Comparative study of OMNIgene®-SPUTUM reagent versus cold-chain for the transportation of sputum samples to GeneXpert® MTB/RIF testing sites in Malawi

Wondwossen A. Alemayehu 1*, S. Neri 1, S. Dalebout 1, R. Nalikungwi 1, A. Trusov 1, E. Ahmed 1, A. Dimba 2, A. Weirich 3, P. S. Curry 3 and C. D. Kelly-Cirino 3

Abstract

Background: The study was conducted in a remote sputum sample collection sites and GeneXpert® MTB/RIF testing centers to detect Mycobacterium tuberculosis in Malawi. The main purpose of the study was to evaluate whether sputum samples stored and transported with OMNIgene®-SPUTUM (OM-S) medium perform comparably to the routine cold-chain stored and transported samples for GeneXpert testing to detect Mycobacterium tuberculosis.

Methods: Two sputum samples from each of 362 tuberculosis suspects were randomly assigned to the OMNIgene treated (OM-S group) or the standard-of-care group (SOC; transported via cold chain). All specimens were tested at regional GeneXpert testing sites using the expectorated (raw) sputum protocol. Demographic, clinical, transport/storage and Xpert data were recorded for each specimen pair. Agreement between the SOC and OM-S groups' Xpert results was evaluated using Cohen's kappa analysis.

Results: Mean patient age was 42.3 years (range 2–79 years), 77% of patients were female, and 80% were HIV-positive. Mean transport/storage time was 6.7 days (range, 0–30 days). The rates of MTB positivity for the OM-S and SOC groups were comparable (11.8 and 11.2%, respectively), inter-test agreement was 'very good' (κ = 0.97), and overall percent agreement was 99%. Two specimen pairs (both mucoid, one 13 days transport, one 1 day transport) had discordant Xpert results.

Conclusion: OM-S-treated sputum specimens can undergo multi-day ambient-temperature storage as well as transport and yield Xpert results comparable to those of cold-chain-transported samples in Malawi.

Keywords: OMNIgene, Sputum, GeneXpert, Mycobacterium tuberculosis, Preservation, Specimen transport medium, Molecular detection

Background

Worldwide, tuberculosis (TB) is one of the top 10 causes of death, and the leading cause of mortality from a single infectious agent [1]. Likewise, it is a major public health concern and cause of mortality in Malawi. The country is also among the top 30 high-burden countries for TB/HIV co-infection. Case notifications for TB peaked at 28,000 in 2003, but detection has trended downward since then [2]. According to WHO’s 2017 report, Malawi’s case notification was 16,853 cases per year [3]. The country’s overall TB cure rate during the past decade has been among the best in Africa, and reached 88% in 2010 [4]; however, case notification has remained virtually unchanged for many years. This has been attributed to several issues, including low sensitivity of smear microscopy and high prevalence of HIV, which is associated with high numbers of negative sputum smears [5].

Difficulties involved in the collection, storage and transportation of sputum samples to diagnostic centers is rampant and significantly affects the overall TB case notification rate in Malawi. The study was designed to evaluate whether sputum samples stored and transported with OMNIgene®-SPUTUM (OM-S) medium perform comparably to the routine cold-chain stored and transported samples for GeneXpert testing to detect Mycobacterium tuberculosis.

Methods: Two sputum samples from each of 362 tuberculosis suspects were randomly assigned to the OMNIgene treated (OM-S group) or the standard-of-care group (SOC; transported via cold chain). All specimens were tested at regional GeneXpert testing sites using the expectorated (raw) sputum protocol. Demographic, clinical, transport/storage and Xpert data were recorded for each specimen pair. Agreement between the SOC and OM-S groups' Xpert results was evaluated using Cohen’s kappa analysis.

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delivery effort. TB diagnostic laboratories usually ne-
cessitate to either process the specimen shortly after col-
lection or store it at very low temperature to inhibit the
growth of contaminating micro-organisms. The latter
procedure entails additional labor and logistics costs for
the processing and conservation of specimens, and re-
ductions in sensitivity. To that end, pre-analytical tech-
nologies that preserve samples at ambient temperature
storage and transport can increase TB detection. Pro-
gram networks can benefit from such tools that maintain
sample integrity, reduce culture contamination, and ease
transport logistics. TB programs in developing countries
where sputum sample collection sites are far from diag-
nostic centers need specimen storage and transport
methods that function seamlessly at ambient temperature.

OMNIgene®-SPUTUM is supposed to serve as conduc-
tive medium for storage and transport of sputum sam-
ple at ambient-temperature and offers the flexibility of
sample batching and reduced laboratory work. OMNI-
gene®-SPUTUM liquefies and decontaminates sputum,
thus eliminating the NaOH/NALC processing step, and
preserves MTb viability for at least 8 days at tempera-
tures up to 40°C. OMNIgene®-SPUTUM treated speci-
mens are directly compatible with the full range of TB
assays, including smear microscopy, liquid culture (BAC-
TEC™ MGIT™ 960 System), solid culture, the Cepheid®
GeneXpert® System, and other molecular tests [6–8]. In
this study, Project HOPE collaborated with Malawi’s Na-
tional TB Program (NTP) to field-test whether sputum
specimens transported/stored in OMNIgene®-SPUTUM
(OM-S) at ambient temperature are comparable to cold-
chain-transported (Standard of Care; SOC) specimens for
Xpert® MTB/RIF (Xpert) testing.

Methods

Project HOPE’s TB REACH program has been operating
in all three regions of Malawi (south, central and north)
and comprises 14 implementation districts. Eight of those
districts were in the southern region with a combined
population of more than three million and more than a
quarter of Malawi’s TB cases [5]. The six remaining im-
plementation districts were in the central and northern
regions of Malawi.

The data collection of this comparative study was com-
pleted in April 2016, and prior ethical clearance was
granted by the Malawi National Health Sciences Research
Committee. Sputum samples were collected at health fa-
cilities in seven implementation districts proportionally
selected across the three regions (four in the south, two
central and two northern).

Patients and sample collection

Patients enrolled in the study were those eligible for Xpert
screening per Malawi’s NTP guideline during the study
period: i) HIV-positive outpatients who had been admitted
to the ward with TB symptoms; ii) patients identified as
having TB symptoms during screening at inpatient or in
special clinics of the health facility; iii) patients with TB
symptoms who were from countries with high multi-
drug-resistant TB (MDR-TB) prevalence. Since
some or most of the study participants or care givers may
not have the literacy level to read and provide written con-
sent, informed verbal consent was sought from all study
participants for consistency purpose.

A maximum sample size of 377 was calculated based
on 5% margin of error and 95% confidence interval. Sample
collection was performed at 29 health facility
sites within the seven selected districts. Each participat-
ing facility tracked its specimen collections until it
reached an established quota. Participants were advised
on how to produce quality sputum specimens. No pa-
tient was assisted by a respiratory therapy technician or
stimulated with hypertonic saline aerosol to produce a
sample. Two sputum specimens were collected per pa-
tient. To minimize frequency of visits and mitigate po-
tential dropouts, samples were collected within one hour
on the same day at most sites (i.e. both “spot” samples).
However, some health facilities collected a spot sample
followed by a “morning” sample the next day, as per
Malawi’s NTP guideline, and some patients’ samples
were both morning samples. Sputum character (e.g., mu-
coid, mucopurulent) was recorded for each participant.
Each individual’s samples were randomly labeled Speci-
men #1 (standard-of-care group, or SOC) or Specimen
#2 (OM-S group), and subjected to different treatments
and procedures.

Treatments

Specimen #1 (SOC group): Each control specimen
was stored and transported in a standard cold-chain
system (2–8°C), and was shipped to the district’s desig-
nated Xpert site (see details below) as soon as possible.
Specimen #2 (OM-S group): Each intervention
specimen was mixed with an equal amount of
OMNIgene®-SPUTUM (i.e., 1:1 volume), re-capped
tightly, and then stored and transported at ambient
temperature (14–28°C). These samples were also
shipped to the designated Xpert site as soon as
possible.

Smear microscopy and Xpert testing

Per Malawi’s Xpert testing algorithm, not all sputum spec-
imens must be evaluated by smear microscopy; those from
HIV positive suspects, person from high MDR-TB burden
countries and contacts, critically sick patients in the in-
patient or special clinics including re-treatment cases are
eligible for direct Xpert testing. As such, most patients in
the study qualified for direct Xpert testing. If smear microscopy was performed, this was done at the collection facility on the day of sputum collection and the technique varied by site: Ziehl-Neelsen (14%) and fluorescence microscopy (11%). Sinear results were recorded as positive or negative. In each study district, one central hospital with a functioning GeneXpert machine was designated as the district’s Xpert site. All specimens for each district were tested in the same GeneXpert machine, and any machine errors and codes were recorded. Xpert testing methods for each group are described below.

**Specimen #1 (SOC group) Xpert testing:** The cold-chain-transported specimens were processed according to Cepheid’s Xpert protocol for expectorated sputum [9]. Briefly, each Xpert MTB/RIF cartridge was labeled with the specimen ID. A sterile transfer pipette was used to transfer approximately 1 mL of raw sputum to a conical, screw-capped tube. Alternatively, the entire specimen was processed in its original sputum collection container. A separate sterile pipette was used to add 2x volume of the manufacturer’s Sample Reagent (SR buffer) to the sputum (i.e., 2:1 volume ratio). The cap/lid was then re-secured and the mixture was either shaken vigorously (10–20 times) or vortexed for 30 s. The tube/container was rested upright for 5 min at room temperature, then shaken vigorously again (10–20 times) or vortexed for 30 s, and finally rested for another 10 min at room temperature. Each specimen was inspected to ensure liquefaction (i.e., no visible sputum clumps). Any sample not fully liquefied was shaken/vortexed and rested repeatedly until liquefaction was complete.

**Specimen #2 (OM-S group) Xpert testing:** The OM-S-treated samples were also assayed directly (no additional processing) per Cepheid’s Xpert protocol for expectorated sputum [9]. Each specimen was mixed with 2x volume SR buffer (i.e., 4 mL of SR buffer was added to a typical 2 mL OM-S-treated sample). The mixture was then shaken/vortexed, incubated, and assessed for liquefaction as described for Specimen #1.

**Analysis and clinical results**

All data from collection facilities and Xpert sites were entered into statistical software (SPSS v.20) and each specimen was assigned a unique identifier to link the data. Descriptive statistics were calculated and the groups’ TB case detection rates were compared. Transport/storage time was calculated as the interval between sputum collection and the Xpert assay date. Inter-test agreement was analyzed to derive Cohen’s kappa coefficient (κ) and percent-agreement values. The resultant κ value was assigned to a “strength of agreement” category [10]: 0.01–0.20 poor; 0.21–0.40 fair; 0.41–0.60 moderate; 0.61–0.80 substantial; 0.81–1.00 very good.

Cases were clinically managed according to Malawi NTP guidelines. There were two types of discordant test outcomes: i) OM-S-negative and SOC-positive, in which case the patient was considered clinically positive, and ii) OM-S-positive and SOC-negative, in which case the patient produced a new sputum sample for separate confirmatory testing. Only original Xpert results (not confirmatory results) were analyzed in the study.

**Results**

In total, 362 specimen pairs were collected (96% of target sample size). Of these, 49 (13.5%) were excluded from analysis because of incomplete or miscoded patient/specimen ID or Xpert data. The study followed strict inclusion and exclusion criteria. All samples with less than 2 mL of solution were excluded. Table 1 summarizes the main demographic and clinical findings for the 313 participants.

Mean patient age was 42.3 ± 13.5 years (range, 2–79 years), most participants (61%) were female, and the large majority (80%) were HIV-positive. Roughly 10% had had a prior TB diagnosis or had been treated for TB. One hundred sixty-nine patients (54%) were from districts in southern region, 85 (27%) were from the central region, and 59 (19%) were from the north. Most patients provided one or both samples as spot specimens (78%), 24% provided one or both as morning specimens, and only 7% had spot/morning combination. Of the 305 samples with sputum characteristics recorded, most (43%) were mucoid, 19% were non-specific character, 14% were mucopurulent, and 13% were classified as saliva.

Regarding the Xpert results, 37 (11.8%) of the OM-S samples were MTB-positive and 269 (85.9%) were MTB-negative; the corresponding results for the SOC samples were 35 (11.2%) MTB-positive and 270 (86.3%) MTB-negative. All MTB-positive samples were also rifampicin-sensitive. Only two specimens had discordant MTB findings: Specimen PH037 (mucoid, transport time 13 days, not smear-tested) and Specimen PH268 (mucoid, transport time 1 day, not smear-tested) were both OM-S-positive and SOC-negative. Smear testing status/information was recorded for 309 patients; however, the majority (77%) were not smear-tested and only six patients (8.6% of the 70 examined) were smear-positive. In four of these cases, the SOC and OM-S Xpert results were also both positive; in the two other smear-positive cases (Specimens PH294 and PH297; sputum character mucopurulent; transport/storage time 4 days), the SOC and OM-S Xpert results were both MTB-negative. Of the 64 patients that were smear-negative, only two (Specimens PH250 and PH255; saliva and mucopurulent, respectively;
transport/storage 5 days) had Xpert results that disagreed with smear. In each case, the SOC and OM-S results were both MTB-positive. Statistical analysis revealed “very good” strength of agreement ($\kappa = 0.97$) between the OM-S and SOC Xpert tests, and 99% overall agreement of these results.

Eight SOC samples (2.6%) and seven OM-S samples (2.2%) had machine run error issues. These occurred in 10 specimen pairs: PH005, PH010, PH012, PH042, PH154, PH162, PH215, PH239, PH240, and PH290. Transport/storage time for these pairs ranged from 1 to 10 days, with seven at $\leq 6$ days. Eight of the 10 specimens were saliva or non-specific sputum character, and four of those eight specimen pairs had “error” or “invalid” outputs for both the SOC and OM-S specimen; one SOC specimen (PH042) had “no result”. The remaining two of the 10 specimens were purulent (SOC MTB-negative, OM-S Error) and mucoid (SOC Error, OM-S MTB-negative), respectively. Error codes 5007 and 2127 were recorded; some codes were not specified. According to the recorded data, no errors/issues were directly caused by the treatment methods.

Regarding transport/storage time analyses, nine of the 313 specimen pairs with Xpert results had to be excluded due to inconsistent or missing data. Mean transport/storage time for the 304 analyzed pairs was 6.7 ± 5.0 days (median, 6 days; range, 0–29 days) (Table 1). Approximately 50% of the specimen pairs were Xpert-tested within 5 days of collection, 36% between 6 and 10 days, and 15% at 11 days or later (Table 2).

**Table 1** Patient demographic and clinical characteristics

| Age (yrs) | Total n | Median | Range |
|-----------|---------|--------|-------|
| 313       | 42      | 42.3 ± 13.5 | 2–79 |

| Sex n (%) | SOC | OM-S |
|-----------|-----|------|
| Males     | 122 (39%) | 191 (61%) |
| Females   | 191 (61%) | 122 (39%) |

| Region of Malawi | SOC | OM-S |
|------------------|-----|------|
| Southern         | 169 (54%) | 85 (27%) |
| Central          | 85 (27%) | 169 (54%) |
| Northern         | 59 (19%) | 85 (27%) |

| HIV status n (%) | SOC | OM-S |
|------------------|-----|------|
| Pos              | 250 (80%) | 31 (10%) |
| Neg              | 31 (10%) | 250 (80%) |
| Unknown          | 31 (10%) | 31 (10%) |

| Prior TB diagnosis n (%) | SOC | OM-S |
|-------------------------|-----|------|
| Yes                     | 21 (9%) | 284 (91%) |
| No                      | 284 (91%) | 21 (9%) |

| Prior TB treatment n (%) | SOC | OM-S |
|-------------------------|-----|------|
| Yes                     | 31 (10%) | 280 (90%) |
| No                      | 280 (90%) | 31 (10%) |

| Collection type | SOC | OM-S |
|----------------|-----|------|
| Spot sample n (%) | 245 (78%) | 68 (22%) |
| Morning sample n (%) | 74 (24%) | 238 (76%) |
| Both types collected n (%) | 21 (7%) | 291 (93%) |

| Sputum character n (%) | SOC | OM-S |
|-----------------------|-----|------|
| Mucoid                | 130 (43%) | 28 (9%) |
| Purulent              | 28 (9%) | 130 (43%) |
| Mucopurulent          | 44 (14%) | 4 (1%) |
| Bloody                | 4 (1%) | 44 (14%) |
| Saliva                | 41 (13%) | 58 (19%) |
| Other                 | 58 (19%) | 41 (13%) |

| Transport/storage time (days) | SOC | OM-S |
|-------------------------------|-----|------|
| Median                        | 6   | 6    |
| Mean ± SD                     | 6.7 ± 5.0 | 6.7 ± 5.0 |
| Range                         | 0–29 | 0–29 |

**Discussion**

Xpert TB detection rates were comparable in the SOC and OM-S groups (both approximately 11% positivity) even though 80% of patients were HIV-positive and sputum from such individuals often contains low numbers of bacilli. Our findings suggest that sputum transported/stored in OM-S was compatible with Xpert and does not affect the sensitivity of the assay, even when a high proportion of samples were low-positive. The Xpert results were concordant with available smear results, and statistical analysis revealed 99% overall agreement between

**Table 2** Xpert and machine issue/error results listed by transport/storage time category

| Transport / Storage Time | Total n (%) | MTB + n (%) total n | Xpert Discrep\(\ast\) | Machine Run / Error Issues |
|--------------------------|-------------|---------------------|------------------------|---------------------------|
|                          | SOC | OM-S | Xpert | SOC | OM-S |
| 0–5 days                 | 148 (49%) | 15 (10%) | 16 (11%) | 1 | 2 | 3 |
| 6–10 days                | 110 (36%) | 10 (9%) | 10 (9%) | 0 | 6 | 4 |
| 11–15 days               | 24 (8%) | 5 (21%) | 6 (25%) | 1 | 0 | 0 |
| 16–29 days               | 22 (7%) | 3 (14%) | 3 (14%) | 0 | 0 | 0 |
| TOTALS\(\ast\)           | 304 (100%) | 33 (11%) | 35 (12%) | 2 | 8 | 7 |

\(\ast\) Discrep discrepancy of Xpert results (both SOC sample MTB-neg, OM-S sample MTB-pos), \(\ast\) MTB Mycobacterium tuberculosis, OM-S OMNigene\(\ast\)-SPUTUM, SOC standard of care

HIV human immunodeficiency virus, TB tuberculosis. Results of interest agreement analysis of the use of utilizing Standard-of Care (SOC) Xpert had a positive agreement of 97% ($\kappa = 0.97$)
the OM-S and SOC Xpert results, as well as 0.97 Cohen's Kappa Index (κ) value signaling strongest category of inter-test agreement [10].

The two specimens with discordant Xpert results were mucoid with transport/storage duration of 1 and 13 days. In both cases, the discordance could be due to low-positive TB status; however, this can only be speculated without supportive smear data. Machine run issues/ errors were comparable in the two groups, and most were associated with sputum samples characterized as saliva or non-specific nature. As observed in another evaluation of OM-S transport to Xpert testing in Nepal [11], the Xpert error codes reported in this study revealed no cartridge is- dependent of the sample treatment method.

The mean transport/storage time for specimens was 6.7 days and approximately 85% were Xpert-tested between 1 and 10 days post-collection; however, almost 50 samples were tested between 11 and 29 days. The results indicate that sputum transport or batching in OM-S for two weeks or more may not interfere with the Xpert assay.

Conclusions
Sputum samples can be collected at remote locations, transported/stored with OMNIgene®·SPUTUM as a medium at ambient temperature for 7–8 days, and then successfully Xpert-tested at hub sites using Cepheid’s ex- pectorated sputum protocol [9]. Statistical analysis indicated that OMNIgene®·SPUTUM treated samples (OM-S) perform comparably to cold-chain-transported samples in this protocol.

Treating raw sputum samples with OMNIgene®·SPU- TUM as storage and transportation medium can potentially help TB programs diagnostic endeavors through: i) shipping/storage at ambient temperature from remote collection sites to GeneXpert diagnostic centers, ii) eliminating additional time needed for decontamination steps in the laboratory, and iii) a larger time window for sputum testing. This study recommends broader investigations of OM-S as a sputum transport medium for culture, microscopy and other types of TB diagnostics.

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Availability of data and materials
The raw data supporting the results reported in this article can be shared to interested individuals or institutions based on written request through the corresponding author.

Authors’ contributions
WA as a principal investigator, designed and conducted the study, analyzed and summarized the data, and led the preparation of the manuscript. SN planned, oversaw study and assisted with preparing the manuscript. SD assisted with the planning of the study and analysis of data. AT assisted with the planning and design of the study. RN coordinated the study at country level. EA assisted with the summarization of the data. AW planned and coordinated the study and assisted with preparing the manuscript. PC analyzed the data and assisted with preparing the manuscript. AD field coordination and supervision of the study. C-DK-C planned and coordinated the study. All the authors have reviewed, provided inputs and approved the manuscript.

Ethics approval and consent to participate
Study participants were informed about the purpose, process, and possible outcomes of the study by the health workers collecting the sputum samples. Anticipating most of the study participants or care givers may not have the literacy level to read and provide written consent, informed verbal consent was sought from all study participants for consistency purpose. For children under 18 years old, consent was obtained from their parents or guardians accompanying them after they were informed about the study by the health workers in the facility. There was no personal identifier in the data collection tool except the sample unique identification code. Any data obtained from the study participant was not shared or disclosed in a manner that breaches confidentiality. Project HOPE has got prior ethical clearance from National Health Sciences Research Committee (NHSRC) to conduct the study.

Consent for publication
Since there are no individual images or clinical data with personal identifier consent for publication is not applicable.

Competing interests
DNA Genotek provided financial support to Project HOPE - Malawi in the form of reimbursement for consumables and reagents used during the study. A. Weirich, P.S. Curry and C.D. Kelly-Cirino were employed by DNA Genotek during the course of the study.

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3. World Health Organization (WHO). Tuberculosis country profile, Malawi. 2018. https://www.who.int/tb/country/data/profiles/en/. Accessed 20 Dec 2018.
4. Stop TB Partnership. High-burden countries lists, World Health Organization. http://www.stoptb.org/countries/tbdata.asp. Accessed 12 Feb 2017.
5. Stop TB Partnership. Malawi TB Reach information sheet. Malawi Ministry of Health and project HOPE – Malawi. 2016. http://www.stoptb.org/assets/documents/global/awards/tbreach/Malawi%20MH&M%20HOPE.pdf. Accessed 12 Feb 2017.
6. DNA Genotek Product Data Sheet PD-BR-00195. www.dnagenotek.com. Accessed 12 Feb 2017.
7. Niles J, Ray B, Curry P, Kelly-Cirino C. Compatibility of OMNIgene®-SPUTUM and prepIT®-MAX with molecular assays for tuberculosis: real-time PCR and Hain Lifescience GenoType MTBC line probe assay. DNA Genotek Whitepaper PD-WP-00051. www.dnagenotek.com. Accessed 16 Mar 2018.
8. Kelly-Cirino CD, Curry PS, Marola J, Helstrom NK, Sallinger M. Novel multi-day sputum transport reagent works with routine tuberculosis tests and eliminates need for cold chain: preliminary study of compatibility with the Xpert® MTB/RIF assay. Diagn Microbiol Infect Dis. 2016;86:273–6.
9. Cepheid. GeneXpert® Xpert® MTB/RIF Assay package insert. 2015. http://www.cepheid.com/administrator/components/com_productcatalog/library-files/3a9ca7cfa6f9bd8d6c736e5a43741eb3a-Xpert-MTB-RIF-ENGLISH-PackageInsert-301-1404-Rev-C.pdf. Accessed 12 Feb 2017.
10. Landis JR, Koch GG. The measurement of observer agreement for categorical data. Biometrics. 1977;33:159–74.
11. Maharjan B, Kelly-Cirino CD, Weirich A, Curry PS, Hoffman H, Avar K, Shrestha B. Evaluation of OMNIgene® SPUTUM-stabilised sputum for long-term transport and Xpert MTB/RIF® testing in Nepal. Int J Tuberc Lung Dis. 2016;20(12):1661–7.