An International Interlaboratory Quality Control program for Bio-Analysis of TB Drugs

Aarnoutse RE¹, Sturkenboom MGG², Robijns K³, Harteveld A³,
Greijdanus B², Uges DRA², Touw DJ³, Alffenaar JW²

¹ Radboud University Nijmegen Medical Centre, Nijmegen
² University Medical Centre Groningen, Groningen
³ Association for Quality Assessment in TDM and Clinical Toxicology, The Hague

The Netherlands
Introduction

- Numerous analytical methods for TB drugs published - many methods not published

- Methods used for quantitation of TB drugs in
  - PK + PD research: all phases of drug development
  - Patient care: Therapeutic Drug Monitoring

- Chromatographic methods
Introduction - Quality control in the bio-analysis of drugs

• *intralaboratory (internal) QC*
  
  - validated equipment
  - trained technicians
  - method validation: accuracy, precision, specificity etc
  - QC samples

• *interlaboratory (external) QC*
Introduction - Association for Quality Assessment in TDM and Clinical Toxicology, The Netherlands

- Active since 1979

- Anti-epileptic drugs, antimicrobials, cardiac drugs, benzodiazepines, psycho-active drugs, various other drugs, ‘experimental’ (including INH)

- International programs:
  antiretroviral drugs (> 50 participants, 10th anniversary)
  antifungal drugs (> 50 participants)
Methods

• Spiking of drug-free human serum with isoniazid, rifampicin, pyrazinamide, ethambutol, moxifloxacin, linezolid to obtain 2 QC samples each with 6 drugs

<table>
<thead>
<tr>
<th>Drug</th>
<th>Low conc. (mg/L)</th>
<th>Intermediate / high conc. (mg/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Isoniazid</td>
<td>0.48</td>
<td>6.47</td>
</tr>
<tr>
<td>Rifampicin</td>
<td>1.41</td>
<td>5.60</td>
</tr>
<tr>
<td>Pyrazinamide</td>
<td>18.79</td>
<td>58.61</td>
</tr>
<tr>
<td>Ethambutol</td>
<td>0.93</td>
<td>4.78</td>
</tr>
<tr>
<td>Moxifloxacin</td>
<td>0.31</td>
<td>2.65</td>
</tr>
<tr>
<td>Linezolid</td>
<td>0.91</td>
<td>11.09</td>
</tr>
</tbody>
</table>
Methods

- drugs with high and specified purity & serum from regional blood bank
- independently calibrated balances
- samples were freeze dried
- confirmative check with validated HPLC method

Analysis of samples within 6 weeks after dispatch

20% limits around true values as thresholds for satisfactory measurements
Results

- 7 laboratories participated in this first round
- Measurement of all 6 drugs: 2 laboratories
  - 5 drugs: 1 laboratory
  - 4 drugs: 2 laboratories
  - <4 drugs: 2 laboratories
- All analyses: 48/58 (83%) satisfactory results
- Analyses, subdivided by drug:
  - Isoniazid: 14/14 (100%)
  - Rifampicin: 5/12 (42%) Systematic error? Dissolution?
  - Pyrazinamide: 10/10 (100%)
  - Ethambutol: 9/12 (75%)
  - Moxifloxacin: 6/6 (100%)
  - Linezolid: 4/4 (100%)
Results

• No effect of concentration level on accuracy
• 3/7 laboratories performed all analyses within 20% inaccuracy limits
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• 3/7 laboratories performed all analyses within 20% inaccuracy limits
Conclusions

• An interlaboratory QC program for TB drugs was developed
• Program alerted some laboratories to previously undetected problems & enabled them to optimize their assays or procedures
• Extension of the program to more drugs?
  Survey of possible sources of error?
• The program will be open for more laboratories to participate

Contact:  R.Aarnoutse@akf.umcn.nl or J.W.C.Alffenaar@umcg.nl
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<table>
<thead>
<tr>
<th>Drug</th>
<th>No of measurements</th>
<th>Conc. level</th>
<th>Measured conc. relative to true conc. (%)</th>
<th>Absolute inaccuracy (%)</th>
<th>No and % of measurements with acceptable accuracy</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Median (min-max)</td>
<td>Median (min-max)</td>
<td></td>
</tr>
<tr>
<td>isoniazid</td>
<td>7</td>
<td>Low</td>
<td>95 (84-105)</td>
<td>5 (3-16)</td>
<td>7/7</td>
</tr>
<tr>
<td></td>
<td>7</td>
<td>Interm./high</td>
<td>104 (80-108)</td>
<td>7 (3-20)</td>
<td>7/7</td>
</tr>
<tr>
<td>rifampicin</td>
<td>6</td>
<td>Low</td>
<td>78 (38-116)</td>
<td>26 (6-62)</td>
<td>3/6</td>
</tr>
<tr>
<td></td>
<td>6</td>
<td>Interm./high</td>
<td>87 (49-135)</td>
<td>27 (1-51)</td>
<td>2/6</td>
</tr>
<tr>
<td>pyrazinamide</td>
<td>5</td>
<td>Low</td>
<td>108 (104-117)</td>
<td>8 (4-17)</td>
<td>5/5</td>
</tr>
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<td></td>
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<td>Interm./high</td>
<td>110 (101-118)</td>
<td>10 (1-18)</td>
<td>5/5</td>
</tr>
<tr>
<td>ethambutol</td>
<td>6</td>
<td>Low</td>
<td>107 (75-137)</td>
<td>14 (4-37)</td>
<td>4/6</td>
</tr>
<tr>
<td></td>
<td>6</td>
<td>Interm./high</td>
<td>105 (98-142)</td>
<td>4.5 (1-42)</td>
<td>5/6</td>
</tr>
<tr>
<td>moxifloxacin</td>
<td>3</td>
<td>Low</td>
<td>98 (95-105)</td>
<td>5 (2-8)</td>
<td>3/3</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>Interm./high</td>
<td>105 (87-106)</td>
<td>6 (5-13)</td>
<td>3/3</td>
</tr>
<tr>
<td>linezolid</td>
<td>2</td>
<td>Low</td>
<td>95 (90-95)</td>
<td>5.5 (1-10)</td>
<td>2/2</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>Interm./high</td>
<td>94 (89-98)</td>
<td>6.5 (2-11)</td>
<td>2/2</td>
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