

<b>Name</b> : MD AMZAD HOSSAIN	<b>Age</b> : 41 Years
<b>Lab No.</b> : 430431996	<b>Gender</b> : Male
<b>Ref By</b> : DR. ZULFIQUAR SERAJI	<b>Reported</b> : 15/4/2023 5:59:35PM
<b>Collected</b> : 11/4/2023 10:09:00AM	<b>Report Status</b> : Final
<b>A/c Status</b> : P	<b>Processed at</b> : LPL-NATIONAL REFERENCE LAB
<b>Collected at</b> : WALK IN DHAKA-GULSHAN Ta-517(New), Ta-99(Old), Gulshan Badda Link Road, Gulshan, Dhaka-1212	<b>National Reference laboratory, Block E, Sector 18, Rohini, New Delhi -110085</b>



**Test Report**

<b>Name of Donor</b> : MD JABEL PRAMANIK	<b>Donor Lab No.</b> : 430431997	<b>Age</b> : 33 Years	<b>Gender</b> : Male
<b>Relation</b> : Cousin brother	<b>Diagnosis</b> : Not mentioned		

**T AND B CELL FLOW CYTOMETRY CROSS MATCH (FCXM)\*\***  
 (Flow Cytometry)

PARAMETER	Ratio	Result	Reference Ratio
Autologous T Cell FCXM	1.1	Negative	1.1
Autologous B Cell FCXM	1.3	Negative	1.3
Patient T Cell FCXM	1.1	Negative	1.1
Patient B Cell FCXM	11.4	Positive	1.3

**Impression**

Flow cytometric cross match shows negative reaction with donor T lymphocytes and positive reaction with donor B lymphocytes. Results must be considered in conjunction with other laboratory testing and clinical risk factors prior to transplant.

**Interpretation**

The criteria defined for interpretation of cutoff values is specific to Dr Lal PathLabs, based on the reference given in the ASHI Manual, Volume 2. The ratio is derived using the negative control median channel values and the patient median channel values.

RESULT	RATIO	REMARKS
T Cells	≤1.1	Negative
T Cells	>1.1	Positive
T cells	1.2 to 1.5	weak Positive
B cells	≤1.3	Negative
B cells	>1.3	Positive
B cells	1.4 to 1.7	weak Positive

**Test Information**

<b>INSTRUMENT</b>	FACSLyric Flow cytometer
<b>SOFTWARE</b>	FACSuite 1.5
<b>METHOD</b>	Immunophenotyping by Flow cytometry (mononuclear cell)



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### Test Report

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<b>Relation</b> : Cousin brother	<b>Diagnosis</b> : Not mentioned		
----- separation, wash and stain) -----			
GATING STRATEGY	CD45 vs. SSC & CD3 vs. CD19		
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- Note:**
1. Test performed on serum & whole blood
  2. The FCXM can independently evaluate T and B cell activity in mixed cell populations.

#### Comment

The test detects both complement and non-complement binding anti HLA class I and II IgG antibodies. A positive flow cross match suggests increased risk of allograft rejection but is not a contraindication for transplantation. Patients undergoing antibody modulation by means of IVlg therapy and having recent treatment by antibody based immunosuppressant drugs may present with a false positive flow cytometric cross match while lacking donor specific HLA antibodies.

#### References

1. Charles W Hamrick and Lauralynn K Lebeck. Flow cytometric T and B cell crossmatching. ASHI manual VI.B.4.
2. Bray RA, Flow cytometry in the transplant laboratory. Annls. N.Y. Acad.Sci.677:138,1993.
3. Garovoy Mr, Rheinschimt MA, Bigos M, et.al., Flow cytometry analysis: a high technology crossmatch technique facilitating transplantation. Transplantation procesding 15:1939,1983.



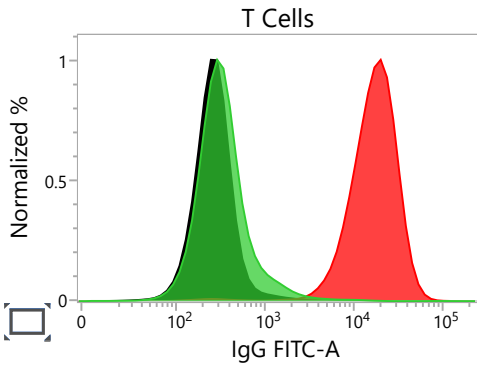
Sample ID: 430431996+997

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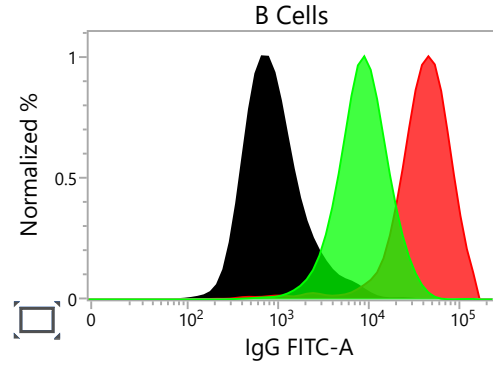
**430431996+997:Patient RunPointerStatistics**

Acquisition Date (Patient): 4/14/2023

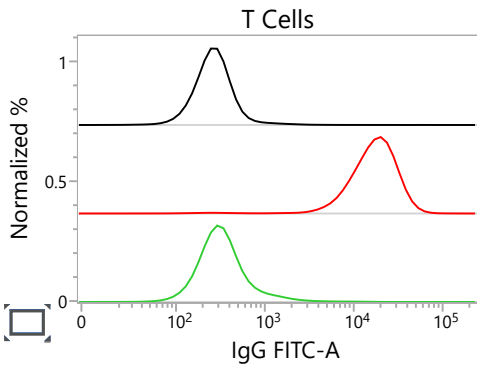
Name	All Events	Lymphs	T Cells	B Cells
Events	22,205	22,143	18,581	1,507
% Total	100.00	99.72	83.68	6.79



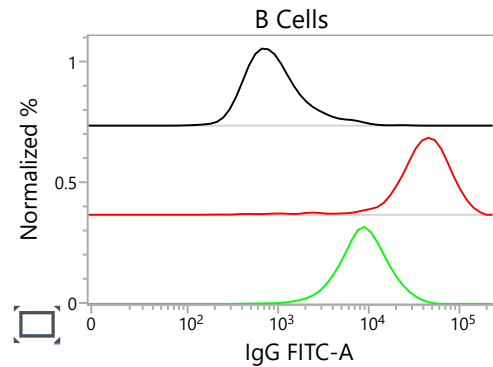
Negative Control-T Cells (PDS) IgG FITC-A —  
 Positive Control-T Cells IgG FITC-A —  
 Patient-T Cells IgG FITC-A —



Negative Control-B Cells (PDS) IgG FITC-A —  
 Positive Control-B Cells IgG FITC-A —  
 Patient-B Cells IgG FITC-A —



Negative Control-T Cells (PDS) IgG FITC-A —  
 Positive Control-T Cells IgG FITC-A —  
 Patient-T Cells IgG FITC-A —



Negative Control-B Cells (PDS) IgG FITC-A —  
 Positive Control-B Cells IgG FITC-A —  
 Patient-B Cells IgG FITC-A —

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Sector 18, Rohini, New Delhi -110085


### Test Report

Test Name	Results	Units	Bio. Ref. Interval
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**HLA T & B CELL LYMPHOCYTE CROSS MATCH  
AHG (ANTI HUMAN GLOBULIN) AUGMENTED**  
(Complement Dependent Cytotoxicity with DTT  
treatment)



Dr (Prof) Jasmeet Kaur  
MD, Pathology; PhD Transplant  
Immunology & Immunogenetics  
Technical Director - Advanced  
Histocompatibility &  
Immunogenetics  
NRL - Dr Lal PathLabs Ltd



Dr Beena Chandrasekhar  
PhD, Life Sciences  
Technical Director - Flowcytometry  
NRL - Dr Lal PathLabs Ltd

-----End of report-----



#### IMPORTANT INSTRUCTIONS

•Test results released pertain to the specimen submitted. •All test results are dependent on the quality of the sample received by the Laboratory.  
•Laboratory investigations are only a tool to facilitate in arriving at a diagnosis and should be clinically correlated by the Referring Physician. •Report  
delivery may be delayed due to unforeseen circumstances. Inconvenience is regretted. •Certain tests may require further testing at additional cost for  
derivation of exact value. Kindly submit request within 72 hours post reporting. •Test results may show interlaboratory variations. •The Courts/Forum  
at shall have exclusive jurisdiction in all disputes/claims concerning the test(s) & or results of test(s). •Test results are not valid for medico legal  
purposes. •This is computer generated medical diagnostic report that has been validated by Authorized Medical Practitioner/Doctor. •The report does  
not need physical signature.

(#) Sample drawn from outside source.

If Test results are alarming or unexpected, client is advised to contact the Customer Care immediately for possible remedial action.

Tel: +91-11-49885050, Fax: - +91-11-2788-2134, E-mail: lalpathlabs@lalpathlabs.com

**National Reference lab, Delhi, a CAP (7171001) Accredited, ISO 9001:2015 (FS60411) & ISO 27001:2013 (616691) Certified laboratory.**



**Name of Patient** : Mr. Md Amzad Hossain      **Age** : 41 Yrs.      **Gender** : Male  
**Name of Donor** : Mr. Md Jabel Pramanik      **Age** : 33 Yrs.      **Gender** : Male  
**Referred by** : Dr. Zulfiqar Seraji      **Collection date** : 11.04.2023  
**Lab No** : 23 430461996/997      **Reporting Date** : 15.04.2023  
**Report at** : WALK IN DHAKA GULSHAN      **Diagnosis** : Not Mentioned  
**Claimed Relationship of the Donor with the Recipient:** Cousin Brother

## T AND B CELL CROSS MATCH REPORT

(Complement Dependent Cytotoxicity Report with DTT Treatment and AHG Augmentation)

### Auto Cross Match (Recipient cells and Recipient Serum)

IgM Antibodies      Negative

	CDC with DTT treatment and AHG Augmentation		Reference Range (ASHI Guidelines)
	At 4°C (% dead cells)	At 37°C (% dead cells)	
T Cell Cross Match	10	10	<20 (%dead cells)
B Cell Cross Match	10	10	

### Final Result:

#### T-Cell Crossmatch:

CDC T-cell Crossmatch with DTT Treatment and AHG Augmentation is Negative at both 4°C and 37°C.

#### B-Cell Crossmatch:

CDC B-cell Crossmatch with DTT Treatment and AHG Augmentation is Negative at both 4°C and 37°C.



RECIPIENT



DONOR

Prof. DR. JASMEET KAUR

MD (Path), PhD (Transplant - Immunology & Immunogenetics)  
Director (Tech.) Dept. of Histocompatibility & Transplant Immunology

### Interpretation Guide:

Interpretation	% dead cells	Score
Not readable		0
Negative	0-10	1
Doubtful negative	11-20	2
Weak positive	21-50	4
Positive	51-80	6
Strongly positive	81-100	8

Score reactions are estimated by calculating the percentage of cell death beyond the negative control. Results recorded as per ASHI standards (ASHI Manual 4<sup>th</sup> Edition- The basic lymphocyte Micro cytotoxicity tests Standard and AHG Enhancement)

**Information about test:**

- 1) CDC with DTT Treatment
  - i) Inactivation of autoantibodies may be critical to the accuracy of crossmatches and antibody screens ii) Antibodies of either IgG or IgM isotype can bind cells and activate the complement pathway resulting in cell lysis iii) Antibodies in transplant can be alloantibodies or autoantibodies.
    - (a) Alloantibodies are those antibodies directed against HLA antigens on the cells of others formed in response to sensitization by transplant, transfusion or pregnancy and are typically (but not always) IgG isotype
    - (b) Autoantibodies are those directed against self-antigens and are typically IgM isotype
    - (c) In spite of lymphocytotoxic activity of IgM autoantibodies are not believed to be damaging to transplanted organs
  - iv) Autoantibodies are detectable in antibodies screen and crossmatches yet irrelevant to solid organ transplant outcome v) DTT treatment is typically done for inactivation of the IgM antibodies to distinguish them from IgG alloantibodies
- 2) CDC with AHG Augmentation
  - i) The antiglobulin variation of the basic micro cytotoxicity test is used only for crossmatch and antibody screening.
  - ii) Where antigen-antibody reactions occur, the conformational change exposing the site on Fc portion of antibody requires that both Fab portions of the antibody molecule are bound to antigen.
  - iii) Complement activation requires binding of complement to 2Fc regions in close proximity to each other. If there is insufficient antibody, Fc regions adjacent antibody molecules may be too far apart.
  - iv) Also, not all Ig classes activate complement with equal efficiency
  - v) The binding of the antiglobulin to the antibody previously attached to the cellular antigens gives the complement a large number of binding sites and can enhance the strength of the reactions. This enhancement lowers the amount of antibody necessary in a sample for cytotoxic detection.
  - vi) AHG is used in crossmatch or antibody screening assays to enhance the sensitivity i.e. to detect low levels of anti HLA antibody and non- complement binding antibodies.
- 3) Incubation Temperature: The temperature of incubation of cells and serum can be selected to elicit a desired degree of reactivity in the lymphocytotoxicity test. Temperatures of 4° C, Room temperature 20-25° C, and 37° C are most often used. 37° C is the most sensitive temperature.
- 4) Donor lymphocytes showing more than 20% cell death over the background constitutes a positive cross match.
- 5) Donor lymphocytes negative with negative control but positive in any dilution constitutes a positive cross match.
- 6) The test is valid for 72 hours only, and ideally Lymphocyte cross match must be repeated if patient is exposed to a sensitizing event like blood transfusion, infections, pregnancy etc.

**Note:** Test conducted on serum and ACD whole blood.

\* This test has been developed and validated at National Reference Lab, Dr. Lal PathLabs, New Delhi.