UK NEQAS
International Quality Expertise

Histocompatibility
& Immunogenetics

Director: *Dr MT Rees*
Deputy Director: *Mrs D Pritchard*
Operations Manager: *Miss A De'Ath*

Tel: *+44 (0) 1443 622185*
Email: *ukneqashandi@wales.nhs.uk*
Web: *www.ukneqashandi.org.uk*

Correspondence to:
UK NEQAS for H&I
Welsh Blood Service
Ely Valley Road
Talbot Green
Pontyclun
CF72 9WB

Interpretive Educational Scheme (iED)
Clinical Scenario 2/2020 – Haematopoietic Stem Cell Transplantation

Dispatched on 20th October 2020

Summary of Results

A total of 49 responses were received, 19 from laboratories based in the UK and Ireland (UK&I) and 30 from Laboratories based in the rest of the world (RoW) participants.

A 49 year old female (ABO: O RhD positive, CMV: negative) weighing 80kg with AML is referred to your laboratory. The patient has five potential related donors: one full sibling and four children. All are sent to the laboratory for HLA genotyping and an unrelated search is initiated.

Patient's HLA type:

	HLA-A*		HLA-B*		HLA-C*		HLA-DRB1*		HLA-DQB1*		HLA-DPB1*	
Patient	02:01	25:01	56:01	57:01	01:02	06:02	04:01	07:01	03:01	03:03	04:01	13:01

1) What aspects of the patient's HLA type make this a challenging unrelated donor search?

Responses Included:
The patient's 2 haplotypes are likely to be derived from 2 different populations making it challenging to find a matched unrelated donor.
Patient has one common European haplotype (A2 B57 Cw6 DR7 DQ9) and a rare Russian/Eastern European haplotype (A25 B56 Cw1 DR4 DQ7). This combination may not be well represented on donor registries.
HLA-A*25 is not common and also not commonly found in association with either HLA-B*56 or HLA-B*57.
Low frequency antigens HLA-A*25:01 and -B*56:01. HLA-A*25 usually seen in haplotype with -B*18 and -C*12
B*56:01 has a lot of HLA-C associations making it harder to find a match when the C type is not stated from the registry.
DRB1*04:01 and DQB1*07:01 have multiple potential DQ associations e.g. DRB1*04:01 is in LD with both DQB1*03:01 and DQB1*03:02. Registry data may lack DQ genotyping.
The HLA-DRB3/4/5 genotype is not available and the provided type suggests this patient has the less common DR7, DR53N, DQ9 association rather than the prevalent DR7, DR53, DQ2 combination.
DRB1*04:01 usually seen 50/50 with either DQB1*03:01 or *03:02, so potential for DQB1*mm
HLA-DRB1*07:01 is present in less common haplotype with DQB1*03:03 (likely including DRB4*01:03:01:02N allele).
HLA-DRB1*04:01 also present with HLA-DQB1*03:02 (almost equal frequencies as DQB1*03:01). DRB1*07:01 can also be present with DQB1*02:02, but in the patients genotype it is part of a frequent haplotype (A*02:01, B*57:01, C*06:02, DRB1*07:01, DQB1*03:03, DRB4*01:03:01:02N) within Caucasian population.
DQB1*03:02 is more commonly associated with the haplotype B*56:01-C*01:02-DRB1*04:01 than DQB1*03:01. Therefore, finding 12/12 donors is unlikely.
HLA-DPB1*13:01 is present in only 3% of local BBMR donors of European origin. (Brown et al.).
Patient has high expression HLA-DPB1*13:01 variant (could be a target if donor is homozygous for low expression DPB1 variants).
No particular concerns. Common B/C and DR/DQ associations.

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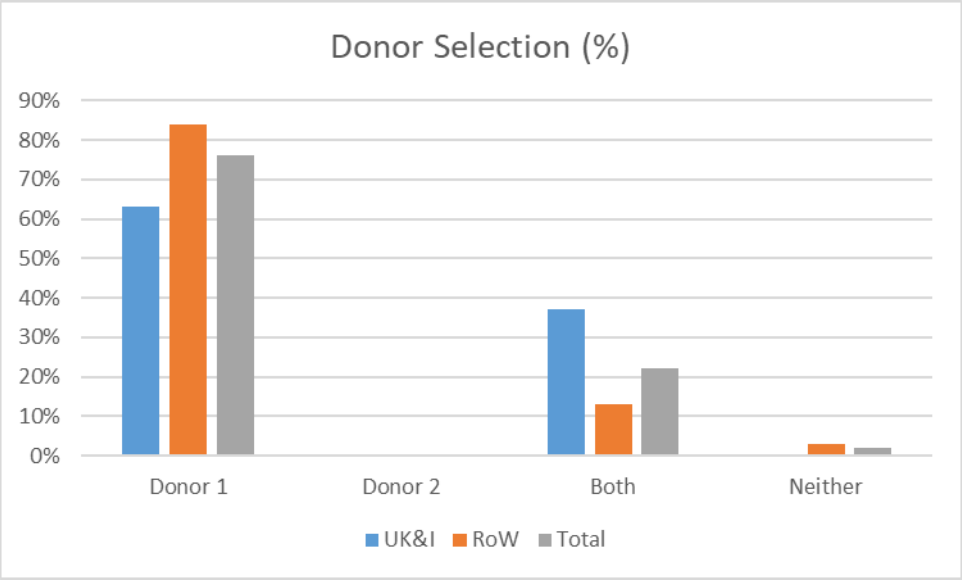
The unrelated donor search revealed only two potential fully matched donors:

Donor	Registry	M/F	Age	ABO	CMV	HLA-A*	HLA-B*	HLA-C*	HLA-DRB1*	HLA-DQB1*	HLA-DPB1*
1	DE-ZKRD	M	21y	AB+	N	02:01; 25:01	56:01; 57:01	01:02; 06:02	04:01; 07:01/07:79	03:01; 03:03	04:01; 13:01
2	BR-REDOME	M	59y			02; 25	56; 57		04; 07		

2) Would you pursue either donor listed? Provide a reason for your answer.

	UK&I	Percentage (n=19)	RoW	Percentage (n=30)	Total	Percentage (n=49)
Donor 1	12	63%	25	84%	37	76%
Donor 2	0	0%	0	0%	0	0%
Both	7	37%	4	13%	11	22%
Neither	0	0%	1	3%	1	2%



Reasons for Making Selection	
Donor 1	Young (<30) male donor.
	Full 10/10 match.
	Possibility of 12/12 match grade to 2nd field (pending confirming ambiguity on DR).
	CMV matched.
	Major ABO mismatch.
	Need to confirm the ABO antibody titre status of the patient before proceeding.
	Donor is from a reliable and rapid Registry, especially important in patients with progressive diseases like AML.

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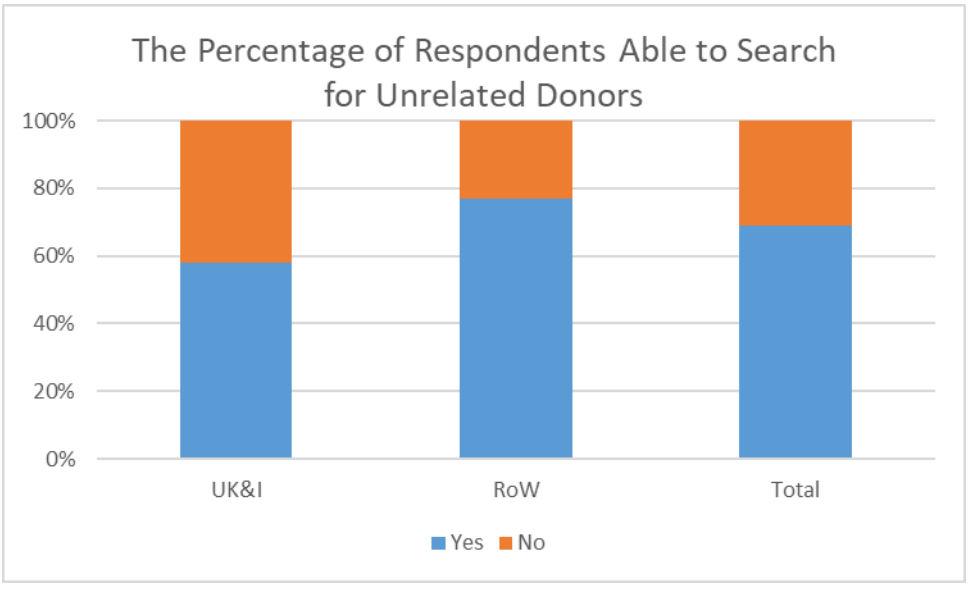
Donor 2	Donor's age (59) is not desirable. Potential co-morbidities.
	HLA typing incomplete (HLA-C & DQ not typed) / low resolution.
	Potential mismatch.
	Brazilian Registry is not very responsive. Risk of Zika virus.
	CMV status unknown.
Both Donors	Donor 1 preferable. Donor 2 backup.
	We would request both as there are only two options available.
	Selecting both will provide a choice for the clinician between old donor or ABO incompatible. Depends also on urgency.
	As the patient has a rare HLA type we would test both donors.
Neither	Wait for the results of HLA typing of full sibling before pursuing an unrelated donor.

3) If you are able, run the patient on a search programme. Are there any potential donor options that may be recommended to the transplant consultant? If yes, give details and the reasons for your selection of your two most preferred donors?

Are you able to run a search request?

	UK&I	Percentage (n=19)	RoW	Percentage (n=30)	Total	Percentage (n=49)
Yes	11	58%	23	77%	34	69%
No	8	42%	7	23%	15	31%



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Most Common Results of Search	
First Preference	**6939DKM0012331311817 - 11/12, DPB1 matched, CMV and ABO matched 22 year old male with an allelic DQB1*03 mismatch with no DSA.**
	1 A-MM (GvH), DPB1 permissive, male, 26yrs
	9/10 in GvH direction (donors homozygote A*02:01) with permissive DPB1*.
	No potential fully matched donors but we would consider a 9/10 match, preferably at HLA-A or HLA-DQ.
	6/6 with no mismatch in A, B and C low resolution loci, but the rest of the genotyping is unknown and the donor is a women of 51y with a CMV+ status.
	9/10 matches one 27y old male with a DQB1mm (03:02 vs 03:01).
	12/12 match, male, age 21y, CMV negative.
Second Preference	**9/10 (10/12 HLA-DPB1 permissive) HLA-A mismatch; CMV Negative (last tested 2019); ABO blood group mismatch; Male 28.**
	Female, 25, CMV Neg, ZKRD, HLA 9/10 DRB1 mismatch (DRB1*11:01), DP permissive
	1 A mismatched (bidirectional), 1 DPB1 mismatched (GvH) German donor. Male, 28yrs. CMV negative (matched). However, ABO major mismatch.
	9/10 A mm, permissive DP mm, young male with recent CMV Neg status
	Mismatch at HLA-A, CMV negative, young male donor
	9/10, permissive DPB1 mismatched, ABO and CMV matched 25 year old female with a DR mismatch with no DSA.
	Male donor 5/6 with no mismatch in A and B loci and one mismatch in C, but the rest of the genotyping is unknown as well as the CMV status and the blood group
	26y old male donor with a HLA_A mm(direction of the Mm gvH, homozygous for -A02:01)
	Female 36 years with 1 non permissive DPB1 mismatch (HvG) and with potential pregnancy. Blood group and CMV unknown.
	1 A-MM, DPB1 permissive, female, 24yrs.
Other Comments	**WMDR search did not return any 10/10 matched donors.**
	The majority of the donors were 9/10 or 8/10 match with a mismatch on the HLA-A locus was the most common.
	No suitable donors.
	found in BMDW but at least four 8/8 with DQ8 instead of DQ7
	recommend either haplo-identical donor or 9/10 donor with HLA-A mismatch, preferred one unidirectional mismatch

*Note: Most common response highlighted in **bold** text*

Both donors were unfortunately deleted from the registry so a cord search was carried out:

Donor	Cord bank	HLA-A*	HLA-B*	HLA-C*	HLA-DRB1*	HLA-DQB1*	TNC (x10^7)	CD34 (x10^5)	Vol (ml)	Blood group	AABB/FACT accredited
1	SE - Cord	02; 02	44; 57		04:01; 07:01		314	90	25	B RhD pos	Yes
2	RU - Samara	02:06/10; 25	56; 57		07:01; 14:03		236	79	23	A RhD pos	No
3	ES - Malaga	01; 02	56:01; 57		01; 07		234	117	162	A RhD pos	Yes
4	BE - Leuven	02; 68	56:01; 57		01:01; 07:01		173	48	25		Yes
5	US-Durham	02; 25	18:01; 57		04:01; 07:01		151	63	25	A RhD pos	Yes

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4) Which, if any, of these cord units would you recommend to the Transplant Consultant and provide a reason for your decision?

	UK&I	%	RoW	%	Total	%	Summary of Reasons for Selection
Donor 1	8	28%	14	39%	17	28%	• Accredited cord bank • Potential 4/6 match • 5/8 in GvH. 6/8 in HvG direction • DRB1 match • High TNC dose • Relatively high CD34 count • Low volume • Suitable for single unit cord transplant
Donor 2	2	7%	1	3%	3	5%	• No DSA • Potential 6/8 • A802:06 an unacceptable mismatch • DR mismatch • Not accredited • Suitable for single unit cord transplant
Donor 3	4	14%	5	14%	9	15%	• 6/8 • DR mismatch • Volume too high (likely RBCs) • Suitable for single unit cord transplant
Donor 4	1	3%	1	3%	2	3%	• 6/8 • Low CD34 count • Low TNC dose
Donor 5	4	14%	3	8%	7	12%	• Accredited cord bank • Potential 5/6 match • DRB1 match • Low volume • Low CD34 count • Low TNC dose • US based units often expensive
Donors 1+5	5	17%	6	16%	11	18%	• Double unit required due to patient weight • DRB1 matched • Good combined TNC and CD34 dose
Donors 1+2	1	3%	0	0%	1	2%	• High TNC dose • High CD34 count
Donors 2+3	0	0%	1	3%	1	2%	• Good cell dose
None	4	14%	5	14%	9	15%	• All units have HLA matching grade of 4/6 to 5/6 • Units do not provide the minimum recommended dose of TNC or CD34 for the adult patient in this case • Require further typing of the units • HLA antibody testing of the patient required • Haploidentical donor preferable

Note: Some labs chose multiple donors

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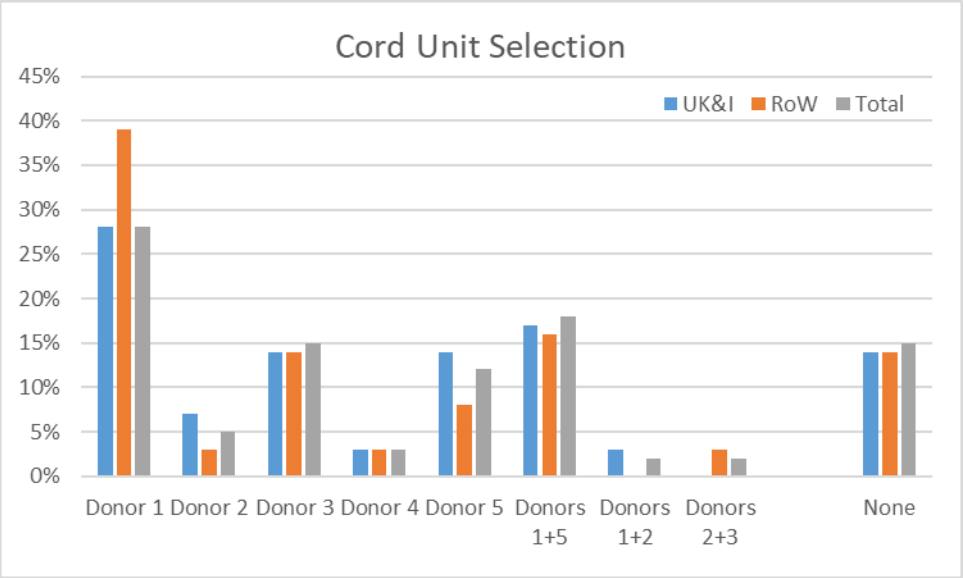
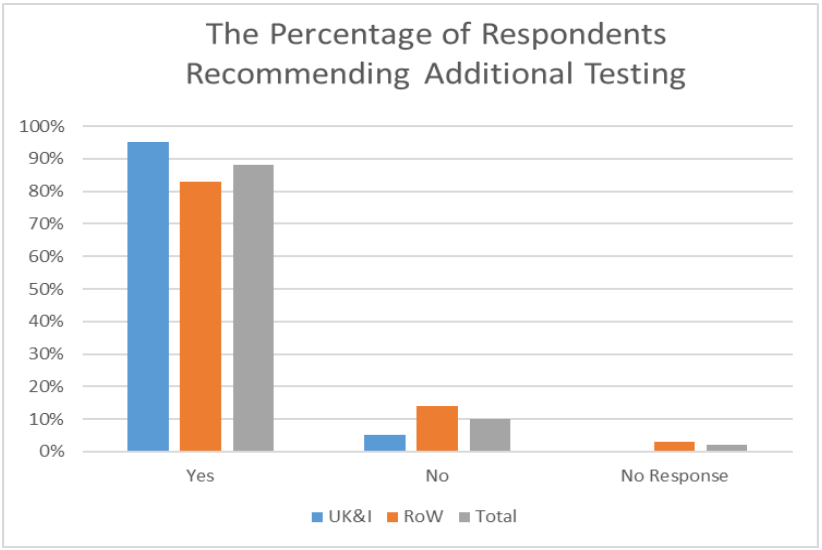


5) Would you recommend any additional testing of these cord units?

	UK&I	Percentage (n=19)	RoW	Percentage (n=30)	Total	Percentage (n=49)
Yes	18	95%	25	83%	43	88%
No	1	5%	4	14%	5	10%
No Response	0	0%	1	3%	1	2%



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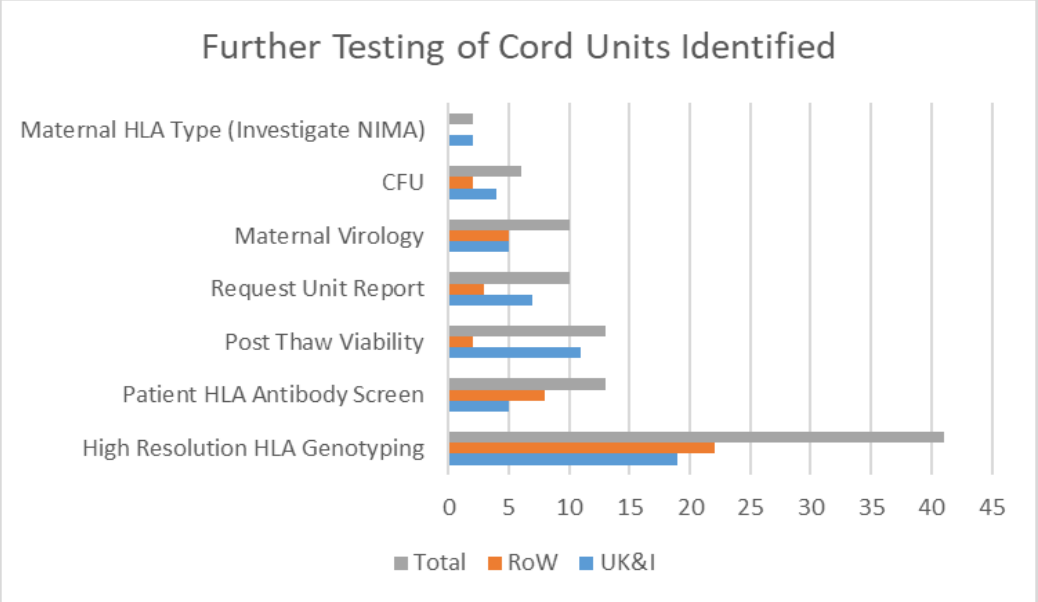
Testing Identified	UK&I	RoW	Total
High Resolution HLA Genotyping	19 (36%)	22 (52%)	**41 (40%)**
Patient HLA Antibody Screen	5 (9%)	8 (19%)	**13 (14%)**
Post Thaw Viability	11 (21%)	2 (5%)	**13 (14%)**
Request Unit Report	7 (13%)	3 (7%)	**10 (11%)**
Maternal Virology	5 (9%)	5 (12%)	**10 (11%)**
CFU	4 (8%)	2 (5%)	**6 (6%)**
Maternal HLA Type (Investigate NIMA)	2 (4%)	0	**2 (2%)**



The Transplant Consultant decides not to use a MUD or cord; a haploidentical transplant is considered next. HLA typing of suitable family members is provided along with HLA Class I antibody screening results for the patient (Class II HLA antibody negative).

	HLA-A*		HLA-B*		HLA-C*		HLA-DRB1*		HLA-DQB1*		HLA-DPB1*	
Patient	02:01	25:01	56:01	57:01	01:02	06:02	04:01	07:01	03:01	03:03	04:01	13:01
	HLA-A*		**HLA-B***		**HLA-C***		**HLA-DRB1***		**HLA-DQB1***		**HLA-DPB1***	
Sibling	02:01	#	56:01	58:01	01:02	03:02	04:01	13:02	03:01	06:09	04:01	104:01
Child 1	02:01	24:02	13:02	56:01	01:02	06:02	04:01	10:01	03:01	05:01	04:01	#
Child 2	02:01	24:02	27:05	56:01	01:02	02:02	01:01	04:01	03:01	05:01	02:01	04:01
Child 3	24:02	25:01	13:02	57:01	06:02	#	07:01	10:01	03:03	05:01	04:01	13:01
Child 4	24:02	25:01	27:05	57:01	02:02	06:02	01:01	07:01	03:03	05:01	02:01	13:01

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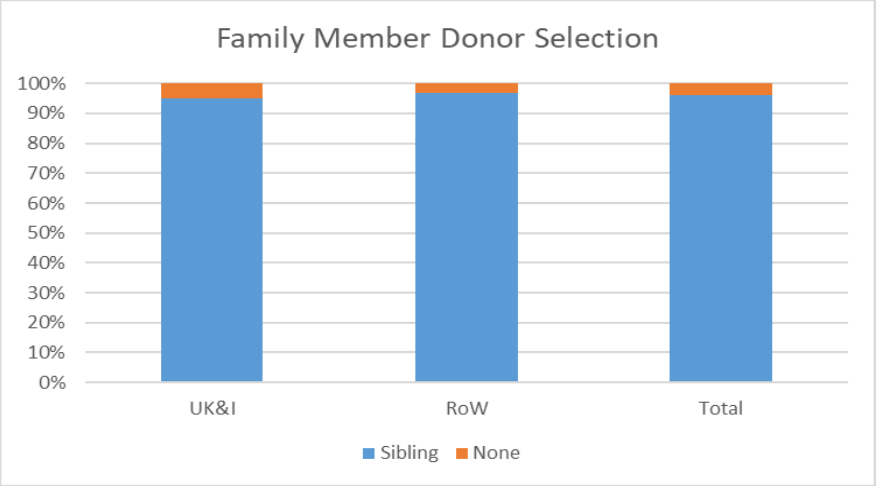
Summary of relevant HLA antibodies

HLA Class I Potential Donor Specific Antibodies	Date of Sample and MFI	
Specificity	26/06/2020	28/07/2020
A*24:02	17,510	18,018
B*13:02	25,004	24,791
B*27:05	19,675	19,387
B*58:01	*Negative*	*Negative*
C*02:02	3445	3064
C*03:02	4036	3962

6) Which donor would you suggest as being the favourable option and give your reasons for selection?

	UK&I	Percentage (n=19)	RoW	Percentage (n=30)	Total	Percentage (n=49)
Sibling	18	95%	29	97%	47	96%
None	1	5%	1	3%	2	4%



Most Common Reasons for Selection	
Sibling	Shares one haplotype - 6/12 match (GvH direction) and 7/12 match (HvG direction)
	Lowest cumulative DSAs of options available (C*03:02)
	DSA against HLA-C which has low expression
	HLA-DPB1 mismatch is non-permissive
	Potential issue around GVH direction homozygous HLA A2
	Desensitisation would likely be effective against a DSA at this level
	More likely to be matched at minor histocompatibility alleles
None of the Donors	Very high, paternal origin DSA against children. Intermediate DSA against sibling.
	Unknown whether children are above 18 years old and age of sibling unknown.

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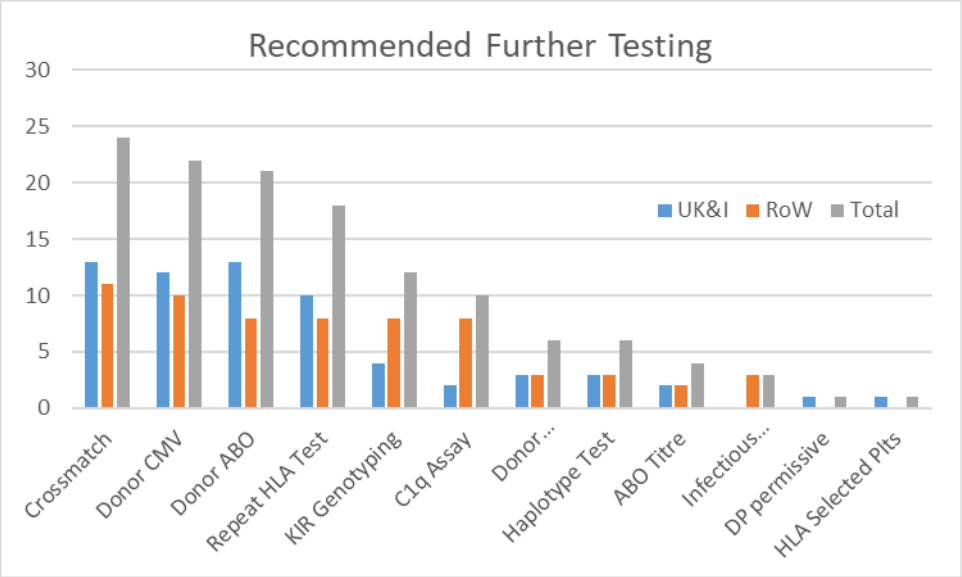
What, if any, further testing would you recommend to assess the risk of the transplant?

Testing Identified	UK&I	RoW	Total
Crossmatch	13 (20%)	11 (17%)	**24 (19%)**
Donor CMV	12 (19%)	10 (15%)	**22 (17%)**
Donor ABO	13 (20%)	8 (12.5%)	**21 (16%)**
Repeat HLA Antibody Screen	10 (15%)	8 (12.5%)	**18 (14%)**
KIR Genotyping	4 (7%)	8 (12.5%)	**12 (9%)**
C1q Assay	2 (3%)	8 (12.5%)	**10 (8%)**
Donor Age/Gender	3 (5%)	3 (5%)	**6 (5%)**
Haplotype Determination	3 (5%)	3 (5%)	**6 (5%)**
ABO Titre	2 (3%)	2 (3%)	**4 (3%)**
Infectious Markers	0 (0%)	3 (5%)	**3 (2%)**
DP permissive/non-permissive	1 (1.5%)	0 (0%)	**1 (1%)**
HLA selected platelets if required	1 (1.5%)	0 (0%)	**1 (1%)**



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7) Any further comments?

UK&I:

- Our team would recommend one of the single locus mismatched MUDs in this instance.
- Matched cells if transfusion required and repeat DSA monthly until transplant. Plasma exchange may be indicated to reduce HLA-C antibody further.
- Consider plasma exchange for patient pre-transplantation with sibling. Gender of related donors and ethnic background of patient would be useful.
- Size of the patient relative to the donor is considered, we wouldn't use a donor less than 2/3 the weight of the recipient. The donor would need to have a full health check. The antibody may need to be monitored post-transplant if delayed engraftment.
- If the patient is receiving HLA selected products a request could be made that HLA-B*58 and -C*03:02 should be avoided in the selected units to avoid sensitisation.
- Based on local experience we would expect a CDC and FC crossmatch to be negative. However, if it were to be positive, we would recommend 2 rounds of plasma exchange, followed by post-transplant antibody monitoring and early chimerism monitoring.
- We would need to consider the siblings age and fitness to transplant. We would still prefer to transplant using a 9/10 DSA negative VUD donor.
- Always important to discuss the clinical urgency as part of the MDT so that HR typing of sibling and children could potentially be initiated early if required.
- Depending on XM result antibody removal could be recommended to better ensure engraftment of the selected donor. Monitor antibody in run up to and post graft for prompt treatment if antibody continues prior to full chimerism.
- Concern about likelihood of disease relapse with haploidentical donor source.
- If the sibling is unsuitable we would crossmatch the children and perform antibody removal if required.

RoW

- We recommend desensitization of the recipient before transplant because of the presence of anti-HLA antibody.
- Desensitization with plasmapheresis.
- We would go for 1MM MUD donor.
- Plasmapheresis could be done on patient to reduce antibody level.
- A potential desentisation could be proposed to ensure a correct engraftment and avoid a prolonged aplasia after allograft.
- Desensitization of HLA antibodies against HLA-C*03:02 (MFI 3962) before transplantation process.

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Comments from UK NEQAS for H&I:

This scenario was based on a real case.

The unrelated donor options in question 2 were a true reflection of this patient's very limited options. Although both donors were investigated, both were unavailable.

The clinical team were unwilling to perform a mismatched transplant for this patient despite the availability of potential 9/10 mismatched donors (HLA-A* homozygote, 9/10 in GvH direction only). As such alternative transplant options were pursued.

Upon review of the cord search, the only units put forwards for consideration were Donor 2 and Donor 4 – these were, however, dismissed due to the cord bank not being accredited and cell dose respectively. The other cord units listed in this question were fictitious.

The clinical team decided to proceed with a haploidentical transplant and due to the strength of DSAs, the sibling was chosen as the best option.

A wet crossmatch was considered for this patient-donor pair but due to the sibling living in another country and logistical difficulties in getting fresh cells to the laboratory to perform a crossmatch, it was decided instead to perform a virtual crossmatch using two different samples.

The patient is now 4 months post-transplant and has been reported at 100% donor chimerism in the whole blood sample, and myeloid and T-lymphocyte subsets.