



Kings Medium A Base

M1543

Kings Medium A is recommended for non-selective isolation, cultivation and pigment production of *Pseudomonas* species

Composition**

Ingredients	Gms / Litre
Proteose peptone	20.000
Potassium sulphate	10.000
Magnesium chloride, anhydrous	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 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.

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

M1543: Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

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

Storage and Shelf Life

Store below 30°C in tightly closed container and prepared medium at 2-8°C. Use before expiry period on the label

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., Tenover F. C., Tenover R. H., (Eds.), 8th Ed., 2003, Manual of Clinical Microbiology, ASM, Washington, D.C.

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