

# PENICILLIN AND PIMARICIN AGAR (PPA)

## ENUMERATION OF *PSEUDOMONAS* SPP

### 1 INTENDED USE

Penicillin and Pimaricin Agar (PPA) is a selective medium for the enumeration of pigmented and non-pigmented psychrotrophic *Pseudomonas* in milk, milk products and in dairy environment samples.

The typical composition responds to that defined in the standard XP ISO/TS 11059.

### 2 PRINCIPLES

Pancreatic digest of gelatin and Tryptone are the nutrient substrates required for the rapid multiplication of *Pseudomonas*.

Magnesium chloride and potassium sulfate maintain the osmotic balance of the medium and stimulate the production of pyocyanin (a blue, non-fluorescent pigment soluble in water and in chloroform) of *Pseudomonas*.

The penicillin G and pimaricin concentrations inhibit most contaminating bacteria contaminants.

### 3 TYPICAL COMPOSITION

The composition can be adjusted in order to obtain optimal performance.

For 1 liter of complete media :

- Pancreatic digest of gelatin ..... 16,0 g
- Tryptone ..... 10,0 g
- Potassium sulfate ..... 10,0 g
- Magnesium chloride ..... 1,4 g
- Penicillin G ..... 10<sup>5</sup> IU
- Pimaricin ..... 10,0 mg
- Bacteriological agar ..... 12,0 g

pH of the ready-to-use media at 25 °C : 7,2 ± 0,2.

### 4 INSTRUCTIONS FOR USE

- Prepare the initial suspension of the product to be analyzed following the XP ISO/TS 11059 standard instructions.
- To the surface of the pre-poured media plates (BM156), transfer 0,1 mL of the stock solution and successive serial dilutions.
- Spread the inoculum over the surface with the help of a sterile triangle or « hockey stick ».
- Incubate at 25 ± 1 °C for 48 ± 2 hours.

✓ **Inoculation :**  
0,1 mL on surface

✓ **Incubation :**  
48 ± 2 h at 25 ± 1 °C

### 5 RESULTS

Only count plates containing no more than 150 colonies maximum.

Randomly pick 5 colonies on each plate to perform their purification on 2% Nutrient Agar (BK185HA or BM11808) before confirmation of the genus *Pseudomonas* with the oxidase and glucose fermentation tests (Glucose agar : BM09908).

*Pseudomonas* species are characterized by the positive reaction to the oxidase test and the absence of glucose fermentation.

## 6 QUALITY CONTROL

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**Complete media** : whitish agar.

Typical culture response on complete media after 48 hours of incubation at 25 °C (NF EN ISO 11133) :

Microorganisms		Growth (Productivity Ratio : $P_R$ )
<i>Pseudomonas aeruginosa</i>	WDCM 00025	$P_R \geq 50$ %, characteristic colonies
<i>Pseudomonas fluorescens</i>	WDCM 00115	$P_R \geq 50$ %, characteristic colonies
<i>Escherichia coli</i>	WDCM 00013	Inhibited

## 7 STORAGE / SHELF LIFE

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**Pre-poured media in plates** : 2-8 °C.

The expiration date is indicated on the label.

## 8 PACKAGING

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**Pre-poured media in Petri plates (Ø 90 mm) :**

20 plates ..... BM15608

## 9 BIBLIOGRAPHY

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XP ISO/TS 11059. Octobre 2009. Lait et produits laitiers. Méthode de dénombrement des *Pseudomonas* spp..

## 10 ADDITIONAL INFORMATION

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The information provided on the labels take precedence over the formulations or instructions described in this document and are susceptible to modification at any time, without warning.

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