# YERSINIA CIN AGAR ACCORDING TO ISO 10273:2017

## INSTRUCTION FOR USE READY-TO-USE PLATED MEDIA

**For professional use**

**Intended use: Yersinia CIN Agar is used for the selective isolation of *Yersinia enterocolitica.***

| Ref.: | Type of medium: | Packaging: |
|---|---|---|
| **201090** | ready-to-use medium-plate | 1x20 pcs (90 mm) |

**1. Principle:** enzymatic digest of gelatin, enzymatic digest of casein, and enzymatic digest of animal tissue provide the nitrogen and amino acids. Yeast extract is the vitamin source in this formula. Selectivity of Yersinia CIN Agar is due to the presence of sodium deoxycholate, crystal violet, and irgasan that markedly inhibit growth of Gram-positive and many Gram-negative organisms. Supplementation with cefsulodin and novobiocin improves inhibition of normal enteric organisms. Differentiation is based on mannitol fermentation. Organisms capable of fermenting mannitol lower the pH around the colony, allowing absorption of neutral red and turning the colony a pink colour. Due to the localized pH decrease, a zone of precipitated bile may also be present. Organisms that do not metabolize mannitol to acid end products will form colourless, translucent colonies. Sodium chloride maintains the osmotic balance of the medium. Sodium pyruvate and magnesium sulfate stimulate organism growth. Agar is the solidifying agent.

**2. Formula/Liter:**

| | |
|---|---|
| Enzymatic digest of casein and animal tissue | 3.0 g |
| Enzymatic digest of gelatin | 17.0 g |
| Yeast extract | 2.0 g |
| Mannitol | 20.0 g |
| Sodium deoxycholate | 0.5 g |
| Sodium chloride | 1.0 g |
| Magnesium sulfate heptahydrate | 0.01 g |
| Sodium pyruvate | 2.0 g |
| Neutral red | 0.03 g |
| Agar | 12.0 g |
| Crystal violet | 0.001 g |

**Supplements/Liter:**

| | |
|---|---|
| Cefsulodin | 0.015 g |
| Irgasan | 0.004 g |
| Novobiocin | 0.0025 g |

**3. pH:** 7.4 ± 0.2 at 25°C.

**4. Appearance:**

**Prepared Appearance:** prepared medium is red-violet.

**5. Sample:** all samples in which *Yersinia enterocolitica* are expected.

**6. Test procedure:** if the agar plate has been refrigerated, allow to warm to room temperature before inoculation. After incubation in PSB Broth or ITC Broth take specified amount of sample and inoculate Yersinia CIN Agar (according to ISO 10273:2017) Streak the specimen for isolation onto the surface of the medium. Incubate plates aerobically at 30±1°C for 24 ± 2 hours. Plates incubate in an inverted position.

**7. Results:** after incubation observe growth of particular microorganism. Pathogenic *Y. enterocolitica* appears as small circular, smooth colonies with entire edge. The colonies have a small, deep red sharp bordered center. The surrounding rim is translucent and non-iridescent and finely granular. Identification of the microorganism should be confirmed by additional tests

**8. Quality control:** perform quality control testing for both negative and positive reaction by inoculating a representative sample of plates with pure cultures of stable control organisms that produce known, desired reactions according to ISO 11133:2014.

| Microorganism: | Method of control: | Criteria: | Appearance of colony: |
|---|---|---|---|
| *Yersinia enterococlictica* WDCM 00038 | productivity: qualitative | good growth (2) | colourless to pink colonies ± bile ppt |

| | | | |
|---|---|---|---|
| *Staphylococcus aureus* WDCM 00034 | selectivity: qualitative | growth or partial inhibition (0-1) | — |
| *Escherichia coli* WDCM 00013 | selectivity: qualitative | growth or partial inhibition (0-1) | — |

**9. Precautions:** due to nutritional variation, some strains may be encountered that grow poorly or fail to grow on this medium. Further tests are necessary for confirmation of *Yersinia* spp. Some strains of normal enteric organisms may be encountered that are not inhibited or only partially inhibited on complete medium, such as *Citrobacter freundii*, *Serratia liquefaciens*, and *Enterobacter agglomerans*. Growth of *Yersinia frederiksenii*, *Y. kristensenii*, *Y. pseudotuberculosis* and *Y. intermedia* is not inhibited on complete medium. Colonies of these organisms must be differentiated from *Y. enterocolitica* on the basis of additional characteristics.

**10. Disposal of waste:** after use, all plates and any other contaminated materials must be sterilized or disposed of in line with appropriate internal procedures and in accordance with local legislations. Plates can be destroyed by autoclaving at 121°C for at least 20 minutes.

**11. Storage:** on receipt, store plates at 2-12°C away from direct sun light in an inverted position. Do not overload a refrigerator with excessive amounts of plates to avoid water condensation on the lids during storage. Plates must not come into direct contact with the inner walls of refrigerator, as the media may freeze, invalidating the tests. Prepared plates, stored in their original sleeve wrapping at 2-12°C until just prior to use, may be inoculated up to the expiration date and incubated for recommended incubation times. Plates from opened stacks of 10 plates should be used for two weeks when stored in a clean area at 2 to 12° C. Do not use plates if they show evidence of microbial contamination, discoloration, drying, cracking or others signs of deterioration. Allow the medium to warm to the room temperature before inoculation.

All microbiological media containing dyes or light-sensitive components should be protected from light and stored in the dark.

Note that shelf life of the growth media changes after the addition of supplements. Complete media containing protein supplement tend to degrade faster than basal media alone.

**12. Shelf life:** 3 months.

**13. Required supplements not supplied together with medium base:** not applicable.

**14. References:** available on request.

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