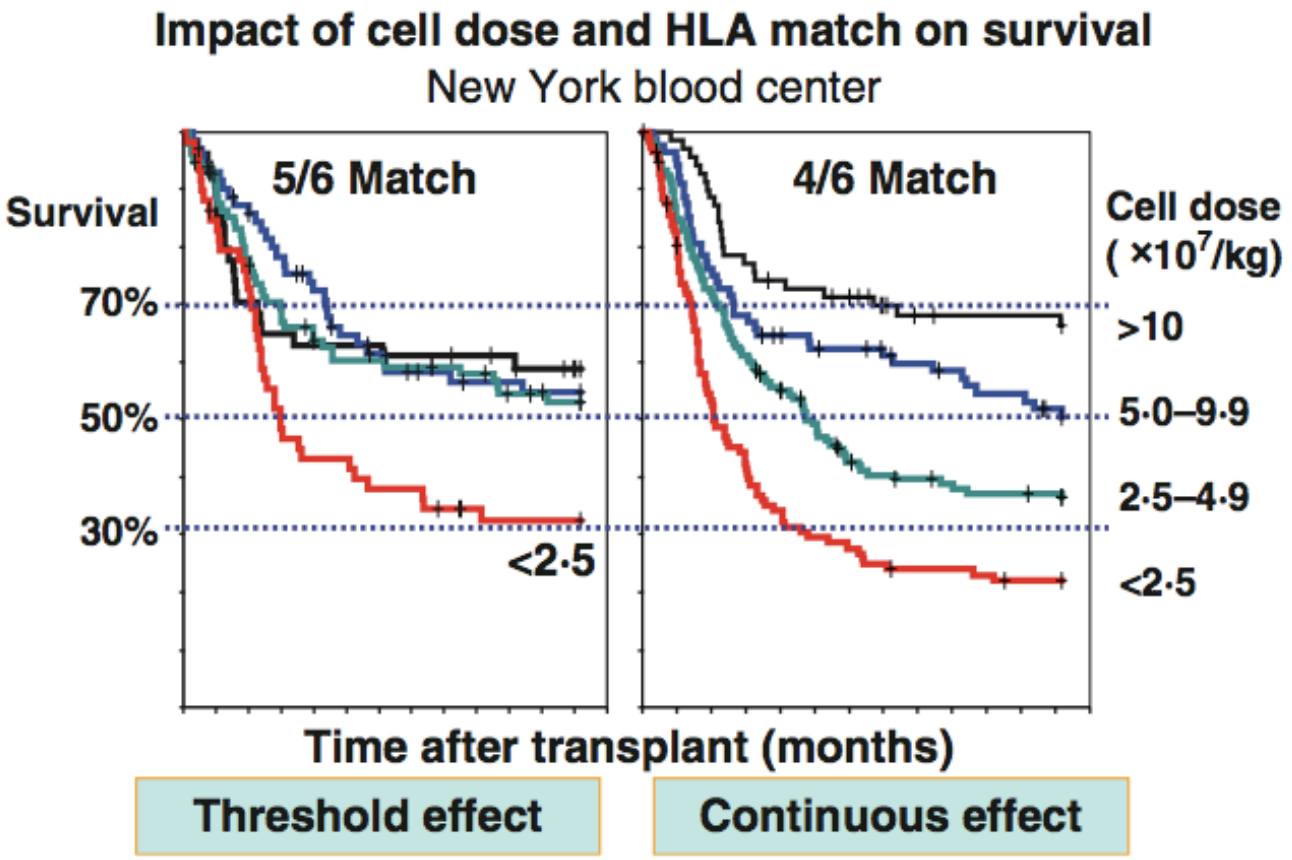
Year	*Number of units provided*	*Total nucleated cells (median, 10^8)*	*Total CD34+ cells (median, 10^6)*
1996–1998	26	11	3.8
1999	26	14	4.3
2000	17	18	5.2
2001	25	17	4.4
2002	17	15	6.0
2003	21	18	6.1
2004	36	15	6.2
2005	44	16	6.1
2006	52	17	6.8
2007	68	17	6.8
2008	94	17	7.5
2009 until October	114	21	9.3

Querol S et al. Bone Marrow Transplantation (2010) 45, 970–978

CBU categories

Category	NCx10e7 (CD34 x10e5)	Concordia	Tx 2009
A (15%) 65kg*	162 (65)	9%	50%
B1 (20%) 50kg (1.5=83Kg)	125 (32)	28%	28%
B (40%) 20Kg	50 (12)	52%	16%
C (15%)	Quality issues	12%	6%

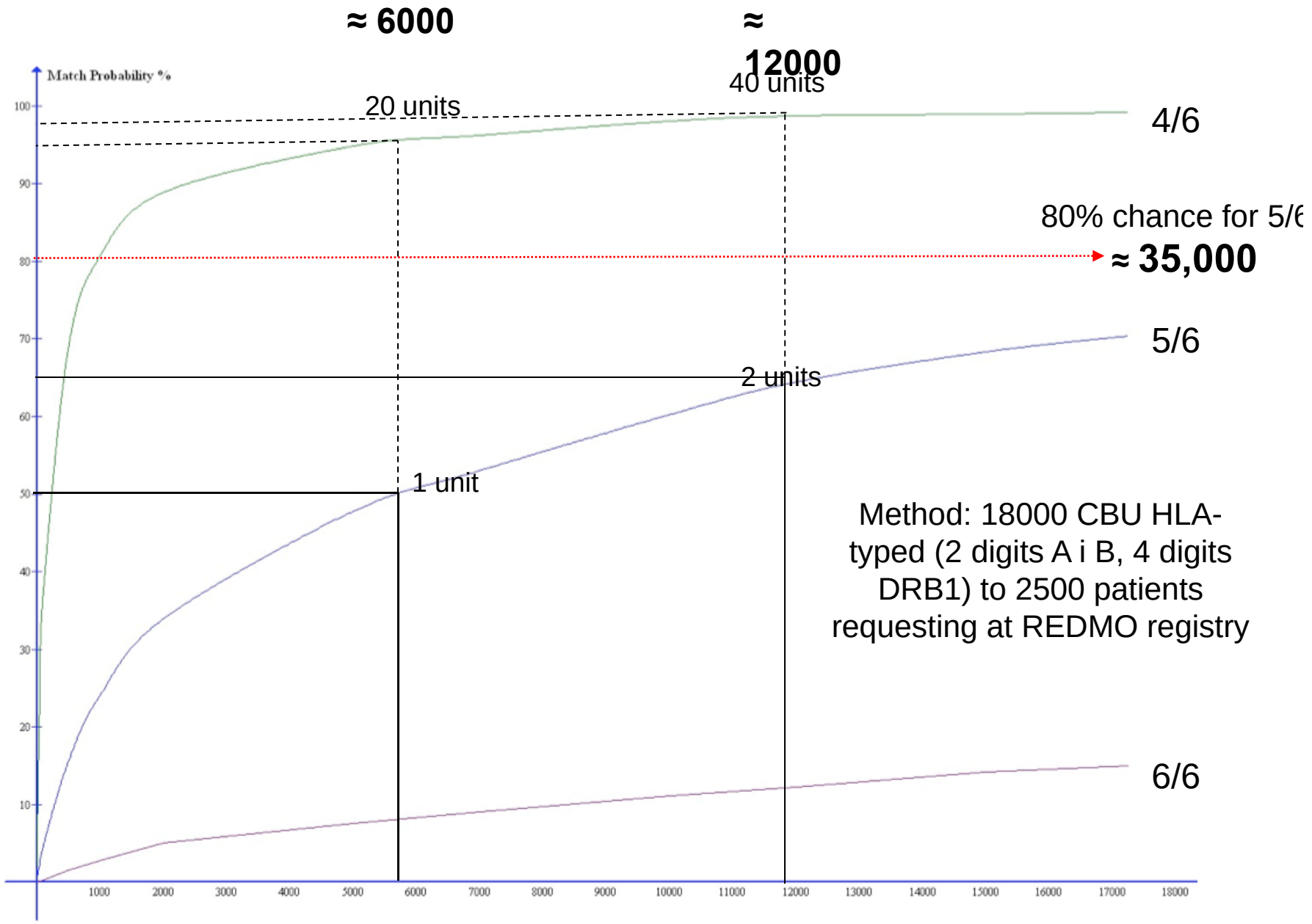
* Median bw of last 2,500 patients requesting unrelated donors in Spain



Assumption for size calculation:

Units 5/6 with more than 2.5 x 10^7 NC/kg will have selection advantage in the future.

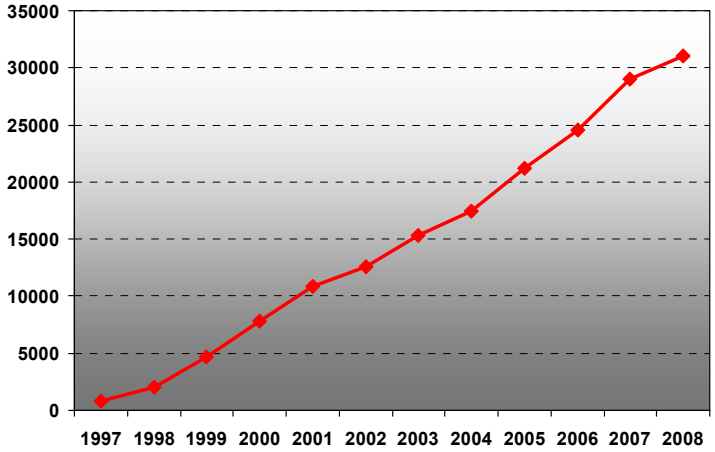
Size for Spain



The Spanish Network:

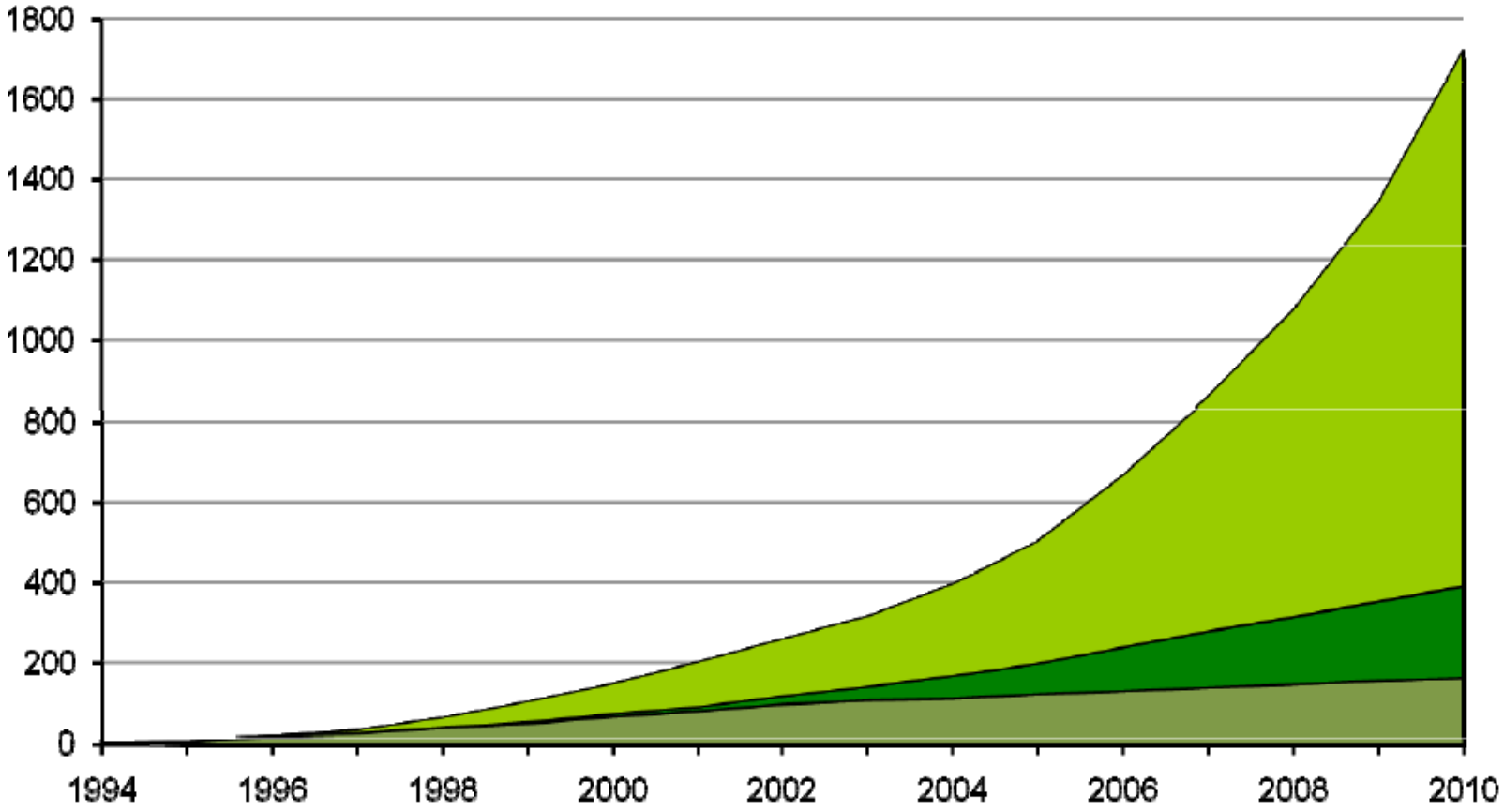
FUNDACIÓ INTERNACIONAL JOSEP CARRERAS PARA LA LUCHA CONTRA LA LEUCEMIA

- Seven regional CBBs and 17 donation programs: **Barcelona** (Catalonia, Balears, Aragon, Navarra, Extremadura, Cantabria), **Madrid** (Madrid, La Rioja**), Malaga** (Andalucia, Castilla La Mancha. Murcia), **Santiago** (Galicia, Castilla-Leon, Asturias), **Tenerife, Valencia, San Sebastian.**



It includes more than **40,000 stored CBU** more than **900 transplanted**.
Barcelona CBB has been one of the NETCORD founders.
Málaga and Santiago CBB are NETCORD members.
They operate in collaboration with the Spanish BMD registry: REDMO

Incidencia acumulada de donaciones españolas

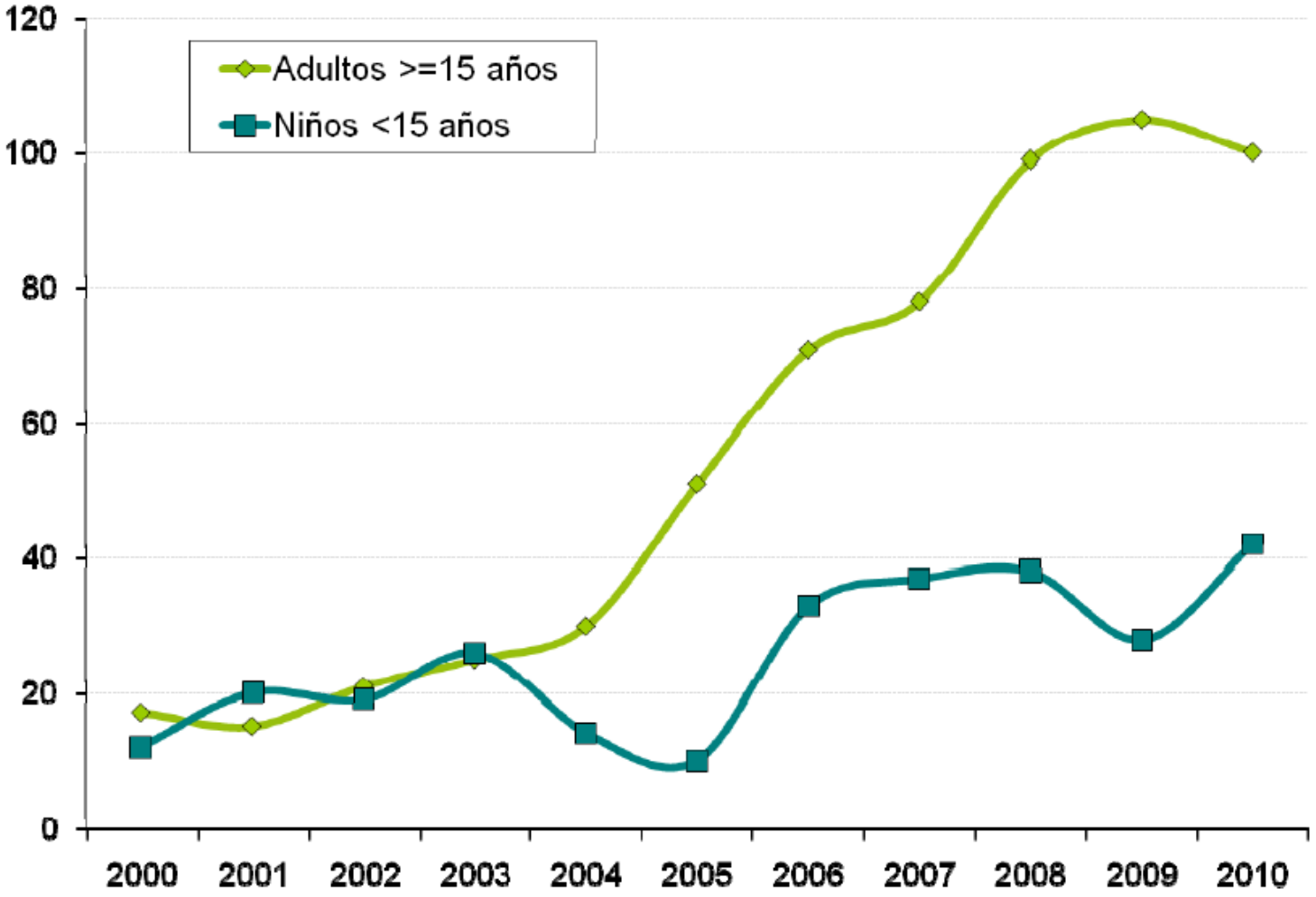


Total donaciones: 1 721

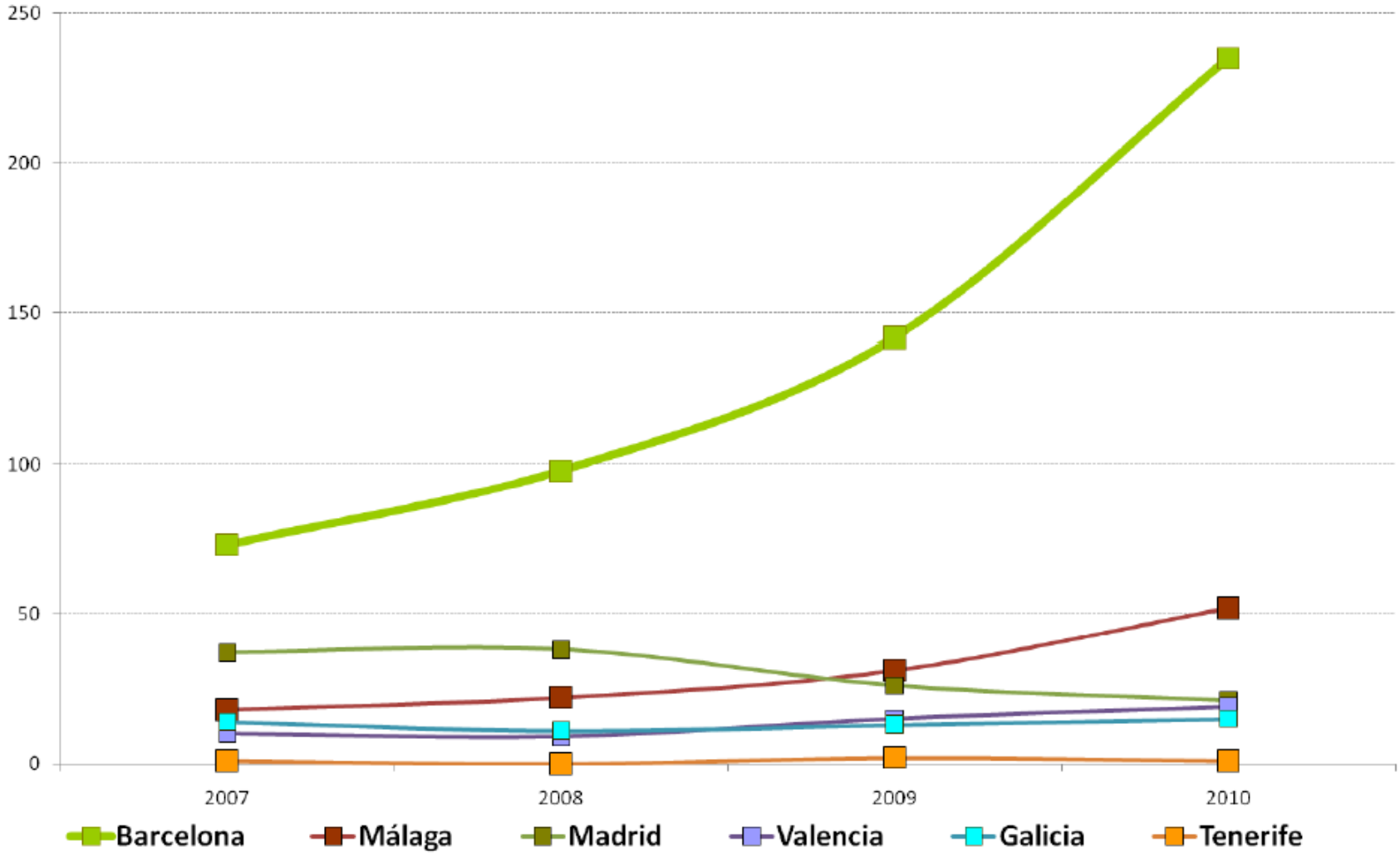
SCU (n= 1 332)[343] SP (n= 230)[32] MO (n= 159)[6]

(total) [2010]

Actividad TPH de SCU España (relación niños / adultos)



Evolución de las unidades SCU suministradas



NETCORD Members Inventory and Use Quarter I 2010

	CB Bank	Inventory	Released for Transplant	Children	Adults
	NETCORD/FACT accredited				
●	Barcelona	13299	679	242	411
●	Besancon	4259	488	140	348
●	Düsseldorf	17075	694	343	311
●	Durham	22889	1147		
●	Helsinki	3039	28	15	13
●	Houston	10551	300	119	181
●	Leiden	3642	80	35	45
●	Liege	2084	125	51	74
●	London	12120	294	151	143
●	Milan	7853	406	209	197
●	New York	50936	3275	1972	1303
●	Pavia	2600	125	49	76
●	Tel Hasomer	1463	18	10	8
	SUM	151810	7659	3336	3110
	not NETCORD/FACT accredited				
●	Athen	1987	7	6	1
●	Bratislava	605	5	1	4
●	Brussels	1335	28	9	19
●	Dresden	2385	8	6	2
●	Dubai	250	0		
●	Firenze	1117	82	36	46
●	Gauting	2810	66	25	41
●	Gent	1810	55	11	24
●	Göteborg	1322	5	1	3
●	Leuven	8542	148	75	73
●	Louvain	1909	101	37	64
●	Málaga	15062	123	44	79
●	Mannheim	1791	71	20	51
●	Mexico City	1719	183	126	57
●	Padova	1509	63	20	43
●	Pescara	413	5	2	2
●	Prague	3310	47	18	29
●	Roma Lazio	1410	58	26	32
●	Santiago de Compostela	5140	56	30	26
●	Tokyo	5646	951	237	714
	SUM	60072	2062	730	1310
	TOTAL	211882	9721	4066	4420

● CBUs in VO

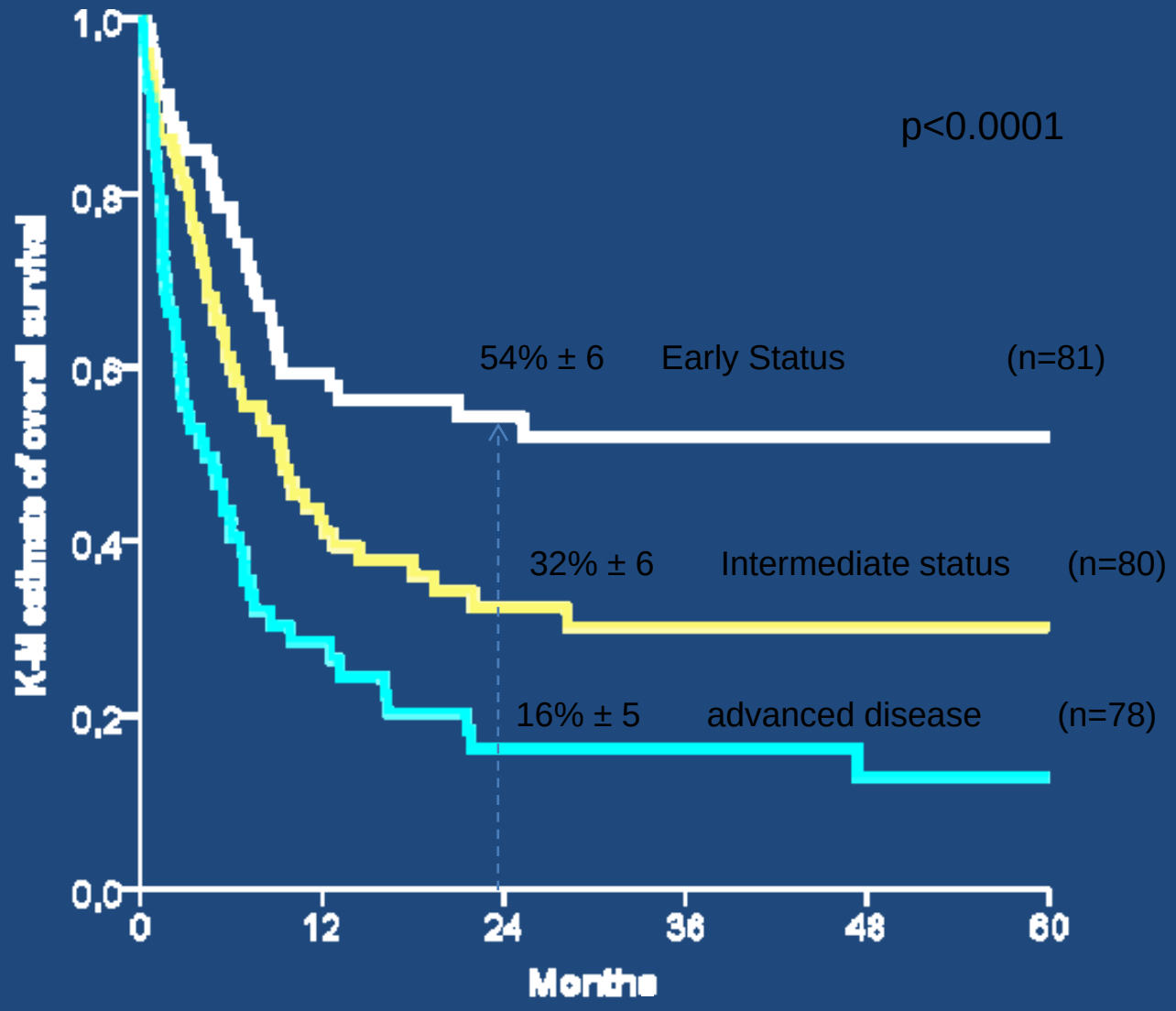
● CBUs NOT in VO

Official NETCORD Collaborators:

Banks in Black have updated recently

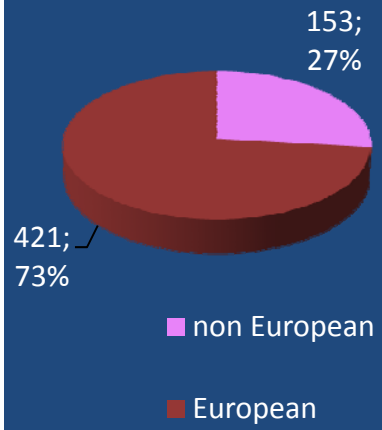
thermogenesis® biosafe

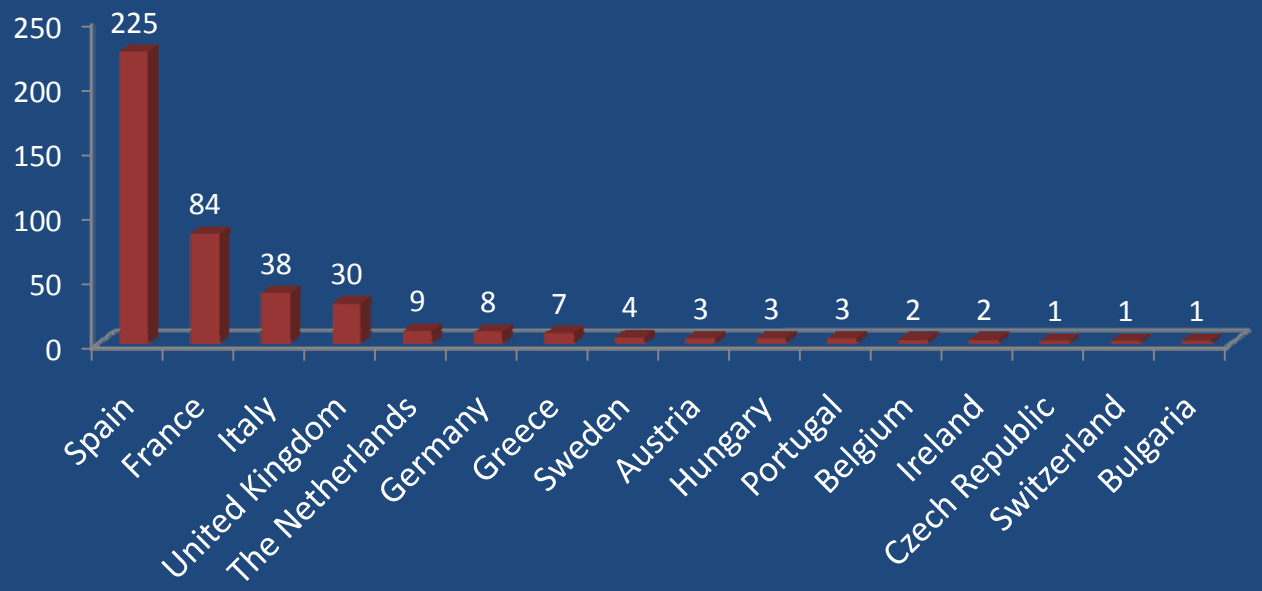
Overall survival by disease status at transplant for patients with malignant disorders n=249

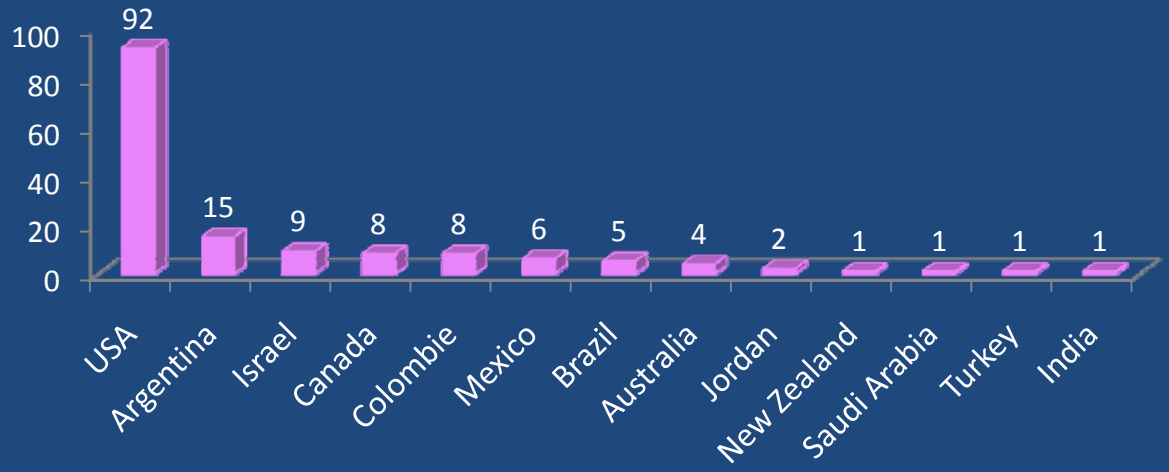


Countries of exportation of the unrelated Barcelona units

Number of UR CB transplants per country (n= 574)







Autologous

- 10-20 AutoTx reported
- AutoTx in leukemia, medulloblastoma (risk of carrying genetic defects)
- Clinical research:
  - DM I
  - Cerebral palsy
- Private banking: issues
  - No possible in Spain unless donor HLA shared with REDMO
  - Informed Consent
  - Quality


Dr Susana G Gomez
Robert Davy
Roger Horton
Laura Fry
Daniel Gibson
Kieran Herrity
Hazel Forde
Richard Duggleby
James Devitt
Aurore Saudemont
Daniel Figueroa-Tentori
Lia Zambetti
Mehri Daryouzeh
Michelle Escobedo
Dunia Jawdat
Prof Tony Dodi and Dr Steve McEwan†
ANTHONY NOLAN
BE A MATCH. SAVE A LIFE
Prof Steven G Marsh
Dr Brownwen Shaw
Dr Sameer Tulpule
Prof John Goldman
Prof Alejandro Madrigal
Terie and all KingsCord donors
Dr Nichola Cooper (ICH)
Prof John Martin (UCL)
Prof Manuel N Fernandez (CPH, Madrid)
Dr Guillermo F Sanz (H La Fe, Valencia)
Prof Robert Rees (NTU, Nottingham)
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Dr Sergio Querol