Pavlova lutheri (Droop) Green

50092™

Description

Strain designation: NEPCC 242 [ATCC 50094]
Deposited As: *Pavlova lutheri* (Droop) Green
Type strain: No

Storage Conditions

Product format: Test tube
Storage conditions: See handling procedure

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 1405: HESNW medium
Instructions for complete medium: Grown with mixed bacteria
Temperature: 20-25°C
Atmosphere: Aerobic

Handling Procedures

Handling of Live Culture

This strain is routinely shipped as a growing culture in a glass 16 x 125 mm screw-capped test tube. The volume of the cell suspension is approximately 5-6 mL. When the culture arrives remove it promptly from the shipping container. **Do not store the**

culture at refrigeration temperatures before handling. To assure viability, immediately loosen the test tube cap and incubate under light (~50 mEinsteins/m^2/s irradiance) at a 15° horizontal slant at 20-25°C for at least one hour before observing the culture. There should be numerous active trophozoites in suspension. If the numbers are low the culture may have been exposed to temperature extremes in transit. Regardless of the state of the culture, seal and agitate the culture, then aseptically transfer a 0.5 mL aliquot to a 16 x 125 mm screw-capped test tube containing 5 mL of sterile ATCC medium 1405. Incubate the parent and daughter cultures at a 15° horizontal slant at 20-25°C with the caps on loosely for air exchange. Maintain under a 14 hour light (~50 mEinsteins/m^2/s irradiance)/10 hour dark cycle.

Culture maintenance:

Subculture every two weeks to a fresh tube of medium in the following manner:

1. Seal and agitate the parent culture to suspend cells, then aseptically transfer 0.5 mL to a 16 x 125 mm screw-capped test tube containing 5 mL of sterile ATCC medium 1405.
2. Incubate the parent and daughter cultures at a 15° horizontal slant at 20-25°C with the caps on loosely for air exchange. Maintain under a 14 hour light (~50 mEinsteins/m^2/s irradiance)/10 hour dark cycle.

Note: Addition of penicillin G at 50-75 U/mL and streptomycin at 50-75 µg/mL to the culture may be necessary if the bacterial density in the culture becomes high enough to affect growth.

Reagents for cryopreservation: Cryoprotective Solution
Glycerol 2.4 mL
Fresh growth medium w/o bacteria 7.6 mL
Cryopreservation:

1. Mix the components of the cryoprotective solution in the order listed.
2. Harvest cells from a culture that is at or near peak density by centrifugation at 800-1000 x g for 5 min.
3. Adjust the concentration of cells to at least 2 x 10^7/mL in fresh medium.
4. Mix the cell preparation and the cryoprotective solution in equal portions.
5. Dispense in 0.5 mL aliquots into 1.0 - 2.0 mL sterile plastic screw-capped cryules (special plastic vials for cryopreservation).
6. Place vials in a controlled rate freezing unit. From room temperature cool at -

1°C/min to -40°C. If freezing unit can compensate for the heat of fusion, maintain rate at -1 C/min through heat of fusion. At -40°C plunge ampules into liquid nitrogen. Alternatively, place the vials in a Nalgene 1°C freezing apparatus. Place the apparatus at -80°C for 1.5 to 2 hours and then plunge ampules into liquid nitrogen. (The cooling rate in this apparatus is approximately -1°C/min.)

7. Ampules are stored in either the vapor or liquid phase of a nitrogen refrigerator.
8. To establish a culture from the frozen state place the vial in a 35°C water bath. Immerse the vial to a level just above the surface of the frozen material. Do not agitate the vial. Immediately after thawing, do not leave in water bath, aseptically remove the contents of the ampule and inoculate into a 16 x 125 mm screw-capped test tube containing 5 mL of sterile ATCC medium 1405. Immediately seal and agitate the culture to evenly suspend cells, then aseptically transfer a 0.5 mL aliquot to a second, identical tube of medium.
9. Incubate the parent and daughter cultures at a 15° horizontal slant at 20-25°C with the caps on loosely for air exchange. Maintain under a 14 hour light (~50 µEinsteins/m^2/s irradiance)/10 hour dark cycle.
10. Follow the protocol for maintenance of culture.

Notes

This xenic culture contains the original bacterial flora present when the alga was first isolated.

Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: *Pavlova lutheri* (Droop) Green (ATCC 50092)

References

References and other information relating to this material are available at

www.atcc.org.

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