Comparison between conventional tube method and gel card technique for blood crossmatching: A study in a tertiary care center of north India

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ABSTRACT

The cross-matching of blood can be performed by various methods like saline tube method and gel card technique. Saline tube method is most commonly used in blood banks, whereas the latest method used is Gel card technique for cross-matching. Both techniques have their advantages and disadvantages. The main aim of this study was the comparison of gel card and conventional tube test for sensitivity and specificity, time and efficacy. This prospective study was done at the Department of Blood Transfusion, BPS GMC Khanpur Kalan, Sonipat, Haryana. A total of 500 samples collected at the Department of Blood Transfusion, were cross-matched using conventional tube method (spin tube method) with and without using AHG and Gel card Method. 497 (99.4%) samples were compatible, and 3 (0.6%) samples were incompatible with Gel card method, but by test tube method 492 (98.4%) samples were consistent, and 5 (1.6%) were false positive (FP). Incompatibility of those 5 samples (FP) disappeared after incubation with AHG reagent at 37°C saline tube method. The sensitivity and specificity of both gel card method and saline tube method with AHG methods is 100% whereas the specificity of saline tube method without AHG is 98.9%. The average time taken by Gel card method was 20-30 minutes for a single compatibility test whereas in conventional spin tube method with the use of AHG (IAT) average time required was 90 minutes and without AHG it was 45-60 minutes. The sensitivity and specificity of the spin tube method and the gel card method are comparable to each other. But the gel card method is easy to perform, rapid, reliable procedure and results can be recorded. In contrast, spin saline tube method is more time consuming, and results cannot be preserved. Thus gel card technique can be preferred over the spin tube method.

INTRODUCTION

In the entire transfusion process, one of the most important factors is pre-transfusion compatibility testing to ensure blood safety (South et al., 2012). The main goal of blood transfusion services (BTS) is to make sure that blood and blood products are safe, and the supply is adequate (Singh, 2018). In pre-transfusion testing, a series of actions are pulled out to ensure that donor and recipient blood groups (ABO and Rh system) are compatible with each other and to rule out any antibodies in the
recipient’s serum that could react with transfused blood (Downes and Shulman, 2011). The main aim of the cross match is to select blood components that will have acceptable survival when transfused and prevent the occurrence of immune-mediated hemolytic transfusion reaction with the transfusion of incompatible donor red cells (Shulman et al., 2001). The compatibility testing method includes ABO and Rh grouping of both (recipients and donors), then cross-matching between recipient’s serum and donor RBCs in indirect antiglobulin (IAT) phase to detect the presence of any clinically significant antibodies (Downes and Shulman, 2011), (Carlson TH, 2011). Spin tube method is the conventional and most commonly used method for blood grouping and cross-matching, which includes saline tube method and indirect coombs method. Gel technique was introduced by Lapierre, which is based on controlled centrifugation of red cells through Sephadex gel contained within microtube (Lapierre, 1988). The gel card technique can be used for ABO and Rh typing, cross-matching Direct and Indirect Antiglobulin Tests (DAT and IAT) and identification of alloantibodies (Letich et al., 1993), (Kaur et al., 2003). Though conventional tube technique (CTT) is still considered a gold standard, it has some limitations like elution of low-affinity antibodies during washing, variability in the results due to variations in the cell serum ratio, and lack of consistency in reporting the results due to inter-observer variability (Bajpai et al., 2012). Rumsey and Ciesielski, 2000. Gel card is less time consuming with advantages as results can be read easily, recorded for a long time, easy handling and disposal but costlier than CTT (Gond et al., 2016), (Singh et al., 2017). The present study was done for comparison between gel card and conventional tube test for sensitivity and specificity, time and efficacy.  

MATERIALS AND METHODS

A total of 500 samples were collected, and compatibility testing was done using CTT and Gel card Method. Donors samples were collected from the pilot tubes of the blood donations from healthy donors with >45 kg and negative serology of HIV, HbsAg, HCV, VDRL and Malaria. Patient samples were received from the ward with the blood requisition form. For cross-matching following steps were followed, Blood grouping of patient blood and donor blood with the help of antisera A, B, D. After matching of blood group we proceeded to perform cross-matching of the blood of both donor and patient’s blood by two methods. Spin saline tube method without AHG and with AHG reagent, the second method is the Matrix Gel card method. In CTT without AHG 1 drop of 2–5% suspension of red donor cells was mixed with two drops of recipient plasma. The mixture was incubated for 30 min at 37°C and washed three times with 0.9% saline, followed by centrifugation at 1000 rpm for 1 min and observed for agglutination. In CTT with AHG major Indirect Antiglobulin Tests (IAT), cross-match on the same samples was performed using polyspecific antiglobulin reagent. After mixing of patient’s serum and red cells of the donor and kept in incubation for one hour at 37°C, one drop of antiglobulin reagent was added, then centrifuged, and observed for agglutination. The strength of positive (incompatible) reactions was recorded. In Gel Card technique we used Matrix Gel Card incorporated with AHG reagent (each plastic card containing six microtubes), ID incubator, ID centrifuge of Gel Card, Diluent- low ionic salt solution (LISS), test tubes and micropipettes. Firstly a 0.8% donor red cell suspension was prepared by adding 10µl of packed red cells of the donor in 1ml LISS in a clean test tube. 50µl of 0.8% donor red cell suspension was added to the microtube, followed by 25 µl of the patient’s serum. The card was incubated at 37°C for 15 minutes, then centrifuged in ID centrifuge for 10 minutes at preset/ 950 rpm, at the end of centrifuge result was read. If gel card shows, pellets of RBCs settled at the bottom of particular microtube means no agglutination or negative reaction indicating that donors blood is compatible with the recipient and suitable for transfusion. The presence of agglutination means positive reaction, which indicates Incompatibility. A positive result shows grading into 1+ to 4. In 4+ reaction, indicated if a solid band of red blood cells (RBCs) is on top of the gel card’s microtube, 3+ reaction displays if agglutination of RBCs in the upper half, 2+ reaction is indicated by RBCs agglutinate dispersed throughout the microtube. In contrast, a 1+ reaction shows if RBCs aggregate in the main lower half part of the microtube with a dotted structure in the column.

RESULTS AND DISCUSSION

In this study, 500 blood samples were cross-matched by using Spin saline tube method with and without using AHG and Matrix Gel Card. We compared both methods of cross-matching for sensitivity & specificity, the accuracy of results, and time is taken. 497 (99.4%) samples were compatible, and 03 (0.6%) samples were incompatible in Gel card method. In CTT 492 (98.4%) samples were compatible, and 05 (1.6%) were false positive (FP). Incompatibility of 05 samples (FP) disappeared after incubation with AHG reagent at 37°C saline tube method. The sensitivity and specificity of both gel card method and saline tube method
Table 1: Observations

<table>
<thead>
<tr>
<th>Method used</th>
<th>Samples</th>
<th>Compatible</th>
<th>Incompatible</th>
</tr>
</thead>
<tbody>
<tr>
<td>Spin saline tube method without AHG (RT)</td>
<td>500</td>
<td>TN FP</td>
<td>FN</td>
</tr>
<tr>
<td>Spin saline tube method with AHG (37°C)</td>
<td>500</td>
<td>49205</td>
<td>03 00</td>
</tr>
<tr>
<td>Matrix Gel Card (37°C)</td>
<td>500</td>
<td>497 00</td>
<td>03 00</td>
</tr>
</tbody>
</table>

Table 2: Comparison of sensitivity, specificity and accuracy of various methods

<table>
<thead>
<tr>
<th>Method used</th>
<th>Samples</th>
<th>Sensitivity(%)</th>
<th>Specificity(%)</th>
<th>Accuracy(%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Spin saline tube method without AHG (RT)</td>
<td>500</td>
<td>100</td>
<td>98.9</td>
<td>99</td>
</tr>
<tr>
<td>Spin saline tube method with AHG (37°C)</td>
<td>500</td>
<td>100</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>Matrix Gel Card (37°C)</td>
<td>500</td>
<td>100</td>
<td>100</td>
<td>100</td>
</tr>
</tbody>
</table>

with AHG methods is 100% whereas the specificity of saline tube method without AHG is 98.9% (Table 2). The average time taken by Gel card method was 20-30 minutes for a single compatibility test whereas in conventional spin tube method with the use of AHG (IAT) average time required was 90 minutes and without AHG it was 45-60 minutes. The results and comparative sensitivities of various methods are shown in Tables 1 and 2, respectively.

CTT is the most commonly used method for pre-transfusion compatibility testing, but it is associated with certain disadvantages like being laborious and time-consuming method. These can be overcome by a gel card method. The gel card technology was developed by (Lapierre, 1988). The basic principle of the gel test is that instead of a test tube, the serum and cell reaction takes place in a microtube. Six of such microtubes filled with specific gel medium are embedded in a plastic card to allow ease of handling, testing, reading and disposal. In our study, 500 blood samples were cross-matched by using Spin saline tube method with and without using AHG and Matrix Gel Card. The sensitivity and specificity of both gel card method and saline tube method with AHG methods are 100% respectively, whereas the specificity of saline tube method without AHG is 98.9%. The average time taken by Gel card method was 20-30 minutes for a single compatibility test whereas in conventional spin tube method with the use of AHG (IAT) average time required was 90 minutes and without AHG it was 45-60 minutes. Thus the gel card method has advantages over CTT. These findings are similar to the study done by (Kaur et al., 2003). They reported that the Diamed gel card system is easy to use and more sensitive than the conventional tube agglutination technique. Also, studies done by reported that gel card method is better than traditional spin tube method because it is easy to perform, results are stable, dispensation of controls, no need of wash phase with comparable sensitivity and specificity which is in agreement with our study. In our study, 492 (98.4%) samples showed compatibility, and 4 were false positive (FP) by CTT method, and no false positives were reported in gel card method. (Bromilow et al., 1991), in their study, said that ID gel system has fewer chances of false positive or false negative results because of its simplicity, sensitivity, a less time-consuming method for detection of antigen-antibody reaction and with the absence of wash phase, thus reducing the possibility of elution of weakly bound antibodies from red blood cells. Our study showed that results of gel card test and tube method with AHG are comparable to each other which is in agreement to the research done by (Rumsey and Ciesielski, 2000), stating that gel test is at least as sensitive as an IAT tube test, with a better balance of sensitivity and specificity. The advantages of matrix gel card are that it gives a reliable, reproducible and easy interpretation of results from microtube, which can be recorded for a long time and easy disposal by incineration (Col et al., 2008), (Malyska et al., 1993). Another study by (Nathalang and Kuvanont, 1993) concluded that Matrix gel card system is better than Spin saline test tube method and simple to perform with less exposure of blood bank personal to blood with reduced risk of HIV, HbsAg and HCV infections. (Prakash and M, 2006) in their study proposed that matrix gel card test is a better alternative to the Spin saline
tube test for blood cross-matching as well as coombs tests (Direct and Indirect). In the present study, it is observed that the gel card method is less time-consuming as compared to CTT, which is an advantage for blood transfusion in life-saving cases and has sensitivity and specificity of 100%. But the only limitation is that the gel card method is not cost-effective, and there is a requirement of separate incubator and centrifuge.

CONCLUSIONS
The results of cross-matching by both methods are comparable. Gel card method is still preferable over conventional spin tube method, because it takes only 15-20 minutes, simple to perform, results can be preserved, absence of wash phase whereas tube method of cross-matching is a laborious and a time-consuming process. It depends on the technician’s techniques, and there is a risk of observer variations. We recommend its usage as a standard procedure for routine compatibility testing in the transfusion department of all hospitals.

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Conflicts of interest
The authors declare that they have no conflict of interest for this study.

REFERENCES