

Actinobacteria: *Rhodococcus erythropolis***Source**

- ATCC
- Catalogue #: 15903

Growth Parameters

Note: The condition(s) tested below is not necessarily optimal for growth of this host. Rather, it represents a “basic” growth medium on which this host can grow.

- Growth media: PYCa media, supplemented with 4.5 mM CaCl₂ in top agar and enrichment culture
- Temperature tested: 30°C
- Streak Plate Time: 4 days
- Liquid Culture Time: 2 days
- Lawn Growth Time: 2 day
- Colony Color: creamy tan
- Colony Morphology: Round, convex, slightly raised, slightly rough, dry

Additional Notes:

- Start fresh cultures every 7 – 14 days.

Streak Plate Image

Background

Rhodococcus erythropolis is a non-motile Gram positive Actinobacterium in the family Nocardiaceae. Various strains have been [sequenced](#) and they are reported to have a length between 6.3 and 7.3 Mb with a GC% of 62%. It has been found in soil and water. Some strains are known to degrade phenol-derivatives, so it is a great bioremediation candidate. It has been reported to degrade N-acyl-homoserine lactones (AHL) and limit the symptoms caused by the plant pathogen *Pectobacterium atrosepticum* on potato tubers.

Host Information

- BSL1
- Found in soil and sludge

References

Genome Sequence of *Rhodococcus erythropolis* Strain CCM2595, a Phenol Derivative-Degrading Bacterium. Strnad H, et al. *Genome Announc* 2014 Mar 20

Genome Sequence of the Quorum-Quenching *Rhodococcus erythropolis* Strain R138. Kwasiborski A, et al. *Genome Announc* 2014 Mar 27

Cirou A, Raffoux A, Diallo S, Latour X, Dessaux Y, Faure D. 2011. Gamma-caprolactone stimulates growth of quorum-quenching *Rhodococcus* populations in a large-scale hydroponic system for culturing *Solanum tuberosum*. *Res. Microbiol.* 162:945–950.

SEA Faculty Pilot Data

The following are observations and tips from SEA Faculty that have worked with this host. These observations have yet to be validated by the SEA Team at HHMI and the University of Pittsburgh.

Host Growth

- Cells settle in flask but are easily resuspended.
- Cells begin clumping after ~ 2 weeks, upon which complete resuspension is difficult to achieve.
- Store working stocks, liquid or streak plate, up to 1 week at room temperature or up to 2 weeks at 4°C
- Note smell and color to detect contaminants.

Phage Isolation

- **Source of phage:**
Phage has successfully been isolated from soil of compost, horse stable and Boston [area with average annual temperature of ~ 15°C].
- **Plating Efficiency:**
Direct plating: No success reported
Enrichment: ~ 10 % success rate
- **Enrichment:**
Vary enrichment culture incubation, plating for plaques after 1 – 3 days.
- **Phage Lysate Titer and Stability:**
 - Titers vary from 10^5 – 10^{10}
 - If titers from individual plaques are too low to subsequently obtain web plates, considering suspending the agar directly underlying the plaque (plaque-plug) in buffer. This will allow for phage within the plaque, as well as within the underlying agar, to be collected.
 - Titers of turbid plaques may decrease with time and manipulation. In such circumstances, consider: resuspending phage in liquid media used for enrichment instead of phage buffer; removing bacterial cells and debris from the resuspension by centrifugation instead of filtration.

- Web plates of turbid plaques may appear as a bacterial lawn without plaques. Back-calculate from plates with individual plaques to help identify web plates.

Genome Extraction

- If excessive degradation of genomes occur, as determined by large smears on agarose gels, considering shortening the incubation period of phage and nuclease mix [e.g. 30 min on ice instead of overnight at 4°C]