OCCURRENCE OF NONTUBERCULOUS MYCOBACTERIA IN AQUATIC SOURCES OF SRI LANKA AND EFFICACY OF CHLORINATION AND IRRADIATION FOR DECONTAMINATION

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Nontuberculous mycobacteria (NTM) have been reported to cause opportunistic infections with increasing frequency, especially in immunocompromised patients. Water plays a major role in the epidemiology of NTM infection in humans, as it is one of the natural sources for transmission of this group of organisms. The current study focused on determining the occurrence of NTM in different aquatic sources of Sri Lanka by using phenotypic tests and polymerase chain reaction – restriction fragment length polymorphism (PCR – RFLP) analysis of the RNA polymerase beta subunit ($rpo\beta$) gene. Furthermore, the resistance of NTM against chlorine and ultraviolet irradiation was evaluated.

A total of 302 water samples were collected from all 25 districts of Sri Lanka, from four different water sources namely; surface water (n=88), ground water (n=76), aquarium water (n=70) and chlorinated tap water (n=68). The number of samples collected from the districts varied from 4 - 19 and water samples from 19 districts yielded positive results for NTM. Twelve surface water samples were excluded from the study owing to the overgrowth of contaminants. Of the remaining 290 samples, 45 (15%) were positive for NTM on culture. The percentage of NTM identified at species level by phenotypic tests and PCR - RFLP analysis of the $rpo\beta$ gene were 44% (20/45) and 73% (33/45), respectively. The frequency of isolation of mycobacteria for aquarium water, surface water, ground water and chlorinated water were 29% (20/70), 26% (20/76), 5% (4/76) and 1% (1/68), respectively. Eleven different NTM species were identified by PCR - RFLP namely, M. fortuitum type I (n=8), M. fortuitum type II (n=4), M. phlei (n=6), M. marinum (n=3), M. gordonae type I (n=2), M. gordonae type II (n=3), M. malmoense (n=2), M. terrae (n=2), M. avium (n=1), M. smegmatis (n=1), and M. celatum type II (n=1). The RFLP profiles of 11 NTM isolates did not match any known mycobacterium species and might represent mutants or previously undescribed NTM species. M. fortuitum type I was the most frequently isolated organism from all four water sources (8/45, 18%) as well as the predominant NTM isolate of aquarium water (8/17, 47%).

Efficacy of chlorination and irradiation on inhibition of *M. fortuitum* and *M. marinum* (two waterborne strains) revealed that *M. fortuitum* was more resistant than *M. marinum* to calcium hypochlorite and UV irradiation. Furthermore, UV irradiation (17,000 μ W/cm²) was the more effective method of disinfection as it reduced the number of detectable *M. fortuitum* and *M. marinum* to zero within one minute of contact time.

The current study suggests that water is an environmental source harboring NTM, a potential public health hazard especially for those with immunodeficiency. It also emphasizes that UV irradiation is more effective and a valid alternative to chlorination in elimination of aquatic NTM.