

SUPPORTING INFECTIOUS DISEASE RESEARCH

Product Information Sheet for NR-166

Coccidioides posadasii, Δcts2/Δard1/Δcts3

Catalog No. NR-166

For research use only. Not for human use.

Contributor and Manufacturer:

University of Toledo, Toledo, Ohio and California State University, Bakersfield Foundation, Bakersfield, California

Product Description:

Classification: Mitosporic Onygenales, Coccidioides

Species: Coccidioides posadasii Strain/Isolate: Δcts2/Δard1/Δcts3

<u>Original Source</u>: NR-166 was derived from *Coccidioides* posadasii (C. posadasii) strain C735 by deletion of two chitinase genes (cts2 and cts3) and a D-arabinitol 2-dehydrogenase gene (ard1).¹

Comment: This attenuated strain can transform into first-generation spherules, but it is incapable of endosporulation, and therefore, cannot reproduce in the parasitic form of the fungus, rendering it avirulent. Avirulence of the Δcts2/Δard1/Δcts3 strain was confirmed through a murine model of coccidioidomycosis. The whole genome shotgun sequence of *C. posadasii* C735 ΔSOWgp [a transgenic strain of *C. posadasii* C735 that has had the spherule outer wall glycoprotein (SOWgp) gene deleted] is available (GenBank: ΔCFW00000000).

C. posadasii is a pathogenic fungus that, along with C. immitis, can result in mild to fatal respiratory disease (coccidioidomycosis, San Joaquin Valley fever, desert rheumatism) due to inhalation of spores in both immunocompetent and immunocompromised individuals. C. posadasii resides in soil in parts of southwestern United States, northern Mexico, and other areas in South and Central America unlike C. immitis which is primarliy found in California.

Material Provided:

Each vial of NR-166 contains approximately 1 mL of fungal culture containing 10% DMSO.

Packaging/Storage:

NR-166 was packaged aseptically in cryovials and is provided frozen on dry ice. The product should be stored at -70°C or colder.

Growth Conditions:

Media:

GYE (1% glucose, 0.5% yeast extract) broth or agar. Note: The addition of 100 mM KCl may enhance growth as the culture is osmotically fragile. This strain is resistant to hygromycin. Addition of hygromycin is optional as the insertion of the gene into the genome is considered

a stable insertion. Hygromycin can be added to a final concentration of 75 ug/mL, if desired.

Incubation:

Temperature: 27°C to 35°C Atmosphere: Aerobic

Propagation:

- 1. Keep vial frozen until ready for use; thaw rapidly.
- 2. Transfer the entire thawed aliquot into GYE broth.
- Incubate with shaking at 27°C to 35°C for 2 days.
 Growth in the presence of hygromycin may take 4-7 days.

Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: Coccidioides posadasii, Δcts2/Δard1/Δcts3, NR-166."

Biosafety Level: 2

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the following publication: U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control and Prevention, and National Institutes of Health. <u>Biosafety in Microbiological and Biomedical Laboratories</u>. 5th ed. Washington, DC: U.S. Government Printing Office, 2009; see www.cdc.gov/biosafety/publications/bmbl5/index.htm.

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NR-166 is claimed in U.S. Patent Number 7,332,324 and the continuations, continuations-in-part, re-issues and foreign counterparts thereof.

References:

- Xue, J., et al. "A Genetically Engineered Live Attenuated Vaccine of Coccidioides posadasii Protects BALB/c Mice against Coccidioidomycosis." <u>Infect. Immun.</u> 77 (2009): 3196-3208. PubMed: 19487479.
- Hector, R. and G. W. Rutherford. "The Public Health Need and Present Status of a Vaccine for the Prevention of Coccidioidomycosis." <u>Ann. N. Y. Acad. Sci.</u> 1111 (2007): 259-268. PubMed: 17344529.
- Cole, G. T., et al. "A Vaccine against Coccidioidomycosis is Justified and Attainable." <u>Med. Mycol.</u> 42 (2004): 189-216. PubMed: 15283234.

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