PM 7/151 (1) Considerations for the use of high throughput sequencing in plant health diagnostics

EPPO Workshop for Heads of Laboratories 19/20th April 2023

EPPO diagnostic team

Charlotte Trontin

Françoise Petter
Valérie Grimault
Madeleine McMullen
Baldissera Giovanni



Why an EPPO Standard on HTS?

2019

Recommendation on: Preparing to use high -throughput sequencing (HTS) technologies as a diagnostic tool for phytosanitary purposes

https://www.ippc.int/static/media/files/publication/en/2019/05/R_08_En_2019-05-06_Post-CPM-14.pdf



Why an EPPO Standard on HTS?

2019

Recommendation on: Preparing to use high -throughput sequencing (HTS) technologies as a diagnostic tool for phytosanitary purposes

https://www.ippc.int/static/media/files/publication/en/2019/05/R_08_En_2019-05-06_Post-CPM-14.pdf

RECOMMENDATIONS

The Commission notes that there are existing challenges and further work is needed on HTS technologies for pest detection and identification as the basis for applying phytosanitary regulations. Findings based on HTS technologies of an unknown microorganism need to be further investigated to demonstrate the potential of that microorganism to be a pest that would qualify as a regulated pest.

Before a contracting party proposes to use HTS technologies and their results as the basis for appropriate phytosanitary regulations, the Commission *encourages* contracting parties to:

- (a) establish guidelines on what phytosanitary actions, if necessary including pest risk analysis, should be taken after detection of an unknown organism (e.g. fungi, bacteria or virus) or detection of non-viable organisms in plant material
- (b) ensure that appropriate infrastructure and investments in Information Technology and bioinformatics, and education and training on bioinformatics, are in place for the appropriate data storage and interpretation of test results, and that there is effective implementation of these technologies
- (c) standardise and apply best-practice operational guidelines for HTS, including proper interpretation of results and quality control measures (e.g. procedure controls) that ensure HTS data outputs are robust and accurate, have biological significance in a phytosanitary context, and are implemented in a harmonized way
- (d) validate the reliability and accuracy of HTS by conducting trials comparing HTS against other existing diagnostic platforms
- (e) communicate information on the interpretation of HTS results, especially regarding conclusions about the phytosanitary risk of organisms detected, to the NPPO of the exporting country

PM 7/151 (1) Considerations for the use of high throughput sequencing in plant health diagnostics



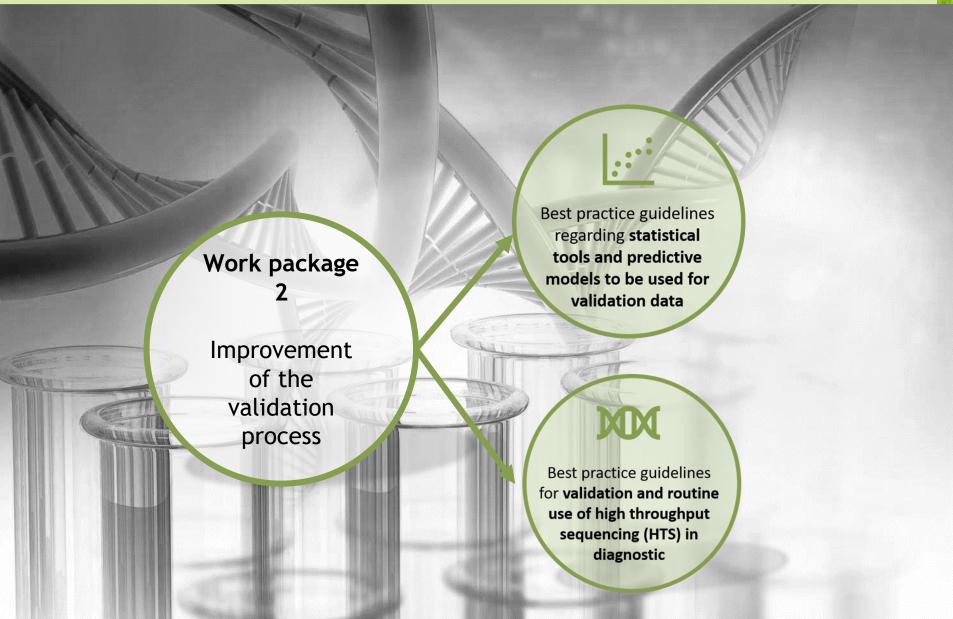
Validation of diagnostic tests to support plant health

- H2020 EU funded project (2018-2021)
- Consortium of 16 partners (research institutes, NPPOs, diagnostic companies, EPPO)
- 9 workpackages





Work package 2



MM

Best practice guidelines for validation and routine use of high throughput sequencing (HTS) in diagnostic



Based on literature review

Reviewed by VALISTEST partners

alitest



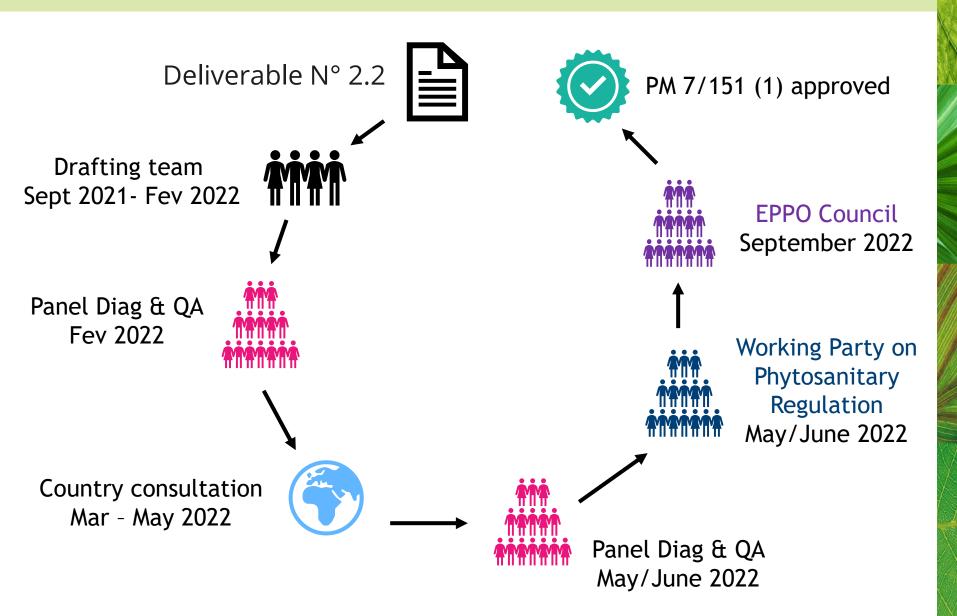
Reviewed by an international panel of 50+ experts from 18 countries

PM 7/151 (1) Considerations for the use of high throughput sequencing in plant health diagnostics

Outcome of alitest

- DELIVERABLE N° 2.2: "Best practice" guidelines for validation and routine use of non-targeted techniques in diagnostic setting which could serve as a basis for a new EPPO Standard
- Lebas et al. Facilitating the adoption of high-throughput sequencing technologies as a plant pest diagnostic test in laboratories: A step-by-step description. (2022) EPPO Bulletin, 52, 394–418. Available from: https://doi.org/10.1111/epp.12863
- Massart, et al. Guidelines for the reliable use of high throughput sequencing technologies to detect plant pathogens and pests. (2022) Peer Community Journal, Volume 2, article no. e62. Available from:
 - https://doi.org/10.24072/pcjournal.181
- Webinars (recordings available on EPPO youtube at: https://youtube.com/playlist?list=PLoVf4Pt04Db4aCrCOzZ33QMzDEa1eMtYZ

PM 7/151 (1) Considerations for the use of high throughput sequencing in plant health diagnostics

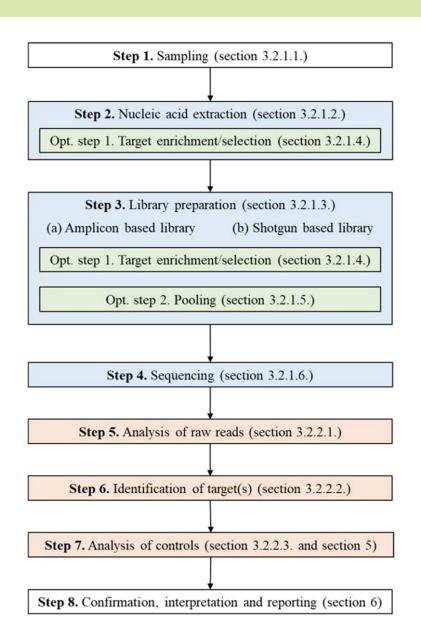


Specific scope of PM 7/151

- This Standard describes elements to take into consideration for the use of high throughput sequencing (HTS) tests, including
 - validation,
 - quality control measures and
- interpretation and reporting of results to ensure HTS test results are robust and accurate, have biological significance in a phytosanitary context, and are implemented in a harmonized way.
- This Standard applies to all plant pest groups and HTS technologies.

Technical content of PM 7/151

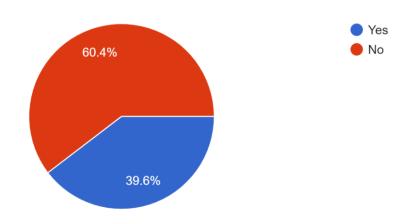
- 1. Introduction
- 2. Definition
- 3. Technical requirements to perform HTS tests
 - 1. General requirements
 - 2. Step specific requirements
- Validation and verification of HTS tests
- 5. Ensuring the validity of HTS test results
- 6. Confirmation, biological interpretation and reporting



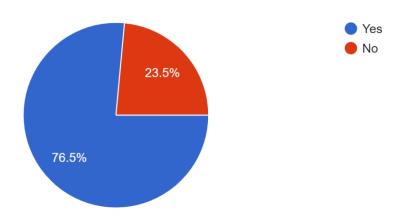
The use of HTS in plant health: next steps for EPPO

- Workshop for Heads of Laboratories 2023 NOW!
- EPPO and Euphresco involved in the organisation of sessions on HTS at the next ICPP meeting
- Use of HTS in plant pest diagnostics is a new developing area, consequently the standard will be revised in 2024 based on experience following its use in laboratories until this date.
- The Panel on diagnostics in virology is investigating the possibility to prepare a Diagnostic Protocol on virus detection and identification using HTS.
- Need for other disciplines?

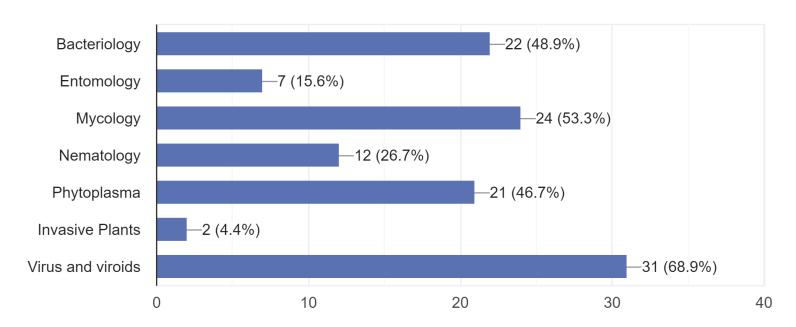
1. Do you use HTS in your laboratory? 53 responses



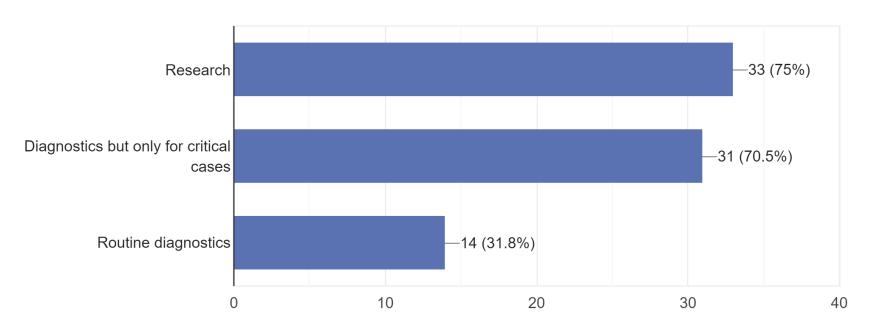
2. If you do not use HTS, do you intend to use it in the future? 34 responses



3. If you answered yes to 1) or 2), for which discipline / intended use do you use or plan to use HTS? 45 responses

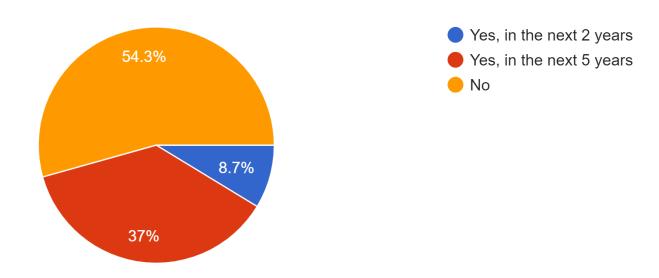


4. If you answered yes to 1) or 2), do you, or will you, use it for ? 44 responses



6. Are you planning to request ISO 17025 accreditation for HTS tests?

46 responses



Thank you for your attention!





The content of this presentation represents the views of the author only and is his/her sole responsibility; it cannot be considered to reflect the views of the European Commission and/or the Research Executive Agency or any other body of the European Union. The European Commission and the Agency do not accept any responsibility for use that may be made of the information it contains.