

Sporulation Assay

Streak strains for colonies on rich media

Inoculate 4mL DSM liquid media with a single colony

Incubate for 24 hours

**Turn on the water bath about 3 hours before you need it, or add pre-heated water

Perform a serial dilution, $10^{-1} - 10^{-6}$, in DSM liquid media

Make a 1mL dilution series with 100 μ L culture in 900 μ L 1X T-Base.

Before heat shock, plate 100 μ L of the last three dilutions, 10^{-4} , 10^{-5} , 10^{-6} , on DSM plates

Heat shock all dilutions at 80C for 20min.

Plate 100 μ L of all heat shocked dilutions, $10^{-1} - 10^{-6}$, on DSM plates

Incubate O/N (~18 hours)

(# after heat kill) / (# before heat kill) = % spores in total population