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Randomized Trial of Metformin With Anti-Tuberculosis Drugs for Early Sputum Conversion in Adults With Pulmonary Tuberculosis

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Background. Metformin, by reducing intracellular *Mycobacterium tuberculosis* growth, can be considered an adjunctive therapy to anti-tuberculosis treatment (ATT). We determined whether metformin with standard ATT reduces time to sputum culture conversion and tissue inflammation in adults with pulmonary tuberculosis (PTB).

Methods. In a randomized, 8-week, clinical trial, newly diagnosed, culture-positive PTB patients were randomized to standard ATT (HREZ = control arm) or standard ATT plus daily 1000 mg metformin (MET-HREZ = Metformin with Rifampicin [METRIF] arm) for 8 weeks during 2018–2020 at 5 sites in India. The primary end point was time to sputum culture conversion by liquid culture during 8 weeks of ATT. Plasma inflammatory markers were estimated in a subset. A Cox proportional hazard model was used to estimate time and predictors of culture conversion.

Results. Of the 322 patients randomized, 239 (74%) were male, and 212 (66%) had bilateral disease on chest radiograph with 54 (18%) showing cavitation. The median time to sputum culture conversion by liquid culture was 42 days in the METRIF arm and 41 days in the control arm (hazard ratio, 0.8; 95% confidence interval [CI], .624–1.019). After 8 weeks of ATT, cavitary lesions on X-ray (7, 5.3% vs 18, 12.9%; relative risk, 0.42; 95% CI, .18–.96; *P* = .041) and inflammatory markers were significantly lower in the METRIF arm. Higher body mass index and lower sputum smear grading were associated with faster sputum culture conversion.

Conclusions. The addition of metformin to standard ATT did not hasten sputum culture conversion but diminished excess inflammation, thus reducing lung tissue damage as seen by faster clearance on X-ray and reduced inflammatory markers.

Clinical Trials Registration. Clinical Trial Registry of India (CTRI/2018/01/011176).

Keywords. host-directed therapy; immunomodulation; metformin; pharmacokinetics; tuberculosis.

Currently, the treatment regimen for drug-sensitive tuberculosis (TB) consists of 4 drugs for 6 months, with a treatment success rate of around 80%–85% [1]. The long duration of such regimens combined with adverse drug reactions leads to poor adherence to treatment, especially after the subsidence of symptoms. Highly potent treatment regimens could allow the treatment duration to be shortened, thus improving adherence and reducing adverse drug effects. However, conventional pathogen-targeted strategies suffer from the serious disadvantage of fostering microbial resistance.

To circumvent this problem, a new paradigm in drug discovery involves the modulation of host cell responses to improve pathogen eradication [2]. Studies have shown that pulmonary TB patients with bilateral or cavitary disease have increased plasma levels of proinflammatory cytokines interferon (IFN)-γ, tumor necrosis factor (TNF)-α, and interleukin (IL)-17A, as well as elevated acute phase proteins such as C-reactive protein and serum amyloid protein [3-5]. Host-directed therapies (HDTs) enhance the host immune defenses by targeting these inflammatory pathways and interfering with the host mechanisms used by the pathogens [6-8]. Among potential drugs for HDT, metformin is one such commonly prescribed drug for TB patients [9]. Singhal et al reported that metformin inhibits the intracellular growth of *Mycobacterium tuberculosis* in infected mice, restricts disease immunopathology, and enhances the efficacy of conventional anti-TB drugs [10, 11]. Metformin with its antiinflammatory and antioxidant properties has been used in other nondiabetic conditions [12, 13]. Retrospective studies of TB patients on metformin for the management of diabetes mellitus showed fewer cavities on chest X-ray, less severe TB disease with better clinical outcomes, and reduced relapse

rates and mortality when compared with those who were on non-metformin treatment [10, 14]. However, these studies were either retrospective or had a small sample size and were in the context of metformin being used as antidiabetic therapy.

The Metformin with Rifampicin (METRIF) trial was a randomized, phase 2B, efficacy trial of metformin added to daily anti-TB treatment (ATT) to evaluate the time to and proportion of sputum culture conversion in adults with pulmonary TB. Plasma drug concentrations and proinflammatory cytokine biomarkers were also measured after 8 weeks of ATT in the 2 groups.

METHODS

Study Design
This study was a randomized, open-label, parallel-arm clinical trial conducted at 5 sites in India: Indian Council of Medical Research (ICMR)-National Institute of Research in Tuberculosis (NIRT), Chennai, Madurai, and Vellore in South India; All India Institute of Medical Sciences, New Delhi, in North India; and ICMR-National AIDS Research Institute, Pune in West India. The scientific advisory committees and the ethics committees of participating institutes approved the study. The study is registered with the Clinical Trial Registry of India (CTRI/2018/01/011176), and the full study protocol has been previously published [15].

Study Participants
Adults enrolled in the study were aged ≥18 years with newly diagnosed sputum smear-positive PTB and susceptible to rifampicin as detected by cartridge-based nucleic acid amplification test. Patients were excluded if positive for human immunodeficiency virus or hepatitis B or C, pregnant or breastfeeding, or had abnormal liver enzymes, diabetes mellitus, or exposure to ATT for more than 7 days.

All eligible participants were counseled by a trained counselor and provided with detailed study information. For those willing to provide written informed consent, sputum was tested for acid-fast bacilli (AFB) by smear, culture by both solid (Lowenstein Jensen [LJ]) and liquid media (mycobacteria growth indicator tube [MGIT]), and drug susceptibility testing for isoniazid, rifampicin, and ethambutol. The growth of mycobacteria on either the liquid or solid medium was considered a positive culture. Blood was drawn for hemogram, liver and renal function tests, and blood sugar levels.

Randomization and Stratification
Eligible patients were randomized to either of the study regimens in a 1:1ratio. Permuted block randomization was done centrally using a computer-generated list of random numbers stratified by the presence or absence of cavities on chest X-ray and highest sputum smear grading (≤2 or >2). For all sites, allocation codes were generated centrally by a team of statisticians

at NIRT at the time of study admission and communicated to the site physician by e-mail for treatment initiation.

Study Procedures
For the initial 8 weeks, participants received either the standard ATT (HREZ, control arm) or standard ATT plus metformin given orally as 500 mg once daily for 1 week followed by 1000 mg of metformin daily for the remaining 7 weeks (HREZ + metformin, METRIF arm) under supervision. This dose of metformin was selected based on previous animal studies and its equivalent human dose, known adverse events of metformin, and profile of TB patients in the country [10, 16]. The standard ATT regimen consisted of daily HREZ based on weight bands per national guidelines [17]. After the initial 8 weeks, all patients continued the standard continuation phase with isoniazid, rifampicin, ethambutol (HRE) for 16 weeks to complete the full course of ATT of 6 months (Figure 1).

During treatment, sputum samples were collected once a week during the first 8 weeks and once a month for the remaining 16 weeks and tested for AFB by smear, MGIT, and LJ cultures. Hemogram and liver and renal function tests were repeated at 4 weeks and 8 weeks. Plasma proinflammatory cytokines, considered markers for disease severity and bacterial load in PTB, were measured in a subset of individuals at baseline (n = 83) and at 8 weeks (n = 74) of ATT. Plasma levels of acute phase proteins (APPs) alpha-2 macroglobulin, C-reactive protein (CRP), haptoglobin (Hp), and serum amyloid P (SAP) and inflammatory markers IFN-γ, TNF-α, IL-17A, and IL-1β were measured in a subset of individuals (n = 44) for APPs using a Milliplex MAP Human CVD Panel Acute Phase magnetic bead panel 3 (Millipore) and for inflammatory markers using the Luminex Human Magnetic Assay kit (R&D Systems) according to the manufacturers' instructions. Various pharmacokinetic parameters including maximum concentration (C_{max}) and partial area under the concentration-time curve (AUC) over 12 hours (AUC0-12) were also estimated by study regimen in a subset (n = 34) of study participants. The primary end point of the study was the time to sputum culture conversion, defined as the interval between the treatment initiation date and the date of acquisition of the first of at least 2 consecutive negative cultures without an intervening positive culture by MGIT taken at least 8 days apart during the 8 weeks of ATT.

Statistical Analyses
In a study by Wang et al, the median time to sputum culture conversion by liquid medium with daily ATT was shown to be 32 days [18]. With the addition of metformin, the assumption was a 30% reduction in the time to sputum culture conversion, that is, 22 days. To achieve this target at 80% power, 0.05 level of significance, and a 5% loss to follow-up, 158 patients were required in each treatment arm, totaling 316 patients for the study (approximately 320 patients). The 30% reduction was an
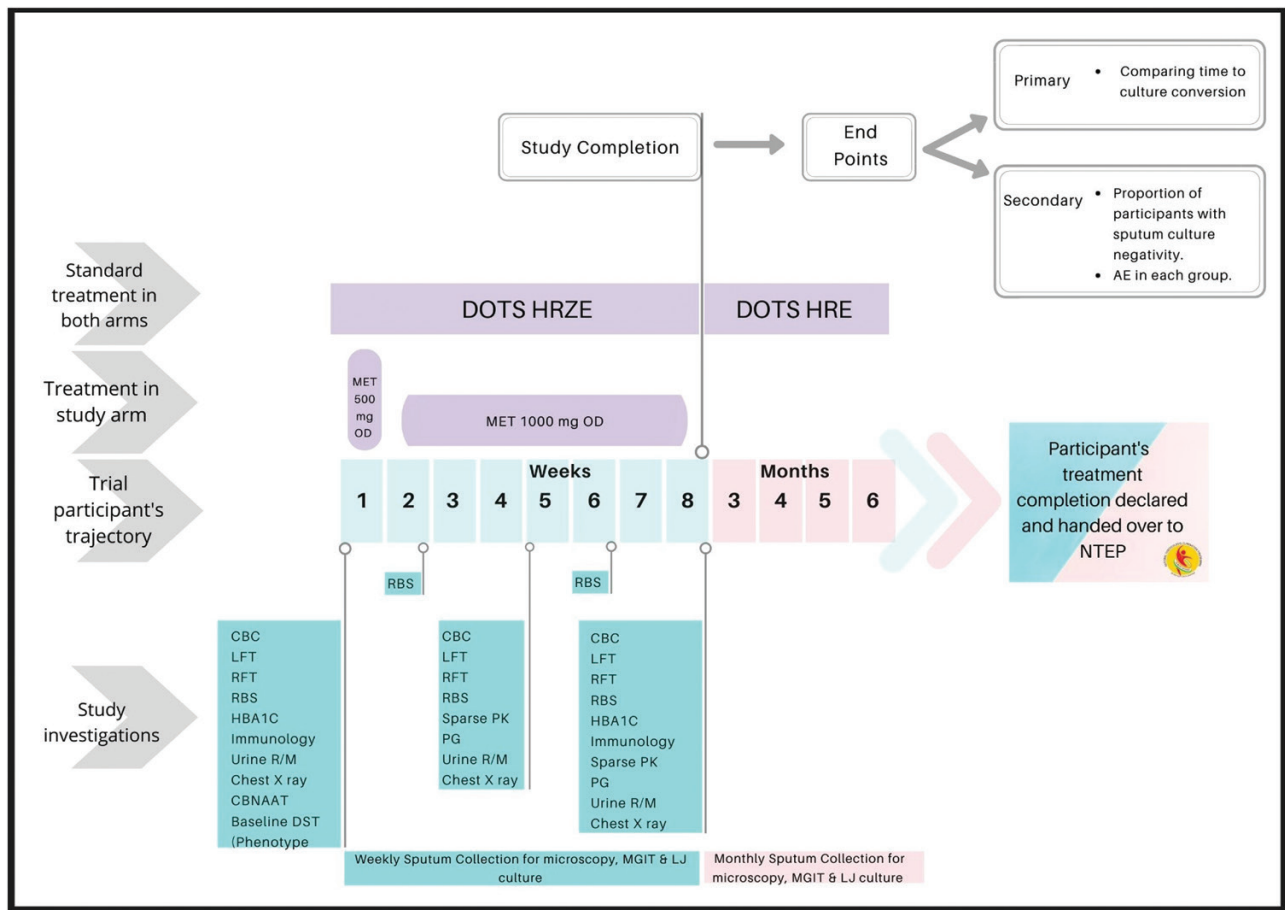
Figure 1. Study diagram of the METRIF clinical trial. Abbreviations: AE, adverse events; CBC, complete blood count; CBNAAT, cartridge based nucleic acid amplification test; DOTS, directly observed treatment short course; DST, drug susceptibility test; HbA1C, glycosylated heamoglobin; HRE, isoniazid, rifampicin, ethambutol; HRZE, isoniazid, rifampicin, pyrazinamide, ethambutol; LJ, Lownstein Jensen; LFT, liver function test; MGIT, Mycobacteria Growth Indicator tube; MET, metformin; METRIF, metformin with rifampicin; NTEP, National Tuberculosis Elimination Programme; OD, once daily; PG, pharmacogenomics; PK, pharmacokinetic; RBS, random blood sugar; RFT, renal function test; R/M, routine and microscopy.

assumption that we made considering a clinically meaningful reduction to keep the time to conversion within 1 month. Data were captured using REDCap software, and analyses were performed with SPSS, version 21.0.

Demographic, clinical, bacteriological, and blood parameters of the 2 study groups were summarized as frequency, percentage, mean (standard deviation), and median (interquartile range). The Shapiro-Wilk test was used to test the normality of data. Log transformation was performed for nonnormal data (pharmacokinetic parameters), then parameters were estimated and expressed by applying antilog (exp(x)). An independent sample t test was performed and expressed as 95% confidence intervals (CIs) with a corresponding mean difference. The Mann-Whitney U test with Holm's correction for multiple comparisons was performed for acute-phase proteins and cytokine parameters on the log-transformed data. Analyses were performed using Graph-Pad PRISM, version 9.0. Chest X-ray results were compared at baseline and at 8 weeks of treatment. Two readers read the X-rays independently, and any

discrepancies were resolved by an umpire reader. The Kaplan-Meier method was used to estimate culture conversion (time to event) and hazard patterns between the 2 groups; the significance was analyzed using the log-rank (Mantel-Cox) test. The Cox proportional hazard model was performed to estimate the hazard ratio (HR) for those variables that were clinically significant from univariate analysis. Statistical significance was expressed as a P value, calculated using the χ^2 test to determine the proportion with culture conversion between the 2 groups. To assess safety, clinical drug adverse events were classified based on systemic examination, while the laboratory-related parameters were classified based on division of AIDS criteria [19].

RESULTS

From a pool of 550 new sputa smear-positive PTB patients screened at the study sites between 15 June 2018 and 7 March 2020, 322 were enrolled in the trial; 160 were allocated to the METRIF arm and 162 to the control arm (Figure 2). Of these,
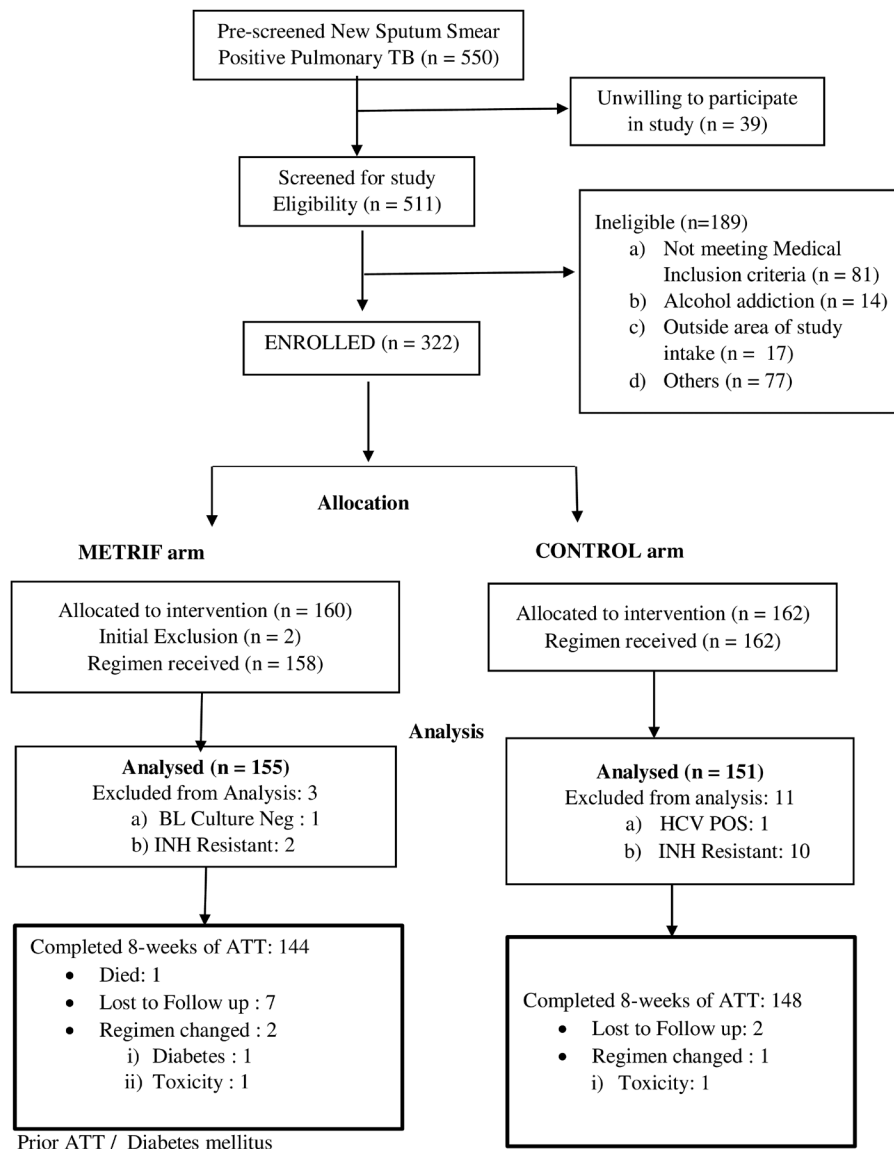
Figure 2. Enrollment and end point at 8 weeks: profile of trial participants. Abbreviations: ATT, anti-tuberculosis treatment; BL, baseline; HCV, hepatitis C virus; INH, isoniazid; METRIF, metformin with rifampicin; POS, positive; TB, tuberculosis.

155 in the METRIF arm and 151 in the control arm completed the study and were included in the final analysis. Enrolled participants with *M. tuberculosis* isolates resistant to isoniazid were excluded from the final analysis.

The clinical characteristics of the trial participants are given in Table 1. The majority of participants were male; productive cough and weight loss were the most common presentations. Three-fourth of the participants had advanced disease with high bacillary load in the sputum and bilateral disease on chest X-ray. Peripheral smear in 207 participants revealed normocytic normochromic anemia in 70%.

Sputum Culture Conversion

The median time to sputum culture conversion by MGIT was 42 days (95% CI, 39.67–44.33) in the METRIF arm and 41 days (95% CI, 38.03–43.97) in the control arm (*P* = .058; Figure 2). However, a significant difference was noticed in the time to culture conversion by LJ medium (35 days; 95% CI, 33.58–36.42) in the METRIF arm and 29 days (95% CI, 25.24–32.76) in the control arm (*P* = .05). Also, there was a significant difference in the proportion of patients who showed culture negativity by MGIT at the end of 8 weeks—72% in the METRIF arm and 86% in the control arm (*P* = .004; Table 2). This difference was also noticed in LJ culture (86% vs 94%, *P* = .03) but not in sputum smear conversion (76% vs 77%, *P* = .79).

Cox proportional hazards regression analysis for MGIT time to stable culture conversion after 8 weeks of treatment revealed an HR of 0.80 (95% CI, .62–1.03; *P* = .082) for the METRIF arm after adjusting for age, body mass index, gender,

Table 1. Baseline Characteristics of the Intention-to-Treat Population

Variable	All (n = 322)	METRIF Arm, 2 MetHREZ$_7$/4HRE$_7$ (n = 160)	Control Arm, 2HREZ$_7$/4HRE$_7$ (n = 162)
Mean age (SD), years	32.7 (11)	33.3 (12)	32.1 (11)
Mean weight (SD), kg	45 (7)	46 (7)	44 (7)
Mean body mass index (SD), kg/m^2	17.5 (3)	17.9 (3)	17.1 (3)
	n (%)	n (%)	n (%)
Male gender	239 (74)	123 (77)	116 (72)
Ever smoker	101 (31)	56 (35)	45 (28)
Current smoker	32 (32)	20 (36)	12 (27)
Sputum smear status			
2+ and 3+	251 (78)	124 (78)	127 (78)
1+ and scanty	71 (22)	36 (22)	35 (22)
L-J culture			
2+ and 3+	214 (67)	111 (69)	103 (64)
1+ and colonies	100 (31)	45 (28)	55 (34)
Negative	8[a]	4	4
Median time to positivity, days	7.15 (5.0–14.0)	6.65 (4.9–14.0)	7.65 (5.1–14.0)
Cavity on chest X-ray	54 (18)	27 (19)	27 (18)
X-ray involvement			
Unilateral	106 (33)	53 (33)	53 (33)
Bilateral	212 (66)	106 (66)	106 (65)
Chest X-ray zones			
3 and below involved	183 (57)	95 (59)	88 (54)
>3 zones involved	134 (42)	63 (39)	71 (44)
Peripheral smear			
Normocytic normochromic	143 (70)[b]	73 (70)	70 (68)
Microcytic hypochromic	54 (27)	26 (25)	28 (28)

Abbreviations: L-J culture, Lownstein Jensen culture; MetHREZ, Metfromin, Isoniazid, Rifampicin, ethambutol, pyrazinamide daily; 2MetHREZ/4HRE, 2 months of MetHREZ/ 4 months of HRE; SD, standard deviation.

[a]Patients were positive on cartridge-based testing.

[b]Of the 322 participants, peripheral smear was done for 207 (105 in the METRIF arm and 102 in the control arm).

smoking, alcohol consumption, and smear grading (Figure 3). After 8 weeks of treatment, the independent risk factor associated with longer time to sputum culture conversion included consumption of alcohol (HR, 0.61; 95% CI, .44–.86; P = .005) and higher sputum smear grading (HR, 0.56; 95% CI, .42–.74; P <.0001), whereas higher body mass index led to faster culture conversion (HR, 1.05; 95% CI, 1.001–1.097; P = .046).

Radiological Improvement

At baseline, cavity on chest X-ray was in an equal proportion of patients in the study arms, that is, 27 (19%) patients in the

Table 2. Proportion of Patients Showing Sputum Culture Conversion in the MGIT Liquid Medium During the 8-Week Treatment Period

	MGIT (Liquid Media) Culture Conversion					
	Metformin (N = 155)			Control (N = 151)		
Day of Conversion	Pos	Neg	% Neg	Pos	Neg	% Neg
7	141	13	8.44	137	13	8.67
14	135	16	10.60	125	22	14.97
21	121	26	17.69	114	35	23.49
28	108	36	25.00	100	45	31.03
35	88	52	37.14	83	60	41.96
42	72	66	47.83	62	84	57.53
49	56	81	59.12	41	99	70.71
56–60	40	104	72.22	21	127	85.81
	P = .0043					

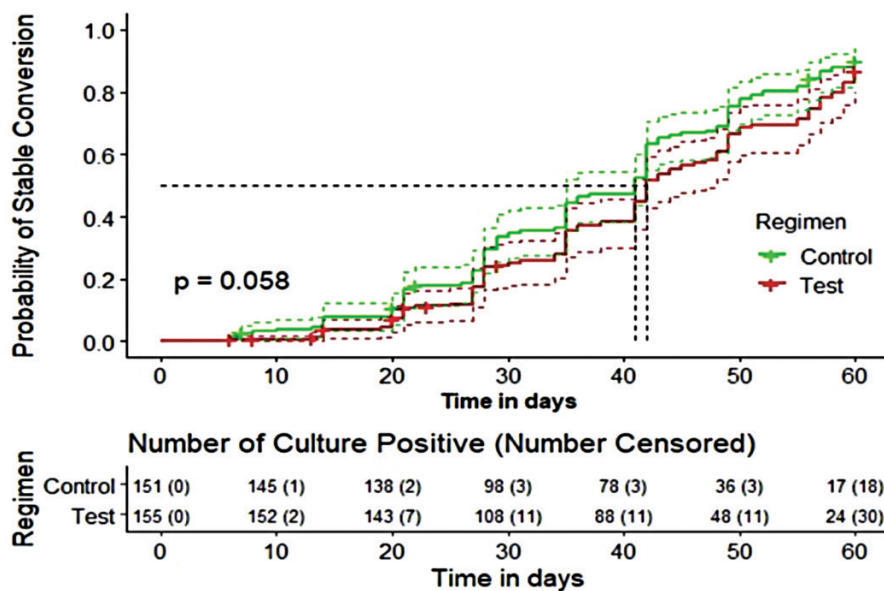Abbreviations: MGIT, Mycobacteria Growth Indicator Tube; Neg, negative; Pos, positive.

Figure 3. Time to MGIT stable culture conversion in the METRIF arm and control arm (with 95% confidence interval). Abbreviations: METRIF, metformin with rifampicin; MGIT, Mycobacteria Growth Indicator tube.

METRIF arm and 27 (18%) in the control arm. By the eighth week, cavity on chest X-ray was noticed in 7 (5.3%) patients in the METRIF arm and 18 (13%) patients in the control arm. The relative risk was 0.42 (95% CI, .18–.96), showing a statistically significant difference between the 2 arms (P = .041). Clinically, a higher proportion of patients in the control arm continued to be symptomatic (especially cough) at the fourth week and the eighth week compared with the METRIF arm. However, this was not statistically significant (Supplementary Table 1).

Adverse Events

Nausea and vomiting (30 vs 4, P < .001) or vomiting alone (47 vs 13, P < .001) were seen more frequently in the METRIF arm compared with the control group. Episodes of hemoptysis were frequent in the METRIF arm (11 vs 8, P = .46), while arthralgia was noticed less in the METRIF arm than in the control arm (16 vs 22, P = .32). Both were not statistically significant (Table 3).

Serious Adverse Events

There were 11 serious adverse events (SAEs) —1 death and 10 hospitalizations reported in the study. Of the 11, 8 were in the METRIF arm and 3 were in the control arm. The death occurred early in the course of treatment, during the second week of ATT, and was attributed to suicide and not related to the study regimen. The remaining 10 SAEs included hospitalization due to acute abdominal pain (3), blood-streaked sputum (2), vomiting (1), pyrexia of unknown origin (1), acute breathlessness (1), deranged liver function tests (1), and intractable hiccups (1). All patients were conservatively managed after hospitalization and were discharged upon improvement. Treatment

had to be changed in 2 patients due to SAEs secondary to incessant vomiting and abdominal pain.

Acute Phase Proteins and Inflammatory Cytokines

Plasma levels of acute-phase proteins CRP, a2M, SAP, and Hp were similar in both arms at baseline (Supplementary Table 2a). At the eighth week of ATT, patients in the METRIF arm exhibited significantly diminished circulating levels of these acute-phase proteins compared with the control arm: CRP (P = .0094), a2M (P = .0032), and Hp (P = .0236) (Figure 4A). Similarly, the plasma levels of proinflammatory markers IL-17A, IL-1β, TNF-α, and IFN-γ, which were similar in both arms at baseline (Supplementary Table 2b), exhibited significantly diminished plasma levels in the METRIF arm compared with the control arm after 8 weeks of treatment (Figure 4B).

Pharmacokinetics

The Cmax values for rifampicin and isoniazid were not significantly different between arms (Table 4). Clearance of both rifampicin and isoniazid was higher in the METRIF arm compared with the control arm. Overall, 85% of all participants had rifampin Cmax values below the therapeutic target in both arms, while the Cmax values for isoniazid and pyrazinamide were within therapeutic range.

DISCUSSION

Although the study suggests that addition of metformin to the standard ATT did not significantly shorten the time to sputum culture conversion, metformin addition was associated with faster resolution of the cavity on chest X-ray and

Table 3. Adverse Events in Study Participants During the First 8 Weeks of Treatment

Systemic Classification	Adverse Event	METRIF Arm	Control Arm
Gastrointestinal tract	Oral ulcer/Angular cheilitis	-	3 (1)[a]
	Ascites	1	-
	Nausea and epigastric pain	2 (1)[b]	1
	Epigastric pain	6	1
	Nausea	6	3
	Vomiting + loose stools	1[a]	-
	Nausea + vomiting	30 (2)[a]	4
	Nausea + lower abdominal pain	-	1
	Abdominal pain	3	2
	Loose stools	1	1
	Vomiting + epigastric pain	-	1
	Vomiting	47 (3)[a]	13 (3)[a]
	Jaundice	-	1
General system	Loss of appetite	2 (1)[a]	2 (1)[b]
	Fever	1[a]	4
	General symptoms	1	4 (1)[a]
Musculoskeletal system	Arthralgia	16 (2)[a]	22 (3)[a]
	Myalgia	3 (1)[a]	10 (1)[a]
	Myalgia + arthralgia	1[a]	-
Renal system	Urinary tract infection	1	2
	Bile pigments in urine	2	-
	Elevated uric acid + arthralgia	4 (1)[b]	3
	Pedal edema + arthralgia	-	1
	Pedal edema	-	2 (1)[a]
Respiratory system	Hemoptysis	11 (1)[a]	8 (2)[a]
	New-onset cough + dyspnea	-	1
	Pleural effusion	1	-
Skin and subcutaneous system	Itching	6	5
	Rash	-	1
	Itching + rash	1	-
	Pustule over back	1	-
	Papular skin lesions	2	1
Cardiac system	Chest pain	3	2
Ear, nose, and throat	Ear discharge	-	1
Peripheral nervous system	Peripheral neuropathy	-	1
Genital system	Menorrhagia	1	-
Opthalmologic system	Redness of both eyes	1	-

Abbreviation: METRIF, metformin with rifampicin.

[a]Moderately severe.

[b]Severe.

reduction in the level of circulating plasma proinflammatory cytokines after 8 weeks of treatment in patients with PTB. This suggests that a metformin-containing 5-drug regimen may reduce the infectiousness and transmissibility in patients with the cavitary disease. It may also play a role in reducing the post-TB lung sequelae, such as fibrosis or structural damage, by a faster decline in the proinflammatory cytokines. However, it did not affect the 2-month culture conversion rate at the dose used in this study and hence cannot suggest shortening the duration of treatment. The findings suggest exploration of a higher dose or extended use of metformin in culture conversion in addition to its antiinflammatory property.

The results are consistent with a preclinical mouse model study that showed that mice that received metformin in combination with the first-line ATT had a similar bacillary burden in the lung compared with control mice that received only ATT [20]. This contrasts with Singhal et al who used the same mouse model (BALB/c) and metformin dose of 250 mg/kg and showed decreased bacillary load in lungs compared with untreated or isoniazid-treated mice, which formed the basis of this clinical trial [10]. The discrepancy between these 2 studies exists because Singhal et al tested metformin as an adjunctive agent in combination with a single anti-tubercular drug (isoniazid or ethionamide), whereas Dutta et al studied the activity in combination with the more potent first-line regimen
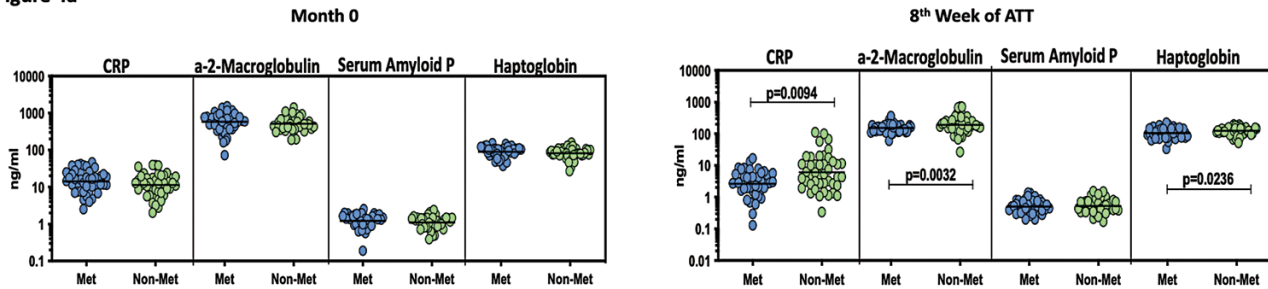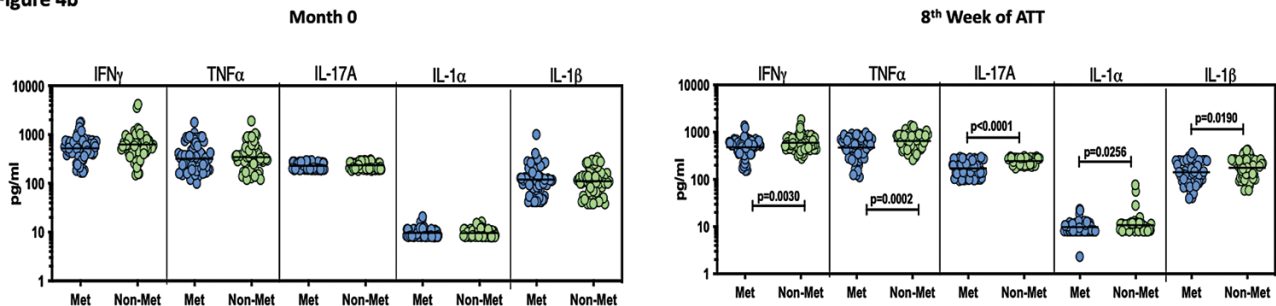
Figure 4. *(A)* Baseline (pretreatment) and 8 weeks of anti-tuberculosis treatment (ATT) plasma levels of acute phase proteins in pulmonary tuberculosis (PTB) patients in the METRIF arm and control non-metformin arm (n = 44). *(B)* Baseline and 8 weeks of ATT plasma levels of proinflammatory cytokines in PTB patients in the METRIF arm (n = 82) and non-metformin arm (n = 81). The data are represented as scatter plots with each circle representing a single individual. Abbreviations: ATT, anti-tuberculosis treatment; CRP, C-reactive protein; IFN, interferon; IL, interleukin; Met, metformin; METRIF, metformin with rifampicin; TNF, tumor necrosis factor.

RHZE, which might mask an adjunctive role for metformin. Also, the inclusion of rifampicin may have altered the pharmacokinetics of metformin as rifampicin may accelerate the metformin clearance or vice versa and reduce drug exposures [21]. Commonly cited therapeutic ranges for rifampin, isoniazid, and pyrazinamide are 8–24 µg/mL, 3–6 µg/mL, and 20–50 µg/mL, respectively. In our study, the Cmax of rifampicin was near the lower limit of the therapeutic range in both arms, and

Table 4. Pharmacokinetic Parameters of Study Participants in Both Arms of the Trial

	Study Regimen		Mean Difference
	METRIF Arm (n = 31)	Control Arm (n = 32)	(Exponentiated 95% Confidence Interval for Mean Difference)
Parameter	Mean + SD	Mean + SD	
Rifampicin PK parameters			
C_{Max}, µg/mL	3.77 + 2.08	4.86 + 1.84	−1.09(0.55–1.09)
T_{Max}, hours	3.11 + 1.71	3.29 + 1.52	−0.18(0.74–1.21)
Elimination rate constant, h^{-1}	0.23 + 1.91	0.25 + 1.64	−0.02(0.71–1.27)
Half-life, hours	2.96 + 1.91	2.80 + 1.64	+0.16(0.79–1.41)
$AUC_{(0-t)(µg/mL \cdot h)}$	19.39 + 2.04	27.31 + 1.89	−7.92(0.51–1.00)
$AUC_{(0-inf)(µg/mL \cdot h)}$	23.86 + 1.88	30.47 + 1.96	−6.61(0.56–1.09)
Clearance, mL/min	19.24 + 1.81	13.71 + 1.80	+5.53(1.04–1.89)[a]
Isoniazid PK parameters			
C_{Max}, µg/mL	3.11 + 2.37	3.87 + 1.94	−0.76(0.55–1.73)
T_{Max}, hours	2.05 + 1.62	2.22 + 1.91	−0.17(0.70–2.01)
Elimination rate constant, h^{-1}	0.19 + 1.84	0.15 + 1.99	+0.04(0.88–2.41)
Half-life, hours	3.69 + 1.84	4.50 + 1.99	−0.81(0.59–1.81)
$AUC_{(0-t)(µg/mL \cdot h)}$	16.43 + 2.52	21.82 + 1.87	−5.39(0.51–1.66)
$AUC_{(0-inf)(µg/mL \cdot h)}$	20.84 + 2.77	30.37 + 2.02	−9.53(0.44–1.56)
Clearance, mL/min	12.27 + 2.86	7.36 + 1.99	+4.91(1.07–2.91)[a]

Abbreviations: AUC, area under the concentration-time curve; METRIF, metformin with rifampicin; PK, pharmacokinetic; SD, standard deviation.

[a]$P < .005$ significant.

the clearance of both isoniazid and rifampicin was higher in the METRIF arm. The faster clearance of drugs could explain the failure to notice the adjunctive role of metformin in the given dose in our study.

Sputum culture conversion at 8 weeks is an accepted marker of sterilizing activity of treatment regimens [22]. A correlation has been shown between the time to culture positivity and smear microscopy grading during ATT [23, 24]. Sputum smear positivity at the end of 8 weeks of ATT indicates infectiousness and an increased chance of drug resistance [25, 26]. In our cohort, 109 (76%) patients in the METRIF arm and 114 (77%) in the control arm showed sputum smear negativity, while 104 (72%) in the METRIF arm and 127 (85%) in the control arm were MGIT culture-negative at the end of 8 weeks of ATT. Unlike Abinaya et al [27] and Marupuru et al [28], we did not find additional benefit in terms of bacterial conversion by adding metformin to ATT.

Systemic inflammation is a hallmark of TB, typically characterized by elevations in the levels of APPs, including CRP, a2M, Hp, and SAP [29, 30]. Also, persistently increased levels of proinflammatory cytokines can prolong the underlying immune pathology in TB disease [3]. The association between diminished production of inflammatory cytokine and increased phagocytic activities has been shown when *M. tuberculosis*–infected cells were treated with metformin [31]. Our data are in agreement with data from previous studies and show a reduction in the plasma APPs and proinflammatory cytokines after 8 weeks of ATT in the METRIF arm compared with the control arm. Patients in the METRIF arm had an almost 60% higher chance of closure of the lung cavity by 8 weeks, similar to a study in Singapore [10]. These strengthen the role of metformin as an adjunct therapy to ATT by faster healing and less damage to the lung parenchyma, resulting in fewer post-TB sequelae.

A higher frequency of mild nausea and vomiting that subsided after a week was encountered in the metformin arm. As metformin is known to cause gastrointestinal adverse events, it is suggested that a loading dose be given for a week before the full dose is started.

Limitations of our study include use of a single dose of metformin as adjuvant therapy with ATT. The smaller dose used here may have failed to detect any association with time to sputum culture conversion. Higher doses could influence the time to culture conversion. Second, as metformin is pitched as host-directed adjuvant therapy to ATT, estimating inflammatory markers and its effect on structural (computed tomography scan) and functional changes (pulmonary function test) in the lung could have been studied in a larger group.

In conclusion, this study showed that adding 1g of metformin to the standard ATT for 8 weeks did not shorten the time to sputum culture conversion in adult sputum-positive PTB patients. However, it was associated with faster resolution of the cavity on chest X-ray and reduced plasma inflammatory markers, which may result in fewer post-TB sequelae. Adjunctive therapy with host-modulating agents such as metformin should be useful in shortening the duration of TB treatment if a balance can be struck between the antimicrobial effect and host inflammation that can mitigate TB sequelae.

Supplementary Data

Supplementary materials are available at *Clinical Infectious Diseases* online. Consisting of data provided by the authors to benefit the reader, the posted materials are not copyedited and are the sole responsibility of the authors, so questions or comments should be addressed to the corresponding author.

Notes

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