

# *Burkholderia thailandensis* Isolated from Infected Wound, Arkansas, USA

## Technical Appendix

### Materials and Methods

DNA was extracted using the Maxwell RSC Cultured Cells DNA kit on the Promega Maxwell RSC Instrument per the manufacturer's instructions (Promega, Fitchburg, Wisconsin). The sample was eluted in PCR-grade water and RNase A, filtered through a 0.1 µm filter, and checked for sterility before submission for whole genome sequencing (1).

The sequence for BtAR2017 was determined from paired-end Illumina reads which were generated on an Illumina MiSeq (Illumina, Inc., San Diego, California). Genomic DNA was sheared to a mean size of 600 bp using a Covaris LE220 focused ultrasonicator (Covaris Inc., Woburn, MA). DNA fragments were Ampure (Beckman Coulter Inc., Indianapolis, IN) cleaned and used to prepare dual-indexed sequencing libraries using NEBNext Ultra library prep reagents (New England Biolabs Inc., Ipswich, MA) and barcoding indices synthesized in the CDC Biotechnology Core Facility. Libraries were analyzed for size and concentration, pooled and denatured for loading onto flowcell for cluster generation. Sequencing was performed on the Illumina MiSeq using 2 × 250 bp cycle paired-end sequencing kits. On completion, sequence reads were filtered for read quality, basecalled and demultiplexed using bcl2fastq (v2.19). Raw read data were archived at NCBI as SRA accession SRP135864.

The genome was submitted to the *Burkholderia pseudomallei* MLST Web site (<http://pubmlst.org/bpseudomallei>) to identify the sequence type (2,3). Core SNPs were analyzed using Parsnp in the Harvest 1.3 suite (4). The dendrogram was generated in MEGA7 (5). Average nucleotide Identity (ANI) was calculated at <http://enve-omics.ce.gatech.edu/ani/index> (6). The Active Melioidosis Detect kit (inBios, Seattle, WA) was used to test BtAR2017 for the presence of *B. pseudomallei* capsular polysaccharide as per manufacturer's instructions (7).

## References

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