

SEROPREVALENCE OF RICKETTSIOSES AMONG HUMAN POPULATION OF SRI LANKA: A STUDY IN FOUR REGIONS

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Introduction

Rickettsioses are considered as paradigms of emerging infections due to their increasing incidence and expanding geographical ranges. They are obligate intracellular coccobacilli living in association with eukaryotic cells and are vectored by arthropods such as ticks, flies and mites. Antigenically they are differentiated into three groups; spotted fever group, scrub typhus group and typhus group. These vary from each other in regard to pathogenesis and clinical manifestations. In Sri Lanka, the presence of the above groups of rickettsia have been shown in clinical patients using IFAT (Kularathne *et al.*, 2001).

However, a study on seroprevalence rickettsial infection in apparently normal population may be helpful to determine the risk factors among the community. The only survey on seroprevalence carried out in Sri Lanka dates back to 1976 where <6% of the population was found to have antibodies against scrub typhus (Van Peenen *et al.*, 1976). Therefore this study was carried out to detect the presence of anti-rickettsial IgG in human population in various regions of Sri Lanka.

Materials and Methods

Serum samples

Ethical clearance was obtained from the Committee on Research and Ethical Clearance, Faculty of Medicine, University of Peradeniya.. The consent was taken from all subjects prior to blood collection and permission was also obtained from the guardians of minors.

A 2 ml sample of venous blood was drawn from each of the 83 residents who lived at Rajawatta in the Kandy district, and from 118 residents of Hemmathagama in the Kegalle district, 34 residents from Tissa Katharagama in the Hambanthota district and from 50 residents from Mahiyanganaya in the Badulla district. Serum samples were separated and stored at -70° C till further analysis. Demographic data such as gender, age, occupation were obtained from all individuals except from from Tissa Katharagama where details of age and occupation were not received. All sera were screened for presence of rickettsia specific IgG antibodies by the Indirect Immunofluorescence Antibody Assay (IFAT) set up at the Parasitology Division, Faculty of Veterinary Medicine and Animal science, University of Peradeniya, in

collaboration with CDC, Atlanta. Cultured, whole bacteria in lyophilized form were used as the antigen in the study. In this study, antigens prepared from *Rickettsia conorii* (Malish) of spotted fever group, *Rickettsia typhi* (Karp) of typhus group and *Orientia tsutsugamushi* (Wilmington) of scrub typhus group were used.

Sera were screened at 1/32 and positive samples were titered to the endpoint using two fold dilution series. Slides were examined via ultraviolet epifluorescence under 40x magnification and an IFA titer \geq 1/64 was considered positive.

Statistical analysis

Statistical analysis was done using Minitab 15 version.

Results

This study included 285 subjects of whom 142 (49.8%) were males and 143 (50.2%) were females. A near 1:1 gender ratio was observed in Tissa Kataragama and Mahiyanganaya, but in Rajawatta higher male participants and in Hemmathagama higher female participants were observed. The age group of the subjects varied from 13 to 88 years (Tissa-Katharagama excluded).

The mean prevalence of rickettsiosis in all four regions was 56.5 ± 23.3 % with seropositive rates ranging from 37.3-85.2 %. Antibodies against *Rickettsia conorii* (RC) were the most prevalent type, except in Mahiyanganaya, with mean prevalence of 47.1 ± 28.7 %. Overall seroprevalence for *Orientia tsutsugamushi* (OT) was 21.4 ± 19.84 % with Tissa- Katharagama exhibiting the highest prevalence of 47 %. None of the subjects of any region exhibited antibodies against *Rickettsia typhi*

(RT). Overall seroprevalence of the four regions ranged from 16 -76.4 % for RC and 1.6-47 % for OT. Tissa-Katharagama showed highest seropositivity for both RC and OT.

A total of 159 (55 %) subjects had IgG antibodies to one or more rickettsial agents. Of these 137 (48 %), 39 (14 %) subjects were positive for RC and OT, respectively. One hundred and thirty six (47.7 %) subjects were seropositive to a single rickettsia type, with 40.7 % positive for RC and only 7 % seropositive for OT. Twenty one (7.3 %) subjects had antibodies against both RC and OT.

There was no significant association between antibody prevalence and gender ($P > 0.05$). Antibodies against RC were the most common in both genders.

In general, seropositivity to rickettsiae seems to increase with age. However, the age group > 60 showed deviation from the pattern with higher seropositivity. Since each group was not represented by an equal number, the results obtained may be biased.

Males exposed to the field environment such as farmers, army personals, laborers had higher antibody prevalence to rickettsiae (50 %). Among the affected females, the larger proportion was found to be housewives and showed the highest seropositivity (59%).

Discussion

The study showed the presence of rickettsial specific antibodies among asymptomatic population in Sri Lanka and the prevalence of the immunogenic strains of spotted fever group and scrub typhus group of rickettsiae. However, agent of murine typhus seems to be absent or least

transmitted, as no antibodies were detected in any of the four areas.

All areas, except Mahiyanganaya, showed predominant prevalence for spotted fever group. Further studies would be required to reason out this difference. Though cross reactions between typhus and spotted fever group has been reported due to their broad antigenic relationship, cross reactions of scrub typhus with other groups have not been reported. Hence, 14 % of seroprevalence observed for both spotted fever group and scrub typhus group is more likely the result of past exposure or co-infection with these two rickettsiae.

Our studies revealed equal gender susceptibility to rickettsiae. Various studies showed increasing antibody levels for rickettsia with age. Decreasing prevalence can be possibly due to waning of antibodies with age. Men with occupations involving direct exposure to vector habitats were proven to be more prone to rickettsial infection (Tay *et al.*, 2000) and this is in agreement with the above findings. Females who were housewives or unemployed were found to show a very high prevalence rate (59%) against rickettsiae, possibly because females venture into vector habitat jungles to engage in cultivation and firewood collection. However further studies are required to prove the above hypotheses.

Conclusion

The study provided serological confirmation of the presence of rickettsial pathogen in human population in Sri Lanka suggesting that there are high rates of transmissions among populations.

Further epidemiological and vector studies need to be carried out in order for Sri Lankan health authorities to determine the geographical distribution of rickettsia. This would help the Ministry of Health to develop a strategy for diagnosis and prevention of rickettsial infection in Sri Lanka.

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References

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