HISTORICAL ASPECTS OF BLOOD BANK TECHNIQUES

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The earliest history of blood transfusion involved the direct transfer of blood from one person to another. Direct transfusion was used mainly due to the initial lack of effective anticoagulation resulting in the inability to successfully store donor blood. The success of this technique was somewhat 'hit or miss' depending upon the (unknown) ABO blood group compatibility of the donor and patient.

Karl Landsteiner identified that differences existed between the bloods of different people based on *in vitro* reactions, which led to his publication in 1901 of the identification of three distinct 'blood groups', which he eventually called A, B and O. This discovery was made possible by the presence of (what were originally called) 'naturally occurring' anti-A and anti-B antibodies in the plasma of normal people. The fourth ABO blood group AB was identified by von Decastello and Sturli in 1902 and the ABO blood groups were subsequently identified to be inherited characteristics. The significance of the ABO blood groups in the practice of transfusion was subsequently identified by the fact that *in vitro* red cell agglutination is mirrored by *in vivo* reactions resulting in the destruction of transfused red cells.

However, the discovery of the ABO blood groups was not the beginning of the use of blood grouping techniques. There are a number of reasons for this including the fact that Landsteiner did not expand his original findings and moved into other areas of research resulting in a lack of appreciation of the significance of his original observations. In fact, Jansky in Poland (1907) and Moss in America (1910), having no knowledge of Landsteiner's work (that was originally published in German), independently identified the ABO blood groups but confusingly used nomenclatures based on different roman numerals, i.e. Jansky: I (O), II (A), III (B), IV (AB) and Moss: I (AB), II (A), III (B), IV (O).

As well as these very real nomenclature difficulties, which were not effectively resolved until 1922, there were a number of practical difficulties that impeded the introduction and routine use of ABO blood grouping techniques, not least of which was the variability in strength and reactivity of the human derived grouping sera that were used. These reagents were initially procured 'in-house' and were subject to variable deterioration during storage. The routine use of ABO grouped blood was also not well accepted by surgeons due not only to the lack of reliability of the technology used but also to the time it took to get the donor blood tested (which could be a number of hours) related to the potential urgent need of the patient. These various factors were instrumental in the preferential use of 'group O universal donor' blood, which could be used without the need for pre-transfusion ABO grouping of the blood donor and the patient.

Initially manual ABO grouping was performed on glass slides or in small glass (precipitin) tubes using weak suspensions of red cells from the donor / patient suspended in isotonic saline, mixed with aliquots of known (human derived – polyclonal) grouping sera, i.e. anti-A, anti-B and anti-A,B obtained from group B, A and O donors respectively (Fig.1). This procedure was eventually 'controlled' by the use of a 'reverse group' whereby an aliquot of the serum of the donor / patient was mixed with aliquots of red cells of known blood group (i.e. group A, B and O).

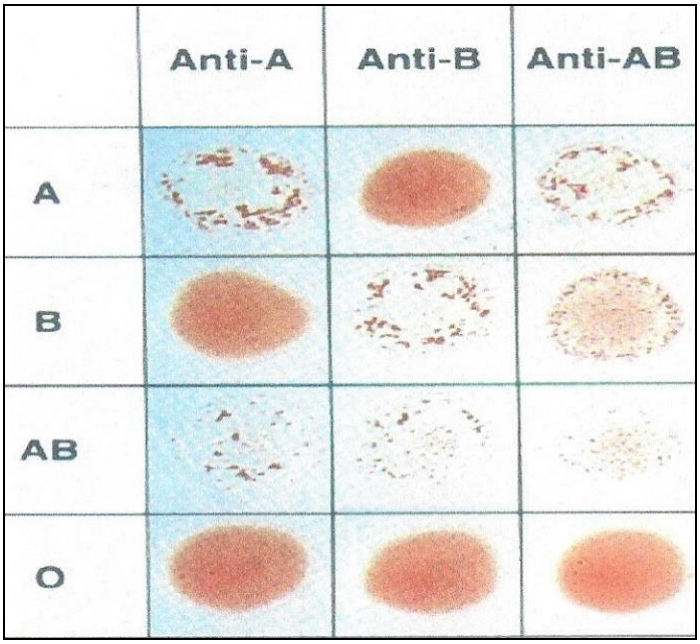

Fig.1: Manual ABO grouping results on a white tile of the four ABO groups showing agglutinated and un-agglutinated red cell reactions

The basic blood group serology techniques were therefore based on direct agglutination being produced by mixing an aliquot of a dilute red cell suspension with an aliquot of serum (or plasma) which were then left together ('incubated') for a period of time, usually at room temperature. A Pasteur pipette was used to produce standardised drop volumes (of approximately 30μl), the pipette barrel being held vertically between the second and third fingers and the teat between the thumb and first finger (Fig.2). Though simple to use, effective cleaning of the pipette, by repeated rinsing / washing in saline during use was essential to avoid cross-contamination ('carry-over') of sample material. Testing was completed by careful visual examination of the red cells for agglutination with the test results being written onto a worksheet. Variations in technique methodology between different laboratories were common.

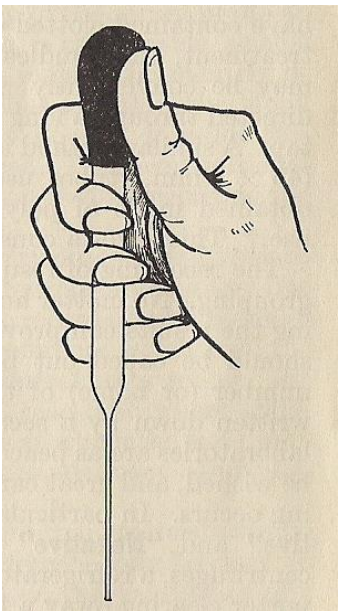

Fig.2: Holding a Pasteur pipette

The initial pre-transfusion ‘cross-match’ was introduced to supplement / check the accuracy of the donor / patient ABO grouping and initially involved only a direct incubation at room temperature of an aliquot of donor red cells and patient’s serum.

Prior to WW2 there was little real advance in the knowledge of blood groups or the development of improved techniques that were capable of detecting clinically significant antibodies related to patient care. The usefulness of blood grouping improved with the introduction of ‘incomplete’ techniques that were capable of detecting IgG blood group antibodies (especially Rh) via the introduction of the bovine albumin test methods (1945), enzyme treatment methods (1946-47) and especially the anti-human globulin (AHG) technique (1945) which was pivotal in enabling the detection of IgG immune antibodies present in the patient’s plasma pre-transfusion. Even so, the methodology of performing the manual ‘AHG test’ (in kahn tubes) with multiple manual red cell washing stages using a bench centrifuge was time consuming and subject to potential error that resulted in a false negative result. However, the development of these different laboratory techniques and technologies eventually enabled the pre-transfusion detection of clinically significant antibodies in the patient and the development of ‘compatibility testing’ (Fig.3).

Fig.3: Performing manual blood grouping techniques (c.1960-1970s)

The sophistication of blood grouping methodologies also mirrored the identification of different blood groups (especially the importance of Rh in Haemolytic Disease of the Foetus / Newborn) and the need for automation, resulting in the development of microplate and column (‘card’) technologies. Improved accuracy, speed, reliability and reproducibility of blood bank techniques was achieved only following the development and use of commercial monoclonal grouping reagents, the introduction of standardised quality assured techniques and more sensitive alternative technologies.

For information about the early history of blood bank techniques and the problems outlined above see: ‘Blood Group Serology – the first four decades (1900 – 1939)’ by A.D. Farr. *Medical History* 1979, 23: 215-226
https://www.cambridge.org/core/journals/medical-history/article/blood-group-serologythe-first-four-decades-19001939/2E501E82CEF61F22BDFABF526D7DC515