



Spongomonas minima **Dangeard**

50404™

Description

Strain designation: AZ-3

Deposited As: *Spongomonas minima* Dangeard

Type strain: No

Storage Conditions

Product format: 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 802: Sonneborn's Paramecium medium

Instructions for complete medium: ATCC Medium 802 inoculated with *Enterobacter aerogenes* (ATCC 13048)

Temperature: 25°C

Handling Procedures

Culture maintenance:

1. Agitate a culture at or near peak density by inverting several times. Encysted cells must be detached from the flask surface with a sterile cell scraper or cotton swab.
2. Aseptically transfer a 0.3 ml aliquot to a T-25 tissue culture flask containing 10 ml

of fresh bacterized medium.

3. Screw cap on tightly and incubate at 25°C.

4. Subculture every 10-15d.

Cryopreservation:

1. Harvest cysts from several cultures in stationary phase of growth. Detach cysts using a sterile cell scraper or cotton swab.

2. Aseptically transfer the cyst suspension to 15 ml plastic centrifuge tubes.

3. Centrifuge at ~800 x g for 5 min.

4. While cysts are centrifuging, prepare a 20% solution of DMSO in bacterized ATCC Medium 802. Cool on ice.

5. Remove the supernatant and pool the cell pellets to one-half the final volume desired with fresh growth medium.

6. Combine the cell suspension with an equal volume of 20% DMSO cryoprotectant solution (prepared in step 4) to yield a final concentration of 10% DMSO.

7. Dispense in 0.5 ml aliquots to 1.0-2.0 ml Nunc vials (special plastic vials for cryopreservation).

8. Place the vials in a controlled rate freezing unit. From room temperature cool at -1°C/min to -40°C. 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.).

9. Store ampules in a liquid nitrogen refrigerator until needed.

10. To establish a culture from the frozen state, place a frozen ampule in a 35°C water bath just enough to cover the frozen material. Allow the ampule to thaw completely (2-3 min).

11. Immediately after thawing, aseptically remove the contents and transfer to a T-25 tissue culture flask containing 10 ml of fresh ATCC medium 802 inoculated with *Enterobacter aerogenes* (ATCC 13048).

12. Screw the cap on tightly and incubate at 25°C. Subculture every 10-15d.

Notes

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

While every 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.

ATCC recommends that individuals contemplating commercial use of any culture first contact the originating investigator to negotiate an agreement. Third party distribution of this culture is discouraged, since this practice has resulted in the unintentional spreading of contaminated cultures.

Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: *Spongomonas minima* Dangeard (ATCC 50404)

References

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

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