



Antimicrobial susceptibility of *Burkholderia pseudomallei* isolates in Northern Vietnam

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ARTICLE INFO

Article history:

Received 12 July 2018

Received in revised form 6 January 2019

Accepted 16 January 2019

Available online 24 January 2019

Keywords:

Burkholderia pseudomallei

Antimicrobial susceptibility

Melioidosis

Vietnam

ABSTRACT

Objectives: The antimicrobial susceptibility pattern of clinical *Burkholderia pseudomallei* (*B. pseudomallei*) in Vietnam has not been reported since the first publication in 2008. The present study aimed to determine the antimicrobial susceptibility pattern of *B. pseudomallei* isolated in a tertiary referral centre in Hanoi from January 2012 to December 2017.

Methods: A total of 312 *B. pseudomallei* isolates obtained from melioidosis patients admitted to a 2000-bed general hospital were analysed by the Etest method. Interpretation of the susceptibility testing results were reported using Clinical and Laboratory Standards Institute guidelines.

Results: All isolates were susceptible to ceftazidime, imipenem and amoxicillin-clavulanate (100%) with MIC_{90s} relatively low (2 µg/mL). Two isolates had intermediate resistance to doxycycline (0.6%) and 34 isolates were resistant to trimethoprim/sulfamethoxazole (10.9%).

Conclusion: The results of this study suggest that currently recommended antibiotics for melioidosis treatment can be empirically used, but continuously monitoring antimicrobial susceptibility should be a concern.

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1. Introduction

Southeast Asia and northern Australia are regarded as endemic areas of melioidosis. In northeast Thailand, melioidosis is the third most common infectious cause of death after HIV/AIDS and tuberculosis; the annual incidence in 2006 was 21.3/100 000 [1]. In Australia, the latest publication for the disease revealed that melioidosis is the most common cause of fatal community-acquired septicaemia pneumonia and yearly incidence rates range between 5.4 and 50.2/100 000 [2]. Melioidosis is a potentially fatal infection cause by *Burkholderia pseudomallei* (*B. pseudomallei*), a saprophytic soil and Gram-negative bacillus [3]. Disease manifestations include subclinical infections, localised abscesses, severe pneumonia, and fulminant sepsis. The mortality of melioidosis ranges from 14 to 40% and may be as high as 80% if effective antibiotics are not given [4]. The appropriate therapy is pivotal to patient outcome from severe melioidosis and relapse of infection. Many reports on antimicrobial susceptibility patterns of

B. pseudomallei from hotspots for melioidosis such as Thailand, Australia, and Malaysia [5–7].

Vietnam is known as an endemic area of melioidosis, but since the first study in northern Vietnam 10 years ago, which was performed on a limited number of *B. pseudomallei* (25 isolates) [8], studies on antimicrobial susceptibility of *B. pseudomallei* to date have been surprisingly negligible. Currently, there are no official or national guidelines on diagnosis and treatment of melioidosis in Vietnam. In most clinical laboratories, the simple algorithm for presumptive identification of *kB. pseudomallei* is used – including oxidase-positive, Gram-negative bacilli, resistant to gentamicin and colistin but sensitive to amoxicillin/clavulanic acid – [9] and the international consensus guidelines for treatment are applied. The treatment guidelines includes two steps: parenteral drugs in the acute phase to prevent death from severe sepsis, followed by oral drugs for the eradication phase to prevent relapse. Ceftazidime is the first choice of acute phase treatment and carbapenem is an alternative choice in cases of severe melioidosis for a minimum 2 weeks, followed by a minimum 3-month eradication with oral trimethoprim/sulfamethoxazole [10,11].

Therefore, the present study aimed to determine the antimicrobial susceptibility profile of *B. pseudomallei* isolated in Bach Mai Hospital, which is a tertiary referral centre in Hanoi and one of the largest hospitals in Vietnam that receives severely ill patients from

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provincial hospitals in northern Vietnam. It also provides a database for national guidelines for the treatment of melioidosis.

2. Materials and methods

A total of 312 *B. pseudomallei* isolates (232 isolated from blood, 30 from sterile body fluids, 26 from respiratory specimens, 22 from pus, one from urine, and one from cerebrospinal fluid) of melioidosis patients diagnosed and treated at Bach Mai Hospital over a 6-year period (January 2012 to December 2017) were identified based on characteristics of colony morphology, Gram stain, oxidase, resistance to colistin and gentamicin, susceptibility to amoxicillin-clavulanate, and API 20NE. Among the 312 isolates, two isolates came from two patients with relapse of infection. Antimicrobial susceptibility testing was performed by Etest (Biomérieux) as per the manufacturer's instruction. Interpretation of the susceptibility testing results were reported using Clinical and Laboratory Standards Institute guidelines [12].

3. Results

A total of 312 *B. pseudomallei* were susceptible to ceftazidime, imipenem and amoxicillin-clavulanate (100%), and MIC_{90s} were relatively low (2 µg/mL). Two isolates had intermediate resistance to doxycycline (0.6%) and 34 isolates were resistant to trimethoprim/sulfamethoxazole (10.9%). However, almost all isolates were susceptible to all five tested antibiotics at low concentrations. Both of the two isolates from patients with infection relapse had the same antimicrobial resistance patterns as the original isolates. One patient had infection again after 3 years and the other after 3 months. The patient who had the 3-month relapse of infection was poorly adherent to eradication treatment.

4. Discussion

Burkholderia pseudomallei is considered to be a stubborn pathogen because of difficulty eradicating it from the host. Additionally, it is not susceptible to some antimicrobial agents that are often empirically used to treat infectious diseases. Since the first study on antimicrobial resistance patterns in northern Vietnam 10 years ago, which was performed on a limited number of *B. pseudomallei* (25 isolates) [8], there have been no data available on *B. pseudomallei* susceptibility characteristics.

The current study tested a relatively high number of isolates. All 312 isolates showed 100% susceptibility to both ceftazidime and imipenem. *Burkholderia pseudomallei* resistance to ceftazidime and carbapenem, to date, is rare but reported in some studies from Thailand [5], Malaysia [7] and Singapore [13]. With high sensitivity, it is believed that these antimicrobial agents are the empirical choice for acute phase treatment of melioidosis in Vietnam while pending culture results. For the eradication phase, trimethoprim/sulfamethoxazole with or without doxycycline is preferred to prevent infection relapse. Amoxicillin/clavulanic acid is in reserve for those who either do not tolerate trimethoprim/sulfamethoxazole, or are children or pregnant women. No strain was resistant to amoxicillin/clavulanic acid in present study, so it was an effective choice for treatment. The simple algorithm for presumptive identification of *B. pseudomallei* may still work well in limited-resource laboratories in Vietnam. By using this simple algorithm from June to December 2015, four provincial general hospitals in north-central Vietnam successfully detected 94 suspected *B. pseudomallei* isolates. Three of four of those hospitals have never previously isolated *B. pseudomallei* [14]. In Thailand [5] and Malaysia [7], a small proportion of *B. pseudomallei* isolates were found to be resistant to amoxicillin/clavulanic acid (2% and 18%, respectively). To prevent missing melioidosis cases, care

should be taken to use a simple and cheap algorithm for identifying *B. pseudomallei* in those countries.

The rate of primary resistance to trimethoprim-sulfamethoxazole was observed in this study. Thirty four of 312 isolates (10.9%) were resistant to trimethoprim-sulfamethoxazole. That is in line with the rate of resistance to trimethoprim-sulfamethoxazole in clinical isolates from Thailand (10–13%) [15] and higher than that in Australia (0–3%) [16]. More than half of trimethoprim-sulfamethoxazole MICs were in the range of 1/19 to 2/38 µg/mL, which was close to the susceptible breakpoint. In a study published 10 years ago from the same hospital, Phuong et al. did not find any isolate resistant to trimethoprim-sulfamethoxazole [8]. Resistance to doxycycline has occurred (0.6%), but it is relatively rare. This antibiotic is an alternative choice for eradication treatment. It is worthy of note that an MIC-based method is required to test susceptibility for *B. pseudomallei* and there are currently no disc diffusion susceptibility criteria for this organism. In studies published in Malaysia, the reported antimicrobial resistance might be inaccurate because of the rate of using the disc diffusion method [7,13]. The right testing method and accuracy of results should be considered to provide clinicians with maximum assistance for decision-making.

In summary, based on current results, it is proposed that empirical ceftazidime and carbapenem for acute phase treatment, and trimethoprim-sulfamethoxazole and doxycycline for eradication treatment of melioidosis in Vietnam are acceptable. However, whenever possible, culture results should be referred and emerging resistance to trimethoprim-sulfamethoxazole impacts in melioidosis therapy need to be monitored regularly.

Funding

This study was supported by funds from Vietnamese Ministry of Science and Technology and German Federal Ministry of Education and Research (RENOMAB project; 01DP13007).

Competing interests

None declared.

Ethical approval

This study was approved by the scientific and ethics committee of Bach Mai hospital (Hanoi, Vietnam) [1004/QD-BM 2014].

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