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International Quality Expertise

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Interpretive Educational Scheme (iED)
Clinical Scenario 2/2021 – Haematopoietic Stem Cell Transplantation

Dispatched on 31st August 2021

Summary of Submitted Responses

A total of 47 responses were received, 18 from UK & Ireland (UK&I) based laboratories and 29 from Rest of the World (RoW) based laboratories.

Background:

A 35 year old male patient suffering from Acute Myeloid Leukaemia (AML) received a haploidentical transplant from their 47 year old brother in 2018. HLA types for patient and sibling donor are given below.

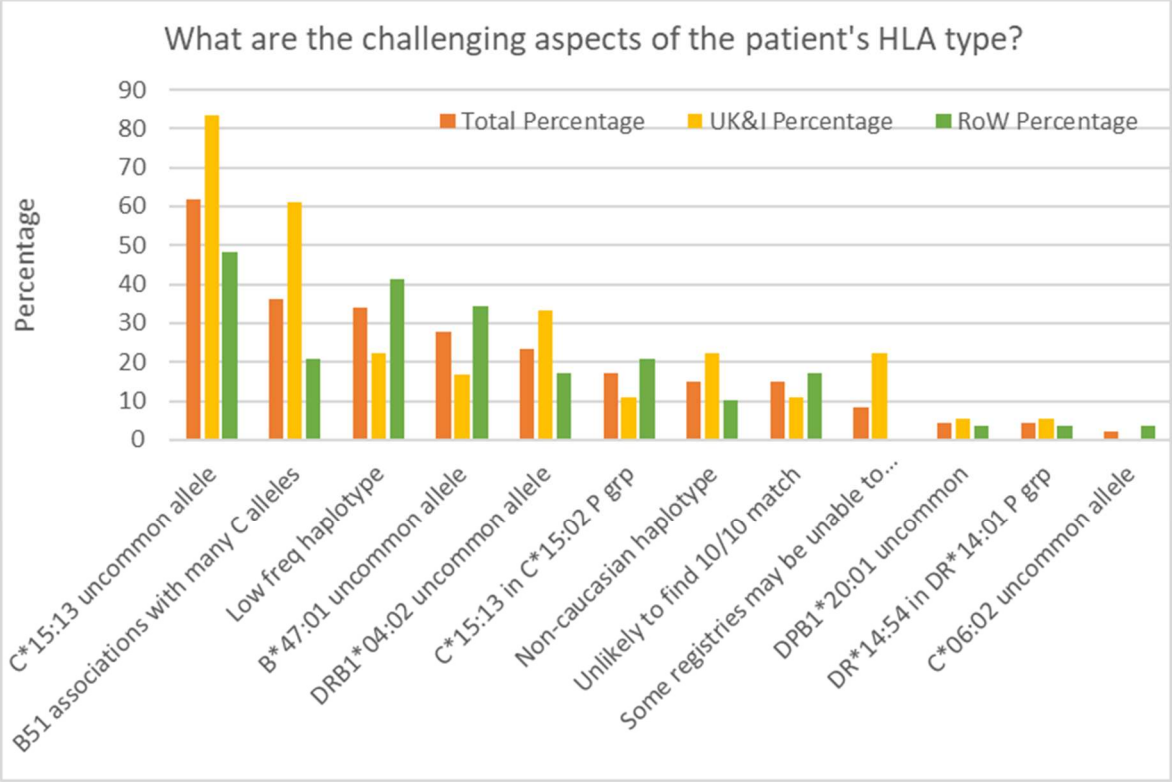
Patient's HLA Type:
HLA-A*02:01, A*03:01; B*47:01, B*51:01; C*06:02, C*15:13; DRB1*04:02, DRB1*14:54; DRB3*02:02, DRB4*01:03; DQB1*03:02, DQB1*05:03; DPB1*04:01, DPB1*20:01

Brother's HLA Type:
HLA-A*02:01, A*-; B*51:01, B*-; C*14:02, C*15:13; DRB1*04:02, DRB1*04:03; DRB4*01:03, DRB4*-; DQB1*03:02, DQB1*-; DPB1*04:01, DPB1*-

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

Reason	Total		UK&I		RoW	
	Number	%	Number	%	Number	%
C*15:13 uncommon allele	29	**62**	15	**83**	14	**48**
B51 associations with many C alleles	17	**36**	11	**61**	6	**21**
Low frequency haplotype	16	**34**	4	**22**	12	**41**
B*47:01 uncommon allele	13	**28**	3	**17**	10	**34**
DRB1*04:02 uncommon allele	11	**23**	6	**33**	5	**17**
C*15:13 in C*15:02 P grp	8	**17**	2	**11**	6	**21**
Non-Caucasian haplotype	7	**15**	4	**22**	3	**10**
Unlikely to find 10/10 match	7	**15**	2	**11**	5	**17**
Some registries may be unable to distinguish DR*14:01/DR*14:54	4	**9**	4	**22**	0	**0**
DPB1*20:01 uncommon	2	**4**	1	**6**	1	**3**

DR*14:54 in DR*14:01 P grp	2	**4**	1	**6**	1	**3**
C*06:02 uncommon allele	1	**2**	0	**0**	1	**3**



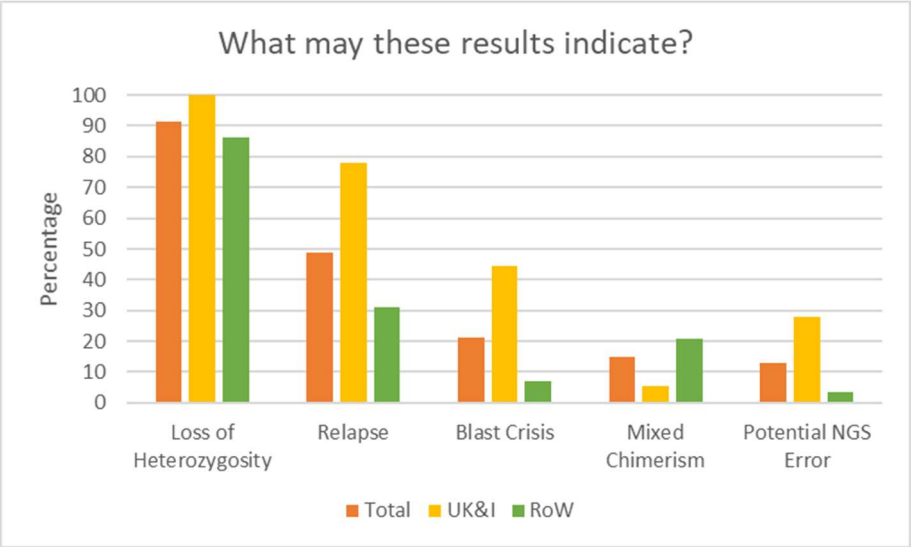
In November 2020, a post-transplant peripheral blood sample was received in the laboratory for HLA typing with suspected relapse.

HLA typing of the patient was performed post-transplant on a peripheral blood sample using next generation sequencing:

HLA-A*02:01, A*-; B*51:01, B*-; C*15:13, C*-; DRB1*04:02, DRB1*-; DRB4*01:03, DRB4*-; DQB1*03:02, DQB1*-

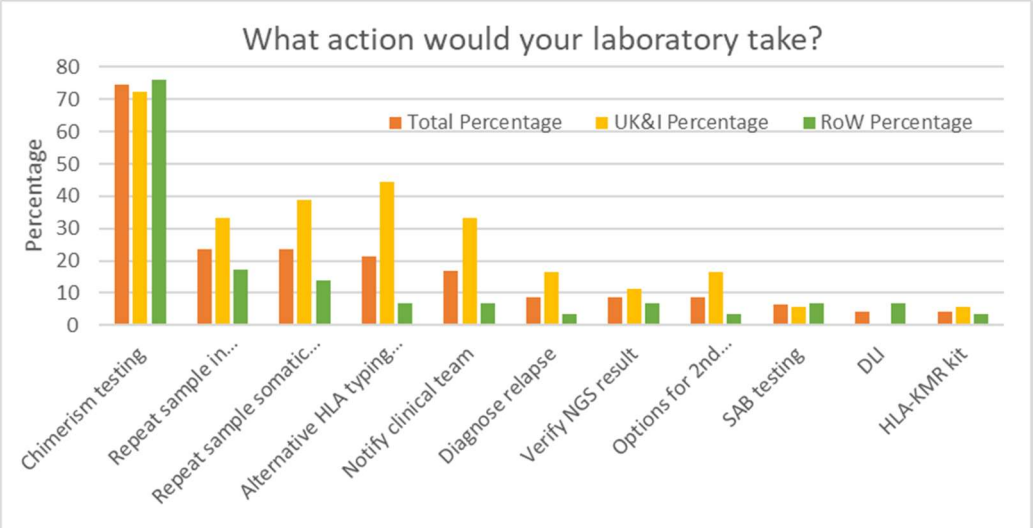
Question 2.1 - What may these results indicate?

Reason	Total		UK&I		RoW	
	Number	%	Number	%	Number	%
Loss of Heterozygosity	43	**91**	18	**100**	25	**86**
Relapse	23	**49**	14	**78**	9	**31**
Blast Crisis	10	**21**	8	**44**	2	**7**
Mixed Chimerism	7	**15**	1	**6**	6	**21**
Potential NGS Error	6	**13**	5	**28**	1	**3**



Question 2.2 - What action would your laboratory take?

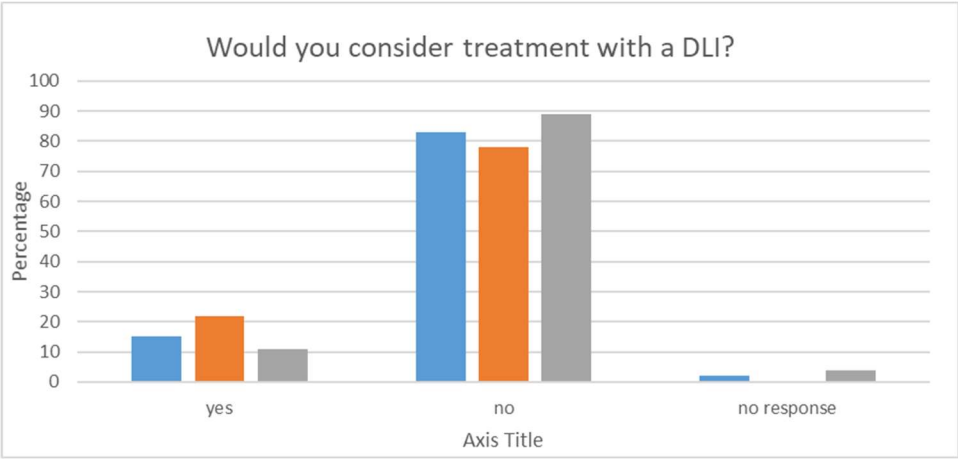
Reason	Total		UK&I		RoW	
	Number	%	Number	%	Number	%
Chimerism testing	35	**74**	13	**72**	22	**76**
Repeat sample in remission	11	**23**	6	**33**	5	**17**
Repeat sample somatic cells	11	**23**	7	**39**	4	**14**
Use alternative HLA typing method	10	**21**	8	**44**	2	**7**
Notify clinical team	8	**17**	6	**33**	2	**7**
Diagnose relapse	4	**9**	3	**17**	1	**3**
Verify NGS result	4	**9**	2	**11**	2	**7**
Investigate options for 2nd transplant	4	**9**	3	**17**	1	**3**
Perform single antigen bead testing	3	**6**	1	**6**	2	**7**
Donor lymphocyte infusion	2	**4**	0	**0**	2	**7**
HLA-KMR kit	2	**4**	1	**6**	1	**3**



Post-transplant chimerism results for the patient showed 20% donor cells in the whole blood samples and 95% donor cells in the T cell sample.

Question 3.1 – Would you consider treatment with donor lymphocyte infusions for this patient? Give reasons for your answer.

Decision	Reasons	Total		UK&I		RoW	
		Number	%	Number	%	Number	%
Yes	To increase GvL effect	7	**15**	4	**22**	3	**11**
	Aim to achieve complete chimerism						
	To avoid relapse						
	More useful for graft failure than active relapse						
	May be less effective as no mm for donor cells to target						
	Considered for all relapse patients unless have GvHD						
No	Increased risk of GvHD	39	**83**	14	**78**	25	**89**
	No benefit - no mismatch for donor cells to target						
	A second allograft from another donor with mismatched haplotype should be considered over DLI (EBMT Guidelines)						
	Relapse is extensive so unlikely to respond to DLI						
	95% of T cells are donor derived so no benefit of DLI						
	DLI only indicated if the lymphocyte fraction low						
	Relapse in myeloid line						
	Patient could benefit from NK cell cellular immunotherapy						
	Decision made by clinical team						
No response	We do not give advice to clinicians regarding post-transplant treatment	1	**2**	0	**0**	1	**4**



The patient is treated with chemotherapy and a second haplo-identical transplant is planned.

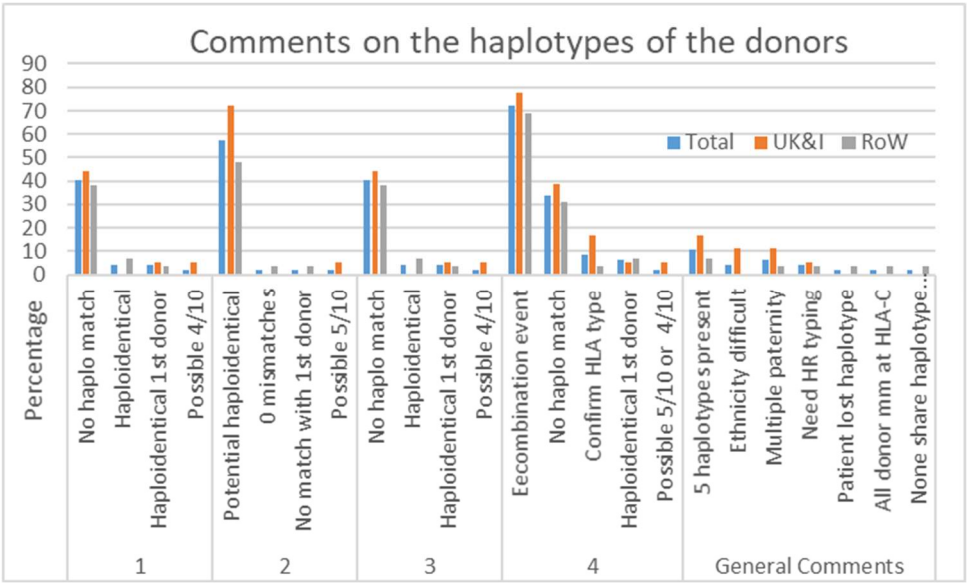
HLA typing reports from four further siblings are received from the family's country of origin:

ID	Gender	Age	HLA Type
Patient	*Male*	*35*	*A*02:01, B*51:01, C*15:13, DRB1*04:02, DQB1*03:02* *A*03:01, B*47:01, C*06:02, DRB1*14:54, DQB1*05:03*
1st Donor (Brother)	*Male*	*50*	*A*02:01, B*51:01, C*15:13, DRB1*04:02, DQB1*03:02* *A*02:01, B*51:01, C*14:02, DRB1*04:03, DQB1*03:02*
Donor 1	Male	50	A*02, B*51, C*14, DRB1*04, DQB1*03 A*24, B*38, C*12, DRB1*13, DQB1*06
Donor 2	Female	46	A*03, B*47, C*06, DRB1*14, DQB1*05:03 A*24, B*38, C*12, DRB1*13, DQB1*06
Donor 3	Female	47	A*02, B*51, C*14, DRB1*04, DQB1*03 A*24, B*38, C*12, DRB1*13, DQB1*06
Donor 4	Female	49	A*02, B*38, C*12, DRB1*13; DQB1*06 A*02, B*51, C*14, DRB1*04, DQB1*03

Question 4.1 – Comment on the haplotypes of the donors?

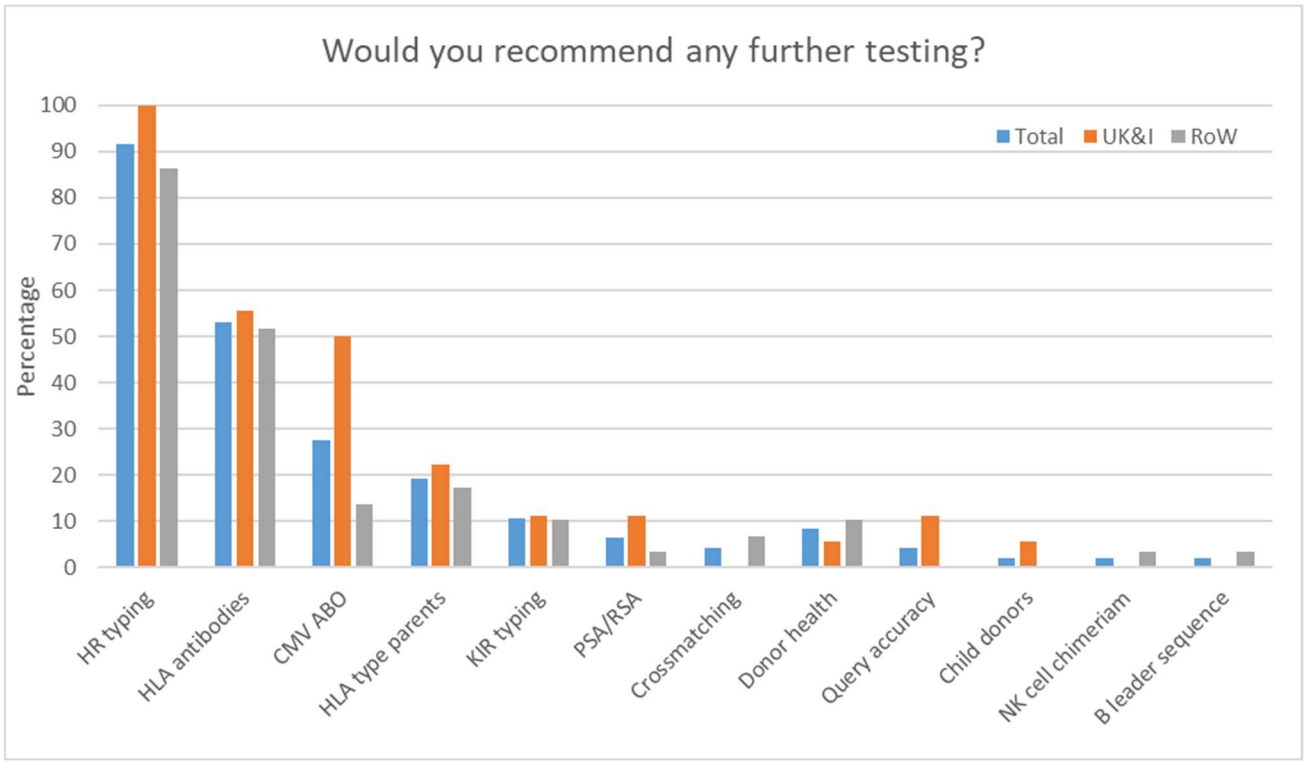
Donor	Comments	Total		UK&I		RoW	
		Number	%	Number	%	Number	%
1	Does not share a haplotype with patient	19	**40**	8	**44**	11	**38**
	Haploidentical	2	**4**	0	**0**	2	**7**
	Shares haplotype with 1st donor	2	**4**	1	**6**	1	**3**
	Possible 4/10	1	**2**	1	**6**	0	**0**
2	Potentially shares a haplotype with patient	27	**57**	13	**72**	14	**48**
	0 mismatches	1	**2**	0	**0**	1	**3**
	No match with 1st donor	1	**2**	0	**0**	1	**3**
	Possible 5/10	1	**2**	1	**6**	0	**0**
3	Does not share a haplotype with patient	19	**40**	8	**44**	11	**38**
	Haploidentical	2	**4**	0	**0**	2	**7**
	Shares haplotype with 1st donor	2	**4**	1	**6**	1	**3**
	Possible 4/10	1	**2**	1	**6**	0	**0**
4	Potential recombination event HLA-A	34	**72**	14	**78**	20	**69**
	Does not share a haplotype with patient	16	**34**	7	**39**	9	**31**
	Confirm HLA type	4	**9**	3	**17**	1	**3**
	Haploidentical with 1st donor	3	**6**	1	**6**	2	**7**
	Possible 5/10 HvG or 4/10 GvH direction	1	**2**	1	**6**	0	**0**

General Comments	5 haplotypes present	5	**11**	3	**17**	2	**7**
	Ethnicity difficult to define	2	**4**	2	**11**	0	**0**
	Potential multiple paternity	3	**6**	2	**11**	1	**3**
	Need high resolution typing	2	**4**	1	**6**	1	**3**
	Patient lost haplotype	1	**2**	0	**0**	1	**3**
	All donor mm at HLA-C with patient post-tx	1	**2**	0	**0**	1	**3**
	None share haplotype between patient and 1st donor	1	**2**	0	**0**	1	**3**

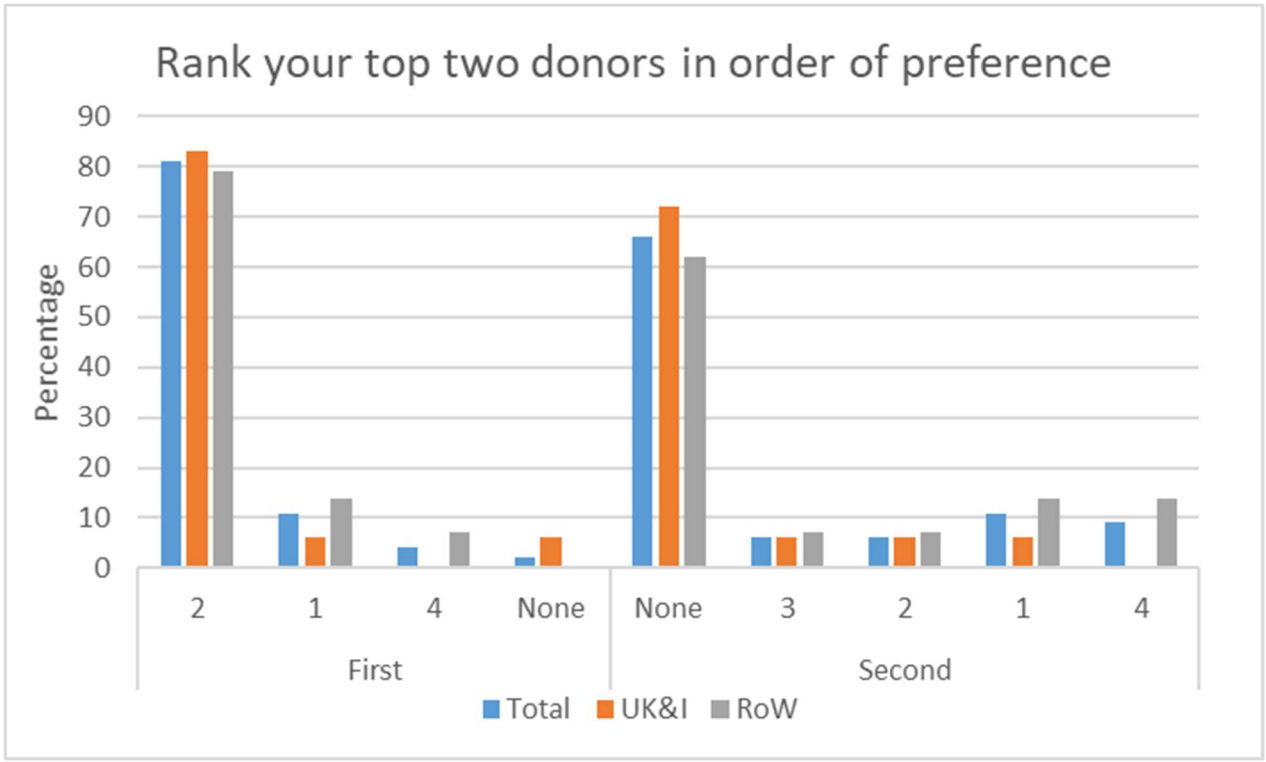


Question 4.2 – Would you recommend any further testing?

Further Testing	Total		UK&I		RoW	
	Number	%	Number	%	Number	%
High resolution typing of donors	43	**91**	18	**100**	25	**86**
Test patient for HLA antibodies	25	**53**	10	**56**	15	**52**
CMV and/or Blood Group	13	**28**	9	**50**	4	**14**
HLA type parents	9	**19**	4	**22**	5	**17**
KIR typing	5	**11**	2	**11**	3	**10**
Test donors for patient specific HLA antibodies	3	**6**	2	**11**	1	**3**
Crossmatching	2	**4**	0	**0**	2	**7**
Donor health / pregnancy info	4	**9**	1	**6**	3	**10**
Query accuracy of donor HLA typing	2	**4**	2	**11**	0	**0**
Enquire if patient has children able to donate	1	**2**	1	**6**	0	**0**
NK cell chimerism	1	**2**	0	**0**	1	**3**
B leader sequence of donor and patient	1	**2**	0	**0**	1	**3**

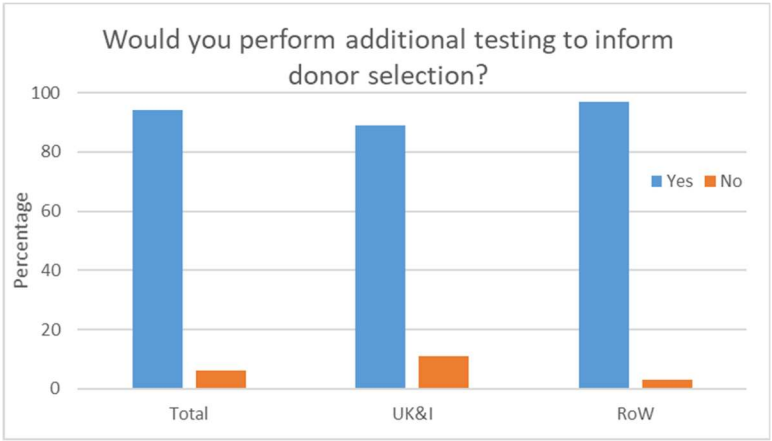


Question 4.3 – Based on the information provided so far, rank the top 2 related donors in the order of preference and outline your reasons.



Choice	Donor	Reason	Total		UK&I		RoW	
			Number	%	Number	%	Number	%
First	**2**	Potential shared haplotype	38	**81**	15	**83**	23	**79**
		Mismatch haplotype different to lost haplotype						
		Possible enhanced GvL effect						
		Youngest donor						
	1	Male donor	5	**11**	1	**6**	4	**14**
		Haploidentical						
		Likely less sensitising events						
	4	Closest match to first donor	2	**4**	0	**0**	2	**7**
		Least mismatches of all donors						
	None	Wait until NGS typing performed	1	**2**	1	**6**	0	**0**
Second	**None**	No other haploidentical donors	31	**66**	13	**72**	18	**62**
		All other donors HLA-C mismatched						
		Re-graft original donor						
		Use matched unrelated donor						
		Wait until NGS typing performed						
		Don't use original donor due to LoH						
	3	Haploidentical	3	**6**	1	**6**	2	**7**
		Younger than Donor 4						
		Avoid donor-recipient mismatch linked with GvHD						
	2	Best HLA match - one mismatch at HLA-C	3	**6**	1	**6**	2	**7**
		Youngest donor						
	1	Male donor	5	**11**	1	**6**	4	**14**
		Antigen match						
		May get NK alloreactivity						
	4	Possibility of NK cell reactivity	4	**9**	0	**0**	4	**14**
		Almost haploidentical						

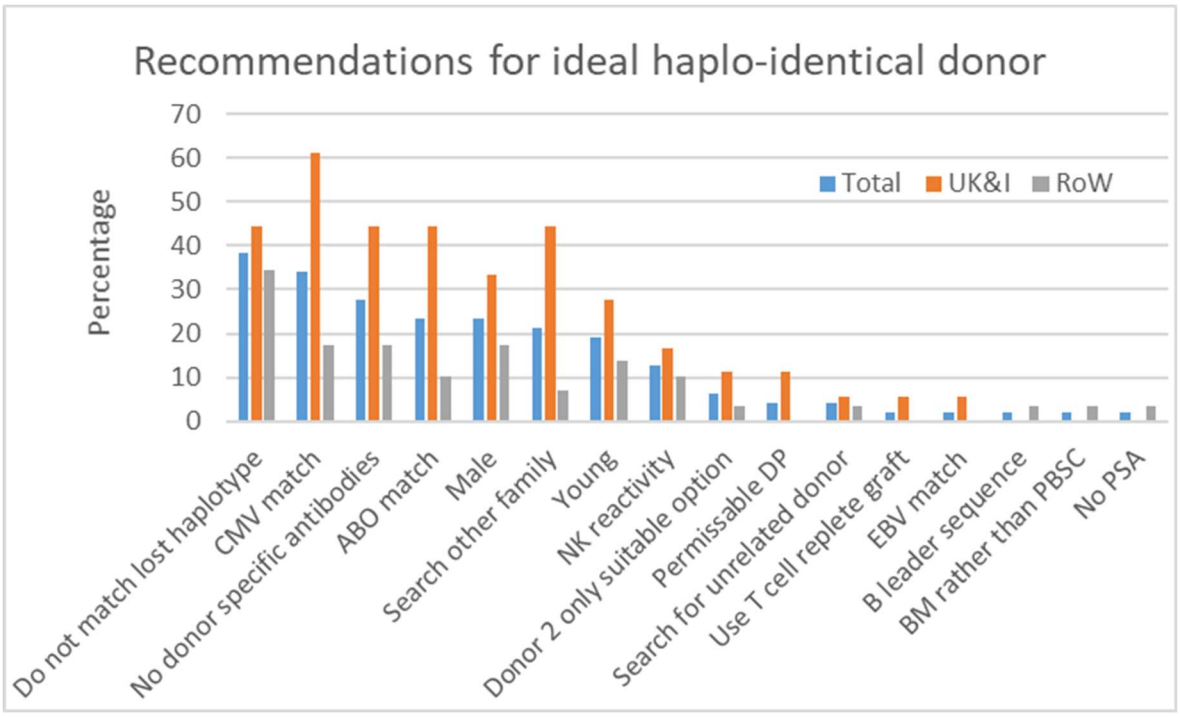
Question 5.1 – Would you perform additional testing or require any further information to inform selection of the related donor? Give reasons for your answer.



Decision	Reason	Total		UK&I		RoW	
		Number	%	Number	%	Number	%
Yes	High resolution HLA genotype / family pedigree	44	**94**	16	**89**	28	**97**
	KIR genotyping						
	ABO and/or CMV testing						
	Screen patient for HLA antibodies						
	Sensitisation history including pregnancies						
	Medical fitness incl. IDM, weight						
	Crossmatching if DSA present						
	Chimerism testing						
	Perform search for an unrelated donor						
	Test donor for patient specific HLA antibodies						
	Check B leader sequences						
	Consider NK cell immunotherapy						
No	Donor 2 the only acceptable related donor	3	**6**	2	**11**	1	**3**
	Due to limits donor options additional testing of limited benefit						

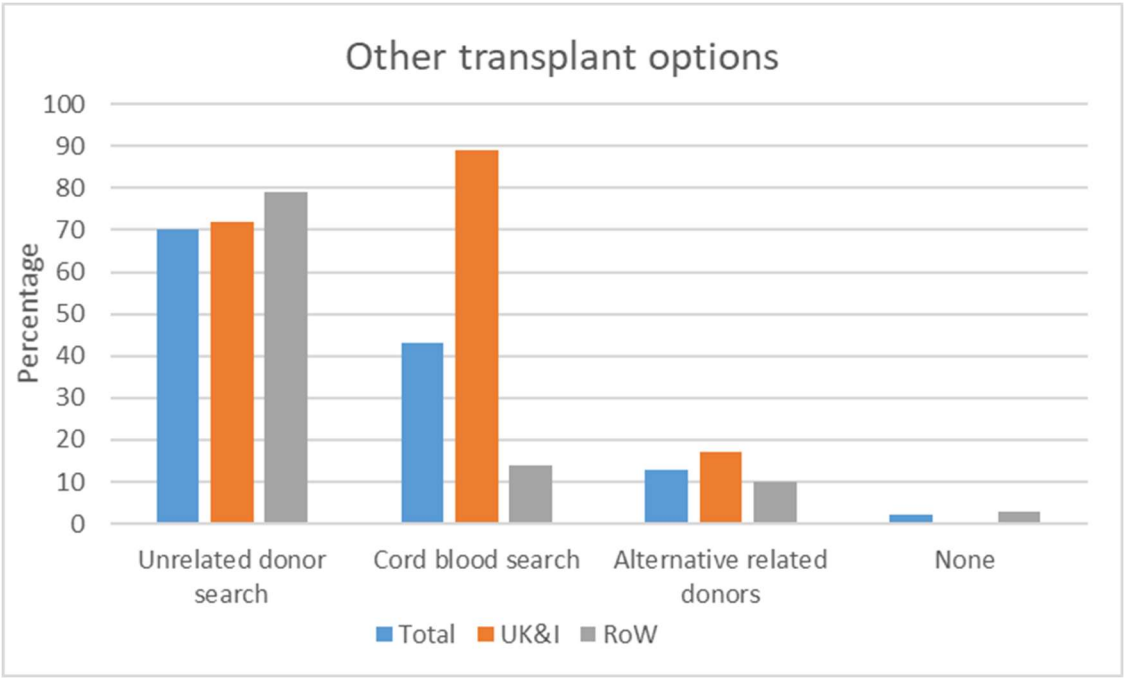
Question 6 – What recommendations (if any) would you give regarding the ideal haploidentical donor for this patient?

Reason	Total		UK&I		RoW	
	Number	%	Number	%	Number	%
New haploidentical donor should not match the lost haplotype	18	**38**	8	**44**	10	**34**
CMV match	16	**34**	11	**61**	5	**17**
No donor specific antibodies	13	**28**	8	**44**	5	**17**
ABO match	11	**23**	8	**44**	3	**10**
Male	11	**23**	6	**33**	5	**17**
Search other family members (NIMA or NIPA)	10	**21**	8	**44**	2	**7**
Young	9	**19**	5	**28**	4	**14**
NK reactivity	6	**13**	3	**17**	3	**10**
Donor 2 only suitable option	3	**6**	2	**11**	1	**3**
Permissable DP	2	**4**	2	**11**	0	**0**
Search for unrelated donor	2	**4**	1	**6**	1	**3**
Use T cell replete graft for GvL effect	1	**2**	1	**6**	0	**0**
EBV match	1	**2**	1	**6**	0	**0**
B leader sequence compatible	1	**2**	0	**0**	1	**3**
BM rather than PBSC	1	**2**	0	**0**	1	**3**
No patient specific HLA antibodies	1	**2**	0	**0**	1	**3**



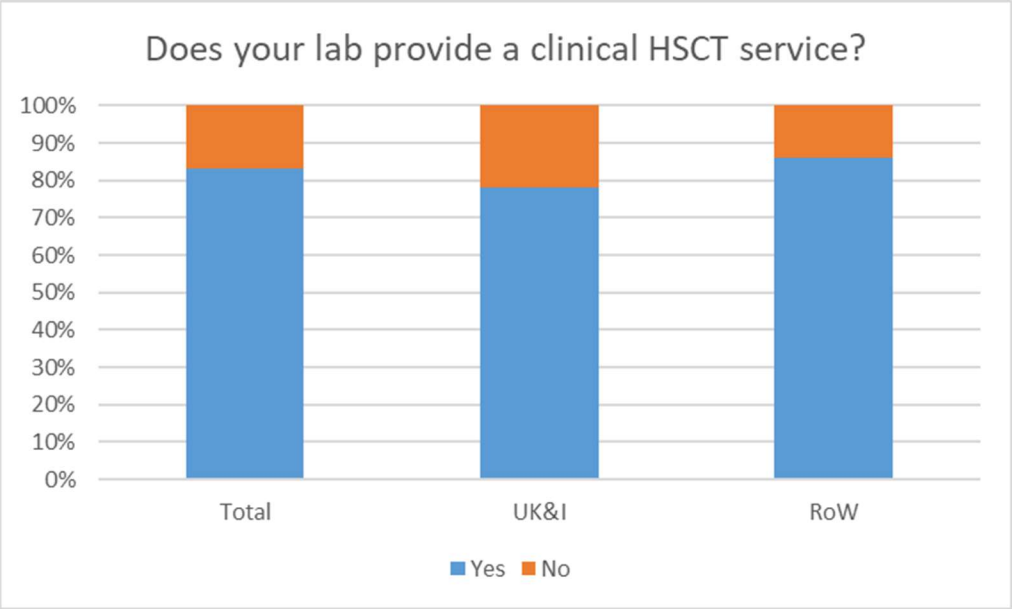
Question 7 – Would you consider any other transplant options?

Option	Total		UK&I		RoW	
	Number	%	Number	%	Number	%
Unrelated donor search	33	**70**	13	**72**	23	**79**
Cord blood search	20	**43**	16	**89**	4	**14**
Alternative related donors	6	**13**	3	**17**	3	**10**
None	1	**2**	0	**0**	1	**3**



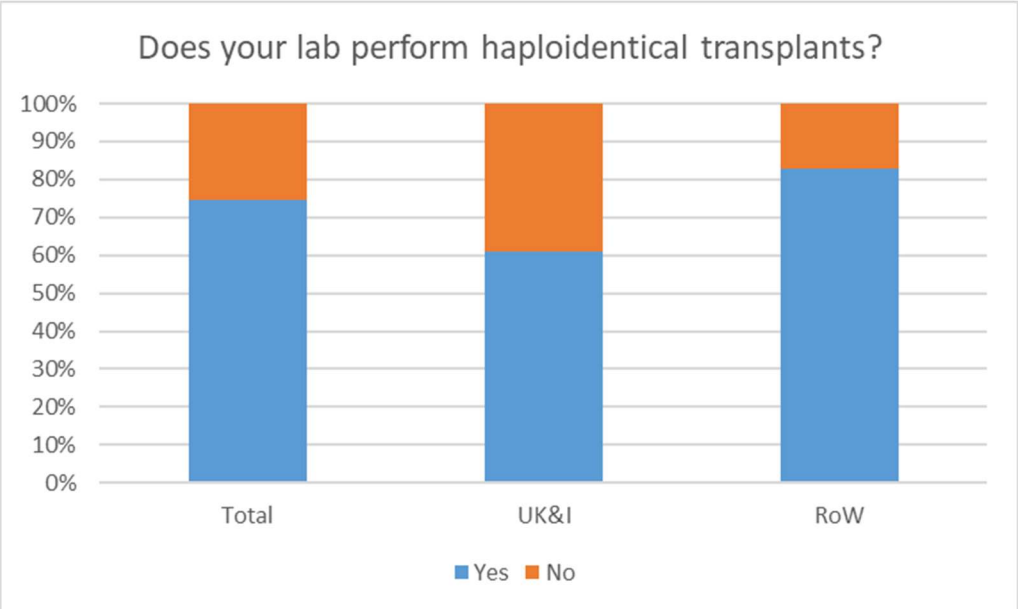
Question 8 – Does your laboratory provide a clinical HSCT service?

Response	Total		UK&I		RoW	
	Number	%	Number	%	Number	%
Yes	39	**83**	14	**78**	25	**86**
No	8	**17**	4	**22**	4	**14**



Question 9.1 – Does you laboratory routinely perform haploidentical transplants?

Response	Total		UK&I		RoW	
	Number	%	Number	%	Number	%
Yes	35	**74**	11	**61**	24	**83**
No	12	**26**	7	**39**	5	**17**



Question 9.2 – Any further comments?

- Aware of the potential use of NK cells in haplo transplantation, and their use in triggering graft vs leukaemia effect, in AML. However, this would not be undertaken in our routine setting.
- Are they all full siblings? There is a large age gap >10 years between patient and siblings, who are all of a similar age. If parents available would request samples to confirm family haplotypes.
- Haploidentical transplants are performed at this centre. Haploidentical transplant would be the option selected when the patient does not have a suitable matched sibling or 10/10 unrelated donor.
- HLA DP information is not recorded in the patient's post-transplant PB sample.
- In recent years, haploidentical transplants are being used more frequently, but they are not the preferred transplant option if a suitable HLA matched related, HLA matched unrelated or 9/10 unrelated donor is available.
- We only assist for HLA typing, Ab detection, KIR genotyping and time to time for donor selection (depending on the responsible physician) The HSCT performed within the Hematology clinics
- We do not have experience of allogenic bone marrow transplant. We only perform familial HLA typing study.
- Our laboratory does not perform chimerism and does not have any physician or contact with patients. Both are done by the hospital.
- We found this really confusing as we only perform haploidentical transplants when they are haplo at all loci. The C locus typing was a challenge to us as 3 0f the 4 donors were a complete mismatch.
- We do not perform chimerism in our lab and do not give advice to clinicians regarding post-transplant treatment.
- While we don't perform transplants ourselves, or make decisions regarding donor selections, we provide HLA typing, antibody screening and clinical data for transplantation. We don't perform all post-transplant monitoring, but we do perform post-transplant HLA antibody monitoring as well as platelet transfusion support. We do a lot of testing for haploidentical HSCT transplants, but only for a few hospitals.

General Comments

- Longitudinal chimaerism results would have been useful, rather than a snapshot, so that any trends could be spotted.
- This laboratory does not perform chimerism testing- it would be part of the transplant protocol in suspected relapse.
- Is there a mistake in the sibling HLA types?

Comments and suggested responses from the H&I experts providing scenario

Question 1
B47 is generally low frequency in our local population. The highest known frequencies are found in Central and Western Africa. This will likely mean it is challenging to find a 10/10 donor for this patient.

Question 2
The results indicate a possible loss of heterozygosity (LoH) post-transplant with the loss of mismatched haplotype as part of GVL immune evasion by leukemic stem cells.
It would be good practice to request a buccal swab sample from the patient to confirm LoH. If this is the case you would expect the HLA type from the buccal swab to correspond to pre-transplant HLA type. Also, information on proportion of blast cells can be used to confirm LoH.

Question 3
An attempt to induce remission by the infusion of donor T-lymphocytes (DLI) would be expected to be ineffective against the leukemic cells due to the LoH and therefore mismatched HLA target antigens. A DLI may also potentially be harmful to the patient due to the risk of inducing GvHD.

Question 4
Donor 4 has a HLA-A*02 which could be attributed to a recombination event based on segregation of haplotypes. We believe Donor 2 would be the preferred option as this donor is haploidentical but the mismatched haplotype differs to the haplotype lost by the leukemic cells signifying a potential graft versus leukemic (GvL) effect. This donor is also the youngest sibling.

Question 5
Further tested is recommended. This could include testing for HLA antibodies, high resolution HLA typing including HLA-DPB1, verification typing and KIR typing.

Question 6
We would recommend a second transplantation from a different HLA-haploidentical donor, selected for being mismatched against the HLA haplotype retained by leukemic blasts.

Patient Update
HLA antibody screening of the patient revealed he was donor specific antibody negative for all the potential sibling donors. Following chemotherapy to reduce disease burden, he was transplanted with Donor 2 (female, 46 years old) who was chosen to harness the potential GVL effect against HLA retained by the leukemic stem cells which experienced LoH to prevent another relapse. The patient had full donor chimerism 100 days post-transplant.