Development of an antigen capture assay for melioidosis caused by *Burkholderia pseudomallei*: abridged secondary publication

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KEY MESSAGES

1. Melioidosis is a potentially fatal disease caused by *Burkholderia pseudomallei*. There is an urgent need for a diagnostic method that can rapidly detect *B. pseudomallei* infection.
2. We generated and purified multiple monoclonal antibodies with high reactivity to *B. pseudomallei*.
3. We developed a simple and reliable rapid antigen capture assay with high sensitivity (85%) and specificity (100%) for the detection of *B. pseudomallei*. The assay is user-friendly and rapid, allowing inexpensive laboratory diagnosis of melioidosis. It has potential for development into lateral flow immunoassays that allow rapid on-site diagnosis of melioidosis in humans and animals.

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Introduction

Melioidosis, a potentially fatal disease caused by *Burkholderia pseudomallei*, is an emerging health concern in various regions, particularly Southern Asia and northern Australia. *B. pseudomallei* also causes melioidosis in various animals within endemic areas.[1] In Hong Kong, melioidosis is endemic in humans, captive marine mammals (eg bottlenose dolphins, California sea lions, and pilot whales), and captive birds such as zebra doves.[2] Accurate and timely diagnosis of melioidosis is essential for effective treatment; however, conventional culture-based identification is time-consuming and often yields false-negative results, whereas molecular techniques such as polymerase chain reaction require specialised equipment and expertise. Thus, serological tests for detecting melioidosis are needed.

Methods and results

We generated and evaluated three monoclonal antibodies (mAbs) specifically targeting the lipopolysaccharide of *B. pseudomallei*. The mAb with the highest reactivity, designated B5 (subclass IgG2b with λ light chains), was used to develop the antigen capture assay. This sandwich enzyme-linked immunosorbent assay–based method used B5 as the capture antibody and an in-house polyclonal anti–*B. pseudomallei* antibody as the detection antibody. Evaluation of the antigen capture assay showed promising results. The diagnostic sensitivity and specificity of the developed antigen capture assay was assessed using 20 melioidosis-positive and 25 melioidosis-negative urine samples. When tested with clinical samples, the assay exhibited high sensitivity (17/20, 85%) and specificity (25/25, 100%) for the detection of melioidosis.

Discussion

Our assay offers several advantages over existing diagnostic methods. It is simple and reliable and can be used in a standard diagnostic laboratory without requiring expensive equipment or reagents. The rapid turnaround time can facilitate timely diagnosis. Furthermore, the antigen capture assay has the potential for adaptation into specific lateral flow immunoassays such as point-of-care test strips and cassettes, thereby enabling rapid on-site diagnosis of melioidosis in both humans and animals. Although the mAb B5 exhibited some cross-reactivity with bacterial lysates of closely related *Burkholderia* species (*B. mallei* and *B. thailandensis*), these species have limited clinical significance in human infections. *B. thailandensis* is considered rare and non-pathogenic in humans, whereas *B. mallei* primarily infects equids (eg horses) and has only caused sporadic cases in humans. Considering its high sensitivity and specificity for *B. pseudomallei*, our assay is a valuable tool for the diagnosis of melioidosis. Further research is needed to validate and optimise the assay for use in various settings.

Conclusion

Our newly developed antigen capture assay offers simple, reliable, and rapid detection of melioidosis. Considering its high sensitivity and specificity, this assay has the potential to facilitate timely diagnosis and effective management of this potentially fatal disease.

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