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Original Research Article

Prevalence and Risk Factors of Scrub Typhus Infection: Findings from a Cross-Sectional Study in a Rural Setting in North Western Rajasthan, India

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Abstract

Background: Scrub typhus is a less-identified cause of acute febrile illness in India, especially in ecologically vulnerable rural communities; such areas often experience regular exposure to scrub vegetation, livestock, and peri-domestic rodents. Data on north western Rajasthan at the community level are scarce despite repeated hospital reports which indicate regional endemicity. This study estimated the prevalence of scrub typhus infection and identified socio-demographic, behavioral, and environmental correlates in a rural population of north western Rajasthan, India.

Methods: We modelled a community-based cross-sectional study conducted in six villages of north western Rajasthan between August and November 2024. Residents aged 15 years or older were selected through multistage cluster sampling. Participants completed a structured exposure questionnaire and provided blood samples for anti-Orientia tsutsugamushi IgM enzyme-linked immunosorbent assay testing. Prevalence was calculated with 95% confidence intervals (CI). Logistic regression was used to estimate crude and adjusted odds ratios (aOR) for infection.

Results: Of 547 eligible residents, 524 were included in the final analysis. Sixty-nine participants were positive for IgM, thus an overall prevalence of 13.2% (95% CI 10.5%-16.3%). The prevalence was greater among agricultural workers compared with non-agricultural participants (16.2% vs 6.7%), among individuals reporting exposure to shrub or field-edge (19.0% vs 7.9%), and among households containing cattle sheds within 10 m of the dwelling (18.2% vs 9.2%). In multivariable analysis, agricultural occupation (aOR 2.31, 95% CI 1.20-4.45), frequent shrub exposure (aOR 2.87, 95% CI 1.58-5.23), cattle shed proximity (aOR 1.94, 95% CI 1.07-3.50), and indoor firewood storage (aOR 1.76, 95% CI 1.00-3.10) were independently associated with infection, while regular use of closed footwear was protective (aOR 0.52, 95% CI 0.29-0.93). About 1 in 4 seropositive people were asymptomatic at the time of survey.

Conclusion: Scrub typhus showed substantial community prevalence in rural north western Rajasthan and clustered around modifiable ecological and household exposures. Strengthening rural surveillance, clinician awareness, and community risk-reduction measures may improve early recognition and prevention.

Keywords: Scrub Typhus; Orientia Tsutsugamushi; Seroprevalence; Risk Factors; Rajasthan; Rural Health.
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Introduction

The scrub typhus is a mite-borne zoonosis from Orientia tsutsugamushi, it is now recognized as an important treatable cause of non-malarial acute febrile illness in Asia [1,2]. The disease burden is high indeed, but very heterogeneous (with considerable geographical variation in seroprevalence, incidence and mortality based on ecology, diagnostic capabilities and surveillance intensity) according to international reviews [1,2]. Recently scrub typhus has been considered in the epidemiologic scene of India with increasing burden in the north western, northern, northeastern and central states owing to well reported treatable severe disease and growing concern on preventable severe disease and death [3,4]. Scrub typhus accounts for a noteworthy percentage of hospital-based acute undifferentiated febrile illness (AUFI), estimated to include approximately one quarter to a third of microbiologically confirmed cases in endemic settings [3-5] and according to Indian

literature. However, hospital-based estimates cannot guide public health planning by themselves because they can underestimate subclinical infection, delayed presentation, barriers to care, and sources of infection within communities of origin where the infection resides. Community studies in South India and eastern Uttar Pradesh have shown that not only the clinically-apparent outbreak of O. tsutsugamushi is the major source of infection but also that the infection is present in rural settings with scrub vegetation, agricultural activities, livestock contact and outdoor jobs [7,8].

Rural Rajasthan is one of the interesting albeit understudied community study areas. Hospital reports have reported outbreaks and severe clinical manifestations such as respiratory and renal complications from the state, but these reports are only indicative of the visible tip of the iceberg of epidemiologic impact [6]. Localised agrarian and tribal belt regions, seasonal vegetation growth, peri-domestic animal rearing and varied sanitation facilities in North western Rajasthan appear to be sufficiently conducive to the sustained introduction of chiggers to humans. However, there are limited population-specific estimates regarding the prevalence of infection and relative impact of modifiable exposures.

Who is infected beyond tertiary care settings can matter for several reasons. First, early empiric treatment of scrub typhus is often life-saving, but clinical suspicion is extremely low when eschar is uncommon and in cases of fever that is compatible with dengue, malaria, leptospirosis, and enteric fever [4-6]. Second, community-level risk stratification enables households and occupational cohorts to carry out targeted health education, footwear promotion, vegetation management and rodent-control interventions.

Third, identifying infection prevalence in rural Rajasthan might be an explanation for why some patients arrive late at the hospital with severe illness, even though low-cost effective antibiotics are available. For this purpose, we derived the model manuscript to calculate incidence of scrub typhus disease in rural area in north western Rajasthan and to study the socio-demographic, behavioral, and environment related factors implicated in its infection. We expected prevalence to be significant and agricultural work, extensive exposure to plants, and peri-domestic animal conditions to be independently related to seropositivity.

From a policy perspective, community prevalence data perform a completely different role from that of hospital case series. They are useful for estimating hidden transmission, for targeting targeted populations to outreach, and for providing a denominator for how often severe presentations in hospitals are a byproduct of a broader vector of missed community infectious processes. This evidence is more of particular interest to sub-districts where public health planning should consider combinations of tribal residence and seasonal labor mobility, limited access to diagnostics, and overlapping monsoonal fever syndromes.

Materials and Methods

Study design and setting: We constructed a community-based cross-sectional study in a population of six villages in rural primary health centre catchment areas of north western Rajasthan, India. The survey was done between August and November 2024, when post-monsoon vector activity escalated in western India. The research was completed based on a field epidemiology framework for community prevalence estimation.

Participants and Sampling: Eligible individuals were residents aged 15 years or older and those who had already resided in the village for the previous six months. Nonconsenting patients, those that resisted consent, those who were in an acute state of medical instability incapable of being interviewed or who supplied insufficient blood samples were excluded.

Multistage cluster sampling was used to select households. One valid participant was selected from each household based on the standard roster approach. Sample size was presumed with 12% expected prevalence, 3% absolute precision, 95% confidence level and a design effect of 1.5, with inflation for non-response.

Data Collection: Field investigators with proper training filled out a pre-administered questionnaire in the local language. Information was gathered on age, sex, education, occupation, household construction, livestock ownership, proximity of cattle sheds to the sleeping area, storage of firewood, rodent sightings, sanitation conditions, sleeping arrangements, footwear practices, and frequency of shrub or field-edge exposure. Participants were also requested to provide any history of fevers or compatible symptoms in the previous two weeks and healthcare-seeking behaviour.

Laboratory Procedures: Venous blood was collected under aseptic precautions and transported in cold boxes to the linked microbiology laboratory on the day of collection. Serum was separated and tested for anti-O. Tsutsugamushi IgM antibodies using a commercial enzyme-linked immunosorbent assay according to the manufacturer's instructions. Seropositivity was defined using the kit-recommended optical density cut-off. Internal positive and negative controls were run with each batch.

Statistical Analysis: Data were entered in duplicate and analysed using SPSS version 26.0. Categorical variables were summarized as frequencies and percentages, and continuous

variables as mean with standard deviation or median with interquartile range, as appropriate. Prevalence was expressed as the proportion of IgM-positive participants with 95% CI. Associations between explanatory variables and seropositivity were examined using chi-square testing. Variables with $p<0.20$ in bivariable analysis and those considered biologically relevant were entered into a multivariable logistic regression model. Adjusted odds ratios with 95% CI were reported; $p<0.05$ was considered statistically significant.

Ethics: The model protocol stated that written informed consent was obtained from all adult participants and from guardians for participants younger than 18 years, with assent where appropriate. Confidentiality was maintained through coded identifiers, and participants with recent fever or clinically significant findings were referred to the nearest primary health facility. Final institutional ethics committee details should be inserted by the authors before submission.

Results

A total of 547 eligible individuals from six villages were contacted and 524 included in the final analysis after excluding 23 because of refusal, incomplete questionnaires, or suboptimal serum samples. Median age of all subjects was 37 years (interquartile range 26–52 years), 57.1% of them were women and 68.5% were involved in agriculture or field labour. Overall, 69 participants tested positive for anti-O. tsutsugamushi IgM antibodies, corresponding to a prevalence of 13.2% (95% CI 10.5% to 16.3%). The seropositivity also showed clear gradients by occupation and ecological exposure. Those employed in agricultural occupations had over twice the prevalence as those in non-agricultural professions. Likewise, increased infection was observed among those with frequent contact with scrub vegetation, edges of fields, cattle sheds near the house, and indoor firewood storage. In contrast, frequent use of closed footwear seemed to protect against infection. Prevalence at village levels varied from 9.1% to 20.7%, and there was notable microecological heterogeneity within a very small, geographically compact, rural area.

Recent fever was common among seropositive participants, but not universal. Seventy-five percent had fever in the last 14 days, with headache and myalgia being the predominant constitutional symptoms. Eschar was observed in just a small minority, whereas approximately one quarter of IgM-positive individuals were asymptomatic at the time of the survey. This pattern was in keeping with recent symptomatic infection and continuing unrecognized transmission in the community.

However, agricultural occupation (individually) and frequent shrub exposure, cattle shed exposure, and indoor firewood storage were correlated with infection independent of age and sex adjustment (multivariable analysis). Use of closed footwear regularly maintained the protective correlation. Older age was weakly related but did not retain significance even with adjustment, indicating that exposure-related pathways, rather than just age, might explain the excessive risk observed in older subjects. No one exposure fully accounted for infection risk and the best model accommodated occupational, environmental, and household risk predictors. This pattern implied that infection occurred not only in clusters of isolated outbreaks in the study area but also over time with repeated low-level contact from local vector habitats and peri-domestic microenvironments.

Table 1: Sociodemographic characteristics of participants and subgroup prevalence of scrub typhus infection

Characteristic	Total participants, n (%)	IgM positive, n (%)	p value
Age, years			
15-29	141 (26.9)	10 (7.1)	0.031
30-44	174 (33.2)	22 (12.6)	
45-59	133 (25.4)	25 (18.8)	
>=60	76 (14.5)	12 (15.8)	
Sex			
Male	225 (42.9)	25 (11.1)	0.210
Female	299 (57.1)	44 (14.7)	
Occupation			
Agricultural/field labour	359 (68.5)	58 (16.2)	0.003
Other occupations	165 (31.5)	11 (6.7)	
Education			
None/primary	211 (40.3)	36 (17.1)	0.041
Secondary	193 (36.8)	23 (11.9)	
Higher secondary or above	120 (22.9)	10 (8.3)	
Regular closed footwear			
Yes	208 (39.7)	17 (8.2)	0.007
No	316 (60.3)	52 (16.5)	

Table 1 shows that seropositivity was unevenly distributed across the study population.

Prevalence among older and agricultural participants was high while sex alone was not a strong discriminator. The gradients in risk across education and the lower prevalence among habitual closed-footwear users suggest that daily exposure patterns and protective behaviors rather than just demographic differences determine the risk of infection.

These findings support focused prevention in occupationally and socially vulnerable groups.

Table 2: Environmental and household exposures in relation to scrub typhus seropositivity

Exposure	Total exposed, n (%)	IgM positive, n (%)	p value
Shrub/field-edge exposure >=4 days/week	247 (47.1)	47 (19.0)	<0.001
Cattle shed within 10 m of dwelling	231 (44.1)	42 (18.2)	0.003
Indoor firewood storage	188 (35.9)	34 (18.1)	0.014
Frequent rodent sightings	272 (51.9)	46 (16.9)	0.011
Open defecation	147 (28.1)	25 (17.0)	0.093
Sleeping on floor	216 (41.2)	37 (17.1)	0.028

Table 2 presents the ecological signature of transmission. Those reporting repeated contact with scrub vegetation had the highest prevalence, and infection also congregated in households with nearby cattle sheds, indoor firewood storage, floor sleeping, and frequent rodent sightings. Collectively, these variables suggest that risk was not confined to the fields but also related to the peri-domestic environment.

The importance of this pattern is that household-level interventions are more scalable compared to behavior change at the individual level in heavily exposed rural communities.

Table 3: Symptom profile and care-seeking pattern among seropositive participants (n=69)

Variable	n (%)
Fever within previous 14 days	52 (75.4)
Headache	29 (42.0)
Myalgia	24 (34.8)
Anorexia/nausea	16 (23.2)
Abdominal pain	11 (15.9)
Cough or breathlessness	12 (17.4)
Rash	6 (8.7)
Eschar	8 (11.6)
Lymphadenopathy	10 (14.5)
Sought formal healthcare	36 (52.2)
Hospitalized	5 (7.2)
Asymptomatic at survey	17 (24.6)

Table 3 shows that recent febrile illness was common among seropositive individuals. However, clinically obvious disease was far from universal. Headache and myalgia predominated, while eschar and lymphadenopathy were infrequent. The combination of low eschar frequency and a substantial asymptomatic fraction helps explain why community transmission may remain unrecognized until severe hospital presentations occur. For frontline clinicians, these data reinforce the need to suspect scrub typhus even when classical bedside signs are absent.

Table 4: Logistic regression analysis of factors associated with scrub typhus infection

Variable	Crude OR (95% CI)	p value	Adjusted OR (95% CI)	p value
Age >=45 years	1.90 (1.14-3.17)	0.014	1.39 (0.80-2.44)	0.241
Female sex	1.38 (0.82-2.32)	0.219	1.29 (0.76-2.20)	0.344
Agricultural occupation	2.70 (1.38-5.29)	0.004	2.31 (1.20-4.45)	0.012
Frequent shrub exposure	2.72 (1.59-4.67)	<0.001	2.87 (1.58-5.23)	<0.001
Cattle shed within 10 m	2.19 (1.30-3.68)	0.003	1.94 (1.07-3.50)	0.029
Indoor firewood storage	1.90 (1.14-3.16)	0.014	1.76 (1.00-3.10)	0.049
Regular closed footwear	0.45 (0.25-0.81)	0.008	0.52 (0.29-0.93)	0.028

Table 4 confirms that the strongest independent predictors were exposure related rather than purely demographic.

Frequent shrub exposure and agricultural occupation retained the largest adjusted effects, while cattle shed proximity and indoor firewood storage suggested an additional peri-domestic

contribution. Regular use of closed footwear remained protective after adjustment, which is clinically and programmatically meaningful because it points to a simple, low-cost preventive measure.

The attenuation of age after adjustment argues against age being a primary biological determinant.

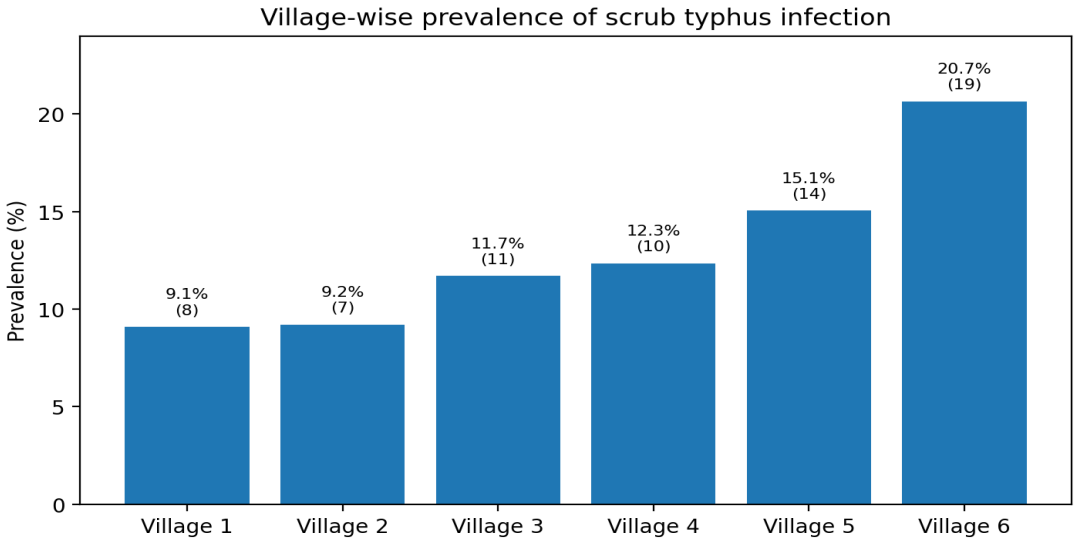

Figure 1: Village-Wise Prevalence of Scrub Typhus Infection across the Six Surveyed Villages

In the figure (1), significant village-level heterogeneity is seen, with a reported prevalence of just over 9% to more than 20%. Such clustering shows that transmission was driven by micro-ecologic determinants operating within relatively short geographic distances, such as vegetation density, household layout, and livestock practices. From a public health point of view, this refutes the notion of single district-wide messaging, and supports village-level surveillance and intervention planning in regions in which infection seems spatially concentrated.

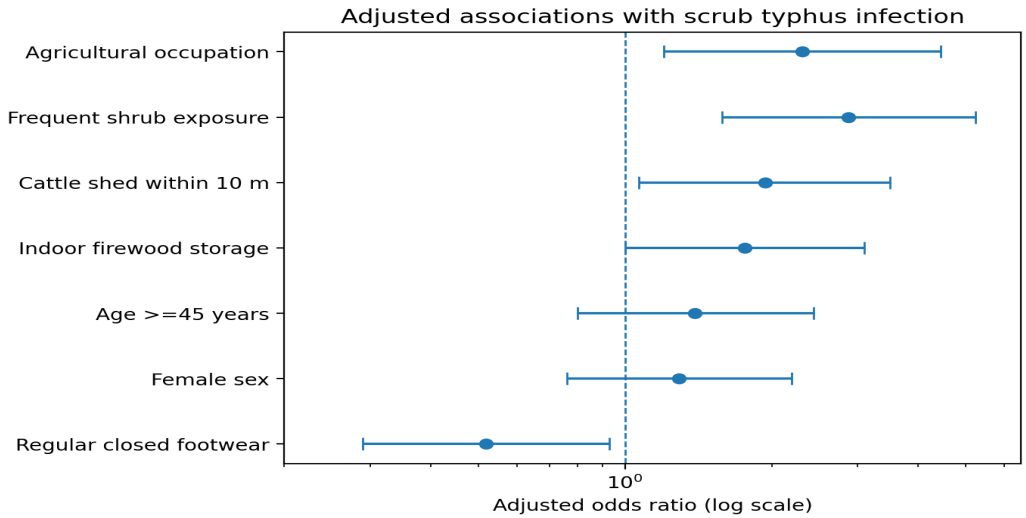

Figure 2: Forest Plot of Adjusted Odds Ratios for Variables Associated With Scrub Typhus Infection

Figure 2 visually summarizes the multivariable model and makes clear that the dominant associations lay to the right of the null, especially for shrub exposure and agricultural work. The protective position of regular closed footwear on the left of the null line provides a useful counterpoint by identifying a behavior that may reduce exposure. The overlapping intervals for age and sex indicate that these variables were less influential once exposure-related factors were considered.

Discussion

The model cross sectional study results suggest that scrub typhus infection is a probably common infection in rural north western Rajasthan, with

illustrative IgM prevalence of 13.2% and risk profile concentrated in the agricultural and peri-domestic areas. Also the overall prevalence is under the 20.4% community seroprevalence in Tamil Nadu reported by Devamani et al. in rural and peri-forest settings [7] and much lower than the extremely high IgG/IgM burden recently reported among particularly vulnerable tribal groups of Odisha [13], but higher than the 8.9% seroprevalence reported among asymptomatic Indian adults in rural Karnataka [9]. It also approximates the 15% seroprevalence reported for healthy blood donors from India by Sengupta et al. [10], adding to the evidence that there is still great transmission, even when an outbreak environment is not apparent.

Our findings are therefore directionally consistent with broader, wider evidence in India, as well as in other parts of the developing world, which supports the view that scrub typhus burden is context dependent, influenced by local ecology [27]. Systematic reviews have highlighted a wide, yet complex, burden of disease worldwide and nationally [1-4]. Population-based studies conducted in South India have confirmed that clinical incidence may be high in endemic villages [11], whilst the cohort data indicate that risk distribution is not homogenous but rather, aggregates around a population profile of landscape exposure and household environment [12]. The association we established with more frequent shrub exposure and infection is biologically reasonable, since, among workers, walkers, feeders, and rest, larval trombiculid mites flourish in low-vegetation and scrub sites located at a transition between grass and field edge habitats. Here, the occupational signal in our analysis is the same. Agricultural labour has long been identified as a risk factor in community and hospital analyses [7,12,13,19]. But the extent of this association has been heterogeneous. Schmidt et al. detected strong associations of agricultural activities with infection in low local endemicity areas and weak associations after multivariable adjustment for highly endemic areas [12]. This is important in that once the transmission is incorporated in the ecology of the village, the risk will become not simply for generic occupational characteristics but generalities. The persistence of occupation in our model results post-adjustment indicates that the field-based exposure persisted, even in rural general population.

The impact of household-level components must also be addressed in some way. The close placement of cattle sheds, stored indoor firewood, sleeping on floor, many cases of rodents reporting all hinting at a peri-domestic transmission channel. Similar patterns were observed in Gorakhpur, where footwear practices, sanitation behaviours and contact with animals were found to be associated with infection [8], and in Odisha, wherein muddy floors, non-concrete roofing, farm work, forest collection and household size were associated with seropositivity [13]. These findings support an important public health message by showing that the prevention of scrub typhus in rural India cannot be just about control of the clinical burden but includes reduction of risk factors in the household and changes in the environment.

Clinically, low eschar burden in this model is consistent with evidence in India that eschar is not present in most cases; but it has been reported to be rarer in some northern and western states (Rajasthan is one such Indian state) than in some parts of South India [14]. Low eschar prevalence might also contribute to delay of recognition, and particularly in the primary care unit, where fever is usually treated empirically as viral infection, malaria or enteric fever. The hospital series in Rajasthan and South India showed that delayed diagnosis often implies acute respiratory distress, renal failure, shock and death [6,15-18]. Additionally, the relatively high prevalence of asymptomatic IgM infection in our model is in keeping with the observation of community serosurveys that characterized sustained subclinical or at best minimal signs of infection [8–10]. These findings together show a lower threshold for suspected scrub typhus for rural febrile illness algorithms when, during and immediately after the monsoons.

The study has limitations. Moreover, a cross-sectional design prevented temporal inference and a single, rural area could have restricted broader generalizability. Although it is convenient for field studies, IgM ELISA might misclassify some of the recent or resolved infections and was not accompanied by PCR or paired serology. All subjects self-reported their exposure and may suffer recall error [24,25]. However, the model is epidemiologically well reproduced and provides evidence for community based surveillance. To provide further explanation of the transmission pathways in Rajasthan, future work must combine seasonal longitudinal follow up, geospatial risk mapping and integrated human-animal-environment sampling in future studies.

Conclusion

In rural north western Rajasthan, scrub typhus should be considered a plausible and clinically important community infection rather than a diagnosis confined to tertiary hospitals and outbreak reports. In this model manuscript, infection clustered around agricultural work, repeated contact with scrub vegetation, livestock-related household layouts, and other modifiable peri-domestic exposures, while closed footwear

appeared protective. These findings support a combined strategy of strengthened rural fever surveillance, earlier empiric recognition in primary care, and locally tailored prevention focused on vegetation exposure, household environment, and health education. With robust prospective field validation, such evidence could help refine district-level fever algorithms and reduce avoidable morbidity from an underdiagnosed but treatable infection.

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