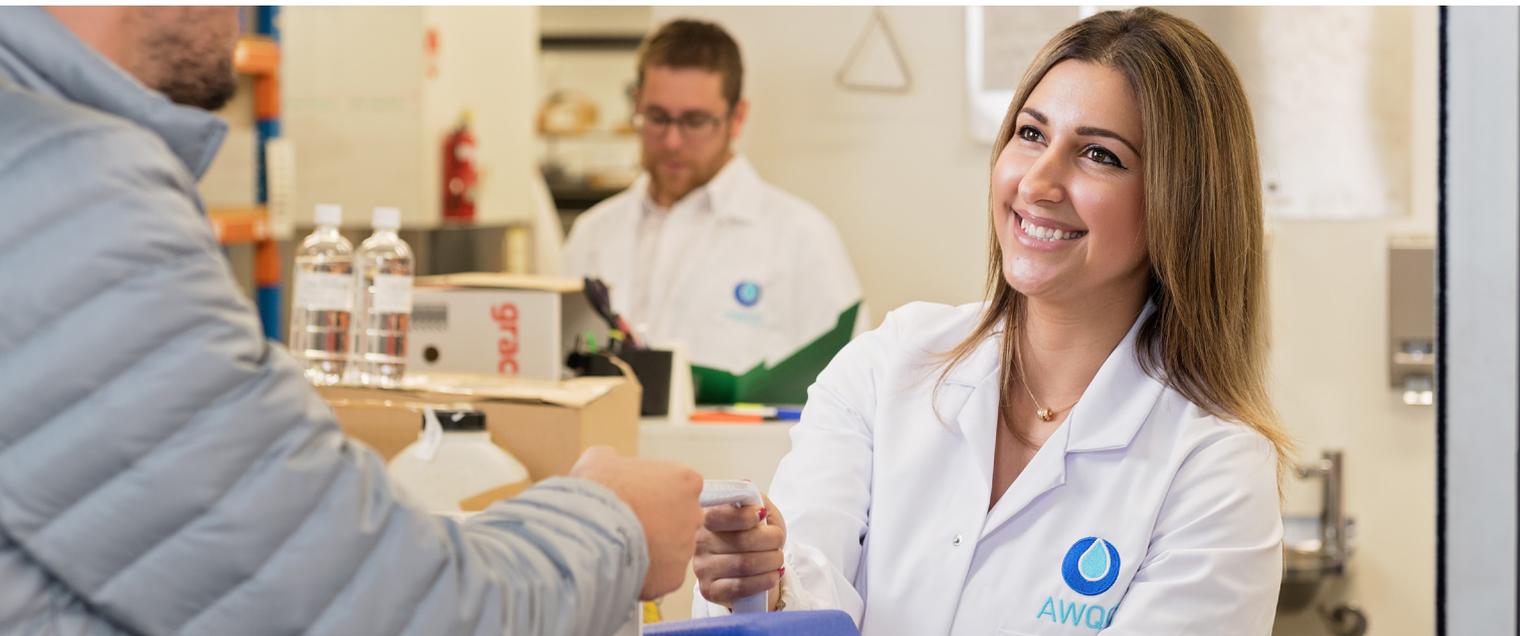


Burkholderia pseudomallei



The Australian Water Quality Centre (AWQC) is dedicated to ensuring and responding to the public health requirements relating to the provision of water and wastewater services for communities in Australia and across the world.

- Specialist water services
- Ensuring public health

Testing for *Burkholderia pseudomallei*

Overview

Burkholderia pseudomallei is a motile, gram-negative, aerobic bacterium of the Burkholderiaceae family. *B. pseudomallei* is commonly found in soil and muddy water in the tropical regions of Australia. It can infect humans and animals and cause the disease Melioidosis.

Melioidosis is a potentially fatal disease affecting both humans and animals. It is endemic in Northern Australia and Southeast Asia. Common symptoms of the disease are pneumonia, skin abscesses or ulcers, abscesses in internal organs and unusual neurological illnesses such as brainstem encephalitis and acute paraplegia (NHMRC 2011). The greatest risk of exposure to *B. pseudomallei* occurs when humans or animals come in contact with contaminated soil or muddy water, generally after heavy rainfall (NHMRC 2011). Transmission of the bacterium most commonly occurs via skin abrasions coming in contact with contaminated soil or water. Human to human transmission, inhalation of contaminated dust or water droplets, and contact with contaminated groundwater have also been reported but are less likely to occur (Foong et al. 2014).

Application and benefits

AWQC's molecular method allows direct detection of *B. pseudomallei* from water, sludge and soil samples with no culture required. The probe PCR-based technique provides a rapid, cost-effective, highly specific and sensitive tool for the qualitative environmental surveillance of *B. pseudomallei*. Both viable and non-viable organisms are detected.

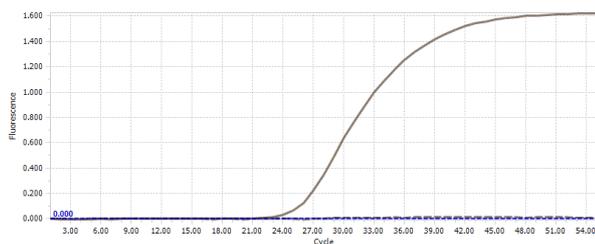
Management of microbial risks in water supplies traditionally focuses on harmful microorganisms that originate from sewage or animal waste, rather than environmental organisms which occur naturally in soil or water. AWQC's analysis allows for accurate and early detection of *B. pseudomallei* to facilitate timely disinfection and process control management, including risk mapping of melioidosis. AWQC's molecular method is more sensitive than traditional culture-based methods.

Method

The AWQC's culture-independent, DNA molecular method allows direct detection of *B. pseudomallei* from water, sludge and soil samples. The assay detects two specific, independent gene targets that are only present in *B. pseudomallei*. The two gene targets are a "type three secretion system 1 single-copy gene" and a hypothetical protein gene only present in *B. pseudomallei* (Knappik et al. 2015, Limmathurotsakul et al. 2012, Trung et al. 2011). The assay for these genes has been shown to be equivalent to direct culture of *B. pseudomallei*, including an internal DNA extraction control, probe polymerase chain reaction (PCR) efficiency and reaction controls (Knappik et al. 2015, Limmathurotsakul et al. 2012, Trung et al. 2011).

The AWQC analysis includes the broadest detection profile possible whilst remaining specific to the *B. pseudomallei* genome. The primers and probe sequences have 100% homology with a broad range of *B. pseudomallei* sequences based on a comprehensive bioinformatics analysis and on publication reviews.

Our PCR-based system provides a highly specific and sensitive tool for environmental surveillance of *B. pseudomallei*.



Detection of *Burkholderia pseudomallei* by probe PCR

	1	2	3	4	5	6	7	8	9	10	11	12	
A			Red			Red			Red				Positive
B													Negative
C			Red			Red			Red				Invalid
D													N/A
E			Red			Red			Red				Positive
F													Negative
G						Green			Red				Invalid
H			Green			Green							Positive

Please contact the AWQC to discuss your requirements and request a competitive quotation for your testing needs.

References

- Foong Y.C., Tan M., Bradbury R.S. (2014). Melioidosis: a review. *Rural and Remote Health* 14: 2763.
- Knappik M., Dance D.A.B., Rattanavong S., Pierret A., Ribolzi O., Davong V., Silisouk J., Vongsouvat M., Newton P.N., Ditttricha S. (2015). Evaluation of molecular methods to improve the detection of *Burkholderia pseudomallei* in soil and water samples from Laos. *Applied and Environmental Microbiology* 81(11):3722-3727.
- Limmathurotsakul D., Wuthiekanun V., Amornchai P., Wongsuwan G., Day N.P.J., Peacock S.J. (2012). Effectiveness of a simplified method for isolation of *Burkholderia pseudomallei* from soil. *Applied and Environmental Microbiology* 78(3): 876-877.
- National Health and Medical Research Council (2011). Australian Drinking Water Guidelines 6. Fact Sheets - Microorganisms (Bacteria): *Burkholderia pseudomallei*, pp. 277-278.
- Trung T.T., Hetzer A., Göhler A., Topfstedt E., Wuthiekanun V., Limmathurotsakul D., Peacock S.J., Steinmetz I. (2011). Highly sensitive direct detection and quantification of *Burkholderia pseudomallei* bacteria in environmental soil samples by using real-time PCR. *Applied and Environmental Microbiology* 77(18):6486-6494.

Component:

- Burkholderia pseudomallei*

Limit of reporting (LOR):

- Detected / Not Detected per litre or per gram.

Sampling Requirements:

- Sludge matrix: one 300mL sodium thiosulphate dosed sterile container, triple bagged (requires a minimum of 1g sludge).
- Water matrix: two 600mL sodium thiosulphate dosed sterile containers, triple-bagged.
- Air gap required for all containers.
- Transport on ice.
- Samples submitted for *B. pseudomallei* analysis samples should arrive adequately labelled by the customer, i.e. chain of custody documentation attached to the outside of eskies and eskies clearly marked as containing hazardous samples.

Holding Times:

- Sludge matrix: 24 hours.
- Water matrix: 24 hours.

Turnaround Time (TAT):

- Standard: 10 days; emergency: 3 days

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