Easybact®

Intended Use
Easybact® is a differential, semi-quantitative bacteriuria collection and screening system.

Summary
Urinary tract infection is one of the most common infections encountered in clinical practice. Cultivation and isolation of microorganisms from pathological urine samples is many a times the key to identification of underlying pathogens. With ever increasing strains of resistant microorganisms, susceptibility testing to antimicrobial agents for selecting appropriate drugs / drug regimens is routinely indicated. Easybact® a ready to use C.L.E.D. agar slants, fills this long felt need using a unique proprietary technology for routine microorganisms in urine screening.

Principle
Easybact® is a ready to use, pre-calibrated C.L.E.D. medium with a pH indicator, for easy isolation, identification and enumeration of microorganisms within 18 to 24 hours. Colonies obtained from this medium can be further processed for antibiotic sensitivity testing directly using the plate method. The standard medium is slightly opalescent greenish / grey in colour. Easybact® is suitable for urinary microbiology and supports the isolation and growth of most common urinary tract pathogens such as *E. coli*, *Proteus*, Streptococci, Staphylococci, and *Enterococcus*. As the urine sample is voided / placed in the Easybact® vial and subsequently emptied, organisms if present gets seeded onto the medium and start growing. Easybact® has been calibrated to yield colony counts similar to the standard plate methods. Easybact® has a double indicator system that allows differentiation of various urinary pathogens by differential colour formation within the colony as well as the medium. Study of colony morphology allows further identification. Since swarming of *Proteus* species is prevented on this media, this media is convenient for colony count.

Formula

Ingredients	**g/L**
Lactose	10.0
Tryptone	4.0
Peptone	4.0
Beef Extract	3.0
L- Cystine	0.128
Andrade Indicator	0.10
Bromothymol Blue	0.02

Additional Material Required
Incubator (35°C-37°C), blotting / filter paper, activated 2% glutaraldehyde solution.

Specimen Collection and Preparation
As pathogens accumulate in the patient's bladder overnight, first morning voided urine samples provide the best yield. Aseptically collect midstream clean catch urine or first morning catheterisation / suprapubic taps in sterile containers. Fresh urine specimen is recommended for testing. Samples may be tested upto 3 hours when stored at 2°C-8°C. If the patients can be explained clearly, Easybact® vial may be used to directly collect the midstream clean catch samples. (Refer Notes for collection of midstream clean catch urine).

Procedure
1. Retrieve the required number of Easybact® vials from the carton.
2. Bring the slants to room temperature (25°C-30°C) prior to testing.
3. Label the Easybact® vials appropriately with the patient's I.D.
4. Open the Easybact® vial observing aseptic conditions.
5. Directly collect clean catch midstream urine into the Easybact® vial OR add urine samples from catheterisation / suprapubic into the Easybact® vial, right up to the brim.
6. Empty the urine sample from the Easybact vial immediately, observing aseptic conditions.
7. In case small amount of urine is retained in the vial, drain the excess urine by gently tapping the mouth of the Easybact® vial on to a fresh clean blotting / filter paper. This is done to ensure no excess urine remains on the slant / in the bottle.

8. Recap the vial immediately.
9. Incubate the vial in an incubator, pre-set at 35°C-37°C for 18 to 24 hours, in an inverted position with the cap facing downwards.
10. Read the results at the end of the incubation period.

Quality Control

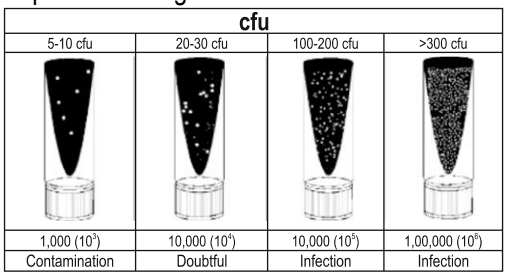
Appearance: Blue-green coloured smooth slant.
Cultural Response: Cultural characteristics observed after an incubation of 18-24 hours at 35°C-37°C.

Organisms (ATCC)	Growth	Colour of the Colony
Escherichia coli (25922)	Good	Pink colonies with pink medium
Proteus hauseri (13315)	Good	Blue-green transluscent colonies
Staphylococcus aureus (25923) subsp. *aureus*	Good	Golden yellow colonies with pink medium
Pseudomonas aeruginosa (27853) Strain Boston 41501	Good	Colourless Colonies

Interpretation of Results

Identification of Bacteria:
The pathogenic organisms can be identified as per the colour chart provided with the Easybact® kit.

Density Count:
Read the density of the colonies as per the chart given below:



1. If approximately 5-10 cfu observed on the Easybact® slant, read the density as 10^3 cfu/mL of urine sample- CONTAMINATION (No Infection).
2. If approximately 20-30 cfu observed on the Easybact® slant, read the density as 10^4 cfu/mL of urine sample- DOUBTFUL INFECTION and has to be repeated with three consecutive sample.
3. If approximately 100-200 cfu observed on the Easybact® slant, read the density as 10^5 cfu/mL of urine sample- INFECTION.
4. If >300 cfu observed on the Easybact® slant, read the density as 10^6 cfu/mL of urine sample - INFECTION.

Remarks

1. Discoloured, dislodged, or contaminated medium should not be used.
2. Ensure that clean catch midstream urine samples are used so that surrounding external microbial flora does not give discrepant results.
3. Observe aseptic conditions while performing the test to avoid contamination with non-pathogenic bacteria.
4. Urine specimens should be collected and cultured immediately. Prolonged storage of urine samples at room temperature (25°C-30°C) may result in multiplication of contaminating organisms and raise the microbial count leading to discrepancies.
5. Cultures of three consecutive first morning urine specimens are universally recommended since the reliability of the test increases to approximately 100%.
6. Whenever possible indicate if the patient is on antibiotic therapy, since urine samples of patients on antibiotic therapy may not show microbial growth or may have low colony count.
7. Treat the specimens and used slants by immersing in 2% activated glutaraldehyde for at least two hours before incineration and disposal.
8. For confirmation and identification of the isolated organism, it is recommended to perform Gram stain, biochemical or serological studies.
9. *Citrobacter* spp. were commonly misidentified as *E. coli.* Improved differentiation between *Citrobacter* spp. and *E. coli* has been previously demonstrated by Chagla *et al.,* using PYR hydrolysis test.
10. Good laboratory practices and hazard precautions must be observed at all times.

11. When performed correctly, Easybact® results correlate with standard plate method using calibrated loop.
12. Optionally calibrated loops can also be used for inoculating the sample. Eg: If 1 µL of the sample is plated on the slant and the count observed is > than 100 cfu it is infection.

Notes

Procedure for collection of clean catch midstream urine samples. The objective is to collect a specimen, which will reflect as much as possible only the urine present in the urinary bladder. Thus, a clean midstream void is recommended. Instruct the patient as follows: Wash and clean the private parts with a dilute soap. Remove all traces of soap by washing with large quantity of water. Wipe dry. Void out, into the toilet, the first stream of urine. This will flush out dead epithelial cells of the urinary bladder, microparticulates and normal microbial flora, which may have collected in the urine. Then hold the remaining urine in the bladder.
Next, void the second stream of urine aseptically into the Easybact® vial right upto the brim. Again, hold the remaining urine in the bladder. Lastly, void out the remaining third stream, into the toilet.
This procedure ensures that the urine is voided as three discreet segments. (First stream to flush out contaminants, second stream as the clean midstream for test). Follow instructions as mentioned in the Test Procedure for performance of the test.

Storage and Stability

1. Store the Easybact® slants at 2°C-8°C, away from light. DO NOT FREEZE.
2. Stability of the unused slants is as per the expiry date mentioned on the vial / carton labels.
3. Avoid jerks and vibrations while storage and incubation.
4. Upon opening, the medium must be put into use immediately.

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. Basic Laboratory Procedures in Clinical Bacteriology, J. Vandepitte, K. Engbaek, P. Piot, C.C. Heuck, W.H.O. Geneva, 1991.
2. Diagnostic Microbiology, Bailey & Scott, 9th Ed., Mosby 1994, Ellen Jo Baron, Lance R. Peterson.
3. Practical Medical Microbiology, Mackie & McCartney, Vol. 1, Microbial Infections, 13th Ed., Churchill Livingston 1978, Edited J.P. Duguid, B.P. Marmion, R.H.A. Swain.
4. Practical Medical Microbiology, Mackie & McCartney, Vol. 2, 13th Ed., Churchill Livingston 1989, Edited by J.G. Collee Duguid, A.G. Fraser, B.P. Marmion.
5. Handbook of Microbiological Media, Ronald M. Atlas, Lawrence C. Parks, 2nd Ed., 1997.
6. Detection, Prevention and Management of Urinary Tract Infections, C.M. Kunin, 4th Edition, 1987
7. Data on file: Microxpress®, A Division of Tulip Diagnostics (P) Ltd.

Product Presentation:

Cat. No.	Product Description	Pack Size
203050280012	Urine Culture Test	12 Tests