

Technical Data

KBM002 HiMotility[™] Biochemical kit for Salmonella

Introduction

KBM002 is a comprehensive test—system that can be used for identification of gram-negative Salmonella species. KBM002 identification kit can be used for screening pathogenic organisms from feces, urine, blood and other relevant clinical specimen. It can also be used for validating known laboratory strains. The complete list of organisms that can be identified with this system is given in the identification index provided with the kit.

Principle

Each KBM002 kit is a standardized colorimetric identification system utilizing seven conventional biochemical tests including motility and four carbohydrate utilization tests. The tests are based on the principle of pH change and substrate utilization. On incubation Salmonella exhibit metabolic changes which are indicated by a spontaneous color change in the media that can be either interpreted visually.

Kit contents

- 1. Each kit contains sufficient material to perform 10 tests.
- 2. 10 kits of KBM002.
- 3. Technical product insert.
- 4. Result Interpretation Chart and Result Entry Datasheet.

Instructions for use

1. Preparation of inoculum

- KBM002 cannot be used directly on clinical specimens. The organisms to be identified have to be first isolated and purified. Only pure cultures should be used.
- Isolate the organism to be identified on a common medium like Nutrient Agar (M001) or a differential medium like MacConkey Agar (M082). Pick up a single well isolated colony and inoculate in 5ml Brain Heart Infusion broth and incubate at 35-37°C for 4-6 hours until the inoculum turbidity is ≥ 0.1 OD at 620nm or 0.5 Mcfarland standard. Alternatively, a homogeneous suspension made in 2-3 ml sterile saline can be used for inoculation. The density of the suspension should be adjusted to 0.10D at 620nm or 0.5 Mcfarland standard.
- Note: Erroneous false negative results may be obtained if the inoculum turbidity is less than 0.1 OD.
 - Results are more prominent when enriched culture is used instead of suspension.

2. Inoculation of the kit:

- Open the kit aseptically. Peel off the sealing foil.
- Stab inoculate the 1st well. DO NOT INOCULATE THE 2ND WELL.
- Inoculate the remaining kit (well no.3-12) by stabbing each individual well (except well no. 2) with a loopful of inoculum. Inoculum should reach the bottom
 of the wells.
- 3 **Incubation**: Temperature of incubation: 35 37°C. Duration of incubation: 18 24 hours.

Interpretation of results

Interpret results as per the standards given in the Result Interpretation Chart.

Motility: Well No. 1

Motility is seen as movement of pink growth from 1st well to 2nd well.

Identification Index

Tests	Group I Strains	Motility	Motility	Citrate utilization	Urease	Arginine	Lysine	H₂S production	ONPG	Arabinose	Lactose	Maltose	Trehalose
Most serotypes		+	+	+	-	٧	+	+	-	+	-	+	+
Serotype Typhi		+	+	-	-	-	+	+	-	-	-	+	+
Serotype Choleraesu	iis subsp. choleraesuis	+	+	+	-	٧	+	+	-	+	-	+	+
Serotype Paratyphi A	4	+	+	-	-	٧	-	-	-	+	-	+	+
Serotype Gallinarum	1	-		-	-	-	+	+	-	V	-	+	V
Serotype Pullorum		-		-	-	-	+	+	-	+	-	-	+
S. serotype Typhimu	ırium	+	+	+	-	V	+	+	-	+	-	+	+
S. choleraesuis subs	sp. arizonae	+	+	+	-	V	+	+	+	+	٧	+	+
S. choleraesuis subs	sp. diarizonae	+	+	+	-	V	+	+	+	+	٧	+	+
S. choleraesuis subsp. houtenae		+	+	+	-	V	+	+	-	+	-	+	+
S. choleraesuis subsp. indica		+	+	V	-	V	+	+	V	+	V	+	+
S. choleraesuis subsp. salamae		+	+	+	-	+	+	+	V	+	-	+	+

Tests	Strains	Motility	Motility	Citrate utilization	Urease	Arginine	Lysine	H ₂ S production	ONPG	Arabinose	Lactose	Maltose	Trehalose
S. enterica subsp. salamae	Group II	-	ŀ	+	-	+	+	+	٧	+	-	+	+
S. enterica subsp. arizonae	Group IIIa	-	+	+	-	٧	+	+	+	+	V	+	+
S. enterica subsp. diarizonae	Group IIIb	-	ŀ	+	-	٧	+	+	+	+	٧	+	+
S. enterica subsp. houtenae	Group IV	-	+	+	-	٧	+	+	-	+	-	+	+
S. bongori	Group V Strains	-	+	+	-	+	+	+	+	+	-	+	+
S. enterica subsp. indica	VI Strains	-	+	٧	-	V	+	+	٧	+	V	+	+

Note: Based on % strains showing reactions following symbols have been assigned from laboratory results and standard references.

+ = Positive (more than 90%) - = Negative (more than 90%) V = Variable (11-89%)

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Result Interpretation chart

No.	Test	Reagents to be added after incubation	Principle	Original colour of the medium	Positive reaction	Negative reaction
1	Motility	_	_	Light pink	Dark pink growth	Light pink
2	Motility	_	Detects motility	Light pink	Movement of dark pink growth form 1 st well to 2 nd well	Light pink
3	Citrate utilization	_	Detects capability of organism to utilize citrate as a sole carbon source	Green	Blue	Green
4	Urease	_	Detects Urease activity	Orangish yellow	Pink	Orangish yellow
5	Arginine utilization	_	Detects arginine decarboxylation	Olive green to Light Purple	Purple / Dark Purple	Yellow
6	Lysine utilization	_	Detects Lysine decarboxylation	Olive green to Light Purple	Purple / Dark Purple	Yellow
7	H ₂ S production	_	Detects H ₂ S production	Orangish yellow	Black	Orangish yellow
8	ONPG	_	Detects β – galactosidase activity	Colourless	Yellow	Colourless
9	Arabinose	_	Carbohydrate utilization	Pinkish Red / Red	Yellow	Red / Pink
10	Lactose	_	Carbohydrate utilization	Pinkish Red / Red	Yellow	Red / Pink
11	Maltose	_	Carbohydrate utilization	Pinkish Red / Red	Yellow	Red / Pink
12	Trehalose	_	Carbohydrate utilization	Pinkish Red / Red	Yellow	Red / Pink

Important points to be taken into consideration while interpreting the result

- 1. In case of Carbohydrate fermentation test some microorganisms may show weak reaction. In this case record the reaction as ± and incubate further upto 48 hours. Orange colour after 48 hours of incubation should be interpreted as a negative reaction.
- 2. In case of Lysine, Arginine utilization, incubation up to 48 hours may be required.
- 3. At times organisms give contradictory result because of mutation or the media used for isolation, cultivation and maintenance.
- 4. The identification index has been compiled from standard references and results of tests obtained in the laboratory.

Precautions

- Clinical samples and microbial cultures should be considered potentially pathogenic and handled accordingly.
- Aseptic conditions should be maintained during inoculation and handling of the kits.

Disposal of used material

After use, kits and the instruments used for isolation and inoculation (pipettes, loops etc.) must be disinfected using a suitable disinfectant and then discarded by incineration or autoclaving in a disposable bag.

Storage and Shelf-life

On receipt store between 2-8 °C. Shelf-life is 12 months.

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