Technical Data

HiNeisseria™ Identification Kit

KB008

Intended use

KB008 is a combination of 12 tests for identification of *Neisseria* species from clinical specimen using pure isolate. It can also be used for validating known laboratory strains.

Kit contents

1. Each kit contains 5/10/20 kits of KB008 sufficient material to perform 5/10/20 tests. Kit contains sterile media for Urease, ONPG, Voges Proskauer's, Oxidase, Catalase, Nitrate reduction, Glucose, Maltose, Lactose, Sucrose, Fructose, Mannose

2.Sulphanilic Acid 0.8% (R015) for Nitrate test
3.N,N-Dimethyl-1-Napthylamine Reagent (R009) for Nitrate test
4.Barritt reagent A (R029) for VP test
5.Barritt reagent B (R030) for VP test
6.Gordon McCleod Reagent (R026) for Oxidase test
7.Result Interpretation Chart and Result Entry Datasheet
8.Identification Index.
9.Technical product insert

Material Required but not supplied

1. McFarland standard
2. Inoculation loops, pipettes
3. Enrichment medium / Isolation media

Direction

Preparation of inoculum :

- Isolate the organism to be identified on a selective medium like Thayer Martin Medium Base (M413) or Chocolate Agar (M103).
- Pick up a single isolated colony and inoculate in 5 ml BHI Broth (M210). Incubate at 35-37°C for 4-6 hours in 5 -10% CO_2 and 70% humidity till the inoculum density is greater than or equal to 0.1 OD at 620nm or 0.5 Mcfarland standard.

Inoculation of the kit :

1. Open the kit aseptically. Peel off the sealing foil.
2. Inoculate each well with 50 µl of the above inoculum by surface inoculation method.
3. Alternatively, the kit can also be inoculated by stabbing each individual well with a loopful of inoculum.

Incubation :

Temperature of incubation : 35 - 37°C in 5 -10 % CO_2 and 70% humidity Duration of incubation : 18 - 24 hours.

Interpretation of results :

Interpret results as per the standards given in the Result Interpretation Chart. Addition of reagents in well no 3,4,5 and 6 should be done at the end of incubation period that is after 18 - 48 hours.

Principle

Each KB008 is a standardized colorimetric identification system utilizing six conventional biochemical tests and six carbohydrate utilization tests. The tests are based on the principle of pH change and substrate utilization. On incubation, organisms undergo metabolic changes which are indicated by a colour change in the media that is either visible spontaneously or after addition of a reagent.

Type of specimen

Pure isolate from clinical specimen.

Specimen collection and handling

Refer direction

Warning and Precautions

In Vitro diagnostic Use. For professional use only. Read the label before opening the pack. Wear protective gloves/ protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling clinical specimens. Aseptic conditions should be maintained during inoculation and handling of the kits. Reagents should not come in contact with skin, eyes or clothing. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Allow the reagents to come to room temperature after removal from the refrigerator .
2. In case of carbohydrate fermentation test some microorganisms 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.
3. At times organisms give conflicting 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 carried out in the laboratory.
5. Erroneous false negative results may be obtained if the inoculum turbidity is less than 0.1 OD.
6. Results are more prominent if an enriched culture is used instead of a suspension.
7. It cannot be used directly for clinical specimens. The microorganisms to be identified have to be first isolated on appropriate isolation media. Only pure cultures should be used.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Sterile white opaque strip with 12 wells containing media of Urease, ONPG, Voges Proskauer's, Oxidase, Catalase, Nitrate reduction, Glucose, Maltose, Lactose, Sucrose, Fructose, Mannose

Quantity of medium

0.8 ml of medium in each well.

Sterility Check

Passes release criteria

Interpretation of results :

Interpret results as per the result interpretation chart.

1.Urease Test : Well No. 1

- Positive test is indicated by a colour change to pink colour.
- No colour change indicates a negative reaction.

2.ONPG Test:Well No. 2

- Medium changes from colourless to yellow if the test is positive.
- Medium remains colourless if the test is negative.

3.Voges-Proskaeur's Test: Well No.3

- Add 2-3 drops of Baritt reagent A (R029) and 1 drop of Baritt reagent B (R030).
- Positive test is indicated by a development of pinkish red colour in 5 - 10 minutes.
- No colour change or a copper colour (due to reaction of Reagent A and Reagent B) indicates a

4.Oxidase Test: Well No. 4

- Add 1-2 drops of Gordon McCleod Reagent (R026).
- Change in colour from colourless to deep purple colour within 10 seconds indicates positive reaction.
- Colourless after 60 seconds indicates negative reaction.

5.Catalase test: Well No. 5

- Add 2-3 drops of Hydrogen peroxide reagent.
- Effervescence indicates positive reaction.
- No effervescence indicates negative reaction.

6.Nitrate Reduction Test : Well No. 6

- Add 1-2 drops of Sulphanilic acid (R015) and 1-2 drops of N,N-Dimethyl-1-Napthylamine Reagent (R009).
- Immediate development of pinkish red colour on addition of reagent indicates positive reaction.
- No change in colour indicates a negative reaction.

7.Carbohydrate fermentation Test : Well No. 7-12

- Positive test is indicated by a colour change to yellow colour.
- Red or no colour change indicates a negative reaction.

Result Interpretation chart

No.	Test	Reagents to be added after incubation	Principle	Original colour of the medium	Positive reaction	Negative reaction
1	Urease	—	Detects Urease activity	Orangish yellow	Pink	Orangish yellow
2	ONPG	—	Detects β–galactosidase activity	Colourless	Yellow	Colourless
3	Voges Proskauer's	1-2 drops of Baritt reagent A and 1-2 drops of Baritt reagent B	Detects acetoin production	Colourless/light yellow	Pinkish red	Colourless/ slight copper
4.	Oxidase	1-2 drops of Gordon McCleod Reagent (R026)	Detects Cytochrome oxidase production	Colourless	Deep purple	No change in or purplish blue colour after 60 seconds
5.	Catalase	3% H_2O_2	Detects Catalase activity	Colourless	Effervescenence coming out from the loop	No effervescence
6	Nitrate reduction	1-2 drops of sulphanilic acid and 1-2 drops of N, N-Dimethyl-1-Napthylamine	Detects Nitrate reduction	Colourless	Pinkish red	Colourless
7	Glucose	—	Glucose utilization	Pinkish Red /Red	Yellow	Red / Pink
8.	Maltose	—	Maltose utilization	Pinkish Red /Red	Yellow	Red / Pink
9	Lactose	—	Lactose utilization	Pinkish Red /Red	Yellow	Red / Pink
10.	Sucrose	—	Sucrose utilization	Pinkish Red /Red	Yellow	Red / Pink
11.	Fructose	—	Fructose utilization	Pinkish Red /Red	Yellow	Red / Pink
12.	Mannose	—	Mannose utilization	Pinkish Red /Red	Yellow	Red / Pink

Result Entry Datasheet

No.	Test	1	2	3	4	5	6	7	8	9	10
1	Urease										
2	ONPG										
3	Voges Proskauer's										
4	Oxidase										
5	Catalase										
6	Nitrate reduction										
7	Glucose										
8	Maltose										
9	Lactose										
10	Sucrose										
11	Fructose										
12	Mannose										

Identification Index of various *Neisseria species*

Tests	Urease	ONPG	Voges Proskauer's	Oxidase	Catalase	Nitrate reduction	Glucose	Maltose	Lactose	Sucrose	Fructose	Mannose
N. animalis	–	–	–	+	+	–	+	–	–	+	+weak	Nd
N. canis	–	–	–	+	+	+	–	–	–	–	–	–
N. cinerea	–	–	–	+	+	–	–	–	–	–	–	–
N. dinetrificans	–	–	–	+	+	–	+	–	–	+	+	+
N. elongata subsp. elongata	–	–	–	+	–	–	V	–	–	–	–	–
N. flavescens	–	–	–	+	+	–	–	–	–	–	–	–
N. gonorrhoeae	–	–	–	+	+	–	+	–	–	–	–	–
N. iguanae	–	–	–	+	+	–	V	–	–	–	Nd	Nd
N. lactamica	–	+	–	+	+	–	+	+	+	–	–	–
N. macacae	–	–	–	+	+	–	+	+	–	+	+	Nd
N. meningitidis	–	–	–	+	+	–	+	+	–	–	–	–
N. mucosa	–	–	–	+	–	+	+	+	–	+	+	–
N. perflava	–	–	–	+	+	–	+	+	–	+	+	Nd
N. polysaccharea	–	–	–	+	+	+	+	+	–	V	–	Nd
N. sicca	–	–	–	+	+	–	+	+	–	+	+	–
N. subflava	–	–	–	+	+	–	+	+	–	V	V	–
N. weaveri	–	–	–	+	+	–	–	–	–	–	Nd	Nd

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% positive)
Nd = Not detected.

Storage and Shelf Life

On receipt store between 2-8°C. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

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 disposal bag. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (2,3).

Reference

1.Bergey's Manual of Systematics of Archaea and Bacteria (BMSAB), 2015.
2.Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
3.Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

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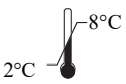
IVD ***In vitro* diagnostic medical device**



Storage temperature

EC REP CEpartner4U, Esdoornlaan 13,
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CE **CE Marking**

Do not use if package is damaged

Do not re-use

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