New Developments in Blood Group Serology

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The last issue of INFUSIONSTHERAPIE UND TRANSFUSIONSMEDIZIN included four articles covering a wide range of developments in serology. These included a review of monoclonal antibodies used in routine and research [1], a study of antibodies to high-frequency red blood cell antigens [2], a contribution to isotypes of cold agglutinins with discussion on the rare occurrence of IgG antibodies to the Pr antigens and the rarity of Ig class switching in cold agglutinins [3] and a paper on a microplate solid-phase method for typing red blood cells [4].

In addition to the important impact of monoclonal antibodies in routine and research, the other most significant advances have been seen with the introduction of the new systems and technologies for blood typing, antibody screening and compatibility testing.

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These new techniques must be shown by evaluation and clinical trials to be at least as good as existing techniques [5, 6] when in routine use. If new methods have greater sensitivity than standard tube tests, then this must be meaningful and cost effective, and not the detection of clinically non-significant antibodies that may not be cost effective or that may delay the issue of blood in urgent circumstances. Laboratories have adopted new technologies to help improve aspects of performance such as to reduce work time or to handle increased work loads in conditions of financial restraints; especially with automated or semi-automated systems staff numbers can be reduced.

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gy with simple endpoints that were easier to read than tube tests, e.g. micro-column and solid-phase systems, in wide use in our hospitals just after we had solved the major common problems of classical spin tube techniques. Certainly it is true that less skilled staff can use the new technology systems, especially those with automated reading devices.

However, serologists know that there is no perfect system for the detection of all antibodies. Thus, the micro-column systems (DiaMed and BioVue) for example may miss weak ABO incompatibility that is detectable by a rapid saline spin tube test [16]. Also micro-column techniques have been demonstrated to miss examples of other weak antibodies such as anti-Fy, anti-Jk, anti-S and anti-K [10, 11, 17]. These failures in detecting certain weak antibodies have been shown [17] to be due to shear forces breaking down weak agglutination as it is forced through the gel or bead matrix by centrifugation forces. For example, in one trial of 100 Gp B plasmas × A2B red blood cells the DiaMed gel antiglobulin test missed 68 incompatibilities while the BioVue bead antiglobulin test missed 76, compared to a 5-min spin tube saline test that only missed 8 and a LISS spin tube antiglobulin test that only missed 7 [17].

However, these failures have to be put in perspective within a clinical setting. Even when performed properly, the rapid saline direct spin tube test does not always demonstrate ABO incompatibility. In addition, the frequency of an incompatibility occurring between a group B patient and an A2B donor owing to limitations of the direct agglutination test in a laboratory where donor units are selected randomly regardless of recipients’ ABO group is very rare. Also the missed incompatibilities are predominantly anti-A1 cold antibodies i.e. titres less than 16 with A1 cells and little reaction with A2 cells, thus they are essentially not dangerous at 37°C with A2B cells that have even less reaction with low titre anti-A than A2 cells.

A far more important issue is that new technology systems, just like classical tube systems, are only as good as the quality of the screening red blood cell panels used for antibody detection. The drive to achieve a cost-effective antibody screening of patients’ specimens causes all laboratory practices to be reviewed, particularly when new technologies are introduced such as the DiaMed and Ortho micro-column tests. The standardisation of these tests and their reliable reading, together with the need for cost saving, led Lillevang et al. [18] to evaluate the use of pooled red blood cell samples for antibody screening using the DiaMed antiglobulin micro-column test. In a study of 5,446 antenatal and pretransfusion EDTA plasma samples, it was concluded that the use of two pools, each of two red cell samples, maintained transfusion safety and compared favourably with their previous testing regime. There pools contained C, D, c, e, k, Fya, Fyb, Jk, Kp, M, N, s, Le, Leb and Lu in the homozygous state and E, K, P1, Lu and others present in the heterozygous state.

However, this was challenged by Eggington et al. [19] who reported on the use of pools of two and three red blood cell samples in the gel micro-column antiglobulin test using 2,000 antenatal serum samples respectively. This study used pools that contained at least one red blood cell sample with a homozygous expression of C, D, c, e, Fya, Fyb, N, s, Le and Leb. The K antigen was present in the heterozygous state. The authors concluded that the use of pooled red blood cells led to a decrease in sensitivity which caused difficulty in the interpretation of results, and that the detection of known antibodies was reduced. Similar conclusions were reached by Bombail-Girard et al. [20].

Microcolumn tests are able to separate unagglutinated from agglutinated red blood cells and, in theory, offer the ability to use pooled red blood cells to investigate the loading capacity of the gel or glass bead column matrix. Neither of the investigators used red blood cells with a homozygous expression of the K antigen, antibodies of which can be difficult to detect in low-ionic-strength antiglobulin tests.

The current guidelines for the UK Blood Transfusion Services [21] state that pooled red blood cells should not be used for the screening of patients specimens. This is based on long experience with tube test methods showing that the use of pooled red blood cell results in a down-grading of the reaction (by the presence of a mixture of red blood cells, some with and some lacking the corresponding antigen to antibody present in the serum), and presents reading difficulties in the interpretation of mixed-field reactions.

A far better move to cost-effective cell screening can be achieved by proper management of screening, selection and use of high-quality two-cell panels by a policy of having large-scale or national reagents laboratories. Methods are now available for the frozen storage and retrieval of matched pairs of reagent red blood cell units, containing antigens of probable clinical significance, without significant loss of these antigens over a usable period appropriate to screening cells. Such matched pairs of R1R1 and R2R2 donations with homozygous expression of all antigens of probable clinical significance but with heterozygous K occur with a frequency of some 10 matched pairs in 100,000 donors. Using them results in considerable resource and investment. However, their use, compared with a three red blood cell sample screen, could cause a saving of some 1.4 million GBP per year in consumable costs over the UK and a 30% saving in work load [22].
References


