QUOTIENT

Evaluation of a Novel Serology-Based Platform for Irregular Blood Group Antibody Identification (Project MosaiQ™)

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Background

Screening of donor and patient samples for ABO system and irregular blood group antibodies is an essential part of pre-transfusion testing. Currently, ABO reverse typing and an antibody screening test will be conducted on each sample using an automated system. If the antibody screen is positive, an antibody identification investigation will subsequently be performed, often manually, before suitable blood can be selected for cross-matching. Identifying the antibody in the initial test would offer time and cost savings in the provision of blood for patients.

A new antibody-antigen microarray technology in development, known internally as MosaiQ™, has the capability to combine screening and identification, using only 50 µL sample volume, in one step, while simultaneously performing comprehensive blood group typing. The technology has been developed to meet the sensitivity levels required for donor and patient testing and of current state-of-the-art systems, without unwanted positives. This work reports an evaluation of the technology to perform antibody identification on 1030 samples.

Methods

Printing of red blood cell microarrays for antibody identification

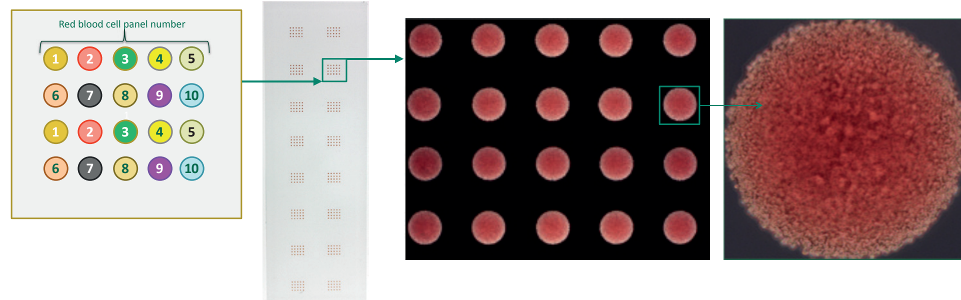

Figure 1: Quotient 10 cell panel red blood cells, suspended in a proprietary print buffer, are printed to a modified substrate. The 16 grids per slide are then subdivided into wells using a super-structure.

Processing of microarrays to form preserved monolayers

- Red cells immobilise to slides and are then processed to form preserved monolayers (Figure 2).
- These arrays can then be assayed as per Figure 3.

Figure 2: Printed red cells are then processed to form a uniform monolayer of cells which are then preserved.

Schematic representation of the MosaiQ™ antibody identification assay

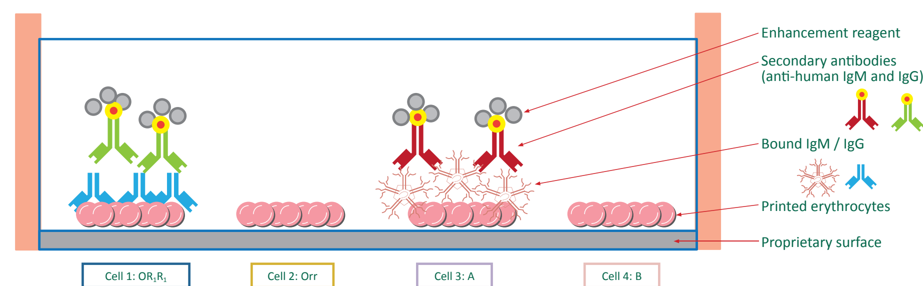

Figure 3: Diluted patient plasma is incubated with processed, immobilized red cells. After washing a secondary antibody cocktail is applied. Upon further washing an enhancement reagent is used to visualize a positive result (grey / black). White spots indicated a negative reaction.

Comparative testing methodology

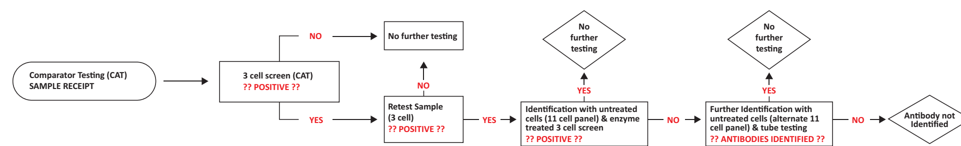

Figure 4: In addition to microarray testing all samples were tested in parallel with column-agglutination technology (CAT). The flowchart outlines the testing / retesting regime used to screen and identify the antibodies.

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Results

Proof of detection of all available specificities

- 23 specificities (IgG/IgM) detected using MosaiQ™.
- This included Anti-A, B, D, C, c, E, e, C^w, V, K, k, Kp^a, Kp^b, Fy^a, Fy^b, Jk^a, Le^b, Xg^a, Lu^a, S, s, M and P1

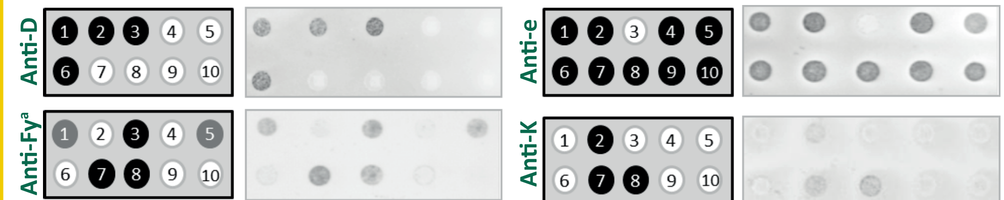

Figure 5: Example images when samples of Anti-D, Anti- Fy^a, Anti-e and Anti-K were tested using the MosaiQ™ system against a printed 10 cell panel and detected with an anti-human IgG & IgM cocktail of secondary antibodies.

1000 Random Donor (EDTA) Plasma Sample Study

99.9% Correlation to Comparator Achieved (Table 1)

Samples Tested	Negative by Comparator	Negative by MosaiQ™	Positive by Comparator	Positive by MosaiQ™
1000	996	997	4 after retesting (3 false positives)	3

Table 1: 1000 samples were tested on MosaiQ™ and with comparator systems. Seven positives were identified initially on the comparator system. After retest four of them were confirmed to be positives, of which MosaiQ™ detected three of these (3 out of 4). Of the negatives, 996 were identified by comparator systems, while MosaiQ™ found 997 negative.

Sample #0236 – Anti-K identified (11 cell panel) on CAT (0.5+ reaction). Not detected on MosaiQ™ although current work is focused on optimizing and automating the assay to enhance sensitivity.

Sample #0313 – Anti-M identified (11 cell panel) on CAT (0.5 + to 2+ reaction). Detected by MosaiQ™ (IgM).

Sample #0314 – Anti-C^w identified (11 cell panel) on CAT (2+ reaction). Detected by MosaiQ™ (IgM).

Demonstration of specificity detection of multiple Anti-D samples (n=15)

12 samples containing anti-D antibodies were sourced and tested on MosaiQ™. These included plasma and sera from the same donor, and weak control reagents manufactured by Quotient. Good correlation was shown between signals from MosaiQ™ arrays and 'potency' (determined by tube/CAT).

Sensitivity limit determination

Anti-D sensitivity level determined to be 0.05 iu/mL with current assay.

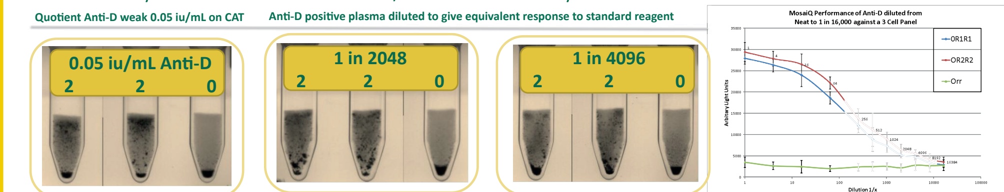

Figure 6: Anti-D positive plasma was diluted and tested on CAT until an equivalent response was obtained to the 0.05 iu/mL standard. This dilution series was then tested on MosaiQ and signals from positive cells were clearly differentiated from negative cells.

Conclusions

The study demonstrated that MosaiQ™ can be used for irregular blood group antibody identification in one step. The platform showed the required level of sensitivity against weak antibodies, whilst demonstrating no unwanted positives. Development work is continuing on additional antibody specificities. Adaptations of this format have also successfully demonstrated that blood typing, direct antiglobulin testing and virology assays can be performed simultaneously. The MosaiQ™ platform offers an efficient antibody identification system which challenges current serologic systems.