**HARDYCHROM™ LISTERIA**

| Cat. no. G317 | HardyCHROM™ Listeria, 15x100mm Plate, 19ml | 10 plates/bag |

**INTENDED USE**

HardyCHROM™ Listeria is a chromogenic medium recommended for the selective isolation, differentiation, and enumeration of *Listeria monocytogenes* from food and environmental samples by colony color and appearance.

This product is not intended to be used for the diagnosis of human disease.

**SUMMARY**

*Listeria* are gram-positive, non-sporing bacilli. They are ubiquitous in nature and can be isolated from soil, vegetables, and natural waters as well as from healthy animals and humans. While many species are non-pathogenic, *Listeria monocytogenes* is a well-established food poisoning risk. It can be found in uncooked meats and vegetables, as well as unpasteurized dairy products. Its ability to cause disease is due, in part, to the bacterium's ability to survive and grow at refrigerated temperatures. Clinical symptoms can range from flu-like illness to more serious conditions including meningitis, pneumonitis, septicemia and endocarditis. *Listeria monocytogenes* infections mainly occur in neonates, pregnant women, the elderly and immunocompromised individuals. Infections in pregnant women are a documented cause of spontaneous abortions and still births.

HardyCHROM™ Listeria is a chromogenic medium that allows for the rapid and reliable detection of *Listeria monocytogenes*. Current isolation methods for *L. monocytogenes* require multiple media types and can require up to 10 days of incubation. With HardyCHROM™ Listeria, there is only a single broth enrichment step for a total incubation time of 48-72 hours.

HardyCHROM™ Listeria contains specific chromogenic substrates that result in all *Listeria* species producing turquoise colored colonies when the substrate is hydrolyzed by specific enzymes. Further, this medium is able to detect the phospholipase activity specific to the two pathogenic *Listeria* species: *L. monocytogenes* and *L. ivanovii*. These two species will produce turquoise colored colonies surrounded by an opaque white halo within 48 hours. While *L. ivanovii* is rare in clinical samples, further tests are needed to definitively differentiate between these two species. Organisms other than *Listeria* are inhibited or grow as colorless or turquoise colonies without halos.

**FORMULA**

Ingredients per liter of deionized water:*

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Amount</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peptones</td>
<td>24.0gm</td>
</tr>
<tr>
<td>Yeast Extract</td>
<td>10.0gm</td>
</tr>
<tr>
<td>Lithium Chloride</td>
<td>10.0gm</td>
</tr>
<tr>
<td>Sodium Chloride</td>
<td>5.0gm</td>
</tr>
</tbody>
</table>
**Disodium Phosphate** | 2.5gm
---|---
**Chromogens and Selective Agents** | 2.2gm
**Glucose** | 2.0gm
**Sodium Pyruvate** | 2.0gm
**Magnesium Glycerophosphate** | 1.0gm
**Magnesium Sulfate** | 0.5gm
**Agar** | 15.0gm

Final pH 7.2 +/- 0.2 at 25ºC.

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

**STORAGE AND SHELF LIFE**

Storage: Upon receipt, store away from direct light at 2-8ºC. away from direct light. Media should not be used if there are any signs of deterioration (shrinking, cracking, or discoloration), contamination, or if the expiration date has passed. Product is light and temperature sensitive; protect from light, excessive heat, moisture, and freezing.

The expiration dating on the product label applies to the product in its intact packaging when stored as directed. The product may be used and tested up to the expiration date on the product label and incubated for the recommended quality control incubation times.

Refer to the document "Storage" for more information.

**PRECAUTIONS**

This product may contain components of animal origin. Certified knowledge of the origin and/or sanitary state of the animals does not guarantee the absence of transmissible pathogenic agents. Therefore, it is recommended that these products be treated as potentially infectious, and handle observing the usual universal blood precautions. Do not ingest, inhale, or allow to come into contact with skin.

This product is for laboratory use only. It is to be used only by adequately trained and qualified laboratory personnel. Observe approved biohazard precautions and aseptic techniques. All laboratory specimens should be considered infectious and handled according to "standard precautions." The "Guidelines for Isolation Precautions" is available from the Centers for Disease Control and Prevention at [www.cdc.gov/ncidod/dhqp/gl_isolation.html](http://www.cdc.gov/ncidod/dhqp/gl_isolation.html).

For additional information regarding specific precautions for the prevention of the transmission of all infectious agents from laboratory instruments and materials, and for recommendations for the management of exposure to infectious disease, refer to CLSI document M-29: *Protection of Laboratory Workers from Occupationally Acquired Infections: Approved Guideline*.

Sterilize all biohazard waste before disposal.

Refer to the document "Precautions When Using Media" for more information.

Refer to the document SDS Search instructions on the Hardy Diagnostics' website for more information.

**PROCEDURE**

**Sample Collection:** Sample collection and preparation should be performed following appropriate standards and guidelines.
Method of Use: Allow medium to warm to room temperature prior to inoculation.

1. Enrich sample according to US Food and Drug Administration (FDA), US Department of Agriculture (USDA), or ISO Standards appropriate for the sample type.
2. Inoculate HardyCHROM™ Listeria plate with 0.1 ml of the enrichment broth and streak for isolation.
   Incubate aerobically at 35-37°C for 24 hours. Do not incubate in CO₂.
3. Observe plates for characteristic colonial morphology and color at 24 hours. If negative for *Listeria monocytogenes* or *L. ivanovii*, re-incubate for an additional 12-24 hours (for a total of 48 hours) and examine again.

**INTERPRETATION OF RESULTS**

The presence of smooth, round, turquoise colonies 1-1.5 mm in diameter surrounded by an opaque white halo is a presumptive positive test for the presence of *L. monocytogenes* / *L. ivanovii*. Further testing should be done to differentiate *L. monocytogenes* from *L. ivanovii* such as hemolysis, CAMP, rhamnose, xylose or other AOAC-RI approved methods such as Microgen™ Listeria ID (Cat. no. MID67). Colonies which appear colorless or turquoise without halos should be interpreted as negative for *L. monocytogenes* / *L. ivanovii*.

<table>
<thead>
<tr>
<th>Organism</th>
<th>Description</th>
<th>Photo</th>
<th>Color</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>L. monocytogenes</em> and <em>L. ivanovii</em></td>
<td>turquoise colonies surrounded by white halo</td>
<td><img src="image" alt="Photo" /></td>
<td><img src="image" alt="Color" /></td>
</tr>
</tbody>
</table>

**LIMITATIONS**

It is recommended that biochemical, immunological, molecular, or mass spectrometry testing be performed on colonies from pure culture for complete identification.

*Listeria* species should produce turquoise colonies within 24 hours; however, halo development may take as long as 48 hours. *L. ivanovii* is also capable of producing a halo and therefore has a similar colony appearance as *L. monocytogenes*, but *L. ivanovii* is rarely found in food. Any possible *L. monocytogenes* colonies should be differentiated from *L. ivanovii* using appropriate testing.

Rarely, some strains of *B. cereus* give turquoise colonies with a halo, but these are easily differentiated. The colonies have an irregular colony shape and a large halo that is not consistent with the appearance of *L. monocytogenes* colonies.

Rare coryneform or diphtheroid-like bacteria may appear green-turquoise in color.

Refer to the document "Limitations of Procedures and Warranty" for more information.

**MATERIALS REQUIRED BUT NOT PROVIDED**

Standard microbiological supplies and equipment such as loops, swabs, applicator sticks, other culture media, incinerators, incubators, etc., as well as serological and biochemical reagents, are not provided.

**QUALITY CONTROL**

Hardy Diagnostics tests each lot of commercially manufactured media using appropriate quality control microorganisms and quality specifications as outlined on the Certificates of Analysis (CoFA). The following organisms are routinely used for testing at Hardy Diagnostics:

<table>
<thead>
<tr>
<th>Incubation</th>
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<tbody>
<tr>
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<td></td>
<td></td>
</tr>
<tr>
<td>Test Organisms</td>
<td>Inoculation Method*</td>
<td>Time</td>
<td>Temperature</td>
</tr>
<tr>
<td>-----------------------------</td>
<td>---------------------</td>
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<td>-------------</td>
</tr>
<tr>
<td><strong>Listeria monocytogenes</strong></td>
<td>A</td>
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<td>35°C</td>
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<tr>
<td>ATCC® 15313</td>
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<td></td>
<td></td>
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<tr>
<td><strong>Listeria innocua</strong></td>
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<td>24hr</td>
<td>35°C</td>
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<tr>
<td>ATCC® 33090</td>
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<td></td>
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<tr>
<td><strong>Escherichia coli</strong></td>
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<td>24hr</td>
<td>35°C</td>
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<tr>
<td>ATCC® 25922</td>
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<td></td>
<td></td>
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<tr>
<td><strong>Staphylococcus aureus</strong></td>
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<td>24hr</td>
<td>35°C</td>
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<tr>
<td>ATCC® 25923</td>
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<tr>
<td><strong>Candida albicans</strong></td>
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<td>35°C</td>
</tr>
<tr>
<td>ATCC® 10231</td>
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</tbody>
</table>

* Refer to the document "Inoculation Procedures for Media QC" for more information.

**USER QUALITY CONTROL**

End users of commercially prepared culture media should perform QC testing in accordance with applicable government regulatory agencies, and in compliance with accreditation requirements. Hardy Diagnostics recommends end users check for signs of contamination and deterioration and, if dictated by laboratory quality control procedures or regulation, perform quality control testing to demonstrate growth or a positive reaction and to demonstrate inhibition or a negative reaction, if applicable. Hardy Diagnostics quality control testing is documented on the certificates of analysis (CoA) available from Hardy Diagnostics Certificates of Analysis website. In addition, refer to the following document "Finished Product Quality Control Procedures" for more information on QC or see reference(s) for more specific information.

**PHYSICAL APPEARANCE**

HardyCHROM™ Listeria should appear slightly opaque, and straw in color.

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REFERENCES


   http://www.fda.gov/Food/FoodScienceResearch/LaboratoryMethods/ucm2006949.htm

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The Hardy Diagnostics manufacturing facility and quality management system is certified to ISO 13485.

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