Diagnosis of drug-resistant tuberculosis disease in children: A practical approach

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Classification of DR-TB disease according to certainty

(*research definitions* [Seddon et al, 2013])

Confirmed:

- at least 1 of the signs and symptoms suggestive of TB disease, *and*
- detection of *M. tb* from the child with demonstration of genotypic or phenotypic resistance.

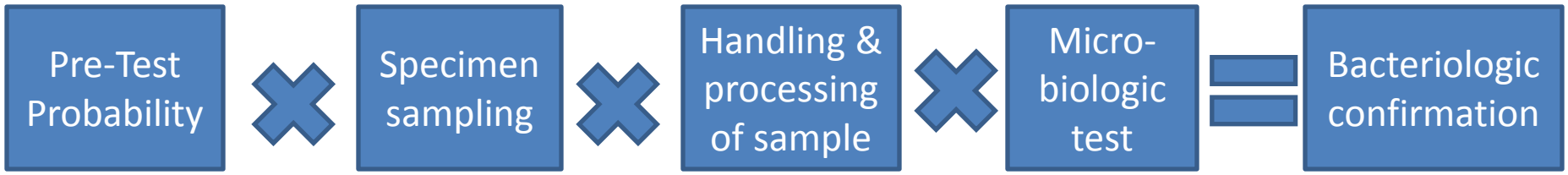
Probable:

- diagnosis of probable TB disease, *and*
- DR-TB contact

Possible:

- diagnosis of probable TB disease, *and*
- either (a) contact of a source case with TB disease who has risk factors for drug resistance, or (b) failure of first-line TB treatment

Determinants of bacteriological confirmation



Pre-Test Probability: Definition of Probable TB disease

(*Research* definitions [Graham et al, 2012])

- At least 1 of the signs and symptoms suggestive of TB disease, *and*
- CXR consistent with intrathoracic TB disease, *and*
- Presence of 1 of the following:
  (a) a positive clinical response to TB treatment
  (b) documented exposure to a source case with TB disease
  (c) immunological evidence of TB infection

M.tb detection & Drug Susceptibility Testing (DST)

Phenotypic: Cultures

- **Preferred Methods**: culture method that detects *M.tb* and DST simultaneously
  - Solid media (eg, Middlebrook 7H10 or 7H11)
  - Liquid media (eg, BACTEC MGIT 960)
- **Minimum threshold for detection**: 10-100 CFU/mL
- **Advantages**
  - Gold-standard for DST to essentially all TB meds
  - Treatment response monitoring
- **Disadvantages**:
  - Labor-intensive, time-consuming, expensive, requires specialized equipment and biosafety level 3 facility.
  - Results may take 2-6 wk; direct DST is preferred, as indirect DST adds ~8 days.

M.tb detection & drug resistance testing (DRT)

Genotypic: Nucleic Acid Amplification Tests (NAATs)

- **Preferred Methods**: automated NAAT that detects *M.tb* complex and DR simultaneously
  - Real-Time Polymerase Chain Reaction (eg, Xpert MTB/RIF)
    - Minimal threshold: 100-150 CFU/mL (c.w. 10,000 CFU/mL for microscopy)
    - Advantages: Useful in smear microscopy (+) & (-) samples; **rapid** (2 hr, vs. 2-8 wks for cultures); very practical (a self-contained, fully integrated, automated, requiring minimal technical expertise to operate); useful in various types of specimens; useful for "ruling in" (not for ruling out")
    - Disadvantages: *Currently* only identifies Rmp (rpoB); not useful for treatment response monitoring (detect dead bacilli)
  - Line Probe Assays (eg, GenoType MTBDRplus *version 2*)

M.tb detection & drug resistance testing (DRT)

Genotypic: Nucleic Acid Amplification Tests (NAATs)

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  - Real-Time Polymerase Chain Reaction (eg, Xpert MTB/RIF)
  - Line Probe Assays (eg, GenoType MTBDRplus *version 2*)
    - **Minimal threshold**: 100-150 CFU/mL (c.w. 10,000 CFU/mL for microscopy)
    - **Advantages**
      - Useful in smear microscopy (+) & (-) samples
      - Rapid (5 hr)
      - Can be done manually or automated
      - Useful for "ruling in" (not for ruling out")
    - **Disadvantages**
      - *Currently* only identifies Rmp (rpoB), low-level INH (inhA), & high-level (katG) resistance
      - not useful for treatment response monitoring (detect dead bacilli)

M.tb detection & drug resistance testing (DRT):
Xpert MTB/RIF vs GenoType MTBDRplus

Study: Barnard et al, 2012 (*J Clin Microbiol* 50(11):3712)

Design: Comparison of performance of Xpert MTB/RIF and GenoType MTBDRplus (v.2.0) on microscopy (+) & (-) patient specimens, using culture as gold-standard.

Setting: National Health Laboratory Service, Cape Town, South Africa.

Results: 282 consecutive specimens were tested by both Xpert MTB/RIF and Genotype MTBDRplus

- Similar sensitivities c.w. MGIT culture, ie, GenoType MTBDRplus (v2) and Xpert MTB/RIF were 73.1% and 71.2%, respectively
- Similar sensitivities c.w. microscopy(-) / culture(+) specimens: 57-58%
- Same specificities (100%) for M.tb detection

M.tb detection & drug resistance testing (DRT): INH-resistance / Rmp-susceptibility

Study: Smith et al 2012 (*Int J Tuberc Lung Dis* 16:203)

Question: *To what extent Rmp resistance is an adequate marker for MDR-TB?* [implications for Tx & surveillance]

Design: Retrospective analysis of data (WHO/The Union Global DRS data 1994–2007) from >81 countries and subnational settings.

Results

- In settings with relatively low MDR-TB prevalence (one third of all countries and subnational settings): >40% of Rmp-resistant isolates from *new* TB cases did not display resistance to INH.
- Among the third of countries or settings in the middle tertile, >24% of Rmp-resistant new TB cases had INH-susceptible TB.

Conclusion: INH susceptibility testing -- in addition to RMP susceptibility testing – may be indicated…

Induced sputa using Xpert MTB/RIF in children

Study: Nicol et al, 2011 (*Lancet Infect Dis* 11:819)

Setting: high burden of both TB and HIV (Cape Town, South Africa)

Study Design: prospective clinical study

Inclusion criteria: hospitalized/inpatient, range up to 15 years, pulm. TB suspected on the basis of having cough for >14 days, plus one of the following:

- TST-positive, or household contact infected with TB; or
- Failure to gain weight, or CXR suggestive of pulm. TB.

Specimens: 2 induced sputum (IS) samples

Number recruited (*n*): 385 with 2 IS; 452 with at least 1 IS

Ages: median 19 months

Results:

Culture(+): **16%** (70/452)

Xpert MTB/RIF(+): **13%** (58/452)

Smear microscopy(+): 6% (27/452)

Incremental yield of 2nd IS by culture: 13.8 % (8/58)

Incremental yield of 2nd IS by Xpert MTB/RIF, *in smear (-) cases*: 27.8%

IS & SS using Xpert MTB/RIF in children

Study: Rachow et al, 2012 (*Clin Infect Dis* 54:1388)

Setting: high burden of both TB and HIV (Mbeya, Tanzania)

Study Design: prospective clinical study.

Inclusion criteria: inpatient *& outpatient*, <14 y.o., with at least 1 of the following symptoms:

(a) Persistent, unremitting cough for 21 days

(b) Repeated episodes of fever within the last 21 days

(c) Weight loss or failure to thrive within the previous 3 months; or,

(d) Signs and symptoms suggestive of extrapulmonary TB.

Specimens: 3 sputum samples – spontaneous (58.5%) & induced (41.5%).

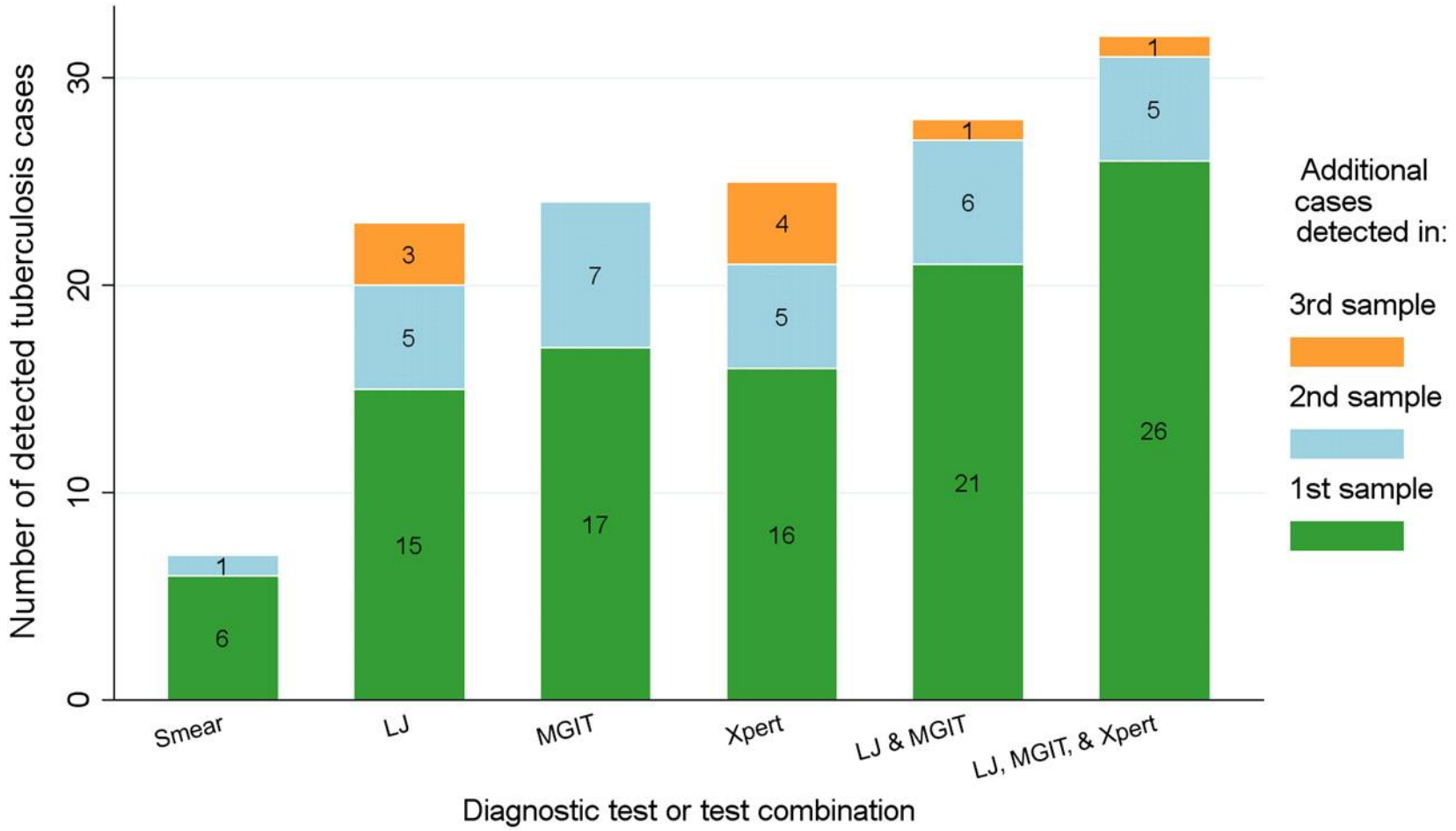
Recruitment: 164; median age 5.8 years

Results: Culture(+): 17.1% (28/164)

Xpert MTB/RIF(+): 15.2% (25/164) -- 21 (75%) of 28 culture(+); *4 of 47 culture(-)*

Smear microscopy(+): 4.3% (7/164)

Bacteriological confirmation: Incremental gain per additional sample analyzed



Rachow et al, 2012 (Clin Infect Dis. 54:1388)

NPA vs. IS using Xpert MTB/RIF in Children

Study: Zar et al, 2012 (*Clin Infect Dis* 54:1388)

Setting: high burden of both TB and HIV (Cape Town, South Africa)

Study Design: prospective clinical study

Inclusion criteria: hospitalized/inpatient, up to 15 y.o., pulm. TB suspected on the basis of having cough for >14 days, plus one of the following:

- TST-positive, or household contact infected with TB; or
- Failure to gain weight, or CXR suggestive of pulm. TB.

Specimens: Paired nasopharyngeal aspirate (NPA) and induced sputum (IS)

Number recruited (*n*): 535 (21.9% HIV-infected) at least one pair; 396 two pairs; median age 19 months

NPA vs. IS using Xpert MTB/RIF in Children

Study: Zar et al, 2012 (*Clin Infect Dis* 54:1388)

Results:

- Overall very low bacteriological yield of 1-2 paired NPA + IS
  - Culture(+): 16.3% (87/535)
  - Xpert MTB/RIF(+): 15.1% (81/535)
  - Smear microscopy(+): 5.6% (30/535)
- Of culture(+), IS yield (84/87, 96.6%) higher than NPA yield (61/87, 70.1%) [P < .001].
- Of Xpert(+), IS yield (71%; 45/63) similar to NPA yield (65%; 41/63) [P=0.44].
- Xpert complementary to culture: Bacteriologically confirmed 5 IS & 7 NPA that were culture(-)
- A 2nd sample increases yield: Incremental yield of 2nd IS & of 2nd NPA by:
  - Liquid culture (MGIT): IS 17.6%; NPA 26.3%; NAAT (Xpert MTB/RIF): IS 25%; NPA 36.7%
- Amongst children with two paired specimens, 63 culture-confirmed cases occurred [60 (95.2%) IS vs 48 (76.2%) NPA, P = .002].

GA using Xpert MTB/RIF in children

Study: Bates et al, 2013 (*The Lancet Infectious Diseases* 13:36)

Setting: High burden of both TB and HIV (Lusaka, Zambia).

Study design: Prospective clinical study.

Inclusion criteria: hospitalized/inpatient with suspected pulmonary TB , 15 years or younger

Number Recruited: 930

Specimens: 1 spontaneous sputum (SS) in 142 (15%)
1 gastric lavage aspirate (GLA) in 788 (85%)

Results:

- Overall yield was low
  - cultures were positive in 58 children (6%).
- Xpert MTB/RIF performed similarly on GLA & sputum *(p=0.1649)
  - Xpert MTB/RIF on GLA had a sensitivity of 69% (33/48)
  - Xpert MTB/RIF on SS had a sensitivity of 90%* (9/10)

Stool using Xpert MTB/RIF in children

Study: Nicol et al, 2013 (*Clinical Infectious Diseases* 57:e18)

Setting: high burden of both TB and HIV (Cape Town, South Africa)

Study design: Prospective clinical study

Age: median age 31 months (interquartile range 19–57 months)

Inclusion criteria: hospitalized/inpatient, pulm. TB suspected on the basis of having cough for >14 days, plus one of the following:

- TST-positive, or household contact infected with TB; or
- Failure to gain weight, or CXR suggestive of pulm. TB.

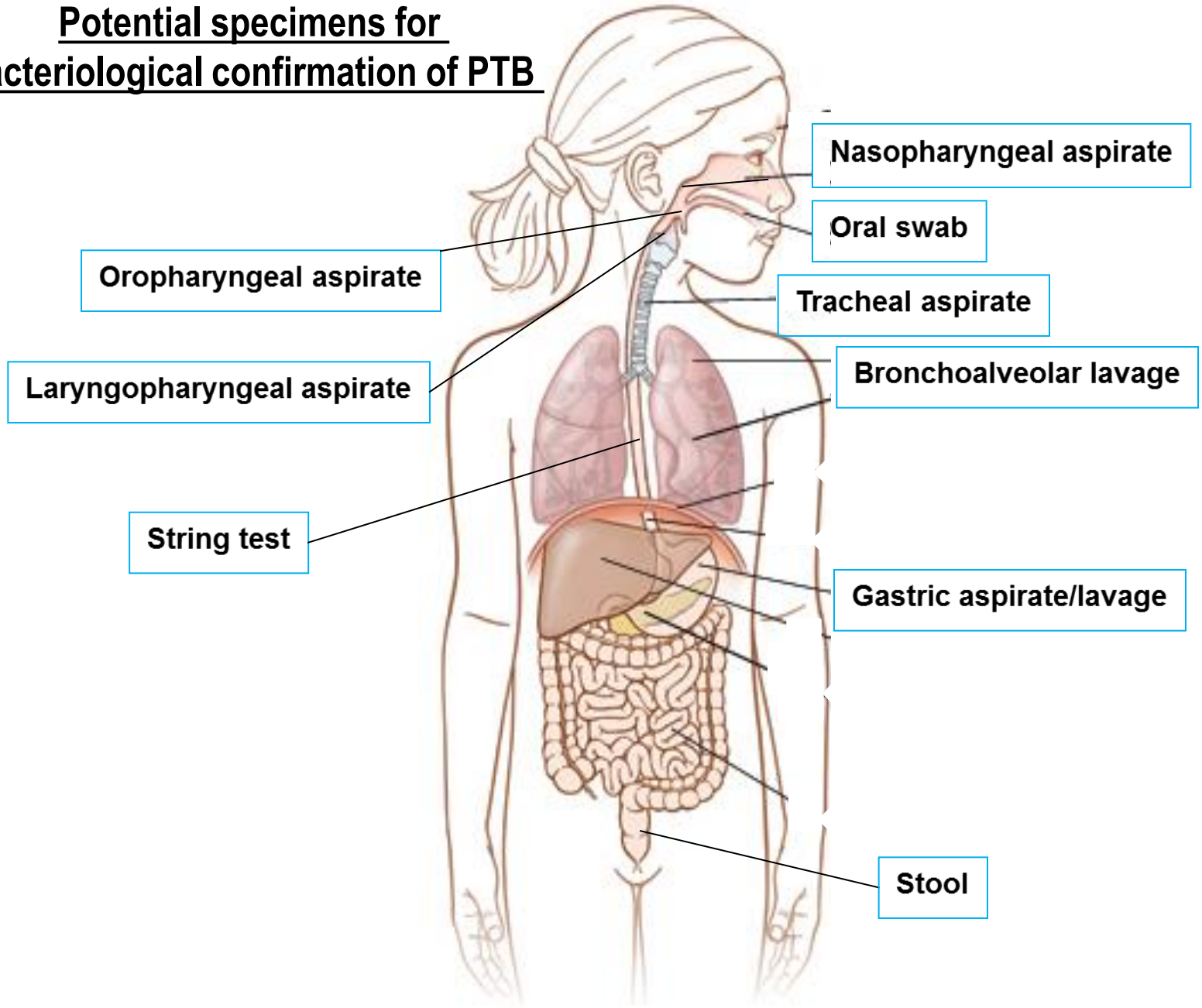
Recruitment: 115 children, of whom 17 (14.8%) were HIV infected and 67 (58.3%) were hospitalized.

Specimens: 1 stool; 2 induced sputa (IS)

Results:

- Overall yield was low
  - cultures were positive in 14.8% (17/115)
- Xpert MTB/RIF c.w. liquid culture (BACTEC MGIT)
  - 1 stool had a sensitivity of 47% (8/17)
  - 2 IS had a sensitivity of 65% (11/17)
  - Sensitivity of Xpert on stool versus IS was not significantly different (p=0.30); *however, sample size very small.*

Potential specimens for bacteriological confirmation of PTB



Variety: Combination of specimens
("intensive" specimen collection)

- Collection of various specimens
  - Gastric aspirate (or gastric lavage)
  - Spontaneous/induced sputum or laryngopharyngeal aspirate
  - Swallowed sputum in esophagus (on string)
  - Nasopharyngeal swab
  - Oral swab
  - Fine-needle aspirate of lymph nodes
  - Cerebrospinal fluid
  - Blood
  - Urine

Types of Specimens

Specimen	Collection procedure	Age group	Minimum volume	Best collection time	Other comments
Spontaneous sputum	Cough up sputum without prior saline nebulization	>7 years	3 ml	Early morning	If unable to produce enough sputum, consider sputum induction
Induced sputum/ laryngo-pharyngeal aspirate	hypertonic saline nebulization before cough up sputum	Any age	3 ml	Early morning	If child unable to cough, consider laryngo-pharngeal suctioning
Gastric aspirate	Nasogastric aspiration of gastric juice containing swallowed sputum	< 7 years	5 ml	Early morning before out of bed	After waking up and sitting and standing, stomach begins to empty, losing volume of aspirate

Types of Specimens

Specimen	Collection procedure	Age group	Minimum volume	Best collection time	Other comments
Gastric Lavage	Nasogastric instillation of solution to wash off and recover sputum adhered to walls of stomach	<7 years	10 ml	Early morning	Use only if at least 3 ml of gastric aspirate can not be obtained
String Test	Esophagogastro-duodenal nylon yarn that can absorb swallowed sputum	> 4 years	N/A	Unknown, duration probably more important	Consider when good quality or quantity of sputum and aspirate can not be obtained
Naso-pharyngeal aspirate	Nasopharyngeal suctioning to collect secretions from URT, but may also collect from LRT if cough reflex is stimulated	< 6 years	2 ml	Unknown, probably higher yield in morning	Yield tends to be similar to or lower than that of induced sputum or gastric aspirate/lavage

Types of Specimens

Specimen	Collection procedure	Age group	Minimum volume	Best collection time	Other comments
Stool	Uncontaminated by toilet bowl or urine	Any age	1 table-spoon (5 g)	Any time	Bacterialogic yield has been lower than that of sputum and gastric lavage and gastric aspirate
Broncho-alveolar lavage (BAL)	Bronchoscopy	Any age	3 ml	Any time	Bacteriologic yield of one sample is not superior to serial induced sputum or gastric lavage or gastric aspirate
Cerebro-spinal fluid	Lumbar puncture	Any age	2 ml	Any time	Submit 3rd or 4th tube for culture to reduce chance of contamination from skin flora

Types of Specimens

Specimen	Collection procedure	Age group	Minimum volume	Best collection time	Other comments
Serosal (pleura, pericardium, peritoneum, synovium)	Serosal fluid aspirate followed by serosal tissue biopsy	Any age	1 ml	Any time	Bacteriologic yield of tissue is much higher than fluid. Biochemical markers useful in all fluids
Urine	Clean catch, mid-stream urine	Any age	2 ml	1st morning urination	Yield low except in urinary tract TB. Lipoarab-inomannan antigen very sensitive in immuno-compromised HIV positive patients
Blood	Phlebotomy	Any age	5 ml	Any time	Yield very low, use in severely ill HIV infected patients
Fine needle aspiration	Fine needle aspiration and/or biopsy	Any age	Based on type	Any time	Useful because histo-pathological features consistent with TB can be diagnostic

Specimen collection Strategies: Pooling of Samples

- Pooling of spontaneous sputum *in adults* [Warren et al, 2000; AJRCCM]
  - By requiring minimum of 5 mL, and pooling daily samples until reached, yield increased 27% (72.5% to 92.0%)
- Pooling of [enhanced] gastric aspirates in children [Loeffler, 2003; *Sem. Resp. Infect.*]
  - GAs, 2 per day (AM & PM), x 3 days (total of 6 samples)
  - Morning sample provided the best culture results
  - Results: At least one positive culture was obtained from 12 of 13 children (ie, 92%).
- Pooling of swallowed sputum on string (preliminary) [Perez-Velez et al 2010, *AJRCCM* 181:A1775]
  - Using "home-brew" PCR as detection method, yield of 3 pooled strings > yield than 1 gastric aspirate or 1 induced sputum/laryngopharyngeal aspirate

Variability in the reported bacteriological confirmation rates of PTB in children: Possible explanations

Most "high-yield" studies tend to have higher thresholds for inclusion with criteria that are more refined:

- Younger children (↑ yield)
- Passive case finding
  - More likely to have CXRs & expert reading (↑ yield)
  - Longer course/history of illness (↑ yield)
- More likely to have CXRs & expert reading (↑ yield)
- More likely to have specimens collected (some with a hospitalization bias) that are…
  - Better quantity & quality (↑ yield)
  - More expeditious processing (↑ yield)

Specimen Collection Strategy

- Variety: Collect multiple samples of different specimens
  - eg, GA (x 2) & IS (x 2) & LN-FNA(s)
- Quantity: Do not self-impose limit to volume
  - eg, 5 mL of gastric aspirate, in early AM, usually possible
- Quality: Collect samples properly
  - Use clean/sterile technique (to minimize contamination)
  - Avoid dilution (eg, GA preferred over GL)
  - Avoid adding preservatives (eg, use sterile water)
  - Neutralize ASAP (eg, < 30 minutes)
  - Avoid prolonged room temperature (eg, place in cooler)

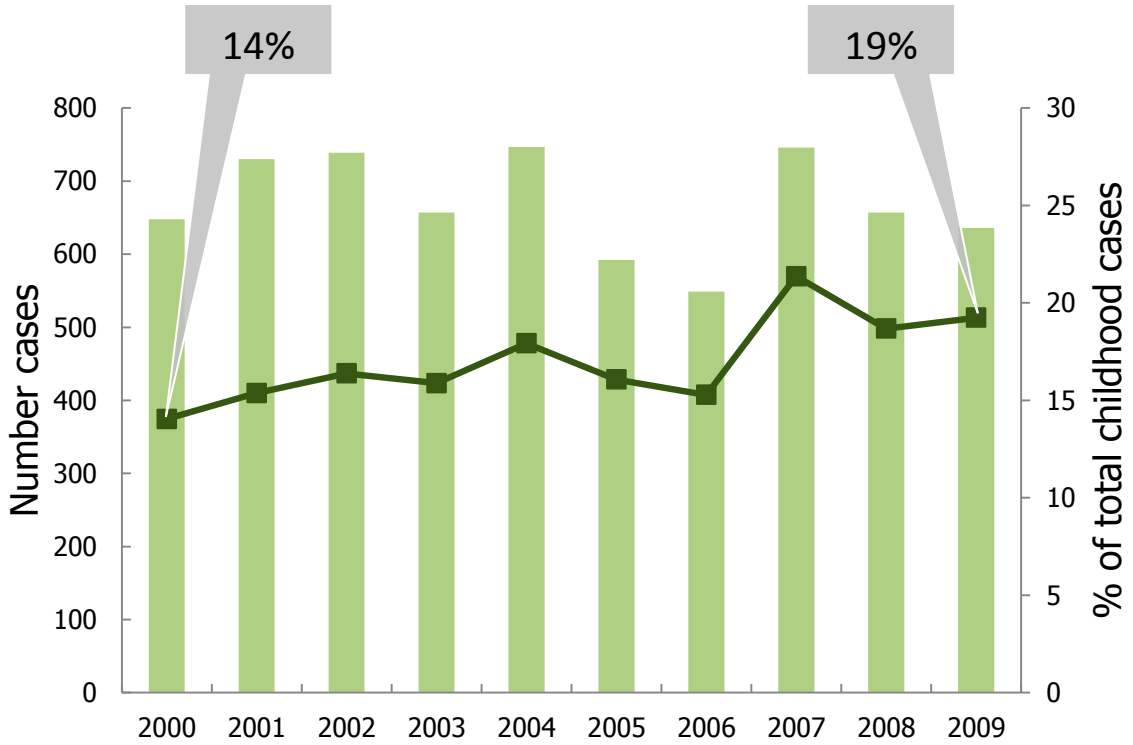
Key points: Bacteriological Confirmation

- Re. Overall bacteriologic confirmation rate of pulm. TB:
  - very low with currently available diagnostic methods.
- Re. Tests: Culture and NAATs (eg, Xpert) are complementary
  - NAATs are far superior to microscopy as rapid tests.
  - NAATs do not replace culture methods.
- Re. Specimens:
  - Variety specimens: although some specimens may have higher yield than others by certain tests, no single specimen type confirms all cases (ie, combinations may be complementary)
  - Quality (eg, GA vs. GL; early morning vs. later)
  - Quantity: ↑ volume → ↑ bacillary load→ ↑ likelihood of surpassing minimum threshold for detection
- Re. Samples: Both by culture and by Xpert, there is an incremental gain from 1 sample to 2 samples, and from 2 to 3 samples

Challenges and barriers to diagnosing TB disease in children

- **Only 19% of childhood TB cases were confirmed by culture in 2009.**
- The trends over the last decade indicate a slight improvement in culture confirmation.
- Only 42% of the nearly 40 000 cases reported were tested by culture. Of these, fewer than 7000 (40%) were culture positive.

Figure: Number of culture-positive childhood TB cases and proportion of total childhood cases, 2000–2009



European Centre for Disease Prevention and Control, 2011

Diagnosis of DR-TB

Question

- *In clinical practice*, should the diagnosis of DR-TB be limited to only bacteriologically confirmed cases of drug resistance?

Diagnosis of DR-TB

Question

- *In clinical practice*, should the diagnosis of DR-TB be limited to only bacteriologically confirmed cases of drug resistance?

- Should patients at *significant risk* for DR-TB be treated as such?

Diagnosis of DR-TB

Case

1 y.o. child, brought by mom to local hospital in Buenaventura, Colombia, with 2-week history of a clinical syndrome consistent with acute meningitis, mother recently diagnosed with smear-positive pulm. TB, *confirmed* to be MDR. Child's CSF predominantly lymphocytic pleocytosis; CXR reveals mediastinal lymphadenopathy & right mid-lung consolidation.

You suspect disseminated (meningeal, pulmonary, & intrathoracic LN) TB disease.

Questions

1. [For clinical purposes] Would you begin empiric coverage for MDR-TB?

Diagnosis of DR-TB

Case

Second-line drugs were not available; a standard regimen of first-line drugs (INH+Rmp+Pza+Emb) was given. After an initial brief clinical improvement, the child soon deteriorated and 2 weeks later. Culture of gastric aspirate grew *M.tb* after 4 weeks; drug-susceptibility testing results were received 8 weeks later from the national reference lab and reported resistance to INH and Rmp.

DR-TB in children:
WHO Guidance (2006)

- ***"Children are as susceptible to drug-resistant as to drug-sensitive TB."***
- *"Drug-resistant TB is a laboratory diagnosis."*
- *"Drug-resistant TB should be suspected if…"*
- *"The diagnosis and treatment of drug-resistant TB in children is complex and should be carried out at referral centres."*

Key Point

- WHO and many NTP guidelines suggest that presumptive diagnosis may be made, and hence respective treatment may be indicated, ***but do not explicitly state so.***

DR-TB in children

Problems

- There are [*too*] many children with DR-TB disease that are not treated because diagnosis not confirmed.
- Prevalence ("market") of DR-TB underestimated.
  - Governments do not proportionately/adequately fund pediatric program within NTP
  - Diagnostics companies not persuaded to develop and study new tests for children
  - Pharmaceutical companies not persuaded to develop pediatric formulations

One of the biggest obstacles to getting children into treatment for MDR-TB is delay in making the diagnosis

- Median delay of 36 weeks seen in a large cohort in South Africa.

[Seddon et al., 2012, J. Trop. Peds.]

- Early diagnosis is linked with improved outcomes.

[Ettehad et al., 2012, Lancet ID]

Challenges in diagnosing TB in children

Challenges for clinical diagnosis

- Clinical presentation:
  - Extrapulmonary disease (eg, *intrathoracic LN disease*)
  - Non-localized Sx/Si: ↓ playfulness; ↓ appetite; failure to thrive; fever
  - "Atypical" respiratory symptoms: eg, wheezing

Challenges for bacteriological confirmation

- Paucibacillary disease
- Difficulties in specimen collection
  - Spontaneous sputum not possible in young children
  - Invasive procedures (eg, gastric lavage/aspiration; sputum induction requiring laryngopharyngeal suctioning; nasopharyngeal aspiration)
    - Culturally not acceptable in some communities (eg, Amerindians; Armenians)
    - Operational barriers in some primary-level centers (trained personnel; equipment; space)

Challenges in diagnosing TB in children

Broad spectrum of clinical syndromes and severity of disease:

Presence and type of clinical manifestations depend on the stage of disease progression which in turn depends on the immune function

- —*the sooner disease is diagnosed and treated, the less morbidity.*

  - **Early stages**:
    - May have very mild symptoms (just constitutional and/or immunological symptoms and signs).
    - But… diagnosis in early stages requires a high index of suspicion.
  - **Later stages**:
    - Localizing signs and symptoms of the affected organ/system/region begin to appear.
      - Intrathoracic TB is the most common clinical syndrome of TB in children.

Clinical Dx: Increasing probability of TB as etiology

- Microbiological studies (Culture; NAATs; Ag detection; microscopy)
- Histopathological studies
- Biological markers
- Immune-based tests
- *Epidemiological risk factors suggestive of exposure*
- *Alternative DDx have been ruled out by appropriate tests*
  - *Comorbidity/coinfection under-recognized (esp. in immunocompromised)*
  - *Acute pneumonia due to M.tb also under-recognized (6% in cohort of young children in Medellin, Colombia—currently ongoing study).*
- *Alternative DDx have failed appropriate therapeutic trials*

Increasing probability of drug-resistance: screening for risk factors

- Features in the source case suggestive of DR-TB
  - contact with a known case of DR-TB
  - remains sputum smear-positive after 3 months of Tx
  - history of previously treated TB
  - history of treatment interruption
- Features of a child suspected of having DR-TB
  - contact with a known case of DR-TB
  - not responding to the TB treatment regimen

DR-TB in Children: Dx & Tx

So… Question

In children…

- with clinical diagnosis of TB disease,
- with risk factors for DR-TB,
- with negative [final] test results (cultures & PCRs-DRT)
- who remain "stable" (ie, without complicated disease)…

…should we wait for complications before considering DR-TB Tx?

…should we initiate Tx for presumed pansusceptible TB?

…should we initiate Tx for presumed DR-TB disease?

DR-TB in Children: Dx & Tx

Question

In children…

- with clinical diagnosis of TB disease,
- with risk factors for DR-TB,
- with negative [final] test results (cultures & PCRs-DRT)
- who remain "stable" (ie, without complicated disease)…

…should we wait for complications before considering DR-TB Tx?

…should we initiate empiric Tx for presumed pansusceptible TB?

…should we initiate empiric Tx for presumed DR-TB disease?

Answer

No evidence base to guide decision making…

DR-TB in Children: Dx & Tx

My **opinion** regarding when to initiate empiric coverage for DR-TB disease…

- In clinically unstable children: initiate ASAP

and

- **In clinically stable, but with *higher risk* for progression to complicated disease**

  – *In 167 children with TBM, the mean period between recognition of first symptoms of TBM and death was 19.5 days*

[Lincoln et al, 1960; *J Pediatr* 57:807]

Risk factors for progression from infection to disease to death
Why are they relevant?

- The identification of risk factors for progression from infection to disease, and from disease to death, is important to:
  - *Increase the index of suspicion* for the Dx of TB disease (both pulmonary and extrapulmonary), and expedite the diagnostic evaluation.
  - *Expedite the initiation of TB Tx* if there is sufficient evidence from findings consistent with TB disease.
  - May affect TB *treatment response or outcome*.

Diagnostic Specimen Management: Principles

- Biosafety:
  - Children can be smear-positive & have a cough effective enough to transmit TB
  - Health care workers should wear N95 masks
- Contamination:
  - Avoid contamination from adjacent secretions or tissues
  - Sterilize all equipment after use
- Quality:
  - Collect respiratory specimen at optimal times (pref. early AM) to maximize yield
  - Avoid Fasting 3-6 hours (depending on diet and age)
  - Avoid gastric lavage if aspirate >2-3 mL
- Quantity: minimum quantity unknown
  - In adults, more than 5 mL increased yield ~more than 25% [Warren et al, 200]
  - Principle: the more organisms collected, the higher the likelihood of detection

Diagnostic Specimen Management: Principles (cont.)

- Minimize risk of false-negatives culture:
  - Collect specimen before starting Tx
  - When TB in DDx, avoid agents with antimycobacterial activity
  - Avoid saline solution with antimycobacterial preservatives
- Preservation:
  - Confirm neutral pH of potentially acidic specimens (eg, gastric aspirate)
  - Use appropriate transport media
  - Minimize transport time
  - Store specimens appropriately

Key Points

1. *We're **not** there yet!...* As long as the majority of DR-TB disease cases in children are not bacteriologically confirmable, presumptive diagnosis of DR-TB disease should be made when risk factors—as stipulated by WHO—are present.
   - should be the "norm"—*not the "exception"*—most especially in TB programs that are not diagnosing (and treating) *a similar proportion of children with DR-TB as adults*
2. Appropriate empiric DR-TB treatment should be…
   - *Initiated* when clinically unstable (and, of course, when Tx failure)
   - *Very seriously considered* when there are risk factors for progression to TB disease and death (eg, immunocompromising conditions, persisting "infecting inoculums", decreasing effectiveness of available Tx options (ie, MDR/XDR).
3. A systematic approach is needed for the *prudent* presumptive diagnosis DR-TB disease
4. Optimize specimen collection strategy: *variety, quantity, quality*
   - In cases of severe/complicated TB disease, do not delay the initiation of *empiric* TB treatment just to collect specimens—do both *concomitantly*!