The anti-D Rh1 (IgM) monoclonal antibodies are in vitro culture

"Anti D Rh, (IgM), monoclonal antibodies for Rh typing

Test done for Rh typing

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METHOD AND MATERIALS:
The study has been taken place in Blood Bank in the Department of Pathology at Saveetha Medical College.

PREVALENCE OF RHD ANTIGEN IN MEDICAL STUDENTS

INTRODUCTION:
Rh blood group system was discovered in 1940 by Karl Landsteiner and Wiener, by immunizing guinea pigs & rabbits with Rhesus monkey blood. Guinea pigs & rabbits produced an antibody that agglutinated 85% of human blood. The "big five" C, D, E, c and e make up 99% of all problems involving Rh system. Antibodies of Rh system are not naturally occurring, they develop by exposure via transfusion. Terms Rh positive and Rh negative refer only to the presence or absence of one antigen i.e Rh D.

Till now about 400 red cells antigen has been identified, the majority of which are inherited by Mendelian fashion.

ABO blood group system was 1 st and Rh blood group system was the 4 th one to be identified. Both of these are most important for blood transfusion purposes.

After ABO blood group system, Rh blood group system is the important blood group system. In 1940 Levine and stenson in a separate study also discovered the Rh system and they divided the system into Rh positive & Rh Negative depending on presence or absence of agglutination of RBC’s with Rh D antisera.

Rh is the most complex and polymorphic blood group system and has major importance in transfusion medicine. The administration of ABO & Rh compatible blood /blood components is primordial for the immunological safety of blood during blood transfusion. ABO & Rhesus (Rh) blood group antigens are hereditary characters and are useful in population genetic studies, in resolving medico legal issues and more importantly in compatibility test in blood transfuse practice.

In 1945 four more types of Rh antigen were discovered. These antithetical antigen C/c and E/e respectively. Presently as many as 50 antigens are recognized in the Rh blood group system.

METHODOLOGY:
Antigens are proteins on the body cells that can cause a response from the immune system. The Rh factor is a type of protein on the surface of the red blood cells. Most people who have the Rh factor are Rh positive. Those who do not have Rh factor are Rh negative.

If the D antigen is present the cells will be sensitized with the anti D contained in the D tube. To determine whether this has occurred. The cells are washed and anti-human globulin is added. If the cells have been sensitized, agglutination, will occur, indicating that the individual is D positive.

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KEYWORDS:

supernatant of hybrids obtained by cellular fusion i.e. hybridoma technology. The anti-D Rh1 (IgM) monoclonal antibodies has following features:

• It agglutinate in saline solution
• Active at room temperature
• Can be used on glass slide as well as in tube.

PRINCIPLE
"Human red blood cells possessing D antigen will agglutinate in the presence of corresponding antibody".

Agglutination of red cells with anti-D Rh (IgM) monoclonal antibodies indicates the presence of D-antigen and hence Rh positive result. It can detect D antigen (Rh1) and high grade D antigen (weaker variant of antigen D).

In case no agglutination has obtained with anti D Rh1 (IgM) monoclonal antibodies, the cells were then checked for the presence of low grade D’ antigen by Coomb's test using anti-D (IgG) and anti human globulin serum (Coomb's reagent). Collection of blood sample and preservation of antiseras are same as that of anti A and anti B antisera.

PROCEDURE
• Prepare a 3–4% suspension of red cells washed in isotonic saline solution.
• Put one drop of anti-D Rh, (IgM) monoclonal antibodies reagent and one drop of 3–4% cells suspension respectively in the tube.
• Shake to homogenise antibodies and red cells suspension, then centrifuged for one min at 1000 rpm.
• The reaction was observed macroscopically by shaking gently the tube so as to loosen the cells pellet.
• If the red cells separate in one or more clumps, the reaction is positive, if the red cells return to a homogenous suspension, the reaction is negative or D’ positive.
• The agglutination is also checked microscopically if the reaction appears to be negative.

• It is a time taking process and the results are calculated after the samples are tested equally and Rh positivity and negativity are shown.
• We can conclude that mostly there is high number of Rh positivity and less number of Rh negativity.
• The Rh factor, Rh positive and Rh negative refer to the RhD antigen present in the body.

PIE CHART:

97%

3%
• The 97% of the sky blue color part in the pie chart indicates the positive presence of RhD antigen in the results obtained by collecting the samples.
• The 3% of the dark blue color part in the pie chart shows the negative presence of RhD antigen in the results obtained.

RESULTS:
Out of 50 samples studied, the incidence of RhD was positive when compared to negative. All the test results show 97% of the positivity of RhD antigen and only 3% of the results indicates the negativity. When we draw a pie chart for the results obtained then we can easily know that the presence of RhD antigen.

CONCLUSION:
Like previous studies, our study also concluded that there is a wide range of racial and geographical variation in the distribution of Rh phenotype and genotype. The Rh blood group system has vital role in population genetic study, in resolving medico legal issues and more importantly in transfusion practice. It also shows the more number of Rh positivity and less amount of Rh negativity.

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