



Kovac's Reagent

RDM-R-01

Principle

The specific bacteria have ability to produce tryptophanase enzyme resulting in the hydrolysis of tryptophan in to the indole and α -aminopropionic acid. Indole produced by such microorganisms forms the red colored ring when reacted with the Kovac's Reagent. The Kovac's reagent is biochemical reagent consisting of p-dimethylaminobenzaldehyde (DMAB), isoamyl alcohol, and concentrated hydrochloric acid.

Use: Recommended for detection of the indole producing microbes for identification of variety of the microbes.

Ingredients

Per 100 ml

p-dimethylaminobenzaldehyde (DMAB)	5.000 gm
Amyl alcohol	75.000 ml
Hydrochloric Acid	25.000 ml

* Formula adjusted for optimum performance and parameters

Directions: Add 0.2 to 0.3 ml of the Kovac's reagent, in the 24-48 h old culture of the microorganism to be studied for the indole production. The development of the red colored ring indicates the positive test for the Indole production.

Specimens types analyzed

Clinical samples, water samples *etc.*

Precautions to be taken

All the handling, experiments, storage, and discarding should be performed with the help of trained and experienced technicians and as per the established guidelines. The material should be disposed only after proper serialization by autoclaving.

Performance and Evaluation

The expected performance of the medium is liable to use as per the direction on the label when stored at optimum conditions and within expiry date.

Quality Control	
Appearance	Green to yellow colored solution.
Solubility	Immiscible in water
Clarity	Clear with no precipitate or turbidity.

Different Microbial Response

Perform the indole production test using tryptophan broth (RDM-TpB-01). Inoculate TpB-01 (inoculum 50-100 CFU) with the desired test organism and incubate at 35 ± 2 °C for 18–24 hours. After incubation, add a few drops of Kovac's reagent and observe the results.

Organism	ATCC	Reaction with Kovac's Reagent
<i>Escherichia coli</i>	8739	Red colored ring formation ant the interface of the medium Positive reaction
<i>Klebsiella aerogenes</i>	13048	No Red colored ring formation Negative reaction

Storage and Shelf Life: The product is highly hygroscopic; keep the container tightly closed at all times and store it properly as per the conditions mentioned on the label. The declared expiry is valid only when stored as per the conditions mentioned on the label.

Note: Sterilize media immediately after reconstitution.

Disposal: To avoid the contamination or propagation of any hazardous microbes the used, unusable or modified preparation of this product must be disposed after autoclaving or incineration after completion of task.

Reference

1. Atlas, R. M. (2004). Handbook of microbiological media. CRC press.
2. Atlas, R. M. (2005). Handbook of media for environmental microbiology. CRC press.
3. American Public Health Association, & American Public Health Association. (2005). Standard methods for the examination of water and wastewater. APHA. American Water Works Association and Water Environment Federation, 21st ed.; American Public Health Association: Washington, DC, USA.

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