Staph Identification Kit

Intended Use
A panel of 12 tests for identification of *Staphylococcus* species (Kit contains sterile medium for Alkaline Phosphatase Detection, Voges Proskauer Test, ONPG Test, Arginine Dihydrolase Test, Urease Detection, and 7 different carbohydrates-Arabinose, Lactose, Mannitol, Sucrose, Raffinose, Trehalose, Maltose).

Summary
Staphylococci are Gram-positive, non-motile and are of major medical significance. Microxpress® Staph Identification Kit is a standardized system for identification of *Staphylococcus* species, which uses miniature biochemical tests. The complete list of organisms that is possible to identify with this system is given in the identification index provided at the end of this package insert.

Principle
Microxpress® Staph Identification Kit is a standardized identification system, comprising 12 miniature biochemical tests for identification of *Staphylococcus*. This kit contains sterile media for colorimetric identification using biochemical test and carbohydrate utilization tests based on principle of pH change and substrate utilization designed to identify various metabolic properties of different bacterial species. On incubation for an appropriate period, the media are examined for colour change. The results of these tests on the suspected organism are then compared to known standards to confirm its identification.

Kit Contents
1. 1 Kit of Staph Identification Kit
2. Technical Product Insert with Result Interpretation Chart, Result Entry Data Sheet and Identification Index
3. Barritt Reagent A (B-A) for Voges Proskauer Test
4. Barritt Reagent B (B-B) for Voges Proskauer Test
5. Creatine (CR) for Voges Proskauer Test

Note: Microxpress® Staph Identification Kit contains sufficient material to perform one test.

Biochemical Tests
Microxpress® Staph Identification Kit is a reagent set for laboratory use only.
Kit comprises of sterile test medium for:

a) Alkaline Phosphatase Detection (V16)
b) Voges Proskauer Test (V18)
c) ONPG Test (V11)
d) Arginine Dihydrolase Test (V1)
e) Urease Detection (V17)
f) Arabinose Utilization (V21)
g) Lactose Utilization (V28)
h) Mannitol Utilization (V30)
i) Sucrose Utilization (V 37)
j) Raffinose Utilization (V33)
k) Trehalose Utilization (V38)
l) Maltose Utilization (V29)

Additional Materials Required
0.9% saline, micropipettes, culture media, activated 2% glutaraldehyde solution, sterile test tubes, incubator / water bath at 35°C-37°C, 40% sodium hydroxide, sterile mineral oil.

Directions

Preparation of Inoculum:

1. Isolate the organism to be identified on Soyabean Casein Digest Agar (201190210500) or Nutrient Agar (201140030500).
2. Pick up 1-3 well isolated colonies and make a homogenous suspension in 2-3 mL sterile saline.
3. Match the turbidity of this suspension to McFarland standard number 0.5.

Note: Erroneous false negative results may be obtained if the inoculum turbidity is less than McFarland standard number 0.5.

Inoculation of the Vials:

1. Bring the kit components to room temperature before testing.
2. Open the kit aseptically.
3. Inoculate each vial with 100 µL of the above-prepared inoculum by surface inoculation method.
4. Overlay test vials V1 (for Arginine Dihydrolase Test) with sterile mineral oil.
5. Incubate at 35°C-37°C and read the result at 18-24 hours of incubation.
6. Alternatively, the kit can be inoculated by stabbing each individual well with a loopful of inoculum.

Voges Proskauer Test

1. Add 1 - 2 drops of Creatine, 2 - 3 drops of Baritt Reagent A and 1 - 2 drops of Baritt Reagent B to the test vial V18.
2. Development of pinkish red colour within 5-10 minutes a positive test.
3. No colour change or slight copper colour (due to reaction of Barritt reagent A with Barritt reagent B) indicates a negative test.

Alkaline Phosphate Test

1. Add 1-2 drops of 40% sodium hydroxide to the test vial V16.
2. Development of bright pink colour within a few seconds indicates a positive test.
3. No colour development indicates a negative test.

Identification Index

Organisms / Tests	Voges Proskauer Test	Alkaline Phosphatase Test	ONPG Test	Urease Detection	Arginine Dihydrolase Test	Mannitol Utilization	Sucrose Utilization	Lactose Utilization	Arabinose Utilization	Raffinose Utilization	Trehalose Utilization	Maltose Utilization
Staphylococcus aureus	+	+	-	W	+	+	+	+	-	-	+	+
Staphylococcus epidermidis	+	+	-	+	W	-	+	d	-	-	-	+
Staphylococcus haemolyticus	d	-	-	-	+	d	+	d	-	-	+	+
Staphylococcus lugdunensis	+	-	-	d	-	-	+	+	-	-	+	+
Staphylococcus saprophyticus subspecies *saprophyticus*	+	-	d	+	W	d	+	d	-	-	+	+
Staphylococcus schleiferi subspecies *schleiferi*	+	+	d	-	+	-	-	-	-	-	d	-

Organisms / Tests	Voges Proskauer Test	Alkaline Phosphatase Test	ONPG Test	Urease Detection	Arginine Dihydrolase Test	Mannitol Utilization	Sucrose Utilization	Lactose Utilization	Arabinose Utilization	Raffinose Utilization	Trehalose Utilization	Maltose Utilization
Staphylococcus arlettae	ND	(+)	ND	-	-	+	+	+	+	+	+	+
Staphylococcus auricularis	d	-	(d)	-	d	-	d	-	-	-	+	+
Staphylococcus capitis	d	-	-	-	d	+	(+)	-	-	-	-	-
Staphylococcus caprae	+	+	-	+	+	d	-	+	-	-	+	d
Staphylococcus cohnii	d	-	-	-	-	d	-	-	-	-	+	(d)
Staphylococcus hominis	d	-	-	+	d	-	(+)	d	-	-	d	+
Staphylococcus simulans	W	W	+	+	+	+	+	+	-	-	d	W
Staphylococcus warneri	+	-	-	+	d	d	+	d	-	-	+	(+)
Staphylococcus xylosus	d	d	+	+	-	d	+	d	+	-	+	+
Staphylococcus caseolyticus	-	-	-	-	V	-	V	+	-	-	+	+
Staphylococcus carnosus subspecies *carnosus*	+	+	+	-	+	+	-	d	-	-	d	-
Staphylococcus chromogens	-	+	-	d	+	d	+	+	-	-	+	d
Staphylococcus delphini	-	+	ND	+	+	+	+	+	-	ND	-	ND
Staphylococcus equorum	ND	+	+	+	-	+	+	d	+	-	+	+
Staphylococcus felis	-	+	+	+	+	d	d	+	-	-	+	-
Staphylococcus gallinarum	-	+	W	+	-	+	+	d	+	+	+	+
Staphylococcus hyicus	-	+	-	d	+	-	+	+	-	-	+	-
Staphylococcus intermedius	-	+	d	+	d	(d)	+	d	-	-	+	(W)
Staphylococcus kloosii	ND	+	ND	d	-	+	-	d	d	W	+	+
Staphylococcus sciuri	W	d	-	-	-	+	+	d	d	-	+	(d)

Key:

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

+: 90% or more strains are positive; -: 90% or more strains are negative; ND: Not Determined; V: Variable; d: 11-89% of strains are positive; ( ): Delayed reaction; W: Weak reaction.

Result Interpretation Chart

Code	Test	Reagent to be added	Principle	Original colour of medium	Positive reaction	Negative reaction
V18	Voges Proskauer Test	1-2 drops of Creatine, 2-3 drops of Barritt reagent A and 1-2 drops of Barritt reagent B	Detects acetoin production	Colourless to cream	Pinkish red within 5-10 minutes	Colourless / Slight copper
V16	Alkaline Phosphatase Test	1-2 drops of 40% NaOH	Detects ability of organism to produce sufficient phosphatase enzyme	Cream	Pink	Cream
V11	ONPG Test	-	Detects β-galactosidase activity	Colourless to cream	Yellow	Colourless to cream
V17	Urease Detection	-	Detects urease activity	Orangish yellow	Pink	Orangish yellow
V1	Arginine Dihydrolase Test	-	Detects arginine decarboxylation	Reddish purple	Purple	Yellow
V30	Mannitol Utilization	-	Detects mannitol utilization	Red	Yellow	Red / Pink
V37	Sucrose Utilization	-	Detects sucrose utilization	Red	Yellow	Red / Pink
V28	Lactose Utilization	-	Detects lactose utilization	Red	Yellow	Red / Pink
V21	Arabinose Utilization	-	Detects arabinose utilization	Red	Yellow	Red / Pink
V33	Raffinose Utilization	-	Detects raffinose utilization	Red	Yellow	Red / Pink
V38	Trehalose Utilization	-	Detects trehalose utilization	Red	Yellow	Red / Pink
V29	Maltose Utilization	-	Detects maltose utilization	Red	Yellow	Red / Pink

Result Entry Data Sheet

Sample Number	V18 Voges Proskauer Test	V16 Alkaline Phosphatase Test	V11 ONPG Test	V17 Urease Detection	V1 Arginine Dihydrolase Test	V30 Mannitol Utilization
Sample Number	V37 Sucrose Utilization	V28 Lactose Utilization	V21 Arabinose Utilization	V33 Raffinose Utilization	V38 Trehalose Utilization	V29 Maltose Utilization

Interpretation of Results

1. Add the reagents in the required vials at the end of incubation period.
2. Interpret results as per the standards given in the result interpretation chart.

Remarks

1. Microxpress® Staph Identification Kit is an *In vitro* diagnostic kit for laboratory and professional use only. Not for medicinal use.
2. This kit cannot be used directly on clinical specimens. Only pure cultures should be used to obtain optimum results.
3. Do not use damaged or leaking kits. Avoid contact of reagents with skin and eyes.
4. Erroneous false negative results may be obtained if inoculum turbidity is less than McFarland standard number 0.5.
5. At times, the organism may give contradictory results because of mutation or media used for isolation, cultivation and maintenance. Results are prominent when fresh and enriched culture is used.
6. In case of carbohydrate fermentation some microorganisms may show weak reaction. Incubate further for 48 hours. Orange colour seen after 48 hours should be a negative reaction.
7. In case of arginine dihydrolase test, incubation up to 48 hours may be required.
8. Identification index has been compiled based on standard references and results of tests obtained in the laboratory.
9. Clinical samples and microbial cultures should be considered as pathogenic biohazard and handled accordingly. Good laboratory practices and hazard precautions must be observed at all times.

Storage and Stability

1. Store the kit at 2°C-8°C. Do Not Freeze.
2. Stability of the kit is as per the expiry date mentioned on the label.

Warranty

This product is designed to perform as described on the label and package insert. The manufacturer disclaims any implied warranty of use and sale for any other purpose.

Reference

1. Practical Medical Microbiology, Mackie & McCartney, 13th edition 1989, Edited by J. G. Collee, J. P. Duguid.
2. Clarke P.H. And S.T. Cowan, Biochemical Methods for Bacteriology, J. Gen. Microbiol., 1952, Vol. 6: 187-197.
3. A. L. Barry and K. L. Feeney, Two quick methods for Voges-Proskauer test, Applied Microbiology, Sept. 1967, p.: 1138-1141.
4. Coblentz, L.H 1943, Rapid detection of the production of acetyl-methyl-carbinol, Am. J. Pub. Health 33:815-817.
5. Rapid Arginine Decarboxylase Test For The Identification Of Enterobacteriaceae, 1972, Applied Microbiol; 23, p 710-713.
6. Five Test Simple Scheme For Species-Level Identification Of Clinically Significant Coagulase Negative Staphylococcus, 2003, Adriana N.De Paulis *et al.,* J.Clin. Microbiol; 41:P 1219-1224.
7. Bascomb,S And M.Manafi, 1998. Use Of Enzyme Tests In Characterization And Identification Of Aerobic And Facultative Anaerobic Gram-Positive Cocci, Clin. Microbial.Rev; 11: 318-340.
8. Bergey's Manual of Determinative Bacteriology, 9th edition 1994; Edited by John G. Holt, Noel R. Krieg.
9. Murray, P. R. and *et al.,* Manual of Clinical Microbiology Vol. 1, ASM, 8th Edition, 2003.
10. Koneman. E. W and *et al.,* Color Atlas and Textbook of Diagnostic Microbiology lippincoh, 6th Edition, 2006.
11. Bergey's Manual of Systematic Bacteriology, The Firmicutes, 2nd edition, Vol. 3.
12. Data on file: Microxpress®, A Division of Tulip Diagnostics (P) Ltd.

Product Presentation:

Cat. No.	Product Description	Pack Size
203190850001	Biochemical Identification Kit	1 Kit (1 Test)