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A Systematic Review of Reported Cost for Smear and Culture Tests during Multidrug-Resistant Tuberculosis Treatment

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Abstract

Background: In 2011, World Health Organization revised its recommendation for microbiological monitoring during treatment for multidrug-resistant tuberculosis (MDR-TB) by increasing the frequency of culture examination from quarterly to monthly after culture conversion. Implementing the recommendation requires substantial additional investment in laboratory infrastructure. The objective of this review is to provide cost evidence that is needed for national TB programs to budget for optimal monitoring strategies.

Methods and Findings: We conducted the first systematic literature review on unit cost estimates of three monitoring strategies: 1) smear only; 2) culture only; 3) combined smear and culture. 26 peer-reviewed studies were selected by searching 10 databases in English and Chinese for literature published between 1995 and 2012. Cost estimates were converted into 2010 constant USD and international dollars. We assessed the quality of the estimates using a matrix with five essential elements and provided a cost projection for the combined smear and culture tests where the data were available. The 26 studies reported the cost estimates in 16 predominantly high- or middle-income countries from 1993 to 2009. The estimated unit cost for smear, culture, and combined tests ranges from \$0.26 to \$10.50, \$1.63 to \$62.01, and \$26.73 to \$39.57, respectively. The ratio of culture to smear costs varies from 1.35 to 11.98. The wide range of estimates is likely attributable to using different laboratory methods in different regions and years and differing practices in collecting and reporting cost data. Most studies did not report information critical for generalizing their conclusions.

Conclusion: The paucity and low quality of unit cost estimates for TB monitoring in resource-poor settings impose technical challenges in predicting the resources needed for strengthening microbiological monitoring. To improve the validity and comparability of the cost data, we strongly advocate the data collection, estimation, and reporting follow protocols proposed by WHO.

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Introduction

Management of multidrug-resistant tuberculosis (MDR-TB) requires extensive monitoring of patients using bacteriologic testing. This is necessary to evaluate interim response to treatment; determine if patient isolation, regimen change, or adjunct therapy is required; and to classify patient treatment outcomes. In order to optimize the ability to detect non-response to treatment, recent changes to World Health Organization (WHO) Guidelines for the Programmatic Management of Drug-Resistant TB increased the frequency of sputum culture monitoring from quarterly to monthly after sputum culture conversion [1]. This recommendation was the result of a systematic analysis, which observed increased delays in detection of treatment failure with bi-monthly or quarterly culture screening, and with exclusive reliance on smear [1]. The available evidence, which was based on observational data and modeling, is considered to be of low quality [2], implying that new evidence would be very likely to change the recommendation.

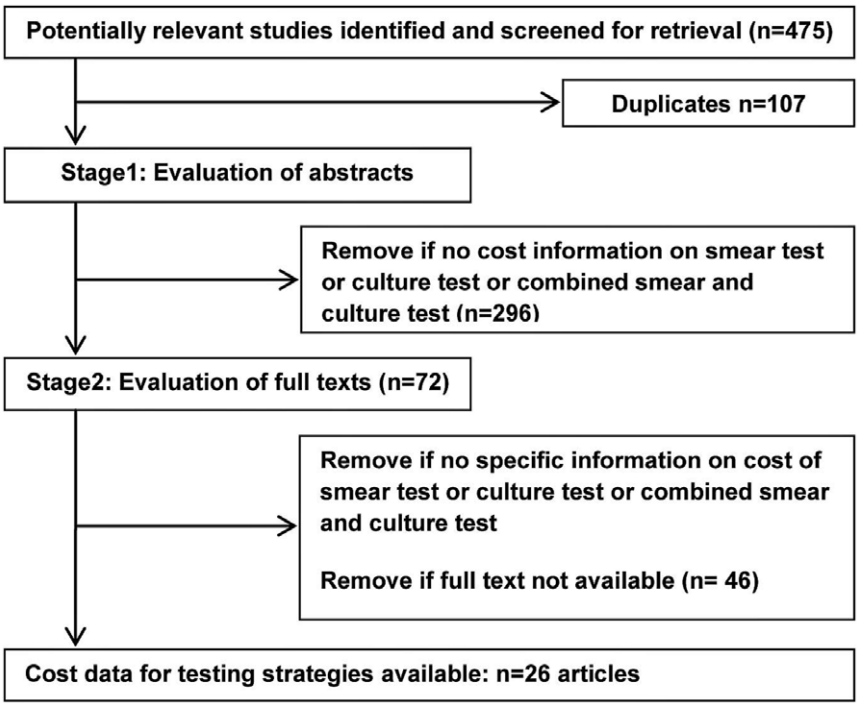

Figure 1. Study selection procedure for peer-reviewed literature from 1995–2012.
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One important consideration of implementing the recommendation is the increased cost required to assure monthly culture, in addition to smear. This will require substantial additional investment in laboratory infrastructure since current capacity of conventional laboratory is insufficient in many low-resource settings. In 2010, eight of the 22 high-burden countries (HBCs) that account for 80% of global TB cases did not meet the target of one microscopy center per 100,000 people. Among the 36 countries with the highest burden of TB and MDR-TB in the world, 20 had less than the recommended capacity of one laboratory to perform culture examination per 5 million people [3]. In order for national TB programs to budget for implementation of optimal monitoring, or to make decisions about the implementing alternative monitoring strategies, information on costs of these strategies is essential. Although new molecular tests have been validated and approved by WHO for diagnostic purposes [4], to date, these tests have no role in monitoring treatment. Consequently, this study focuses exclusively on sputum smear microscopy and sputum culture for tuberculosis.

The purpose of this study is to provide cost estimates for the different MDR-TB monitoring strategies recommended by WHO. We conducted a systematic literature review of the published cost estimates for three strategies to monitor bacteriologic response of patients on MDR-TB treatment: 1) smear only; 2) culture only; 3) combined smear and culture. Our objectives are to (1) provide a comprehensive list of published cost estimates for the three testing strategies, (2) assess the quality and limitation of the published cost estimates, (3) project the cost of combined testing when data are available, and (4) compare costs across monitoring schedules and methods when data are available.

Methods

Literature Search Strategy

We searched the literature published in peer-reviewed journals from 1995 to 2012 in both English and Chinese through 10 databases: Pubmed, Embase, Web of Knowledge, Health Economic Evaluation and Database (HEED), Econlit, National Health Service Economic Evaluation Database (NHSEED), Cost-Effectiveness Analysis Registry (CEA), Research Papers in Economics (RePEc), European Network of Health Economic Evaluation Database (EURONHEED), China National Knowledge Infrastructure (CNKI), Google Scholar and WHO. We also searched grey literature from System for Information on Grey Literature in Europe (OpenSIGLE), Healthcare Management Information Consortium (HMIC) database, National Technical Information Service (NTIS), and Biological Abstracts (BIOSIS).

We refined the search strategy in consultation with experts from Harvard Countway Library of Medicine and used a combination of three parts of keywords (e.g. "costs/economics/expenditure/price", "Tuberculosis," and "smear/culture/diagnosis/laboratory") when searching through databases. The detailed key words used in the search can be found in Table S1. The search was conducted between 23 March and 25 April, 2012. Citations were collected and managed electronically using EndNote X5. A total of 475 citations were selected in the search. 107 duplicates were automatically identified by EndNote and removed. This left a total of 368 studies, which were screened in two phases (Figure 1). First, we excluded 296 articles that did not contain cost information on MDR-TB diagnosis strategies in their abstracts. In the second phase, the full texts of the 72 remaining articles were evaluated and 46 were excluded because they do not have specific cost information for testing strategies. Our final study includes 26 articles [5–30]. One study [26] reported unit cost estimates for three countries and we listed the estimates separately in results. No protocol exists for systematic review of this topic.

Quality Assessment

We constructed a matrix to assess the quality of collected cost estimates. In order for the cost estimates to be useful and comparable, we sought at least the following information from

Table 1. Summary of published studies by income group, as per World Bank classification [5–30].

Country	Author (publication year)	Testing type[a]	Data collection year	Level of estimates	Data sources	Included items in cost estimation
High-income countries						
Canada	Menzies. et al. (2006) [5]	S; C: solid & liquid media, liquid media only; SC: smear & liquid media only	2005–2006	National	Interim Federal Health Fee Schedule	Labor, equipment, supplies, and overhead
Estonia	Floyd et al. (2012) [6]	S; C	2001–2002	National	Sources of data included expenditure records, interviews with staff and patients, project records and databases, clinical records, the social insurance system	Not specified
Finland	Rajalahti et al. (2004) [7]	SC	2000	Local(Pirkanmaa and Varsinais-Suomi)	Pirkanmaa Hospital District	Not specified
Italy	Migliori et al. (1999) [8]	S; C	1995	National	Nationwide, 41 TB-reporting units self-selected into participating in the study	Buildings, diagnostic facilities, salaries, overhead, and direct examination costs
UK	Dinnes et al. (2007) [9]	C: standard culture, rapid culture, culture and first-line sensitivity on solid media	Not available	National	Price list from the Public Health Laboratory Mycobacterium Reference Unit	Not specified
USA	Heymann et al. (1997) [10]	C: combined radiometric broth and solid medium; SC: smear and conventional/ radiometric culture	Not available	National	National Jewish Center for Immunology and Respiratory Medicine in Denver and Massachusetts state public health laboratory	Not specified
	Wurtz et al. (1998) [11]	SC	1993	Local(Chicago)	A public hospital 's 1993 Medicare Schedule C charges and a state university hospital located in close proximity to the study hospital	Not specified
	GA for TB Drug Development (2001) [12]	C	2000	National	Medical Resource Based Relative Value Scale Reimbursement Schedule	Not specified
Upper-middle-income countries						
Brazil	Dowdy et al. (2008) [13]	C: LJ, MGIT	2006–2008	Local(Rio de Janeiro)	29 municipal health clinics and hospitals randomly selected	Culture tubes and media, decontamination reagents, cryovials for pellet storage, lab supplies and equipment and personnel; fixed costs: transportation, and automated MGIT 960 reader
	Scherer et al. (2009) [14]	S: ZN; C: LJ; SC: ZN+LJ	2003–2004	Local(Porto Alegre City)	Public Reference Laboratory, Centro de Desenvolvimento Científico e Tecnológico and Fundação Estadual de Produção e Pesquisa em Saúde	Laboratory running costs and patient costs (including costs for travel, food and income loss)
China	Chen et al. (2011) [15]	S; C	Not available	National	Cited from the websites on health expenditure	Not specified
Thailand	Kamolratanakul et al. (2002) [16]	S; C	1996–1997	National	Four referral centers were randomly selected from four geographical regions (Eastern, Southern, Northern and Northeastern)	Overhead costs and materials costs
	Sohn et al. (2008) [17]	S: ZN, FM	2007–2008	National	National Tuberculosis Reference Laboratory	Capital assets(e.g., building space, equipment, staff), laboratory consumables and chemicals, and recurrent costs
Peru	Suárez et al. (2002) [18]	S: ZN; C: LJ	1997–1999	National	MDR-TB unit in Lima	Not specified
Russia	Floyd et al. (2012) [6]	S; C	2001–2002	Local(Tomsk Oblast)	Sources of data included expenditure records, interviews with staff and patients, project records and databases, clinical records	Not specified

Table 1. Cont.

Country	Author (publication year)	Testing type[a]	Data collection year	Level of estimates	Data sources	Included items in cost estimation
	WHO Policy Brief (2005) [19]	S; C	2003	Local(Vladimir Oblast)	Clinical diagnostic laboratory of general health care; level II clinical diagnostic laboratory within primary health care services	Not specified
	Balabanova et al. (2009) [20]	C: MGIT, LJ	2006–2008	Local(Samara Oblast)	Central TB laboratory of Samara Region	Decontamination(including specimen transportation costs), prep LJ, overhead, building, equipment, staff, medical supplies
South Africa	Sinanovic et al. (2003) [21]	S; C	1998–1999	Local(Guguletu and Nyanga)	Cape Town City Council, the South African Institute for Medical Research, the TB Care Association, local equipment suppliers, car dealers, staff interviews and patient survey.	Not specified
	Albert (2004) [22]	S: ZN; C: BACTEC 460TB	2003	Local(Cape Town)	National Health Laboratory Service(NHLS), Cape Town	Not specified
	Hausler et al. (2006) [23]	S; C	Not available	Local(Cape Town)	Three public primary health care facilities	Not specified
	Chihota et al. (2010) [24]	C: LJ, MGIT, MGIT+LJ	2006–2007	Local(Johannesburg)	National Health Laboratory Services regional TB laboratory in Johannesburg	Capital costs (buildings, furniture, medical equipment, non-medical equipment), recurrent costs (staff costs, medical supplies, non-medical supplies, overhead)
	Whitelaw et al. (2011) [25]	S: LED, ZN	2009	Local(Cape Town)	Two primary care clinics in Cape Town and NHLS	Direct examination costs, capital costs (laboratory space and equipment), overhead costs (staff costs and time, and space and infrastructure utilized to each test)
	Vassall et al. (2011) [26]	S: LED; C: MGIT	Not available	National	Urban or periurban primary care health centers in South Africa	Building, overhead, staff, equipment and consumables, quality control and maintenance, and calibration inputs
Lower-middle income countries						
India	Muniyandi et al. (2006) [27]	S	2002	Local(Tamil Nadu)	All the government health facilities, including subcenters situated in a TB unit of a rural district of Tamil Nadu	Staff salary, costs incurred for reagents, drugs, maintenance, stationery and fuel etc.
	Vassall et al. (2011) [26]	S: LED; C: LJ	Not available	National	Urban or periurban primary care health centers in India	Building, overhead, staff, equipment and consumables, quality control and maintenance, and calibration inputs
Zambia	Mueller et al. (2008) [28]	C: HLJ, CLJ, MMGIT, AMGIT	2006	National	Zambia National TB Reference Laboratory	Overhead costs, running costs(rent of the building, utilities, vehicle running, staff management), culture-specific costs(equipment, consumables, staff costs)
Low income countries						
Kenya	Kivihya-Ndugga et al. (2003) [29]	S: FM, ZN	2000–2001	Local(Nairobi)	Nairobi City Council Chest Clinic	Labour costs, investment costs and running costs
Uganda	Okello et al. (2003) [30]	S	1995–1999	Local(Kiboga)	Kiboga district hospital and two Masindi district hospitals	Not specified

Table 1. Cont.

Country	Author (publication year)	Testing type[a]	Data collection year	Level of estimates	Data sources	Included items in cost estimation
	Vassall et al. (2011) [26]	S: LED; C: LJ, MGIT	Not available	National	A central hospital in Uganda	Building, overhead, staff, equipment and consumables, quality control and maintenance, and calibration inputs

S: smear test alone, C: culture test alone, SC: combined smear and culture test; ZN: Ziehl-Neelsen, FM: fluorescence microscopy, LED: *light-emitting diode*, LJ: Lowenstein-Jensen, MGIT: Mycobacteria Growth Indicator Tube, HLJ: Homemade Löwenstein-Jensen, CLJ: Commercially Löwenstein-Jensen, MMGIT: Manually Mycobacteria Growth Indicator Tube, AMGIT: Automated Mycobacteria Growth Indicator Tube, FIND: Foundation of innovative New Diagnostics, BD: Becton Dickinson.

[a]For studies with detailed information on diagnostic tests, we listed their specific type; otherwise, it's not available.

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each study: (1) the year of cost data being collected; (2) the level of estimates (national or regional), (3) the specific diagnostic methods and materials used; (4) the sources of data; and (5) the components included in cost estimation. We treated each category as binary and assigned values "0" or "1". For instance, if a study reported the year of data being collected, "Data collection year" takes value of 1, 0 otherwise. In tables and figures, we used publication year as a proxy if the studies did not report information data collection year. If the estimate is national, "National estimate" takes value 1, 0 otherwise. If test methods (such as light-emitting diode [LED], Ziehl-Neelsen [ZN], etc) were reported in the paper, "Specification of test methods" takes value of 1, 0 otherwise. If cost data were directly collected from health facilities, "direct data sources" takes value of 1. If cost data were obtained from published price list, "direct data sources" takes value of 0. If the cost components included in estimating the unit cost of tests was reported by the paper, "specification of cost items" takes value of 1, 0 otherwise. We then summed the scores across the five categories for each estimate with 0 representing the weakest quality and 5 the best. When information was available, we also listed cost components in Table 1 so the readers could identify which cost components were included in cost estimation.

Projection of Unit Costs of Combined Smear and Culture Tests

When smear and culture costs were reported separately in the same study and the cost for combined tests was not available, we imputed the unit cost of the combined tests by adding the unit costs of the two testing strategies. The imputed value may serve as an upper bound estimate for the combined test. Total costs for combined tests may be lower than the imputed value due to a single set of procedures being performed for both tests (e.g., for sputum collection, transport, and processing). All cost estimates were converted into 2010 constant USD using an exchange rate and GDP deflator from International Monetary Fund [31]. To adequately represent the distinction of costs across different countries, the international dollar is preferable since it adjusts the distortion effect of non-traded goods compared to single US$ value [32]. When detailed cost information was available to identify the cost of traded and non-traded goods, we also converted the cost estimates to 2010 international dollars using purchasing power parity [33].

Analyzing Existing Data

When papers provided unit cost estimates for both the culture and smear tests, we calculated the cost ratio of culture to smear. For studies with the values of the cost components, we first classified the components into two categories, traded and non-traded goods, based on the definition from the WHO guideline for cost-effectiveness analysis. Traded goods (e.g. equipment, supplies and pharmaceuticals) are available on the international market and available to all countries at an international market price. Personnel, utilities, buildings and domestic transport are treated as non-traded goods [32]. We then calculated the share of the two types of goods in unit cost.

Results

Assessing Existing Studies

26 studies published between 1995 and 2012 reported cost estimates in 16 countries (Table 1). Of these, 22 studies were conducted in high-income or upper-middle income countries. Five studies reported unit cost estimates in four low and lower-middle income countries (India, Zambia, Kenya and Uganda) [26–30]. 17

Table 2. Quality assessment of the studies (1 = yes; 0 = no).

Author	1) Data collection year	2) National estimate	3) Specification of test type	4) Direct data source	5) Specification of cost items	Sum
Mueller et al. [28]	1	1	1	1	1	5
Sohn et al. [17]	1	1	1	1	1	5
Dowdy et al. [13]	1	0	1	1	1	4
Kamolratanakul et al. [16]	1	1	0	1	1	4
Balabanova et al. [20]	1	0	1	1	1	4
Menzies. et al. [5]	1	1	1	0	1	4
Suárez et al. [18]	1	1	1	1	0	4
Vassall et al. (South Africa) [26]	0	1	1	1	1	4
Vassall et al. (India) [26]	0	1	1	1	1	4
Vassall et al. (Uganda) [26]	0	1	1	1	1	4
Whitelaw et al. [25]	1	0	1	1	1	4
Scherer et al. [14]	1	0	1	1	1	4
Chihota et al. [24]	1	0	1	1	1	4
Kivihya-Ndugga et al. [29]	1	0	1	1	1	4
Migliori et al. [8]	1	1	0	1	1	4
Albert [22]	1	0	1	1	0	3
Heymann et al. [10]	0	1	1	1	0	3
Muniyandi et al. [27]	1	0	0	1	1	3
Dinnes et al. [9]	0	1	1	0	0	2
WHO Policy Brief [19]	1	0	0	1	0	2
GA for TB Drug Development [12]	1	1	0	0	0	2
Rajalahti et al. [7]	1	0	0	1	0	2
Wurtz et al. [11]	1	0	0	1	0	2
Okello et al. [30]	1	0	0	1	0	2
Floyd et al. (Estonia) [6]	1	1	0	0	0	2
Hausler et al. [23]	0	0	0	1	0	1
Chen et al. [15]	0	1	0	0	0	1
Sinanovic et al. [21]	1	0	0	0	0	1
Floyd et al. (Tomsk Oblast) [6]	1	0	0	0	0	1

Notes: We treat each category as binary and assign values "0" or "1". 1) "data collection year": whether or not the data collection year was provided in the study. If yes, "data collection year" = 1, 0 otherwise; 2) "national estimate": whether or not the cost was estimated at national level. If yes, "national estimate" = 1, 0 otherwise; 3) "specification of test type": whether or not the test type was provided in the study, e.g. ZN/FM, MGIT/LJ. If yes, "specification of test type" = 1, 0 otherwise; 4) "direct data source": whether or not the cost was directly collected from health facilities (e.g. hospital, clinic, laboratory etc.). If yes, "direct data" = 1, 0 otherwise; 5) "specification of cost items": whether or not the study described the components included in cost estimation. If yes, "specification of cost items" = 1, 0 otherwise. All the studies are ranked by the summation of five scores from highest to lowest.

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of the selected studies reported unit cost estimates for smear test alone. 19 studies reported unit cost estimates for culture test alone. Five studies reported cost estimates for combined smear and culture test in four middle-upper or high income countries.

The quality of reported data varied considerably among 26 studies. Five of them did not report which year the cost data were collected, 12 of them reported national estimates, 14 of them specified the methods used for test, 20 of them obtained data directly from health facilities, and 13 of them provided the cost components that were used to estimate of unit costs (Table 2). Components of cost estimates were not reported in a standardized way. Some studies only included costs for materials and overhead, while others included costs on building, equipment, or even patients' spending on travel, food and income loss due to sick leave. Using our quality scale, two studies scored 5 points, 11 studies scored 4, and 13 studies scored 3 or below (Table 2).

Cost Estimates

Estimated costs of smear microscopy, presented in Figure 2 in constant 2010 USD, vary across countries from $0.26 in Tamil Nadu, India (2002) [27] to $10.50 in Thailand (1996–1997) [16]. Unsurprisingly, unit costs for sputum smear differed in a given country and year when different microscopy methods were used. For example, in Cape Town, South Africa, unit costs for smear with light-emitting diode (LED) microscopy and Ziehl-Neelsen (ZN) in 2009 were $1.64 and $2.11 respectively [25].

The estimated unit cost for mycobacterial culture is between $1.63 in Vladimir Oblast, Russia (2003) [19] and $62.01 in the United Kingdom (2007) [9] (Figure 3). Estimates in the same country and same year unsurprisingly vary when different media were used and follow-up tests were required. For example, in Brazil, unit costs of culture vary from $18.48 to $35.14 during the same period (2006–2008). The former value is the cost per

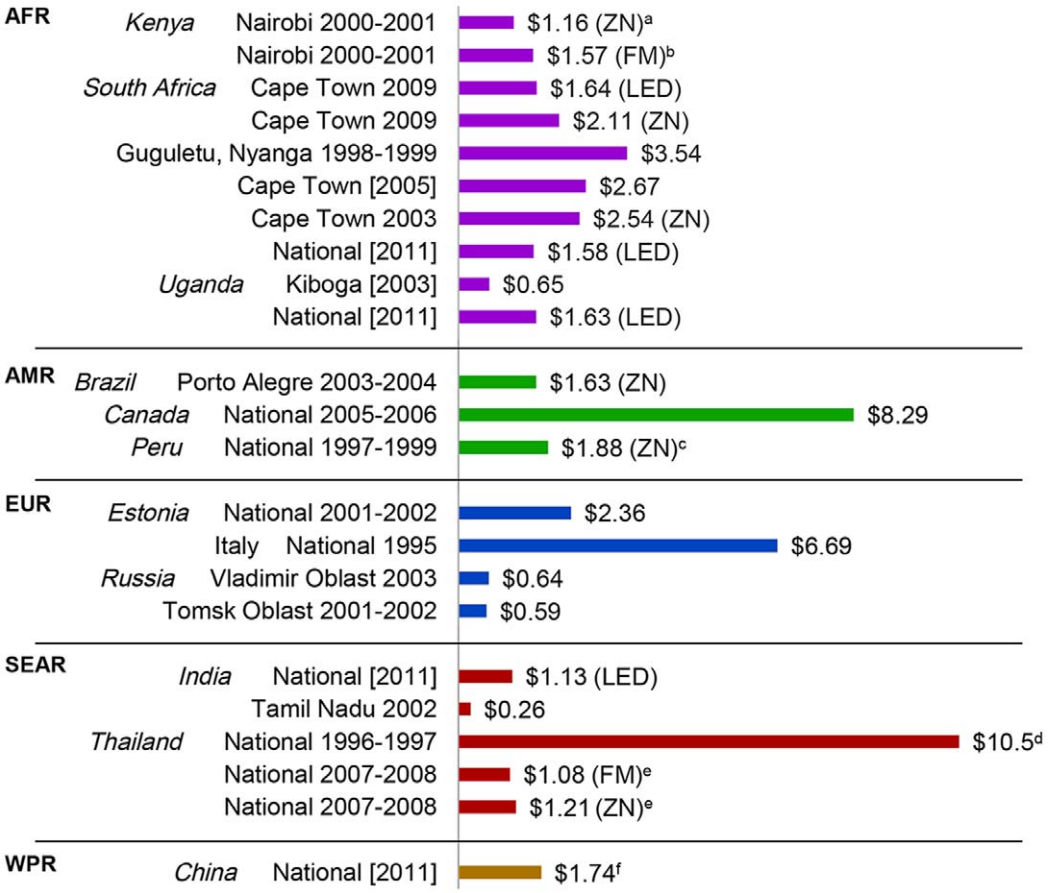

Figure 2. Unit cost in 2010 USD for smear test alone. (1) Cost data were sorted by WHO regions: African Region (AFR), Region of the Americas (AMR), Eastern Mediterranean Region (EMR), European Region (EUR), South-East Asia Region (SEAR) and Western Pacific Region (WPR). (2) For studies with available information on test methods, we labeled them at the end of each bar. (3) [] indicates publication year when data collection year is not available. (4) ZN: Ziehl-Neelsen; FM: fluorescence microscopy; LED: light-emitting diode. [a]$1.16 is the average laboratory costs on 1000 subjects and three specimens. [b]$1.57 is the average laboratory costs on 1000 subjects and three specimens. [c]$1.88 is the total cost $26.27 divided by 14 sputum smears. [d]Sum of the overhead cost ($10.4) and the material cost ($0.1). [e]For the examination of three sputum specimens, the cost per patient evaluated is $3.24 for FM and $3.59 for ZN. [f]The unit cost is the average over six regional estimates. For detailed information of the six regional estimates, see Table S2.
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negative culture using solid media for eight patients per week; the latter value is the cost per positive culture using MGIT for eight patients per week [13].

Limited data were available for the cost of combined testing. Results have only been reported in four countries and the values ranged from $26.73 in Canada (2005–2006) [5] to $39.57 in USA (1997) [10] (Figure 4). The imputed unit cost of combined tests ranged from $2.27 in Vladimir Oblast, Russia (2003) [19] to $48.23 in Thailand (1996–1997) [16]. The majority of imputed estimates lies between $10 and $30.

The distribution of cost estimates for sputum smear is right-skewed, with a median of $1.67 (Figure 5). The cutoff points for the 25th and 75th percentiles are $1.21 and $2.54 respectively. The median of cost estimates for culture tests is $18.48 with $11.08 as the 25th percentile and $33.33 as the 75th percentile. For combined testing, the median cost is $16.82, the 25th percentile is $10.62 and the 75th percentile is $26.81.

12 studies reported cost estimates for both smear and culture tests performed separately. The ratio of estimated costs for culture to smear varies from 1.35 to 11.98 (Table 3); most are larger than 1.6, the ratio that has been used previously in the context of cost and cost-effectiveness studies for drug-susceptible TB [34]. The median ratio is 3.75. Notably, the ratio is available for only one low-income (Uganda, 2011) [26] and one lower-middle income country (India, 2011) [26]. Studies conducted between 1998 and 2011 in South Africa reported ratios from 2 to 11.98 [21,22,23,26].

Eight studies broke down estimated costs by traded inputs (i.e. supplies and equipment) and non-traded inputs (such as labor). A large variation is observed in the percentage of costs attributed to traded inputs (Table 4): for smear tests, it ranges from 0.95% [16] to 70.87% [17], and for culture test, it ranges from 21.16% [13] to 75.39% [20]. Unit cost estimates did not change significantly in 2010 international dollars (I$): I$ 1.34–19.24 for smear testing and I$ 15.32–38.84 for culture testing.

Discussion

The existing unit cost estimates for smear, culture, and combined smear and culture tests are very limited, especially in low or lower-middle income countries. Nevertheless, a wide range of published unit cost estimates was observed. For smear alone, the estimated unit cost is between $0.26 and $10.5. For culture alone, the estimated unit cost is between $1.63 and $62.01. For combined

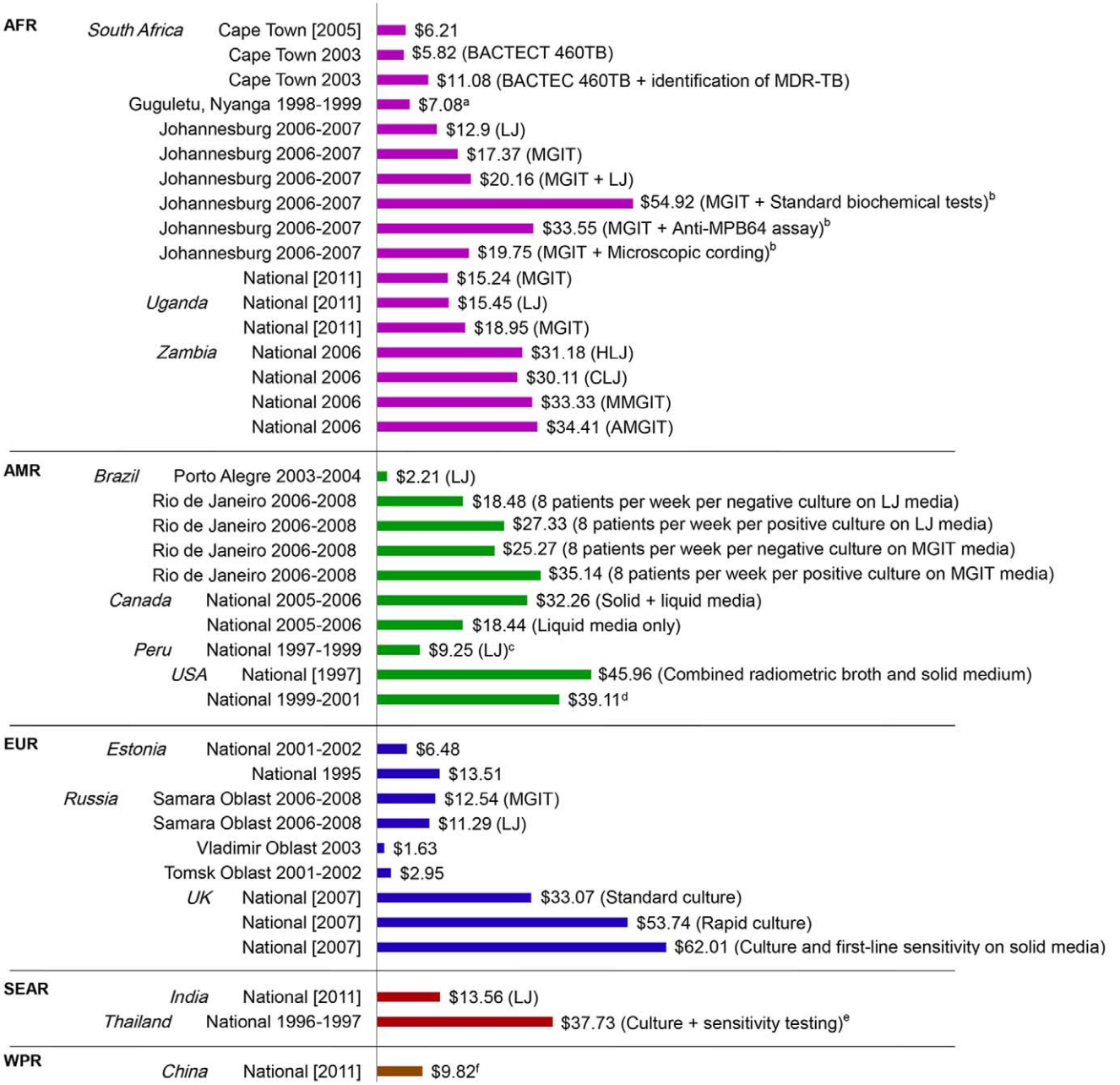

Figure 3. Unit cost in 2010 USD for culture test alone. (1) Cost data were sorted by WHO regions: African Region (AFR), Region of the Americas (AMR), Eastern Mediterranean Region (EMR), European Region (EUR), South-East Asia Region (SEAR) and Western Pacific Region (WPR). (2) For studies with available details on test methods, we labeled them at the end of each bar. (3) "[]" indicates publication year when data collection year is not available. (4) LJ: Löwenstein-Jensen; MGIT: Mycobacteria Growth Indicator Tube; HLJ: Homemade Löwenstein-Jensen; CLJ: Commercially Löwenstein-Jensen; MMGIT: Manually Mycobacteria Growth Indicator Tube; AMGIT: Automated Mycobacteria Growth Indicator Tube; FIND: Foundation of innovative New Diagnostics; BD: Becton Dickinson. [a]$7.08 is the average costs between negative and positive tests. [b]The paper indicates cost for organism identification per positive culture on MGIT was $37.55 for using standard biochemical tests, $16.18 for anti-MPB64 assay and $2.38 for cording; we added each of them to the cost per MGIT ($17.37) for calculating the cost for positive culture. [c]$9.25 is the total cost of $85.07 divided by 9.2 sputum cultures. [d]Sum of the cost for sputum collection ($19.12) and the cost for bacterial culture ($19.99). [e]Sum of the overhead cost ($10.4) and the material cost ($27.33). [f]The unit cost is the average over six regional estimates. For detailed information of the six regional estimates, see Table S2.
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smear and culture testing, the estimated unit cost is between $2.27 and $48.23. Adjustment for purchasing power parity does not fully explain the wide range of unit cost estimates we observed.

The wide variability of unit costs is partly due to using different materials and methods in testing, or conducting the study in different years or regions, partly due to non-standardized practice in unit cost defining, data collecting, and reporting. For example, for those with cost components available, the reported components vary greatly across studies, from only including material and overhead cost to covering the costs of building, equipment, and even the spending of patients. Cost data were obtained from different sources, including citing figures from a price list,

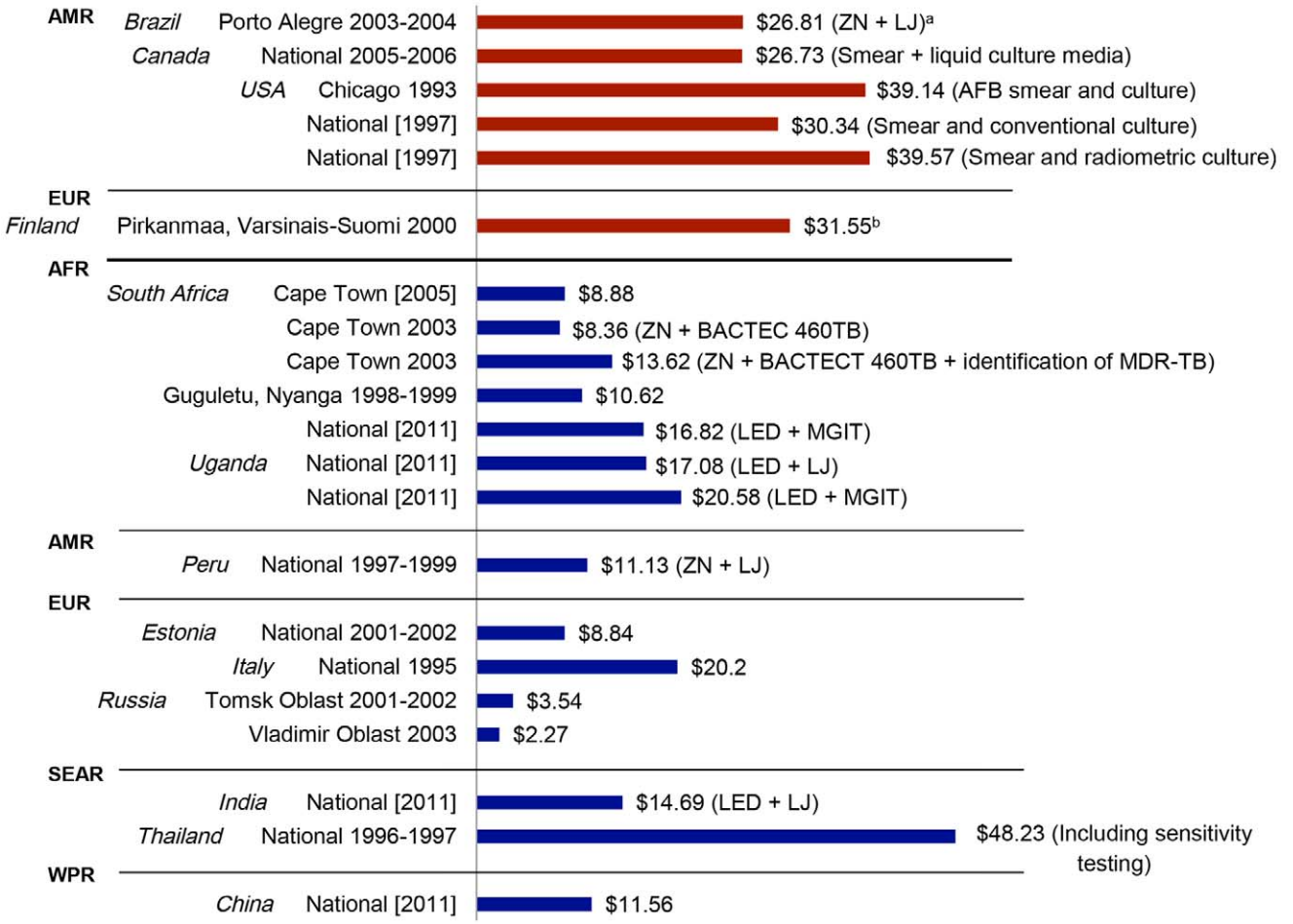

Figure 4. Unit cost in 2010 USD for combined smear and culture test. (1) Directly obtained cost data are in red; imputed cost data are in blue. (2) Cost data were sorted by WHO regions: African Region (AFR), Region of the Americas (AMR), Eastern Mediterranean Region (EMR), European Region (EUR), South-East Asia Region (SEAR) and Western Pacific Region (WPR). (3) For studies with available details on test methods, we labeled them at the end of each bar. (4) [] indicates publication year when data collection year is not available. (5) AFB: acid-fast bacillus; LJ: Löwenstein-Jensen; MGIT: Mycobacteria Growth Indicator Tube. [a]Laboratory running cost is $14.34. Estimated costs incurred by patients are $12.47 (assuming that for taking an examination, a patient has to miss one-day work, take two-way transportation and have one meal outside). [b]$31.55 is the total cost of $94.66 divided by three combined smear and culture tests.
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collecting data from a single health facility in a specific area of a country, and aggregating data from all regions in a country. Non-standardized cost estimates make it very difficult for cross-setting comparison and making meaningful inference.

The quality of the estimates is a concern. About one fifth of the selected studies did not even report the year in which cost data were collected. Half of the selected studies did not specify test methods (Migliori, or Kamolratanakul, or Qunfei, for example) used in reported smear or culture tests. Almost half of the studies did not report what components were included in cost estimation. Since we know these factors have a significant bearing on cost estimates, the lack of standardization–and low quality overall–in cost data collecting and reporting present major challenges for improving our knowledge of unit costs of various MDR-TB monitoring strategies.

The calculated unit cost ratio for culture tests to smear tests from existing studies is greater than the 1.6, a number which was previously generated from cost data collected from a government laboratory in South Africa [34]. The extent to which this ratio varies between countries will likely depend on the relative weight of non-traded inputs in the cost of each test. The cost of non-traded inputs such a labour is more sensitive than the cost of traded inputs to the income level of a given country. Therefore, if the share of non-traded inputs in total cost is smaller for cultures than it is for smears, we would expect the ratio to be higher in the lowest income countries and lower in the highest income countries.

The new recommended strategy of monthly–rather than minimum of quarterly–culture test after culture conversion, would cost more. If smear and culture were done quarterly, only 6 combined tests would be required (in addition to 14 monthly smears). According to the current recommendations of monthly smear and culture, 20 combined tests would be required. Smear and culture both have limited ability to predict poor treatment response [1,35]. Culture, however, is much more accurate than smear in detecting the presence of viable mycobacteria. Smear microscopy sensitivity estimates range from 40 to 76%, with lower sensitivity in children and HIV-coinfected patients [36–40]. As the Guidelines note, "high value was placed on outcomes such as preventing death, decreasing the transmission of MDR-TB that could result from its delayed diagnosis, and avoiding increased use of resources. [1]" Consequently, increased costs associated with more frequent culture test may be justified because of the

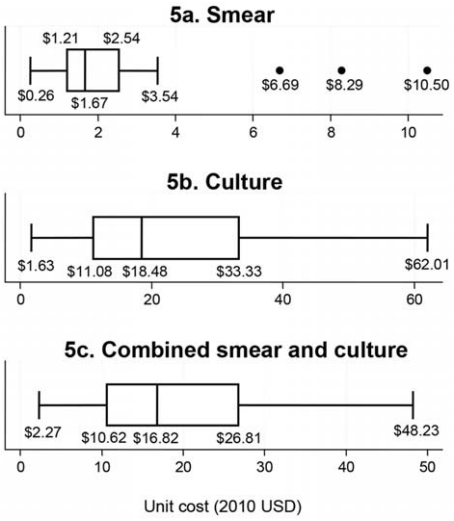

Figure 5. Summary of estimates of the three types of tests. (1) In each boxplot, dots represent outliers which are beyond the interval of (Q1–1.5*IQR, Q3+1.5*IQR): Q1 is the 25th percentile, Q3 is the 75th percentile, IQR is the interquartile range (75%–25%). (2) The five values listed beside each boxplot represent upper adjacent value (maximum value after excluding outliers), 75th percentile, median (50%), 25th percentile, and lower adjacent value (minimum value after excluding outliers), respectively. For instance, in Plot 5a, for estimates of smear test alone, $3.54 (upper adjacent value) is the maximum value excluding three outliers. $2.54 is the value at the 75th percentile. $1.67 is the value of median. $1.21 is the value at the 25th percentile. $0.26 (lower adjacent value) is the minimum value excluding outliers. (3) For Plot 5c, the estimates of combined test include the imputed values.
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importance of early detection of risk for these negative outcomes and the possibility of implementing interventions to avert them. While it remains important for patients on treatment for MDR-TB to have access to good quality culture for their proper monitoring, our findings highlight a high cost difference between culture and smear testing. It is noteworthy that one factor contributing to this difference in low-resource setting may be the relatively infrequent use of culture compared to smear at the time of data collection or publication. These prices may be expected to decline once the initial outlay associated with expanding culture laboratories has been discounted.

Even within the same monitoring method, certain methodological differences may result in cost differences, but also in sensitivity and timing. For example, culture performed in liquid medium, using the MGIT system is known to increase the detection of viable mycobacteria over culture performed in solid LJ or Ogawa medium, while decreasing from 8 weeks to 6 weeks the time required to confirm a culture as negative. Although there was additional cost associated with MGIT in at least 3 studies that compared cost of culture in liquid and solid medium ([13], [24], and [28]), these cost differences may be justified since they accelerate the time to detection and intervention and increase the sensitivity of the test. There are similar differences among the cost and sensitivity of smear microscopy methods [40].

Lastly, three studies ([13], [22], and [24]) reported variation in unit cost of culture depending on whether the result was negative or positive. This highlights another possible source of variability in estimates that was not explicitly reported in the other studies.

This study is the first systematic review of cost estimates for tests commonly used to monitor MDR-TB treatment. We reviewed the

Table 3. Ratio of unit cost for culture to smear.

Author	Site	Time period	Ratio of culture to smear	Methods on smear/culture
Scherer et al. [14]	Brazil	2003–2004	1.35	ZN (S)
Sinanovic et al. [21]	South Africa	1998–1999	2[a]	Not available
Migliori et al. [8]	Italy	1995	2.02	Not available
Menzies et al. [5]	Canada	2005–2006	2.22	Liquid media (C)
Albert [22]	South Africa	2003	2.3	ZN (S); BACTEC 460TB (C), (−)
Hausler et al. [23]	South Africa	[2005]	2.32	Not available
WHO Policy Brief [19]	Russia	2003	2.58	Not available
Floyd [6]	Estonia	2001–2002	2.75	Not available
Kamolratanakul et al. [16]	Thailand	1996–1997	3.6[b]	Not available
Menzies et al. [5]	Canada	2005–2006	3.89	Solid+liquid media (C)
Albert [22]	South Africa	2003	4.38[c]	ZN (S); BACTEC 460TB (C), (+)
Suárez et al. [18]	Peru	1997–1999	4.93	ZN (S); LJ (C)
Floyd [6]	Tomsk Oblast	2001–2002	5.2	Not available
Chen et al. [15]	China	[2011]	5.61	Not available
Vassall et al. [26]	Uganda	[2011]	9.47	LED (S); LJ (C)
Vassall et al. [26]	India	[2011]	9.67	LED (S); LJ (C)
Vassall et al. [26]	Uganda	[2011]	11.61	LED (S); MGIT (C)
Vassall et al. [26]	South Africa	[2011]	11.98	LED (S); MGIT (C)

S: smear test alone; C: culture test alone; ZN: Ziehl-Neelsen; (+): positive result; (−): negative result; TB: tuberculosis; LJ: Löwenstein-Jensen; LED: light-emitting diode; MGIT: Mycobacteria Growth Indicator Tube.
[a]The original unit cost for culture is the average cost between negative and positive tests;
[b]The original unit cost for culture includes the cost for sensitivity testing;
[c]The original unit cost for positive culture includes the cost for MDR-TB identification.
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Table 4. Tradable cost and non-tradable cost from eight studies.

Author	Site (year of study)	Test type	Tradable cost components	Non-tradable cost components	Tradable cost (%)	I$2010
Smear						
Kamolratanakul et al. [16]	Thailand (1996–1997)	Not available	Material costs: 2.04 baht	Overhead cost: 212.19 baht	0.95%	19.24
Sohn et al. [17]	Thailand (2007–2008)	ZN	Equipment: $0.08; reagents and chemicals: $0.04; consumables: $0.22	Overhead: $1.06; building space: $0.01; staff: $0.69	16.19%	2.06
		FM	Equipment: $0.08; reagents and chemicals: $0.25; consumables: $0.40	Overhead: $0.15; building space: $0.01; staff: $0.14	70.87%	1.34
Whitelaw et al. [25]	South Africa (2009)	ZN	Equipment: $0.02; reagents and chemicals: $0.33; consumables: $0.40	Overhead: $0.21; building space: $0.01; staff: $0.19	64.66%	2.44
		LED	Equipment: $0.08; reagents and chemicals: $0.04; consumables: $0.22	Overhead: $0.81; building space: $0.01; staff: $0.47	20.86%	2.21
Culture						
Kamolratanakul et al. [16]	Thailand (1996–1997)	Culture and sensitivity testing	Material cost: 557.90baht	Overhead cost: 212.19baht	72.45%	23.68
Muller et al. [28]	Zambia (2006)	HLJ	Consumables: $5.52; equipment: $4.6; consumables, equipment and furniture(included in the overheads): $2.04	Staff: $1.44; overheads(excluding consumables, equipment and furniture): $15.68	41.53%	35.51
		CLJ	Consumables: $4.62; equipment: $4.12; consumables, equipment and furniture(included in the overheads): $2.05	Staff: $1.31; overheads(excluding consumables, equipment and furniture): $15.69	38.83%	34.49
		MMGIT	Consumables: $7.14; equipment: $4.05; consumables, equipment and furniture(included in the overheads): $2.19	Staff: $1.73; overheads(excluding consumables, equipment and furniture): $16.35	42.53%	37.88
		AMGIT	Consumables: $7.13; equipment: $5.48; consumables, equipment and furniture(included in the overheads): $2.18	Staff: $1.24; overheads(excluding consumables, equipment and furniture): $16.24	45.83%	38.84
Chihota et al. [24]	South Africa (2006–2007)	MGIT	Furniture: $0.35; medical equipment: $2.03; non-medical equipment: $0.09; medical supplies: $5.02; non-medical supplies: $0.21	Buildings: $0.52; staff costs, culture: $6.53; staff costs, non culture: $1.73; overheads: $0.15	46.3%	21.48
		LJ	Furniture: $0.45; medical equipment: $0.45; non-medical equipment: $0.09; medical supplies: $2.29; non-medical supplies: $0.22	Buildings: $0.52; staff costs, culture: $6.44; staff costs, non culture: $1.82; overheads: $0.08	28.32%	16.98
		MGIT+LJ	Furniture: $0.45; medical equipment: $2.05; non-medical equipment: $0.09; medical supplies: $6.28; non-medical supplies: $0.23	Buildings: $0.52; staff costs, culture: $7.60; staff costs, non culture: $1.93; overheads: $0.15	47.15%	24.86
Dowdy et al. [13]	Brazil (2006–2008)	Solid Media (8 patients per week per negative culture)	Culture tubes and media: $0.59; decontamination reagents: $0.83; lab supplies (e.g., pipette tips, centrifuge tubes): $0.53; lab supplies (e.g., mini-pipettes, vortex machine): $1.17; lab equipment (e.g., incubator, freezer): $0.59	Transportation: $9.61; laboratory personnel: $4.21	21.16%	19.08
		Solid Media (8 patients per week per positive culture)	Culture tubes and media: $0.59; decontamination reagents: $0.83; lab supplies (e.g., pipette tips, centrifuge tubes): $0.53; lab supplies (e.g., mini-pipettes, vortex machine): $1.17; lab equipment (e.g., incubator, freezer): $0.59; Confirmation/speciation: $7.90	Transportation: $9.61; laboratory personnel: $ $4.21	45.65%	27.95

Table 4. Cont.

Author	Site (year of study)	Test type	Tradable cost components	Non-tradable cost components	Tradable cost (%)	I$2010
		MGIT (8 patient per week per negative culture)	Culture tubes and media: $3.00; decontamination reagents: $0.83; cryovials for pellet storage: $0.81; lab supplies (e.g., pipette tips, centrifuge tubes): $0.53; automated MGIT 960 reader: $4.62; lab supplies (e.g., mini-pipettes, vortex machine): $1.00; lab equipment (e.g., incubator, freezer): $0.39	Transportation: $8.57; laboratory personnel: $3.75	47.57%	25.82
		MGIT (8 patient per week per positive culture)	Culture tubes and media: $3.00; decontamination reagents: $0.83; cryovials for pellet storage: $0.81; lab supplies (e.g., pipette tips, centrifuge tubes): $0.53; automated MGIT 960 reader: $4.62; lab supplies (e.g., mini-pipettes, vortex machine): $1.00; lab equipment (e.g., incubator, freezer): $0.39; confirmation/speciation: $9.18	Transportation: $8.57; laboratory personnel: $3.75	62.3%	35.69
Balabanova et al. [20]	Russia (2006–2008)	MGIT FIND-BD	Decontamination: $2.76; equipment: $1.45; medical supplies: $4.58	Overhead: $1.94; building: 0.42; staff: $0.51	75.39%	15.32
		LJ	Decontamination: $2.76; prep LJ: $0.10; equipment: $0.24; medical supplies: $0.05	Overhead: $4.95; building: $1.07; staff: $1.34	29.97%	18.41
Combined smear and culture						
Menzies et al. [5]	Canada (2005–2006)	AFB smear & culture on liquid media only	Supplies: C$10.07	Labour costs: C$21.83	31.57%	23.88

Notes: For studies with detailed information of cost for the included test components, we separated them into two parts: tradable costs and non-tradable costs. All the costs are the original data from selected studies without any conversion and standardization. If the detailed information for overheads is not available, we treated the overheads as non-tradable cost item.

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relevant literature in Chinese. China has the highest prevalence of TB after India and the availability of Chinese cost data provides critical information for scaling up the monitoring tests for this large at-risk population. Using existing cost data, we also projected the unit cost of combined tests which could serve as useful reference to policy makers.

We propose a framework for evaluating the quality of unit cost data for TB monitoring tests. The five categories included in the quality score are crucial for determining the generalizability and validity of the cost data. They, may not, however, cover all important aspects. For instance, we only distinguished between the availability and absence of cost components, but did not consider the comprehensiveness of cost components. We assigned each category with the same weight and this may oversimplify the evaluation.

The paucity and low quality of unit cost estimates for TB monitoring in developing countries impose technical challenges in predicting the resource needed for strengthening microbiologic monitoring. As new molecular tests are being rapidly introduced globally to diagnose patients with presumptive TB and drug-resistant TB in one step, evaluation of the costs associated with the change in diagnostic practices – which was not the object of this paper - will be necessary. High quality cost data is especially important for the regions with high incidence of tuberculosis and MDR-TB, where scarce resources must be allocated efficiently. We strongly advocate that more data are collected from these regions, and that cost data collection, estimation, and reporting should follow the protocols proposed by the WHO [34] to improve the validity and comparability.

Supporting Information

Table S1 Databases and search terms of the search strategy.
(DOCX)

Table S2 Cost for smear/culture in different regions in China (2010 USD).
(DOCX)

Author Contributions

Conceived and designed the experiments: CL CDM. Senior authors: CL CDM. Analyzed the data: CL QL. Wrote the paper: QL CL AS CF DF CDM.

References

1. WHO website. Available: http://whqlibdoc.who.int/publications/2011/9789241501583_eng.pdf. Accessed 2012 April 20.
2. Guyatt GH, Oxman AD, Vist GE, Kunz R, Falck-Ytter Y, et al. (2008) GRADE: an emerging consensus on rating quality of evidence and strength of recommendations. BMJ 336: 924–926.
3. WHO website. Available: http://www.who.int/tb/publications/global_report/2011/gtbr11_main.pdf. Accessed 2012 April 20.
4. WHO website. Available: http://whqlibdoc.who.int/publications/2011/9789241501569_eng.pdf. Accessed 2012 April 20.
5. Menzies D, Lewis M, Oxlade O (2008) Costs for tuberculosis care in Canada. Can J Public Health 99: 391–396.
6. Floyd K, Hutubessy R, Kliiman K, Centis R, Khurieva N, et al. (2012) Cost and cost-effectiveness of multidrug-resistant tuberculosis treatment in Estonia and Russia. Eur Respir J 40: 133–142.
7. Rajalahti I, Ruokonen EL, Kotomaki T, Sintonen H, Nieminen MM (2004) Economic evaluation of the use of PCR assay in diagnosing pulmonary TB in a low-incidence area. Eur Respir J 23: 446–451.
8. Migliori GB, Ambrosetti M, Besozzi G, Farris B, Nutini S, et al. (1999) Cost-comparison of different management policies for tuberculosis patients in Italy. AIPO TB Study Group. Bull World Health Organ 77: 467–476.
9. Dinnes J, Deeks J, Kunst H, Gibson A, Cummins E, et al. (2007) A systematic review of rapid diagnostic tests for the detection of tuberculosis infection. Health Technol Assess 11: 1–196.
10. Heymann SJ, Brewer TF, Ettling M (1997) Effectiveness and cost of rapid and conventional laboratory methods for Mycobacterium tuberculosis screening. Public Health Rep 112: 513–523.
11. Wurtz R, White WD (1999) The cost of tuberculosis: utilization and estimated charges for the diagnosis and treatment of tuberculosis in a public health system. Int J Tuberc Lung Dis 3: 382–387.
12. Rouse DJE (2001) Economics of TB Drug Development: New Data for New Research. New York, NY: The Global Alliance for TB Drug Development.
13. Dowdy DW, Lourenco MC, Cavalcante SC, Saraceni V, King B, et al. (2008) Impact and cost-effectiveness of culture for diagnosis of tuberculosis in HIV-infected Brazilian adults. PLoS One 3: e4057.
14. Scherer LC, Sperhacke RD, Ruffino-Netto A, Rossetti ML, Vater C, et al. (2009) Cost-effectiveness analysis of PCR for the rapid diagnosis of pulmonary tuberculosis. BMC Infect Dis 9: 216.
15. Chen Q WL, Li Y, Yang X, Li D (2011) Interferon-γ Release Assays Screening for Latent Tuberculosis Screening: A Cost-Effectiveness Analysis. Chin J Evid-based Med 11: 768–774.
16. Kamolratanakul P, Hiransuthikul N, Singhadong N, Kasetjaroen Y, Akksilp S, et al. (2002) Cost analysis of different types of tuberculosis patient at tuberculosis centers in Thailand. Southeast Asian J Trop Med Public Health 33: 321–330.
17. Sohn H, Sinthuwattanawibool C, Rienthong S, Varma JK (2009) Fluorescence microscopy is less expensive than Ziehl-Neelsen microscopy in Thailand. Int J Tuberc Lung Dis 13: 266–268.
18. Suarez PG, Floyd K, Portocarrero J, Alarcon E, Rapiti E, et al. (2002) Feasibility and cost-effectiveness of standardised second-line drug treatment for chronic tuberculosis patients: a national cohort study in Peru. Lancet 359: 1980–1989.
19. WHO (2005) The efficiency of TB Laboratory services in the Russian Federation. WHO Policy Brief.
20. Balabanova Y, Drobniewski F, Nikolayevskyy V, Kruuner A, Malomanova N, et al. (2009) An integrated approach to rapid diagnosis of tuberculosis and multidrug resistance using liquid culture and molecular methods in Russia. PLoS One 4: e7129.
21. Sinanovic E, Floyd K, Dudley L, Azevedo V, Grant R, et al. (2003) Cost and cost-effectiveness of community-based care for tuberculosis in Cape Town, South Africa. Int J Tuberc Lung Dis 7: S56–62.
22. Albert H (2004) Economic analysis of the diagnosis of smear-negative pulmonary tuberculosis in South Africa: incorporation of a new rapid test, FASTPlaqueTB, into the diagnostic algorithm. Int J Tuberc Lung Dis 8: 240–247.
23. Hausler HP, Sinanovic E, Kumaranayake L, Naidoo P, Schoeman H, et al. (2006) Costs of measures to control tuberculosis/HIV in public primary care facilities in Cape Town, South Africa. Bull World Health Organ 84: 528–536.
24. Chihota VN, Grant AD, Fielding K, Ndibongo B, van Zyl A, et al. (2010) Liquid vs. solid culture for tuberculosis: performance and cost in a resource-constrained setting. Int J Tuberc Lung Dis 14: 1024–1031.
25. Whitelaw A, Peter J, Sohn H, Viljoen D, Theron G, et al. (2011) Comparative cost and performance of light-emitting diode microscopy in HIV-tuberculosis-co-infected patients. Eur Respir J 38: 1393–1397.
26. Vassall A, van Kampen S, Sohn H, Michael JS, John KR, et al. (2011) Rapid diagnosis of tuberculosis with the Xpert MTB/RIF assay in high burden countries: a cost-effectiveness analysis. PLoS Med 8: e1001120.
27. Muniyandi M RR, Balasubramanian R (2006) Estimating provider cost for treating patients with tuberculosis under revised national tuberculosis control programme (RNTCP). Indian J Tuberc 53: 12–17.
28. Mueller DH, Mwenge L, Muyoyeta M, Muvwimi MW, Tembwe R, et al. (2008) Costs and cost-effectiveness of tuberculosis cultures using solid and liquid media in a developing country. Int J Tuberc Lung Dis 12: 1196–1202.
29. Kivihya-Ndugga LE, van Cleeff MR, Githui WA, Nganga LW, Kibuga DK, et al. (2003) A comprehensive comparison of Ziehl-Neelsen and fluorescence microscopy for the diagnosis of tuberculosis in a resource-poor urban setting. Int J Tuberc Lung Dis 7: 1163–1171.
30. Okello D, Floyd K, Adatu F, Odeke R, Gargioni G (2003) Cost and cost-effectiveness of community-based care for tuberculosis patients in rural Uganda. Int J Tuberc Lung Dis 7: S72–79.
31. International Monetary Fund website. Available: http://www.imf.org/external/ns/cs.aspx?id = 28. Accessed 2012 April 2.
32. WHO website. Available: http://www.who.int/choice/publications/p_2003_generalised_cea.pdf. Accessed 2012 April 2.
33. WHO website. Available: http://whqlibdoc.who.int/hq/2002/who_cds_tb_2002.305a.pdf. Accessed 2012 April 2.
34. Horne DJ, Royce SE, Gooze L, Narita M, Hopewell PC, et al. (2010) Sputum monitoring during tuberculosis treatment for predicting outcome: systematic review and meta-analysis. Lancet Infect Dis 10: 387–394.
35. Lagrange PH, Thangaraj SK, Dayal R, Despande A, Ganguly NK, et al. (2012) A toolbox for tuberculosis diagnosis: an Indian multicentric study (2006–2008): microbiological results. PLoS One 7: e43739.

36. Cuevas LE, Yassin MA, Al-Sonboli N, Lawson L, Arbide I, et al. (2011) A multi-country non-inferiority cluster randomized trial of frontloaded smear microscopy for the diagnosis of pulmonary tuberculosis. PLoS Med 8: e1000443.
37. Kanaujia GV, Lam PK, Perry S, Brusasca PN, Catanzaro A, et al. (2005) Integration of microscopy and serodiagnostic tests to screen for active tuberculosis. Int J Tuberc Lung Dis 9: 1120–1126.
38. Matee M, Mtei L, Lounasvaara T, Wieland-Alter W, Waddell R, et al. (2008) Sputum microscopy for the diagnosis of HIV-associated pulmonary tuberculosis in Tanzania. BMC Public Health 8: 68.
39. Cattamanchi A, Dowdy DW, Davis JL, Worodria W, Yoo S, et al. (2009) Sensitivity of direct versus concentrated sputum smear microscopy in HIV-infected patients suspected of having pulmonary tuberculosis. BMC Infect Dis 9: 53.
40. Marais BJ, Brittle W, Painczyk K, Hesseling AC, Beyers N, et al. (2008) Use of light-emitting diode fluorescence microscopy to detect acid-fast bacilli in sputum. Clin Infect Dis 47: 203–207.