Short Research Article

Feasibility for Same Day Tuberculosis Diagnosis Using the Smear Microscopy Approach in Rural South Western Uganda

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ABSTRACT

Background:

Current WHO guidelines require that TB suspects submit 2 sputum samples on 2 different days which is time consuming, needs multiple visits leading to a high dropout. We studied the feasibility of same day sputum smear microscopy in rural refugee settlement in South Western Uganda.

Methods:

Sputum specimens were collected from tuberculosis suspects at two health centers in Nakivale refugee settlement in South Western Uganda. Patients submitted 2 spot samples one hour apart and an early morning sample was submitted the next day. All samples were stained by Ziehl-Nielsen stain. Results for the two spot samples were given on day one and the morning sample results were given on day 2. Patients found to have TB were referred for treatment on reception of their results

Results:

Of the 316 TB suspects, 190 (60.1%) were males and 126 (39.9%) were females. The mean age of the TB suspects was 40years. Overall smear positivity rate was 46/ 316 (15.0%). Of the 40 smear positive TB cases, 38(95.0%) were positive on the spot1, 35 (92.5%), on spot 2 while 31(85.0%) were positive on the early morning specimen. Only one TB suspect had negative sport1 sample that was positive with spot2 and early morning. Three TB suspects who had positive spot1 result did not return with spot2 sample. Six (15%) patients with a positive spot 1 did not bring the early morning sample. Though the bacillary load differed on the spot2 and morning samples the difference was not significant p-value > 0.05, all samples that were positive on morning sample were also positive on the spot 2. There was no association between HIV and TB infection.

Conclusion:

Same day smear microscopy for diagnosing tuberculosis is feasible in a rural setup by examining two spot samples.

Keywords: Same day microscopy, Tuberculosis, ZN,

1. INTRODUCTION

In most TB high burden countries, like Uganda, the infrastructure for the diagnosis of infectious diseases are inadequate. Despite numerous technical advances, microscopy remains the cornerstone of TB diagnosis, particularly in developing countries.[1, 2] Owing to low sensitivity of sputum smear microscopy; the diagnosis of TB requires repeated sputum examinations on several days [3]. The only diagnostic technique for TB, suitable to peripheral levels of health services, is serial sputum smear microscopy with Ziehl Neelsen (ZN) staining. Patients submit sputum samples over multiple days incur considerable costs. Direct sputum smear examination by Ziehl Neelsen (ZN) stain is a simple, economical tool that is widely used not only for diagnosis of tuberculosis and monitoring treatment of tuberculosis in several countries. It is also a key component of the DOTs and DOTS plus strategies for management of tuberculosis. With the current World Health Organization (WHO) policy on PTB diagnosis (spot-morning)[4], there is a need for a patient to make a minimum of two visits to the health facility in a bid to submit all required sputum samples, and then acquire laboratory results and treatment. This protracts the diagnostic process. Although TB services offered in Nakivale refugee settlement are free of charge there is a likelihood that patient dropout will be high because of the cost incurred to complete the diagnostic process. Failing to complete the diagnosis, therefore, is a major obstacle to accessing treatment in these settings.

The WHO has recently recommended that in countries where WHO-recommended external microscopy quality assurance systems are in place and good-quality microscopy results have been documented, it is sufficient to examine two specimens per patient and that these specimens can be collected in an accelerated scheme (called front-loaded or same-day microscopy). This study was carried out to establish the feasibility of same day tuberculosis diagnosis using sputum smear microscopy in a Refugee settlement in rural South Western Uganda

2. MATERIAL AND METHODS

The study was conducted at Nakivale Health Centre III and Kibengo Health Centre II in Nakivale refugee settlement located in Isingiro district, South Western Uganda. The settlement hosts refugees mainly from the Democratic Republic of Congo, Rwanda, Somalia, Burundi, Ethiopia and Eritrea. Sputum specimens were collected from 316 randomly selected tuberculosis suspects who attended at the Health units. Patients submitted 2 spot samples one hour apart and an early morning sample was submitted the next day [5]. All the samples received were given different numbers and stained by acid fast Ziehl-Neelsen (ZN) stain. The first two samples were examined on the first day and the morning sample was examined on the second day. ZN staining technique and smear grading were done as per the Uganda national guidelines. For quality control, all the positive slides and randomly examined, 5% of negative slides were screened by another technician at Mbarara University of Science and Technology. Patients were referred for treatment as soon they received their results. We performed data analysis using Stata Version 12.1 (Stata Corp., College station Texas). Percentages and proportions were calculated and a p-value < 0.05 was regarded as significant

Ethical clearance was obtained from the faculty research and ethics committee of the faculty of medicine of Mbarara University of Science and Technology and the institutional review board of Mbarara University of Science and Technology

3. RESULTS

Out of the 316 patients recruited in the study, 190 (60.1%) were males and 126 (39.9%) were females with a mean age of 40years. The 316 patients submitted the first sample, 262 submitted the second spot ample while 253 submitted the morning sample (Table1). Of the 316 TB suspects, 54 (17.0%) and 63 (20.0%) did not did not submit the spot2 and early morning samples respectively

The overall smear positivity rate was 46/ 316 (15.0%. Out of the 46 smear positives TB cases, 45(97.8 %) were positive on the spot1, 42 (91.3%) were positive on the spot 2 while 35(76.1%) were positive on the early morning specimen. (Table1)

Only one TB suspect had negative sport1 sample that was positive with spot2 and early morning. Of the 45 Smear positive TB cases on spot1, 10(22.2 %) patients did not bring the early morning sample. There were 7 TB suspects who had spot1 results but did not return with spot2 sample (Table 1)

Table 1: Timing of specimen collection and positivity results

	Spot1 (%)	Spot2 (%)	Early morning (%)
Negative	271 (85.02)	225(85.66)	219(86.95)
Positive	45(14.98)	42(14.34)	35(13.08)
Total	316	262 (82.9)	253(80.1)

Although the bacillary load reduced in spot2 and morning samples the difference was not significant P > 0.05. All the samples that were positive on morning sample were also positive on the spot 2. One sample was negative on spot 1 but positive on spot2 and morning and one sample was positive on spot1 (1-9/100) but negative on spot2 and morning. The patient who had a scanty positive ZN result on spot1 was also HIV positive (Table 2).

Table 2: Bacillary load of the samples collected at the different times

Types of sample	**Bacilli load**				
	1-9/100	**1+**	**2+**	**3+**	**Total**
spot1	4	7	7	27	**45**
spot2	2	6	7	27	**42**
Morning	0	4	7	24	**35**

Of the 316 TB suspects who submitted the first sample 9.2 %(29/ 316) did not submit the second spot and 19.0 % (60/316) failed to submit the morning sample. Three TB suspects

that had a positive ZN smear for the spot1 did not submit the second spot and four TB suspects that had a positive ZN smear for spot2 did not submit the morning sputum sample.

Among those who were sputum positive, 24 were HIV negative and 21 were HIV positive (Table3). There was no association between HIV and TB Infection. $x2 = 0.49$, $P = 0.48$

Table 3: Relationship between TB and HIV status

TB status	HIV status		Total n(%)
	Neg n(%)	Pos n(%)	
Neg	131 (56.73)	107(43.27)	238(100.00)
Pos	25(54.35.)	21(45.65)	46 (100.00)
Total	156(55.67)	128(44.33)	284 (100.00)

4. DISCUSSION

Many patients screened for TB abandon the diagnostic process and 22% of the participants in the present study failed to bring the second day specimens.

As patients screened with spot-morning and spot-morning-spot specimens receive all test results together, when all specimens have been examined, patients not returning the second day fail to start treatment even if their first smear was positive. Under routine conditions, this number is much more[6-8].Examination of two spot smears enables us to identify most smear-positive cases on the first day they consult which is beneficial in our setting and would lead to more TB suspects completing the diagnostic process and commencing treatment thereby limiting transmission.

Most smear-positive patients were identified by the first smear. This study shows that the spot-spot and spot-morning schemes have similar yields. This indicates that front-loaded TB diagnostic services are feasible and would not be associated with significantly less yield than the equivalent standard approach.

Although it was widely accepted that overnight specimens were more likely to contain more bacilli, it was also acknowledged that in less favorable circumstances, it was more practical to obtain specimens at the time the patient was attending the service. Though the sputum sample collected in the morning is 10% more sensitive compared to spot sample because of higher bacillary load[9], in our study there was no big difference in the yields of the spot2 sample and morning sample. Therefore if two spot samples are properly collected on the same day, morning samples may not be necessary as the difference is not statistically significant.

Studies conducted in India on the comparison of same day versus Conventional microscopy have yielded differing results. One study conducted Rajahmundry; India [9] had similar results to our study whereas two other studies in India [10, 11] reported different results. The difference in these findings, it might be related to the differences in patient characteristics and the spectrum of severity of TB in populations, which could be assessed indirectly using the distribution of smear quantification among smear positive TB patients [12].

Further, this same day diagnostic approach for PTB can help to initiate therapy on the same day and can save time as well as resources of the patients.

5. CONCLUSION

According to the findings of this study there is sufficient generalizable evidence that same-day diagnosis using the spot spot alogarithm (microscopy of two consecutive spot-spot sputum specimens) is equivalent, in terms of diagnostic accuracy, to conventional case-finding strategies by microscopy. Same day smear microscopy for diagnosing tuberculosis is feasibly in a rural setup as the two spot samples on the same day yielded comparable results and there was no much gain from early morning sample.

COMPETING INTERESTS

The authors declare that they have no competing interests

CONSENT

"All authors declare that 'informed consent was obtained from all the participants.

ETHICAL APPROVAL

This study was approved by the Institutional Ethics review committee of Mbarara University of Science and technology and the Uganda National Council for Science and Technology

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