Melioidosis in India

Amy Tishler
Lake Forest College
Lake Forest, Illinois 60045

Melioidosis, also known as Whitmore's disease, needs more funding for scientific research and healthcare. Melioidosis infects approximately 165,000 people each year and kills 90,000 (Chen, 2016; Dunachie et al., 2017; Morici et al., 2019). Death rates can go up to 70%, and the caseload creates a healthcare burden equivalent to that of rabies (Chen, 2016). The number of deaths is roughly the same as that of measles, and several times that of dengue (Chen, 2016; Mukhopadhyay et al., 2018). In addition, recurrence of the disease occurs in 9% of patients (Stone et al., 2014). The disease is caused by the gram-negative and rod-shaped bacterium, *Burkholderia pseudomallei* which is found in contaminated soil and water. It is predominantly a disease of tropical climates, existing twenty degrees north and south of the equator (CDC, 2021; Pandey et al., 2010). It is widespread and can be found spanning Australia, Asia, Africa, and Latin America (CDC, 2021; Morici et al., 2019). It is present in as many as eighty-two countries (Morici et al., 2019). It is capable of infecting both animals and humans (CDC, 2021). The disease was initially recognized by British pathologist, Alfred Whitmore, and his research assistant, Krishnaswami while working in Myanmar in 1911 (Mukhopadhyay et al., 2018; Pandey et al., 2010).

Melioidosis is transmitted to humans and animals through inhalation, ingestion, and inoculation through direct contact with skin abrasions. There is also the possibility that it can be transmitted sexually, but there is no definitive proof. The disease can be passed perinatally from mother to infant (CDC, 2021; Cheng and Currie, 2005). It is extremely rare for the disease to be transferred from person to person or from animal to human (Lipsitz et al., 2010).

Burkholderia pseudomallei has several virulent factors. It produces a biofilm of glycocalyx polysaccharide allowing for the formation of microcolonies in a protective environment where the bacterium is phenotypically altered for antibiotic resistance. Pili are used to adhere to the eukaryotic cells (Cheng and Currie, 2005). Adhesins mediate attachment to non-phagocytic cells and cell contact triggers T3SS-3. Once inside the host cell phagocyte, with the help of T3SS-3, the bacterium escapes the phagosome into the cytoplasm where it can replicate. It then polymerizes host cell actin with BimA, facilitating actin-based mobility and actin tail formation which spreads from cell to cell. VirAG is activated due to the limited iron environment, and senses a signal that activates T6SS-1 gene expression which is critical for the formation of multinucleated giant cells created by induced host fusion. TssM interferes with host cell signaling and activates pattern recognition receptors which help the bacterium escape host cell autophagy (Stone et al., 2014). Alkyl hydroperoxide reductase interferes with reactive nitrogen intermediate (RNS) by helping the bacterium evade being killed by RNS. Other mechanisms that contribute to virulence include transposon mutations and the loss of the ability to assimilate arabinose (Chen and Currie, 2005).

The virulence of *Burkholderia pseudomallei* would make it a good bioweapon, particularly if spread by aerosol since the lethal dosage is lower with that route of transmission (Chen, 2016). The higher the dosage one is infected with, the shorter time until symptoms appear (Lipsitz et al., 2010). In addition, the bacterium is extremely hardy, to the point that Thai scientists mixed it with distilled water and tested the water and soil for the presence of the bacterium every year since the experiment began in 1993. The bacterium was able to survive for sixteen years without anything to sustain it (Chen, 2016). Melioidosis is resistant to most antibiotics, and currently there are no approved vaccines for human usage (Lipsitz et al., 2010; Morici et al., 2019; Stone et al, 2014). A close relative to the bacterium is B. mallei, known for causing glanders. Glanders was used as a bioweapon during the World Wars to injure and kill horses and mules. Inadvertently, human handlers also fell ill (Chen, 2016). Both *B. mallei* and *B. pseudomallei* are listed by the CDC as having Class B Bioterrorism potential (Menon et al., 2021).

Incubation is generally one to twenty-one days, but the disease can also remain latent for months and years before symptoms emerge (CDC, 2021). The longest incubation period has been 62 years for a World War II veteran (Goodyear et al., 2013; Lipsitz et al., 2010). The disease can either be acute or chronic. There are several types of melioidosis, each with its own set of symptoms. Localized infections involve local pain or swelling, fever, ulcerations, and abscesses. Pulmonary infections include a cough, chest pain, high fever, headache, and lack of appetite. Bloodstream infections typically consist of fever, headache, respiratory distress, abdominal discomfort, joint pain, and disorientation. Disseminated infections exhibit fever, weight loss, stomach or chest pain, muscle or joint pain, headache, central nervous system/brain infection, and seizures (CDC, 2021). Melioidosis is an opportunistic infection that is unlikely to kill a healthy person so long as they are diagnosed early and have the resources to receive proper care and antibiotics (Currie et al., 2010

Melioidosis is an emerging endemic disease in India (Chen, 2016; Halim et al., 2021). Most cases come from southwestern coastal Karnataka and northeastern Tamil Nadu. This most likely reflects the presence of research centers capable of diagnosing and treating melioidosis. Currently, India has no surveillance data for melioidosis, so there are no exact numbers of cases. It is known that the first indigenous case from India was in Mumbai in 1991, and a serosurvey confirmed that there was an epidemic in the mid-nineties (Mukhopadhyay et al., 2018; Pandey et al., 2010). The Indian isolates have novel allelic profiles compared to the Australian and Southeast Asian isolates (Mukhopadhyay et al., 2018). The genomic global analysis discovered that Australia was the early reservoir for the disease before entering southeast Asia. It disseminated to the south and east. Isolates from Africa appeared in Central and South America during the period of the slave trade (Morici et al., 2019).

Melioidosis is difficult to diagnose. The predominant clinical presentations of melioidosis differ based on their location. Pneumonia and bacteremia are the most common clinical presentations that occur. However, other presentations include ulcers, skin lesions, gastrointestinal ulceration, sepsis, and infections and abscesses involving internal organs (Lipsitz et al., 2010; Morici et al., 2019). In Thailand, spleen and liver abscesses are the most common, but in Australia prostate abscesses are more common (Currie et al., 2010). Melioidosis is known as "the great mimicker" since the symptoms are so non-specific. Frequently, the disease is mistaken for tuberculosis or common forms of pneumonia (Currie et al., 2010). Misdiagnosis is a common problem in areas of the world where the disease isn't well known, such as in emerging disease areas like India, Brazil, and Indonesia (Inglis et al., 2006). Melioidosis can be diagnosed by using the patient's bodily fluids to grow cultures (Chen, 2016). Unfortunately, this process is time-consuming, and it may take up to seven days to confirm a diagnosis (Lowe et al., 2016). It is possible that patients will die before the disease is confirmed to be melioidosis, due to likely ineffective antibiotics. The disease can kill within 48 hours (Chen, 2016).

Recently, researchers have been able to design multiplexing four single plex polymerase chain reaction assays that can identify various strains that fall under the *Burkholderia pseudomallei* complex. This method is more reliable than growing cultures. It is widely accepted that the *B. mallei* evolved as a pathogen from the *B. pseudomallei* complex (Lowe et al., 2016). The pathogenic *B. mallei* and *B. cenocepacie* are closely related to B. psuedomallei, as well as the non-pathogenic *B. thailandensis* (Inglis et al., 2006). In many cases, people live in rural areas with little access to healthcare and are therefore unable to be diagnosed (Vandana et al., 2015).

The type of infection and type of treatment influence the long-term outcomes. Melioidosis can be treated effectively if it is diagnosed early on. Treatment starts with at least two weeks of intravenous antimicrobial therapy consisting of either ceftazidime or meropenem. This is then followed up by three to six months of oral antimicrobial therapy consisting of either trimethoprim-sulfamethoxazole or amoxicillin/clavulanic acid (CDC, 2021). With the proper antibiotics and medical care, fatalities drop to one out of ten. There is a 5.9% resistance to trimethoprim/sulfamethoxazole and a 2.6% resistance to doxycycline. In some cases, surgeries are necessary, such as for abscesses (Currie et al., 2010; George et al., 2020). The type of infection can also influence the long-term outcome. For example, fatality rates are higher in patients with acute melioidosis. The death rate of sepsis-associated melioidosis is 50-90%. A proportion of acute infections are fatal, and those at the highest risk have meningoencephalitis (Inglis et al., 2006). In addition, a quarter of septicemic patients relapse despite taking antibiotics, which led to the concept of having a second round of eradication treatment (Inglis et al., 2006). The recurrence of melioidosis is mostly attributed to poor adherence to therapy (Currie et al., 2010).

There are general health risk factors that increase morbidity. Risk factors for contracting melioidosis include pre-existing conditions such as diabetes mellitus, alcohol usage, chronic lung disease, and chronic renal disease (Currie et al., 2010). Surprisingly, HIV is not included as a risk factor (Morici et al., 2019). India has the second highest number of cases of diabetes mellitus in the world after China. It used to be considered a disease

of affluence, but now it is also impacting those of lower socioeconomic status. 77 million people in India have diabetes (Martin, 2021). The most common comorbidity of melioidosis is type two diabetes mellitus (Menon et al., 2021). People with melioidosis face more risk factors based on some clinical presentations. Patients with acute melioidosis frequently have respiratory issues and bacteremia or septic arthritis (Koshy et al., 2019). There are some patients with chronic melioidosis where the condition worsens due to bacteremia and the inflammatory immune response. This usually occurs in elderly patients and those with diabetes (Koshy et al., 2019).

Climate change and weather play roles in morbidity. Outbreaks are most common after heavy rainfall, typhoons, monsoons, and flooding in general. The association of acute melioidosis with the wet season in certain regions concurs with other studies that there is a connection between illness and rainfall intensity (Koshy et al., 2019; Vidyalakshmi et al., 2012). Extreme weather events can cause the bacteria in the soil to become aerosolized (Lipsitz et al., 2010). Climate change means that natural disasters will be more common, leading to more opportunities for bacterial infection.

Melioidosis appears to affect more men than women based on clinical studies. Also, the highest prevalence is recorded as amongst the middle-aged male working population (Vandana et al., 2015). This points towards possible occupational exposure. Based on the research, the people most likely to encounter the bacterium are military personnel, construction workers, farmers, fisherman, forestry rangers, adventure travelers, and ecotourists (Currie et al., 2010). With increased agricultural work, there is more soil erosion and water turbidity. Soil erosion and higher turbidity levels are associated with a greater prevalence of the bacterium. Higher abundances of the bacterium are found in the groundwater and at depths greater than 30 cm (Ribolzi et al., 2016; Manivanh et al., 2017). The presence of *Burkholderia pseudomallei* is found in acrisol and luvisol soil types. Acrisol is a major soil group that has poor drainage but is often used for irrigating rice. However, this means water flow and gas renewal in the soil are drastically reduced. Luvisols are another major soil group that causes the mobilization of iron. Ferralsols are associated with the absence of *B. pseudomallei* (Ribolzi et al., 2016).

Some risk factors have impacts on mortality. Most of India's population in rural settings have limited access to hospitals and clinics where they could be diagnosed (Vandana et al., 2015). The disease has failed to gain the attention of the Ministries of Health and the office of WHO (Mukhopadhyay et al., 2018). People who can't afford intensive treatment are discharged early against medical advice (Mukhopadhyay et al., 2018). The likelihood of mortality can be estimated based on the amount of time it took for the cell culture to look positive (Lipsitz et al., 2010). The mortality rates of people diagnosed early on range from 10-25%. An early diagnosis lowers the chances of dying (George et al., 2020). The fatality rate is higher in patients with acute melioidosis (Koshy et al., 2019). Some other predictors of mortality are tachypnea, tachycardia, hypotension, organ failure assessment, bacteremia, respiratory involvement, hypoalbuminemia (Koshy et al., 2019), intensive care unit admission, and mechanical ventilation. Mortality rates are 40% in northern Thailand, 39% in Singapore, and 19% in Australia, while the overall prevalence in India is unknown (Lowe et al., 2016).

Seropositivity rates may represent exposure to the less pathogenic and generally avirulent *B. thailandensis*. *B. pseudomallei* differs from *B. thailandensis* by the ability to assimilate arabinose, despite having a similar genotype (Cheng and Currie, 2005). In parts of northeast Thailand, up to half the population has been infected by *B. thailandensis* at some point and carries antibodies in their blood. This suggests that most infections are asymptomatic. There is limited knowledge of melioidosis in India because people from poor, rural regions are underdiagnosed. In Udupi, which is on the southwestern coast of India, the seroprevalence was 29%. Females were twice as likely to be seropositive compared to males, not reflecting the numbers diagnosed in clinics. It is possible that women contract melioidosis while washing clothes in the river and gardening and were unable to get diagnosed at a clinic. There were no factors that indicated a relationship with seropositive status, including farmers and non-farmers. Also, seroprevalence didn't vary significantly between different age groups, though children were not included in this study (Vandana et al., 2015). In east India, over 75% of Bihar's population is employed in the agricultural sector, and they are among the largest producers of rice in the country. There is speculation that India will lead the global melioidosis map by 2030 with the highest predicted cases due to the number of diabetics in the country (Halim et al., 2021).

Melioidosis has the potential to spread to areas where it isn't typically found. Travelers who visit regions with the bacterium can contract the disease and become hospitalized back in their home country. Each year, there are about a dozen cases in the United States among travelers and immigrants. In 2021, there were four linked cases of melioidosis in the states due to imported aromatherapy (CDC, 2021; Sreenivas, 2022).

The establishment of preventative measures can decrease the incidences of melioidosis. Preventative measures against the bacterium are to wear waterproof gear and boots when performing agricultural work. Meat cutters should regularly disinfect their knives and gloves. Those who have a higher risk and people with skin abrasions should avoid contact with soil and standing water (CDC, 2021).

Currently, there are researchers working on a vaccine, but they have yet to gain approval to try it on humans. Five protein antigens are immunodominant in survivors: BopE, AhpC, PilO, BPSL2520, and BPSS1385. Survival was associated with enhanced T-cell responses to the antigens. In diabetics it was found that T-cell immunity was specifically impaired (Dunachie et al., 2017). Diabetic individuals have a twelve-fold increased risk. More than half of all melioidosis patients have diabetes (Morici et al., 2019). GroEL proteins are explored as vaccine candidates, but there is still a lack of consensus about the qualities needed for vaccine candidates (Dunachie et al., 2017). Lipopolysaccharides of *B. pseudomallei* are an important virulence factor that allows the bacterium to evade human immune responses during the early stages of infection. Lipopolysaccharides could be a possible vaccine candidate since it reduces the severity of disease in animals (Shaw et al., 2019). A vaccine would be of great value if used in high-risk populations, even if it did not seem to be worth the cost at first. There is limited evidence that CD8+T cells play a role in the protection. A multi-component vaccine is essential due to the virulence mechanisms and strain heterogeneity. There have been consistent findings with vaccine candidates tested on mice. Researchers can induce sterilizing immunity at low-challenge doses and extend the time of death at higher-challenge doses (Morici et al., 2019). Other animal models have been used as well. They range from mice, rats, and hamsters to invertebrate models and non-human primates. Overall, most models are rodents that who are experimented on having various routes of infection and different challenge doses (Morici et al., 2019).

In conclusion, melioidosis should receive funding for scientific research and healthcare applications. Funding would be used to set up a surveillance program for melioidosis in India as well as to start a grassroots education campaign. The funding would also be used to create a vaccine for melioidosis and to monitor antibiotic resistance of the bacterium *Burkholderia pseudomallei.*

References

CDC (2021). Melioidosis. Centers for Disease Control and Prevention.

Chen, Angus (2016). This Germ Can Live Decades in Distilled Water, Kill Humans in 48 Hours. *National Public Radio.*

Cheng and Currie (2005). Melioidosis: Epidemiology, Pathophysiology, and Management. *Clinical Microbiology Reviews* p. 383-416.

Currie et al., 2010. The epidemiology and clinical spectrum of melioidosis: 540 cases from the 20 year Darwin prospective study. 4(11), e900-.

Dunachie et al. (2017). Infection with Burkholderia pseudomallei-immune correlates of survival in acute melioidosis. *Scientific Reports* 7:12143.

George, A.J. et al. (2020). Surgical management of Melioidosis: 10-year retrospective data from a tertiary hospital. In India. *Bulgarian Association of Young Surgeons.*

Goodyear, A. et al. (2013). An Improved Selective Culture Medium Enhances the Isolation of Burkholderia pseudomallei from Contaminated Specimens. *American Journal of Tropical Medicine and Hygiene* 89(5):973-982.

Halim et al. (2021). Melioidosis in Bihar, India: unearthing the first of many? *Access Microbiology*.

Inglis et al. (2006). Melioidosis in the Americas. *American Society of Tropical Medicine and Hygiene*, 75(5):947-954.

Koshy et al. (2019). Clinical Manifestations, Antimicrobial Drug Susceptibility Patterns, and Outcomes in Melioidosis Cases, India. *Emerging Infectious Diseases* 25(2).

Lipsitz et al. (2012). Workshop on Treatment of and Postexposure Prohphylaxis for Burkholderia pseudomallei and *B. mallei* infection, 2010. *Emerging Infectious Disease* 18(12).

Lowe et al. (2016). A Quadruplex Real-Time PCR Assay for the Rapid Detection and Differentiation for the Most Relevant Members of the B. pseudomallei Complex: *B. mallei*, *B. pseudomallei*, and *B. thailandensis*. *PLoS ONE* 11(10).

Manivanh, L. et al. (2017). Burkholderia pseudomallei in a lowland rice paddy: seasonal changes and influence of soil depth and physico-chemical properties. *Scientific Reports* 7:3031.
Martin, Lauren (2021). What to know about diabetes in India. *Medical News Today.*
Menon, R. (2021). Risk Factors for Mortality in Melioidosis: A Single-Centre, 10-YearRetrospective Cohort Study. *The Scientific World Journal.*
Morici et al. (2019). Novel multi-component vaccine approaches for *Burkholderia pseudomallei*. *Clinical and Experimental Immunology*, 196: 178-188.
Mukhopadhyay et al. (2018). Melioidosis in South Asia (India, Nepal, Pakistan, Bhutan and Afghanistan). *Tropical Medicine and Infectious Disease* 3, 51;
Pandey et al. (2010). *Burkholderia pseudomallei* musculoskeletal infections (melioidosis) in India. *IJO* 44(2).
Ribolzi et al. (2016). Land use and soil type determine the presence of the pathogen *Burkholderia pseudomallei* in tropical rivers. *Environmental Science Pollution Research* 23:7828-7839.
Shaw et al. (2019) *PLOS Neglected Tropical Diseases*.
Stone et al. (2014). Melioidosis: Molecular Aspects of Pathogenesis. *Expert Rev Anti Infect Ther.* 12(12): 1487-1499. Doi:10.1586/14787210/2014.970634.
Sreenivas, Shishira (2022). Melioidosis: What to Know. WebMD.
Vandana, K. E. et al. (2015). Seroprevalance of Burkholderia pseudomallei among Adults in the Coastal Areas in Southwestern India. *PLOS. Neglected Tropical Diseases.*
Vidyalakshmi et al. (2012). Emerging clinico-epidemiological trends in melioidosis: analysis of 95 cases from western coastal India. *International Journal of Infectious Diseases.*