INTENDED USE

BD Yersinia Selective Agar (=CIN Agar, Cefsulodin-Irgasan-Novobiocin Agar) is a selective differential medium for the isolation of Yersinia enterocolitica, and BD Aeromonas Yersinia Agar is a selective differential medium for the isolation of both Yersinia enterocolitica and Aeromonas spp. from clinical specimens.

PRINCIPLES AND EXPLANATION OF THE PROCEDURE

Microbiological method.

BD Yersinia Selective Agar was first described by Schiemann as an alternative to MacConkey Agar and other commonly used media for isolation of Yersinia enterocolitica, a causative agent of gastroenteritis.¹ The medium has been found to be superior to MacConkey, SS, CAL or Y agars.² BD Aeromonas Yersinia Agar is a modification of BD Yersinia Selective Agar that, in addition to Yersinia enterocolitica, supports growth of Aeromonas species (which also may cause enteritis) since it has a reduced cefsulodin content.³,⁴

In both media, peptones provide nutrients. Fermentation of mannitol in the presence of neutral red results in a characteristic "bull's-eye" colony of Y. enterocolitica, colorless with red centers on both media. Selective inhibition of gram-negative and gram-positive organisms is obtained by means of crystal violet, sodium desoxycholate and the antimicrobial agents, cefsulodin, Irgasan® (Triclosan), and novobiocin. On BD Aeromonas Yersinia Agar, Aeromonas species produce pale colonies that have a rose to red center, similar to Yersinia. Both organisms may be differentiated from each other by means of the oxidase reaction (positive for Aeromonas only).³,⁵

BD Yersinia Selective Agar and BD Aeromonas Yersinia Agar may also be used for the isolation of Yersinia species other than Y. enterocolitica, e.g., for Y. pseudotuberculosis.⁵

REAGENTS

Formulas* Per Liter Purified Water

<table>
<thead>
<tr>
<th>BD Yersinia Selective Agar</th>
<th>Pancreatic Digest of Gelatin 10.0 g</th>
<th>Magnesium Sulfate 0.001 g</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Peptic Digest of Animal Tissue 5.0 g</td>
<td>Crystal Violet 0.001 g</td>
</tr>
<tr>
<td></td>
<td>Beef Extract 5.0 g</td>
<td>Neutral Red 0.03 g</td>
</tr>
<tr>
<td></td>
<td>Yeast Extract 2.0 g</td>
<td>Cefsulodin 0.015 g</td>
</tr>
<tr>
<td></td>
<td>Mannitol 20.0 g</td>
<td>Irgasan 0.004 g</td>
</tr>
<tr>
<td></td>
<td>Sodium Pyruvate 2.0 g</td>
<td>Novobiocin 0.0025 g</td>
</tr>
<tr>
<td></td>
<td>Sodium Chloride 1.0 g</td>
<td>Agar 12.0 g</td>
</tr>
<tr>
<td></td>
<td>Sodium Desoxycholate 0.5 g</td>
<td></td>
</tr>
</tbody>
</table>

pH 7.4 +/- 0.2

Instead of 0.015 g of cefsulodin in BD Yersinia Selective Agar, BD Aeromonas Yersinia Agar contains only 0.004 g of cefsulodin per liter medium.

*Adjusted and/or supplemented as required to meet performance criteria.

PRECAUTIONS

For professional use only. ☹

Do not use plates if they show evidence of microbial contamination, discoloration, drying, cracking or other signs of deterioration.

Consult GENERAL INSTRUCTIONS FOR USE document for aseptic handling procedures, biohazards, and disposal of used product.
STORAGE AND SHELF LIFE
On receipt, store plates in the dark at 2 to 8°C, in their original sleeve wrapping until just prior to use. Avoid freezing and overheating. The plates may be inoculated up to the expiration date (see package label) and incubated for the recommended incubation times. Plates from opened stacks of 10 plates can be used for one week when stored in a clean area at 2 to 8°C.

USER QUALITY CONTROL
Inoculate representative samples with the following strains (for details, see GENERAL INSTRUCTIONS FOR USE document). Incubate plates aerobically at 25 +/- 2°C or 35 +/- 2°C and read after 20 to 24 and 42 to 48 hours.

<table>
<thead>
<tr>
<th>Strains</th>
<th>BD Yersinia Selective Agar (CIN Agar)</th>
<th>BD Aeromonas Yersinia Agar</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Aeromonas hydrophila</em></td>
<td>Inhibition partial to complete</td>
<td>Growth fair to good; colorless to pale rose colony with a rose to red center</td>
</tr>
<tr>
<td>ATCC™ 7966</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Yersinia enterocolitica</em></td>
<td>Growth good to excellent; pale rose colony with a dark red center (“bull’s eye” colony)*</td>
<td>Growth good to excellent; pale rose colony with a dark red center (“bull’s eye” colony)*</td>
</tr>
<tr>
<td>ATCC 9610</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Escherichia coli</em> ATCC 25922</td>
<td>Inhibition complete</td>
<td>Inhibition complete</td>
</tr>
<tr>
<td><em>Enterococcus faecalis</em></td>
<td>Inhibition partial to complete</td>
<td>Inhibition partial to complete</td>
</tr>
<tr>
<td>ATCC 29212</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Proteus mirabilis</em> ATCC 14153</td>
<td>Inhibition partial to complete</td>
<td>Inhibition partial to complete</td>
</tr>
<tr>
<td>ATCC 27853</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Pseudomonas aeruginosa</em></td>
<td>Inhibition partial to complete</td>
<td>Inhibition partial to complete, fair growth acceptable</td>
</tr>
<tr>
<td>ATCC 25923</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Staphylococcus aureus</em></td>
<td>Inhibition partial to complete</td>
<td>Inhibition partial to complete</td>
</tr>
<tr>
<td>ATCC 25923</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Uninoculated</td>
<td>Light pink, slightly opalescent</td>
<td></td>
</tr>
</tbody>
</table>

* May be completely pink after 42 to 48 hours of incubation.

PROCEDURE
Materials Provided
BD Yersinia Selective Agar (CIN Agar) or BD Aeromonas Yersinia Agar, both provided in 90 mm Stacker™ plates. Microbiologically controlled.

Materials Not Provided
Ancillary culture media, reagents and laboratory equipment as required.

Specimen Types
BD Yersinia Selective Agar is a differential selective medium for the isolation of *Yersinia enterocolitica*, and BD Aeromonas Yersinia Agar is a selective differential medium for the isolation of *Aeromonas* species and *Yersinia enterocolitica* from human stool specimens or rectal swabs (see also PERFORMANCE CHARACTERISTICS AND LIMITATIONS OF THE PROCEDURE).

Test Procedure
Streak the specimen as soon as possible after it is received in the laboratory. The streak plate is used primarily to isolate pure cultures from specimens containing mixed flora. Alternatively, if material is being cultured directly from a swab, roll the swab over a small area of the surface at the edge; then streak from this inoculated area. *Aeromonas* and *Yersinia* species grow well between 25 and 37°C. The lower temperature is optimal for *Yersinia* and is recommended for primary isolation. Incubate plates at 25 to 32°C for 24 to 48 h.
A less selective medium, such as BD MacConkey II Agar, should also be inoculated with the specimen (and incubated at 35 +/- 2° C) for the detection of other pathogens involved in the infection.

A "cold enrichment" procedure may occasionally be necessary for the isolation of Yersinia enterocolitica, from stool specimens and other materials such as food: Inoculate about 1 gram of sample into 8 to 12 ml phosphate-buffered saline (pH 7.2) and hold at 4°C for up to 21 days. Enrichment of Aeromonas can be performed in alcaline peptone water. Periodically subculture from the respective enrichment onto plates of either medium described in this document, streaking for isolation. Incubate plates as stated above.

Results
Typical Yersinia enterocolitica colonies will have deep-red centers surrounded by a transparent, pale border giving the appearance of a "bull's-eye" on BD Yersinia Selective Agar and BD Aeromonas Yersinia Agar after 24 hours of incubation. After 42 to 48 hours of incubation, they are often completely pink. Yersinia pseudotuberculosis usually lacks the transparent zone around the colonies. Aeromonas produces paler colonies which also have a rose to red center and are oxidase positive. Aeromonas can be easily differentiated from Yersinia and other Enterobacteriaceae by a standard oxidase test (only Aeromonas spp. will give a positive result). Biochemical and serological confirmation is necessary for a complete identification of suspicious isolates.

PERFORMANCE CHARACTERISTICS AND LIMITATIONS OF THE PROCEDURE
BD Yersinia Selective Agar (CIN) and BD Aeromonas Yersinia Agar are suitable for the isolation of Yersinia enterocolitica from human stool specimens or other materials. In addition, BD Aeromonas Yersinia Agar is used for the isolation of Aeromonas. Although Yersinia can be recovered by direct plating, specimens with low viable counts may require cold enrichment (4°C) in phosphate-buffered saline. However, cold enrichment may not be practical because of the long incubation time and because it selects for nonpathogenic strains of Y. enterocolitica and other Yersinia species. Enrichment of Aeromonas in alcaline peptone water may be helpful to isolate the organism form populations that shed low numbers of organisms, e.g., from carriers or convalescent-phase patients.

On both media described here, also other Yersinia species, such as Y. pseudotuberculosis, Y. frederiksenii, and Y. intermedia will grow. The pathogenic potential of the latter two species is controversial, but at this time, isolates should not be disregarded.

The formulation of BD Aeromonas Yersinia Agar but not BD Yersinia Selective Agar is recommended for the selective isolation of Yersinia pestis. Enterobacteriaceae other than Yersinia may grow on these media, especially Citrobacter species. Serratia and Citrobacter cannot always be reliably differentiated from Yersinia by colony morphology alone. Therefore, biochemical and serological tests are necessary for confirmation and complete identification of the isolates.

Certain strains of Aeromonas may produce weak growth on BD Aeromonas Yersinia Agar. It is recommended to include additional isolation media for a recovery of the total population of these organisms.

REFERENCES


PACKAGING/AVAILABILITY

BD Yersinia Selective Agar (CIN)
Cat. No. 254056 Ready-to-use Plated Media, cpu 20
Cat. No. 254088 Ready-to-use Plated Media, cpu 120

BD Aeromonas Yersinia Agar
Cat. No. 254443 Ready-to-use Plated Media, cpu 20

FURTHER INFORMATION

For further information please contact your local BD representative.

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http://www.bd.com/europe/regulatory/

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