



Food and Agriculture  
Organization of the  
United Nations



International  
Plant Protection  
Convention



Department  
for Environment  
Food & Rural Affairs

Six millions of SARS-Cov-2 genomes sequenced...

How can plant health scientists transfer high throughput sequencing technologies toward plant pest diagnostics ?

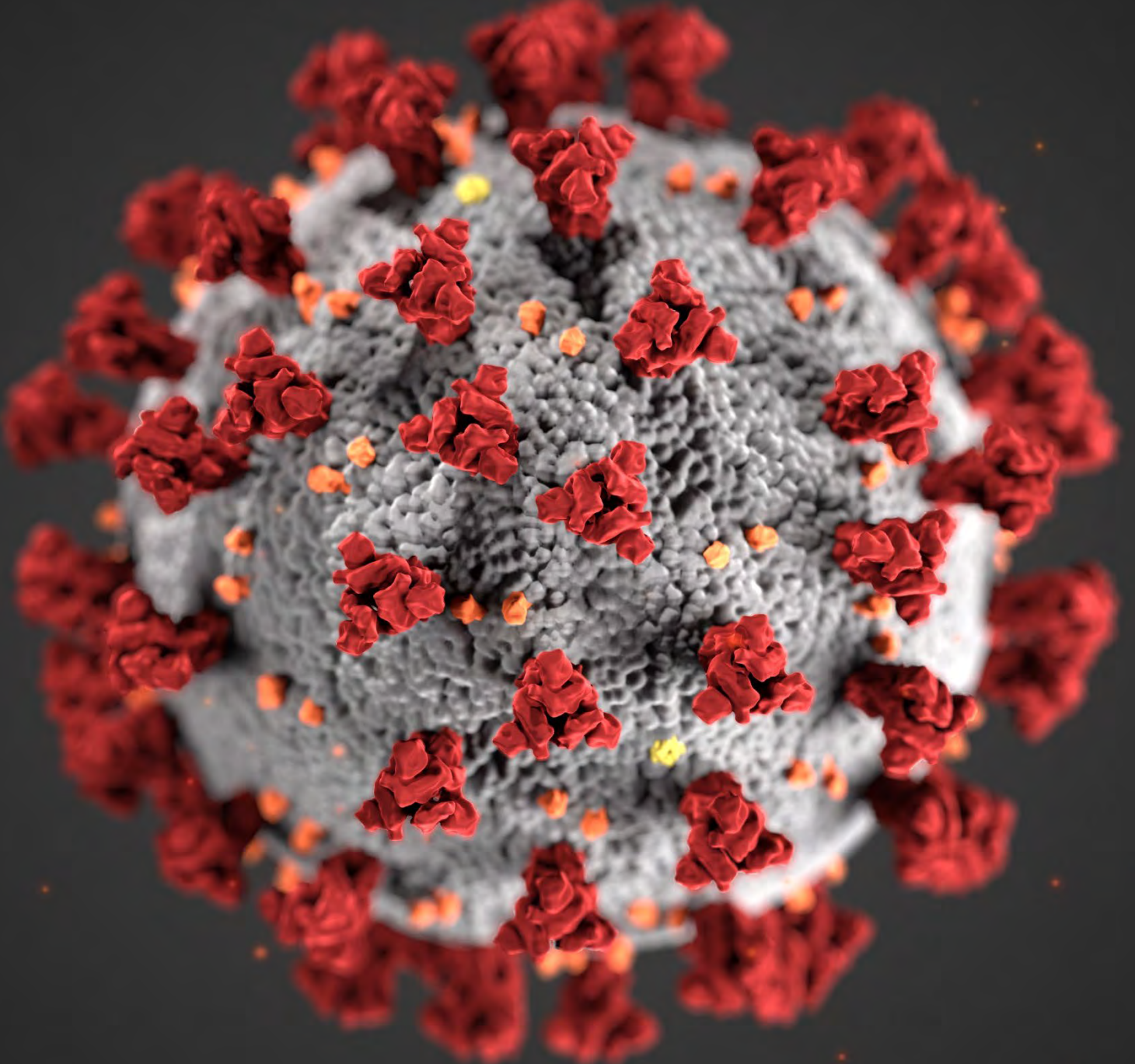
Prof. Sébastien Massart – Liège University – Gembloux AgroBio Tech - Belgium

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London, 21 – 23 September 2022

**International Plant Health Conference**







# NCBI SARS-CoV-2 Resources

## Quick Navigation Guide

- Sequence Submission
- Literature
- Sequence-Related Resources

## SARS-CoV-2 Data

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## Diagnostics for SARS-CoV-2 infections

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Wang, X. et al. *Nature* 602, 691–695 (2022).  
DOI: 10.1038/s41586-022-03441-1  
Published online 7 February 2022  
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### Sequence-Related Resources

**ClinicalTrials.gov**

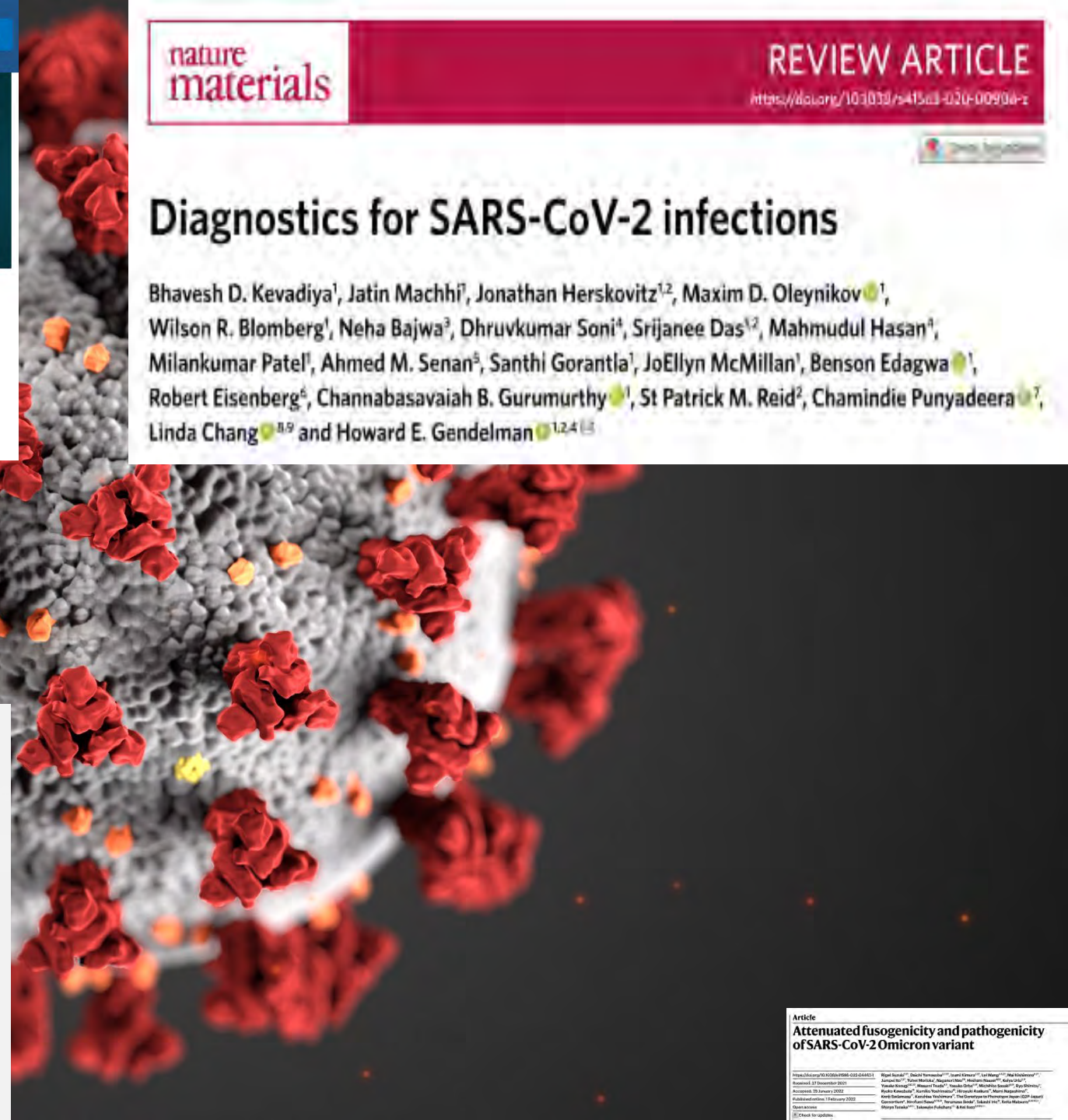
Bhaves D. Kevadiya<sup>1</sup>, Jatin Machhi<sup>1</sup>, Jonathan Herskovitz<sup>1,2</sup>, Maxim D. Oleynikov<sup>1</sup>,  
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# Resources

[ClinicalTrials.gov](https://clinicaltrials.gov)



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## Article

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<https://doi.org/10.1038/s41586-022-04462-1>

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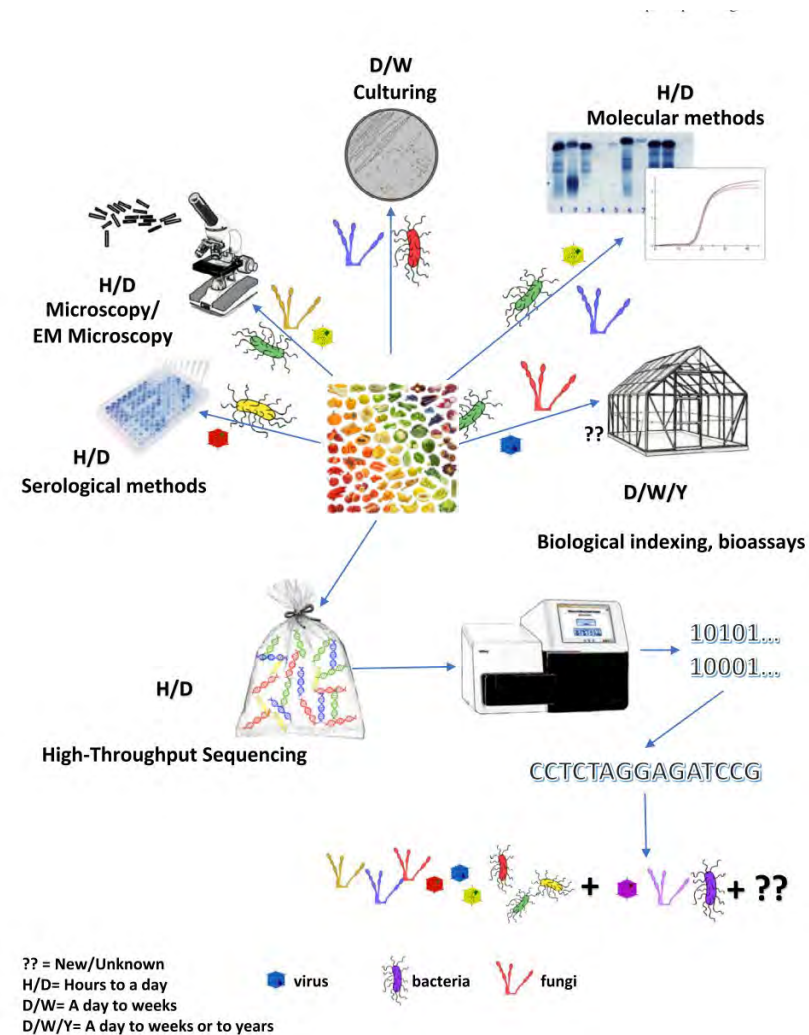
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Rigel Suzuki<sup>1,7</sup>, Daichi Yamashita<sup>2,3,7</sup>, Izumi Kimura<sup>2,3,7</sup>, Lei Wang<sup>1,4,7</sup>, Mai Kishimoto<sup>6,7</sup>, Junpei Ito<sup>2,7</sup>, Yuhei Morioka<sup>1</sup>, Naganori Nao<sup>1,8</sup>, Hesham Nasser<sup>1,9</sup>, Kelya Uriu<sup>1,10</sup>, Yusuke Kosugi<sup>2,12,13</sup>, Masumi Tsuda<sup>4,5</sup>, Yasuko Orbe<sup>6,12</sup>, Michihito Sasak<sup>1,12,13</sup>, Ryo Shimizu<sup>11</sup>, Ryoko Kawabata<sup>13</sup>, Kumiko Yoshimatsu<sup>10</sup>, Hiroyuki Asakura<sup>17</sup>, Mami Nagashima<sup>17</sup>, Kenji Sadamasu<sup>17</sup>, Kazuhisa Yoshimura<sup>17</sup>, The Genotype to Phenotype Japan (G2P-Japan) Consortium\*, Hirofumi Sawa<sup>6,7,14</sup>, Terumasa Ikeda<sup>8</sup>, Takashi Irie<sup>15</sup>, Keita Matsuno<sup>1,14</sup>, Shinya Tanaka<sup>1,15</sup>, Takasuke Fukuhara<sup>1,15</sup> & Kei Sato<sup>1,15</sup>

# What about us ?





# What about us ?

What about plant health and its stakeholders ?

**Transfer toward diagnostics :**

Eur J Plant Pathol  
<https://doi.org/10.1007/s10658-018-1570-0>



SI: PLANT PATHOLOGY FOR INNOVATIVE AGROECOLOGY

## The impact of high throughput sequencing on plant health diagnostics

Ian P. Adams • Adrian Fox • Neil Boonham • Sébastien Massart •  
Kris De Jonghe

2018



Source: <https://genetideiteracyproject.org>



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2022



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**Transfer toward diagnostics :**





# From research to diagnostics : technical verification or validation

INVITED REVIEW  Free Access

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## Evaluation of sensitivity and specificity in RNA-Seq-based detection of grapevine viral pathogens

Gabriele Di Gaspero <sup>a</sup>, Slobodanka Radovic <sup>b</sup>, Elisa De Luca <sup>c</sup>, Alessandro Spadotto <sup>b</sup>, Gabriele Magris <sup>d</sup>, Luigi Falginella <sup>c</sup>, Federica Cattonaro <sup>b</sup>, Fabio Marroni <sup>d,e</sup>

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RESEARCH

Open Access

## Towards the validation of high-throughput sequencing (HTS) for routine plant virus diagnostics: measurement of variation linked to HTS detection of citrus viruses and viroids





Rachelle Bester<sup>1</sup>, Glynnis Cook<sup>1</sup>, Igharim E. J. Breytenbach<sup>2</sup>, Chanel Steyn<sup>3</sup>, Rochelle De Bruyn<sup>1,4</sup> and Hans J. Maréchal<sup>1,\*</sup> 

## High-Throughput Sequencing of Small RNAs for the Sanitary Certification of Viruses in Grapevine

Leonardo Velasco<sup>1\*</sup> and Carlos V. Padilla<sup>2</sup>



REVIEW

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Alexander M. Piper <sup>1,2,\*</sup>, Jana Batovska <sup>1,2</sup>, Noel O.I. Cogan<sup>1,2</sup>, John Weiss<sup>1</sup>, John Paul Cunningham <sup>1</sup>, Brendan C. Rodoni<sup>1,2</sup> and Mark J. Blacket <sup>1</sup>

Article

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Nourolah Soltani <sup>1,†</sup> , Kristian A. Stevens <sup>2,3,4,†</sup>, Vicki Klaassen <sup>2</sup>, Min-Sook Hwang <sup>2</sup>, Deborah A. Golino <sup>1</sup> and Maher Al Rwahnih <sup>1,\*</sup> 









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# Writing international guidelines for using HTS to detect plant pests



**Guidelines for the selection, development, validation and routine use of high-throughput sequencing analysis in plant diagnostic laboratories**

Coordination: [Lebas B.](#), [Massart S.](#)

Contributors: [Adams I.](#), [Al Rwahnih M.](#), [Baeyen S.](#), [Bilodeau G.](#), [Blouin A.](#), [Boonham N.](#), [Bruinsma M.](#), [Candresse T.](#), [Chandelier A.](#), [De Jonghe K.](#), [Fox A.](#), [Gentit P.](#), [Gaafar Y.](#), [Haegemans A.](#), [Ho W.](#), [Hurtado-Gonzales O.](#), [Kutnjak D.](#), [Jonkers W.](#), [Kreuze J.](#), [Landa B.](#), [Leite Vicente C.](#), [Liefting L.](#), [Liu M.](#), [Maclot F.](#), [Malapi-Wight M.](#), [Maree H.](#), [Martoni F.](#), [Mehle N.](#), [Minafra A.](#), [Mollov D.](#), [Moreira A.](#), [Nakhla M.](#), [Petter F.](#), [Piper A.](#), [Ponchart J.](#), [Rae R.](#), [Remenant JP.](#), [Rivera Y.](#), [Rodoni B.](#), [Roenhorst A.](#), [Rollin J.](#), [Saldarelli P.](#), [Santala J.](#), [Souza-Richards R.](#), [Spadaro D.](#), [Studholme D.](#), [Sultmanis S.](#), [van der Vlugt R.](#), [Tamisier L.](#), [Trontin C.](#), [Wetzel T.](#), [Ziebell H.](#)



*This project has received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement N° 773139*



55 co-authors

>1,500  
revisions



Any pest

Any  
technology



# Writing international guidelines :

First publication: « building the house »

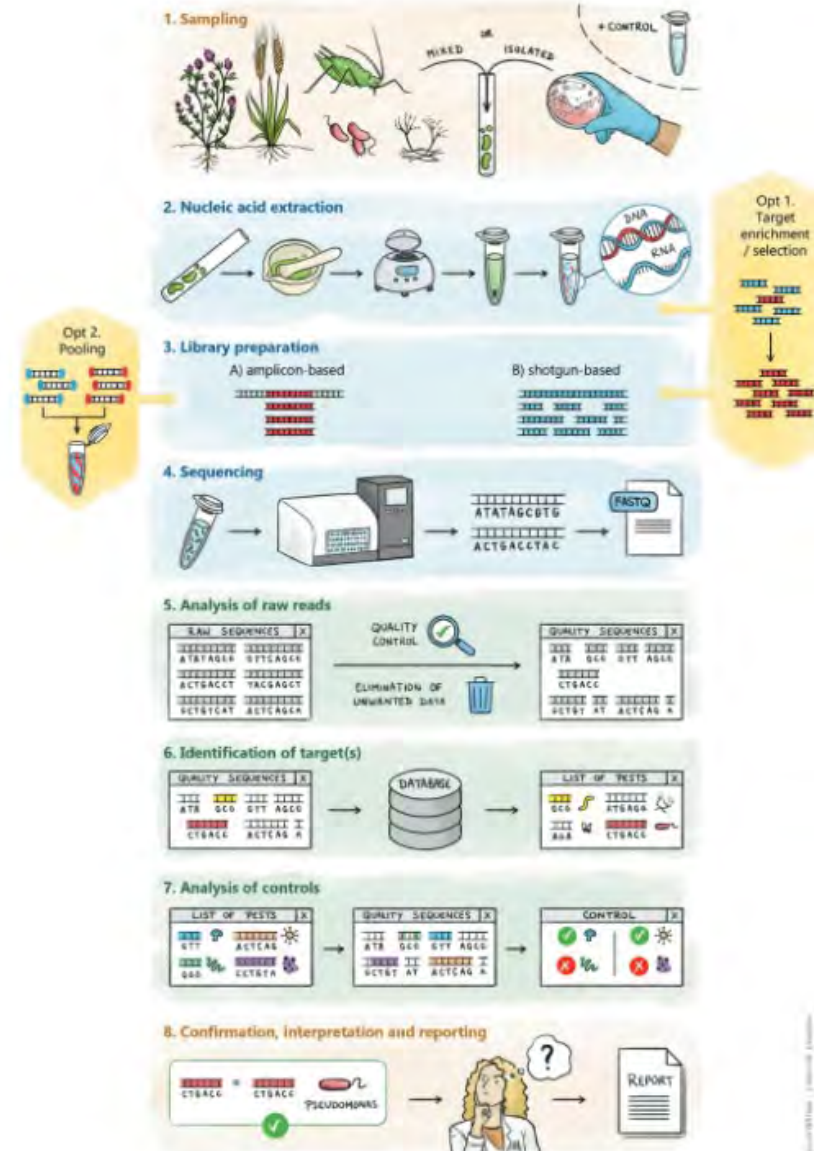


ORIGINAL ARTICLE | [Free Access](#)

## Facilitating the adoption of high-throughput sequencing technologies as a plant pest diagnostic test in laboratories: A step-by-step description

Benedicte Lebas, Ian Adams, Maher Al Rwahnih, Steve Baeyen, Guillaume J. Bilodeau, Arnaud G. Blouin, Neil Boonham, Thierry Candresse, Anne Chandelier, Kris De Jonghe, Adrian Fox, Yahya Z. A. Gaafar, Pascal Gentit, Annelies Haegeman, Wellcome Ho, Oscar Hurtado-Gonzales, Wilfried Jonkers, Jan Kreuze, Denis Kutnjak, Blanca Landa, Mingxin Liu, François Maclot, Martha Malapi-Wight, Hano J. Maree, Francesco Martoni, Natasha Mehle, Angelantonio Minafra, Dimitre Mollov, Adriana Moreira, Mark Nakhla, Françoise Petter, Alexander M. Piper, Julien Ponchart, Robbie Rae, Benoit Remenant, Yazmin Rivera, Brendan Rodoni, Johanna W. Roenhorst, Johan Rollin, Pasquale Saldarelli, Johanna Santala, Rose Souza-Richards, Davide Spadaro, David J. Studholme, Stefanie Sultmanis, René van der Vlugt, Lucie Tamisier, Charlotte Trontin, Ines Vazquez-Iglesias, Claudia S. L. Vicente, Bart T. L. H. Vossenbergh, Thierry Wetzels, Heiko Ziebell, Sebastien Massart ✉ ... [See fewer authors](#) ^

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# Writing international guidelines :

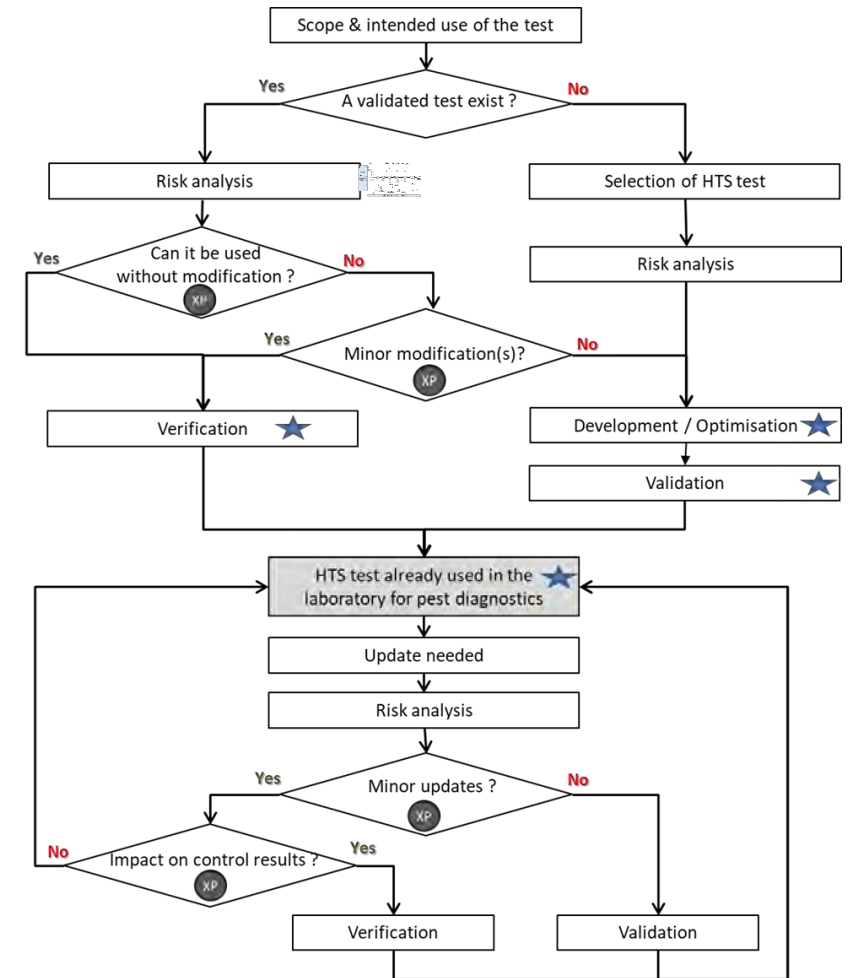
Second publication: « running the tests»



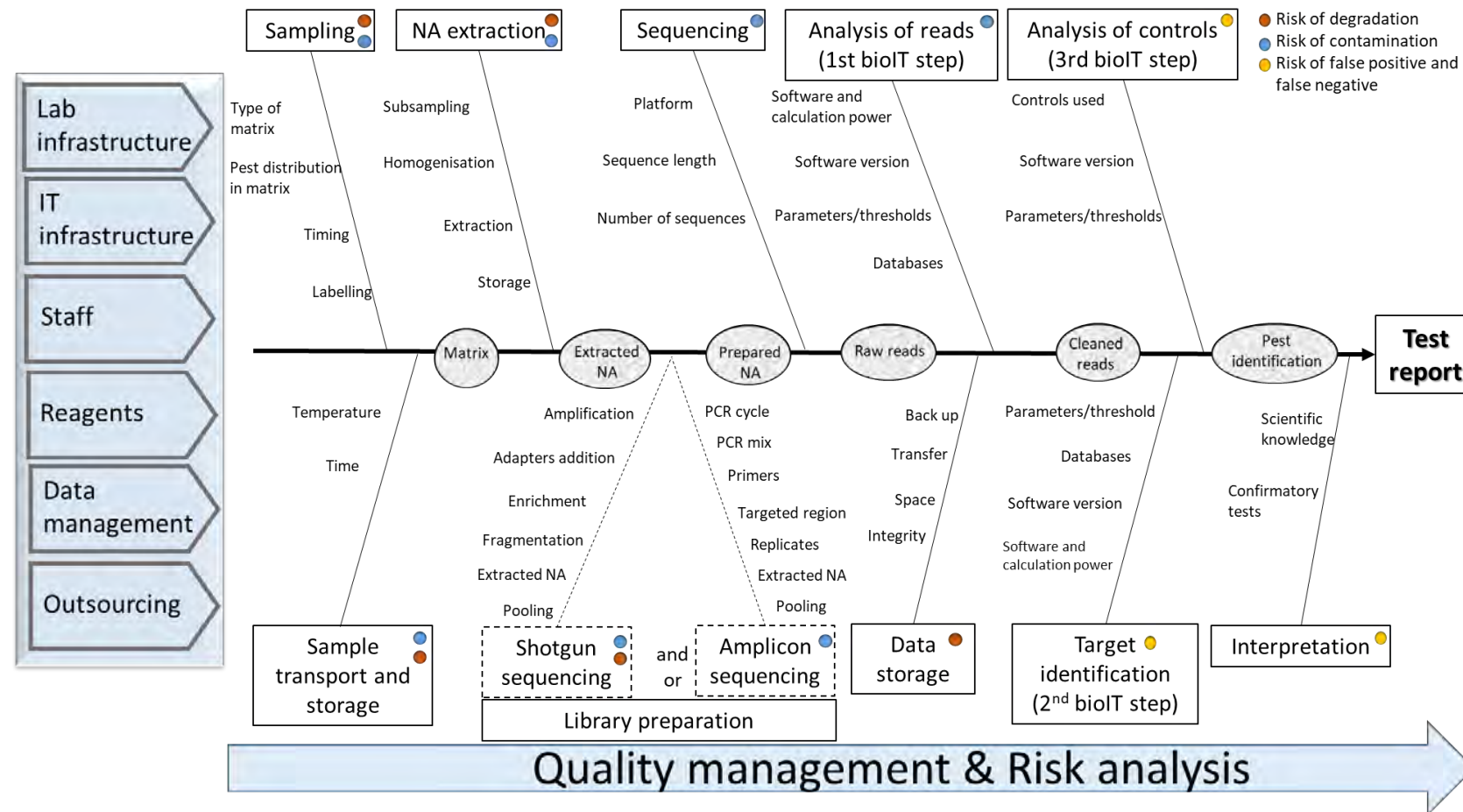
RESEARCH ARTICLE



**Guidelines for the reliable use of high throughput sequencing technologies to detect plant pathogens and pests**

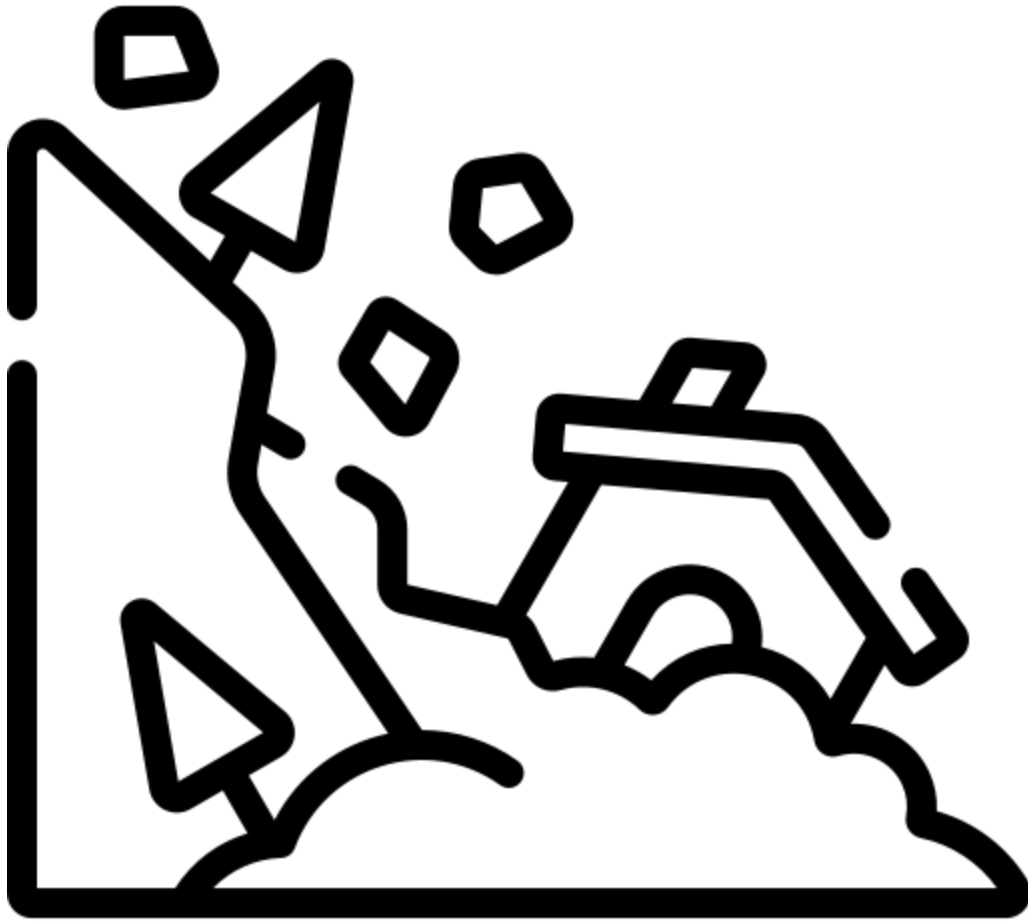


# Writing international guidelines :

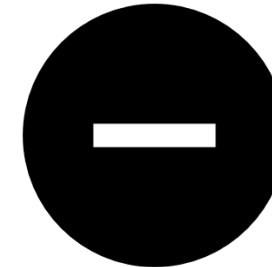
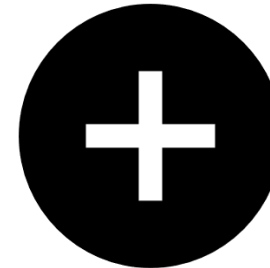




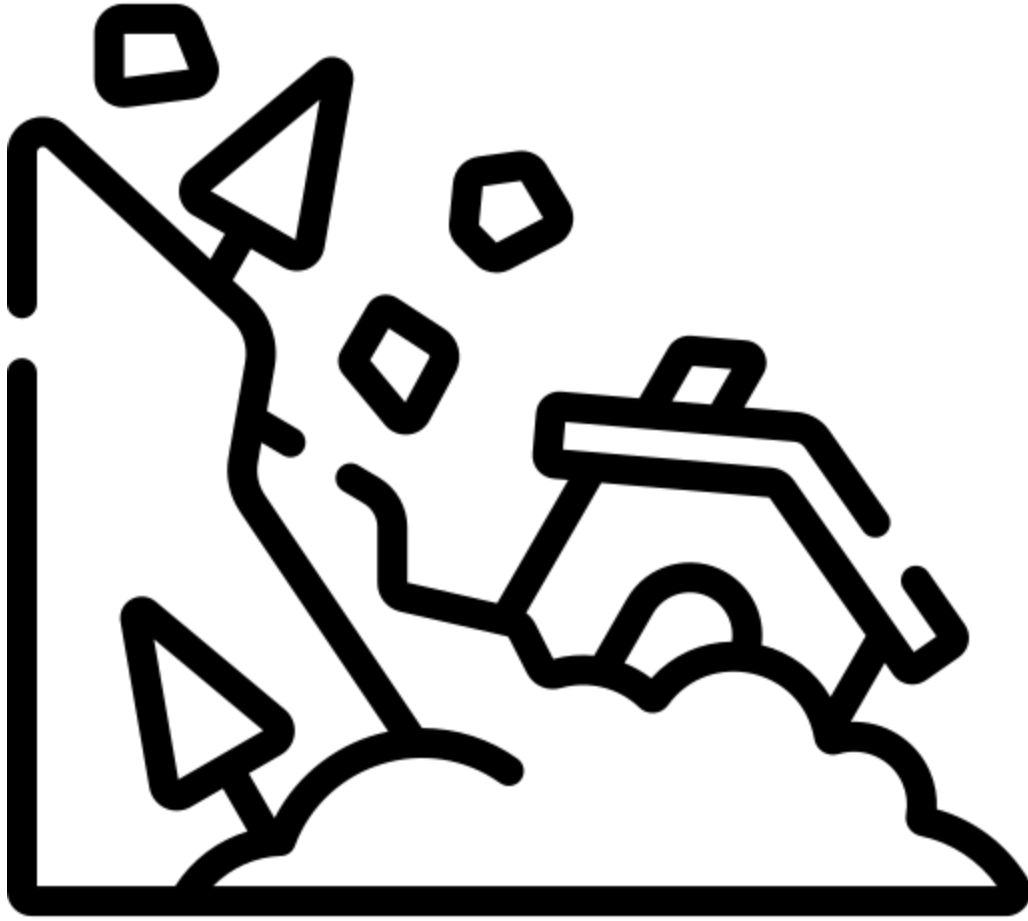
## Writing international guidelines :



Landslide on external control ?

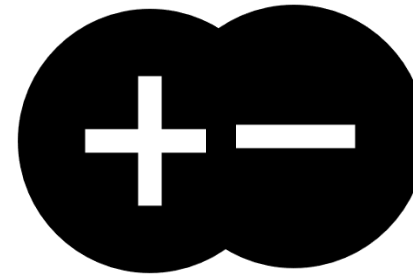


## Writing international guidelines :



### Landslide on external control ?

- Positive control also negative control
- Negative control often useless
- New alien control !





# From guidelines to an official standard :



**COUNTRY CONSULTATION – Deadline 2022-05-05**

**European and Mediterranean Plant Protection Organization**  
**Organisation Européenne et Méditerranéenne pour la Protection des Plantes**

**22-27406**

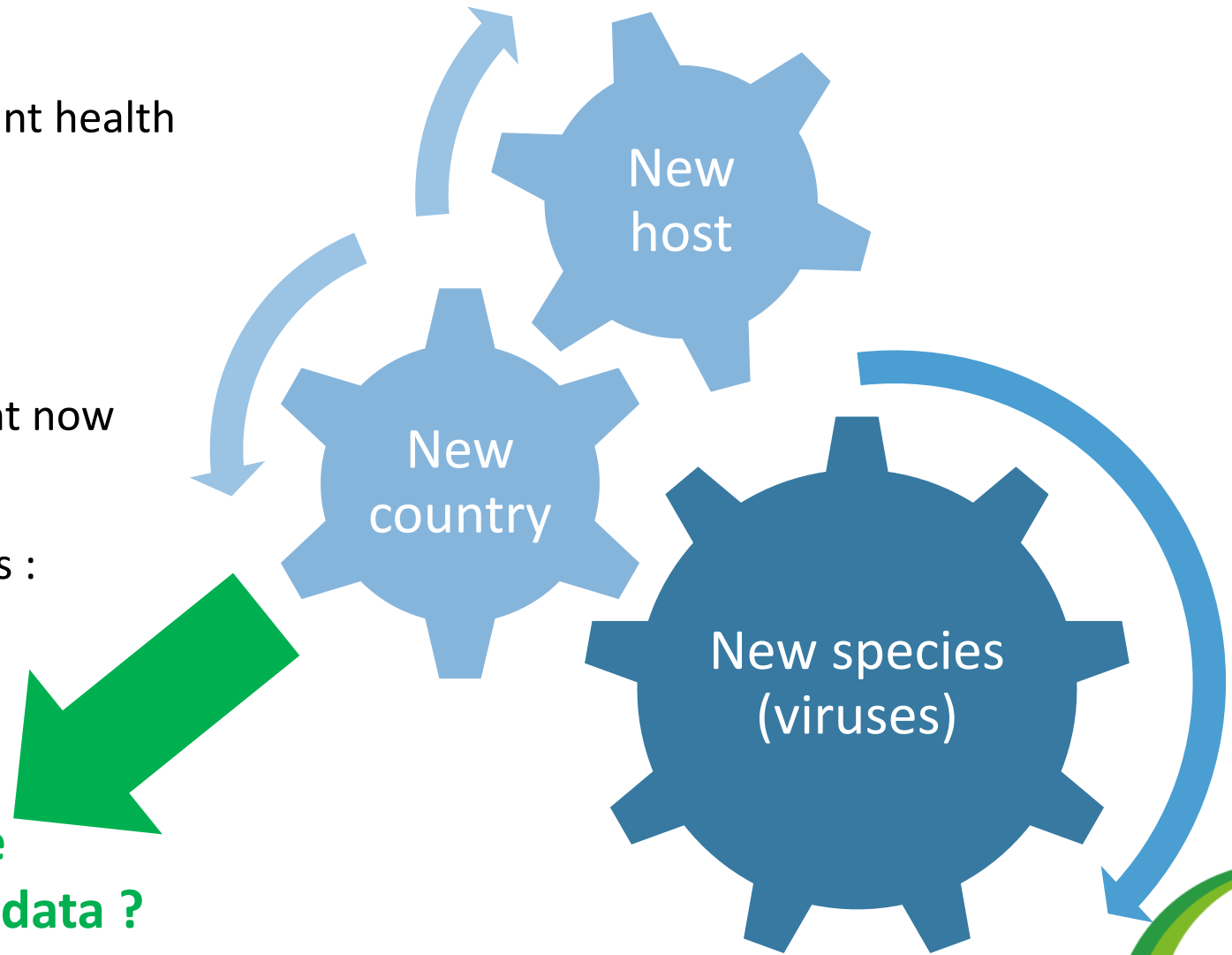
**Diagnostics**

**PM 7/XX**

**PM 7/XX Considerations for the use of High Throughput Sequencing in plant health diagnostics**

## Conclusion & challenges

- From research to diagnostics in plant health
- Guidelines to guide this transition
- ISO17025 accredited HTS tests right now
- Scientific and regulatory challenges :



**How to deal with the  
information and share the data ?**





Food and Agriculture  
Organization of the  
United Nations



International  
Plant Protection  
Convention



Department  
for Environment  
Food & Rural Affairs

Thank you for your attention  
and let's exchange on this !

London, 21 – 23  
September 2022

**International  
Plant Health  
Conference**

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**Sebastien Massart**

*Professor – Liège University – Gembloux AgroBio Tech -Belgium*

