



A Cryptic ABO Discrepancy

Meagan Wilson | St Vincent's Hospital,
Sydney



Two methodologies – two different results

- 51 year old female for investigation of MDS
- History of ALL treated with auto PBSC 20 years ago.
- The following results were obtained:

	Forward Group			Reverse Group				Interp
	Anti-A	Anti-B	Anti-D	A1 cells	A2 cells	B cells	O cells	
Biovue	0	4	0	4	-	0	-	B Neg
Tube	4 MF	4	0	3	3	0	0	??

- Reverse group consistent with B
- Forward group Bneg or ABneg??

Two methodologies – two different results

- Potential causes of discrepancy:
 - **A?B subtype with Anti-A1?**
 - A1 lectin pos?? – does not explain neg reaction with anti-A in Biovue
 - **Review patient history - ? Allogeneic group mismatch transplant**
 - Confirmed auto PBSC
 - **Recent transfusion –**
 - Performed blood group using cells from top and bottom of the cell pellet - identical results - only one cell population
 - Patient denied any recent hospital presentations

The answer

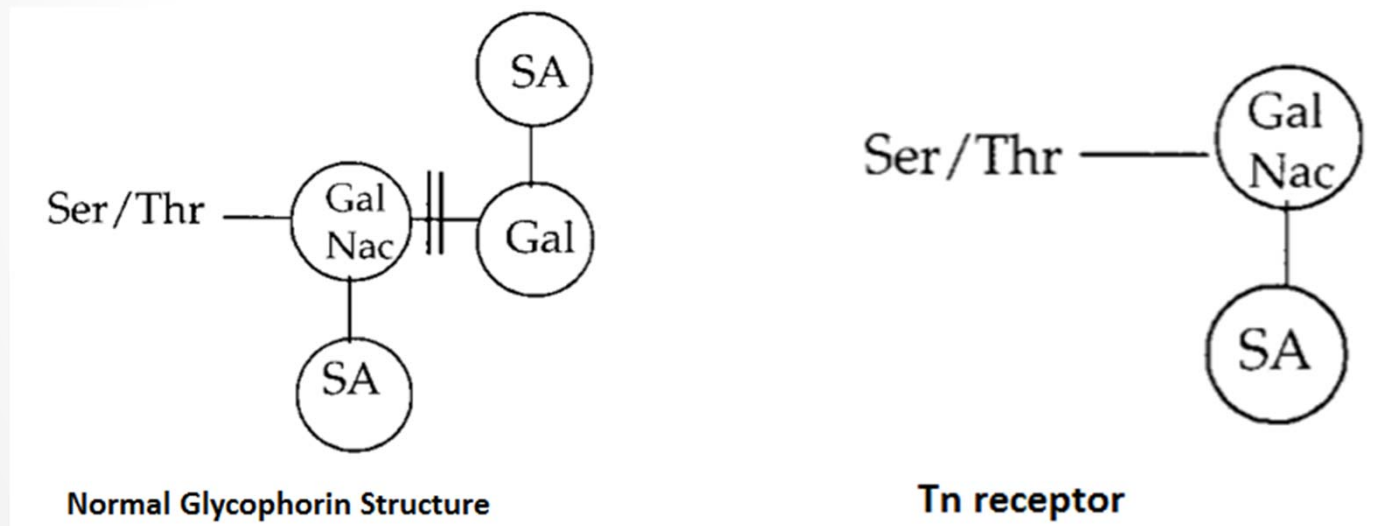
- A review of package inserts revealed different clones used in two monoclonal Anti-A reagents.

Reagent	Anti-A clone	Tn Polyagglutination
Ortho BioVue	MHO4 and 3D3	Does not react with Tn polyagglutinable cells
Epiclone (anti-A and anti A1)	4E7 and 8F2	May react with strong examples of Tn activated cells

- Confirmed by ARBCS reference laboratory to be Tn activation

Tn Activation

- Mutation in a stem cell clone results in some cells (including RBCs) to have altered expression of carbohydrate residues. Exposure of previously concealed Tn antigen ("cryptantigen")



https://4.bp.blogspot.com/-Dl12f17pjGk/Vx14iWfuMsI/AAAAAAAAACLw/GuqOWEo_WLM6jtu16WYztyJLTPLk7WSvgCLcB/s1600/TN%2Bpoly.png

Lab findings

- Pre-formed anti-Tn found in all adult sera.
- Some monoclonal antisera share epitopes with A glycoporphin and Tn resulting in false positive reactions in vitro in Tn activated cells.
- Mixed population of Tn activated RBCs and normal RBC results in typical mixed field reactions whereby only activated cells agglutinate

Lab findings

- Reactivity with different plant lectins permits differentiation of T activation.

Lectin / reagent	T	Th	Tk	Tx	Tn
<i>Arachis hypogaea</i> *	+	+	+	+	-
<i>Glycine soja</i> *	+	-	-	-	+
<i>Vicia cretica</i>	+	+	-	-	-
<i>Medicago disciformis</i>	+	+	-	-	-
<i>Salvia sclarea</i> *	-	-	-	-	+
<i>Salvia horminum</i> *	-	-	-	-	+
<i>Bandeiraea simplicifolia II</i>	-	-	+	-	-
<i>Vicia hircanica</i>	+	+	+	-	-
<i>Cord Serum</i>	-	-	-	-	-
<i>Polybrene</i>	-	-	+	-	-
<i>Papain & A hypogaea</i>	+	-	++	-	-

* lectin is a component of the Gamma Lectin System (Gamma Biologicals Inc.)
+ denotes agglutination occurs
++ denotes increased agglutination compared with untreated cells

Significance of Tn Activation

- Tn activation, when found, is often associated with MDS and acute leukaemia in adults. *In vitro* effects only observed and no special transfusion requirements are necessary.
- All plasma products contain preformed anti-T antibodies. In adults, exposure to similar environmental epitopes means that immune tolerance to T antigen occurs. *In vivo* antigen/antibody reactions not clinically significant in these individuals.
- Other forms of T activation (eg. T_{classical}, T_h, T_k, T_x) have been associated with varying degrees of intravascular haemolysis and are particularly significant in infants as often have a bacterial cause and ubiquitous T antibodies are not yet formed.

Concluding remarks

- Tn activation presents a unique ABO discrepancy in the lab. The advent of monoclonal reagents means that this is a rarely encountered observation.
- Benefits of using multiple reagents for performing ABO typing.
- Value of reading the package insert.
- Tn activation is the most benign form of T activation. Significant challenges for transfusion are presented in other forms of T activation such as T_{classical} as this is often associated with a bacterial cause and, when found is often found in young infants.

References

- Baldwin M.L, Barrasso C , Ridolfi R.L (1979); Tn-Polyagglutinability Associated with Acute Myelomonocytic Leukemia. *American Journal of Clinical Pathology* Vol. 72 pp 1024 - 1027
- Manno CS (2001); Does red cell T-activation matter? *British Journal of Haematology* Vol 114 pp115-120
- Berger E.G (1999); Tn Syndrome. [*Biochimica et Biophysica Acta \(BBA\) - Molecular Basis of Disease*](#) Vol 1455 pp 255 – 268
- Chaffin J (2011) *Perils of Poly(agglutination)* [Blog] That Blood Bank Guy. Available at: <http://bbguy.blogspot.com.au/2011/03/perils-of-polyagglutination.html> (accessed 14/09/2016)
- Massey E (2011) *Diagnosis and management of T antigen activation*. Available at: <http://hospital.blood.co.uk/media/2178/f62ef923-2fa0-4bea-82c1-c43989b7e111.pdf> (accessed 14/09/2016)
- Walker P S (2005) Polyagglutination. In: D.M Harmening; *Modern Blood Banking & Transfusion Practices* 5E: Philadelphia: F.A Davis Company pp. 508 -515
- Red Cell Reference Report ID:S144663 [2016, May][Raw Data], Australian Red Cross Blood Service, Alexandria, NSW