Michigan Blood
MI blood saves lives.
PART OF VERSITI™

Case Studies and Working in an Immunohematology Reference Laboratory (IRL)

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IRL - Manager
Michigan Blood

Job Description

Detectives

Michigan Blood
 MI blood saves lives.
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Staff Training/Education/Locations

- Most are med techs with blood bank experience of 1 or more years
- Often BB or SBB certified
- An appreciation/desire to be challenged by antigen/antibody problems
- Need to think outside the box – antibodies don’t read textbooks
- Willing to relocate? Most IRL’s are located in metropolitan areas that have Blood Centers or in large teaching hospitals

Donor Testing

- Resolve ABO/Rh discrepancies
  - Most common are A subgroups containing anti-A1
  - A cold reactive antibody ( -M, -Lea or anti-PP, Pk) affect the reverse grouping
  - Rarely there are incorrect entries at registration where the ABO/Rh does not match the history at all

- Antigen testing
  - IRL’s "bread and butter", our biggest source of revenue
  - Consumes most of lab "downtime" (no samples)
  - O+, O- (our "Universal donor), A+ and some A-

Donor Testing

- Antigen Testing
  - Start with the 4 Rh (C, E, c, e)
    - In the Rh+'s looking for R1's (E-c-); R2's (C-e-) Ro's (C-E-)
    - In the Rh-'s looking for rr's (C-E-)
  - Then K, Fy?, Fy*, Jk*, Jk, S and s
  - Molecular genotyping (performed at BloodCenter of Wisconsin – 36 nucleotide polymorphisms per donor
  - over 74,000 antigens entered in 2016
Patient Testing

- SOPs – Michigan Blood IRL staff use over 40 different SOPs to resolve difficult serologic donor and patient problems
- Antibody detection methods range from the most basic (saline indirect antiglobulin test) to the highly sensitive and specific Solid Phase (SP) and gel technologies
- Special chemicals for treating red cells such as enzymes and dithiothreitol (DTT)
- Adsorption (autologous and allogeneic) and elution techniques

Patient Testing

- Rare reagent red cells and antisera (collected in-house over the years or shared between IRL’s across the US and the world (e.g. S.C.A.R.F.)
- Molecular testing – not performed at Michigan Blood but it is available to us from our Versiti partner at the Blood Center of Wisconsin
- Grand Rapids/Livonia IRLs rotate 3 staff members on call 24/7, 365 days a year
- MD available via telephone for consultation
- American Rare Donor (ARDP) member

Versiti

- Members include Heartland Blood Center, Indiana Blood Center and the Blood Center of Wisconsin
- Allows us a greater ability to share resources (red cells/plasma/platelets)
- Increases our ability to locate antigen negative red cells for our difficult serologic patients
- Assist with antibody resolution – each site has a unique collection of rare antisera and red cells
CASE STUDIES - IRL

Case Study #1 – A.T.

- 32 year old female
- Currently pregnant
- 2 previous pregnancies (1-full term, 1-miscarriage)
- No known transfusions
- Referring hospital observed positive panel with negative autocontrol

Initial Screen – A.T.
Patient A.T.

- Negative cell is Fy(a-b-) and Lu(a+b-)
- Chose some additional cells based on that finding
Patient A.T.

- Probable anti-Lu^b^b
- Molecular genotyping to confirm patient is Lu^-b^b
- Complete rule outs - use more Lu^- cells or allogeneic adsorptions

Michigan Blood Rule Out Rule:
Require at least one homozygous expression of antigen and a minimum of one heterozygous (ex. to rule out anti-Fya need one Fy(a+b-) and one Fy(a+b+)

Alloadsorbed Panel - A.T.

Patient A.T. - Conclusion

- Anti-Lu^b^ only
- >99% random population
- Can cause mild to moderate hemolytic transfusion reactions
- Very moderate cause of hemolytic disease of the fetus and newborn (Why? – Lutheran antigens poorly developed on cord cells and placental tissues may adsorb antibody)
A.T. Prenatal Care and Transfusion Recommendations

- Monitor anti-Lu<sup>b</sup> as any other red cell antibody with intervention as needed
- Locate a source of red cells that are Lu<sup>b</sup>- for delivery
  - Test patient’s siblings as this is an inherited trait
  - Approach the ARDP for assistance
- Future Needs Post Delivery
  - Consider autologous donation – Red cells may be stored for up to 10 years in glycerol

Case Study #2 – J.H.

- 96 year old male
- Diagnosis: anemia, dizziness
- Hgb: 5.5 g/dL
- Not transfused in the last 3 months
- Referring hospital observed non-specific reactivity and a positive autocontrol

Initial Screen – J.H.
Initial Panel – J.H.

<table>
<thead>
<tr>
<th>Patient</th>
<th>ABO</th>
<th>Rh</th>
<th>Rh Phenotype</th>
<th>DAT</th>
<th>Anti-IgG</th>
<th>Anti-C3</th>
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<tbody>
<tr>
<td>J.H.</td>
<td>O</td>
<td>-</td>
<td>C-E-c+e+</td>
<td>2+</td>
<td>2+</td>
<td>0</td>
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</table>

Eluate: panagglutinin, no specificity

Patient J.H.

- Patient ABO: O Rh Negative  Rh Phenotype: C-E-c+e+
- DAT: Polyspecific = 2+, Anti-IgG = 2+, Anti-C3 = 0
- Eluate: panagglutinin, no specificity

Patient J.H. – Current Findings

- Eluate contains an autoantibody with no specificity
- Serum appears to have an alloantibody that is reactive in gel only
Patient J.H. – Autoadsorbed Plasma x2

<table>
<thead>
<tr>
<th>Gel of Sample</th>
<th>Autoadsorbed Plasma</th>
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<tbody>
<tr>
<td>1</td>
<td>-</td>
</tr>
<tr>
<td>2</td>
<td>+</td>
</tr>
<tr>
<td>3</td>
<td>+</td>
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</table>

Patient J.H. - Conclusions

- Patient serum contains an autoantibody reacting best in gel - demonstrated specificity to the f(ce) antigen
- Two 37°C autoadsorptions removed this reactivity
- No underlying alloantibodies to common clinically significant red cell antigens detected at this time

Transfusion Recommendations:
Provide ABO/Rh compatible and crossmatch compatible or least incompatible red cells if needed. Since autoantibody was nonreactive in LISS, it may be beneficial to use this method for crossmatching.
Case Study #3 – G.A.

- 86 year old female
- Diagnosis: Anemia
- Hgb = 7.6 g/dL
- Transfused one unit 2 months ago, no antibodies at that time
- Referring hospital observed reactivity with all reagent red cells in gel
- Negative autocontrol
Patient G.A. - Initial Panel

<table>
<thead>
<tr>
<th>Type of Testing</th>
<th>Test Results</th>
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<tbody>
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Patient G.A. – Selected Cells

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<thead>
<tr>
<th>Type of Testing</th>
<th>Test Results</th>
</tr>
</thead>
<tbody>
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Patient G.A. - Conclusions

- Anti-Ch(Chido) only, no underlying alloantibodies
- High Titer Low Avidity (HTLA) – titers to a greater dilution than common clinically significant antibodies, does not adhere to red cells well, often cannot be removed by adsorption, most are weakened/destroyed by 0.2M DTT
- Clinically insignificant, benign antibody, "nuisance antibody"

Transfusion Recommendations:
Provide ABO/Rh compatible and crossmatch compatible or least incompatible red cells if needed.
Paroxysmal Cold Hemoglobinuria (PCH)

- Diagnostic test known as the Donath-Landsteiner (DL)
- Detects a bi-phasic hemolysis directed towards the P antigen
- Originally associated with syphilis patients
- More commonly observed in children 1-3 weeks post viral infection
- Acute hemolysis and resulting anemia
- Causative antibody IgG in nature that attaches in cold to bind complement then dissociates in warm leading to red cell lysis

Paroxysmal Cold Hemoglobinuria (PCH)

- Nine tubes
- Patient serum/Fresh Normal Serum (FNS)/50% suspension of Group O P1+ red cells
- One set of tubes incubated for 30 minutes in an ice bath followed by 60 minutes at 37C
- One set of tubes incubated for 90 minutes in an ice bath
- One set of tubes incubated for 90 minutes at 37C
- Spin and check for hemolysis

Donath-Landsteiner Test
Positive DL

Thank you for your time and attention
If you have questions…please contact me

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