

WELLCOLEX COLOUR SALMONELLA TEST

I. PRINCIPLE

The Wellcolex Colour Salmonella Test is a rapid method to identify *Salmonella* serogroups using color-coded polystyrene latex particles, each coated with specific rabbit antibody to the different serotypes. A bacterial suspension is mixed with the two latex reagents allowing agglutination of the bacterial antigen and the antibody coated latex particles. Agglutination results in a colored aggregate. The serotype is determined by the color of the aggregated particles and contrasting background.

II. REAGENTS

Store the following reagents at 2-8°C in the dark. Do not freeze or leave in direct sunlight. Bring to room temperature before use.

Wellcolex Latex Reagent 1 - 50 tests per vial.

Red latex	<i>Salmonella</i> Group B
Blue latex	<i>Salmonella</i> Group C
Green latex	<i>Salmonella</i> Group D

Wellcolex Latex Reagent 2 - 50 tests per vial.

Red latex	Vi
Blue latex	<i>Salmonella</i> Groups E and G
Green latex	<i>Salmonella</i> Group A

Positive Controls - three vials containing killed bacterial suspensions in preservative.

Red vial	<i>Salmonella</i> group B and Vi
Blue vial	<i>Salmonella</i> groups C and E
Green vial	<i>Salmonella</i> groups A and D

III. MATERIALS

Disposable suspension cups
Disposable pipettes
Disposable reaction cards
Wooden applicator sticks
Sterile saline
Flat bed rotator 150 rpm
Disinfectant for material disposal

IV. SPECIMEN REQUIREMENTS

One or two non-lactose-fermenting colonies (1-2mm) from overnight primary selective media (XLD) or pure subcultures should be used. Suspected *Salmonella* colonies should be H₂S positive and urea negative.

V. PROCEDURE

1. Bring the reagents to room temperature (15-30°C).
2. Place 200ul of saline in the suspension cup using a marked pipette.
3. Pick one or two suspected colonies (enough to cover the flat end of a wooden stick), and emulsify in the saline.
4. Resuspend latex reagent 1 and 2 by shaking vigorously. Hold the tip down vertically, and dispense one drop (30ul) into separate circles on the reaction card. Do the same with the Positive Controls.
5. Dispense one drop (40ul) of bacterial suspension onto each of the two circles using a kit pipette.
6. With a wooden stick, mix and spread the reagent drop with the bacterial suspension to cover the circle. Avoid bubbles.
7. Rotate for two min at 150 rpm on the rotator. Leave the card on the rotator, and interpret the results using the color chart.
8. Discard the card, pipettes, sticks, and cups into the disinfectant.

VI. QUALITY CONTROL

Using the same techniques as above, mix each of the three positive controls (Red, Blue, Green) with each of the two latex reagents (six card circles used). SHAKE VIGOROUSLY before using the controls. QC must be done with each new kit or lot number.

VII. INTERPRETATION

Positive

A colored latex aggregate with contrasting background color should match one of the color chart selections. A mixed *Salmonella* culture may show two colors in one reagent or single color may agglutinate in both reagents. These should be easily distinguished.

Negative

A smooth gray brown suspension in both reagents indicates a negative result. Faint granularity may occur.

Nonspecific

Grey brown clumps on a clear background indicate questionable results. Re-subculture the isolate to XLD, and retest when growth appears.

Debris

Lumps are caused by debris from the broth sample. Remove by filtration or centrifugation, and retest sample.

NOTE: If Vi antigen is detected, emulsify suspected colonies in saline, boil for 30 min, and retest with Reagent 1. Vi antigen obscures O antigens and may be removed by heating.

VIII. LIMITATIONS

1. Color blind individuals cannot interpret the test results.
2. The latexes do not cover the full range of serogroups but include 98% of the strains of *Salmonella* involved in human infection. This kit does not detect *S. arizoneae*.
3. An insufficient concentration of *Salmonella* in broth culture can cause false negative reactions.
4. False positive results can occur due to shared antigens among heterologous species or genera.
5. Vi antigens can be found in organisms other than *Salmonella*.

IX. REPORTING RESULTS

1. Confirm with Micro ID.
2. Enter into the computer: "*Salmonella* species. Sent to State Lab for Speciation".
3. Call report to doctor or floor.
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 in the lab until the results are received from the State lab. Label the slants with patient name, date inoculated, accession number, and organism identification.
6. Notify Epidemiology and the Public Health Dept.

REFERENCES

Wellcolex Color Salmonella Test Package Insert, May 1998.

Effective 06/1999

Reviewed by Microbiology Director, Dr. Ann Robinson: 03/29/2000

Reviewed by Medical Director, Dr. Joseph Schappert: 03/10/2010

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Updates and Revisions: