WELLCOLEX COLOUR SHIGELLA LATEX TEST

I. Principle

A suspension of bacteria is mixed with latex reagents to serotype an organism that is *Shigella* based on biochemical testing. Each colored latex is coated with an antibody specific for a different *Shigella* species. In the presence of homologous antigen, one of the colored latex reagents will agglutinate. The identity of the antigen is indicated by the color of the aggregated particles with a contrasting change in the color of the background. Each reaction is readily distinguished from the other reactions, including a negative result in which the particles remain in smooth suspension and the occasional non-specific result in which all of the particles agglutinate into purple aggregates against a clear background.

II. Reagents

A. Wellcolex Latex Reagent 1 - 50 tests per vial
- Red latex *Shigella sonnei* (Forms I and II)
- Blue latex *Shigella flexneri* (Types 1-6, X and Y)

B. Wellcolex Latex Reagent 2 - 50 tests per vial
- Red latex *Shigella dysenteriae*
- Blue latex *Shigella boydii*

C. Wellcolex Positive Controls - two vials containing killed bacterial suspensions in a buffer with preservative.
- Red vial *Shigella sonnei* and *Shigella dysenteriae*
- Blue vial *Shigella flexneri* and *Shigella boydii*

III. Materials

A. Provided In Kit:
1. Disposable suspension tubes
2. Disposable sampling sticks
3. Disposable sample dispensers

B. Sterile saline
C. Flat-bed rotator, 150 rpm, orbital diameter
D. Disinfectant for material disposal

IV. Specimen Requirements

One or two (1-2 mm) suspected *Shigella* colonies from selective medium (XLD) should be used. Test colonies should be lactose negative and nonmotile.

V. Procedure

1. Bring reagents to room temperature.
2. Place 200 ul of saline in the suspension cup using the line on the provided pipette as a volume guide.
3. Pick one or two suspected colonies, and emulsify in saline.
4. Resuspend latex Reagents 1 and 2 by shaking vigorously for a few sec. Hold

the tip down vertically, and dispense one drop (30 ul) into separate circles on the reaction card.

5. Dispense one drop (40 ul) of the bacterial suspension onto each of the two circles containing Reagents 1 and 2. Do the same for the positive control on an additional two circles.
6. Mix the contents of each circle using a sample stick, and spread the mixture until the area of the circle is covered. Avoid bubbles.
7. Place the card on a flat-bed rotator for two min at 150 rpm. Leave the card on the rotator, and interpret the results by comparing them with the color chart.
8. Discard the card, pipettes, sticks and cups into the disinfectant.

VI. Interpretation

1. Positive: A colored latex aggregate with contrasting colored background should match one of the color chart selections. A mixed culture of *Shigella* may show two colors in one reagent, or single color may agglutinate in both reagents. These should be easily distinguished.
2. Negative: A smooth purple suspension indicates the isolate is not *Shigella*. A faint granularity may occur.
3. Nonspecific: Purple clumps against a clear background indicate a nonspecific result that cannot be interpreted as either positive or negative.

VII. QC Procedures

The performance of the latex reagents should be checked with each new kit by using the positive control provided.

VIII. Limitations

1. Color blind individuals cannot interpret the results of this test.
2. This product only identifies *Shigella* isolates to the species level and does not type within a species.
3. False positive results due to shared antigens with organisms from other species, particularly *E. coli,* can occur. Therefore, biochemical identification of the organism is essential.
4. Envelope antigens can inhibit the agglutination of O Antigens. The suspension may be heated at 100^{o}C for 2 h to inactivate the envelope antigens.

IX. Reporting Results

1. Confirm the identification with Micro ID.
2. Enter into the computer "*Shigella* sp." (as indicated by the test identification). Comment: "Sent to State Lab for Epidemiological purposes".
3. Call the report to the doctor or floor and document to whom and when it was called.
4. Perform susceptibility testing, and report.
5. Inoculate two BHI slants for stock cultures. One is sent to the State Lab and

the other is kept as a stock culture until the results are received from the State lab. Label the BHI slants with patient name, date of inoculation, accession number, and organism identification.

6. Notify Epidemiology and the Public Health Dept.

X. References

Wellcolex Color Shigella Test Package Insert, July 1997.

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Reviewed by Microbiology Director, Dr. Ann Robinson: 03/30/2000
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