PRO-LAB DIAGNOSTICS

PROLEX™ *E. coli* O157 LATEX TEST REAGENT KIT

(for *in vitro* diagnostic use)

PRODUCT CODE PL.070B Σ **50**

PRODUCT CODE PL.071B Σ **100**

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INTENDED USE

The Prolex™ *E. coli* O157 Latex Test Reagent Kit is an agglutination test kit for the presumptive identification of *Escherichia coli* serogroup O157.

SUMMARY AND EXPLANATION

Escherichia coli serotype O157:H7 is a Shiga toxin-producing pathogen.[1,2] This serotype has been reported as an etiological agent in sporadic and outbreak cases of haemorrhagic colitis.[3,4,5] It is also associated with haemolytic uraemic syndrome.[6] Certain *E. coli* serotypes other than O157:H7 also produce Shiga toxin.[7,8,9] However, the diarrhoea caused by these other serotypes is not usually bloody. Additionally, *E. coli* serotype O157:H7 does not ferment sorbitol, whereas most other serotypes do.[10,11] Therefore, if Sorbitol MacConkey Agar is used as a primary screen, the colonies of *E. coli* serotype O157:H7 appear colourless (non-sorbitol-fermenting colonies [NSFC]) while colonies of the other serotypes appear characteristically pink (sorbitol-fermenting colonies [SFC]).[11]

PRINCIPLE OF THE TEST

The blue polystyrene latex particles used in the kit are coated with an antibody against the *E. coli* O157 somatic antigen. When these latex particles are mixed with fresh colonies of *E. coli* serogroup O157, the bacteria will bind to the antibody causing the latex particles to agglutinate (positive reaction). Bacteria that are not *E. coli* O157 will not bind to the antibody and will not agglutinate (negative reaction).

MATERIALS PROVIDED

Prolex™ *E. coli* O157 Latex Reagent (PL.072B / PL.073B):

- One dropper bottle containing 3.1 ml (PL.070B) or 6.2 ml (PL.071B) of latex particles coated with purified rabbit IgG that reacts with *E. coli* serogroup O157. Latex particles are suspended in a buffer containing 0.098% sodium azide as a preservative.

Prolex™ *E. coli* O157 Positive Control (PL.074B / PL.075B):

- One dropper bottle containing 1.5 ml (PL.070B) or 3.0 ml (PL.071B) of Positive Control suspension containing *E. coli* serotype O157:H7 antigen produced by harvesting and inactivating *E. coli* serotype O157:H7 colonies grown on agar medium. The antigen is suspended in a buffer containing 0.095% sodium azide as preservative.

Prolex™ *E. coli* O157 Negative Control Latex Reagent (PL.077B / PL.076B):

- One dropper bottle containing 1.5 ml (PL.070B) or 3.0 ml (PL.071B) of latex particles coated with purified rabbit IgG that does not react with *E. coli* serogroup O157. Latex particles are suspended in a buffer containing 0.098% sodium azide as a preservative.
- Test cards
- Mixing sticks
- Instructions for use

MATERIALS REQUIRED BUT NOT PROVIDED

- Normal saline
- 12 x 75-mm test tubes
- Inoculating loop or needle
- Pasteur pipettes

STABILITY AND STORAGE

Reagents should be stored at 2°C to 8°C. Reagents stored under these conditions will be stable until the expiration date shown on the product label. **Do not freeze.**

PRECAUTIONS

1. The kit is intended for *in vitro* diagnostic use only.
2. Do not use the reagents after the expiration date shown on the product label.
3. The reagents contain ≤ 0.098% sodium azide. Sodium azide can react explosively with copper or lead plumbing if allowed to accumulate. Although the amount of sodium azide in the reagents is minimal, large quantities of water should be used if reagents are flushed down the sink.
4. Specimens and reagents should be considered potentially infectious, and universal precautions should be observed when performing the test.
5. Do not use the latex reagents if autoagglutination is visible. This would appear as agglutination of the Prolex™ *E. coli* O157 Latex Reagent in the absence of a test isolate or agglutination of the Negative Control Latex Reagent in the presence of Positive Control Antigen or the test isolate.
6. The procedures, storage conditions, precautions, and limitations specified in these directions must be followed to obtain valid test results.
7. Some reagents contain materials of animal origin and should be handled as a potential carrier and transmitter of disease.

PREPARATION OF CULTURES

Clinical specimens should be cultured on Sorbitol MacConkey Agar. Non-sorbitol-fermenting colonies (NSFC) may be tested directly or from a subculture on a non-selective agar medium. Colonies from an overnight culture (18-24 hrs) must be cleanly removed from the agar surface for testing using a sterile loop or needle. Young, fast-growing cultures typically give the best results.

TEST PROTOCOL

1. Allow all of the reagents to come to room temperature before use.
2. Using a pipette, transfer 0.2 ml normal saline into a 12 x 75-mm test tube.
3. Using a sterile loop or needle, pick off sufficient colonies from the plate and suspend them in the saline to achieve turbidity corresponding to a 3-5 McFarland Standard.
4. Place one drop of the Prolex™ *E. coli* O157 Latex Reagent in a test circle on one of the test cards provided. Using a Pasteur pipette add one drop of the test suspension into the same test circle and mix using one of the mixing sticks provided.
5. Rock the card gently and examine for agglutination for up to two minutes.
6. Isolates that give a positive result with the test latex must be tested further by repeating the procedure using the Prolex™ Negative Control Latex Reagent.

QUALITY CONTROL PROCEDURES

The Prolex™ *E. coli* O157 Latex Reagent and Prolex™ Negative Control Latex Reagent must be tested with the Prolex™ Positive Control before running the test isolates. There must be agglutination with the Prolex™ *E. coli* O157 Latex Reagent within two minutes and no agglutination with the Prolex™ Negative Control Latex Reagent.

INTERPRETATION OF RESULTS

1. The following table shows how the results obtained with the Prolex™ *E. coli* O157 Latex Reagents and the Prolex™ *E. coli* O157 Positive Control should be interpreted:

O157 LATEX REAGENT	NEGATIVE CONTROL LATEX REAGENT	REMARKS
+	-	Kit performance is satisfactory.
-	-	Potency is too low. Discard reagents.
+	+	Autoagglutination: Discard reagents

2. Agglutination of latex reagents with test specimen is interpreted as shown below:

O157 LATEX REAGENT	NEGATIVE CONTROL LATEX REAGENT	REMARKS
+	-	Presumptive for *E. coli* serogroup O157.
+	+	Autoagglutinating or cross-reacting strain present. Perform further testing to rule out *E. coli* O157.
-	not done	Indicates absence of *E. coli* sergroup O157.
stringy or mucoid appearance	not done	Uninterpretable. Make fresh suspension of colonies in saline and allow clumps to settle out. Retest supernatant.

LIMITATIONS OF THE PROCEDURE

1. Test only colonies that exhibit typical colonial morphology on Sorbitol MacConkey Agar (non-sorbitol-fermenting).
2. Positive test results should be confirmed using routine biochemical testing.
3. This reagent was developed to detect the presence of *E. coli* serogroup O157 antigen. Some other *E. coli* O157 strains (e.g., O157:H16) that are non-sorbitol-fermenting also produce a positive result with this test.[1,12,13]
4. Although this test has been specifically developed to reduce the normal cross-reactivity of *Escherichia hermanii* (12), rare strains can cross-react.

PERFORMANCE CHARACTERISTICS

Clinical performance of the Prolex™ *E. coli* O157 test kit was evaluated at a hospital microbiology laboratory. Blood-stained stool specimens from 474 patients diagnosed with diarrhoea, haemorrhagic colitis or haemolytic uraemic syndrome were cultured. Of these 474 specimens, 47 produced sorbitol-negative colonies and tested positive for *E. coli* strain O157 by a commercially available latex test. These results were confirmed by conventional biochemical testing. All 47 of the these isolates gave a positive result when tested using the Prolex™ *E. coli* O157 Latex Reagent Kit (47\47 = 100% sensitivity).

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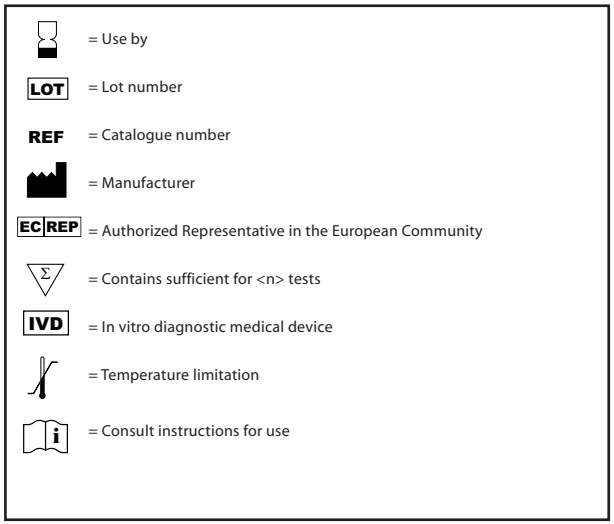

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