31401 DEV Lactose Peptone MUG Broth

For the enrichment and titer determination of coliform bacteria in connection with the bacteriological examination of water. The presence of *E. coli* can be demonstrated by UV fluorescence and a positive indole test.

Composition:

Ingredients	Grams/Litre
Meat peptone	10.0
Lactose	10.0
Sodium chloride	5.0
Bromocresol purple	0.01
Tryptophan	1.0
Methylumbelliferyl-β-D-glucuronide	0.1

Final pH 7.0 +/- 0.2 at 37°C

Store prepared media below 8°C, protected from direct light. Store dehydrated powder, in a dry place, in tightly-sealed containers at 2-25°C.

Directions:
Dissolve 26.1 g in 1 litre distilled water and pour into test-tubes fitted with Durham tubes. Autoclave at 115°C for 20 minutes.
Gas production from lactose fermentation is indicated by using inverted Durham tubes. Inoculate at least 1 ml sample in a tube containing 10 ml of DEV Lactose Peptone MUG Broth and a Durham tube. Incubate at 37°C for 16-24 hours. In case of gas formation the Durham tubes rise or/and show bubbles. If no gas has formed in the inverted tube, reincubate and reexamine after 48 hours. Turbidity of the medium accompanied by formation of gas within 48 hours is a positive presumtive test for the presence of E. coli and/or other coliform organisms.
In addition check the tubes under UV light at about 360-370 nm. Slightly blue fluorescence indicates the presence of *E. coli*. If no fluorescence occurs after 24 hours of incubation do not add Kovacs reagent (Fluka 60983) for checking the indole reaction as this alcoholic reagent will destroy the growth conditions for cultures. Continue incubation for another 24 hours and check again for fluorescence and indole reaction. For confirmation add Kovacs reagent to the tubes (5 mm layer). If the reagent layer becomes cherry red after 1-2 minutes, the presence of *E. coli* is confirmed.

Principle and Interpretation:
Meat peptone provide carbon, nitrogen, minerals, vitamins, trace elements and other essential nutrients for growth. Sodium chloride is for the osmotic balance. Lactose is the carbohydrate source. Bromocresol purple is a pH indicator which has a yellow color below pH 5.3 and a purple color above pH 6.7. Due to the fermentation of lactose and the attendant acid production the color changes to yellow. The addition of tryptophan improves the indole reaction.
β-D-glucoronidase produced by *E. coli,* cleaves 4-Methylumbelliferyl-β-D-glucuronide to 4-methylumbelliferone and glucuronide. The fluorogen 4-methylumbelliferone can be detected under a long wavelength UV lamp. In addition the indole test with Kovac's reagent (Fluka 60983) can be performed.

Cultural characteristics after 24-48 hours at 35°C.

Organisms (ATCC)	Growth	Color change to yellow	Gas formation	Fluorescence	Indole reaction
Escherichia coli (25922)	+++	+	+	+	+
Enterobacter aerogenes (13048)	+++	+	+	-	-
Enterococcus faecalis (11700)	++	+/-	-	-	-
Klebsiella pneumoniae (13883)	+++	+	+	-	-
Aeromonas hydrophila (7966)	++	-	-	-	-
Salmonella typhimurium (14028)	+++	-	-	-	-

References:

1. K. Kolbeck, et al., Supervision of the hygienic quality of bathing water by detection of E. coli after a 7-day salt water stress period, Zbl. Hyg., 193, 31437 (1992)
2. Deutsche Einheitsverfahren zur Wasser-, Abwasser- und Schlammuntersuchung, VCH Verlagsgesellschaft, D-6940 Weinheim
3. Verordnung über Trinkwasser und über Wasser für Lebensmittelbetriebe vom 12. Dezember 1990, Bundesgesetzbl., Teil I; 2613 (1990)