LOSS OF RhD ANTIGENS

How *A Positive* Became *A Negative*

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CASE STUDY – A.M.

CLINICAL

- 73 year old female
- Known essential thrombocytosis (ET)
- Presented with worsening headache and papilloedema
- Extensive thromboses despite warfarin therapy
- Bone Marrow Trephine:
  - consistent with known essential thrombocytosis
  - no evidence of transformation to myelofibrosis
- Bone Marrow Cytogenetics:
  - no abnormalities at <300 band resolution
  - normal 46,XX[40] karyotype
- Diagnosed with cerebral venous sinus thrombosis

TRANSFUSION

- Grouped as A Positive at Nowra, 2011.
- Grouped as A Negative at Wollongong, 2012.
- Antibody screen consistently ‘Nil Detected’.
- Clinical notes did not provide any obvious explanation for the discrepancy.
- Collection error ruled out by two recollects.
- Tube RhD and weak-D typing confirmed RhD Negative (rr) result.
- A.M. confirmed to be the same patient that had been previously tested in Nowra
- A.M.’s group changed to A Negative in Cerner.

LITERATURE

1. Loss of Rh Antigen Associated With Acquired Rh Antibodies and a Chromosome Translocation in a Patient With Myeloid Metaplasia

2. Shift from Rh-positive to Rh-negative phenotype caused by a somatic mutation within the RHD gene in a patient with CML

3. Mosaicism due to myeloid lineage-restricted loss of heterozygosity as a cause of spontaneous Rh phenotype splitting

CONCLUSIONS

- In Caucasians, the deletion of the whole RHD gene is the usual basis for the RhD Negative phenotype, whilst RhD Positive individuals may be homozygous or heterozygous.
- A clear relationship was observed between haematological diseases and Rh antigen loss.
- The main reason for the loss of Rh antigens, after excluding transfusions, transplants, and inherited chimerism, was a loss of *RHD* heterozygosity.
- This was found to occur in three main ways:
  - the recombination of genetic material in chromosome 1 and the subsequent proliferation of abnormal stem cell clones (mosaicism) (most common).
  - a single nucleotide deletion in the *RHD* gene.
  - complete deletion of a stretch of chromosome 1, including the *RH* locus (rare).

- Flow cytometry can be used to determine the proportion of RhD Positive to RhD Negative red blood cells
- FISH analysis would enable recombinations of chromosome 1, and particularly the *RHD/RHCE* gene, to be visualised.
- Microsatellite analysis of the *RH* loci would highlight any nucleotide deletions, insertions, substitutions or duplications, and could also be used to provide the Rh genotype of the patient.

WHAT ABOUT A.M.?

- Crossmatching of RhD Negative red blood cells was the safest option.
- No further testing was performed to identify the cause of the RhD antigen loss
- Recent grouping of A.M. has identified weak RhD reactions. Is she returning to her A Positive phenotype, or are we just seeing an increase in the number of RhD Positive cells back up to detectable level?
- The remission of myeloproliferative disorders has been suggested to lead to the return of Rh antigens, however no improvement in A.M.'s ET has been recorded.
- Ongoing monitoring of A.M. may involve the careful observation of her blood film morphology and flow cytometry markers for signs of remission or, alternatively, transformation to myelofibrosis.

REFERENCES

- Cooper, B et al. Loss of Rh Antigen Associated With Acquired Rh Antibodies and a Chromosome Translocation in a Patient With Myeloid Metaplasia. Blood. Vol. 54, No 3, 1979.
- Cherif-Zahar, B et al. Shift from Rh-positive to Rh-negative phenotype caused by a somatic mutation within the *RHD* gene in a patient with CML. *BJH*. Vol. 102, pp. 1263-1270, 1998.
- Kormoczi, G et al. Mosaicism due to myeloid lineage-restricted loss of heterozygosity as a cause of spontaneous Rh phenotype splitting. *Blood*. Vol. 110, No 6, 2007

QUESTIONS?