



Culture and drug susceptibility testing among previously treated tuberculosis patients in the Dominican Republic, 2014

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ABSTRACT

Multidrug-resistant tuberculosis (MDR-TB) is a major public health concern that threatens global progress toward effective TB control. The risk of MDR-TB is increased in patients who have received previous TB treatment.

This article describes the performance of culture and drug susceptibility testing (DST) in patients registered as previously treated TB patients in the Dominican Republic in 2014, based on operational research that followed a retrospective cohort design and used routine program data.

Under the current system of TB culturing and DST, the majority of patients with previously treated TB do not undergo DST, and those who do often experience considerable delay in obtaining their results. The lack of DST and delay in receiving DST results leads to underestimation of the number of MDR-TB cases and hinders the timely initiation of MDR-TB treatment.

Key words

Tuberculosis; tuberculosis, multidrug-resistant; culture techniques; microbial sensitivity tests, standards; cohort studies; operations research; Dominican Republic.

Multidrug-resistant tuberculosis (MDR-TB) is a major public health concern that threatens global progress toward effective TB control (1). The risk of MDR-TB is increased in patients who have received previous TB treatment. Previously treated TB patients are those who have, at some point in the past, received one month or more of antituberculosis drugs. Globally, they comprise a significant proportion of TB notifications (13%) (2).

Previously treated TB patients are further classified into the following subgroups based on the outcome of their most recent course of treatment: 1) relapse (those who completed treatment and develop another episode of smear-positive pulmonary TB); 2) retreatment after loss to follow-up (those who were lost to follow-up on treatment and return to treatment smear-positive); and 3) treatment failure (those who

become or remain smear-positive five or six months into treatment) (3, 4).

MDR-TB is defined as a form of TB caused by bacteria resistant to rifampicin and isoniazid. The risk of MDR-TB is increased in patients who have received previous TB treatment. In 2013, it was estimated that, worldwide, 480 000 people developed MDR-TB and 210 000 died. Early identification of MDR-TB through culture and drug susceptibility testing (DST) and initiation of MDR treatment with second-line antituberculosis drugs increases the chance of a cure and prevents the development and spread of further resistance (5). Therefore, the World

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Health Organization (WHO) recommends that sputum samples be obtained from all patients previously treated for TB for culture and DST (6).

CULTURE AND DRUG SUSCEPTIBILITY TESTING IN THE DOMINICAN REPUBLIC

In the Dominican Republic, 4 200 pulmonary TB cases were registered in 2014, including 3 556 new and 564 previously treated cases. WHO estimated that 6.6% of the new TB cases and 20% of the retreatment cases have MDR-TB. The Dominican Republic is therefore classified as a country with a high MDR-TB burden (4, 7). In line with the WHO guidelines, the Dominican Republic's National Tuberculosis Program (NTP) recommends that sputum samples be collected from all TB retreatment patients for culture and DST for first-line drugs using the proportion method. However, given the large gap between the estimated number of MDR-TB patients (340) and the number of patients initiated on MDR treatment (93), it is likely that this recommendation is not followed, leaving many MDR-TB patients untreated.

A functioning and adequate testing system requires that sputum samples be collected at peripheral health services before the start of TB treatment, that the samples be sent to higher-level laboratories for culture and DST, and that all test results be sent back to the peripheral health services. Lack of proactive follow-up in this process may lead to unnecessary delays in testing, or providing DST results to, patients at risk for MDR-TB (previously treated TB patients). Indeed, studies from other countries have reported that difficulties in obtaining samples, transferring them to central laboratories, getting the results back, and initiating treatment at appropriate times have been principal causes of the gap between estimated and detected MDR-TB cases (8–10).

The authors hypothesize that due to suboptimal follow-up of the MDR diagnostic process, DST results are delayed or not available for a significant proportion of previously treated TB patients in the Dominican Republic, and that this may explain the low number of MDR-TB patients registered as having begun treatment. To improve diagnosis and treatment of patients with MDR-TB, it is important to know which steps in the diagnostic process create barriers to

treatment, such as preventable delays in testing/diagnosis and loss to follow-up.

To help identify these barriers, this research evaluated the performance of the culturing and DST system among previously treated pulmonary TB patients in the Dominican Republic.

Research design, setting, population, and variables studied

This operational research used routinely collected program data and followed a retrospective cohort design. Study implementation and reporting of results followed guidelines developed by the STROBE (STrengthening the Reporting of OBservational studies in Epidemiology) initiative.

The Dominican Republic is an upper-middle-income country in the Caribbean with 10 million inhabitants. TB incidence has declined over the past decade and is currently estimated at 60 per 100 000 per year (4). HIV prevalence among TB patients is 25% (4). The NTP operates at three levels: 1) the central level—the Ministry of Health; 2) an intermediary level—38 provincial or municipal health directorates; and 3) a peripheral level—1 563 health centers. The NTP began implementing the DOTS⁵ strategy in 1998 and gradually scaled up to cover 95% of the total population. The national TB laboratory network consists of one central reference laboratory with DST services; 12 regional laboratories that perform smear microscopy and culture, but not DST; and 193 laboratories that perform smear microscopy only.

To diagnose MDR-TB, a sputum sample for all patients previously treated for TB and other at-risk groups (e.g., health workers, prisoners, HIV-infected people, and MDR contacts) should be submitted for culture and DST. According to national procedures, sputum samples should be sent to a regional laboratory for culture and to the central laboratory for DST. Culture is performed on solid Ogawa–Kudoh or Löwenstein–Jensen media and *Mycobacterium tuberculosis* is identified according to standard techniques. DST for first-line drugs is done using the MGIT™ 960 Mycobacterial Detection System. In the Dominican Republic, DST can be done for rifampicin, isoniazid, pyrazinamide, ethambutol, streptomycin, capreomycin, kanamycin,

amikacin, levofloxacin, and ofloxacin. For external quality control of DST, the National Reference TB Laboratory receives strains every year from the Supranational Reference Laboratory (SRL) in Chile. The strains are processed and results are compared with the results of the SRL.

Both standardized and individual MDR treatment regimens are available in the Dominican Republic. Once a patient is diagnosed with MDR-TB, standardized or individual MDR treatment based on DST results should begin immediately. Patients with treatment failure can begin a standardized second-line treatment while the results of DST are under way. Contacts of known MDR-TB patients can also begin a specific treatment right away. If a contact of an MDR-TB case is diagnosed with TB based on clinical or bacteriological evidence, the patient can begin the same second-line treatment as the index case while waiting for the DST results. A national committee for drug-resistant TB confirms all individual MDR treatment regimens. According to the national MDR-TB guidelines, patients can either start a specific treatment in a specialized MDR-TB unit in a hospital, or they can start ambulatory treatment in one of 15 health centers. The remaining duration of the treatment is continued on an ambulatory basis. There are two specialized MDR-TB units in the country. MDR-TB treatment and services are provided free-of-charge to all patients.

The study population included all patients previously treated for TB that were registered again in 2014 with an active case of TB.

The following patient data were collected: age; sex; HIV status; TB category (relapse, retreatment after loss to follow-up, treatment failure); and the provincial or municipal health directorate where the patient was registered. The research assessed whether or not 1) sputum samples were collected, 2) culture and DST were done, 3) results were available, and 4) patients with MDR-TB had begun treatment. To describe the time taken for each step, the following dates were recorded: registration as a previously treated TB patient, sample collection, results available, and start of treatment. Data sources included 1) the individual patient treatment cards, 2) the regional and central laboratory registers, and 3) the second-line TB treatment register. When there were discrepancies between data sources, the data from the patient treatment cards were used.

⁵ DOTS is the internationally recommended strategy for TB control until 2005, and the foundation of the new Stop TB Strategy introduced in 2006.

Analysis, statistics, and ethics approval

Data were collected using a structured form, entered into EpiData Entry software version 2.2 (EpiData Association, Odense, Denmark), and analyzed using EpiData Analysis version 1.1–2.2.171. Frequencies were calculated to report on demographic and clinical characteristics, patients for whom sputum samples were collected, culture and DST performed, and those who had started MDR-TB treatment. Variables about duration (calculated as number of days between two steps) had abnormal distribution so were expressed as medians and interquartile ranges (IQRs).

Ethics approval was received from the Ethics Advisory Group of the International Union Against Tuberculosis and Lung Disease (“The Union”) (Paris) and permission was obtained from the NTP in the Dominican Republic. Because the study involved routine program data and did not contain patient identifiers, the issue of informed consent did not apply.

RESULTS

A total of 427 TB patients were registered as previously treated patients in the Dominican Republic in 2014. Their demographic and clinical characteristics are summarized in Table 1. Mean age was 41 years (standard deviation: 15 years) and 72% were men. HIV status was known for 350 (82%) patients: 77 were HIV-positive (22%) and 273 were HIV-negative. A total of 259 (61%) of the patients were classified as relapse, 136 (32%) as retreatment after loss to follow-up, and 32 (7%) as failure. A total of 226 (53%) of the previously treated cases were registered in the Metropolitan Regional Health Service.

The proportion of cases that achieved each step in the culture and DST process, along with the timing, is shown in Figure 1. Of the 427 previously treated TB patients, sputum was collected in 324 cases (76%); of those, culture was performed in 252 (78%) and positive cultures were obtained in 105 (42%). For 100 patients with positive cultures, DST was processed, and DST results were available for 82 cases, representing only 19% of the previously treated patients registered during the period. For 68% of cases, the DST results were available in health centers; the remaining DST results were found in the databases of the MDR-TB unit and the national TB laboratory. A total of 40 out of 82 cases with DST

TABLE 1. Demographic and clinical characteristics of previously treated tuberculosis (TB) patients, Dominican Republic, 2014

Characteristic	No.	(%)
Sex		
Men	309	(72)
Women	118	(28)
Age group (years)		
< 15		0
15–34	161	(38)
35–54	193	(45)
≥ 55	70	(16)
Not recorded	3	(1)
TB category		
Relapse	259	(61)
Retreatment after loss to follow-up	136	(32)
Failure	32	(7)
HIV ^a status		
Positive	77	(18)
Negative	273	(64)
Unknown	77	(18)
Regional Health Service		
Metropolitan	226	(53)
Valdesia	36	(8)
Cibao Norte	65	(15)
Cibao Nordeste	14	(3)
Enriquillo	11	(3)
Este	41	(10)
El Valle	14	(3)
Cibao Occidental	12	(3)
Cibao Central	8	(2)

Source: prepared by the authors based on the study results.

^a HIV: human immunodeficiency virus.

results (49%) were sensitive to all first-line drugs, 36 (44%) were MDR-TB, and 6 (7%) had other types of drug resistance. A total of 33 (92%) of the 36 patients diagnosed with MDR-TB began specific treatment for MDR-TB. Five of them started second-line treatment before their DST results were known. In addition, eight patients who never got a DST-based diagnosis of MDR-TB started treatment with second-line drugs.

Among 259 patients classified as relapse, 45 (17%) had DST results available, of which 15 were MDR (33%). Among 136 retreatment patients after loss to follow-up, 19 (14%) had DST results available, of which 4 were MDR (21%). Among 32 patients with treatment failure, 18 (56%) had DST results available, of which 17 were MDR-TB (94%). The proportions of MDR (33% in patients with relapse, 21% in retreatment patients after loss to follow-up, and 94% in patients with treatment failure) differed significantly (Fisher’s exact test: $P < 0.0001$). Among the 77 HIV-positive patients, 15 (19%) had a DST result available: 7 were sensitive to all first-line drugs, 6 had MDR-TB, and 2 had other types of drug resistance.

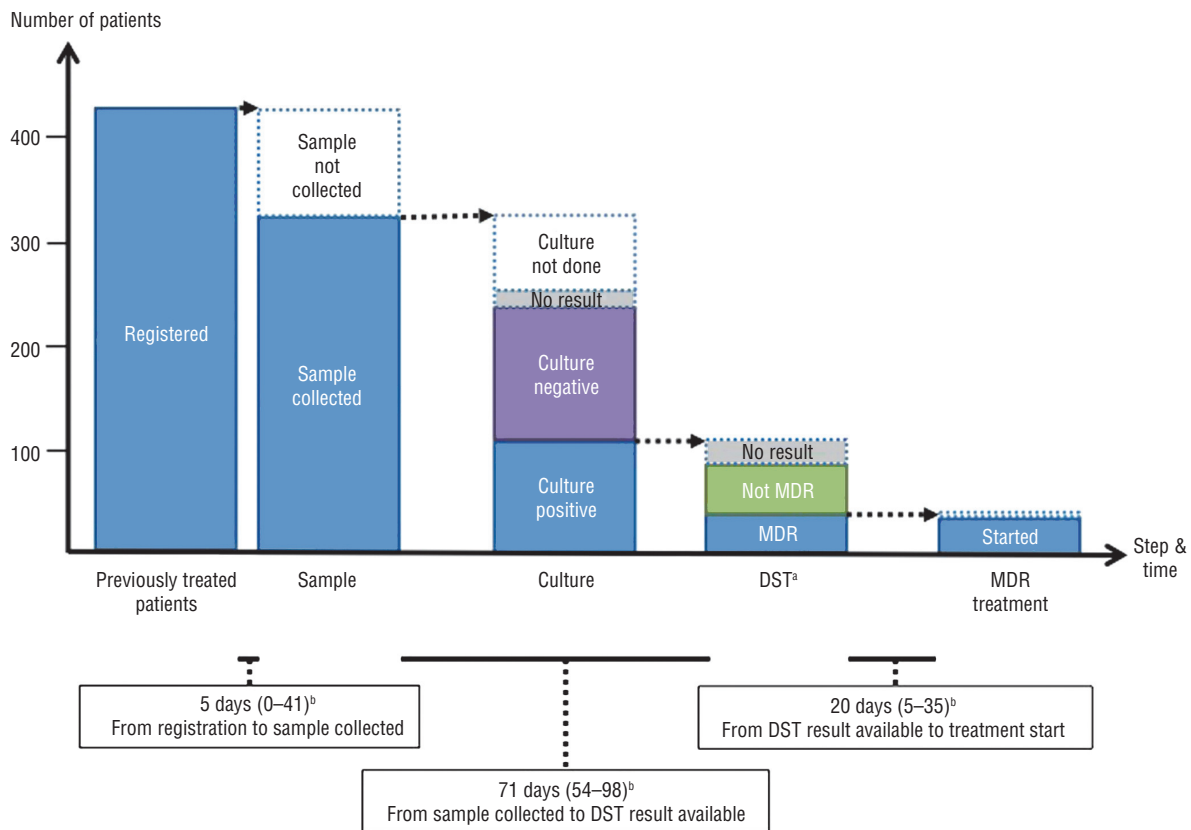
The median duration between registration as a previously treated TB patient and collection of sputum samples was 5 days (IQR: 0–41 days). Culture results were available at the health center level for 82 of the 252 patients for whom a culture was done (33%). DST results were available for 64 of the 100 patients for whom DST was done (64%). The median time between sputum collection and availability of DST result was 71 days (IQR: 54–98 days), and the median time from DST result to start of treatment in those diagnosed with MDR-TB was 20 days (IQR: 5–35). For patients diagnosed with MDR-TB, the median time between registration as a previously treated TB patient and start of MDR-TB treatment was 136 days (IQR: 82–177).

Implications of the research

This is the first study published in the Dominican Republic to describe the process of culturing and DST among patients with previously treated TB. The findings confirm the authors’ hypothesis that management and follow-up of these patients is suboptimal. A significant proportion of patients were lost in the diagnostic process. Failure to collect samples or to get samples to the appropriate laboratory accounts for a considerable part of the loss. The fact that more than half of the obtained samples failed to grow in culture deserves further attention and might be explained by the timing of sample-taking (sometimes after treatment start) or the conditions of sample transport to the laboratories. Overall, less than 20% of registered TB patients actually had DST results available to inform their subsequent management and treatment.

Where culture and DST results were available, about one-fifth of patients retreated after loss to follow-up, one-third of patients who relapsed, and nearly all those with treatment failure had MDR-TB. These figures may overestimate the real proportion of MDR-TB because it is possible that patients with other risk factors for MDR-TB were followed up more proactively and hence more likely to have available DST results. Nevertheless, the fact that among the patients with DST results the proportion of MDR-TB was as high as 44% is an issue of concern.

Among patients diagnosed with MDR-TB, there was a median duration of more than four months between being registered with previously treated TB and starting

FIGURE 1. Diagnostic process from patient registration at health centers to start of multidrug-resistant tuberculosis (MDR-TB) treatment for previously treated TB patients, Dominican Republic, 2014

Source: prepared by the authors based on the study results.

^a DST: drug susceptibility testing.

^b Median number of days (interquartile range).

effective treatment. A considerable part of this delay can be attributed to the diagnostic methods used. Culture by solid media and DST using the MGIT™ 960 Mycobacterial Detection System can take months before it yields results. On an encouraging note, the majority of patients diagnosed as MDR-TB did begin treatment.

The HIV status of 82% of the previously treated TB patients was known, and 18% of them were HIV-positive. In this study, DST availability and results were similar in HIV-positive and HIV-negative patients. Nevertheless, given that the severe implications of MDR-TB in HIV-infected patients are well known, the proportion of TB patients screened for HIV should improve, as well as the availability of DST results in patients with HIV and TB. The findings of this study are considered representative of the situation in the Dominican Republic, because a one-year national sample of patients with previously treated TB was studied. However, the challenges identified through this study are not

confined to that country; studies in China, Malawi, and Nepal revealed similar problems with the management of sputum samples for culturing and DST, with losses to follow-up at each stage and delays between registration and start of MDR-TB treatment (7–9).

These study results have several implications for the TB program and overall public health policy. First, careful monitoring of implementation of the NTP requirement that all previously treated TB patients submit sputum samples for culture and DST before the start of treatment is required. Failure to make diligent efforts to identify MDR-TB cases among this group of patients, particularly those who have failed treatment, is poor public health practice because it has serious health implications for the individual patient and leads to ongoing transmission of MDR-TB in the community. Second, a means of carrying out cultures and DST at one central reference laboratory in the country should be identified. In addition, laboratory capacity should be strengthened and the use

of rapid DST methods such as Xpert MTB/RIF (Cepheid Inc., Sunnyvale, California, United States), decentralized at peripheral centers, should be considered. Third, culture and DST results need to be made available in the health centers more quickly so that appropriate treatment for MDR-TB patients can be initiated sooner. An online system that systematically links data from different health facilities and laboratories is currently being implemented by the NTP and may improve communication and facilitate monitoring.

Limitations

The limitations of this study relate to its operational nature. All information that was analyzed was collected routinely, so some data were missing, especially those related to dates and times of the various steps in the diagnostic process. In addition, the study design did not allow for exploring the reasons behind any non-adherence to national guidelines.

Conclusions

This study shows that under the current system of culturing and DST, the majority of the patients with previously treated TB never obtain a DST result, and others obtain DST results after a considerable delay. The lack of or delay in receiving DST results skews the numbers for MDR-TB cases that are used by the TB program and hinders the timely initiation of MDR-TB treatment.

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RESUMEN

Cultivo y antibiograma en pacientes previamente tratados por tuberculosis en la República Dominicana en el 2014

Palabras clave

La tuberculosis multirresistente (MR) constituye un importante problema de salud pública que amenaza el progreso hacia un control eficaz de la tuberculosis en el mundo. Los pacientes con antecedentes de tratamiento antituberculoso presentan un mayor riesgo de padecer tuberculosis MR. En la presente investigación operativa se describe el rendimiento diagnóstico del cultivo y el antibiograma (prueba de sensibilidad a drogas) en los pacientes registrados como casos de tuberculosis previamente tratados en la República Dominicana en el 2014, mediante un estudio retrospectivo de cohortes, a partir de los datos recolectados rutinariamente por el programa. En el sistema actual, el cultivo y el antibiograma no se practican en la mayoría de los pacientes previamente tratados y, cuando se realizan, el retraso en la obtención de los resultados suele ser considerable. La falta de antibiograma y la demora en la recepción de sus resultados ocasionan una subestimación del número de casos de tuberculosis MR y obstaculizan el inicio oportuno del tratamiento de este tipo de tuberculosis.

Tuberculosis; tuberculosis resistente a múltiples medicamentos; técnicas de cultivo; pruebas de sensibilidad microbiana; estudios de cohortes; investigación operativa; República Dominicana.