# Subcommittee on Plant Health Diagnostics

## SPHD Reference Standard No. 2
## (SPHD RS No. 2)

# *Development of National Diagnostic Protocols*
# *- Procedures for Authors*

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|---|---|
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# CONTENTS

# 1. INTRODUCTION

## *1.1 Scope*

The *Development of National Diagnostic Protocols – Procedures for Authors* (SPHD RS No. 2) is a Subcommittee on Plant Health Diagnostics (SPHD) Reference Standard (RS) providing guidelines for authors on the development of diagnostic protocols (refer to SPHD RS No.1 for definitions). The Reference Standard has been developed to standardise and incorporate relevant information in diagnostic protocols for the identification of emergency plant pests in Australia.

## *1.2 Purpose*

The purpose of this Reference Standard is to provide guidelines and instructions to authors for the development of a diagnostic protocol. Once developed, the diagnostic protocol will be submitted to the Diagnostic Protocols Working Group (DPWG) for review and approval and SPHD for endorsement (with the authority of Plant Health Committee). Once endorsed, the document is recognised as a National Diagnostic Protocol (NDP) and published on the NPBDN website.

If an approved International Plant Protection Convention (IPPC) diagnostic protocol exists, this should be used unless it is shown that the NDP has improved procedures for Australian conditions.

## *1.3 Review*

The SPHD RS No. 2 will be reviewed every five years or earlier if required. The review will be implemented by DPWG (in consultation with SPHD). Changes to the Standard are subject to the endorsement of SPHD members for adoption.

All SPHD Reference Standards can be found on the NPBDN website (https://www.plantbiosecuritydiagnostics.net.au/). Click on the “Resources” tab, and then on the “National Diagnostic Protocols”.

## *1.4 References*

IPPC. 2021. ISPM No. 6 *Surveillance*. Food and Agriculture Organisation of the United Nations, Rome.

IPPC. 2021. ISPM No. 13 *Guidelines for the Notification of Non-compliance and Emergency Action*. Food and Agriculture Organisation for the United Nations, Rome.

IPPC. 2021. ISPM No. 27 *Diagnostic Protocols for Regulated Pests*. Food and Agriculture Organisation for the United Nations, Rome.

IPPC. 2011. *Procedural Manual.* Food and Agriculture Organisation of the United Nations, Rome. September 2011.

IPPC. 2024. ISPM No. 5 Glossary of Phytosanitary Terms. Food and Agriculture Organisation of the United Nations, Rome.

# 2. SPHD PROCEDURE FOR THE DEVELOPMENT OF NATIONAL DIAGNOSTIC PROTOCOLS

## *2.1 Introduction*

The SPHD Procedure for the Development of NDPs has been adapted from the IPPC Procedural Manual, Section 3.6.3. *Technical Panel to Develop Diagnostic Protocols for Specific Pests* (IPPC 2011).

Definitions of terms, acronyms and abbreviations used are contained in the SPHD RS No. 1: *Glossary of Terms* and *ISPM No. 5 Glossary of Phytosanitary Terms* (IPPC 2024).

An overview of the procedure for the development, review and verification, approval and endorsement phases of a NDP is provided in Figure 1.

## *2.2 Procedures*

a) A Subject Matter Expert (SME) approved by DPWG will lead the development of a diagnostic protocol either by adapting an existing diagnostic protocol or by developing a new diagnostic protocol as required. The author uses the Instructions to Authors for NDPs for guidance (SPHD RS No.2, Section 3) and if needed additional instructions may also be given by DPWG and SPHD.

b) A drafted diagnostic protocol should be submitted to the National Protocols Coordinator (NPC), who then checks that it meets the requirements set out here and looks for typos etc before submitting to DPWG for initial evaluation. The NPC can send the draft back to author for corrections before sending it to DPWG.

c) The diagnostic protocol is evaluated by DPWG and may be returned to the author if considered not to contain sufficient detail.

d) Peer review of the diagnostic protocol is undertaken by a SME approved by DPWG (in accordance with SPHD RS No. 4 *Guidelines for Peer Review, Verification and NDP Reviews*).

e) Verification of the diagnostic protocol is undertaken by an independent laboratory approved by DPWG (in accordance with SPHD RS No. 4). *

f) The diagnostic protocol and the associated peer review and verification reports are submitted to DPWG via the NPC for approval (in accordance with SPHD RS No. 3 *Guidelines for the development, Approval and Endorsement Processes of NDPs*), before being submitted to SPHD.

g) The diagnostic protocol is either endorsed by SPHD, or returned to the author, applicant or person nominated by DPWG with comments for further work required before resubmission.

h) Section-9 of the diagnostic protocol will be reviewed (a) along with the rest of the protocol if it is included at the initial drafting stage (b) or separately by the DPWG, if it is added later, after the protocol has already undergone the review and verification stage of the NDP workflow process (Figure 2).

i) Once endorsed, acknowledgment of the reviewer(s), verifying laboratory and original authors of the protocol are included with a citable International Standard Book Number (ISBN).

*An alternative procedure may be where the NDP is developed and/or updated as a part of a national research project and is reviewed and verified within the same project. In this case steps 1 and 4 and 5 are combined and the verification laboratory needs to be in another organisation and/or jurisdiction (refer to Figure 2).

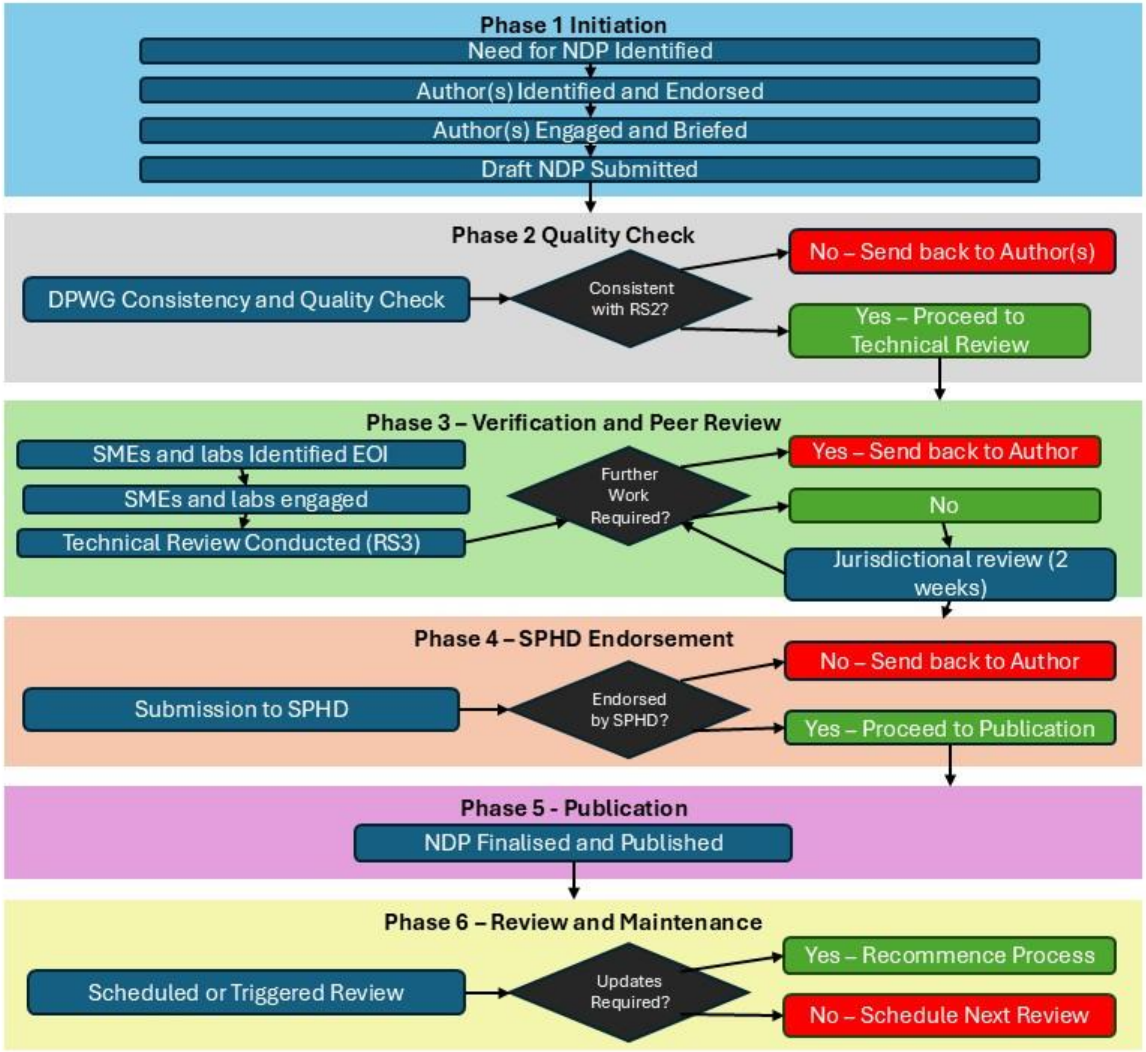


**Figure 1**: Overview of the National Diagnostic Protocol (NDP) six phase process.

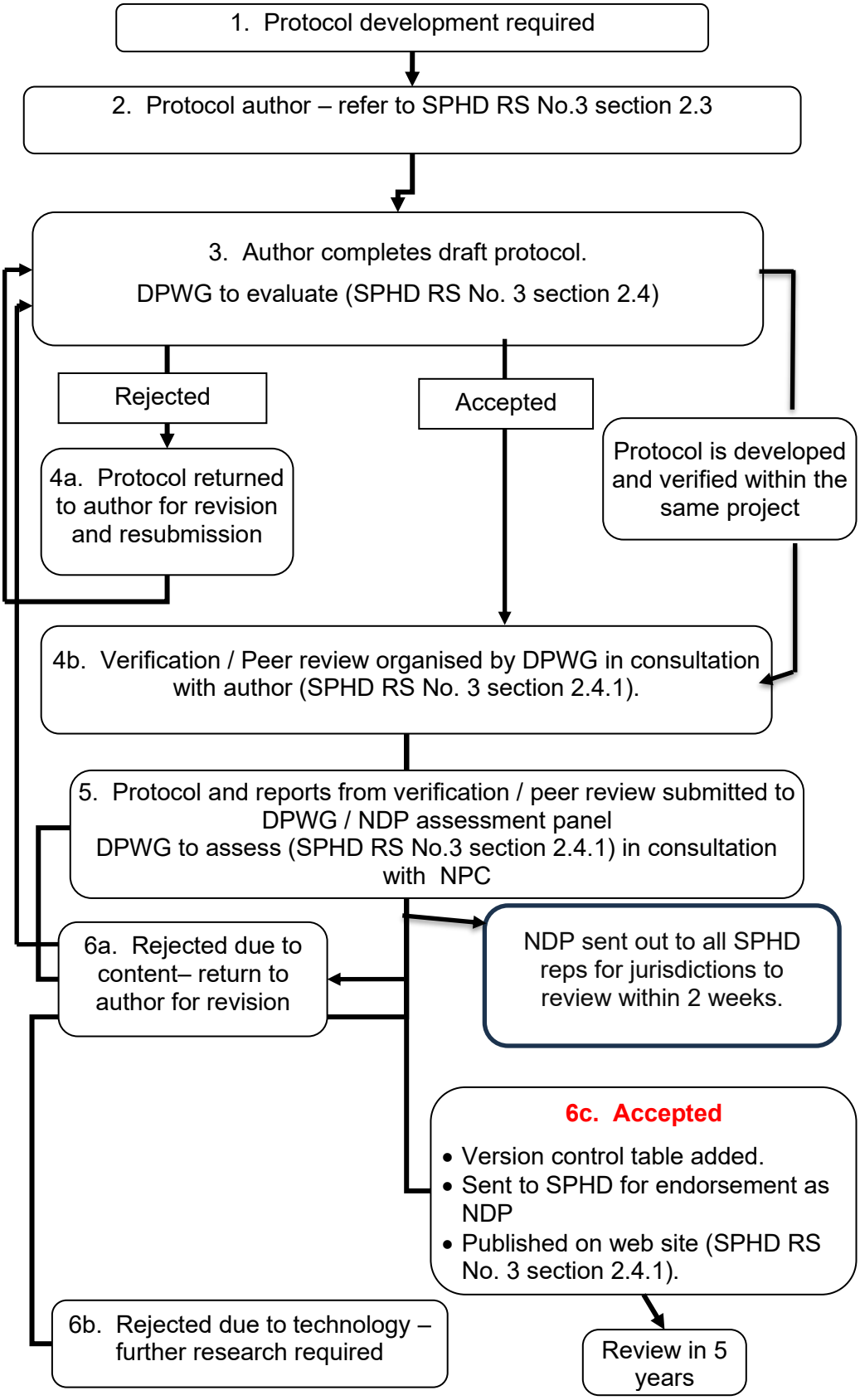


**Figure 2.** National Diagnostics Protocol (NDP) development flowchart.

# 3. INSTRUCTIONS TO AUTHORS OF NATIONAL DIAGNOSTIC PROTOCOLS

## *3.1 Introduction*

These instructions to authors of NDPs) are based on the ISPM No. 27 *Diagnostic Protocols for Regulated* Pests (IPPC 2021) and IPPC *Procedural Manual* section 3.6.3 (*Technical Panel to Develop Diagnostic Protocols for Specific Pests* (TPDP)) and Annex 11 (*TPDP Guidance, Part 1: Guidelines on Formatting of Diagnostic Protocols).*

Definitions of terms, acronyms and abbreviations used are contained in the SPHD RS No. 1: *Glossary of terms* and the *ISPM No. 5 Glossary on Phytosanitary Terms* (IPPC 2024).

Authors can use the proforma in Section 4 of this reference standard and also refer to NDPs on the NPBDN website https://www.plantbiosecuritydiagnostics.net.au/resources/ for examples.

Authors are encouraged, if appropriate, to publish their work in national and international journals prior to endorsement.

## *3.2 Instructions*

### 3.2.1 General requirements

***A protocol should only contain information directly relevant to the accurate taxonomic identification of the organism.***

National diagnostic protocols (NDPs) provide the minimum requirements for diagnostic procedures and methods for the detection and identification of plant pests. Information is provided on the pest, its host and taxonomic status and the methods to detect and identify it based on the best available information.

NDPs may cover a species, an intra-specific taxon, several species within a genus or multiple genera of related pests.

Authors must ensure that all text is appropriately cited and correctly acknowledged. Authors must not violate intellectual property or copyright conventions.

The document -should include page numbers and text font must be Cambria size 11 or Arial 11 (see *Style Guide Checklist for Authors* in Section 5).

### 3.2.2 Images and illustrations

Any images not copyright to authors **must** have written permission from the copyright owner, or be cited if from a published document, for use in the protocol.

All figures **must** be numbered, and the figure caption include the country or region where images were taken and name of the copyright holder.

Images and illustrations **must** be appropriate, in focus, and of sufficient resolution. The images, where possible, shall be made available to upload to Pest and Disease Image Library (PaDIL) https://www.padil.gov.au/.

- Format: for images use JPG or TIFF, for line art/graphics/logos can also use PNG.
- Resolution of images a minimum of 300 dpi at 100%.
- Resolution of line drawings a minimum of 600 dpi at 100%.
- Image size minimum of 500 KB, maximum 5Mb.
- Images in focus.
- Image files cropped as close to the actual image as possible.
- Images of high quality with good contrast of dark and light.
- Include figures in the serial order in the text.

- Include meaning of any symbol or abbreviation

**Note: the resolution will change if you enlarge the image from the original. For example, if you double the size the resolution will be reduced to 150dpi. Pictures can be resized in programs like photoshop to retain the accepted resolution.*

**Do not:**

- Supply files that are optimised for screen use (e.g., GIF, BMP, PICT, WPG); the resolution is below the acceptable minimum.
- Supply files below the minimum resolution.
- Supply files in which colours are not realistic, text is illegible, or images are pixilated.

Authors should follow the requirements outlined in the style guide checklist (Section 5).

#### 3.2.3 Definitions

*Pest detection* in a diagnostic protocol is the information needed to detect the pest in the host material so it can then be identified.

*Pest identification* in a diagnostic protocol is defined as the process of ascertaining the taxonomic identity of an organism.

#### 3.2.4 Methodology

Each protocol should contain the diagnostic procedures and guidance necessary for the named pest(s) to be detected and positively identified by a diagnostician. Authors should select diagnostic procedures based on their accuracy, sensitivity and reproducibility; also consider the availability of equipment, the expertise required for these procedures and their practicality (e.g., ease of use, speed and cost).

Diagnostic protocols should include more than one diagnostic procedure, if appropriate, to consider the varying capabilities of laboratories. Diagnosis of different developmental stages of organisms may require different methodologies. In cases where morphological procedures can be reliably used, but appropriate molecular procedures have been developed, the use of both procedures **must** be described.

Authors should provide information and guidance on diagnostic procedures that either singly or in combination lead to diagnosis of the pest. When several procedures are mentioned, their advantages/disadvantages should be given as well as the extent to which the procedures or combinations of procedures are equivalent. If several procedures are needed for the diagnosis, and/or if many alternative procedures are included, a schematic flow diagram may be necessary.

Only methods relevant for the definitive diagnosis should be included. Techniques suited to high throughput diagnostics (i.e., the ability to process large numbers of samples with fast turnaround time) which would support surveillance, should be included in Section 9 of the NDP.

#### 3.2.5 Structure and content of a diagnostic protocol

Diagnostic protocols should be arranged as per the protocol template provided with the following sections (with additional sub-headings as appropriate):

**Contents**

1. Introduction
2. Taxonomic information
3. Detection
4. Identification
5. Contact points for further information
6. Acknowledgements

7. References
8. Appendices (optional)
9. Sampling and diagnostic procedures to support surveillance

## 1. Introduction

This section should be no greater than 400 words.

Authors should provide brief information on the pest that is directly relevant for diagnosis and should be referenced. This information may include host range, effect on hosts and brief description of mode of transmission, dispersal and dissemination (e.g., vectors).

Host range should be entered in list format: for long hosts lists this can be included in the appendix.

No geographical information, control or risk assessment (entry, spread, establishment potential or consequences of entry) should be included.

Other relevant information can be included in the appendix, e.g., life cycle, full details of transmission, dispersal and dissemination.

Authors should include information on whether other protocols are available, reference them, and comment on why they are not suitable, or how they are related to this protocol.

***If an IPPC protocol is available, and the NDP has improved procedures, the NDP should be used in preference. The NDP should clearly state the reason, and the date of the IPPC diagnostic protocol being referenced.***

## 2. Taxonomic information

The correct scientific name and authority should be given and an overview of the relevant taxonomic hierarchy (Kingdom, Phylum, Class, Order, Family, Genus, Species, relevant subspecific taxon). Include synonyms and relevant former names (these may be taxonomically incorrect, but relevant in relation to the literature) as appropriate. Internationally recognised acronyms should be included.

For protocols that detect and identify groups of organisms (e.g., several species within a genus, several genera within a family), the scope of the protocol should be clearly defined. Where possible, the taxonomic rules used to define this group should be included as well as evidence via refereed papers that demonstrate the diagnostic capacity of the protocol. If appropriate, a list of organisms that have been detected and identified using the diagnostic protocol can be included in an appendix.

Any taxonomic disagreements should be stated along with appropriate references.

A taxonomic description can be included only if this information is not covered in detection.

## 3. Detection

This section contains the information needed to detect the pest so it can then be identified. It is NOT the procedures used for identification.

However, for some pests, particularly invertebrates, there may be significant cross-over of information. If the information for detection is also the method used for identification, place the method in the identification section.

Authors should provide information and guidance on:

- The part(s) of the plant, plant products or other articles on/in which it may be found.

- The likely occurrence of the pest associated with developmental stages of the host(s), climatic conditions and seasonality.
- The signs or symptoms associated with the pest including illustrations/images.
- Differences or similarities with signs and/or symptoms from other pests/causes including illustrations/images.
- The developmental stages of the pest that may be encountered, together with their likely distribution on/in the plants/plant products or other articles.
- Sampling procedures critical for the detection methods and the diagnostic procedures. Any aspects of the sample that may impact on the diagnostic procedure should be included, e.g., age of the sample, chemical sprays applied to the crop prior to sampling, sample storage (sampling procedures for surveillance should not be included).
- Detection procedures for extracting, recovering, and collecting the pest from the plants. These methods should be described in sufficient detail so that the user of the protocol will be able to apply the method without further reference to the literature.
- If the pest is vector borne, include available detection methods in the vector.
- High throughput sequencing (HTS) can be used as a diagnostic procedure. Guidelines/standards for its use was endorsed by SPHD in 2024 and available on NPBDN website.

Subheadings should be included as appropriate, but it is not necessary to include all dot points as subheadings.

Refer to section 3.2.2. for images and illustration.

Images for symptoms should:

- Show typical symptoms, including key characteristics such as necrosis, wilting, sporulation, bacterial ooze, fruiting bodies, etc.
- Clearly show the part of the plant exhibiting the symptom.
- Show correct composition and correct distance for the given symptoms.
- Be shown in context (closeup, macro, symptomatic plant vs. non symptomatic plant(s)).
- Include a point of reference such as an image scale, a coin or any other common object for size comparison, if not of a recognisable whole plant part.

## 4. Identification

In this section, authors should provide information and guidance on diagnostic procedures that either singly or in combination lead to the identification of the pest. ***The minimum requirements for identification must be clearly stated.*** Diagnostic procedures for quick, presumptive indications of identity (which will later need to be confirmed) may also be included. Each diagnostic procedure should be described separately.

For all the diagnostic procedures, information should be provided on their accuracy, specificity and reproducibility, and specifications from multi-laboratory validation trials (if available). For protocols that detect and identify more than one organism, information should be provided that clearly defines the scope of the protocol. If the list of organisms is extensive, this data can be presented as a table in an appendix. This table should include if relevant, key diagnostic characters to differentiate each organism.

Guidance should be provided on positive and negative controls and reference material to be included in each of the procedures. Sources and specifications (technical, commercial, collection entry codes) of controls and reference material should be indicated. Where the inclusion of additional specific controls, including reference material, is essential, this should be indicated in the protocol.

Guidance should be provided on the criteria for the determination of a positive or negative result for each method, and methods to resolve ambiguous results. Guidance should also be provided on resolving possible confusion with similar and related species or taxa.

Where appropriate, diagnostic procedures for detection of pests from asymptomatic plants or plant products should also be given.

Two main types of methodologies are included in diagnostic protocols: those which are based on morphological and morphometric characteristics and those based on biochemical or molecular properties.

**Methodologies based on morphological or morphometric characteristics of a pest.**

Details should be provided, as appropriate, on:

- Methods for extracting, recovering, and collecting the pest from the plants, plant products or other articles, including isolation and culture of the pest and duration of the process.
- Methods to prepare, mount and examine the pest (such as for light microscopy, electron microscopy).
- Identification keys (to family, genus, species and others), with references.
- Descriptions of the morphology of the pest, or of its colonies, including sex of the pest, illustrations of diagnostic characteristics, morphometric data and an indication of any difficulties in seeing particular structures.
- Comparison with similar or related species.
- Relevant reference specimens or cultures, if available.

Images/ illustrations for identification should:

- Be appropriately cited and correctly acknowledged. Authors must not violate intellectual property or copyright conventions.
- Include either macro or micro images to show appropriate diagnostic features used in identification and where necessary annotated with arrow, circle or similar to highlight the specific feature.
- Include scale bar or appropriate point of reference (coin, ruler, etc). Images/ illustrations types for invertebrate identification should also:
- Include images of the whole invertebrate and appropriate life stages.
- Include habitus (how the organism looks in the field) if not shown in symptoms images.
- If the NDP covers more than one species, show the relevant characters for each of the taxa being compared.

**Methodologies based on biochemical or molecular properties.**

- For biochemical or molecular identifications, each method should be described separately in sufficient detail (including equipment, reagents and consumables) to allow performing the diagnostic procedures without further reference to the literature. Where appropriate, reference may be made to methodology described in other diagnostic protocols annexed to this standard. Standard methods for sequencing do not need to be described.
- Where DNA extraction methods used are not standard methods, reasons should be given for their use over a standard kit extraction method. Details of kit methods should not be included unless they are modified. Any modification should be justified.
- Where gene sequences used to develop the tests are not referenced, information on their stability, the alignment from which they are developed, and the population sample used to develop them should be included.
- Where a protocol is based on species-specific or group-specific PCR amplification, sequencing of the amplicon is mandatory for confirmation of a suspected EPP. At least one reference sequence should be nominated and made available in a publicly available nucleic acid or amino acid database (e.g., GenBank) and the expected level (%) of intra-specific sequence variation should be provided. Where used, data from international sequence databases should be derived from appropriately validated specimens and accession numbers, or the appropriate reference should be included.
- Molecular demarcation criteria used for species, subspecies, pathovar or strain discrimination should be provided, according to the latest valid taxonomic classification.
- Where culturing or isolation are necessary components of these methods, details should be provided including the duration of the process if not previously included.

## 5. Contact points for further information

In this section, authors should provide contact details of institutes and individuals (Australian and/or international) with expertise on the pest(s), who may be consulted regarding any questions or for confirmatory diagnosis. Contact details for subject matter experts (SMEs) should include address, email and phone.

***The author must seek permission from the experts to include their details.***

## 6. Acknowledgements

The name and address of the SME, who wrote the first draft are included in this section, together with those of any others who made major contributions including reviewers and verifying laboratories (if not listed as authors). This section also covers additional contributions made to the NDP, such as funding, data, and image sources.

## 7. References

References cited in the protocol **must** be included in this section. Other references to scientific publications, published laboratory manuals and web sites can be included in a separate section if they directly relate to detection or identification.

Use referencing style from *Australasian Plant Pathology* or *Journal of Australian Entomology Society*.

## 8. Appendices

Appendices may be used to provide relevant information such as life cycle, full details of transmission, dispersal and dissemination.

## 9. Sampling and Diagnostic procedures to support surveillance

This section provides information on the sampling and near-field procedures used in the screening, detection or identification of plant pests in a surveillance situation.

The information is intended for use by surveillance practitioners to outline sampling requirements that support laboratory testing; however, it may also be used in the development of a surveillance plan. The near-field procedures included in this section are to be used to support surveillance activities and are NOT to be used as definitive diagnostics in an initial detection.

Section-9 should be a standalone document to provide guidance on the host symptoms and information about the sampling requirements. The section can include the near-field diagnostics procedures that are already described in the NDP for the initial detection and identification of plant pests.

A good example of a section 9 “sampling procedures to support surveillance” can be found in the NDP for Plum pox virus (https://www.plantbiosecuritydiagnostics.net.au/app/uploads/2020/12/NDP-2-Plum-pox-virus-V4.pdf).

The author should include the following information (in quotation marks) in section-9 of the NDP, where applicable.

“It should be noted that, for sampling requirements, surveillance practitioners should consult their state laboratory to discuss (1) the number of samples which can be received by the lab, (2) the expected time until results are available and (3) whether there is time or cost advantage in undertaking near-field diagnostics for triaging purposes. These parameters are laboratory specific depending on equipment, laboratory test methods, and laboratory resourcing. The sample processing time provided below for the near-field tests (section 9.3) are also intended as a general guideline only”.

**Structure and content**

Section 9 should be arranged as per the protocol template provided with the following sections (with additional sub-headings as appropriate):

**Contents**



### 9.1 Introduction

This section should be no greater than 400 words. It may include summary of the host range, symptoms and the near-field tests available for diagnostic purpose. For the host range, an appropriate reference source should be included if the list exceeds 10 species.

### 9.2 Sampling and symptoms

Authors should provide any information for sampling that is critical to the diagnosis. The aim is to provide information for the most useful sample for the diagnostician to run the required test.

If the sample requirements are specific to the test, they should be included in the test information. Any generic sampling information should be included here.

However, this information may also be used to develop the surveillance plan, so the language should be clear and understandable by non-specialists. Be brief, but be specific: for example, “wrap sample in paper”: should the paper be wet, damp, or dry? Paper towel or newspaper?

Sampling information may include:

- Type of sample, e.g., plant part, healthy vs diseased, live vs dead, adult vs juvenile, mid-branch vs early growth etc.
- What not to sample, e.g., no soil with sample.
- Size of the sample, e.g., number, weight, volume.
- When to sample, e.g., best time of the year, growth stage of the crop, persistence in absence of host.
- When not to sample, e.g., if surveillance is necessary at a non-ideal time of year, can we still sample.
- Sample integrity, e.g., whether specific sampling methods or surveillance hygiene procedures may damage the specimen and make it unsuitable for diagnosis.
- Preservation of sample, e.g., storage media, temperature, length of time sample can be left in storage, etc.
- Any specific requirements for transport of sample.
- Any information on hosts, e.g., definite non-hosts, symptomatic hosts, asymptomatic hosts, etc.
- Any data that the diagnosticians may need from surveillance teams to assist with the diagnostic procedure, e.g., host, stage of growth, number of leaves in sample, chemical applications that affect sample quality, etc.

Subheadings should be included as appropriate, but it is not necessary to include all dot points as subheadings.

It would be preferable if images are provided to illustrate the above. Refer to section 3.2.2.

Sampling information could be supplied in a table like illustrated below.

**Plant example:**

| Specification | Description |
|---|---|
| Sample type | Plant tissue that shows typical symptoms. Very important to include the interface between healthy and diseased tissue.<br>If present, include tissue with bacterial ooze.<br>For sampling asymptomatic plants, younger material including floral parts and shoots should be selected as they are more easily colonised. |
| What not to sample | Older non symptomatic plant tissue. |
| Sample size | X cm of tissue, y number of floral parts/shoots. |
| When to sample | The best time to sample is during the growing season when symptoms are most visible. This will depend on the kind of host and geographic location.<br>Ooze most likely to be present in the morning when air humidity is high and temperature low. |
| When not to sample | Sampling in winter on dormant plant material is difficult as cankers may not be visible. |
| Sample integrity | Sample on same day as dispatch, otherwise store in a cool place (preferably not a fridge as low temperatures can cause problems in isolating). |
| Sample preservation | Samples should be wrapped in dry paper towel in a plastic bag. Samples should be processed as soon as possible after collection but can be stored at ~8$^{o}$C for up to 14 days before processing. |
| Sample transport | Important to ensure temperature of samples remain at or below 20$^{o}$C during transport to laboratory. Ensure samples not in transit over the weekend. |
| Host specific sampling | There are no differences between hosts. |
| Additional sample information | |

**Invertebrate example:**

| Specification | Description |
|---|---|
| Sample type | Crawler life stage. |
| What not to sample | Above ground symptomatic leaf material. |
| Sample size | N/A |
| When to sample | Summer to early autumn (December-February). |
| When not to sample | Other times of the year. |
| Sample integrity | N/A |
| Sample preservation | Bottle of 80 % ethanol. |
| Sample transport | Place samples in an esky (or appropriate holding container). Transport as quickly as possible to the laboratory. |
| Host specific sampling | N/A – grapevine only host. |
| Additional sample information | |

### 9.3 Near-field tests

Near-field tests can be used to optimise sampling, reduce samples requiring laboratory testing or provide definitive diagnosis. Therefore, near-field tests do not necessarily need to achieve definitive identification of the pest.

Near-field tests can include image-based diagnostics (e.g., remote diagnostics, good photos), on-line tools and any other procedure that can inform sample triage.

If no near-field tests are available, authors should note this here.

Authors should provide information on the scope, sampling and types of near-field tests included. For each test, authors should provide guidance on:

- The best use of the near-field test.
- Number of samples able to be tested at one time.
- Time from receipt of sample to result under field situation.

This information could be presented in a table as illustrated below:

| **Test** | **Identification Confidence** | **Deployment** | **Required Time** | **Throughput** |
|---|---|---|---|---|
| Test 1 | Low (<90%) | Field | >1 day | Medium (100s) |
| Test 2 | Medium (90-99%) | Field/Laboratory | >1 hour | Medium (100s) |
| Test 3 | High (99%+) | Laboratory | >1 week | Low (10s) |
| Test 4 | High (99%+) | Laboratory | < 1 week | High (1000s) |

Author should also include the following text note (in quotation marks) “Please note, that regardless of the result of near-field testing, samples should be sent to the laboratory for confirmatory testing”.

An example of the above table included in the plum pox virus NDP is as follows.

**Table XX**: Methodology that maybe utilised for initial triage near-field of plum pox virus.

| **Method** | **Identification Confidence** | **Deployment** | **Required Time** | **Throughput (No. of samples)** |
|---|---|---|---|---|
| **Agdia Immunostrip® (9.3.1)** | Medium (90-99%) | Field | <1 h | Medium (10-25) |
| **Agdia AmplifyRP® Acceler8® (9.3.2)** | Medium (90-99%) | Field | <1 h | Medium (10-25) |

Where possible, any near-field tests should be verified by the user prior to use in the field.

Authors should provide the specificity and sensitivity of the tests, including percentage of false negatives/positives where available. Field tests should have been validated in the field and not be purely laboratory based. Tests should have internal controls.

Authors should provide interpretation of results including what samples from the field test should be sent to the laboratory for further confirmation.

If the near-field tests are commercially available kits, the source is required, and an indication of whether they require specific expertise to run.

# 4. PROFORMA FOR NATIONAL DIAGNOSTIC PROTOCOL.

## Instructions to Authors

- This template serves as a guideline to authors in the production of a national diagnostic protocol (NDP).
- A copy of the NDP protocol master template is to be obtained from the National Protocols Coordinator (NPC).
- Instructions from the SPHD Reference Standard No. 2: *Development of National Diagnostic Protocols –Procedures for Authors* are not repeated in this template. Authors should familiarise themselves with the requirements of Reference Standard No. 2 prior to completion of the report.
- Various components (headings) of the template may relate only to specific groups of plant pests or diseases. Authors may select or add headings/sub-headings relevant to their pest organism.
- Authors must ensure that all text and images are appropriately cited and correctly acknowledged. Authors **must** not violate intellectual property or copyright conventions. The country or region where images were taken **must** also be provided.
- Images and illustrations must be appropriate, in focus, and of sufficient resolution. Authors should follow the requirements outlined in the style guide (Section 5).
- Table of Contents **must** reflect the contents of the NDP.
- An electronic copy must be sent to the NPC Coordinator:

  *Email: NDPCoordinator@aff.gov.au*

***A protocol should only contain information directly relevant to the accurate taxonomic identification of the organism.***

# 5. STYLE GUIDE CHECKLIST FOR AUTHORS

When writing the NDP, the author should ensure the following style points are checked.

| | | |
|---|---|---|
| **Overall** | Font size minimum Cambria 11 or Arial 11. | |
| | Formatting with headings numbered. | |
| | Contents page generated from “Table of Contents”. | |
| | Pages numbered. | |
| | Appropriate image on title page. | |
| **Images** | Numbered, captioned, geography and copyright included.<br>E.g. “**Figure 1** Ringspot symptoms on tree in Hungary (Photo J. Bloggs)”<br>See further information in section 5.1 below. | |
| **Introduction** | Length <400 words. | |
| | No risk or geographic information. | |
| | Host range included as list. | |
| **Taxonomy** | Hierarchy included. | |
| **References** | Use style guide from *Australasian Plant Pathology* or *Journal of Australian Entomology Society*. | |

## *5.1 Images or illustrations used in NDPs*

- Format: for images use JPG or TIFF, for line art/graphics/logos can also use PNG.
- Resolution of images a minimum of 300 dpi at 100%.
- Resolution of line drawings a minimum of 600 dpi at 100%.
- Image size minimum of 500 KB, maximum 5Mb.
- Images in focus.
- Image files cropped as close to the actual image as possible. If this means a lot of cropping, do it in another program and save the cropped image at the required resolution.
- Images of high quality with good contrast of dark and light.

**Note: the resolution will change if you enlarge the image from the original. For example, if you double the size the resolution will be reduced to 150dpi.*

**Do not:**

- Supply files that are optimized for screen use (e.g., GIF, BMP, PICT, WPG); the resolution is too low.
- Supply files that are too low in resolution.
- Supply files in which colours are not realistic, text is illegible, or images are pixilated.

## *5.2 Inserting images or illustrations in word docs.*

*Please place images only as follows, otherwise editing is difficult.*

- Insert the image complete, position left and wrap: “In line with text”.
- Enter the caption underneath.

**Do not:**

- Put images in frames or tables. If you want the picture to end up in a table, enter a note in the table such as “image 1 here” and provide as a separate file (preferred naming convention: pest name_FigX.jpg).
- Put multiple images on one line (i.e. side by side).

**Web sites with useful information.**

- https://authorservices.wiley.com/asset/photos/electronic_artwork_guidelines.pdf
- https://www.escienceediting.org/journal/view.php?number=15