

## Specification

Solid medium used for the enumeration of water microorganisms according to ISO standards.

## Presentation

10 Prepared bottle  
Bottle 125 ml  
with: 100 ± 3 ml

### Packaging Details

1 box with 10 bottles 125 ml. Plastic screw inner cap.  
The use of syringes needles with a diameter greater than 0.8 mm is not recommended.

### Shelf Life

16 months

### Storage

8-25 °C

## Composition

Composition (g/l):

Yeast Extract.....	3.00
Tryptone.....	6.00
Agar.....	15.0

## Description /Technique

### Description

This medium, formulated according to ISO Standard 6222 and others is for the enumeration of heterotrophic microorganisms from water. Medium known as "Tryptone Yeast Extract Agar" and "Water Plate Count Agar".

### Technique

Melt the medium contained in bottles in a water bath (aprx. 100°C) or in microwave oven, avoiding overheating, before pouring into Petri dishes when cooled to room temperature.

Using a water sample obtained according to the ISO Standard 5667-2 and 5667-3, make a decimal dilution series (see ISO Standard 6887) using 1/4 Ringer Solution and take aliquots to 2 parallel series of plates. Pour the sterilized Tryptone Yeast Extract Agar cooled to 45°C, and homogenize with the sample (see ISO Standard 8199). Once solidified, incubate one of the series at 36 ± 2°C for 48 ± 2 hours and the other one at 22°C for 3 days (72 ± 4 hours).

In order to achieve a good count, select plates with 30-300 colonies. Express the results as number of colony forming units per millilitre (CFU/mL) of sample for each temperature of incubation. If there are no colonies with the undiluted sample express the results as "none detected in one mL". If there are more than 300 colonies in the highest dilution express the results as ">300 CFU/mL".

Note: The solid mediums can be melted in different ways: autoclave, bath and, if the customer considers appropriate, also the microwave. Whenever the microwave option is chosen, it is necessary to take certain safety measures to avoid breaking of the containers, such as loosening the screw cap and putting the bottle or tube in a water bath in the microwave. The fusion temperature and time will depend on the shape of the container, the volume of medium and the heat source. Avoid overheating as both the heating periods.

**Quality control****Physical/Chemical control**

Color : Yellowish

pH: 7.2 ± 0.2 at 25°C

**Microbiological control**

Melt Medium - Prepare Plates - Spiral Spreading: Practical range 100 ± 20 CFU. min. 50 CFU (productivity)

Analytical methodology according to ISO 11133:2014/A1:2018; A2:2020.

Aerobiosis. Incubation at 36 ± 2 °C, reading at 44±4 h

Reference medium : YEA (validated). Microbiological control according to ISO 11133:2014/A1:2018

**Microorganism****Growth***Escherichia coli* ATCC® 25922, WDCM 00013

Good (≥70%)

*Ps. aeruginosa* ATCC® 27853, WDCM 00025

Good (≥70%)

*Bacillus subtilis* ATCC® 6633, WDCM 00003

Good (≥70%)

*Stph. aureus* ATCC® 25923, WDCM 00034

Good (≥70%)

*Escherichia coli* ATCC® 8739, WDCM 00012

Good (≥70%)

**Sterility control**

Incubation 48 h at 30-35 °C and 48 h at 20-25 °C: NO GROWTH.

Check at 7 days after incubation in same conditions.

**Bibliography**

- ISO Standard 6222 Water Quality - Enumeration of cultivable microorganisms. Colony count by inoculation in a nutrient agar culture.
- ISO Standard 5667-2 (1991) Water Quality - Sampling - Guidance on sampling techniques.
- ISO Standard 5667-3 (1996) Water Quality - Sampling - Guidance on the preservation and handling of samples.
- ISO Standard 6887 (1999) Microbiology - General - Guidance for the preparation of dilutions for microbiological examination.
- ISO Standard 8199 (1988) Water Quality - General guide to the enumeration of microorganisms by culture.
- ISO 11133:2014/ Adm 1:2018. Microbiology of food, animal feed and water. Preparation, production, storage and performance testing of culture media.