Short Report: Melioidosis as a Consequence of Sporting Activity

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Abstract. In the tropical city of Darwin, Northern Territory, Australia, dry season soil sampling cultured *Burkholderia pseudomallei* from 7 (70%) of 10 sports fields. However, during the 23 years of the Darwin Prospective Melioidosis Study, only 5 (0.6%) of 785 melioidosis cases have been attributed to infection from sports fields. In one soccer player with cutaneous melioidosis, *B. pseudomallei* cultured from the player was identical by multilocus sequence typing and multilocus variable-number tandem repeat analysis with an isolate recovered from soil at the location on the sports field where he was injured. Melioidosis is uncommon in otherwise healthy sports persons in melioidosis-endemic regions but still needs consideration in persons with abrasion injuries that involve contact with soil.

Athletes can be infected by pathogens through inoculation events caused by cuts and abrasions resulting from sporting activities.¹ A notable example is the methicillin-resistant *Staphylococcus aureus* outbreak in a professional American football team that was associated with turf-abrasion inoculation.² With the globalization of sporting competitions, the rate of returning athletes with diseases endemic to tropical regions is expected to increase, such as occurred with the Eco-challenge 2000 event in Malaysian Borneo, where leptospirosis developed in > 80 of the endurance athletes after contact with contaminated water and soil.^{3,4} Melioidosis is another disease that is endemic in tropical regions, potentially placing athletes at risk from inoculation events through broken skin and contact with contaminated soil or surface water.^{5,6}

In the early 1970s, Burkholderia pseudomallei, the causative bacterium of melioidosis was found in surface water samples of five sports fields in Singapore⁷ after a potential sporting connection in two of ten cases described from neighboring Malaysia.⁸ In Darwin in northern Australia, B. pseudomallei levels in the upper soil layers increase during the wet season (October-April), and B. pseudomallei is often cultured from surface water.9 The first use of molecular genotyping to link clinical isolates of B. pseudomallei to epidemiologically related environmental strains was published in 1994 and used ribotyping.¹⁰ However, ribotyping has limited specificity in its ability to discriminate between closely related strains of *B. pseudomallei*. Subsequently, pulsed-field gel electrophoresis was shown to be superior to ribotyping and was used to link two separate clonal clusters of melioidosis in northern Australia to contamination of the community water supplies with *B. pseudomallei*.^{11,12} Multilocus sequence typing (MLST) has more recently become the global standard for epidemiologic investigations of melioidosis, with > 1,000*B. pseudomallei* sequence types (STs) identified worldwide.¹³

Early in the 2005 Darwin dry season, a soccer player had chronic cutaneous melioidosis of the lower leg after a grassabrasion incident 2.5 months earlier on a water-logged Darwin soccer field. *Burkholderia pseudomallei* was cultured from the leg ulcer (isolate MSHR2080) and also from pus from incision and drainage of an ipsilateral inguinal lymph node abscess (MSHR2078). The patient made a full recovery after therapy for

*Address correspondence to Bart J. Currie, Menzies School of Health Research, PO Box 41096, Casuarina, Darwin, Northern Territory 0811, Australia. E-mail: bart@menzies.edu.au two weeks with intravenous ceftazidime plus oral trimethoprim/ sulfamethoxazole, followed by three additional months of oral trimethoprim/sulfamethoxazole. Environmental sampling was undertaken as a response to the incident and nine *B. pseudomallei* isolates were cultured from soil samples taken at various locations on the soccer field, including one isolate (MSHR2188) from the location where the patient identified that the abrasion incident had occurred.

Multilocus sequence typing was performed on the strains from this study, and patient strains MSHR2078 and MSHR2080 were typed as ST36. ST36 was also found in four of nine soccer field soil samples, including MSHR2188, and the remaining 5 isolates typed as ST144. ST36 has been found in clinical and environmental samples elsewhere in the Northern Territory. Therefore, to obtain greater genotypic resolution of the soccer field and clinical isolates, we performed multilocus variable-number tandem repeat analysis (MLVA) on the six ST36 isolates by using four-locus MLVA (MLVA-4) as described.¹⁴ Multilocus variable-number tandem repeat analysis can discriminate within a single MLST ST and thus can help further establish the relatedness of isolates that have temporal and spatial proximity to each other.¹⁵ MSHR2078 and MSHR2080 strains from the patient were identical at all four MLVA loci to MSHR2188 retrieved from soil at the location of the abrasion incident. In contrast, the three ST36 environmental isolates from different locations of the same field contained 1-3 MLVA locus mismatches when compared with MSHRs 2078, 2080 and 2188. The 100% MLST and MLVA match between clinical and environmental isolates supports the field soil being the source of the soccer player's infection.

The Darwin Prospective Melioidosis Study began in October 1989, and in the 23 years until October 2012, there have been 785 culture-confirmed cases of melioidosis identified at Royal Darwin Hospital. We previously documented that in 22% of cases there was a specific exposure scenario that was considered the likely infecting event, with most events being abrasion or laceration inoculations.¹⁶ Despite this finding, during 23 years there have been only 5 (0.6%) of 785 cases (including the one described in this report) where the documented suspected infecting event involved an injury on a Darwin sports field: three from soccer, one from rugby, and one from Australian rules football.

Field sports are especially popular during the Darwin dry season, when there is less surface water and *B. pseudomallei* is less abundant at the soil surface.⁹ Nevertheless, irrigation of

sports fields during the dry season is considered likely to increase survival of B. pseudomallei in upper soil layers. We therefore investigated the prevalence of B pseudomallei at ten Darwin grassed sports fields during the dry season of 2010, and collected ten soil samples per field. Using culture,17 we detected B. pseudomallei at 7 (70%) of 10 sports fields. This prevalence was higher than our previous finding of 27% (38 of 141) B. pseudomallei positive sites (environmentally disturbed and undisturbed sites) in the 2006 dry season (P = 0.008, by two-tailed Fisher's exact test), and with 16% (16 of 100) positive sports field soil samples overall, the prevalence was similar to the prevalence we found in soil samples from irrigated gardens (17%, 11 of 65).⁹ The mean pH of each positive field was within the optimal pH range for B. pseudomallei survival and growth $(5.0-6.0)^{18}$ and electrical conductivity measures of salt content were low, which is consistent with previous environmental studies of conditions favorable for *B. pseudomallei*.¹⁷

Although there may be some under-ascertainment of inoculating event histories, we have identified only five cases of sports field-related melioidosis in Darwin in over 20 years, despite the high incidence of melioidosis in Darwin, the high prevalence of B. pseudomallei in sports field soil, and the large numbers of adults and children partaking in organized sports. This low number reflects the opportunistic nature of melioidosis, a disease that predominantly affects those with defined medical risk factors such as diabetes and hazardous alcohol use, and where severe disease is relatively uncommon in healthy persons.¹⁶ Although cutaneous melioidosis without disseminated disease occurs in healthy persons,¹⁹ most melioidosis cases diagnosed in the United States and Europe are in returned travelers or residents from melioidosis-endemic regions who have recognized risk factors such as diabetes and cystic fibrosis.²⁰ The rarity of severe melioidosis in those without risk factors contrasts with leptospirosis, where severe disease is well recognized to occur in exposed healthy travelers participating in outdoor activities such as sports.^{3,4} Nevertheless fatal co-infection with both melioidosis and leptospirosis has also been recently described.²¹

In conclusion, melioidosis is uncommon in otherwise healthy sports persons in melioidosis-endemic regions. However, this disease needs to be suspected in persons with abrasion injuries that involve contact with soil.

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