Prolex™ *E. coli* Non-O157 Identification Kit

(for *in vitro* diagnostic use)

PRODUCT CODE PL.1070

Σ 50

PRO-LAB DIAGNOSTICS

UK CA CE

INTENDED USE

The Prolex™ *E. coli* Non-O157 Identification Kit provides a rapid method to identify six non-O157 *Escherichia coli*: O26, O103, O111, O145, O45 and O121 isolated from cultured specimens.

SUMMARY AND EXPLANATION

Although *E. coli* O157 is the most common cause of STEC illness it has now been recognized that non-O157 strains of *E. coli* cause severe human disease that is comparable with that caused by *E. coli* O157. Of these strains *E. coli* O26 is the most commonly isolated and does cause haemolytic uremic syndrome (HUS) as can several other serotypes [1, 2, 3, 4, 5]. In addition, the following strains are the next most commonly isolated *E. coli* O103, *E. coli* O111, *E. coli* O145, *E. coli* O45 and *E. coli* O121. These serotypes have also been shown to cause significant human disease [6]. In the United States, the CDC has recommended that all clinical laboratories screen for STEC[7].

PRINCIPLE OF THE TEST

Polystyrene beads are sensitized with immunoglobulins against each of the serotype specific *E. coli* somatic antigens (*E. coli* O26, *E. coli* O103, *E. coli* O111, *E. coli* O145, *E. coli* O121 or *E. coli* O45). When the coated polystyrene particles are mixed with fresh organisms of the corresponding *E. coli* serotype the bacteria will bind to the antibody, causing the particles to visibly agglutinate (positive reaction). Bacteria which are not of serotypes O26, O103, O111, O145, O121 or O45 will not bind to the antibody and will not result in agglutination (negative reaction).

MATERIALS PROVIDED

Each kit is sufficient for 50 tests. Materials are supplied ready for use.

- Latex Reagents:
  Six dropper bottles each containing 2.5 ml of latex particles coated with purified rabbit IgG that react with *E. coli* somatic antigens (O26, O103, O111, O145, O121 or O45). Polystyrene particles are suspended in a buffer containing 0.094% sodium azide as a preservative.
- Prolex™ Negative Control Latex Reagent:
  One dropper bottle containing 2.5 ml of polystyrene particles coated with purified normal (non-immune) rabbit IgG. Polystyrene particles are suspended in a buffer containing 0.098% sodium azide as a preservative.
- Test cards
- Mixing sticks
- Instructions for use

All components of this kit are available separately for purchase:

Reagent or Component	Catalogue Number
Prolex™ *E. coli* O26 Latex Reagent	PL.1071
Prolex™ *E. coli* O45 Latex Reagent	PL.1072
Prolex™ *E. coli* O103 Latex Reagent	PL.1073
Prolex™ *E. coli* O111 Latex Reagent	PL.1074
Prolex™ *E. coli* O121 Latex Reagent	PL.1075
Prolex™ *E. coli* O145 Latex Reagent	PL.1076
Prolex™ Negative Control Latex Reagent	PL.1077
Mixing Sticks	PL.091P
Test Cards	PL.092-48

MATERIALS REQUIRED BUT NOT PROVIDED

- Phosphate buffered saline (PBS)
- 12 x 75 mm test tubes
- Inoculating loop or needle
- Pasteur pipettes
- Timer

STABILITY AND STORAGE

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

PRECAUTIONS

1. These reagents are 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 reagent is minimal, large quantities of water should be used if reagents are flushed down the sink.
4. Universal precautions should be taken in handling, processing and discarding all clinical specimens. All test materials should be considered potentially infectious during and after use and should be handled and disposed of appropriately.
5. Do not use any of the reagents if autoagglutination is visible. This would appear as agglutination of the test reagent or negative control in the absence of a 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 media that will facilitate optimal growth, such as MacConkey Agar, Sorbitol MacConkey Agar, Blood Agar, etc. Suspect colonies may be tested directly or from a sub-culture. Colonies from an overnight culture (18-24 hours) 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 PROCEDURE

1. Allow all reagents to come to room temperature before use.
2. Re-suspend the test latex reagents by gently inverting the dropper bottle several times. Examine the dropper bottles to ensure that the latex particles are properly suspended before use. Do not use if the latex fails to re-suspend.
3. Using a pipette transfer 0.3 ml of phosphate buffered saline into a 12 x 75 mm culture tube or equivalent.
4. Select sufficient suitable colonies from the test culture with a loop or needle and prepare a suspension in the phosphate buffered saline corresponding to a 3-5 McFarland Standard.
5. Label the test card with each of the serotypes and then add one drop of each latex reagent into the appropriate test circle.
6. Using a pipette transfer one drop (35 µl) of the test suspension onto each of the test circles.
7. Mix each of the test circles with a separate mixing stick.
8. Rock the card gently and examine for agglutination. A positive reaction (agglutination) will be visible within 30 seconds.
9. Isolates that give a positive test with any of the test reagents must be tested further by repeating the procedure using the Prolex™ Negative Control Latex Reagent.

QUALITY CONTROL PROCEDURE

The Prolex™ *E. coli* Non-O157 Latex Reagents and the Prolex™ Negative Control Latex Reagent must be tested with phosphate buffered saline before running the test isolates. There must be no agglutination in either of the reagents within 30 seconds.

INTERPRETATION OF RESULTS

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

Organisms	Results with Latex Reagents						Results with Negative Control Latex Reagent
	E. coli O26	*E. coli* O45	*E. coli* O103	*E. coli* O111	*E. coli* O121	*E. coli* O145	
E. coli O26	+	-	-	-	-	-	-
E. coli O45	-	+	-	-	-	-	-
E. coli O103	-	-	+	-	-	-	-
E. coli O111	-	-	-	+	-	-	-
E. coli O121	-	-	-	-	+	-	-
E. coli O145	-	-	-	-	-	+	-

Uninterpretable results: If the test isolate agglutinates with both the latex reagent and the Negative Control Latex Reagent, an auto-agglutinating or cross-reacting strain is present. Perform further testing to rule out non-O157 *E. coli*. If the test isolate reacts with more than one of the test reagents, the test is uninterpretable.

LIMITATIONS OF THE PROCEDURES

1. Positive test results should be confirmed using routine biochemical testing.
2. Although this test has been developed to reduce cross-reactivity, rare strains can cross-react. Do not observe for agglutination after 30 seconds.
3. If the test isolate fails to react with any one of the test reagents and you suspect that it is a pathogen please send it to your local reference centre for further study.
4. If the test isolate reacts with more than one of the test reagents please send it to your local reference centre for further study.

PERFORMANCE CHARACTERISTICS

The performance of the reagents was evaluated at a Reference Laboratory. Each of the reagents was tested against 177 different serotypes of *E. coli* including many other STEC serotypes.
The results from this evaluation showed 100% specificity and sensitivity for each of the reagents.

Canada ☎ 800 268 2341 Fax 905 731 0206
20 Mural Street, Unit #4, Richmond Hill, ON, L4B 1K3

U.S.A.☎ 512 832 9145 Fax 512 832 6424
21 Cypress Blvd., Suite 1155, Round Rock, Texas, 78665-1046

U.K.☎ 0151 353 1613 Fax 0151 353 1614
3 Bassendale Road, Bromborough, Wirral, Merseyside, CH62 3QL

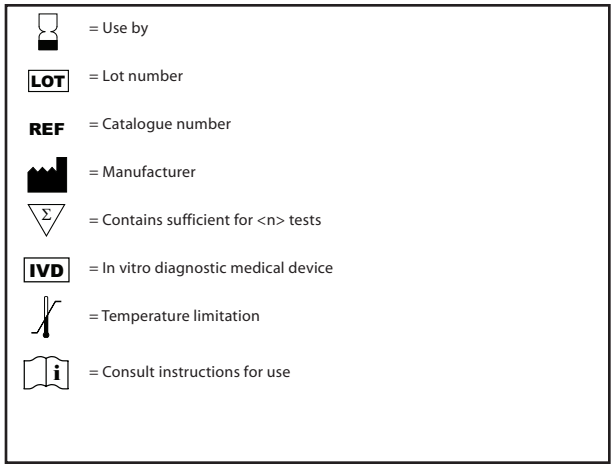
EC REP Advena Ltd. Tower Business Centre, 2nd Flr., Tower Street, Swatar, BKR 4013 Malta

Cross Reactivity
Each of the reagents was tested against the following enteric bacteria including *Shigella* species for cross reactivity. No cross reactions were found.

Organism	Result
Aeromonas hydrophila	Negative
Bacillus cereus	Negative
Bacillus subtilis	Negative
Campylobacter coli	Negative
Campylobacter fetus	Negative
Campylobacter jejuni	Negative
Citrobacter braakii (freundii)	Negative
Enterobacter aerogenes	Negative
Enterobacter cloacae	Negative
Enterococcus faecalis	Negative
Escherichia hermanii	Negative
Klebsiella pneumoniae	Negative
Proteus vulgaris	Negative
Pseudomonas aeruginosa	Negative
Salmonella enteriditis	Negative
Salmonella typhimurium	Negative
Serratia marcescens	Negative
Serratia liquefaciens	Negative
Shigella flexneri	Negative
Shigella dysenteriae	Negative
Shigella sonnei	Negative
Staphylococcus aureus	Negative
Vibrio parahaemolyticus	Negative

REFERENCES

1. **Banatvala N., Debeukelaer M.M., Griffin P.M., et al.** Shiga-like toxin producing *Escherichia coli* O111 and associated haemolytic-uremic syndrome; a family outbreak. Pediatr Infect Dis J 1996 15:1008-11.
2. **Tarr P.I., Fouser L.S., Stapleton A.E., et al.** Haemolytic-uremic syndrome in a six year girl after a urinary tract infection with Shiga-toxin producing *Escherichia coli* O103:H2. N Engl J Med 1996 335:635-8.
3. **Brooks J.T., Sowers E.G., Wells J.G., Greene K.D., Griffin P.M., Hoekstra R.M., Strockbine N.A.** Non-O157 Shiga Toxin-Producing *Escherichia coli* Infections in the United States, 1983-2002. J Infect Dis 2005 192:1422-1428.
4. **Beutin L., Zimmermann S., Gleier.** Human Infections with Shiga-Toxin Producing *Escherichia coli* Other Than Serogroup O157 in Germany. Emerging Infectious Disease 1998 4:635-639.
5. **Hadler J.L., Clogher P., Hurd S., Phan Q., Mandour M., Bemis K., Marcus R.** Ten–year trends and risk factors for non-O157 Shiga toxin-producing *Escherichia coli* found through Shiga toxin testing, Connecticut, 2000-2009. Clin Infect Dis. 2011 53(3):269-76.
6. **Hermos C.R., Janineh M., Han L.L., McAdam A.J.** Shiga Toxin Producing *Escherichia coli* in Children: Diagnosis and Clinical Manifestations of O157:H7 and Non-O157:H7 Infection. J Clin Micro. 2011 49:955-959.
7. **CDC 2009**. Recommendations for diagnosis of Shiga toxin-producing *Escherichia coli* infections in clinical laboratories. MMWR Morb. Mortal. Wkly. Rep 58:1-14.



Revision: 2023 04