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Sero–epidemiology of exposure to *Burkholderia pseudomallei* in the North Central Province

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Melioidosis is a potentially severe, soil bacterial infection caused by *Burkholderia pseudomallei*. At present melioidosis is not considered endemic in Sri Lanka. However, about 21 patients with melioidosis have been reported in Sri Lanka to date. Although manifest disease is rare, subclinical infection with *B.pseudomallei* seems to occur frequently in endemic areas. Diagnosis of previous exposure to the organism can be made by demonstrating antibodies to *B.pseudomallei* by the indirect heamagglutination (IHA) test. The IHA test is the most widely used serological assay for epidemiological surveys of subclinical melioidosis. In the IHA test, sheep erythrocytes sensitized with *B.pseudomallei* antigens are added to dilutions of the healthy blood donor's sera to detect and measure the titre of specific antibody. In this study, sera from 40 healthy blood donors in the North Central Province, 22 sera from Polonnaruwa and 18 sera from Anuradhapura, were collected. Sera were stored and transported at -20 ℃. Analysis was performed at the Department of Microbiology, Faculty of Medicine, Colombo. Antibodies against *B.pseudomallei* were detected by the standard IHA technique where antigen sensitized sheep red cells are reacted with serial dilutions of donor serum. Melioidin antigen prepared by the Department of Microbiology was used in this experiment. Sensitised red cells were made by adding 750 µl of 10% sheep Cells to 10 ml of the diluted antigen (1:80 i.e. 125 µl antigen in 10 ml saline). Unsensitised cells were used as a control to detect non-specific agglutination. All sera were inactivated on a 56 ℃ hot block for 30 minutes in capped labeled micronic tubes and 300 µl of normal saline and 100 µl of 10%Sheep Cells were added to give a 1:5 dilution. The 10% sheep cells were added to remove any non-specific agglutinins. The sera were then reacted with the sensitized cells in V shaped microwell plates to detect any antibodies. Negative reactions were detected as neat buttons. Positive reactions manifested as an agglutinated mat across the bottom of the well. Know sera for the positive control and sera for the intermediate positive control were obtained from positive samples with documented titres and sera for the negative control was obtained from a known negative sample.

In this study, antibody titres of at least 1:10 were considered positive. Three samples (7.5%) were found to have titres of 1:40 or greater. The prevalence of positive antibody titres found in sera from the Anuradhapura district was 22.2% (4/18) and the Polonnaruwa district was 18.2% (4/22). In conclusion, the extent of subclinical infection is such that it can no longer be considered a rare infection in the North Central Province of Sri Lanka.

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