Measuring Trachomatous Inflammation-Intense (TI) When Prevalence Is Low Provides Data on Infection With *Chlamydia trachomatis*

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PURPOSE. Clinical trachoma is the current measure of effectiveness of antibiotic and environmental improvements in trachoma endemic communities. Impact assessments measure only trachomatous inflammation-follicular (TF). Trachomatous inflammation-intense (TI) is not used for decisions on stopping mass drug administration (MDA) or achieving intervention goals. We tested the supposition that TI was not associated with *Chlamydia trachomatis* when disease prevalence is low.

METHODS. In 35 communities undergoing MDA as part of a larger project, 110 children ages 1 to 9 years were randomly selected in each community for surveys at baseline, 6, and 12 months. Both eyelids were graded for TF and TI, and a swab for detection of *C. trachomatis* infection was taken.

RESULTS. Overall TF prevalence was 5% at baseline. Cases of TI alone constituted 15% of trachoma; 37% of TI cases had infection. At 6 and 12 months, the proportion of trachoma cases that had TI only was 13% and 20%; infection rates were similar to the rates in cases with TF alone.

CONCLUSIONS. Despite low prevalence of trachoma, infection rates for TF alone and TI alone were similar at each time point. The exclusion of cases of TI alone when reporting trachoma prevalence discards additional information on infection. Trachomatous inflammation-intense could be considered as part of impact surveys.

Keywords: trachoma, trachomatous inflammation-intense, *Chlamydia trachomatis*, Tanzania

Trachoma, the leading infectious cause of blindness worldwide, is caused by repeated ocular infection with *Chlamydia trachomatis*.[1] Global efforts to eliminate blinding trachoma by 2020 include implementation of the SAFE (surgery, antibiotics, facial cleanliness, environmental improvements) strategy, which involves antibiotics to decrease the pool of infection.[1,2] In order to monitor the effect of antibiotic use in endemic districts and judge the need for further treatment, impact assessments are recommended after 3 to 5 years of program activities.[3] A single clinical disease indicator is used during assessment, trachomatous inflammation-follicular (TF).[3] However, according to the simplified grading scheme of the World Health Organization (WHO), active trachoma can consist of two measures, TF or trachomatous inflammation-intense (TI).[4,5] However, the sign of TI is not measured during mapping or impact assessment as it is thought to also indicate ocular disease that is not related to *C. trachomatis* infection.[6] Some have also suggested that it is less reliably graded, although recent work suggests this is not the case.[7] At present, only TF is reported on impact assessments or for mapping.

Research in endemic areas where both TF and TI have been measured has shown that TI is a less common clinical sign, often occurs with TF, and when present, was associated with infection.[8–16] However, much of this research was carried out in highly endemic communities, and it is possible that measuring TI when the prevalence of trachoma is low might be more problematic, especially if TI is reflective of other diseases and not infection with *C. trachomatis*. In this study, we tested the supposition that when the prevalence of TF is low, cases of TI alone are not common and not associated with infection with *C. trachomatis*.

MATERIALS AND METHODS

Study Population

This study was conducted in 35 communities in Kongwa district of central Tanzania currently enrolled in a surveillance trial. These communities underwent mass drug administration (MDA) for the previous 5 years, and these 35 were slated again to receive MDA following the baseline survey.

A complete census of households in each community was carried out at baseline, and a random sample of 110 children from each community between ages 1 and 9 years was selected

TABLE 1. Characteristics of Children in the 35 Study Communities at Baseline

Number of children in survey	3578
Age of children in survey in years mean (SD)	4.7 (2.5)
Gender of children in survey, %	
Male	48.4
Female	51.6
Household characteristics of survey children, %	
No formal education of head of household	37.5
Has latrine	78.0
More than 30 min away from water source	58.9
Roof made of mud/wood	10.6
Trachoma status, %	
TF, ±TI	5.3

to ensure at least 100 children were in the survey. Details are described below. At 6 months a new random sample was taken of 110 children in a fashion identical to the baseline survey. At 12 months, a census update was carried out, and another random sample of children ages 1 to 9 years was obtained.

### Survey Visit and Trachoma Assessment

Surveys were conducted in identical fashion at baseline, 6, and 12 months. Examination of each everted eyelid was performed by a trained trachoma grader using a 2.5× loupe. The trachoma grader was trained by a Global Trachoma Mapping Project–certified grader[16] (H.M.), and had to have a κ of >0.6 against him for each sign prior to each survey. Trachoma was assessed in both eyes using the WHO simplified grading scheme,[5] which assesses the presence or absence of TF, TI, conjunctival scarring, trichiasis, and corneal opacity. For this study, the relevant signs are TF and TI.[4]

For quality control purposes, photographs were taken of the right upper eyelid of a random 20% sample of children examined (Nikon D-40 camera with 105 mm f/2·8D AF Macro lens; Nikon, Inc., Melville, NY, USA) and all cases of active trachoma. These images were used in the main study to monitor the consistency of grading TF. We used the images to assess the agreement between TI as graded in the field and TI graded on the images. The image grader was masked to the field grade and was presented with a folder of images to grade that represented a 30% sample of all TI cases at baseline and 6 months and a 10% random sample of TI cases at 1 year, plus an overall 10% random sample of eyes that did not have any TI from the photographs in all the surveys. The images were presented randomly, without regard to field grade or survey time.

All children had ocular swabs taken from the right eye for determination of infection, using strict protocols to avoid field contamination. In each village a 5% sample of children had "air swabs" taken to check for field contamination. Samples were stored in a refrigerator and shipped to the Johns Hopkins International Chlamydia Laboratory (Baltimore, MD, USA) within 30 days to be analyzed using a commercial test for *C. trachomatis* (APTIMA ACT; Gen-Probe, Inc., San Diego, CA, USA) and processed according to manufacturer specifications. Lab personnel were masked to the identified study and air swabs. None of the air swabs throughout the study were positive.

Mass Drug Administration

Mass drug administration was provided by trained community treatment assistants, who recorded the treatment provided in a
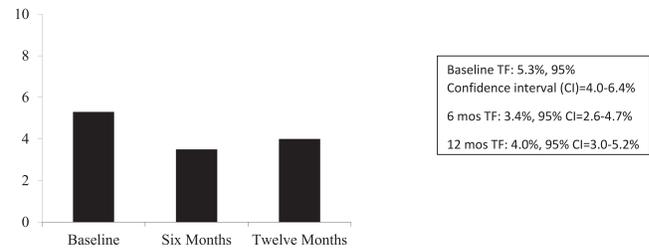
FIGURE 1. Trachomatous inflammation-follicular prevalence at baseline, and 6 and 12 months post baseline.

log book. Azithromycin was administered in a single dose according to standard guidelines: height-based dosing for children older than 1 year and weight-based dosing at 20 mg/kg to children 6 months to 1 year of age. Topical tetracycline ointment was provided to parents to treat children less than 6 months old. Coverage of children was 90% or greater.

Ethical Approval

The research complied with the tenets of the Declaration of Helsinki, and all guardians gave written informed consent for study procedures. Research was conducted with approval from the Johns Hopkins Institutional Review Board and the National Institute for Medical Research of Tanzania.

Data Analyses

For our analyses, TF could exist in the presence or absence of TI. Active trachoma was defined as the presence of either sign, TF and/or TI. The other definitions were TF alone (no presence of TI) or TI alone (no presence of TF).

The overall prevalence of active trachoma in communities was calculated, and the proportion of active trachoma cases that were TF alone and TI alone was calculated. Infection rates were estimated and compared between TF alone and TI alone. The prevalence of TF and corresponding confidence intervals were estimated for each period using the general estimating equation to account for within-village correlations. A test of significance for the differences in infection proportions between cases of TF alone and cases of TI alone at each survey round was calculated using logistic models with infection as the outcome, TF alone as the reference group, and TI alone as the predictor.

RESULTS

A total of 3578 children had both a clinical exam and a swab to test for infection at baseline, 3389 at 6 months and 3582 at 12 months. The response rate to the examination was over 88% at each visit. None of the air control swabs at any of the visits were positive, indicating a lack of contamination in the field. In the random sample of photographs presented to the grader, the field grader had called TI present in 49 right eyes. The image grader confirmed the grade in 45 of the eyes (92%). Among the 204 images where the field grader had called TI absent, the image grader confirmed the absence in 199 (98%) of the eyes.

At baseline, 78% of children lived in households with latrines, and 59% lived more than 30 minutes from a water source. At baseline, the prevalence of TF was 5.3% (Table 1).

After a single round of MDA, there was a small decline in the prevalence of TF to 3.4%, which was 4.0% by the 12-month survey (Fig. 1).

TABLE 2. Proportion of Children With No Trachoma and Trachoma, and of Those With Trachoma (TF or TI), the Proportion With Different Clinical Signs

Trachoma Status	Baseline	6 Mo Post MDA	12 Mo Post MDA
Total children	3575	3389	3582
Without any signs of trachoma (n)	94.2% (3366)	96.1% (3258)	94.9% (3400)
With TF and/or TI (n)	5.8% (209)	3.8% (131)	5.1% (182)
With TF alone (n)	3.7% (134)	2.7% (93)	2.9% (103)
With TF and TI (n)	1.3% (47)	0.7% (25)	1.1% (39)
With TI alone (n)	0.8% (28)	0.4% (13)	1.1% (40)
Active trachoma that was TI alone	13.4%	9.9%	22.0%

A summary of the distribution of clinical signs within the active trachoma cases is shown in Table 2 for each time point. The prevalence of any trachoma sign remained between 5.8% and 3.9%. Trachomatous inflammation-intense alone as a percentage of all the trachoma cases varied from 9.9% to 22%. The most frequent sign was TF alone at all three time points, but particularly 6 months after treatment.

At all points in time, even after MDA, there was infection within the group that had TI alone and at rates similar to that of TF alone (Table 3). No statistically significant difference was found between infection rates in cases with TI alone versus TF alone at any of the three visits ($P = 0.10$, 0.70, and 0.72, respectively). The infection rates in TI cases decreased from baseline (38%) to 6 months post MDA (31%) but returned to pretreatment levels by 12 months (38%).

If TI alone as a sign is not rare (in this study it comprises between 10% and 22% of all trachoma cases), and if the infection rate is similar to that of cases of TF, then what is the implication of adding cases of TI alone to the estimate of prevalence of trachoma? As Figure 2 shows, it increases the estimate approximately 1%.

DISCUSSION

In this study we found that despite a low prevalence of active trachoma in these communities, the proportion with clinical signs that presented as TI alone was non-negligible, anywhere from 10% to 22% of the burden of trachoma. Moreover, we observed a consistent relationship between TI and infection both before and after MDA that was at least as strong as that with TF and infection. The prevalence of infection in the cases of TI was always at least 30% or more. Therefore, our initial suppositions that TI was rare as TF declines and that TI was not associated with infection were not supported by the data in this study.

As the natural course of the disease suggests, intense inflammation or TI can be the first clinical sign that occurs after infection; therefore, the likelihood of being associated with infection would be expected.[4,10,11,16–18] However, in low-endemicity areas, and where MDA has been administered in multiple rounds, clinical signs and infection rate decline and the relationship between clinical signs and infection can change.[8,9,19–21] While the rate of infection we found in the TI cases was lower than that reported from more hyperendemic communities,[8–12,15,16,19,21] it was at least as high as in the TF cases. Moreover, neither TF nor TI is perfectly associated with infection, nor do we expect it to be based on biology. Children with clinical signs may have a negative test for infection as part of the course of disease where infection has cleared but remnant clinical signs remain.[8,10,14,19] Animal models also suggest that frequent reinfection may lead to a very short period of infection but prolonged clinical signs.[22]

Others have argued that the clinical signs of trachoma may be caused by infections due to other nonchlamydial bacteria.[23,24] While this may be the case for some of the disease, the fact is we found rates of infection with *C. trachomatis* in our TF cases that are not dissimilar to the rates found in TF cases from hyperendemic areas, so there is no need to invoke a role for other organisms, even with rates of TF at 4% to 5%.

We observed children who do not have signs of trachoma according to the WHO simplified grading scheme, but who do have infection. Infection rates were very low, 2%. It is possible that these children had the sample collected while in a preclinical phase, where infection is present but the clinical signs are just beginning and do not meet the WHO criteria for TF or TI; this has been reported by others.[16] We do not feel infection without the clinical signs was due to field contamination as contamination under low-prevalence conditions is unlikely and none of our air swabs were positive throughout the study.

There are some limitations to our study. Trachomatous inflammation-intense is a relatively rare sign when overall trachoma prevalence is low, so field assessment of intergrader agreement to ensure quality grading is much harder. We have photographs in a random sample of children at the time of survey and were able to confirm 95% of all field grades of TI in the sample of 49 eyes and confirm absence of TI in 98% of 204 images field graded as no TI. Others have reported good agreement grading TI in interobserver trials in endemic areas, suggesting that TI as defined by WHO can be graded reliably.[7]

What is the potential value of measuring TI? In hyperendemic settings, it is likely a good indicator that infection is

TABLE 3. Proportion of Cases Within Each Clinical Sign Who Have Infection, by Survey Visit

	Proportion of Cases Within Each Clinical Sign That Have *C. trachomatis* Infection*			
Time of Survey	No Clinical Signs, Proportion (95% CI)	TF Alone, Proportion (95% CI)	TI Alone, Proportion (95% CI)	TF and TI, Proportion (95% CI)
Baseline	1.7 (1.3–2.1)	23.9 (16.9–32.0)	39.3 (21.5–59.4)	65.2 (49.8–78.7)
6 Mo	1.4 (0.1–1.8)	25.8 (17.3–35.9)	30.8 (9.1–61.4)	60.0 (38.7–79.2)
12 Mo	2.5 (2.0–3.1)	40.8 (31.2–50.9)	37.5 (22.5–54.2)	56.4 (39.6–72.2)

CI indicates confidence interval.
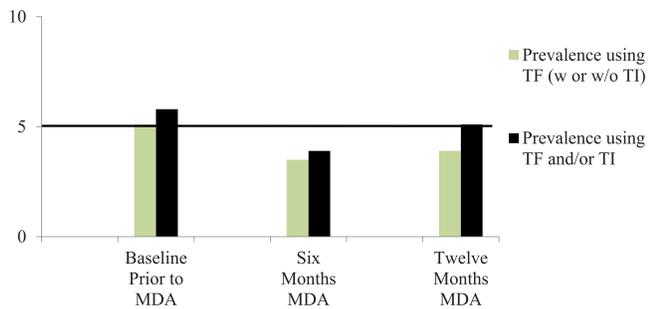* Exact 95% confidence limits.

FIGURE 2. Prevalence of trachoma excluding and including cases of TI alone.

present as TI cases have high rates of infection, but it does not add programmatically to decisions on treatment. The question is more relevant in low-endemic situations like the one in this study, where the prevalence of TF is hovering at 5%. We have shown that TI carries at least as much information about the presence of infection as does TF. By excluding TI alone in calculation of prevalence, the baseline and 12-month data would have suggested trachoma <5% and no MDA indicated. Including TI would suggest at least one more round of MDA would be indicated. A researchable question is whether the value of treating with one more round, based on the inclusion of TI as part of trachoma, would produce any greater impact than the value of not treating with MDA.

In summary, we found that in low trachoma prevalence communities, infection was equally correlated with TI alone as with TF alone. Not including TI in the grading discards information of overall prevalence of trachoma and might be considered in impact assessments or mapping in low-prevalence situations.

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