TM 1363 - KING'S MEDIUM B BASE

INTENDED USE

For non-selective isolation, cultivation and pigment production by *Pseudomonas* species.

PRODUCT SUMMARY AND EXPLANATION

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. Kings Medium B Base is particularly suited for fluorescein.

Kings Medium B Base is based on the formulation of King et al. 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. *Agrobacterium* have been traditionally identified as gram-negative bacteria that do not produce fluorescent pigment on Kings B medium and do produce tumors (or hairy roots) when inoculated onto test plants.

COMPOSITION

Ingredients	Gms / Ltr
Proteose peptone	20.000
Dipotassium hydrogen phosphate	1.500
Magnesium sulphate. heptahydrate	1.500
Agar	20.000

PRINCIPLE

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 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. The production of pigments especially non-fluorescent blue pigment, pyocyanin is readily demonstrated by culturing on Kings Medium B, which contains no added iron. The addition of dipotassium phosphate increases the phosphorus content of the medium thereby enhancing production of fluorescent pigment. For inoculation, use the organisms freshly cultured in Kings Medium A, incubate overnight at 37°C and then at room temperature for 6 days. With Kings Medium B, incubate at 37°C for 6 days.

INSTRUCTION FOR USE

- Dissolve 43.0 grams of dehydrated medium in 1000 ml distilled water containing 15 ml of glycerol.
- Heat to boiling to dissolve the medium completely.
- Mix well. Sterilize by autoclaving at 15 psi pressure (121°C) for 15 minutes.
- Aseptically pour into sterile Petri plates.

QUALITY CONTROL SPECIFICATIONS

Appearance of Powder : Cream to yellow homogeneous free flowing powder.
Appearance of prepared medium : Light yellow coloured, clear to slightly opalescent gel forms in Petri plates.
pH (at 25°C) : 7.2±0.2

INTERPRETATION

Cultural characteristics observed after an incubation.

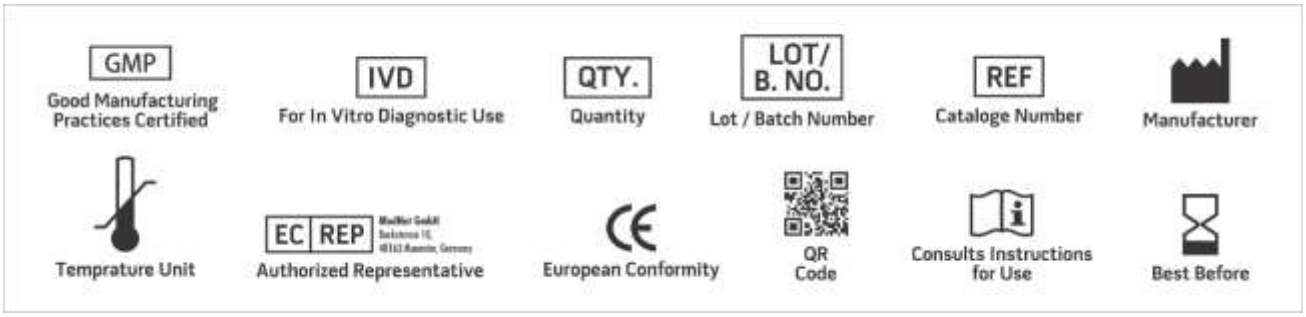
Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Recovery	Pigment production	Incubation Temperature	Incubation Period
Pseudomonas aeruginosa	17934	50-100	Good-luxuriant	>=50%	Greenish yellow	35 - 37°C	18-24 Hours
Pseudomonas aeruginosa	27853	50-100	Good-luxuriant	>=50%	Greenish yellow	35 - 37°C	18-24 Hours
Pseudomonas aeruginosa	9027	50-100	Good-luxuriant	>=50%	Greenish yellow	35 - 37°C	18-24 Hours
Burkholderia cepacia	25609	50-100	Good-luxuriant	>=50%	No pigment	35 - 37°C	18-24 Hours

PACKAGING:
In pack size of 500 gm bottles.

STORAGE
Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers between 25-30°C and protect from direct sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out replace the cap tightly to protect from hydration.
Product Deterioration: Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

DISPOSAL
After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

REFERENCES
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.
3. Ann G., Matthysse, 1998, The Genus Agraobacterium, Chapter 3.1.4. Martin Dworkin, 3rd Ed., The Prokaryotes, An Evolving Electronic Resource for the Micrbiological Community.
4. Todar K., Todars Online Textbook of Bacteriology, University of Wisconsin - Madison, Department of Bacteriology.



NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices.

***For Lab Use Only**

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