



Product Sheet

Spiroplasma leptinotarsae (ATCC® 43213™)

Please read this **FIRST**



Intended Use

This product is intended for research use only. It is not intended for any animal or human therapeutic or diagnostic use.

Citation of Strain

If use of this culture results in a scientific publication, it should be cited in that manuscript in the following manner: *Spiroplasma leptinotarsae* (ATCC® 43213™)

Description

Designation: LD-1

Deposited Name: *Spiroplasma leptinotarsae* Hackett et al.

Propagation

Medium

ATCC® Medium 1188: *Spiroplasma* medium MID (ATCC medium 798) with fresh yeast extract replaced by water

Growth Conditions

Temperature: 30.0°C

Atmosphere: Anaerobic

Propagation Procedure

1. Follow instructions as suggested for the culturing of *Mollicutes*:

PROCEDURES FOR PROPAGATING *MOLLICUTES*:

- a) Open the vial according to the enclosed instructions.
 - b) Using a Pasteur or 1.0 ml pipette, withdraw approximately 0.5 to 1.0 ml from a tube containing 5.0 ml. Rehydrate the pellet.
 - c) Aseptically transfer this aliquot back into the tube. Mix well.
 - d) Make serial dilutions by transferring 0.5 ml from the original tube to a tube containing 4.5 ml. Repeat process by transferring 0.5 ml from the second to a third tube, etc. Dilutions are important, not only for titration purposes, but also to keep culture in varying stages of growth. Many strains will die out rapidly once acid or alkaline conditions are reached. It is recommended to prepare several dilutions from the initial tube as the cryoprotectant used in the freeze-drying process often inhibits growth.
 - e) Use an uninoculated tube of broth to serve as a control.
 - f) Plates may be inoculated to check colonial morphology. You can also spot each dilution on the surface of plate (4 or more/plate) to determine the number of colony-forming units. However, not all strains do well on solid medium.
 - g) Incubate all tubes and plates under the recommended conditions and appropriate temperature. The time necessary for growth will vary from strain to strain. Growth on plates generally requires additional incubation.
 - h) Depending on the medium used, growth will be indicated by increased turbidity, a color change, or both.
2. Tubes are incubated anaerobically at 30°C.
 3. This strain will start to show a color change from orange-red to yellow in 5 to 7 days on primary culturing. No turbidity will be observed.
 4. For long term storage of *Spiroplasma leptinotarsae*, freeze-drying or freezing is recommended. Liquid nitrogen storage is the best method. Optimally grown cells are centrifuged at 9000 rpm for 30 minutes, the supernatant poured off, and the packed cells resuspended in a smaller amount of #1188 broth. To this, add an equal amount of sterile 20% glycerol as a cryoprotectant. This suspension is aliquoted into small plastic vials and stored at -70°C or below.

References

References and other information relating to this product are available online at www.atcc.org.

Biosafety Level: 1

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the current publication of the *Biosafety in Microbiological and Biomedical Laboratories* from the U.S. Department of Health and Human Services Centers for Disease Control and Prevention and National Institutes for Health.

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Disclaimers

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Please see the enclosed Material Transfer Agreement (MTA) for further details regarding the use of this product. The MTA is also available on our Web site at www.atcc.org

Additional information on this culture is available on the ATCC web site at www.atcc.org.
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