51696 PA *ChromoSelect* Broth (Presence/Absence *ChromoSelect* Broth)

PA *ChromoSelect* Broth is recommended for the detection of presence and absence of coliform bacteria in water.

Composition:**

Ingredients	Grams/Litre
Casein enzymic hydrolysate	20.0
Lactose	5.0
Bile salts mixture	1.5
Dipotassium hydrogen phosphate	3.0
Potassium dihydrogen phosphate	1.5
Sodium chloride	5.0
2-Nitrophenyl β-D-galactopyranoside (ONPG)	1.25
4-methylumbelliferyl β-D-glucuronide (MUG)	0.10
Final pH 7.0 ± 0.2 at 25°C	

**Formula adjusted, standardized to suit performance parameters

Store at 2-8°C and the prepared medium at 2-8°C. Use before expiry date on the label.

Appearance:	Faint beige, homogeneous, free flowing powder.
Gelling:	Firm
Color and Clarity:	Light amber colored, clear solution without any precipitate

Directions:
Suspend 37.35 grams in 1000 ml distilled water. Heat if necessary to dissolve the medium completely. DO NOT AUTOCLAVE. Cool to 50°C. Dispense into sterile test tubes.

Principle and Interpretation:
Examination of water for the presence of marker groups such as coliforms is one of the most common tests in food microbiology laboratory, partly because of the relative ease and speed with which these tests can be accomplished. Where it is claimed that water has been processed for safety, the finding of such organism demonstrates a failure of the process (1) PA *ChromoSelect* Broth is a modification of the medium originally devised by Hajna and Perry (2) and is used for the detection of presence and absence of coliform bacteria in water.
The fluorogenic compound 4-Methylumbelliferyl β-D-glucuronide (MUG) is incorporated in the medium for the fluorogenic detection of Escherichia coli, the main indicator organism for the faecal contamination of water. The enzyme β-glucuronidase possessed by Escherichia coli hydrolyses MUG to yield a fluorescent end product 4-Methylumbelliferone; which can be detected when the medium is observed for fluorescence under UV light (3,4) MUG also detects anaerogenic strains which may not be detected in the conventional procedure (3). ONPG test is used to determine the presence or absence of β-galactosidase in organisms (5) and is also important in differentiating Enterobacteriaceae which are commonly classifed according to their ability to ferment lactose. ONPG is similar in structure to lactose. The presence of two enzymes, permease and β-D-galactosidase are required to demonstrate lactose fermentation. True lactose nonfermenters do not possess either of these enzymes. Late lactose fermenting organisms do not have permease but do possess β-galactosidase. If β-galactosidase is present, the colorless ONPG is split into galactose and o-nitrophenol, a yellow compound (6).

Casein enzymic hydrolysate provides essential nutrients. Lactose is the fermentable carbohydrate, sodium chloride maintains osmotic equilibrium. The medium has a strong buffering system to control the pH in the presence of fermentative action. Bile salts inhibit gram-positive bacteria especially Bacillus species and faecal Streptococci. Mostly β-glucuronidase activity occurs within 4 hours but some weakly β-glucuronidase positive strains require overnight incubation (7).

Cultural characteristics after 18-24 hours incubation at 35-37°C.

Organisms (ATCC)	Inoculum [CFU]	Growth	ONPG	Fluorescence at 366 nm
Escherichia coli (25922)	50-100	+++	+ (yellow)	+
Enterobacter aerogenes(13048)	50-100	+++	+ (yellow)	-
Klebsiella pneumoniae (13883)	50-100	+++	+ (yellow)	-
Proteus mirabilis (25933)	50-100	+++	-	-
Salmonella Typhimurium (14028)	50-100	+++	-	-
Enterococcus faecalis (29212)	$\geq 10^3$	-	-	-
*Staphylococcus aureus (*25923)	$\geq 10^3$	-	-	-

References:

1. Corry J.E.L., Curtis G.D.W., and Baird R.M., Culture Media For Food Microbiology, Vol. 34, Progress in industrial Microbiology, 1995, Elsevier, Amsterdam
2. Hajna A.A. and Perry C.A., 1943, Am. J. Public Health, 33:550.
3. Feng P.C.S. and Hartman P.A.S., 1982, Appl. Environ. Microbiol.,43:132.
4. Robinson B.J., 1984, Appl. Environ. Microbiol., 48:285.
5. MacFaddin J.F, 2000, Biochemical Tests for Identification of Medical Bacteria. 3rd Ed. Philadelphia, Lippincott Williams and Wilkins, p. 160-9.
6. Isenberg H.D., (Eds.), Clinical Mirobiology Procedures Handbook, Vol. I, Washington D.C. American Society for Microbiology; 1992, p.l. 19.20-1.19.22.
7. Eaton A.D., Clesceri L.S. and Greenberg A.W.,(Eds.), 2005, Standard Methods for the Examination of Water and Wastewater, 21st Ed., APHA, Washington, D.C

Precautions and Disclaimer
This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.