Current impact and future direction of High Throughput Sequencing in plant virus diagnostics

Sébastien Massart

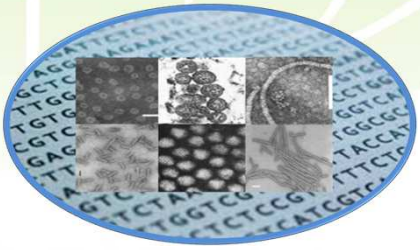
Laboratoire de Phytopathologie – Gembloux Agro-Bio Tech
Université de Liège – Belgique

sebastien.massart@ulg.ac.be

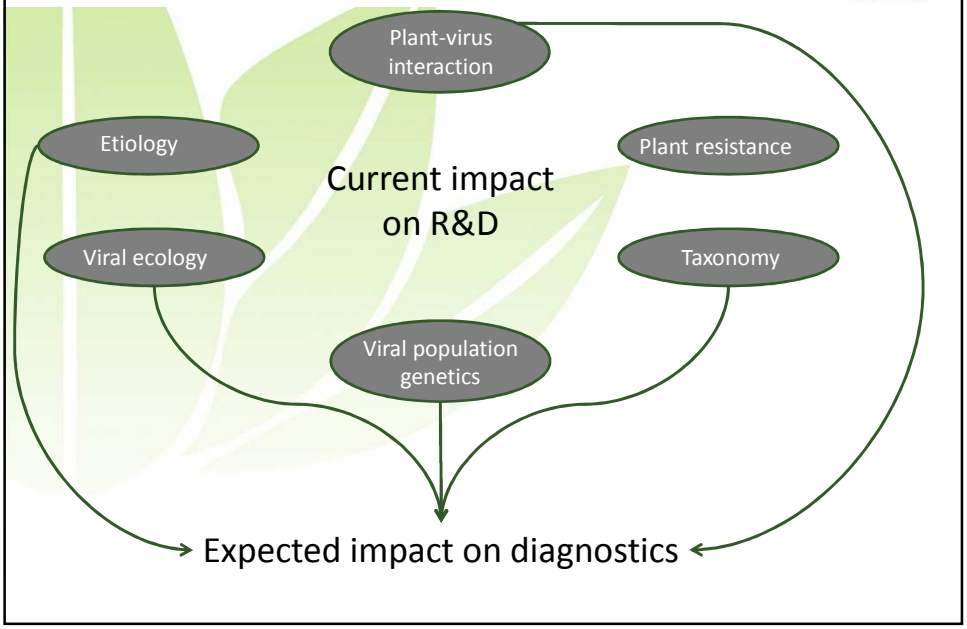
,Antonio Olmos, Haissam Jijakli and Thierry Candresse

Gembloux Agro-Bio Tech
Université de Liège

Driving factors



NGS & Viruses

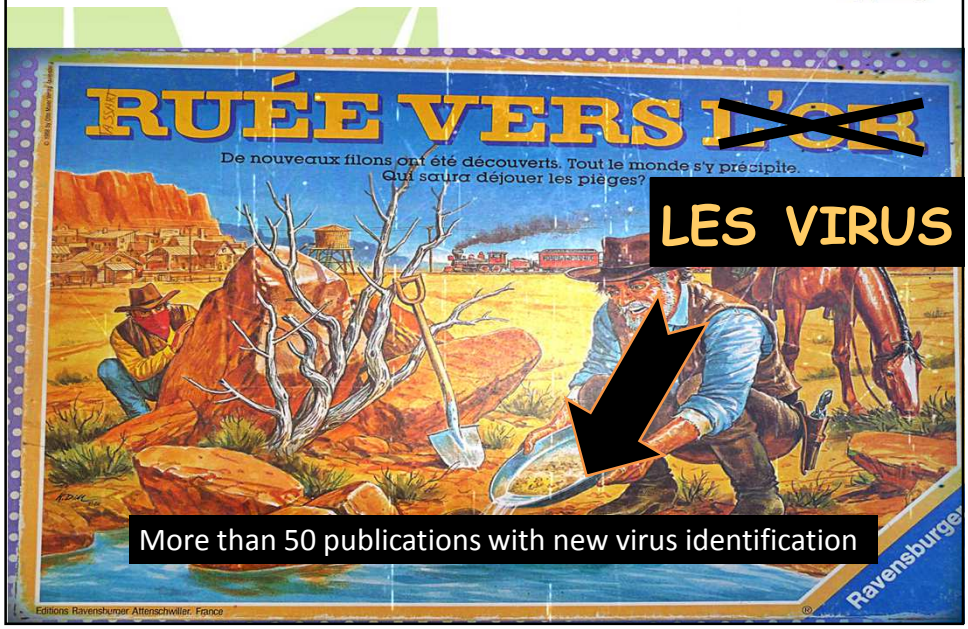


Diagnostic & Etiology

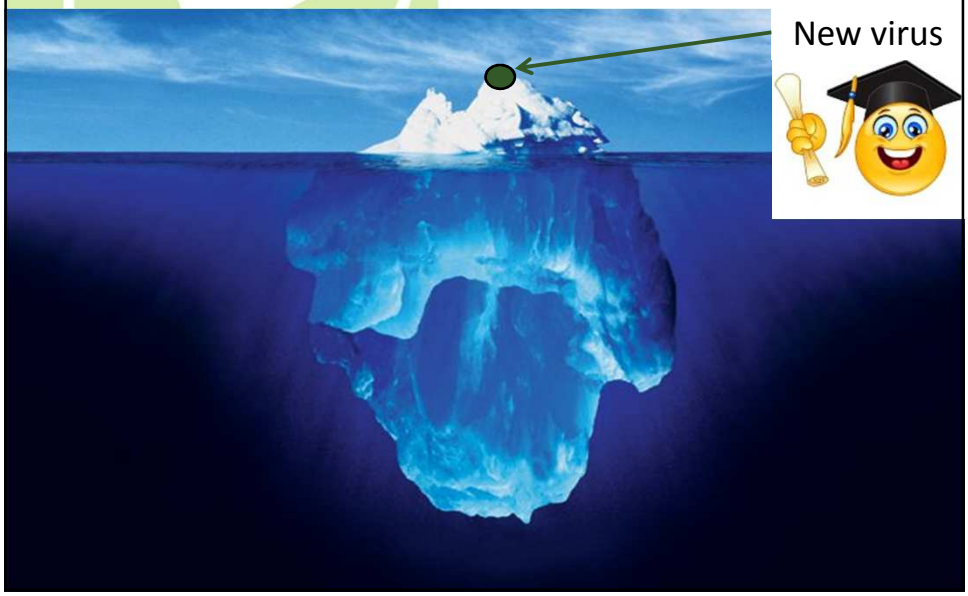
Before NGS

With NGS

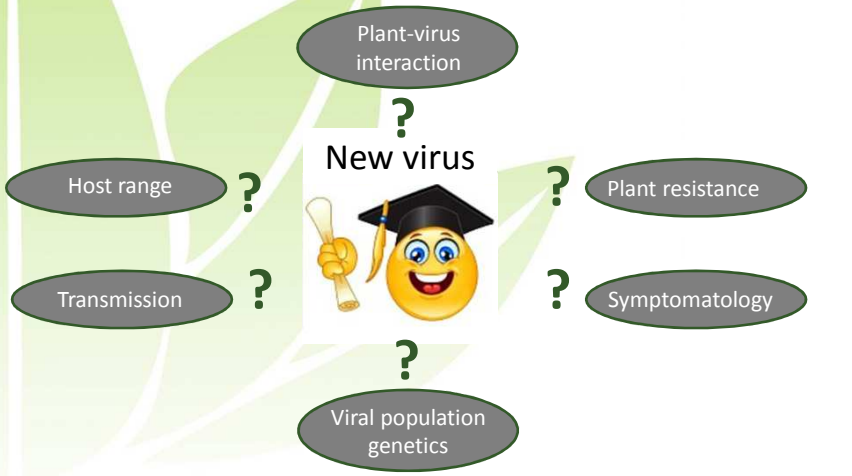
Diagnostic & Etiology



Diagnostic & Etiology



Diagnostic & Etiology



? Threat to the plants & control measures ?

Diagnostic & NGS

Certification, sanitation and quarantine

Field diagnostic and eradication

CLEAN UPSTREAM ⟷ **IMPROVE DOWNSTREAM**

Improved and standardized NGS methodologies for indexing

Deluge of sequences to accelerate and improve rapid-testing

Technical Challenges

1. Preparation & sequencing strategies ?

Extraction protocol
DNA, total RNA, particle isolation, small RNA, dsRNA

2. Sensitivity ?

Dependent on sequencing depth
Similar to PCR
What about a single or a couple of viral reads ?

3. Reproducibility and repeatability ?

Intra and inter-laboratory comparison

Technical Challenges

4. Contaminations ?

Human & beef contigs -> what about viruses ?

Partial or total genome coverage

Strict confinement needed !

5. Adapatation to host species ?

Virus titre and extraction method

→ Practical and technical challenges requiring both fundamental and applied research

Practical impact

Identification of a new virus or a poorly characterized virus by a diagnostician ?

Halt the distribution of the plant material ?

Confirmatory assays (Molecular Koch postulate) ?

Long-term experiment on virus biology ?

→ Technical framework and decision schemes are needed

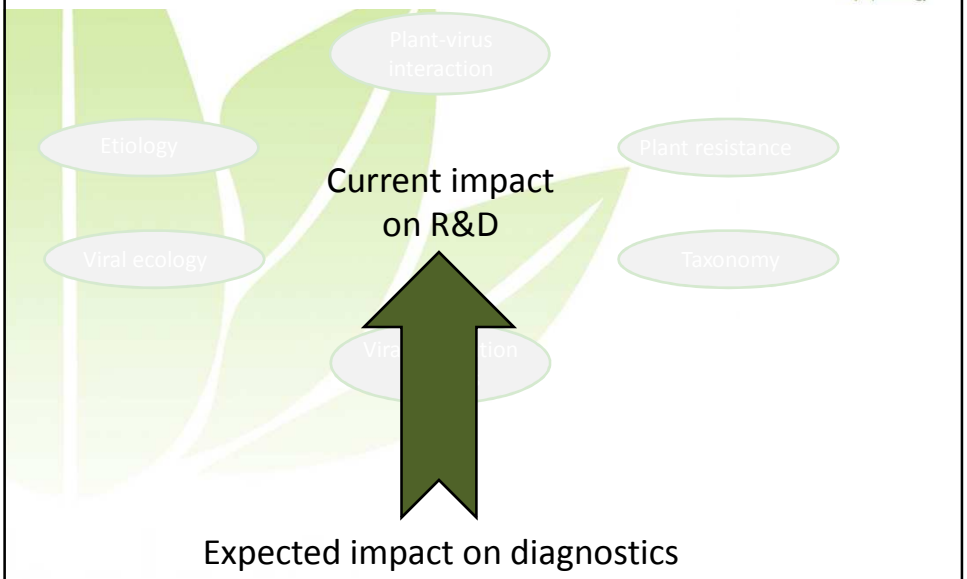
Latent (or symbiotic)viruses discovered ?

Probability of discovering non pathogenic viruses is high

Re-think trophic relationships ?

→ Adaptation of regulatory framework ?

NGS & Viruses



COST Action FA1407

Applied virologists

Fundamental virologists

Diagnostician

Coordinate and raise the European capacity to apply NGS technologies for the **study and diagnosis of viral diseases** in vegetatively propagated plants, seeds and seedlings.

Industries

NPPO/EPPO

Regulatory authorities

cost
EUROPEAN COOPERATION IN SCIENCE AND TECHNOLOGY

COST Action FA1407

NGS FOR THE STUDY AND DIAGNOSIS OF PLANT VIRAL DISEASES IN AGRICULTURE

- WG1 Development of protocols for NGS
- WG2 Etiology and biological impact of new viruses
- WG3 Taxonomy and viral population genetics
- WG4 Regulatory and socio-economic impact

1. Technological improvement
2. New scientific Knowledge
3. Societal innovations

Societal innovations

Conclusion

- ✓ Large impact in R&D
- ✓ Impact ongoing in diagnostic
  - ❖ Scientific & regulatory consequences
  - ❖ Feedback loop to fundamental research
- ✓ Long term :
  - ❖ Safer movement and trade
  - ❖ Better understanding of the virus threat and biology (including the Pathovirome)

Merci – Thanks

For more information:

Massart et al., 2014. Virus Research, 188: 90-96
Massart et al., 2014 Virologie, 18(5):247-250