thermoscientific

Product Specification Sheet

Brilliance™ E. coli / Coliform Selective

Intended Usage: A selective, chromogenic medium for the detection and enumeration of *Escherichia coli* and other coliforms in food and water samples.

For professional use only.

	PO5176A
Version: 12	Revision Date: 15 May 2020

Thermo Scientific™ *Brilliance*™ E. coli / Coliform Selective

Form of Product	Poured plate
Storage	2 – 12°C, dark
Filling weight	17 g ± 5 %
Packaging	10 plates wrapped in film
pH	6.7 ± 0.2
Appearance	Ivory, transparent
Shelf life	8 weeks
Intended Usage	A selective, chromogenic medium for the detection and enumeration of *Escherichia coli* and other coliforms in food and water samples. For professional use only.
Technique	Depends on the different methods. For information see product information Thermo Scientific™ Oxoid™ CM1046.

Typical formulation*	g/l
Peptone	8.0
Disodium hydrogen phosphate	2.2
Sodium chloride	5.0
Potassium dihydrogen phosphate	1.8
Sodium lauryl sulphate	0.1
Chromogenic mix	0.35
Tryptophan	1.0
Agar	10.6

*Adjusted as required to meet performance standards.

Quality Control

1. Control for general characteristics, labelling and printing.

2. Contamination check
   ≥ 72 h @ 20 – 25 °C, aerobic
   ≥ 72 h @ 30 – 35 °C, aerobic

3. Microbiological control

Positive Controls	Growth
Inoculum 50 – 120 colony forming units (cfu), quantitative Incubation conditions: 18 – 24 h @ 36 ± 1°C, aerobic	
Escherichia coli ATCC® 25922™	1 mm, violet colonies.
Colony counts shall be ≥ 50% of the control medium TSA.	
Inoculum 10^3 – 10^4 cfu, qualitative, control medium COL+SB Incubation conditions: 18 – 24 h @ 36 ± 1°C, aerobic	
Citrobacter freundii ATCC® 8090™	Good growth, small rose colonies.

Negative Control	Growth
Inoculum ≥ 10^4 cfu, quantitative, control medium TSA Incubation conditions: 18 – 24 h @ 36 ± 1°C, aerobic	
Staphylococcus aureus ATCC® 25923™	Partial inhibition (≤ 100 cfu).

Note: It is recommended when using Microbact Indole MB1448A to perform the indole test directly onto colonies rather than on filter paper.

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Description

The recovery and enumeration of *Escherichia coli* and coliforms are important indicators of environmental and food hygiene. Detection of ß-glucuronidase activity is widely used to differentiate *Escherichia coli*, as this by the *uid*A gene encoded enzyme is only present in *E. coli* but not in other members of the coliform group. As most coliforms are lactose-positive, ß -galactosidase activity, encoded by the *lac*Z gene, is then used to differentiate this group from other organisms able to grow on the selective medium. This results in purple *E. coli*, as they are able to cleave both chromogens, with other coliforms giving pink colonies as they cleave only the galactoside chromogen (see Table 1).

Brilliance™ E. coli/ Coliform Selective Agar contains two chromogenic agents:

- **Rose-Gal**: detects ß-galactosidase activity
- **X-Glu**: detects ß-glucuronidase activity

The medium also contains sodium lauryl sulphate which acts as a selective agent, inhibiting the growth of Gram-positive organisms. Due to the addition of tryptophan the rapid confirmation of *E. coli* by the formation of indole using a spot indole test is possible.

Table 1: expected results, using *Brilliance*™ E.coli/ Coliform Selective Agar

Organism	β-glucuronidase	β-galactosidase	Colony colour
Escherichia coli	+	+	Purple
Coliforms	-	+	Pink
Other organisms	-	-	Colourless or Blue
	+	-	

Technique

Prepare food samples by diluting 1:5 or 1:10 (as appropriate) with 0.1% (w/v) sterile Peptone Water (OXOID, product code CM0009), and homogenise in a Stomacher or a laboratory blender. Heavily contaminated water samples should first be diluted in Ringers Solution (OXOID, product code BR0052) or Maximum Recovery Diluent (OXOID, product code CM0733) so that the number of colonies to be counted is of a readable number e.g. 20-100 colonies. Potable water should be concentrated either by centrifugation or by using the filter membrane method.

The following techniques may be used:

1. Spread Plate

Dry the surface of the prepared plates. Pipette 0.1ml of the prepared sample onto the plate and spread over the surface with a sterile spreader. Incubate plates for 24 hours at 37ºC.

2. Filter Membrane Method

Dry the surface of the prepared plates. Filter an appropriate volume of sample through the membrane. Place the membrane onto the surface of an agar plate and avoid trapping air-bubbles under the membrane. Incubate for 24 hours at 37°C.

For all methods count the numbers of pink and purple colonies. Multiply the numbers of colonies by the dilution factor and express the result as the number of coliforms and *Escherichia coli* per gram of food or volume of water.

Presumptive identification of *E. coli* can be confirmed using a rapid indole test (DMAC) (e.g. Microbact-Reagent Spot Indole DMACA, product code MB1448A or BactiDrop™ Spot Indole, product code R21550) for same-day results. Do not use Kovacs, as the purple colouration of the colony could make interpretation of results difficult.

References

1. Kilian M., Bulow P. (1976). *Acta Pathol. Microbiol. Scand.* Sect. B 84, pp. 245-251.
2. Kilian M., Bulow P. (1979). *Acta Pathol. Microbiol. Scand*. Sect. B 87, pp. 271-276.
3. Frampton E. W., Restaino L., Blaszko N. (1988). *J. Food Prot.* Vol: 51(5), pp.402-404.