

UREA AGAR BASE (CHRISTENSEN) ISO 6579, ISO 19250

CAT N°: 1110

For the differentiation of Enterobacteria on the basis of urease production

FORMULA IN g/l

Urea	20.00	Gelatin Peptone	1.00
Sodium Chloride	5.00	Dextrose	1.00
Monopotassium Phosphate	2.00	Phenol Red	0.012

Final pH 6.8 ± 0.2 at 25°C

PREPARATION

Dissolve 29 grams of the medium in 100 ml of distilled water. Sterilize by filtration. Separately dissolve 15 grams of agar in 900 ml of distilled water by boiling. Sterilize in autoclave at 121 °C for 15 minutes. Cool to 50°C and add to the 100 ml of the sterile Urea Agar Base. Mix well and dispense aseptically in sterile tubes. Leave the medium to set in a slanted position so as to obtain deep butts. Do not overheat and do not remelt the slanted agar. The prepared medium should be stored at 2-8°C. The color at a pH of 6.8 to 7.0 should have a light pinkish-yellow color.

The dehydrated medium should be homogeneous, free-flowing and orange-red in color. Urea is considered a highly hygroscopic substance. If the medium is not kept under controlled conditions of humidity and temperature, caking of the medium may occur. For this reason, avoid sudden changes in temperature of the dehydrated medium.

USES

UREA AGAR BASE (Christensen) may be used as an aid in the differentiation of microorganisms, particularly enteric Gram-negative Enterobacteria, on the basis of urea hydrolysis, from clinical samples and other materials. The formula is according to ISO 6579, and ISO 19250.

Urea Agar Base, with TSI Agar (Cat. 1046), may be used as a screening medium for the selection of *Salmonella* and *Shigella*. Urea Agar Base is used in spot tests for the rapid detection of urease activity and, when combined with results of other quick screening tests, it is the most common method to detect urease production by Enterobacteria. It is particularly recommended for the differentiation of members of the genus *Proteus* from those of *Salmonella* and *Shigella* in the diagnosis of enteric infections.

Gelatin peptone provides nitrogen, vitamins, minerals and amino acids essential for growth. Dextrose is the fermentable carbohydrate providing carbon and energy. Sodium chloride maintains the osmotic balance. Monopotassium phosphate provides buffering capacity. Urea is a source of nitrogen for those organisms producing urease. Phenol red is the pH indicator.

To obtain good results, inoculate heavily over the slant as the speed of the reaction depends on the relation of organism amount and medium surface. Do not inoculate the butt of this medium as it is used as a negative color control. ISO 6579 recommends to incubate at 35 ± 2°C for 24 ± 3 hours, and ISO 19250 recommends to incubate at 36 ± 2°C for 24 ± 3 hours. A positive test is denoted by a change in color, due to urease production, from yellow (pH 6.8) to a deep purple or bluish-red (pH 8.1) on the slant surface. *Proteus* and a few other organisms give a positive (purple) reaction.

Reincubate all negative cultures daily for up to 7 days for positives such as *Brucella*.

MICROBIOLOGICAL TEST

The following results were obtained in the performance of the medium from type cultures after incubation at a temperature of 35±2° C and observed after 24±3 hours.

Microorganisms	Growth	Urease
<i>Enterobacter aerogenes</i> ATCC 13048	Good	- (no change of color in the medium)
<i>Escherichia coli</i> ATCC 25922	Good	- (no change of color in the medium)
<i>Klebsiella pneumoniae</i> ATCC 13883	Good	+ (red or purple medium)
<i>Proteus vulgaris</i> ATCC 13315 (<i>Proteus hauseri</i> O'hara et al.2000)	Good	+ (red or purple medium)
<i>Salmonella typhimurium</i> ATCC 14028	Good	- (no change of color in the medium)

According ISO 19250 incubate at 36±2° C for 24±3 hours.

BIBLIOGRAPHY

Christensen J. Bact. 52:641. 1946. Thal and Chen J. Bact. 69:10. 1955. Ewing Enterobacteriaceae. USPHS, Publication 734.
Norma UNE-EN-ISO 6579. Microbiology of food and animal feeding stuffs -- Horizontal method for the detection of *Salmonella spp.*
ISO 19250 water quality-detection of *Salmonella spp*



STORAGE

Once opened keep powdered medium closed to avoid hydration.

