

INTRODUCTION OF THE MMA TEST – A PILOT STUDY

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South African Blood Transfusion Congress,
Sun City

30 August 2017



MMA???



Accessed online on 10/08/2016 from www.mutantmma.com



MMA???



Accessed online on 10/08/2016 from www.bleacherreport.com



MMA =

MONOCYTE

MONOLAYER

ASSAY



THE MONOCYTE MONOLAYER ASSAY

- In vitro assay
- Considered to be representative of in vivo survival of sensitized red blood cells
- Therefore, may be used as a predictor of:
 - The clinical severity of red cell alloantibodies
 - The severity of haemolytic disease of the newborn



INTRODUCTION INTO SANBS

- The MMA test was first put into use in the early 1970's – but not ever used in SANBS
- Implementation in SANBS Reference Lab will provide an additional tool to patients' treating physicians regarding the suitability of transfusing their patients with incompatible blood



SANBS APPLICATIONS FOR MMA

- Determining the possible clinical significance of alloantibodies to red blood cell antigens of high frequency where the patient may require transfusion and antigen negative blood is unavailable
- Post transfusion monitoring of cases in which patients with alloantibodies to high frequency antigens have been transfused with incompatible blood due to the lack of compatible or antigen negative blood



SANBS APPLICATIONS FOR MMA

- On request or in studies for determining the possible clinical significance of alloantibodies in haemolytic disease of the foetus and newborn (HDFN) cases
- MMA testing may be applicable when the following antibodies are identified and antigen negative blood is not available: anti-hr^B, -hr^S, -Yt^a, -Lu^b, -Ge:2, -Ge:3, -Lan, -Gy^a, -Hy, -Jo^a, -Jr^a, -LW, -At^a, -In^b, -Mi^a and -Cs^a



TEST PROCEDURE

- Identify the patient's alloantibody
- Select antigen positive RBC to be used in the test – prior to an incompatible transfusion, this will be a sample of the donor RBC
- Obtain 3 – 4 ACD tubes of whole blood from a healthy, willing monocyte donor
- Centrifuge ACD tubes at 150g for 10 minutes, remove the platelet rich plasma and resuspend in PBS



TEST PROCEDURE



Layer remaining
red cell/white
cell/PBS
mixture onto a
solution of
Ficoll



TEST PROCEDURE



TEST PROCEDURE

- The monocytes are transferred into a clean test tube and washed
- Sensitized cells are prepared
 - Patient serum (known antibody specificity) and donor RBC (ag+)
- Sensitized and unsensitized cells are washed and tested



TEST PROCEDURE



Monocytes are placed on a glass chamber slide, incubated and the supernatant removed – adherent monocytes remain on the slide



TEST PROCEDURE



Antibody-sensitized and unsensitized red blood cells added to the chambers and incubated with the monocyte layer



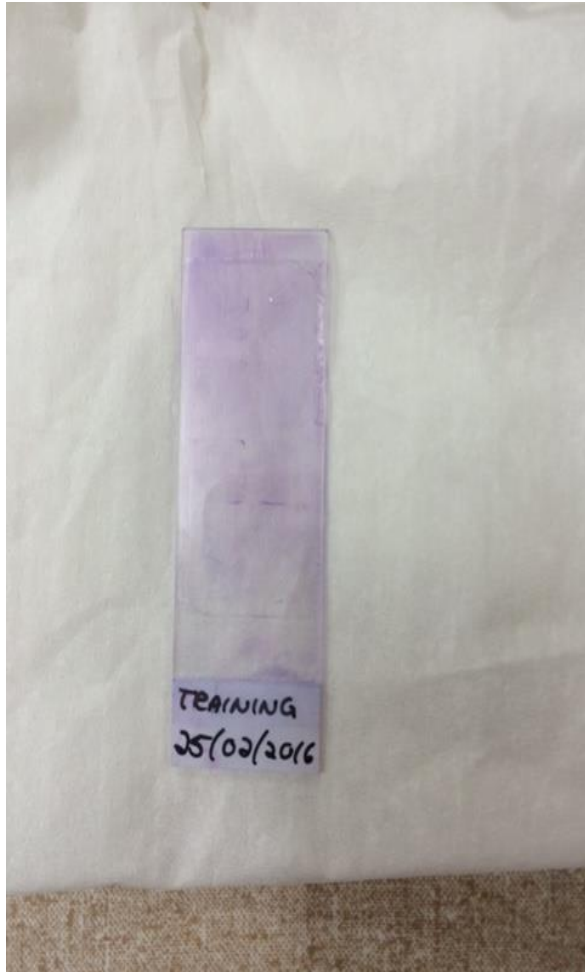
TEST PROCEDURE



The supernatant is removed with a pipette and the chambers separated from the slide



TEST PROCEDURE



- Unattached RBC are removed by washing and the slide is stained
- Slide is read microscopically
- Percentage of monocytes with adherent and/or phagocytosed RBC is determined.



INTERPRETATION OF RESULTS



Accessed online on 10/08/2016 at www.ucdmc.ucdavis.edu



INTERPRETATION OF RESULTS

- %R of $\leq 5\%$ = Incompatible blood could be given with little risk
- %R between 5.1 - 20% = 33% of patients may have clinical signs of a reaction
- %R of $> 20\%$ = 64% of patients may have clinical signs of a reaction



INTERPRETATION OF RESULTS

IMPORTANT:

Although values of $\leq 5\%$ have been associated with only a low risk of an acute haemolytic transfusion reaction, it must be noted that this value does not necessarily guarantee the normal, long-term in-vivo survival of the transfused RBC



SANBS MMA PILOT STUDY

- Challenge: no prior SANBS experience meant that external training would be required
- Training provided at the American Red Cross, Southern California Region
- ARC protocol brought back to SANBS for implementation
- SANBS Immunohaematology Reference Lab staff trained



SANBS MMA PILOT STUDY

- Series of 3 pilot tests were performed to confirm test performance in our laboratory
- Clinically significant antibodies used
 - 2 X anti-D and 1 x anti-K
 - %R >20% anticipated
- Actual results:
 - 56%, 69% and 44% respectively



SANBS MMA PILOT STUDY

- The MMA pilot study indicates that the ARC protocol can be successfully implemented in our laboratory
- Full test validation to be completed and documented
- Full rollout of the MMA in the Immunohaematology Laboratory is anticipated later this year



CONCLUSION

HOWEVER

- The decision to transfuse incompatible blood cannot be based solely on the MMA result – assess clinical need
- Assists the treating physician to make a more informed decision regarding patient treatment



REFERENCES

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