Kings Medium A Base

M1543

Intended Use:

Recommended for the non selective isolation, cultivation and pigment production of *Pseudomonas* species.

Composition**

Ingredients	Gms / Litre
Proteose peptone	20.000
Potassium sulphate	10.000
Magnesium chloride	1.640
Agar	15.000
Final pH (at 25°C)	7.3±0.1

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 46.64 grams in 1000 ml purified / distilled water containing 10 ml of glycerol . Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Aseptically pour into sterile Petri plates.

Principle And Interpretation

Pseudomonas aeruginosa is known to produce two types of pigments, pyocyanin and fluorescein which is a characteristic property and aids in isolation of *Pseudomonas* from clinical material. An additional pigment called as pyorubin was reported by King. Pyocyanin is green while fluorescein is fluorescent yellow and pyorubin is reddish brown. Some strains produce all these pigments while the others produce one or two pigments. *P.aeruginosa* can be identified on Hugh Leifson Medium (M826). Kings Medium A Base is particularly suited for the production of pyocyanin and pyorubin.

Kings Medium A Base is based on the formulation of King et al (1, 2). This medium can be used as a general medium for the non-selective isolation and pigment production of *Pseudomonas* species from foods, cosmetic samples etc.

These media contain proteose peptone, which provides carbonaceous and nitrogenous compounds for the growth of bacteria. Glycerol serves as a source of energy and also enhances pigment production. Magnesium chloride, potassium sulphate and magnesium sulphate also enhances pigment production. Pigments and/ or their derivatives produced by *Pseudomonas* species play a role as siderophores in the iron uptake systems of bacteria, and hence, their production is markedly enhanced under conditions of iron deficiency. For inoculation, use the organisms freshly cultured in Kings Medium A, incubate overnight at 37°C and then at room temperature for 6 days.

Type of specimen

Clinical samples - pus, urine, body fluids, Water samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (3,4).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards.(1)

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

In Vitro diagnostic use. Read the label before opening the container. The media should be handled by trained personnel only. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling clinical specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations :

1. For inoculation, use the organisms freshly cultured in Kings Medium A, incubate overnight at 37°C and then at room temperature for 6 days.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temmperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Gelling

Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Light yellow coloured, clear to slightly opalescent gel forms in Petri plates

Reaction

Reaction of 4.6% w/v aqueous solution (containing 1.0%v/v glycerol) at 25°C. pH : 7.3±0.1

pH

7.20-7.40

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organism	Inoculum (CFU)	Growth	Recovery	Pigment production
Pseudomonas aeruginosa ATCC 17934	50-100	good-luxuriant	>=70%	blue-green
Pseudomonas aeruginosa ATCC 27853	50-100	good-luxuriant	>=70%	blue-green
Pseudomonas aeruginosa ATCC 9027	50-100	good-luxuriant	>=70%	blue-green
Burkholderia cepacia ATCC 25609	50-100	good-luxuriant	>=70%	no pigment

Reference

1.King E. O., Ward M. K. and Raney D. E., 1954, J. Lab and Clin. Med., 44:301-307.
2.Murray P. R., Baron E. J., Jorgensen J. H., Pfaller M. A., Yolken R. H., (Eds.), 8th Ed., 2003, Manual of Clinical Microbiology, ASM, Washington, D.C.

Revision : 1 / 2011