Passenger lymphocyte syndrome causing persistent immune-mediated haemolytic anaemia in a liver transplant recipient

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The patient

- 55 year old male
- Alcoholic liver disease with ascites, encephalopathy, portal hypertension and varices
- Abstinent since 2011
- 6th March 2014: deceased-donor liver transplant
- Female donor
- 4 unit RBC transfusion peri-operatively
- No prior transfusion history

Pre-transplant

	PATIENT	DONOR
GROUP	A RhD+	A RhD-
ANTIBODY SCREEN	Negative	Anti-D Anti-C

Post-transplant course

- Immunosuppression with tacrolimus, MMF and prednisolone
- Early graft dysfunction
- Haemolytic anaemia – Day 11:

Haemoglobin (g/L)	44
Platelets ($x10^9$/L)	418
Reticulocytes ($x10^9$/L)	266.3
Blood film	Leukoerythroblastic
Haptoglobins (g/L)	<0.2
Bilirubin (umol/L)	130
Lactate Dehydrogenase (IU/L)	777
DAT	2+ IgG
Eluate	Anti-C, Anti-D

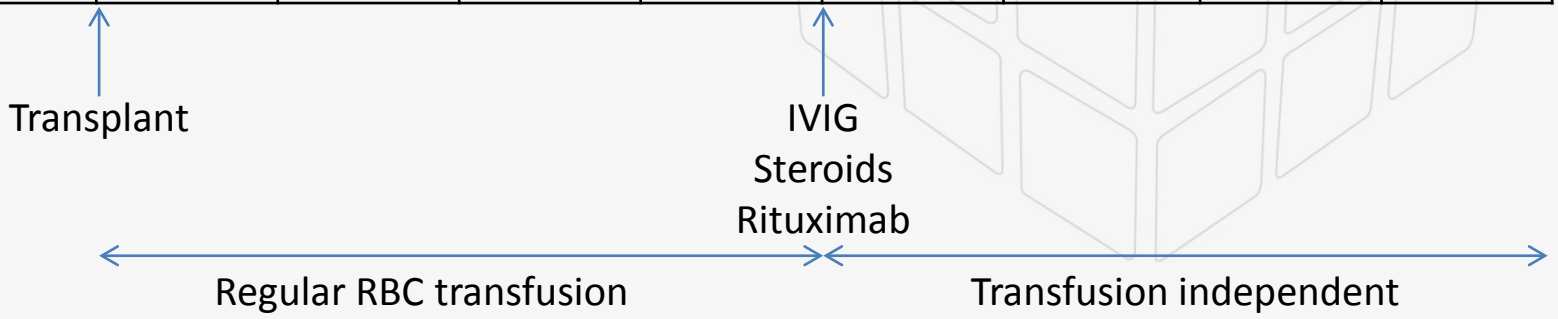
Ongoing haemolysis

- Monthly red cell transfusions post-discharge
- Transfused with ABO compatible, D- C-, E-, K- red cell units
- Persistent DAT+ve, Anti-C, Anti-D

- Seen by haematology at 6 months post-transplant:
  - *Erythropoeitin and folic acid*
  - *IVIG (1g/kg x2) Dec 2014 – No response*
  - *Prednisolone (30mg) Dec 2014 & Jan 2015*
  - *Rituximab (375mg/m^2 x4) Jan 2015*

Clinical course

	14/3/14	13/8/14	1/9/14	11/11/14	21/4/15	18/5/15	21/9/15	3/8/16
Hb (g/L)	44	96	80	82	114	113	115	119
Retics ($x10^9$/L)	266.3	230.5	167	325	382	209	235	
LDH (IU/L)	777	364	302	345	387	387		
Bili (umol/L)	130	31	25	24	16	17	19	13
DAT	IgG 2+	IgG +	IgG +	IgG 4+	IgG 4+	IgG +	IgG 3+	IgG +
Eluate	Anti-C Anti-D	Anti-C Anti-D	Anti-C Anti-D	Anti-C Anti-D	Anti-C Anti-D	Anti-C Anti-D	Anti-C Anti-D	Anti-C Anti-D



Investigations

- Red cell phenotyping:

  Rh phenotype R1R1 (CDe)

- Peripheral blood XY FISH:

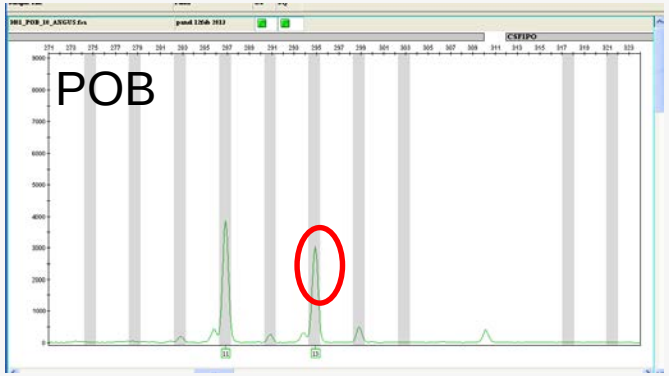
FISH analysis was performed using the CEP XY dual colour probe (Cytocell) on 200 cells from the cell fractions provided.

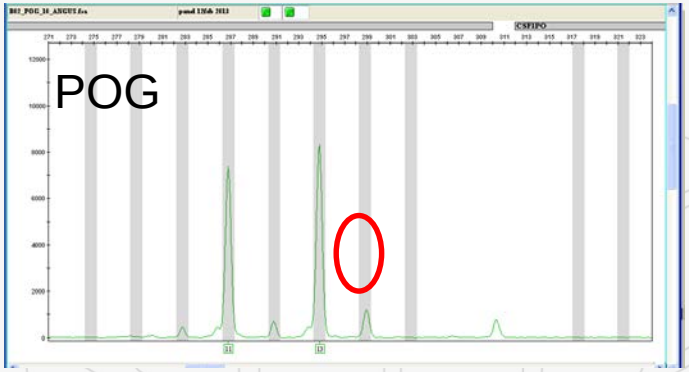
PBMC: 0% female (donor), 100% male (recipient).
Granulocytes: 0% female (donor), 100% male (recipient).
B-cells: 0% female (donor), 10% male (recipient).
T-cells: 0% female (donor), 100% male (recipient).

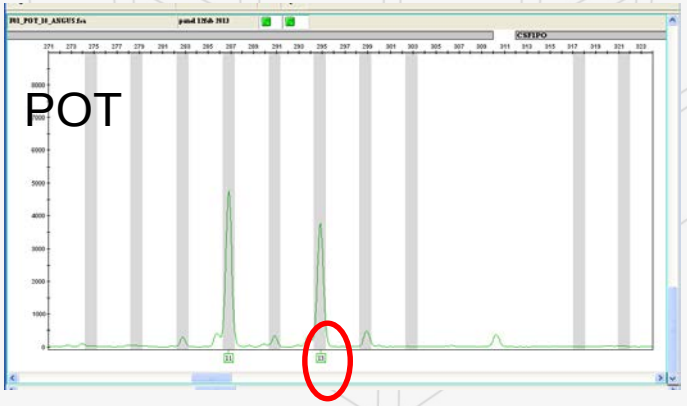
STR Analysis

- 4 recipient cell lysates from peripheral blood analysed to check for presence of donor DNA
  - B-cells, T-cells, granulocytes and mononuclear cells
- No donor tissue available for analysis
- Tested against 15 STR markers
- Extra peak noted with 3 of these markers raising the possibility of the presence of donor DNA

FMR labelled Dye marker D16S539







“The extra peak with D16S539 is present in the bin assigned to allele 14 and so could potentially be the result of the presence of Donor DNA.. There is also an extra peak to its right which does not fit to any bins.”

Passenger Lymphocyte Syndrome

- Graft versus host reaction where transplanted donor memory B-lymphocytes produce a secondary immune reaction against recipient red cells
- Most commonly seen in ABO incompatible transplants
- Onset of haemolysis usually 5-15 days post-transplant and usually resolves within 3 months, although Anti-D may last up to a year
- Use of calcineurin inhibitors post-transplant spares B-cell activity
- Lung>liver>kidney – more lymphoid tissue
- Can rarely be severe causing renal failure, DIC

PLS

Prendergrast et al. Blood 2013

- 1 in 200 solid organ transplants
- First detectable antibody D4-D120
- Duration of haemolysis 0-776 days

Post-transplant AIHA

- Seen in allogeneic SCT setting whereby the engrafted donor immune system reacts against donor red cells, typically 2-25 months post-transplant related to immune reconstitution
- Between 2.5-15% of patients
- More rare in solid organ transplants, but case reports in recipients of intestinal, renal and liver transplants where the recipient's immune system reacts with recipient red cells
- Use of calcineurin inhibitors thought to contribute through sparing of B-cell activity

PLS

- In favour of:
  - Onset at day 14 post transplant with detectable Anti-C, Anti-D
  - Rh incompatibility, Anti-C, Anti-D present in donor pre-transplant
  - Possible donor DNA detection in peripheral blood leukocytes could indicated engraftment of donor stem cells and mixed chimerism

- However:
  - Persistent DAT +ve and persistent Anti-C, Anti-D >2 years post-transplant
  - No definite evidence of mixed chimerism to confirm presence of donor lymphocytes without donor DNA sample

AIHA

- In favour of:
  - Patient is D+, C+
  - Long term persistence
  - Response to treatment with stability post rituximab

- However:
  - Rapid onset post-transplant
  - Donor serological status (Anti-C, Anti-D)

Diagnosis

- Passenger lymphocyte syndrome with mixed chimerism and donor stem cell engraftment
- Could perform biopsy of transplanted liver as a source of donor DNA to confirm diagnosis
- Could imply tolerance of donor cells and allow tapering of immunosuppression

Acknowledgements

- Mallika Sekhar
- Rita Atugonza
- Seble Tekle
- Edmund Lee

References

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- Pendergrast JM et al. Passenger Lymphocyte Syndrome Following Solid Organ Transplantation: Graft Source, Incidence, Specificity, Duration, and Severity Of Hemolysis. Blood 2013 122:37