S&A REAGENTS LAB LTD.,PART.

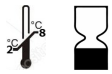
4 Soi Latphraowanghin 28 , Latphrao-wanghin Rd.,
Lat Phrao, Bangkok 10230, Thailand.
Tel. (+662) 538-8622
Fax. (+662) 931-6098
e-mail : saplab1990@gmail.com
www.serotest-thailand.com

Escherichia coli Antisera

E. coli antisera are produced for serological identification of *E. coli*, based on agglutination method. These polyclonal antibodies are prepared by immunizing rabbit with the standard strains.

For high specificity; the non-specific agglutinins have been removed by absorption.

Preservative : 0.1% Sodium azide
Storage condition : 2-8°C / See Exp. Date on package
Do not freeze the antisera.



Complete identifications of *E. coli* requires cultural isolation, biochemical characterization and serotyping. However well defined the serology, the use of serological procedures do not supersede cultural isolation and biochemical characterization.

Principle of the procedure

Serological tests are based on the fact that antibodies in serum, produced in response to exposure to bacterial antigens, will agglutinate with bacteria carrying homologous antigens. Test cultures with polyvalent sera, which are intended for use by the slide agglutination technique only. Both the confluent growth and selected colonies from the primary plate should be examined.

A positive slide reaction with a live culture may be due to the presence of K antigen on the surface of the organisms.

O-antigen testing

A. Slide agglutination of live organisms

1. Place 1-2 drops of saline onto control area and place a drop of polyvalent antiserum onto test area of clean glass slide.
2. Using platinum wire, transfer a portion of a loopful of growth from TSI slant onto the drop of saline and antiserum, mix the cultures and serum or cultures and saline well. Tilt the glass slide back and forth for one minute. If there is clumping in the control, the culture is rough and serological tests cannot be interpreted on slide testing.
3. If the live organisms doesn't give positive reaction with antiserum, mix 2 loops of the culture with 1 ml. of saline then heat at 100°C for 1 hr. then repeat step 1 and 2.
4. Further testing of the isolate should be conducted as described in steps 1-3 with monovalent O antisera to reveal the full O antigenic grouping of the isolate. Always confirm the O grouping by slide agglutination on heat killed organisms, see next step.

B. Slide agglutination of heat killed organisms

If the live organisms give positive reaction with antiserum, mix 2 loops of the culture with 1 ml. of saline then heat at 100°C for 1hr. then repeat the agglutination test as above using monovalent O antiserum on the heated cell suspension. This should be done to identify the O antigen type as distinct from the K antigen.

Slide Agglutination Interpretation

Agglutination should be strong and clearly visible within 1 minute. There should be no visible agglutination in the control suspension ; if agglutination is seen in the control, the suspension is not suitable for testing by this method.

H-antigen Testing

1. Allow the organism to pass through semi-solid medium. Then inoculate the resulting organism into 2 ml of Tryptic soy broth tubes and incubate at 37°C for 6-8 hrs.
2. After incubation period, add 2 ml. of 0.85% saline containing 1% (v/v) Formalin, allow the inoculum at the room temperature for 30 minutes. Transfer the 0.45-0.5 ml of inoculum into duplicate of 12x75 mm tube.
3. For the first tube, add 2 drops of E. coli H7 antiserum (AS546) and not add any antiserum for another tube for Negative control. Allow both tube to stand in water bath at 48-50 °C for 1 hour.
4. After incubation period, observe the result by checking the negative control tube which should shown an even suspension; clumping in the control indicates that the culture is rough and serological tests cannot be interpreted, then observe another tube for the flocculation reaction; that is Positive result. An isolate producing a distinct positive reaction is assumed to be an E. coli bearing the H antigenic factors represented by that antiserum.

E. coli Polyvalent Antisera

Cat. No.	Description	Specific Factors
AS521	E. coli (O&K) Polyvalent I	O25 : K11 O26 : K60 O44 : K74 O55 : K59 O78 : K80 O111 : K58 O114 : K- O119 : K69
AS530	E. coli (O&K) Polyvalent II	O86 : K61 O124 : K72 O125 : K70 O126 : K71 O127 : K63 O128 : K67
AS537	E. coli (O&K) Polyvalent III	O18a O18c : K77 O20a O20b : K84 O28 : K73 O112a O112c : K66

E. coli Monovalent Antisera

Cat. No.	Description
AS522	E. coli O25 : K11
AS523	E. coli O26 : K60
AS524	E. coli O44 : K74
AS525	E. coli O55 : K59
AS526	E. coli O78 : K80
AS527	E. coli O111 : K58
AS528	E. coli O114 : K–
AS529	E. coli O119 : K69
AS531	E. coli O86 : K61
AS532	E. coli O124 : K72
AS533	E. coli O125 : K70
AS534	E. coli O126 : K71
AS535	E. coli O127 : K63
AS536	E. coli O128 : K67
AS538	E. coli O18a O18c : K77
AS539	E. coli O20a O20b : K84
AS540	E. coli O28 : K73
AS541	E. coli O112a O112c : K66
AS545	E. coli O157
AS546	E. coli H7
AS547	E. coli O1 : K1
AS548	E. coli O2

Reference:

1. Kauffmann, F ., Classification of Bacteria 1947
2. Edward, P.R. and Ewing, W.H. , Indentification of of Enterobacteriaceae , Fourth Edition, Burgess Company, Minnesota 1986

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