

## MASTDISCS® ID Nitrate Discs

### D51/D51C

#### Intended use

For the detection of nitrate reductase activity in anaerobes.

FOR IN VITRO DIAGNOSTIC USE ONLY

#### Contents

100 discs in a vial (D51) or a pack of 5 cartridges (D51C), each cartridge containing 50 discs.

#### Formulation\*

Material:	Content per disc:
Potassium nitrate	40%
Sodium molybdate	0.1%

#### Storage and shelf life

Store at 2 to 8°C in the containers provided until the expiry date shown on the pack label. Allow to equilibrate to room temperature before opening.

#### Precautions

For *in vitro* diagnostic use only. Observe approved biohazard precautions and aseptic techniques. To be used only by adequately trained and qualified laboratory personnel. Sterilise all biohazard waste before disposal. Refer to Product Safety Data sheet.

#### Materials required but not provided

Standard microbiological supplies and equipment such as loops, MAST® culture media, swabs, applicator sticks, incinerators and incubators, etc., as well as serological and biochemical reagents and additives such as blood.

#### Procedure

- Using a pure, fresh culture of the test organism, prepare a suspension equivalent in density to a McFarland 2 opacity standard.
- Using a sterile swab spread the suspension uniformly across the surface of a plate containing a medium suitable for the culture of anaerobes (e.g. MAST® Columbia Agar DM115D) supplemented with 5 to 7% lysed blood.
- Using a sterile needle or forceps, place a Nitrate Disc on to the inoculated medium.
- Incubate at 35 to 37°C for 24 to 48 hours in anaerobic conditions
- Remove the disc from the surface of the plate and place it in a clean Petri dish or on a slide.
- Add one drop each of N,N-dimethyl alpha naphthylamine (or 1,6-Cleve's Acid) and sulfanillic acid reagents to the disc.
- Observe for the development of a red colour occurring within 3 to 5 minutes.

- If the results from step 7 are negative, confirm by the addition of a small amount of zinc dust to the disc.
- Observe for the development of a pink/red colour occurring within 5 to 10 minutes.

#### Interpretation of results

Positive - Red or pink colour development after adding the reagents or no colour development after adding zinc dust.

Negative - No colour development after adding the reagents and red colour development after adding zinc dust.

#### Quality control

Check for signs of deterioration. Quality control must be performed with at least one organism to demonstrate a positive reaction and at least one organism to demonstrate a negative reaction. Do not use the product if the reactions with the control organisms are incorrect. The list below illustrates a range of performance control strains which the end user can easily obtain.

Test Organisms	Result
<i>Bacteroides ureolyticus</i> ATCC® 33387	Positive
<i>Bacteroides fragilis</i> ATCC® 25285	Negative

#### Limitations

It is recommended that further biochemical and/or serological tests are performed on colonies from pure culture to confirm identification.

Rapidly growing organisms may cause the nitrate discs to turn a tan colour as a result of haemolysis and/or metabolism. Addition of test reagents may cause only a subtle colour change or no colour change at all. In the event that this reaction occurs, it is recommended that some other means of nitrate reduction testing is employed.

Organisms that produce only light or non-confluent growth may fail to produce a sufficient quantity of nitrate reductase and may therefore produce false negative results.

#### References

Bibliography available on request.