Evaluation of Methodology and Comparative Study between Micro Typing System Gel Card and Conventional Tube Techniques for Cross Matching in a Tertiary Care Centre

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Abstract: _Introduction:_ _Gel card technique is one of the latest technique emerged and due to its high sensitivity and accurate results, it is considered as a good technique for cross matching. The aim of this study is to compare sensitivity of Gel card and conventional saline tube method for cross matching in blood bank of tertiary care centre._ _Method:_ _Comparative study between 600 samples received in the blood bank of tertiary care centre was done to evaluate the efficacy and sensitivity of gel card method and test tube method with and without AHG._ _Result: 600 samples were taken for study, out of which 597 cases were compatible and 3 cases were incompatible with gel card and saline tube with AHG, but all 600 cases came compatible with saline tube method without AHG. This shows specificity of gel card and saline tube method with AHG is 100% and of saline tube without AHG is 99.5%._ _Conclusion: Gel method is rapid procedure as use of controls is not needed. Wash phase was not required in indirect antiglobulin test and sensitivity was also compareable with conventional tube method. So we can conclude that gel card technique can be considered as a better alternative for conventional tube method._

Keywords: Gel card, saline tube, AHG, cross match

1. Introduction

In 1901 ABO system was discovered by Landsteiner and in 1945, antiglobulin test was done first time, but since then researchers are trying to develop more specific serological methods to avoid ABO and Rh incompatibility between blood of donor and recipient[2] , and to find out if any antibody is present in recipient's serum which can react with donor's red cell and can cause serious compilcation after blood transfusion[11].

The purpose of the cross match is to select blood component that will have acceptable survival when transfused and will not cause harm to the recipient. Compatibility testing is done to ensure safe transfusion therapy[10]. Cross matching is an integral part of routine pretransfusion testing. It is done to prevent the incompatible red cell transfusion which may result in immune-mediated hemolytic transfusion reaction[1].

The terms compatibility test and cross-matching are sometimes used interchangeably, it is a part of compatibility test, a cross-match is carried out to ensure there are no antibodies are present in patient's serum that will react with donor cells when transfused[4]. Even if the blood groups of patient and donor are known, it is necessary to perform a cross-match as the final serological test of compatibility as this will also show if any mistakes have been made in the ABO grouping of the patient or donor, remember that it is ABO incompatibility between the patient's plasma and donor red cells that causes fatal hemolytic transfusion reactions.

Whenever possible, an indirect antiglobulin test should be used for crossmatch[6].

The gel card method introduced by Lappiere et al is used for cross matching of blood along with saline tube method. The gel card is a reliable and advantageous method and is suitable in routine use for detection and identification of alloantibodies in a community hospital transfusion service laboratory[14]. The tube technique has been the cornerstone of compatibility testing over last 40 years, but the enhanced sensitivity of the gel card technique has made the interpretation of the tests more objective[15]

Sephadex gel is used in gel cards which holds agglutinate in semisolid medium, this helps in clear visualization of agglutinationthen that of the tube method 5. When RBCs are added in a gel card,this gel acts as a trap, RBCs which agglutinates are seen trapped in gel at bottom of the tube, this agglutination can be seen for hours. For easy handling, reading and testing there are 6 mcro tubes in a single Gel card 2.

Our aim of this study is to compare the accuracy and sensitivity of gel card technique(LISS/COOMBS) and saline tube method, assess the compatibility test by gel card and saline tube method with coomb's and without coomb's test.

2. Material and Method

This is a prospective study in which 600 samples were included which were referred to blood bank of tertiary care

centre for cross matching. All samples were cross matched by two methods , first by conventional spin tube method with AHG and without it and second by Gel card system (ID-Card "LISS/Coombs micro typing system containing specific antihuman globulin with Anti IgG, C3d activity, manufactured by Bio-Red diagnostics P Ltd. The following material and reagent were used: centrifuge, incubator, test tubes, slides, ABO-Rh reagent, Coombs sera, ID-Card "LISS/ Coombs", normal saline. All donor and recipients samples and blood bags were first checked for their blood groups by anti sera A, B, D. After matching of blood groups we proceeds to cross match.

Cross match test is divided into two parts i.e Major cross match (mixing of donor's red cells with recipient's serum) and Minor cross match (mixing of donor's plasma with recipient's red cells).

Saline tube technique is done both for IgM and IgG antibodies. Patient serum or plasma and reagent red cells are combined, then centrifuged, and observed for agglutination.

Tube method without AHG is done by adding 2 drops of patient's serum in tube , add 1 drop of 2-4% saline suspended red cells of donor, mix and incubate for 5-10 minutes , centrifuge at 1000 rpm for 1 minute and see for agglutination. Cells are washed 3-4 times to remove unbound antibodies. Agglutination indicates positive results (incompatibility). For Indirect antiglobulin test AHG (anti human globulin) is added then incubate tube at 37^0 c for 45-60 minutes and then centrifuge at 1000 rpm and see for agglutination. Check cells are used as control for this test. Check cells are IgG coated cells which reacts with AHG in the tube. If check cells are negative , then it means procedure is not done correctly and we should repeat the procedure[14] .

For gel card technique ID-Card "LISS/Coombs included with AHG reagent and C3d (each plastic card contain 6 micro tube), incubator, card centrifuge , diluent-2 LISS, test tubes and micropipette used. 0.8% red cell suspension of donor's red cells was prepared in a test tube.50 µl of this suspension was then added in to micro tube of gel card followed by 25 µl of patient's serum.

The card was incubated for 15 minutes at 37^0 c, then centrifuge in card centrifuge and result was read.
No agglutination:- compatible
Agglutination:- incompatible
Grading of positive result
Grade 4:- indicated by solid band of red blood cells on the top of gel.
Grade 3:- indicated by agglutinated RBCs in the upper half of gel.
Grade 2:- indicated by RBC agglutinates dispersed throughout the column.
Garde 1:-. Indicated by RBCs aggregation in mainly lower half of the column.

3. Result

This study is carried out in the blood bank of tertiary care hospital during the period from 1 February 2019 to 31 July 2019. A total number of 600 blood units were cross matched with 400 patient samples requesting blood. All samples were evaluated by gel card technique and saline tube method (RT and 37^0c).
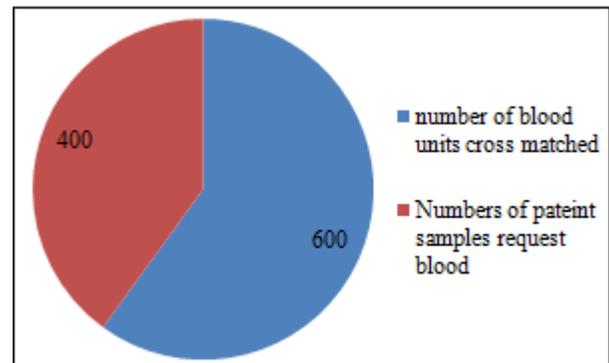

Figure 1: Numbers of blood units cross matched and the number of patient samples requesting blood
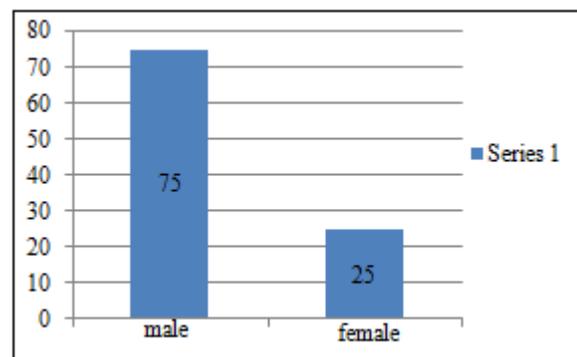

Figure 2: number of patient samples requesting blood based on sex

Blood requests were more for males (75%) then females (25%) i.e. 450 males and 150 females in patient sample requesting blood.(Figure 2)

The highest number of blood group , cross matched was blood group B+(56.7%) and least AB-(0.33) as depicted in **Table 1**.

Table 1: Cases studies based on blood group

Blood group type	Number of samples	Percentage %
B+	339	56.5
O+	123	20.5
A+	55	9.16
AB+	50	8.33
A-	10	1.66
O-	12	02
B-	09	1.52
AB-	02	0.33
Total	600	100%

600 samples were tested in this study , 597 samples (99.5%) showed compatibility and 3 samples (0.5%) showed incompatibility with Gel card technique but all 600 samples(100%) shows compatibility with tube method without AHG , When AHG was added in tube , incubated for 60 minutes at 37 ^{0} c then results were similar as that of gel card method **(Table2)** .This shows that sensitivity and specificity of both test is 100% if use of AHG is included in all test performed by tube method, otherwise the specificity of tube method is 99.5%. positive predictive value is same (100%) for both methods i.e. Gel card and tube method including AHG, but only 99.5% if AHG is not included.

Cross matching can be done within 15-20 minutes by Gel card while it takes 90 minutes if done by conventional spin tube method including AHG and 20-30 minutes if AHG is not included.

Table 2: Result of technique used

Technique used	Number of samples	Negative (compatible)	Positive (Incompatible)
Gel Card Method	600	597	3
Saline tube at RT	600	600	00
Saline tube at 37^0C	600	597	3

4. Discussion

In our study 0.5% samples, which showed agglutination by gel card technique and not by the routine spin tube method, when subjected to 60 minutes incubation at 37°C followed by IAT, showed comparable results. Our findings are in agreement with other studies kaur R et al[9], Novaretti MCZ et al [12] , Bromilow IM et al[15].

It also concluded that matrix gel card test is better alternative to the Spin saline tube test for blood cross- matching as well as coombs tests (Direct and Indirect) (Jai prakash et al., 2006).

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Our study is similar to Jai prakash et al [7] which showed that gel card is better alternative for cross matching and direct and indirect coomb's test .Cate et al[14] found gel system appropriate for detection and identification of antibodies. Our study showed sensitivity and specificity of gel card is 99.5% and that of tube method without AHG is 100% , which is similar to study conducted in south Carolina by John et al [14] which showed sensitivity of gel card 95% and saline tube 99.1% respectively.

Bromilow et al [16] showed in their study that when IT is done by gel method , serum to cell ratio imcreases and as wash phase is not needed in gel method so weakly bound antibodies will not be eluded and possibility of false positive or false negative results reduces.

Rumsey et al [13] concluded that gel test is at least as sensitive as an LISS IAT tube test, with a better balance of sensitivity and specificity which is in agreement with our study.

5. Conclusion

Our study showed that gel card is easier to use and the factors affecting the results are less in it, time consumption is also less in gel card. The advantage of gel card is an easy reading of microtube, handling and disposal. Results of Gel card can be preserved for 3-4 days unlike tube method where results cannot be preserved. Gel card assay appears to be an excellent method for detecting agglutination better than saline tube method and easy to read weak agglutination and it can also detect ABO incompatibility. The performance of saline tube technique requires more experience and highly accuracy due to its long stages and multiple washing. But one disadvantage of gel card method is that gel cards are costly and require separate incubator and centrifuge. To increase efficacy and test results, blood banks should introduce antibody screening of donors and recipients within the routine screening time and students who are studying should be given more training so that they should be knowing gel card method and how to perform. More courses and seminars for blood bank personnel should be done in order to follow new ideas and innovations in the blood bank.

6. Conflict of interest

None

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None

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