Orenia salinaria Moune et al.

700911™

Description

Strain designation: SG3902
Deposited As: *Orenia salinaria* Moune et al.
Type strain: Yes

Storage Conditions

Product format: Freeze-dried

Intended Use

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

BSL 1

ATCC determines the biosafety level of a material based on our risk assessment as guided by the current edition of *Biosafety in Microbiological and Biomedical Laboratories (BMBL)*, U.S. Department of Health and Human Services. It is your responsibility to understand the hazards associated with the material per your organization's policies and procedures as well as any other applicable regulations as enforced by your local or national agencies.

ATCC highly recommends that appropriate personal protective equipment is always

used when handling vials. For cultures that require storage in liquid nitrogen, it is important to note that some vials may leak when submersed in liquid nitrogen and will slowly fill with liquid nitrogen. Upon thawing, the conversion of the liquid nitrogen back to its gas phase may result in the vial exploding or blowing off its cap with dangerous force creating flying debris. Unless necessary, ATCC recommends that these cultures be stored in the vapor phase of liquid nitrogen rather than submersed in liquid nitrogen.

Certificate of Analysis

For batch-specific test results, refer to the applicable certificate of analysis that can be found at www.atcc.org.

Growth Conditions

Medium:
ATCC Medium 2180: Orenia salinaria medium
Temperature: 40°C
Atmosphere: Anaerobic

Handling Procedures

1. Sterilize the top of the Balch tube by spraying it with 70% ethanol and then flaming the top.

2. If needed exchange the gas in the Balch tube for 100% N_2.

3. If the medium is pink (see discussion about resazurin) add 2.0 ml of reducing agent (3% cysteine, stock solution) per 100 ml of medium. Let the medium sit at room temperature for 10 to 20 minutes, until the resazurin becomes colorless, before

inoculating.

4. When the Balch tube is ready to inoculate, open the vial according to enclosed instructions. Take an anaerobic

1.0 ml syringe (*see discussion below*) tipped with a 22 gauge needle and withdraw 0.5 ml of 2180 medium from the Balch tube and rehydrate the freeze dried pellet. Immediately place the rehydrated vial under a stream of sterile gas to maintain anaerobic conditions.

5. Using the same syringe transfer the rehydrated cell suspension back into a tube of #2180 broth. Plate 0.1 ml of the inoculated culture onto a non-selective agar medium and incubate aerobically at 37ºC. Inoculate a nonselective anaerobic and aerobic broth. Transfer 0.5 ml of the rehydrated culture to a second tube of 2180 medium. Incubate the inoculated tubes at 37 to 45ºC.

6. Growth should be detected in the #2180 broth within 24 to 48 hours. No growth should be detected on the aerobic plate, or in the nonselective aerobic or anaerobic broth.

ANAEROBIC CONDITIONS:

a. Balch tubes (available from Bellco Glass, Vineland ,NJ) are specially designed for anaerobic work and use an aluminum crimp cap to hold a rubber stopper in place. Needles can easily be inserted through the stopper, and the tubes can be pressurized to 2 atm. /alternatively, serum vials may be used, or screw cap tubes with butyl rubber stoppers. In the latter case the stopper may be removed and the tube placed under a cannula system that dispenses sterile, oxygen-free gas for the addition of reducing agents or inoculation.

b. Resazurin is a commonly used redox indicator that is pink when the redox potential is above 50 mv., and colorless when the redox potential is below 110 mv. i.e. highly reducing. Most strict anaerobes require this low redox potential for optimum growth.

c. To obtain a fully reduced medium, it is necessary that the medium be anoxic and that a reducing agent be added. Common reducing agents are sodium sulfide, cysteine, dithiothreitol, and titanium citrate.

d. Syringes can be made anaerobic by one of two methods. 1. Displace the

dead space in the syringe with a sterile

Notes

Cells appear as long rods in chains that are weakly motile. As the culture ages the cells will form spheroplasts and long irregular cells. When exposed to oxygen the cells quickly lose their integrity.

In broth the cells form stringy clumps that will settle to the bottom. These clumps are easily re-suspended when the tube is shaken.

Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: *Orenia salinaria* Moune et al. (ATCC 700911)

References

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

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