Exercise 12: Mononucleosis Testing

Objectives:

1. Follow instructions of the reagent package insert to select and evaluate appropriate specimens for Infectious Mononucleosis testing.
2. Perform the Infectious Mononucleosis kit test for the detection of antibodies to obtain control and patient results that match instructor values with 100% accuracy.
3. Evaluate reagent package inserts to determine each of the following:
   a. The substance being detected.
   b. The principle of the procedure.
   c. Samples which are acceptable for testing.
   d. The expected result if the substance is present.
   e. Clinical significance of positive results.
   f. Limitations of the procedure.
   g. Troubleshooting procedures to follow when control results are unacceptable.
4. Accurately record and report results as instructed.
5. Utilize lecture notes, textbook and laboratory (including product inserts) information to answer study questions.

Introduction:

Infectious mononucleosis (IM) is an acute or subacute benign infectious lymphoproliferative disease that is caused by the Epstein-Barr virus (EBV), a herpes DNA virus. The virus is spread by person-to-person contact through saliva. The disease most often affects children and young adults and is characterized by an increase in atypical lymphocytes, enlargement of the lymph nodes and spleen, fever, sore throat, fatigue, malaise and the production of characteristic heterophile antibodies. Heterophile antibody titers rise during the first 2-3 weeks and gradually decline and usually disappear in 8-12 weeks following onset. The results of the IM test should be used by the physician along with clinical signs and symptoms along with other laboratory data, such as a CBC with low white count and high total lymphocyte count, the presence of atypical lymphocytes in the differential, to come to a diagnosis of IM.

Serological testing for mononucleosis became available as a result of the research of Paul and Bunnell on heterophile antibodies. Heterophile antibodies are those that will react (cross react) with any member of a group of similar antigens that can be found in unrelated animals or microorganisms. The antigens, called heterophile antigens, are so similar that an antibody built to one will cross react with the others. Paul and Bunnell noted that during the course of infectious mononucleosis, patients developed heterophile antibodies which were capable of agglutinating erythrocytes from sheep and horses. Davidsohn modified the original Paul-Bunnell test by adding steps to absorb out cross reacting Forssman and serum sickness antibodies leaving behind the IM heterophile antibody to react with sheep red cells. Guinea pig kidney absorb Forssman antibodies but do not affect the heterophile antibodies in IM. Their adaptation, the Davidsohn Differential test, became the classical reference method for laboratory diagnosis of infectious mononucleosis, however it is time consuming to perform.

Recent researchers have developed tests for infectious mononucleosis heterophile antibodies using treated horse or bovine red cells, or extracted antigens from bovine RBCs that have been attached to latex
particles or coated in tubes for enzyme-linked immunoassay testing (EIA).

**Principle:**

Review the principle of the specific kit being used in this laboratory.

**Materials:**

1. Mononucleosis testing kit
2. Patient and control serum specimens.
3. Timer
4. Other materials as directed by reagent product insert.

**Procedure:**

See specific reagent product insert.

**Interpretation:**

See specific reagent product insert.

**Expected Results:**

1. Consult the reagent product insert for interpretation.
2. As with all tests for heterophile antibodies, false positive and negative results do occur, but usually in low numbers (approximately 1%).
3. Detectable heterophile antibody level may be observed as early as four days, but more commonly requires three weeks or more. Some persons, especially young children, will not produce observable heterophile antibody levels although clinical and hematological signs are evident.
4. Positive test results may persist even though clinical symptoms are absent.

**Precautions:**

1. Follow manufacturer’s directions exactly (unless directed by instructor).
2. Reagents and specimens must be at room temperature.
3. **Review the product insert of the specific kit being used in this laboratory for additional information.**
Laboratory 12: Mononucleosis Testing

Results

Name _____________________________________ Date _____________________ Points _____/36

<table>
<thead>
<tr>
<th>KIT INFORMATION</th>
</tr>
</thead>
<tbody>
<tr>
<td>Test Kit Name</td>
</tr>
<tr>
<td>Manufacturer</td>
</tr>
<tr>
<td>Lot Number</td>
</tr>
<tr>
<td>Expiration Date</td>
</tr>
<tr>
<td>Package Insert Revision Date</td>
</tr>
<tr>
<td>Kit Storage Temperature</td>
</tr>
<tr>
<td>Samples which can be tested</td>
</tr>
<tr>
<td>Sample storage temperature</td>
</tr>
</tbody>
</table>

Interpret results as “Pos” for positive or “Neg” for negative.

<table>
<thead>
<tr>
<th>Patient Name</th>
<th>Identification Number</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2.</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Controls

<table>
<thead>
<tr>
<th>Positive Control</th>
</tr>
</thead>
<tbody>
<tr>
<td>Negative Control</td>
</tr>
</tbody>
</table>

INSTRUCTOR USE ONLY
Kit information – 1 point each (8 total)
Results area – 2 points each (16 total)
Study questions – 12 points

Total = 36 points
Using your textbook, lecture, lab and notes, answer the following questions.

1. Based on the control results, can these patient results be reported? (1 point)
   Yes   No (circle one)   If “no”, explain why.

2. What is being detected, antigen or antibody? (1 point)

3. What is the antigen or antibody used for detecting the substance present in the patient sample attached to? (1 point)

4. State at least three conditions which may cause positive results for heterophile antibodies. (3 points)
   a. 
   b. 
   c. 

5. What the name of the organism which causes infectious mononucleosis? (1 point)

6. What clinical and laboratory data must be considered before a diagnosis of IM can be made? (3 points)

7. After reviewing the product insert state the principle of this test kit including the appearance of positive and negative reactions. (In your own words, please.) (2 points)