WL Differential Agar

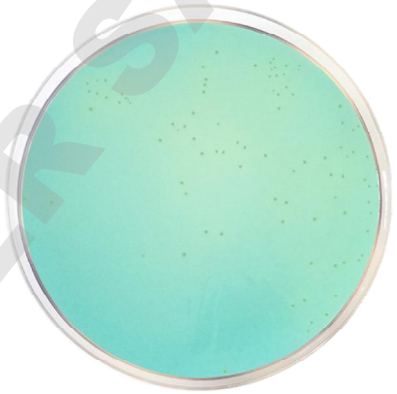
Cat. 1026

Selective medium used in the control of industrial fermention processes, especially in brewery

Practical information

Aplications	Categories
Selective enumeration	Microorganisms of the brewing industry
Selective isolation	Microorganisms of the brewing industry

Industry: Alcoholic beverages



Principles and uses

WL Differential Agar is a selective medium for the isolation and enumeration of microbial flora used together with WL Nutrient Agar (Cat. 1086) for the control of the manufacture of beer and other fermentation processes by yeasts. Both media are widely used in the industries of vinegar, bread yeasts, grape and wine-growing and distilled spirits. In the production of yeasts for the bakery and distillery industries, the pH of the media is adjusted to 6.5.

The medium allows the selective multiplication of yeasts in fermentation liquids, which contain a microflora mix consisting of fungi and bacteria. When the number of yeasts present is relatively small, certain bacteria can also be detected.

Tryptone provides nitrogen, vitamins, minerals and amino acids essential for growth. Dextrose is the fermentable carbohydrate providing carbon and energy. Yeast extract is a source of vitamins, particularly of the B-group. Monopotassium phosphate is the buffer. Potassium, Calcium and Ferric chlorides all provide essential ions for the osmotic balance. Magnesium and Manganese sulfates are sources of divalent cations. Bromocresol purple is the pH indicator. Bacteriological agar is the solidifying agent. The addition of 0.004 grams of Cycloheximide converts the WL Nutrient formula into a differential medium, which inhibits the development of yeasts and molds while permitting the notable proliferation of the bacteria present in the fermentation liquids and subsequent identification and enumeration.

Formula in g/L

Dextrose	50	Bacteriological agar	20
Bromocresol green	0,022	Calcium chloride	0,125
Cycloheximide	0,004	Ferric chloride	0,0025
Magnesium sulfate	0,125	Manganase sulfate	0,0025
Monopotassium phosphate	0,55	Potassium chloride	0,425
Tryptone	5	Yeast extract	4

Preparation

Suspend 80 grams of the medium in one liter of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Sterilize in autoclave at 121°C for 15 minutes.

Instructions for use

-Inoculate initial suspension and/or diluted sample.
- Extend the inoculum with a sterile loop on the agar surface.
- Incubate the plates in an inverted position at a temperature of 35±2 ºC for 24-48 hours.
Time and temperature of incubation are important factors according to the type of yeast. In general, temperatures of 25°C with the beer yeasts and 30°C with the bread and other alcoholic fermentation yeasts are appropriate. The time of incubation varies from 2 to 7 days and up to 14 days, depending on the flora found.

Likewise, the atmosphere chosen for incubating the culture must be appropriate. The bread yeasts are incubated aerobically while the alcoholic fermentation yeasts are incubated anaerobically and in the presence of CO2.

Quality control

Solubility	Appareance	Color of the dehydrated medium	Color of the prepared medium	Final pH (25ºC)
w/o rests	Fine powder	Beige with blue tint	Blue	5,5 ± 0,2

Microbiological test

Incubation conditions: (30 ºC / 24-48 h)

Microrganisms	Specification
Escherichia coli ATCC 25922	Good growth
Proteus mirabilis ATCC 25933	Good growth
Sacharomyces cerevisiae ATCC 9080	Inhibited growth
Lactobacillus fermentum ATCC 9338	Good growth
Saccharomyces cerevisiae ATCC 9763	Inhibited growth

Storage

Temp. Min.:2 ºC
Temp. Max.:25 ºC

Bibliography

Green and Grey. Wallenstein, Lab. Comm. 13:357. 1950. Green and Grey. Wallenstein, Lab. Comm. 14:169, 1951. Applicable to bacteriological investigation in brewing Wallesstein Lab. Commus 13: 357.