A flexible scope on phytosanitary diagnostics

Arjen Werkman
National Reference Centre (NRC)
National Plant Protection Organization
The Netherlands
Basic requirements for quality management in plant pest diagnosis laboratories

Specific scope

This guideline includes specific quality management requirements for laboratories preparing for accreditation according to the ISO/IEC Standard 17025 General requirements for the competence of testing and calibration laboratories (references to relevant parts of ISO/IEC Standard 17025 are included). It should be noted that in EPPO standards the verb ‘should’ carries the highest level of obligation.

1. Introduction

Development of quality management systems (also referred to as management systems or quality systems) historically, the accreditation of laboratories has usually...
## Current accreditation NRC (17025 Fixed scope)

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<table>
<thead>
<tr>
<th></th>
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</thead>
<tbody>
<tr>
<td>1</td>
<td><em>Lycopersicon Esculentum</em> (tomato)</td>
<td>Isolation of <em>Clavibacter michiganensis subsp. michiganensis</em> in symptomatic material (stem) from tomato by plating on semi-selective media Identification of <em>Clavibacter michiganensis subsp. michiganensis</em> by IF, real-time PCR and pathogenity test</td>
<td>A-NRC-BAC-001</td>
</tr>
</tbody>
</table>

2. *Andean potato latent virus*

3. *Phytophthora ramorum*

4. *Thrips palmi*

5. *Ditylenchus dipsaci*
Why the search for phytosanitary based flexible scope

-> NRC is working since 2005 according to a quality system including audits on all processes, tests and diagnostic activities
Why the search for phytosanitary based flexible scope (2)

• Standard Fixed/flex scope do not fit well with the nature of the work
  » Some diagnoses/tests are performed only once every few years
  » Diagnostic process is based on knowledge and can be a combination of e.g. symptoms, literature, tests, morphology

• Extensive validation and 2nd/3rd line controls for each single test performed: not feasible
Phytosanitary based flex scope

Therefore:
- Agreement with Dutch Accreditation Council to come with a proposal for an specific scope
- Project started in the end of 2013
- Audit in October 2015
Principles flex scope NRC

Method: e.g. DAS-ELISA, PCR, morphology
Test: application method to a specific pest (antisera, primers)
Diagnose ≠ diagnosis of new or unknown pests
1) Scope of accreditation

In our work the diagnostic process is leading i.o. tests

• Quality management system is applicable for all tests/diagnoses
• Methods are extensive validated, tests fit for purpose
• Use of quality controls and assurance is method based
• Competence of diagnostician – Expertise Document
1) Scope of accreditation

<table>
<thead>
<tr>
<th>Organism group</th>
<th>Visual</th>
<th>Microscopy (morphological)</th>
<th>Electron microscopy</th>
<th>Extraction</th>
<th>Pathogenicity</th>
<th>Baiting</th>
<th>Indicator plants</th>
<th>Grafting</th>
<th>non-)selective isolation</th>
<th>Fatty acid</th>
<th>Isozym analysis</th>
<th>r-Page</th>
<th>PCR</th>
<th>real-time PCR</th>
<th>Sequencing</th>
<th>ELISA</th>
<th>IF</th>
</tr>
</thead>
<tbody>
<tr>
<td>Arthropoda</td>
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<tr>
<td>Bacteria</td>
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<tr>
<td>Fungi en Oomycota</td>
<td>x</td>
<td>x</td>
<td>x</td>
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<td>x</td>
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<td>Plantae</td>
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<tr>
<td>Nematodes</td>
<td>x</td>
<td>x</td>
<td>x</td>
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<tr>
<td>Viruses, viroids and phyttoplasms</td>
<td>x</td>
<td>x</td>
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<td>x</td>
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</tbody>
</table>

<table>
<thead>
<tr>
<th>No</th>
<th>Material or Product</th>
<th>Type</th>
<th>Methods</th>
</tr>
</thead>
<tbody>
<tr>
<td>2</td>
<td>Plants, plant materials and cultures</td>
<td>identification of plant pathogenic bacteria</td>
<td>IF, (non-) selective isolation, PCR, real-time PCR, Sequencing, Pathogenicity</td>
</tr>
</tbody>
</table>
2) Management of scope

- Adding new activities to scope
- Validation
- Audit process
- Internal approval institute
- Add extension to internal list
- Performing tests and reporting
- Internal List is send to AB
- Regular audit AB
- Evaluation and decision by AB

Day 1

Day 364

Day 365
2) Management of scope

-> Laboratory responsible for management of scope!

(+) Flexibility to add organisms becoming relevant during year
3) Internal list

- Not all tests mentioned in the scope need to be used in each identification, decision by diagnostic specialist (traceable)

- Combination of tests used guarantee the quality of the diagnoses; at least one of the tests is validated

- Validation/verification of tests: relevant performance criteria determined (fit for purpose)
### 3) Internal list (example)

<table>
<thead>
<tr>
<th>Order</th>
<th>Taxon</th>
<th>Stadium</th>
<th>Morphological</th>
<th>PCR</th>
<th>Sequence analysis</th>
<th>Data</th>
</tr>
</thead>
<tbody>
<tr>
<td>Thysanoptera</td>
<td><em>Thrips palmi</em></td>
<td>all stadia</td>
<td>THRPL_20151008_MOR</td>
<td>THRPL_20151008_PCR</td>
<td>THRPL_20151008_SEQ</td>
<td>20151008</td>
</tr>
<tr>
<td>Lepidoptera</td>
<td><em>Spodoptera</em></td>
<td>Adult; egg</td>
<td>SPODG_20151008_MOR</td>
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<td></td>
<td>20151008</td>
</tr>
<tr>
<td>Lepidoptera</td>
<td><em>Spodoptera eridania</em></td>
<td>all stadia</td>
<td>SPODG_20151008_MOR</td>
<td>SPODG_20151008_PCR</td>
<td>SPODG_20151008_SEQ</td>
<td>20151008</td>
</tr>
<tr>
<td>Lepidoptera</td>
<td><em>Spodoptera frugiperda</em></td>
<td>all stadia</td>
<td>SPODG_20151008_MOR</td>
<td>SPODG_20151008_PCR</td>
<td>SPODG_20151008_SEQ</td>
<td>20151008</td>
</tr>
<tr>
<td>Lepidoptera</td>
<td><em>Spodoptera littoralis</em></td>
<td>all stadia</td>
<td>SPODG_20151008_MOR</td>
<td>SPODG_20151008_PCR</td>
<td>SPODG_20151008_SEQ</td>
<td>20151008</td>
</tr>
<tr>
<td>Lepidoptera</td>
<td><em>Spodoptera litura</em></td>
<td>all stadia</td>
<td>SPODG_20151008_MOR</td>
<td>SPODG_20151008_PCR</td>
<td>SPODG_20151008_SEQ</td>
<td>20151008</td>
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<tr>
<td>Coleoptera</td>
<td><em>Monochamus galloprovincialis</em></td>
<td>Adult</td>
<td>MONCGA_20151008_MOR</td>
<td></td>
<td>MONCGA_20151008_SEQ</td>
<td>20151008</td>
</tr>
</tbody>
</table>
4) Assuring the quality

• **Method focussed assurance** (1st/2nd/3rd line)
  - Each test relevant 1st line controls
  - 2nd (blind samples) and 3rd line controls (Proficiency Testing) at method level

• **Process focussed assurance**
  - Individual diagnoses: combination of tests, multiple assessors or second opinion
  - Diagnostic process
    > ‘blind’ samples throughout the whole process

Quality assurance of low or infrequent #samples: possible
## Procedure for addition of new organisms/tests

<table>
<thead>
<tr>
<th>Situation</th>
<th>example</th>
<th>minimal demands:</th>
<th></th>
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</thead>
<tbody>
<tr>
<td>addition of a new method</td>
<td>Malditov</td>
<td>y</td>
<td>-</td>
<td>y</td>
<td>y</td>
<td>y</td>
<td>y</td>
<td>y</td>
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<tr>
<td>addition of a new organism group</td>
<td>Mollusca</td>
<td>y</td>
<td>-</td>
<td>y</td>
<td>y</td>
<td>y</td>
<td>y</td>
<td>y</td>
</tr>
<tr>
<td>addition of a new test in existing</td>
<td>primers x/y in real-time PCR</td>
<td>-</td>
<td>y</td>
<td>y</td>
<td>tbd</td>
<td>tbd</td>
<td>y</td>
<td>n</td>
</tr>
<tr>
<td>method</td>
<td></td>
<td></td>
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</tr>
<tr>
<td>addition of a new organism in existing</td>
<td>Potato virus X with real-time PCR</td>
<td>-</td>
<td>y</td>
<td>y</td>
<td>tbd</td>
<td>tbd</td>
<td>y</td>
<td>n</td>
</tr>
<tr>
<td>method/organism group</td>
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</tbody>
</table>
End of October 2015: audit on flexible scope

A) Reassessment Fixed scope
   -> no major remarks

B) Audit new Flexible scope
   -> best solution for our work
   -> morphological analysis
   -> scope/format internal list
   -> validation & internal audit
Experiences