Laboratory

- Often a bottleneck or a factor to define the velocity of the survey
Lab work

Smear Microscopy

Direct Smear, ZN
LED-FL

Collect, Store, Transport and Put in Culture within 5 days
Sputum collection

• At least two samples: so far- spot+morning
• STAG-WHO recommendations
  – Two spots with 1hr or longer interval
• Difficult to collect quality specimen from non-symptomatic

• No-induced sputum, no laryngeal swab, no gastric tubing
Reverse cold chain from collection to processing the specimen

- 3 days from collection and local storage to a culture lab
- 2 days in culture lab till processing

Do within 5 days, maximum in 7 days
Estimate workload

- Sample size: 50,000
- 90% participation: 45,000
- 12% eligible for sputum exams: 5,400
- 95% of specimens are collected and arrived: 10,500 samples
- If ZN MS: 420 person days, If LED FL MS: 105 person days
- Capacity of culture: Human resources, incubators etc
Smear

- Work Load: ZN 25, LED FL 100/ Per technician day
- Qualified by the EQA by lot sampling does not mean a capacity for study at all
- Smear in local lab may increase a chance of contamination – Open and re-cap
Exam in repeated surveys

• Is it really necessary to keep consistency in exam methods?
  – Which has priority: Having more accurate estimate or simply comparing two survey results?
  – HIV TB: lower bacteriological load - Prevalence of TB detectable by conventional technology may decrease while that of TB with lower bacteriological load in sputum increases
Quality of Sputum samples

- Directly Observed Taking Sputum at least for Spot Specimens
  - Clear Instruction, Visual Aid (posters)

- Don’t discard “saliva” specimens
  - Judgement by naked eyes are not always correct

- One specific specimen: Smear and culture in same lab → “Smear - re-cap – transportation” causes contamination
Quality Assured Lab often failed

- Survey specimens from field are totally different from clinical specimen from TB suspects in medical facility
  - Quality of sputum
  - Volume of a single specimen
  - Bacteriological load
  - Time (days) from collection to examination
  - Quantity (No. of specimens sent one time)

- Limit the number of Labs and technicians to assure quality examinations
VIABILITY CHANGE OF *M. TUBERCULOSIS*
IN SPUTUMS STORED AT DIFFERENT TEMPERATURES

**Culture positive percentage**

<table>
<thead>
<tr>
<th>Duration of storage (days)</th>
<th>0</th>
<th>3</th>
<th>7</th>
<th>14</th>
<th>21</th>
<th>28</th>
</tr>
</thead>
<tbody>
<tr>
<td>4°C</td>
<td>100</td>
<td>94</td>
<td>77</td>
<td>25</td>
<td>15</td>
<td>10</td>
</tr>
<tr>
<td>25°C</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>25-30°C</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
</tr>
</tbody>
</table>

*No. of sputa tested:*

- 41
- 41
- 36

*Kim SJ, et al, 1986*
*Paramasivan CN, et al, 1983*
One culture or two or more

<table>
<thead>
<tr>
<th>number (cases)</th>
<th>1st sputum</th>
<th>total</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>any positive</td>
<td>negative</td>
</tr>
<tr>
<td>2nd sputum</td>
<td></td>
<td></td>
</tr>
<tr>
<td>any positive</td>
<td>151</td>
<td>74</td>
</tr>
<tr>
<td>negative</td>
<td>61</td>
<td></td>
</tr>
<tr>
<td>total</td>
<td>212</td>
<td></td>
</tr>
</tbody>
</table>

At least one: Having additional exam has more yield than expanding the screening criteria
“1/3 of specimen could not be treated within 7 days after collection”

Figure B. Days to Processing Correlated with Contamination Rate and Culture Positivity Rate

Unexpected Problem in a certificated Lab at the beginning
We may have more S(−)C(+) cases

Experiences in National TB Lab in Yangon, 2006

<table>
<thead>
<tr>
<th>Culture Recovery Rate of S(+) Cases</th>
<th>Any positive</th>
<th>Smear (+) Cases</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>number C(+) (%)</td>
<td>number C(+) (%)</td>
</tr>
<tr>
<td>First 16 clusters</td>
<td>50 23 46%</td>
<td>37 23 62%</td>
</tr>
<tr>
<td>Second 14 clusters</td>
<td>25 21 84%</td>
<td>23 21 91%</td>
</tr>
</tbody>
</table>

(Non-eligible participants are included)

Technical problem in sputum treating process (ex.decontamination) was suspected
• Philippines
• Myanmar
• Cambodia
• Ethiopia
WHO recommended method
or
Locally adapted method

Concentration method
  Or
Direct method

"quality of exam affect more than difference by methods does"