Urea Agar Base, Christensen

Intended Use
Urea Agar Base, Christensen is a medium with added urea used for the detection of urease production, particularly by the genus *Proteus*.

Summary
Christensen devised Urea Agar Base for use as a solid medium for the differentiation of Enteric Bacilli. It differentiates between rapid urease-positive organisms (*Proteus* species, *Morganella morganii* subspecies *morganii*, *Providencia rettgeri* and some *Providencia stuartii*) and other urease-positive organisms: *Citrobacter*, *Enterobacter* and *Klebsiella* and bacteria other than *Enterobacteriaceae*, i.e., some *Bordetella* and *Brucella* species. Urea Agar Base is included in the Bacteriological Analytical Manual for food and cosmetics testing, in IP and is recommended by APHA for the examination of foods.
Rustigian and Stuart had originally formulated Urea Broth to differentiate *Proteus* species from other Gram-negative enteric bacilli capable of utilizing urea; the latter were unable to do so because of limited nutrients and the high buffering capacity of the Urea Broth. To provide a medium with greater use Christensen devised Urea Agar Base with the addition of peptone, dextrose and reduced content of buffer to promote rapid growth of many of the *Enterobacteriaceae* and permit a reduction in incubation time.

Principle
Peptone is the source of essential nutrients. Dextrose is the energy source. Sodium chloride maintains the osmotic equilibrium of the medium whereas phosphates serve to buffer the medium. Urea is hydrolyzed to liberate ammonia. Phenol red indicator detects the alkalinity generated by visible colour change from orange to pink.

Formula*
Ingredients	**g/L**
Sodium Chloride	5.0
Disodium Phosphate	1.2
Dextrose	1.0
Peptone	1.0
Mono Potassium Phosphate	0.8
Phenol Red	0.012
Agar	15.0
Final pH (at 25°C)	6.8 ± 0.2

*Adjusted to suit performance parameters.

Storage and Stability
Store dehydrated medium below 30°C in tightly closed container and the prepared medium at 2°C-8°C. Avoid freezing and overheating. Use before expiry date on the label. Once opened keep powdered medium closed to avoid hydration.

Specimen Collection and Handling
Ensure that all samples are properly labelled. Follow appropriate techniques for handling samples as per established guidelines. Some samples may require special handling, such as immediate refrigeration or protection from light, follow the standard procedure. The samples must be stored and tested within the permissible time duration. After use, contaminated materials must be sterilized by autoclaving before discarding.

Directions
1. Suspend 24.01 g of the powder in 950 mL purified / distilled water and mix well.
2. Boil with frequent agitation to dissolve the powder completely. DO NOT OVERHEAT.
3. Sterilize by autoclaving at 115°C (10 psi) for 20 minutes as per validated cycle.
4. Cool to 50°C and aseptically add 50 mL of sterile 40% Urea solution (204210060005) and mix well.
5. Dispense into sterile test tubes and allow to set in a slanting position.
6. Do not reheat the medium after the addition of Urea 40%, as urea decomposes very easily.

Quality Control

Dehydrated Appearance: Light orange coloured, homogenous, free flowing powder.
Prepared Appearance: Yellow orange coloured, clear to slightly opalescent gel forms in tubes as slants.
Cultural Response: Cultural characteristics observed after an incubation of 18-24 hours at 30°C-35°C.

Organism (ATCC)	Growth	Urease
Escherichia coli (25922)	Good	-
Salmonella enterica subsp. *enterica* serovar *Typhimurium* (14028)	Good	-
Klebsiella aerogenes (13048)	Good	-
Proteus mirabilis (25933)	Good	+

Key:
+ = Positive, pink - red colour
- = Negative, no change

Interpretation of Results

1. When organisms utilize urea, ammonia is formed which makes the medium alkaline, producing an intense pink-red colour on the slant.
2. The colour may penetrate into the agar butt; the extent of colour indicates the rate of urea hydrolysis.
3. A negative reaction is no colour change. The agar medium remains pale yellow to buff.

Performance and Evaluation

Performance of the product is dependent on following parameters as per product label claim:

1. Directions
2. Storage
3. Expiry

Precautions/Limitations

1. Do not reheat the medium as urea decomposes very easily.
2. The alkaline reaction produced in this medium after prolonged incubation may not be caused by urease activity. False positive reactions may occur due to the utilization of peptones (*P. aeruginosa* for e.g.) or other proteins, which raises the pH due to protein hydrolysis, and the release of excess amino acid residue. To eliminate possible protein hydrolysis, perform a control test without urea.
3. Urea Agar rapidly detects urease activity of only the urease positive *Proteus* species. For results to be valid for the detection of *Proteus*, the results must be read within first 2-6 hours after incubation.
4. Urease positive *Enterobacter, Citrobacter* or *Klebsiella* in contrast, hydrolyze urea much more slowly, showing only slight reaction into the butt of the medium in 6 hours and requiring 3-5 days to change the reaction of the entire butt.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques.

Warranty

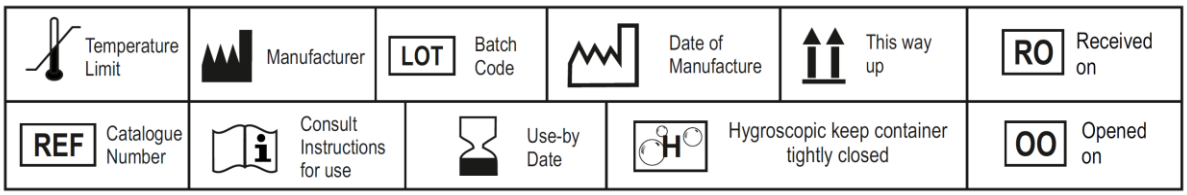
This product is designed to perform as described on the label and package insert. The manufacturer disclaims any implied warranty of use and sale for any other purpose.

Reference

1. Christensen WB; 1946, J. Bact; Vol. 52:461.
2. US Food and Drug Adm; 1998, Bacteriological Analytical Manual, 8th Ed; Rev. A, AOAC, International, Gaithersburg, Md.
3. IP, 1996, Ministry of Health and Family Welfare, Govt. of India, Vol. 2.
4. Downes and Ito (ed.) 2001, Compendium of Methods for The Microbiological Examination of Foods, 4th edition, APHA Washington DC.
5. Rustigian and Stuart, 1941, Proc. Soc. Exp. Bio. Med; 47: 108.
6. Data on file: Microxpress®, A Division of Tulip Diagnostics (P) Ltd.

Product Presentation:

Cat No.	Product Description	Pack Size
201210010100	Dehydrated Culture Media	100 g
201210010500	Dehydrated Culture Media	500 g
203210040012	Ready Prepared Slant	12 Slants



Revision: 0825/VER-03

Disclaimer

Information provided is based on our inhouse technical data on file, it is recommended that user should validate at his end for suitable use of the product.