**编者按**:你可能从来没有听过这种传染病,即使它在全球范围内每年可使约9万人殒命,与麻疹相当;你可能经常去海南旅游,却不知道海南正是它的重灾区;你可能熟悉结核,殊不知香港学者推测大约1/3的当地结核病实则为这种传染病。这种危害严重但不为人熟悉的传染病就是类鼻疽。类鼻疽是由类鼻疽伯克霍尔德菌所致的热带传染病,流行于东南亚和澳大利亚北部等热带地区。我国海南、广东、香港、台湾等地是主要疫区。美国已将其列为Ⅰ类病原体,并且在泰国建立了热带病研究中心。这些提示我们加强类鼻疽的研究有着重要的军事意义和社会价值。鉴于此,本刊特邀请新加坡国立大学Yunn-Hwen Gan副教授就类鼻疽的免疫逃逸、细菌分泌系统等最新研究进展进行评述,并对其诊断防治进行了展望,以期能引起微生物学研究者、临床医师以及管理部门的广泛关注和高度重视。

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# 类鼻疽研究进展

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[**摘要**] 类鼻疽是由类鼻疽伯克霍尔德菌引起的一种新兴传染病,流行于东南亚和澳大利亚北部,目前尚无有效疫苗。类鼻疽菌作为一种兼性胞内致病菌具有庞大基因组,编码多种毒力因子,通过控制宿主细胞生物学途径并且抑制宿主细胞免疫应答过程,促进其在细胞内的增殖。本文对目前类鼻疽菌毒力相关的Ⅲ型和Ⅵ型分泌系统在类鼻疽病发病机制中的作用进行了概述,并讨论了类鼻疽菌新发现的毒力因子在细菌致病过程以及逃逸或破坏宿主免疫应答中发挥的作用。随着基因组学和蛋白组学的进展,对类鼻疽菌与宿主相互作用的分子机制有所加深,但许多问题仍有待探索解决,如疫苗、诊断、有效的治疗药物等。

[**关键词**] 类鼻疽;免疫逃逸;细菌分泌系统

[**中图法分类号**] R378; R515.9 [**文献标志码**] A

# Current state of melioidosis research

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*Burkholderia pseudomallei* is the causative agent of melioidosis, an infectious disease endemic in Southeast Asia, Northern Australia and other tropical regions[1]. Recently, the first evidence-based predictive map of the global incidence and burden of melioidosis estimated 165 000 (95% *CI*: 68 000 ~ 412 000) human melioidosis cases per year worldwide, from which 89 000 (36 000 ~ 227 000) people die[2]. These estimates indicate that melioidosis is severely under-reported in the 45 countries in which it is known to be endemic and it is probably endemic in additional 34 countries that have never reported the disease. Low awareness of the disease is mainly due to 2 reasons. The first is the difficulty of

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diagnosis as the disease has a wide spectrum of clinical presentations, ranging from asymptomatic infection to disseminated septicaemic infection[3]. The second reason is the inadequacy of conventional bacterial identification methods. The only assays sensitive and specific enough for routine clinical use are indirect hemagglutination, latex agglutination and immunofluorescence[4]. To compound the problem, the bacterium is inherently drug-resistant. Current standard treatment typically requires intravenous ceftazidime, the carbapenem antibiotics (imipenem and meropenem), or to a lesser degree amoxicillin-clavulanate for a minimum of 10 to 14 d. This is followed by the eradication phase therapy consisting of antibiotic trimethoprim-sulfamethoxazole, with or without doxycycline, and chloramphenicol for 3 to 6 months[5-7]. However, adherence to the long eradication phase among patients is typically poor and a new treatment guideline developed in Australia proposed a longer intensive phase beyond 2 weeks with a reduction or removal of the eradication phase that could reduce relapse[8]. The bacteria appear to be able to stay dormant in the host for long periods and relapses are not uncommon[3]. There are also no vaccines available. It does not help that many endemic regions are in the developing world and lack the resources to handle the disease. Furthermore, type 2 diabetes mellitus is the strongest risk factor associated with melioidosis, with about 40% ~ 60% of melioidosis patients being diabetic[3,9]. With the rising rate of type 2 diabetes mellitus around the world, including the developing nations where melioidosis is endemic, more cases of melioidosis can be expected. Besides rising numbers, the mortality rate is also unacceptably high. The case fatality rate is 40% in Thailand and 20% in Singapore and Australia[2]. Reliable statistics are not available for other regions.

Therefore, there is an urgent need to raise disease awareness among populations in the endemic as well as the probably endemic regions, and clinicians in these regions have to be trained to be able to diagnose the disease accurately, and to treat it appropriately. In Singapore, melioidosis has been a statutorily notifiable disease since 1989. Thailand and Australia also track their disease burden. It is thus important for endemic regions to gather statistics on their disease burden in order to understand how to implement better disease control and management. The disease is usually transmitted *via* breaks through skin with contaminated soil and water. It can also be inhaled through fomites or droplets, as well as ingested[3]. For example, in Thailand, a public education program among farmers has shown some success to prevent disease[10]. This includes boiling drinking water and putting on protective clothing to prevent soil and water contact with skin. Besides education and prevention, there is also a need to find better treatment regimens to shorten the time of treatment in order to circumvent non-compliance. The development of a vaccine would be helpful in endemic regions with poor clinical treatment facilities[11].

Besides causing an important disease, the bacterium is also a fascinating organism to study because of its versatility in metabolic requirements and the possession of many virulence factors[12]. The gram-negative saprophyte thrives in diverse ecological niches. Besides the soil and muddy water, it seems equally adept at surviving in plants[13], soil organisms such as amoebae[14], nematodes[15] and many mammalian species including marine animals[16]. This versatility is perhaps best reflected by its large genome size of 7.2 Mb[17] encoding a huge repertoire of virulence factors, including a complex array of protein secretion systems. *B. pseudomallei* has 3 Type Ⅲ Secretion Systems (T3SSs)[18-19] and 6 Type Ⅵ Secretion Systems (T6SSs), of which T3SS3 (also known as the bsa cluster)[20] and T6SS1 (also known as T6SS5)[21-22] have been shown to be critical for bacterial pathogenesis in mammalian hosts[23-25]. The bacterium enters the cells through unknown mechanisms and escapes from the phagosomal compartments into the host cytosol, where it multiplies and spreads through inducing host cell fusion. Some diagrams showing the intracellular life cycle could be found here[12,26-27]. So far, the only unambiguous role that has been attributed to T6SS1 *in vitro* is the induction of multi-nucleated giant cell formation and slower intracellular growth. Although actin based motility appears not to be important for infection *in vivo*[28], the *in vivo* significance of cell fusion or MNGC formation to

pathogenesis is less clear. It is critical to determine whether the requirement of T6SS1 in animal infection is due to the requirement of cell fusion for successful spread and establishment of disease. It is unclear what the secreted effectors of T6SS1 are, except for the T6SS1 needle component Hcp1 and the terminal VgrG trimer[29]. Given the absolute requirement for T6SS1 in bacterial pathogenesis in mammalian hosts, future research could target the cluster for the discovery of novel therapeutics or strategies to thwart bacterial virulence.

The T3SS3 in *B. pseudomallei* is also critical for its virulence. It is necessary for the early escape of the bacterium from vesicular and phagosomal compartment to the cytosol in host cells, and therefore affects its ability to multiply successfully in the cytosol[20]. However, it is not clear which exact component of T3SS3 is responsible for this early escape. It is also puzzling that so far, the only identified effectors of T3SS3, are namely BopA, BopC and BopE[30-32]. These are considered few compared to many other gram-negative pathogens with T3SS. Generally they also exhibit only partial and weak phenotypes in mammalian systems. One possibility is that perhaps their functions are more evident in a natural host of the bacterium, such as the amoeba, which it would encounter in the soil. No one is certain what the functions of T3SS1 and T3SS2 are, although there is some evidence that they may be important for plant infection[13].

*B. pseudomallei* has several ways to thwart the host immune response, where it could suppress and evade host immune responses[33-36]. We have also shown previously that the glutathione deficiency in Type 2 diabetic patients have impaired IL-12 and IFN response to *B. pseudomallei* infection, resulting in poorer intracellular bacterial killing[9]. Others have shown neutrophil defects in diabetic patients which could contribute to poorer disease outcomes[37]. Therefore, this is an area that deserves more study to determine the various reasons diabetic patients have such a strong disposition for disease, and how the immune system interacts with the bacteria.

We have come a long way in our understanding of the pathogenesis of *B. pseudomallei*. We know the bacterial capsule is important[12,38], and so are T3SS3 and T6SS1 in causing disease in animals and likely humans. The role of these three virulence factors in human disease can be surmised in the example of a patient in Darwin, Australia, who had chronic asymptomatic carriage of *B. pseudomallei*[39]. A 12-year analysis of the bacteria in the patient revealed several adaptations, including deleterious mutations in the capsule, T3SS3 and T6SS1 loci which likely allowed the bacteria to persist in the patient within the respiratory cavity without spread or disease. However, there are still many unanswered questions in melioidosis research, such as where do the bacteria reside during long periods of latency in the host? What are the exact triggers to enable bacteria to resurface to cause disease? What kind of metabolic adaptations the bacteria undergo in different environments? Therefore, I believe not only is there much to be done to improve disease prevention, diagnosis and treatment, but also understanding fundamental biology of the pathogen and how it interacts with its host will continue to yield many surprising discoveries for us in the future.

**Acknowledgment**: I apologize to colleagues whose work I may not have cited due to the constraints of this short perspective on melioidosis. I refer interested readers to many excellent recent reviews published on melioidosis as well as the original publications detailing many of the findings mentioned in this piece.

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