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Determination of Region-Specific cut off value for Scrub Typhus IgM ELISA at a Tertiary Care Centre, Thanjavur, India

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ABSTRACT

Keywords

Orientia tsutsugamushi, Rickettsial infection, pathogen, antibiotics, monsoon period

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Introduction

Scrub typhus, caused by *Orientia tsutsugamushi* (formerly Rickettsia), is transmitted to humans by arthropod vectors of the Trombiculidae family, such as *Leptotrombidium deliense* and *Leptotrombidium akamushi* (Sayantani Chakraborty and Nilendu Sarma, 2017). It is the most common re-emerging Rickettsial infection in India, including various parts of Tamil Nadu. Thanjavur, located in the delta

Rickettsial infections are primarily diagnosed using serological methods. The cutoff value for scrub typhus IgM ELISA is not standardized and varies depending on the disease prevalence in the geographical location where the kit is used. Therefore, establishing a region-specific cut-off value for diagnostic ELISA is crucial for the initial treatment of patients and distinguishing other febrile diseases in tertiary care settings. In this study, 300 patients from three different categories, including Category I (scrub typhus IgM ELISA positive patients), Category II (Dengue IgM ELISA positive patients), and Category III (100 healthy volunteers), were enrolled. Scrub typhus IgM ELISA (InBioS, International, Inc.) was performed, and the optical density (OD) values of all 300 samples were documented and analyzed using a receiver operating characteristic (ROC) curve. The analysis revealed a diagnostic IgM ELISA cut-off value of 0.728, with a sensitivity and specificity of 94% and 91%, respectively. Considering the significance of scrub typhus diagnosis for treatment and understanding disease dynamics in the Thanjavur district of Tamil Nadu, a cut-off value of >0.728 for diagnostic IgM ELISA can be utilized.

> region, is predominantly an agricultural area with a higher risk of mite bites, particularly during the monsoon period. Therefore, it is essential to determine the region-specific cut-off value based on the endemicity of the pathogen in the area.

> Access to medical care has become more widespread, and with increased use of affordable and accurate rapid tests, a high index of suspicion for scrub typhus is necessary to improve diagnosis

and treatment of this condition, which can be easily managed with antibiotics.

Materials and Methods

Participants

A study was conducted among 100 scrub typhus IgM ELISA positive patients, 100 other fever positive patients & 100 Healthy volunteers at Tertiary care Testing Facility during the period of January 2023 to April 2023 to determine the regionspecific cutoff value for Scrub typhus IgM ELISA using InBios ELISA kit. For cases, 100 patients samples were collected (Category I). For controls, 200 samples were collected {100 samples from other fever cases (Category II) & 100 samples from Healthy volunteers (Category III)}.

Sample Collection

The samples were collected under strict aseptic precautions in accordance with the current Institutional standard operating procedure. Using sterile syringe with needle, Trained Health Care Professional after wearing Personal protective equipment like gloves & apron took the blood. Collected blood was transferred to Red vacutainer and then serum samples was tested for Scrub IgM ELISA.

Test kit and Procedure used

InBioS Scrub Typhus Detect IgM ELISA System

ELISA Procedures were done according to kit literature

Results and Discussion

The Optical Density of all 300 samples including 100 cases and 200 controls were analysed under ROC curve (Receiver Operating Characteristic Curve)

Scrub Typhus is showing a recent resurgence in our country as evidenced by reports from different parts of India in the last two decades. It is a serious acute febrile illness associated with significant morbidity and mortality. A high index of suspicion is needed in patients presenting with fever during the monsoon months. In our tertiary care centre, 40% of tested samples in December 2022 were reported as positive for Scrub typhus. Diagnosis is often missed and delay in initiating treatment owing to this may lead to untoward fatality.

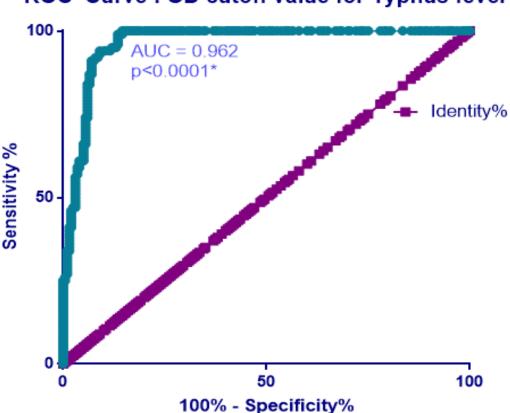
Table.1 ROC statistics

S. No.	Area under the ROC curve	Value
1	Area	0.962
2	Std. Error	0.009586
3	95% confidence interval	0.9454 to 0.983
4	P value	< 0.0001

Table.2 Diagnostic marker value (N=300; Case=100; Control=200)

S. No	Diagnostic marker	Value
1	Cutoff value	>0.728
2	Sensitivity	94%
3	Specificity	91%





ROC Curve : OD cutoff value for Typhus fever

In this study, Category I (Scrub IgM ELISA positive) was considered for the samples having OD Value more than 0.500 as per kit protocol.

Applying this, in category II out of 100 other fever samples 20 were found to be positive whereas in category III out of 100 healthy volunteer samples, 10 were found to be positive. This shows significant amount of cross reactions with the antibodies. Hence determination of the region-specific cut off value is mandatory to increase the sensitivity and specificity of the kit.

References

Sayantani Chakraborty and Nilendu Sarma. 2017 Scrub Typhus: An Emerging Threathttps://www.ncbi.nlm.nih.gov/pmc/articles/P MC5618834/ https://doi.org/10.4103/ijd.IJD_388_17

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