

MAST® ID Indole Agar

IDM34

Intended use

Indole Agar dehydrated culture medium powder is intended for use to produce a medium to aid differentiation of members of the Enterobacteriales family based on their ability to decompose the amino acid tryptophan to indole. When prepared in accordance with the instructions for use, it produces a semi-solid medium capable of supporting the growth of Enterobacteriales. The metabolic action of an organism able to deaminate tryptophan forms indole which accumulates in the medium. When indole is combined with a separate developing reagent, 4-(Dimethylamino) cinnamaldehyde (DMACA) at an acidic pH (the reagent contains hydrochloric acid) a blue coloured product is formed which is interpreted as a positive result. This medium can also be used in conjunction with additional identification products to produce a phenotypic biochemical profile of the bacterial isolate in order to generate specific fingerprints or datasets that can be used, for example, to detect or rule out cross-infection or elucidate bacterial transmission.

Indole Agar is intended to be used in conjunction with other phenotypic tests to aid epidemiological typing of previously isolated and identified pure cultures of members of the Enterobacteriales family derived from, animal, food, environmental or human samples. It is a non-automated, qualitative device, intended to be used by professional, trained laboratory users for *in vitro* use and is not intended for use in the diagnosis of disease or other conditions in humans or as the basis of treatment or case management decisions.

Contents

See pack label.

Formulation*

| Material: | Concentration in medium: |
|---------------------|--------------------------|
| Enzymic casein | 15.0 g/litre |
| Soy peptone | 5.0 g/litre |
| Sodium chloride | 5.0 g/litre |
| Agar | 20.0 g/litre |
| Final pH: 7.3 ± 0.2 | |

Storage and shelf life

All dehydrated culture media containers should be kept tightly closed and stored in a dry place at 10 to 25°C until the expiry date shown on the pack label.

Precautions

For *in vitro* use only. Observe approved hazard 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 (available on request or via MAST® website). Indole Agar is not intended for use in the diagnosis of disease or other conditions in humans.

Observe recommended safe laboratory practices when preparing and handling DMACA reagent.

Materials required but not provided

Standard microbiological supplies and equipment such as loops, MAST® selective supplements, swabs, applicator sticks, incinerators and incubators, etc. Separate reagents: 4-(Dimethylamino) cinnamaldehyde (DMACA), CAS No. 6203-18-5 and solvent, hydrochloric acid.

Procedure

1. Refer to pack label for quantities and volumes required. Prepare MAST® ID Indole Agar (IDM34/A) by suspending the powder in distilled or deionised water. For sachet packs, dissolve the entire contents of the sachet in the volume shown on the label.
2. Sterilise by autoclaving at 121°C (15 p.s.i.) for 15 minutes.
3. Mix well and pour culture plates (15 to 20ml per plate) into Petri dishes which have been labelled using the self-adhesive labels provided. Self adhesive labels are provided in each box of preweighed sachets.
4. Prepared culture plates may be used immediately or stored in plastic bags at 2 to 8°C for up to one week.
5. Prepare a suspension of each organism equivalent in density to a 0.5 McFarland standard. Inoculate the surface of a well-dried plate using a replicating device, e.g. the SCANURIDOT Multipoint Inoculator, to deliver each inoculum onto the agar surface.
6. Allow the inoculum drops to dry before disturbing and incubate plates aerobically for 18 to 24 hours at 35 to 37°C.
7. After incubation of plates localise growth spots using a No. 7 cork borer or scalpel blade to cut the agar. Add 1 drop of DMACA reagent, as prepared below, to each spot.
8. Dissolve 1g of 4-(Dimethylamino) cinnamaldehyde (DMACA), CAS No. 6203-18-5, in 100ml of 10% v/v concentrated hydrochloric acid. The solution can be stored in the dark for up to 2 months at room temperature. DMACA reagent is classified as Irritant.

Interpretation of results

On addition of the DMACA reagent record any colour development occurring within 1 minute. A positive result indicating formation of indole is shown by development of blue colour. A negative result is indicated by a pink colour.

Quality control

Check for signs of deterioration. Quality control must be performed with at least one organism to demonstrate expected performance. Do not use the product if the result with the control organism is incorrect. The list below illustrates a range of performance control strains which the end user can easily obtain.

| Test Organisms | |
|---|----------|
| <i>Escherichia coli</i> ATCC® 25922 | Positive |
| <i>Klebsiella pneumoniae</i> ATCC® 13883 | Negative |

References

Bibliography available on request.