

Serological tests for tuberculosis: the evidence is reviewed

**Karen R Steingart, MD, MPH
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Berlin, 11 November 2010**



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Disclosure

- ▶ I serve as co-chair of the Evidence Synthesis subgroup of Stop TB Partnership's New Diagnostics Working Group
 - ▶ I am a member of the GRADE (Grading of Recommendations Assessment, Development and Evaluation) Working Group
- 

Overview

- ▶ Background
 - ▶ Updated systematic review and meta-analysis
 - ▶ Cost effectiveness model
 - ▶ WHO/TDR Laboratory-based...report, 2008
- 

Serological tests for TB

- ▶ Antibody-based immune assays
- ▶ Have been around for a long time
- ▶ Attractive, especially if made into point of care (POC)
- ▶ Existing serological tests have failed (3 systematic reviews)

OPEN ACCESS Freely available online

PLOS MEDICINE

Commercial Serological Antibody Detection Tests for the Diagnosis of Pulmonary Tuberculosis: A Systematic Review

Karen R. Steingart^{1,2}, Megan Henry³, Suman Laal^{4,5,6}, Philip C. Hopewell^{1,2}, Andrew Ramsay⁷, Dick Menzies^{8,9}, Jane Cunningham⁷, Karin Weldingh¹⁰, Madhukar Pai^{8,9*}

CLINICAL AND VACCINE IMMUNOLOGY, Feb. 2009, p. 260–276
1556-6811/09/\$08.00+0 doi:10.1128/CVI.00355-08
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Vol. 16, No. 2

Performance of Purified Antigens for Serodiagnosis of Pulmonary Tuberculosis: a Meta-Analysis[†]

Karen R. Steingart,^{1*} Nandini Dendukuri,² Megan Henry,^{3,4} Ian Schiller,² Payam Nahid,⁴ Philip C. Hopewell,^{1,4} Andrew Ramsay,⁵ Madhukar Pai,² and Suman Laal^{6,7,8}

A systematic review of commercial serological antibody detection tests for the diagnosis of extrapulmonary tuberculosis

Karen R Steingart, Megan Henry, Suman Laal, Philip C Hopewell, Andrew Ramsay, Dick Menzies, Jane Cunningham, Karin Weldingh, Madhukar Pai

Current situation

- ▶ No serological TB test for clinical use is recommended by international guidelines nor approved by the US Food and Drug Administration
 - ▶ Dozens of commercial serological tests based on detection of antibodies are marketed in many parts of the world, especially in developing countries with weak regulatory systems
- 

Based on a survey of 80+ Indian labs, some preliminary estimates:

- About 50 large and medium private labs alone are doing over 60,000 tests per month
- If all 20,000+ labs/hospitals in India are included, could easily exceed 120,000 per month [~1.5 million per year]
- @ \$10 per test**, market is worth at least \$15 million [this is highly conservative]

** Cost is actually ~\$10 per antibody (e.g. IgG). Combination of all 3 antibodies (IgG, IgA, IgM) is often done at cost of >\$30 per patient; for simplicity, we have used \$10 per patient, a conservative estimate (RNTCP annual budget ~ \$65 million)

Pai et al. Unpublished

Case study from India on how inaccurate serological tests for TB can do harm

- 25 year old male from Chhattisgarh presented with cough, fever, and weight loss for several months
- Seen at community health center where physician advised a TB serology test (USD \$4.50)
- Result was negative for TB

PATHOLOGY LABORATORY
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SWAL
T)

AGE : SEX M-
DATE: 01-05-08

ION	OBSERVED VALUE	NORAMAL VALUE
OBIN (CYAN)	9.2 GM %	F.12-14 M.14-16 GM%
C. COUNT	/ CUMM.	4000-12000/ CUMM.
C. COUNT		
UTROPHILS	%	40-75%
MPHOCYTES	%	20-40%
SINOPHILS	%	01-05
ONOCYTES	%	02-06%

: 8.5MMAT THE END OF 1 ST HOUR

GROUP :

D.R.L :

TIME : 1-3 MTS

TIME : 2-6 MTS

TEST :

FOR TB : Negative

for :

in :

TEST :

of :

TH :

TH :

TH :

SIGNATURE

Case study continued

- Patient was sent home with a Rx for vitamins and cough syrup
- Within a few weeks, his condition rapidly deteriorated
- Sputum smear microscopy was positive for acid-fast bacilli and the patient eventually died of his disease

Date
2/5/08

Rf

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(4) Desojal vain 5ml
⑧ All



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THE POLITICAL SCENE

Hendrik Hertzberg and Ryan Lizza discuss the midterm results.



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Michael Specter answers readers' questions about tuberculosis, **Thursday at 3 P.M. E.T.**

LETTER FROM INDIA

A DEADLY MISDIAGNOSIS

Is it possible to save the millions of people who die from TB?

by Michael Specter

TEXT SIZE: A | A | A

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NOVEMBER 15, 2010



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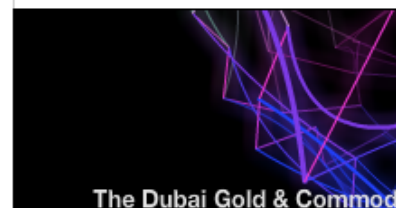
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LETTER FROM INDIA

A DEADLY MISDIAGNOSIS

Is it possible to save the millions of people who die from TB?

BY MICHAEL SPECTER

Every afternoon at about four, a slight woman named Runi slips out of the cramped, airless room that she shares with her husband and their sixteen children. She skirts the drainage ditch in front of the building, then walks toward the pile of hardened dung cakes that people in this slum on the edge of the northeastern Indian city of Patna use for fuel. Dressed in a bright-yellow sari shot with gold threads, Runi is followed by several of her children. Although she can't remember their ages, or her own, Runi must be about forty, because she

of the National TB Control Program, told me when we met in New Delhi. "But there are thousands of labs. Shut one down and the next day ten more appear."

Runi's test was indeed worthless. It determined the presence of antibodies, which show that a body's immune system has begun to respond to an infection. But most TB infections are latent: no more than ten per cent will ever cause illness. This means that ninety per cent of people with antibodies for TB in their blood don't have the disease. Ru-

"Now she really is sick," he continued, explaining that Runi's TB was no longer dormant, and that taking drugs when they are not necessary often makes them ineffective when they are. "This is what happens when tests mislead us. She will need the drugs again. If they don't work properly, she will be in real trouble. She has almost certainly infected some of her children. That makes everything harder, more expensive, more painful."

Tuberculosis strikes vulnerable people with special ferocity. Victims are seized by severe night sweats, wasted by fatigue, and punished by the blood-tinged cough that is the disease's defining symbol. In most cases, tuberculosis affects the lungs, but it can invade almost any organ of the body. When an infectious person coughs, sneezes, spits, or even shouts, he sends minute particles of sputum, or phlegm, into the air—ex-

Commercial serological tests for the diagnosis of tuberculosis: an updated systematic review and meta-analysis

Steingart KR, Flores LL , Dendukuri N , Schiller I, Laal S, Ramsay A, Hopewell PC, Pai M

Population - active TB, all ages, all countries

Intervention - commercial serological test

Comparison - microscopy smear

Outcomes – sensitivity and specificity

Reference standard – culture

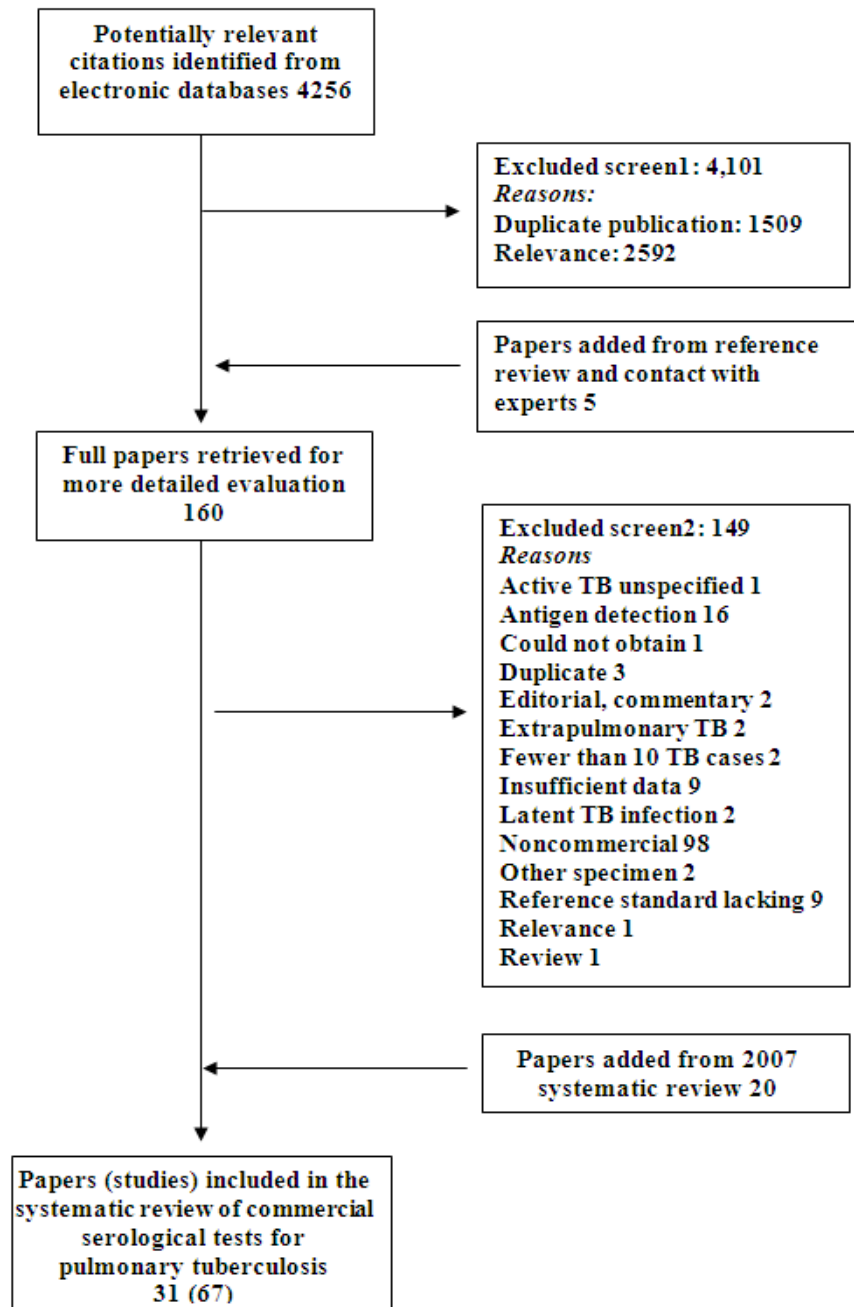
Excluded studies published before 1990 and studies with < 10 TB cases



Results - Pulmonary TB



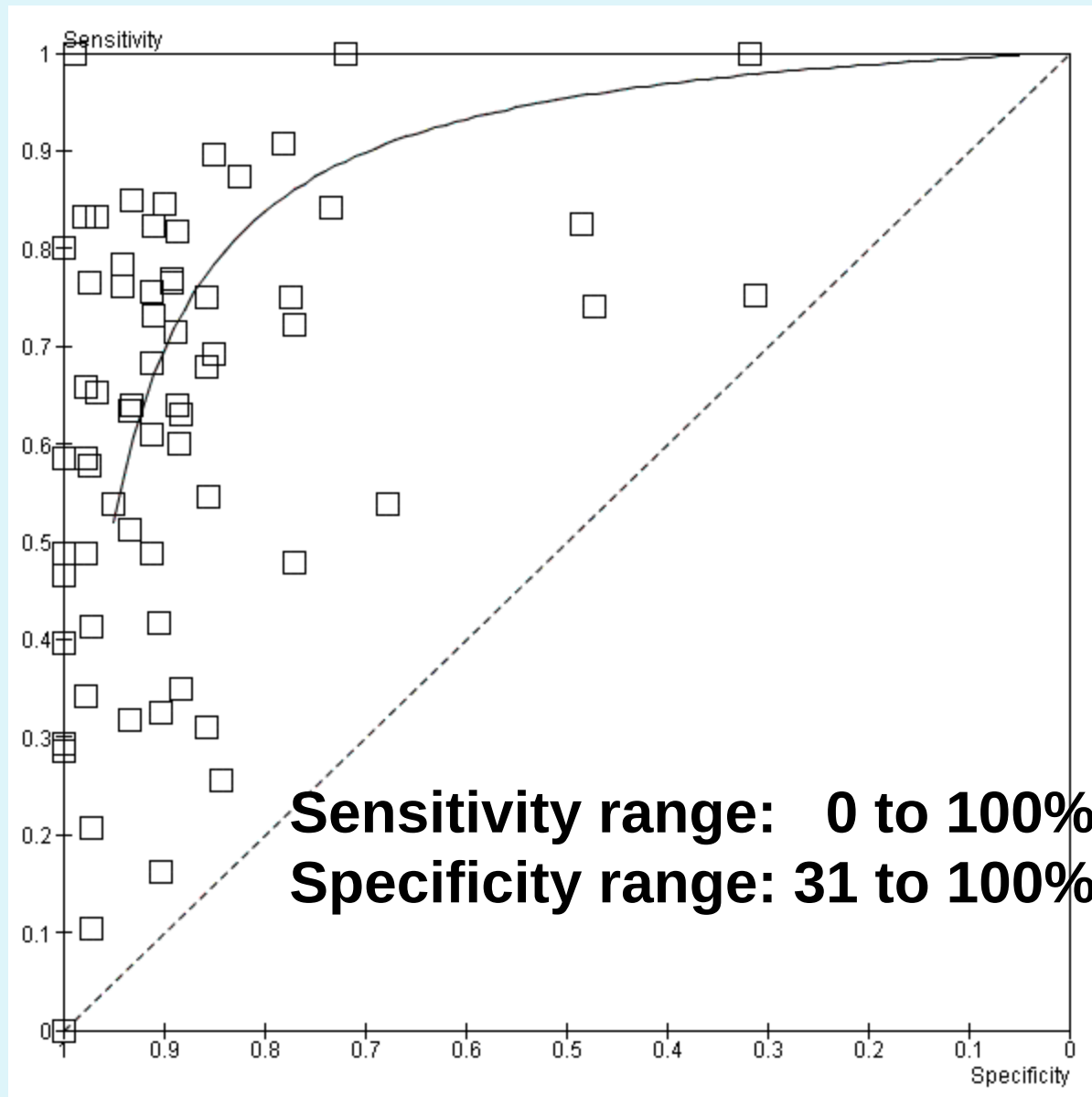
Flow of studies



Study characteristics

- ▶ 67 studies used 18 different serological tests (anda-TB IgG most common, 19% of studies)
- ▶ 32 (48%) studies in low/middle-income countries
- ▶ Zero studies involved children; 1 study involved HIV-infected individuals
- ▶ Median TB patients 41 (IQR 33, 54)
- ▶ No studies reported on patient-important outcomes

Plot of sensitivity versus specificity for all 67 studies in the review



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REF 311909

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IgG
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ELISA test for the determination of anti-mycobacteria
Test ELISA pour la détermination des anticorps anti-mycobactéria
192 Determinations / 192 Déterminations

KIT COMPONENTS

1. Sample Diluent**
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3. Concentrated TMB (10x)
4. TMB Diluent
5. Sulfuric acid
6. Hing, 1,2,4,5,6 U/ml Ref**
7. Weak Positive Control**
8. Strong Positive Control**
9. Concentrated Washing Buffer (20x)
10. Antigen A60-coated plate

anda-tb IgG
No pasteurized
No pasteurized

anda
Biologicals

ELISA test for the determination of anti-mycobacterial antibodies
Test ELISA pour la détermination des anticorps anti-mycobactéria
192 Determinations / 192 Déterminations

KIT COMPONENTS

1. Sample Diluent**
2. Anti-human IgG-labeled Ab**
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6. Hing, 1,2,4,5,6 U/ml Ref**
7. Weak Positive Control**
8. Strong Positive Control**
9. Concentrated Washing Buffer (20x)
10. Antigen A60-coated plate

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**Hing: 0.05% (w/v), 0.05%

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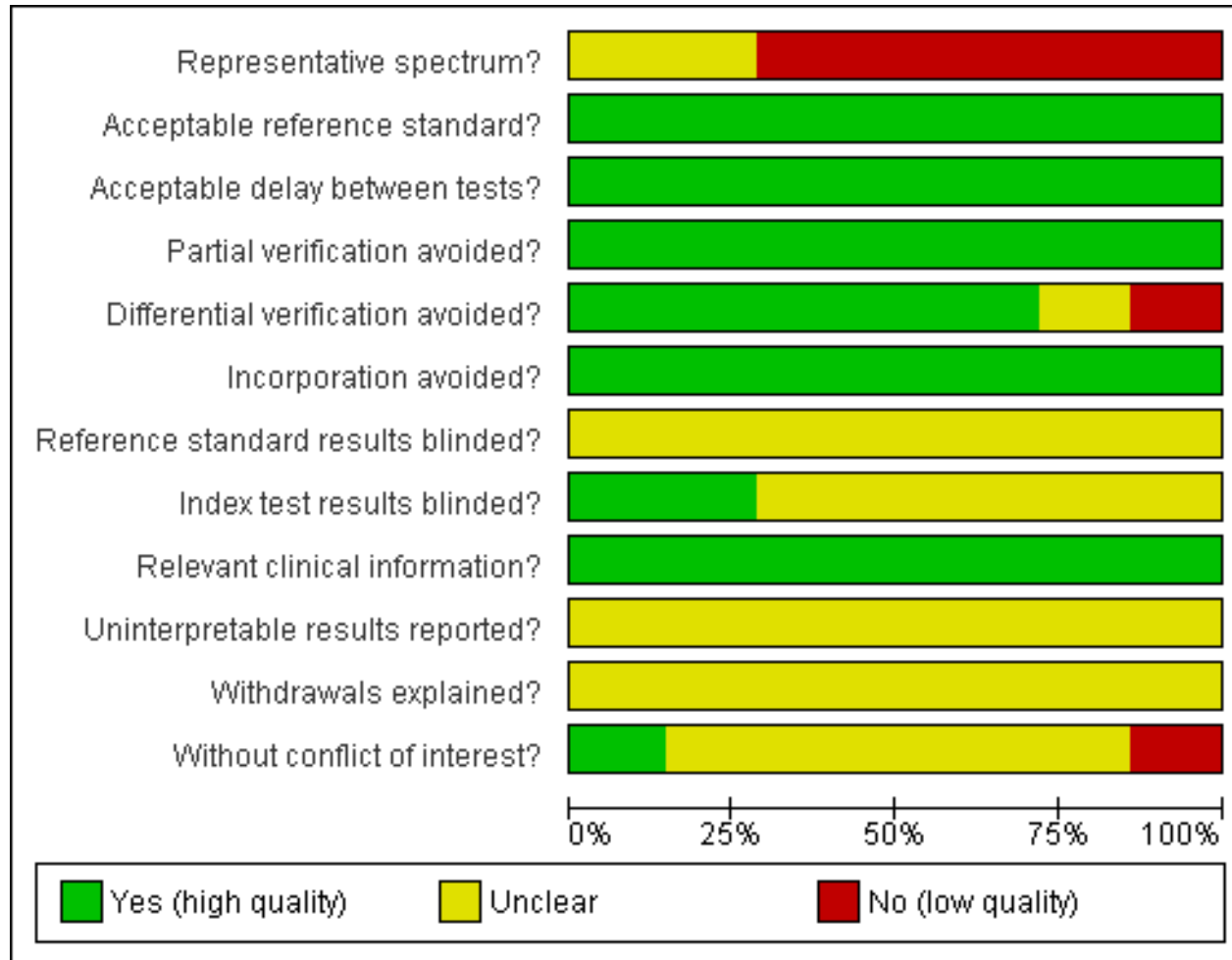
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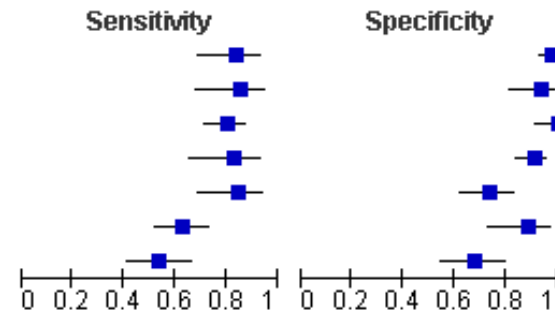
anda-TB IgG (Anda Biologicals, Strasbourg, France) studies involving smear-positive patients



anda-TB IgG

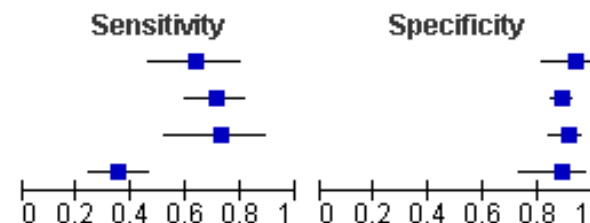
A. Smear +

Study	TP	FP	FN	TN	Sensitivity	Specificity
Alifano 1994	35	2	7	92	0.83 [0.69, 0.93]	0.98 [0.93, 1.00]
Alifano 1996 (a)	28	3	5	41	0.85 [0.68, 0.95]	0.93 [0.81, 0.99]
Kalantri 2005 (a)	84	0	21	40	0.80 [0.71, 0.87]	1.00 [0.91, 1.00]
Okuda 2004 (a)	28	10	6	101	0.82 [0.65, 0.93]	0.91 [0.84, 0.96]
Traunmuller 2005	32	21	6	58	0.84 [0.69, 0.94]	0.73 [0.62, 0.83]
Wu 2004 (a)	58	4	34	30	0.63 [0.52, 0.73]	0.88 [0.73, 0.97]
Wu 2005	35	19	30	40	0.54 [0.41, 0.66]	0.68 [0.54, 0.79]



B. Smear -

Study	TP	FP	FN	TN	Sensitivity	Specificity
Alifano 1996 (c)	23	3	13	41	0.64 [0.46, 0.79]	0.93 [0.81, 0.99]
Luh 1996 (b)	50	33	20	260	0.71 [0.59, 0.82]	0.89 [0.85, 0.92]
Okuda 2004 (b)	19	10	7	101	0.73 [0.52, 0.88]	0.91 [0.84, 0.96]
Wu 2004 (b)	30	4	56	30	0.35 [0.25, 0.46]	0.88 [0.73, 0.97]

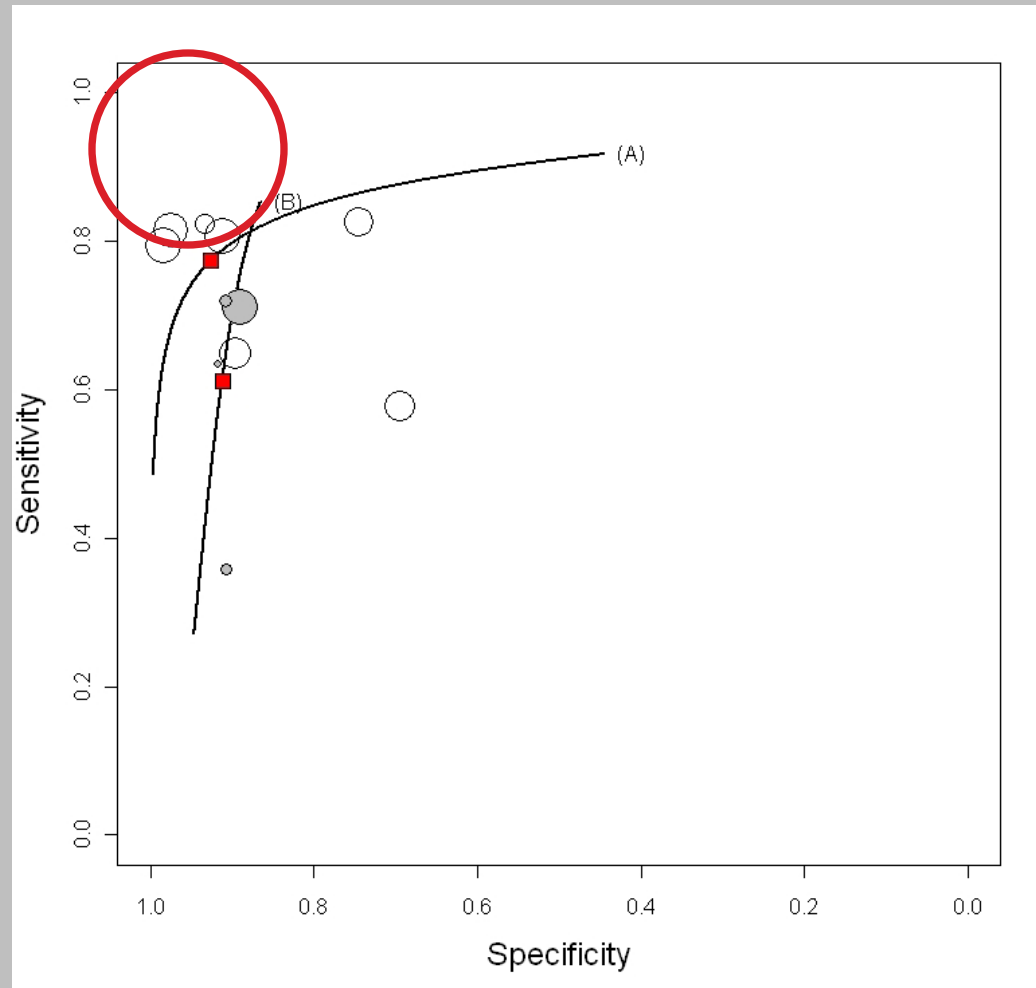


Bivariate meta-analysis pooled estimates

Smear+ Sensitivity = 76% (63,87); Specificity = 92% (74, 98)

Smear- Sensitivity = 59% (10,96); Specificity = 91% (79, 96)

Summary ROC plots for Anda-TB IgG showing better performance in studies of smear-positive patients (A) than in studies of smear-negative patients (B). The red squares are summary sensitivity and specificity



Limitations

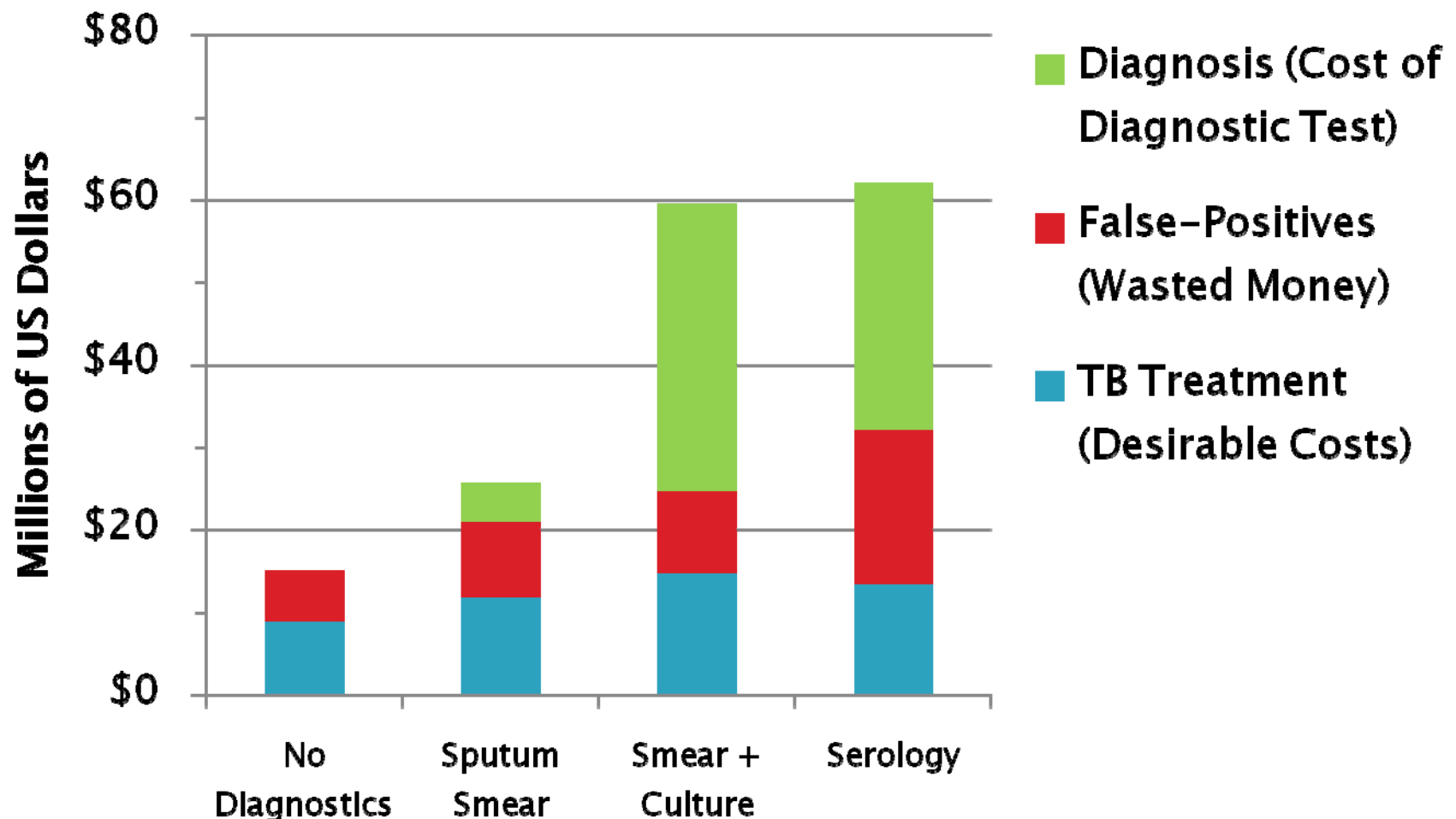
- ▶ Majority studies not representative or blinded
- ▶ Meta-analysis limited by small number of studies for a particular serological test
- ▶ Tests used different cut-points
- ▶ Children & HIV-infected individuals, data insufficient
- ▶ No data on patient-important outcomes

Serologic testing for tuberculosis in India: cost-effectiveness model

Hypothetical “Study Population”

- ▶ 1.5 million TB suspects
 - Conservative estimate of annual volume of serologic tests in India (sensitivity analysis on 3 mil)
- ▶ 1 in 7 actually have TB
 - Estimate from FIND, comparable to other studies
- ▶ Among TB patients, 53% are “highly infectious”
 - Would be diagnosed with 2 sputum smears in an ideal lab
- ▶ 5% HIV prevalence
 - 10% with access to ART (UNAIDS 2009)
 - Does not affect model results

What is the cost for 1.5 million TB suspects who undergo serologic testing in India?



David Dowdy, 2010 unpublished

Table 3. Cost-Effectiveness of Diagnostic Strategies for 1.5 Million TB Suspects in India, Relative

Diagnostic Test	Cost (US\$)	Additional TB Cases Treated	Additional False-Positive Cases Treated	Secondary Cases Averted
Sputum smear microscopy	\$11.9 million	44,000	36,000	443,000
Sputum smear + TB culture	\$45.0 million	71,000	48,000	555,000
Serological testing	\$47.5 million	58,000	157,000	411,000
Rapid molecular testing	\$52.8 million	86,000	12,000	629,000

WHO/TDR Laboratory-based...2008

- ▶ Rapid - test result (< 15 mins)
- ▶ Simple - 1 or 2 steps, minimal training and no equipment
- ▶ Easy to interpret - card or strip format with visual readout
- ▶ Gold standard - culture plus clinical follow-up
- ▶ Archived specimens

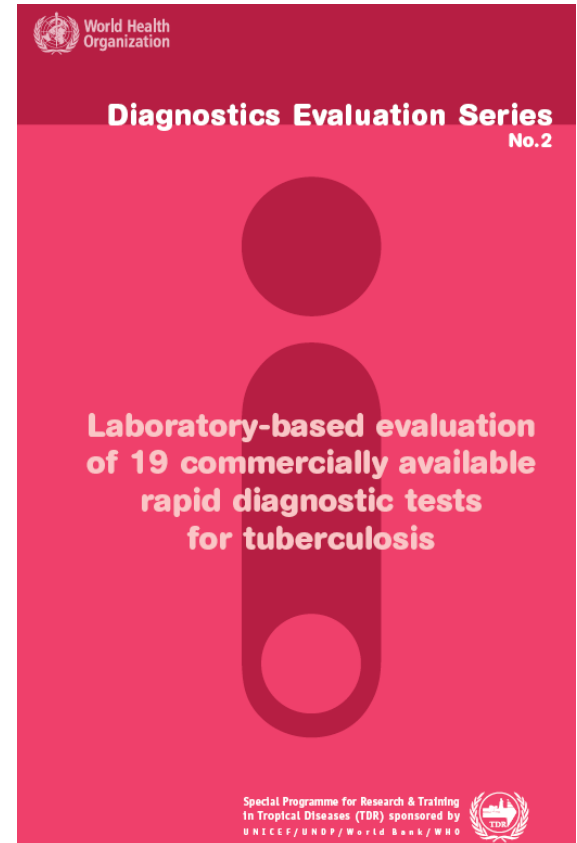
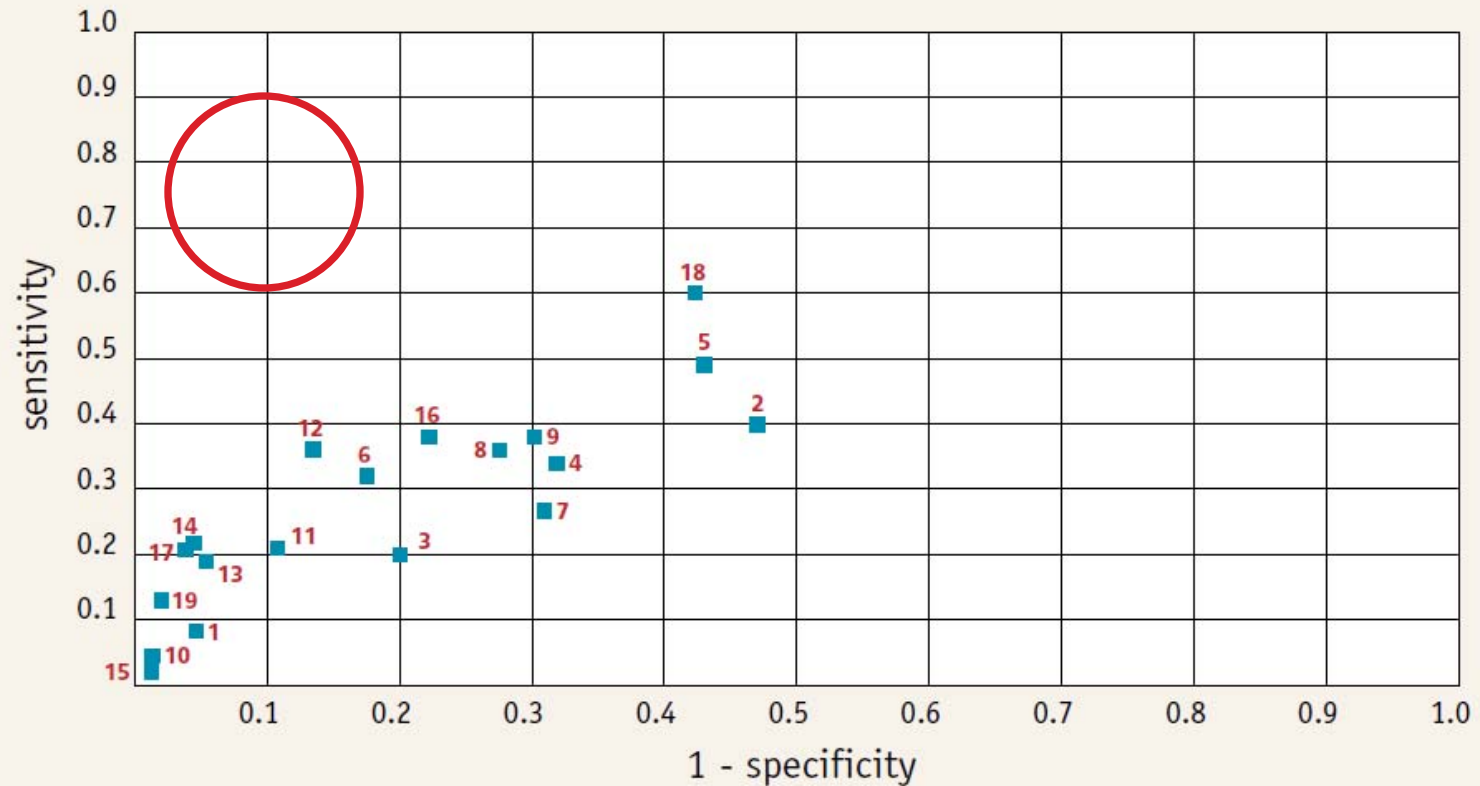


Table 4. Performance of 19 rapid tests for pulmonary tuberculosis

	Manufacturer	Rapid Test	Sensitivity % (95% CI)	Specificity % (95% CI)
1	ABP Diagnostics	Focus Sure Check TB	8 (4-11)	95 (92-99)
2	Advanced Diagnostics	Tuberculosis Rapid Test	40 (33-46)	53 (45-61)
3	American Bionostica	Rapid Test for TB	20 (15-26)	80 (73-86)
4	Ameritek dBest	One Step TB Test	34 (27-40)	68 (61-76)
5	BioMedical Products Corp	TB Rapid Screen Test	49 (42-56)	57 (49-65)
6	Chembio	TB Stat-Pak II	32 (25-38)	83 (76-89)
7	CTK Biotech TB Antibody	Onsite Rapid Screening Test	27 (21-33)	69 (62-77)
8	Hema Diagnostic	Rapid 1-2-3 TB Test	36 (29-42)	72 (65-80)
9	Laboratorio Silanes	TB-Instantest	38 (31-44)	70 (62-77)
10	Millenium Biotechnology	Immuno-Sure TB Plus	2 (0-5)	99 (97-100)
11	Minerva Biotech	V Scan	21 (16-27)	89 (84-94)
12	Mossman Associates	MycoDot	36 (30-42)	87 (81-92)
13	Pacific Biotech	Bioline TB	19 (14-25)	95 (91-98)
14	Premier Medical Corporation	First Response Rapid TB	21 (16-27)	95 (92-99)
15	Princeton BioMeditech	BioSign M tuberculosis	1 (0-2)	99 (97-100)
16	Span Diagnostics	TB Spot ver 2.0	38 (32-45)	78 (71-85)
17	Standard Diagnostics	SD Rapid TB	21 (15-26)	96 (93-99)
18	UniMED International Inc	FirstSign MTB Card Test	60 (53-66)	58 (50-66)
19	Veda Lab	TB Rapid Test	13 (8-17)	98 (96-100)

ROC curve, commercial rapid tests for the diagnosis of pulmonary TB (n=355)



Sensitivity range: 1 to 60%
Specificity range: 53 to 99%

In conclusion

- ▶ Published data on commercial serological tests produce inconsistent and imprecise estimates of sensitivity and specificity
 - ▶ There is no evidence that existing serological assays improve patient-important outcomes
- 

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