ATCC

Product Sheet

Sappinia pedata (ATCC® PRA-237™)

Please read this FIRST

Storage Temp.
Frozen Cultures:
-70°C for 1 week; liquid N_2 vapor for long term storage

Freeze-dried Cultures:
2-8°C

Live Cultures:
See Protocols section for handling information

Biosafety Level
1

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: *Sappinia pedata* (ATCC® PRA-237™)

American Type Culture Collection
PO Box 1549
Manassas, VA 20108 USA
www.atcc.org

800.638.6597 or 703.365.2700
Fax: 703.365.2750
Email: Tech@atcc.org

Or contact your local distributor

Description

Strain Designation: AUK06-2-1
Depositor: FW Spiegel
Isolation: Isolated by MW Brown, Ground Litter Plant material, Auckland Island, New Zealand, 2006.

Propagation

Growth Conditions
Temperature: 15°C to 20°C

Medium
ATCC® Medium 2432: wMY (weak Malt Yeast Extract)

Instructions for Complete Medium
Media: ATCC Medium 2432 wMY inoculated with *Escherichia coli* (ATCC® 23437™).

Protocols

Storage and Culture Initiation
Frozen ampules packed in dry ice should either be thawed immediately or stored in liquid nitrogen. If liquid nitrogen storage facilities are not available, frozen ampules may be stored at or below -70°C for approximately one week. **Do not under any circumstance store frozen ampules at refrigerator freezer temperatures (generally -20°C).** Storage of frozen material at this temperature will result in the death of the culture.

1. To thaw a frozen ampule, place it in a 35°C water bath such that the lip of the ampule remains above the water line. Thawing time is approximately 2 to 3 minutes. Do not agitate the ampule. Do not leave ampule in water bath after it is thawed.
2. Immediately after thawing, aseptically transfer contents to a plate of ATCC medium 2432. Distribute the material evenly over the plate using a spread bar.
3. Wrap the entire edge of the plate with parafilm and incubate upright at 15-20°C. Amoeboid trophozoites and/or stalked fruiting bodies should be seen within 2-5 d.

Culture Maintenance

1. Prepare an ATCC medium 2432 plate with a lawn of *Escherichia coli* (ATCC® 23437™) and incubate at 37°C overnight.
2. Remove an agar block (~5 mm^2) with trophozoites from the edge of an agar plate culture and invert the block at the edge of the freshly prepared plate.
3. Wrap the entire edge of the plate with parafilm and incubate upright at 15-20°C.
4. Repeat steps 1-3 at 7-10 d intervals.

Cryopreservation

Reagents
Cryoprotective Solution
DMSO, 1.5 mL
Fresh growth medium w/o bacteria, 8.5 mL

Harvest and Preservation

1. Mix the components in the order listed. When the medium is added to the DMSO the solution will warm up due to chemical heat.
2. Harvest cells from a culture which is at or near peak density by adding 5 mL ATCC medium 5080 (Dryl's solution) and washing cells into suspension. Rub the surface of the plate with a spread bar to detach adhering trophozoites.
3. Adjust the concentration of cells to at least 2 x 10^4/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

Product Sheet

Sappinia pedata (ATCC® PRA-237™)

Please read this FIRST

Storage Temp.
Frozen Cultures:
-70°C for 1 week; liquid N_2 vapor for long term storage

Freeze-dried Cultures:
2-8°C

Live Cultures:
See Protocols section for handling information

Biosafety Level
1

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: *Sappinia pedata* (ATCC® PRA-237™)

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, aseptically transfer the contents of the ampule to the center of a fresh plate of ATCC medium 2432. Distribute the material evenly over the plate using a spread bar.
9. Wrap the entire edge of the plate with parafilm and incubate upright at 15-20°C.
10. Follow the protocol for maintenance of culture.

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.

ATCC Warranty

The viability of ATCC® products is warranted for 30 days from the date of shipment, and is valid only if the product is stored and cultured according to the information included on this product information sheet. ATCC lists the media formulation that has been found to be effective for this strain. While other, unspecified media may also produce satisfactory results, a change in media or the absence of an additive from the ATCC recommended media may affect recovery, growth and/or function of this strain. If an alternative medium formulation is used, the ATCC warranty for viability is no longer valid.

Disclaimers

This product is intended for laboratory research purposes only. It is not intended for use in humans.

While ATCC uses reasonable efforts to include accurate and up-to-date information on this product sheet, ATCC makes no warranties or representations as to its accuracy. Citations from scientific literature and patents are provided for informational purposes only. ATCC does not warrant that such information has been confirmed to be accurate.

This product is sent with the condition that you are responsible for its safe storage, handling, and use. ATCC is not liable for any damages or injuries arising from receipt and/or use of this product. While reasonable effort is made to insure authenticity and reliability of strains on deposit, ATCC is not liable for damages arising from the misidentification or misrepresentation of cultures.

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.

American Type Culture Collection
PO Box 1549
Manassas, VA 20108 USA
www.atcc.org

800.638.6597 or 703.365.2700
Fax: 703.365.2750
Email: Tech@atcc.org

Or contact your local distributor