Simultaneous Test for Coliform group and E. coli in water

Enzyme Substrate Medium Method (X-GAL - MUG method)

EC Blue 100P

Sterilized, folded dose form, granule medium

No. 1 000 227

EC Blue 100

Sterilized granule medium in bottle

No. 1 000 230

Features and advantages of EC Blue

1. Simultaneous detection of Coliforms and E. coli

Enzyme substrate method (X-GAL - MUG method) enables to detect both Coliform and E. coli simultaneously in one medium.

2. High sensitivity and short time to result

Coliforms and E. coli of 1 CFU/100 ml can be detected in 24 hours. EC Blue also works for stressed cells, damaged by chlorine disinfectant.

3. No needs of sterilization or preparation of medium

No sterilization and weighing of medium is required. The bottles and the folded pouches are sterilized by an electron beam after medium is dispensed into the container.

4. Easy and simple operation

Dispense the medium from the folded pouch into the test water, or pour the test water into a bottle, and incubate them.

5. Clear and distinctive interpretation

Blue/blue-green color occurs when Coliforms are present. Light blue/blue-purple fluorescent color can be observed under ultraviolet light when E. coli is present.

6. Three convenient packages

Available packages are the sterilized folded medium for 50 ml qualitativetest use, the sterilized medium in a bottle for 50 ml qualitative test use and the sterilized medium in a bottle for 10 ml quantitative test use.

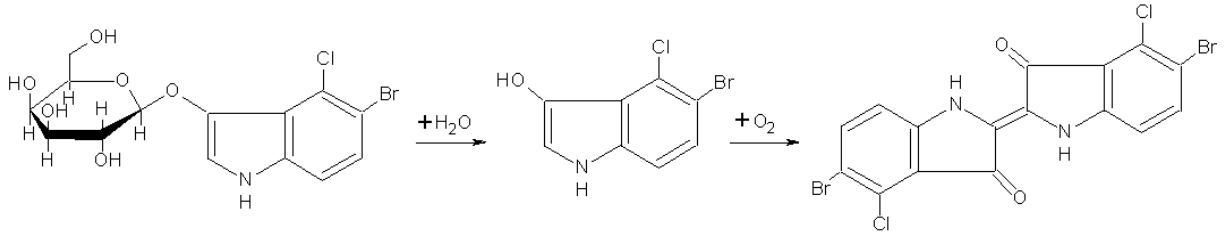
7. Shelf Life

Two years after manufacturing, when stored at room temperature, light shielded.

Mechanism of detection

Coliform bacteria

X-GAL, a synthetic colorimetric enzyme substrate, is decomposed by ß-galactosidase produced by Coliforms into bromo-chloro-indigo which causes blue/blue-green color.



5-bromo-4-chloro-3-indolyl-ß

colorless

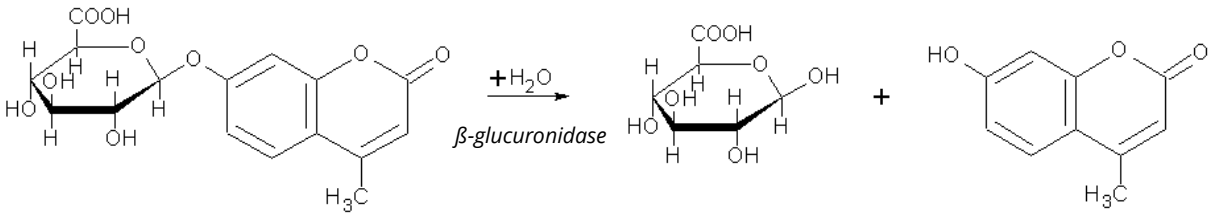
5-bromo-4-chloro-3-indolin

5,5-di-bromo-4,4-dichloro-indigo

blue/blue-green color

E. coli

MUG, a synthetic fluorescence enzyme substrate, is decomposed by ß-glucuronidase produced by E. coli to isolate the fluorescence compound 4-methyl-umbelliferone. Light blue/blue purple fluorescence can be observed under UV light.



4-methyl-umbelliferyl-ß-D-glucuronide (MUG)

Non-fluorescent

ß-D-glucuronic acid

4-methyl-umbelliferone

Fluorescence: light blue/blue-purple

Protocol

EC Blue 100P

For 100 ml Qualitative Test Use:
Add the granule into 100 ml water in a sterilized container. The pouch can be opened without any risks of contamination.

EC Blue 100

For 100 ml Qualitative Test Use:
Simply add 100 ml of the water to be tested into the already prepared bottle that is made of special materials, which have no fluorescence or moisture permeability.

EC BlueQuant MPN can be used for quantitative analysis,

Evaluation of results

Coliform group
The *Coliform* group is present when blue/blue-green color is observed under natural light. *Coliform* group is the major bacteria growing in this medium. *Coliforms* produce ß-galactosidase, which decomposes X-GAL (synthetic colorimetric enzyme substrate) and changes the color into blue/blue-green.

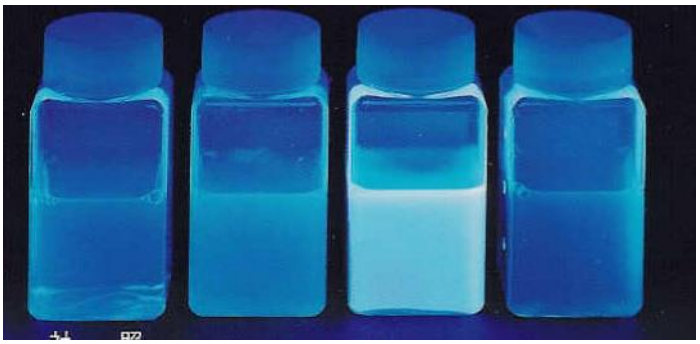
E. coli
The light blue/blue-purple fluorescence under UV light (364nm) indicates the presence of *E.coli* in the sample. About 95 % of *E. coli* produce ß-glucuronidase that hydrolyzes MUG (synthetic fluorescence enzyme substrate) to isolate a fluorescence substance that produces a light blue/blue purple fluorescence.

Pictures demonstrate positive results:

1. Analysis in natural Light:

Control ***Coliform* (-)** ***Coliform (+)*** ***Coliform (++)***

2. Analysis under UV Light, using the UV Lamp for EC Blue:



Control ***E. coli* (-)** ***E. coli* (+)** ***E. coli* (-)**

Formula (in 17.4 g for 1 liter)

Peptone	5.0 g	Sodium Chloride	5.0 g
Sodium Pyruvate	1.0 g	Sodium Lauryl Sulfate	0.1 g
Dipotassium Phosphate	4.0 g	Monopotassium Phosphate	1.0 g
5-bromo-4-chloro-3-indolyl-ß-D-galactopyranoside (X-GAL)	0.1 g	Isopropyl-ß-D(-)-thiogalacto-pyranoside (IPTG)	0.1 g
Potassium nitrate	1.0 g	4-methyl-umbelliferyl-ß-D-glucuronide (MUG)	0.1 g

ph 7.1 ± 0.1

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