18th EAPR Virology Triennal Meeting

Agroscope

Nyon
Switzerland
1-5 June 2025

Program and Abstract Book

eapr
european association for potato research

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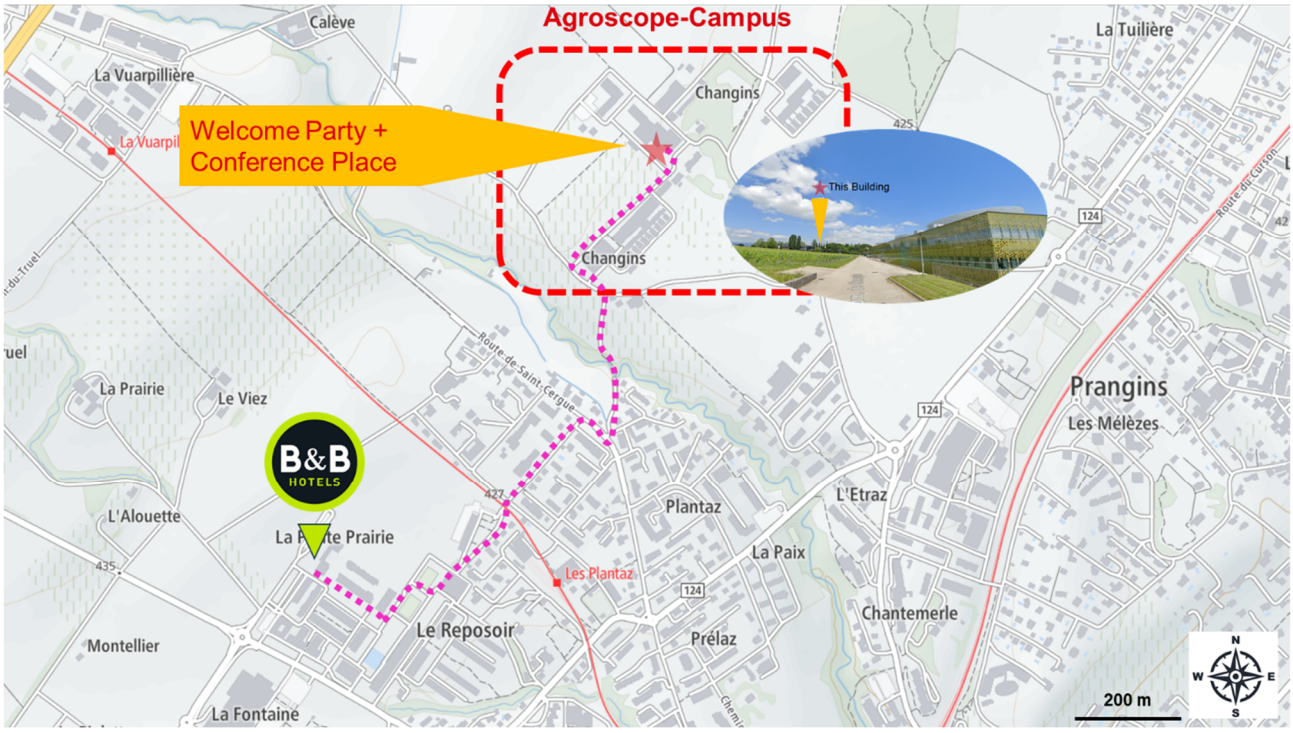

- The conference is a 10-15 minute walk from the conference venue.
  - Guided departure every day 30 minutes before the start of the presentations.
- Shuttle organized every day, 30 minutes before the start of presentations.
- Monday shuttle: 7:45 a.m.

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Keynote speakers

Session "Epidemiology in the context of climate change"

Dr Piotr Trębicki is a researcher at Macquarie University in Sydney, specialising in plant virology and entomology. His research focuses on plant-pathogen-insect vector interactions and the impact of climate change on these relationships. He is particularly interested in how environmental factors such as increased CO_2 and rising temperatures affect virus transmission by insects, especially aphids. Dr Trębicki is a member of the International Committee on Plant Virus Epidemiology, representing Australasia and Oceania. His work contributes to a better understanding of plant disease dynamics in the context of climate change, providing essential insights for the development of pest and disease management strategies.

Session "Stolbur and Arsenophonus in potatoes"

Professor Dr. rer. nat. Michael Kube from the University of Hohenheim in Stuttgart, Germany, specialises in plant pathology, with a focus on Stolbur phytoplasma, an important pathogen of crops such as potatoes and grapevines. His research investigates its transmission by insect vectors, its molecular interactions with host plants and environmental factors that influence disease severity. He has also studied Candidatus Arsenophonus phytopathogenicus and developed advanced PCR detection methods to track its impact on sugar beet and potato crops. His work combines bacteriology, molecular biology and bioinformatics to understand pathogen evolution and virulence.

Session "Vector-Plant interactions"

Dr Patricia Sanches is a postdoctoral researcher in the group of Prof. Consuelo De Moraes at ETH Zurich. Her work focuses on plant–insect–microbe interactions, particularly the influence of endosymbiotic bacteria on insect behavior and plant responses. By exploring how microbial partners shape virus - vector dynamics, her research offers new perspectives on disease ecology in crops. We are delighted to welcome her as an invited speaker in the virus–vector interaction session.

Session "Diagnostic and detection methods"

Dr Adrian Fox is Plant Virologist at Fera Science Ltd, the national reference laboratory for plant health in England and Wales. With over 25 years of experience in regulatory plant virus diagnostics, he began his career identifying virus vectors at Science and Advice for Scottish Agriculture (SASA) in Edinburgh. In 2008, he joined Fera Science Ltd in York as a senior virologist. His team applies innovative diagnostic techniques to improve plant virus detection and conducts research on the epidemiology and management of emerging viruses affecting vegetable crops and greenhouse production. Dr. Fox has co-authored more than 100 publications, including research articles, reviews, book chapters, and disease reports. As a member of the expert panel on Diagnostics in Virology and Phytoplasmology of the European and Mediterranean Plant Protection Organization (EPPO), he contributes to developing diagnostic standards.

PROGRAM SUNDAY, JUNE 1

18:00 21:30 Welcome Party

Drinks / Raclette Dinner

PROGRAM MONDAY, JUNE 2

08:00 08:30 Registration

08:30 08:55 Opening addresses

Chair: Olivier Schumpp

00:05 Alain Gaume: Welcome speech

00:05 Roberto Miglino: Introductive talk

00:15 Christian Bucher: From Field to Fork: Overviewof the Swiss Potato Sector

08:55 10:25 Epidemiology under climate pressure: emerging patterns and vector dynamics

Chair: Brice Dupuis

00:45 **Keynote 1 - Piotr Trębicki** : Adapting to a changing climate: challenges and strategies for controlling plant viruses and their vectors

00:20 Martin Verbeek: Epidemiology of PVY infections, environmental influences on the interactions between plant, vector and virus

00:15 Adrian Fox: Investigating the epidemiology of 'Candidatus Liberibacter solanaceraum' in great britain

00:10 Mohamad Chikh-Ali: Novel approach to study the dynamics of potato virus Y infection pressure –Case study: Colorado, USA

10:25 10:45 Coffee break ----------

10:45 12:25 Stolbur and arsenophonus in potato: epidemiology, impact and diagnostic challenges

Chair: Mout De Vrieze

00:45 **Keynote 2 - Michael Kube**: Stolbur, phytoplasma evolution and host interaction from a genomic perspective

00:20 Nuria Fontdevila Pareta: Comparative genomics unveils the lifestyle transition of phytopathogenic Arsenophonus strains

00:20 Benjamin Klauk: Management options to control symptoms caused by candidatus arensophonus phytopathogenicus and stolbur group pathogens

00:15 Natasha Witczak: The diversity of ‚Ca. Phytoplasma solani' in the german agricultural landscape

00:15 Henri Stopin: Phytoplasma an ancient organism as a modern potato threats. Stolbur risk assessment preliminary data

12:40 14:00 Lunch

14:00 15:45 Stolbur and arsenophonus in potato: epidemiology, impact and diagnostic challenges

Continued, Chair: Arnaud Blouin

00:20 Andreas C. Keiser: Vascular bundle browning in processing potatoes in Switzerland

00:15 Christophe Debonneville: Arsenophonus and Stolbur in Potato Crop in Switzerland: A Threat for the Processing Industry?

00:20 Elmar Schulte-Geldermann: Screening of potato varieties against Candidatus Arsenophonus phytopathogenicus and Stolbur Phytoplasma group pathogens

00:10 Jasmin Wiedmer: Exploring resistance to Candidatus Phytoplasma solani and Candidatus Liberibacter solanacearum in potato

00:20 Simon Schiwek: Current results and insights from interlaboratory comparison studies on the detection of PHYPSO and ARSEPH in potato and sugar beet

00:20 Salma Benaouda: qPCR-based quantification of Candidatus Arsenophonus phytopathogenicus and Candidatus Phytoplasma solani in potato (Solanum tuberosum L.) and their effect on yield parameters in southwest Germany

15:45 16:15 Coffee break ----------

16:15 17:15 Roundtable

Phloem-limited bacterial diseases in potato: impacts, management strategies, and research priorities

Moderator Andreas C. Keiser

PROGRAM TUESDAY, JUNE 3

09:00 10:20 Molecular basis of plant–virus recognition and response

Chair: Roberto Miglino

00:20 Laurent Glais: Identification and characterization of potential resistance genes in potato to control potato virus Y infection: interest of the N hypersensitive genes

00:20 Nie Xianzhou: Saikai 35, a potato cultivar believed to carry Rychc, exhibits the classical characteristics of N-gene-mediated temperature-dependent responses upon potato virus Y infection

00:20 Mout De Vrieze: Study of Variety-Specific Responses to the Necrotic Strain of PVYNTN

00:20 Patrice De Werra: Long-Term Assessment of Variety-Specific Yield Losses in Potatoes Caused by PVY

10:20 10:40 Coffee break ----------

10:40 12:00 Plant–aphid interactions and innovative control measures in potato crop protection

Chair: Martin Verbeek

00:45 **Keynote 3 - Patricia Sanches**: Endosymbionts modulate virus effects on aphid-plant interactions

00:15 Arnaud Blouin: Gut content analysis of Myzus persicae reveals insights into viral reservoirs of sugar beet viruses.

00:20 Brice Dupuis: Controlling Potato Virus Y Spread Through Sunflower Intercropping in Seed Potato Fields

12:00 13:30 Lunch

13:30 14:15 Flash talks

Chair: Olivier Schumpp

14:15 15:10 Long-term surveillance of plant viruses: insights from national monitoring programs

Chair: Laurent Glais

00:20 Cécile Thomas: Increasing rejected areas in swiss seed potatoes production from 2016 to 2024 are influenced by viral infections and climate change

00:15 Piret Van der Sman: Long-term monitoring of potato virus Y in Estonian seed potatoes and an evaluation of susceptibility of potato varieties

00:10 Eva Kovacec: Historical perspective of challenges in potato breeding and seed potato production related to virus testing in Slovenia

00:10 Steyer Stephan: Current status of Potato virus Y (PVY) in the Belgian seed potato sector

15:10 15:50 Coffee break ----------

Preparation for those who wish to participate in the walk at Basse Ruche (not part of the official program)

PROGRAM WEDNESDAY, JUNE 4

9:15 – Coffee in the hall of the AO building , Agroscope Nyon

9:30 – Start of visits

- Tour 1: Laboratory visits (30 min)
- Tour 2: Field demonstration visit (30 min)

10:45 – Departure for Delley

12:00 – Lunch in Delley, offered by Swisssem

13:30 – Visit of post-harvest field with Swisssem representatives

15:30 – Departure for Murten

16:00 – Visit of the old town of Murten

17:30 – Transfer to the boat

18:00 – Boarding and cruise departure (include diner)

21:15 – Arrival in Grandson, end of cruise

22:15 – Arrival in Nyon, B&B Hotel

PROGRAM THURSDAY, JUNE 5

09:00 10:50 Diagnostic tools for viral and phloemien bacterial diseases in potato crops

Chair: Christophe Debonneville

00:45 **Keynote 4 - Adrian Fox**: The detection and diagnosis of potato viruses – are we making progress?

00:20 Mohamad Chikh-Ali: Development of a Duplex IC-RT-qPCR for large-scale detection of Potato Mop-Top Virus in dormant potato tubers

00:10 Stephan Steyer : Simple and efficient PCR-based detection of phytoplasmas: validation and application in potato production systems

00:15 Olivier Schumpp: One Reaction to Rule Them All: A Multiplex RT-qPCR for Strain-Level Discrimination of PVY

00:20 Martin Verbeek: Development of a Luminex xTAG assay for the determination of aphid species and simultaneously detection of plant viruses

10:50 11:10 Coffee break ----------

11:10 12:00 Roundtable

Potato viral diseases: impact, control, and future challenges for research and diagnostics

Moderator: Adrian Fox

12:00 12:30 End of the meeting

Chair: Roberto Miglino

12:00 13:30 Lunch

SUMMARY OF PRESENTATIONS

1. From Field to Fork: Overview of the Swiss Potato Sector

Christian Bucher

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Switzerland is a grassland - 70% of the agricultural land is covered with grass. Potatoes are only grown on one percent of the area. However, the crop has a high added value compared to other products and therefore contributes disproportionately to agricultural gross value added.

Potato production
The area under potato cultivation amounts to approx. 10,700 ha and has been constant to slightly decreasing in recent years (10% of which is farmed organically). The structures in Switzerland are rather small: 3,950 farms currently grow potatoes, which corresponds to an average area of 2.7 ha of potatoes per farm. The average yield (net) in recent years has been 37 t/ha.

Potato market
Total annual production amounts to around 400,000 tons. Most of the volume flows into the packaging and industrial channels, with both segments being roughly the same size. The production volume of seed potatoes is around 20,000 tons. Exports are negligible at 10,000 tons. Switzerland is a net importer of potatoes. On average, 70,000 tons are imported every year. Average potato consumption in Switzerland is 46 kg/person per year and has been stable for some time.

Challenges
The potato industry in Switzerland is faced with a number of challenges. The most important of these are

- **Climate change:** More heat waves, drought and heavy rainfall lead to greater fluctuations in yield. In addition, the quality of potatoes is declining, and new pests are spreading. Tolerant varieties are needed to meet these challenges. Potato producers also need to invest more in irrigation infrastructure.
- **Restrictions on the use of pesticides:** Politicians in Switzerland have launched a reduction pathway for pesticides, which calls for a 50% reduction in risks by 2027. As potato cultivation is heavily dependent on plant protection, this represents a challenge for the sector. In addition to various other measures, the industry has decided to focus on resistant potato varieties in the future. To this end, swisspatat has also concluded a target agreement with the federal government (80% robust varieties by 2040).
- **Arsenophonus in potatoes:** The damage caused by Arsenophonus in industrial potatoes represents a rather new challenge. For the past 3-4 years, frites and chips potatoes have suffered major damage due to brown discoloration. One of the solutions here is also the use of tolerant varieties. However, these varieties are limited, which is why further solutions need to be developed. A great deal of research work is still required for this.
- **Attractiveness of potato production for farmers**: In recent years, it has been more difficult to keep the area under potato cultivation stable. Farmers have increasingly refrained from growing potatoes. There are many reasons for this, such as the high risk of production or generational transitions on farms. To counter this, the potato industry has had to improve the framework conditions in recent years. For example, the target prices have been increased significantly.

Opportunities
In addition to these challenges, the potato also has great opportunities in Switzerland. On the one hand, the potato is a trend product in the kitchen. It is versatile, has hardly any allergens and many positive nutritional and physiological properties. In addition, its carbon footprint is relatively small compared to other products. It can therefore be assumed that the demand for potatoes will remain

stable or even increase slightly. As the 2030 agricultural policy in Switzerland aims to strengthen plant-based production and nutrition, we are seeing positive signals from politicians for better framework conditions for this crop in the future.

Keynote 1

Epidemiology under climate pressure

2. Adapting to a changing climate: challenges and strategies for controlling plant viruses and their vectors

Piotr Trebicki

Applied BioSciences, Faculty of Science and Engineering, Macquarie University, Australia, Email: piotr.trebicki@mq.edu.au

Abstract:
Climate change, driven by increasing greenhouse gas emissions such as carbon dioxide, poses a major threat to agriculture and future food security. Among the critical challenges is the impact of changing environmental conditions on interactions between food crops, plant viruses, their insect vectors, and associated trophic levels. In potato production systems, understanding these interactions is vital for effective disease management under future climate scenarios. Plant viruses and their vectors are highly responsive to shifts in temperature, carbon dioxide levels, and other climatic factors, potentially altering virus, vector and host dynamics. These changes may not only increase the incidence and severity of viral diseases in potato crops and other food systems but also can create conditions that favour the emergence, evolution or colonisation of new pests and pathogens that are currently of minor or no significance. However, despite some case studies, our overall knowledge remains limited. The ability of viruses and insect vectors to rapidly adapt to changing conditions, owing to their short generation times, could lead to faster disease spread, more severe outbreaks and greater pressure on agricultural systems. Given these risks, there is a need to deepen our understanding of how climate change influences pests and diseases, particularly those affecting potato crops. Expanding this knowledge will be essential for designing resilient and sustainable disease management strategies to protect potato production and ensure global food security in an increasingly changing world.

3. Epidemiology of PVY infections; environmental influences on the interactions between plant, vector and virus

Martin Verbeek[1], Klaas van Rozen[2], and René A.A. van der Vlugt[1]

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[2]Wageningen University and Research, Open field crops, Lelystad, The Netherlands
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INTRODUCTION

Potato virus Y (PVY) is the major virological issue in growing seed potatoes in The Netherlands. The Dutch General Inspection Service (NAK) provides export certificates for the seed lots through field inspections and post-harvest laboratory tests. Over the past 10 years, seed potato growers have been confronted with increasing PVY infections resulting in rapidly increasing declassification numbers, especially in years following (very) mild winters. Also the banning of several crop protection agents is shaping a different situation regarding the epidemiology of PVY and other viral diseases.

MATERIALS AND METHODS

In 2019, a four-year project started that investigated several aspects of PVY infection and spread. The research focused on possible source plants of PVY, especially in spring, on aphid flight monitoring and the relation with the winter temperatures and the behaviour of PVY in plants. In total 6000 plants, including weeds surrounding fields where potatoes were grown in the previous year and potato volunteer plants were tested in ELISA. Aphids flights were monitored using three trapping methods: yellow water traps, Rothamsted suction traps, and Ashby funnel traps, during 4 seasons. Caught aphids were identified to species level by the Dutch Inspection Service (NAK). In order to better understand the behaviour of PVY in potato plants, the translocation of viral particles from the leaves to the tuber was studied.

RESULTS AND DISCUSSION

When sampling of weeds and volunteer plants was conducted in spring time, PVY was only found in the volunteer plants. This result suggests that overwintering of PVY in weeds is not a big issue in The Netherlands. As volunteer plants are often emerging before the sown seed-potatoes, and were found to be often infected with PVY, these volunteer plants seem to be the most important virus sources in early spring. Aphid flights occur earlier in the year and aphids occur in higher numbers due to milder winters. Therefore, the chance of early infection of the young and susceptible potato plants is considerable.
For a long time, the translocation speed of PVY was estimated to be approximately 10 days which formed the basis for haulm destruction dates after reaching a certain vector and virus pressure. However, in our studies we determined a translocation speed of 3-4 days. This knowledge is important for being aware of the risk of regrowth after haulm destruction. The new shoots of the regrowth are very susceptible for PVY infections and the virus can reach the potato tubers in just a few days. These late infections are often underestimated, and can still cause unexpected elevated numbers of infected tubers in post-harvest testing.

ACKNOWLEDGEMENTS

This research was financed by the Topsector Tuinbouw en Uitgangsmaterialen under project number TU18049 and partners.

4. Investigating The Epidemiology of 'Candidatus Liberibacter Solanaceraum' In Great Britain

Adrian Fox

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INTRODUCTION

The proteobacterium '*Candidatus* Liberibacter solanacearum' (Lso) is associated with disease in a range of crops, most notably potato, tomato, carrot and celery. Lso is vectored by psyllids, with species such as *Trioza apicalis* (carrot psyllid) and *T. anthrisci* transmitting Lso to apiaceous hosts including carrot (*Daucus carota*) and cow parsley (*Anthriscus sylvestris*). Where crops are infected Lso can lead to significant economic losses. However, despite Lso haplotypes, hosts and vectors all being present, the pathogen is not causing major crop losses in GB. The "CALIBER" interdisciplinary consortium, consisting of researchers from FERA, John Innes Centre, Newcastle University, Rothamsted Research, SASA, and University of Strathclyde, has been working to understand the presence, distribution, host range, ecology, and vector characteristics influencing the distribution of Lso haplotypes in the UK.

DISCUSSION

Field surveillance was conducted to determine the prevalence of Lso haplotypes and vectors in hosts across GB. Sampling follows the principles of a network ecology study, with paired crop and non-crop transects in each sampled field, with approximately 6,000 plants sampled per year. Vector samples were taken directly from plant samples and from sweep netting of sampling transects. In each case samples were tested for the presence of Lso haplotypes, to determine associations between the vectors, plant hosts and Lso haplotypes detected. Initial results indicate that some Lso haplotypes are widespread in hosts such as stinging nettle (*Urtica dioica*), however, the vectors associated with the transmission of apiaceous haplotypes, although present, may have limited distribution in the UK.

With the recent discovery of novel Lso haplotypes in the UK (Sumner-Kalkun et al., 2020), feeding and behaviour studies on inter-host transmission by known vectors of Lso were also being conducted. The outcomes of this work will help to understand the factors which determine if benign infections can become damaging epidemics.

REFERENCES

Sumner-Kalkun, J. C., Highet, F., Arnsdorf, Y. M., Back, E., Carnegie, M., Madden, S., Carboni, S., Billaud, W., Lawrence, Z., & Kenyon, D. (2020). 'Candidatus Liberibacter solanacearum'distribution and diversity in Scotland and the characterisation of novel haplotypes from Craspedolepta spp.(Psylloidea: Aphalaridae). Scientific reports, 10(1), 16567.

5. Novel Approach to Study the Dynamics of Potato Virus Y Infection Pressure –Case Study: Colorado, USA

Jeremy Daniel[1] and Mohamad Chikh-Ali[1,2]

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INTRODUCTION

Potato virus Y (PVY) is the leading cause of seed potato rejection, which imposes a crucial threat to the availability of certified seed potatoes in many potato producing areas. The control of PVY relies on multiple approaches to minimize the seed-borne and current season infection. To reduce the current season infection (primary infection), monitoring the dynamics of PVY infection pressure over the growing season is important to develop a targeted management program and reduce the impact caused by insecticides and crop oil on the environment. In the current study, we developed and tested a novel approach to monitor PVY infection pressure in Colorado, U.S. for the first time. This approach relied on the use of tobacco plants as baits to capture PVY natural infection. This allowed tracking PVY infection on a weekly basis throughout the growing season.

MATERIALS AND METHODS

Healthy sets on tobacco plants (*Nicotiana tabacum* cv. White Burley) were deployed weekly to potato fields in the San Luis Valley, Colorado. By the end of the week exposure window, tobacco plants were treated with insecticides and placed in the greenhouse for 3-4 weeks. Plants were then tested for PVY using ELISA and RT-PCR and the incidences were plotted against the field exposure time. This experiment was repeated over three growing seasons.

RESULTS AND DISCUSSION

Tobacco bait plants developed mosaic and\or vein necrosis 2-3 weeks after being exposed to natural field infection by aphids. PVY infections were confirmed using ELISA and RT-PCR. In the San Luis Valley, most PVY infection occurred during the months of July and August in 2022 and 2023, while it was delayed in 2024 to the month of August. The RT-PCR helped to identify three PVY strains, PVY^{O}, $PVY^{N\text{-}Wi}$ and PVY^{NTN}. The current study demonstrated the usefulness of bait plants to understand PVY epidemiology and develop more targeted control practices of PVY.

ACKNOWLEDGEMENTS

This work was funded by the Colorado Potato Administrative Committee; the USDA-NIFA Hatch Project grant COL00426; and the Colorado State University Agricultural Experiment Station.

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Chikh-Ali, M., Gray, S. M., & Karasev, A. V. (2013). An improved multiplex IC-RT-PCR assay distinguishes nine strains of Potato virus Y. Plant Disease, 97(10), 1370-1374.

Chikh-Ali, M., Maoka, T., Natsuaki, K. T., & Natsuaki, T. (2010). The simultaneous differentiation of Potato virus Y strains including the newly described strain PVYNTN-NW by multiplex PCR assay. Journal of Virological Methods, 165(1), 15-20.

Daniel, J., & Chikh-Ali, M. (2024). Dynamics of Potato Virus Y Infection Pressure and Strain Composition in the San Luis Valley, Colorado. Plant Disease, PDIS-10.

Keynote 2

6. Stolbur, phytoplasma evolution and host interaction from a genomic perspective

Michael Kube

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Phytoplasmas have long been recognised as insect-borne bacterial plant pathogens in various crops. Currently, a stolbur outbreak is threatening sugar beet and potato production in Germany. It is dominated by a recently discovered phytoplasma from the 16SrXII-P subgroup. The cixiid vector *Pentastiridius leporinus* transmits this phytoplasma and the causal agent of Syndrome Basses Richesses, *Candidatus* Arsenophonus phytopathogenicus, to various crops. Comparative and functional analyses of the representative strain GOE and other phytoplasmas illustrate the impact of genomics for understanding these pathogens. Common features of phytoplasmas such as the reductive evolution and an efficient metabolic repertoire were aso confirmed for strain GOE, but also virulence factors involved in immunomodulation and effector-derived manipulation of organogenesis associated with the mobilome. The growing genome database for phytoplasmas provides a differentiated picture of evolutionary adaptations in phytoplasmas.

7. Comparative genomics unveils the lifestyle transition of phytopathogenic Arsenophonus strains

Mathieu Mahillon[1], Christophe Debonneville[1], Nuria Fontdevila Pareta[1], Raphaël Groux[1], David Roquis[2], Justine Brodard[1], Franco Faoro[3], Xavier Foissac[4], Olivier Schumpp[1], Jessica Dittmer[3,5]

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INTRODUCTION

Phloem-infecting bacteria pose an escalating threat to global agriculture, causing significant losses in crops such as sugar beet and potato. Among these, "*Candidatus* Arsenophonus phytopathogenicus" (Ap) has emerged as a notable pathogen and a causal agent of syndrome basses richesses (SBR). These bacteria are challenging to culture *in vitro* yet thrive within the plant's phloem and hemipteran insect vectors. Despite their agricultural importance, the genetic mechanisms underlying the transition of *Arsenophonus* from insect endosymbionts to plant pathogens remained largely unexplored until now.

MATERIALS AND METHODS

To investigate the evolutionary and functional basis of phytopathogenicity in Ap, the genome of Ap was reconstructed using publicly available metagenomic sequencing data obtained from insect vector hosts, such as *Pentastiridius leporinus*. As Ap cannot currently be cultured in vitro, this metagenomic approach provided access to high-quality genomic sequences directly from infected insects. Comparative genomic analyses were conducted to assess the genome size, gene content, and the abundance of viral sequences. Phylogenomic analyses, including multilocus sequence analysis and whole-genome phylogenies, were used to clarify the evolutionary position of Ap within the *Arsenophonus* genus and to determine its relationship to other endosymbionts and plant pathogens. Orthologous gene clustering across available *Arsenophonus* genomes identified genes unique to Ap and potentially associated with its phytopathogenic lifestyle. Functional annotation of these unique genes, together with in silico predictions of protein localization and activity, as well as gene expression profiling, provided insights into their possible roles during plant infection and phloem colonization.

RESULTS AND DISCUSSION

The reconstructed genome of Ap closely resembled those of other facultative endosymbiotic *Arsenophonus* strains associated with sap-sucking hemipterans. Notably, the Ap genome is rich in viral sequences, suggesting a history of frequent interactions with mobile genetic elements. This viral enrichment is significant, as it likely facilitated horizontal gene transfer events that contributed to the acquisition of new functions relevant to plant pathogenicity. Phylogenomic analysis positioned Ap within the *Triatominarum* clade of the *Arsenophonus* genus, a distinct lineage within the *Arsenophonus* genus originally characterized by symbionts of triatomine bugs. The placement of Ap in this clade seems to indicate that Ap evolved from an insect-associated ancestor and only later acquired the ability to infect plants. Within its genome, Ap harbors a small set of orthologous genes not found in non-pathogenic *Arsenophonus* strains. Several of these genes code for putative plant cell wall-degrading enzymes and cysteine peptidases related to xylellain, which is a papain-like peptidase from *Xyllela fastidiosa*. The presence of these unique genes suggests that Ap has acquired the capacity to degrade plant cell walls and overcome plant defense barriers, which are key traits for successful phloem colonization and systemic infection. The likely origin of these genes through horizontal gene transfer from other plant-associated bacteria highlights the importance of

mobile genetic elements in the emergence of new phytopathogens; and, how a limited number of genetic innovations, facilitated by viral elements, can drive the transition from an insect endosymbiont to a plant pathogen. The identification of unique, plant-targeting enzymes in its genome not only sheds light into the evolutionary steps leading to its emergence as a plant pathogen but also suggests potential targets for future disease management strategies in crops affected by SBR and related diseases.

ACKNOWLEDGEMENTS
This research was funded by the European Union's Horizon 2020 research and innovation program Marie Sklodowska-Curie, grant agreement No. 792813 to J.D., and the Swiss Federal Office for Agriculture (grant 2020/33/LES-Z II to O.S.).

8. Management Options To Control Symptoms Caused By Candidatus Arensophonus Phytopathogenicus And Stolbur Group Pathogens

Benjamin Klauk, Benson Kisinga, Elmar Schulte-Geldermann

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INTRODUCTION

In recent years, potato production in south-west Germany has been affected by the emerge of the phytoplasma *Candidatus* Phytoplasma solani (stolbur) as well as *Candidatus* Arsenophonus phytopathogenicus (Lang et al., 2025). In addition to the increase in genetic diversity of the pathogens, particularly *Ca*. Phytoplasma (Toth et al., 2025), the rapid spread of their vector *Pentatstiridius leporinus* threatens profitable potato production. Typical symptoms such as aerial tubers, leaf discoloration, wilting plants and rubbery tubers were discovered in many fields, leading to yield and quality losses (Behrmann et al., 2023). Due to the socioeconomic value of potatoes in affected regions and inefficient direct control to *P. leporinus*, cultural practices should be focused on minimizing the economic loss. However, direct vector control by insecticides, which are approved on the European market, seems unpracticable due to high mobility and extended migration period of the adult planthopper (Pfitzer et al., 2022). Cultivation measures can hypothetically reduce the symptoms and the associated quantity and quality losses in two ways: (1) Reducing contact between the plant and the vector by confusing the vector's olfactory or visual senses. For example, the landing rates of aphids can be significantly reduced by applying straw mulch to potatoes (Winkler et al., 2025), which could also be hypothesized for *P. leporinus*. (2) Increasing the plant's resilience to infection. Noteworthy, the plant's defenses can be induced by exogenous addition of substances such as salicylic acid (Wani et al., 2017). The aim was to evaluate promising management options such as variation in planting date, the use of mulch and catch crops in potatoes and the application of exogenous substances such as salicylic acid to reduce the symptoms of Stolbur and Arsenophonus in potatoes.

MATERIALS AND METHODS

All field trials were carried out in Bingen, Germany (49°58′ N 7°54′ E). *Hyalestes obsoletus* and *P. leporinus* have been detected in Bingen since 2022, with a steady increase in *P. leporinus*. During the trial period from 2022 to 2024, the average annual temperature was around 12 - 12.2 °C. 2022 was the driest year with only 496 L/m² and 2024 the wettest with 600 L/m². All field trials used a completely randomised block design, which included four replications for each treatment. All plots were 3 m (four rows of 0.75 m) and 6 m in size. A mulching trial was carried out in all trial years. For each of three varieties (cv. Valdivia, Annalena, Emiliana), 3 kg of barley straw per m² were applied after the plants had emerged and a treatment without mulch was used as a control. In 2024, three mixtures of catch crops (a mixture of 40 kg ha^{-1} *Lathyrus sativa*, 4 kg ha^{-1} *Plantago lanceolata*, 15 kg ha^{-1} *Linum usitatissimum*; a mixture of 5 kg ha^{-1} *Tagetes patula*, 5 kg ha^{-1} *Calendula officinalis* and a mixture of all five species at 50 % seed rate) were used as underseeding between the ridges three weeks after potato planting. The mixtures were applied to the varieties Emanuelle, Almonda and Cammeo. In addition to salicylic acid, triacontanol and tarin were also tested substances designed to induce the plants defence. Loker SA (2 L ha^{-1}, salicylic acid), Begreen (2 L ha^{-1}, tarin) and Begreen (2 L ha^{-1}, tarin) plus Triaminol (1 L ha^{-1}, triacontanol) was applied weekly (seven times, start eight weeks after planting). The products were tested in the Annalena and Cammeo varieties. Wilting symptoms were visually assessed and rAUDPC was calculated in all trials. After recording tuber yield, a sample of 5 kg tubers were assessed for specific symptoms like rubbery-like tubers and filamentous sprouts. Additionally, under water weight was measured to determine the starch content.

RESULTS AND DISCUSSION

In 2022, higher tuber yield and marketable tuber yield was harvested in the mulched treatment. The proportion of rubbery-like tubers decreased significantly up to 20 %, if mulch was applied. Approximately 42 t ha^{-1} were harvested in the mulched treatment compared to 32 t ha^{-1} in the control treatment in 2023. In the last trial year, no significant differences could be observed between mulched and control treatments in yield and quality of the tubers. The decreasing differences between the treatments correlated with a higher incidence of *P. leporinus*. Consequently, we assume that mulching reduces abiotic stress of plants likely to be the reasons for reduced disease symptoms, especially in low-rainfall years such as 2022.

Using catch crops for underseeding in potatoes led to a higher tuber yield compared to the control treatment, if legumes like *L. sativus* were included. No significant differences were found for the proportion of rubber-like tuber. There was only an observed tendency that the mixture of *T. patula* and *C. officinalis* had a lower number of rubber tubers. Both species flowered, which could be an indication of vector confusion due to the visual alteration. Only marginal differences in tuber yield and quality were measured or assessed for the applied substances compared to the control treatment. In conclusion, the use of mulch reduces abiotic stress, especially in dry years, and strengthens the plants against infection by stolbur pathogens and/or *Ca. Arsenophonus phytopathogenicus*. There were no significant improvements in the first year of trials with undersown catch crops and plant defense inducers. Other measures, such as earlier planting dates and the use of tolerant varieties, need to be implemented as part of a strategy to safeguard potato production in Central Europe.

ACKNOWLEDGEMENTS

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9. The Diversity of Ca. Phytoplasma solani‘ in the German Agricultural Landscape

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INTRODUCTION

Yellowing symptoms in sugar beet (*Beta vulgaris* ssp. *vulgaris*) have been observed with the phloem-feeding planthopper (*Pentastiridius leporinus*) in France (Gatineau et al., 2001) and Germany (Schr¨oder et al., 2012). These symptoms are associated with three phloemrestricted pathogens: The -proteobacterium ’*Candidatus* Arsenophonus phytopathogenicus’ (ARSEPH), 16SrXII phytoplasma ’*Candidatus* Phytoplasma solani’ (PHYPSO), and the newly described PHYPSO-related 16SrXII-P phytoplasma (Duduk et al., 2023; Toth et al., 2025). *Pentastiridius leporinus* transmits these pathogens to sugar beet (Bressan et al., 2011; Duduk et al., 2023; Toth et al., 2025) and potato (*Solanum tuberosum* ssp. *tuberosum*) (Behrmann et al., 2023; Therhaag et al., 2024), causing considerable yield reductions and thus major economic losses for farmers. Since 2024, infection occurred in other vegetable crops. To analyze the genetic variants of the pathogen and the possible spread of PHYPSO across different crops 16SrXII strains from crops, potential reservoir plants, and vectors were characterized genetically. Analyses of phytoplasma populations were conducted across locations to identify possible new epidemiological pathways and to assess risks for crop production.

MATERIALS AND METHODS

Insect and plant sampling

Adult planthoppers and larvae were collected from various locations across Germany using sweeping nets and shovels at different times in 2023 and 2024. Specimens from potato fields and areas in close proximity were used to transfer isolates of 16SrXII phytoplasmas (PHYPSO and 16SrXII-P) and ARSEPH to periwinkle (*Catharanthus roseus*). Deceased insects were identified morphologically according to the keys provided by Biedermann & Niedringhaus (2004) and Kunz et al. (2018). Female Cixiidae and larvae underwent COI barcoding in accordance with the EPPO standard protocol (EPPO, 2021). Plant tissues were collected from weeds and arable crops before storing them at -20 ◦C until DNA extraction.

Transmission

On the day of collection, up to 12 adult specimens of the same species were placed on a single periwinkle plant within a cylindrical acrylic insect-proof cage. The experiments were conducted in a climate-controlled insect chamber with an inoculation access period of 10 days. The cages were checked daily for deceased insects, which were subsequently removed and stored at -20 ◦C for further analysis. Infected plants are stored in a master collection at JKI and serve as references.

DNA extraction and PCR

DNA was extracted from insects using the CTAB method and from plants according to Maixner et al. (1995).Both insect and plant tissue samples were tested for the presence of 16SrXII phytoplasma and ARSEPH. Infection with 16SrX phytoplasmas was evaluated by PCR with specific primers targeting the *tuf* gene (Schneider et al., 1997), and with ARSEPH using *Fra4/5* targeting a part of 16S rDNA (Zreik et al., 1998). PHYPSO positive PCR results were further characterized by MLST

including RFLP based on the marker genes *tuf* and *vmp1*, as well as sequence analysis of the nPCR amplicons of the marker genes *stamp* and *secY*.

RESULTS AND DISCUSSION

The results of the MLST analyses were compared between insect and plant samples. In the insect samples, the 16SrX-P phytoplasma was primarily detected. In the plant samples, various genotypes were identified with the recently described 16SrX-P phytoplasma being predominant (Duduk et al., 2023; Toth et al., 2025). Preliminary data indicate that certain genotypes were detectable across the analyzed cultures.

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10. Phytoplasma an ancient organism as a modern potato threats. Stolbur risk assessment preliminary data.

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Stolbur (*Candidatus Phytoplasma Solani*) is present in Europe for decades. Since 2020, the prevalence of this pathogen is increasing in Western Europe in potato crops. HZPC, has integrated this emerging pathogen into its research programs.
Identification of cultivars that show tolerance and/of resistance would be of value for potato breeding. Therefore, the last two years we have been carrying field experiments with Dutch, French, Swiss, and German teams to be able to identify tolerant cultivars and to learn more about this disease
Furthermore, we have been focusing on determine the risk of planting Stolbur infected tubers. First, a molecular detection method (PCR) has been implemented, in house, to detect and quantify Stolbur and BTW (*Candidatus Arsenophonus Phytopathogenicus*) in tubers as well as aboveground (leaf, stem) samples. This implementation was made possible thanks to the support of Agroscope.

Few knowledge is present in the literature about the vitality of Stolbur infected tubers progeny. Several experiments in partnership with Agroscope have been conducted in 2023 and 2024 to get more knowledge on this topic.

In the last experiment, infected tubers from growing season 2023 have been collected and diagnostic using PCR. Seven lots of 2 cultivars have been planted in May 24, in a field assay. For each lot, infected tubers versus non infected tubers have been compared. Emergence, plant growth and tuber yield have been measured.

For all tubers, full emergence has been observed. The impact of Stolbur infection on the following parameters was significant: 35% height reduction, 41% less soil coverage, 48% less tubers per plant and 55% less yield per plant. Cultivar diversity was not an influencing parameter

The conclusion of the two years experiment is that Stolbur infection does not inhibit the germination and the emergence of tubers, but the result is a weak plant with low yield.

To continue the risk assessment, the focus of 2025 research is to explore the pathogen presence in the tuber progeny: the mother – daughter interaction.
To answer this question, we are currently running greenhouse experiments.

11. Vascular bundle browning in processing potatoes in Switzerland

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INTRODUCTION

In the last years, Swiss processing potatoes have increasingly been scoring badly in the compulsory baking test. The test consists in frying potato slices and evaluating the browning of the tissue. The rejected potato batches present an unacceptable browning of the vascular bundles. Affected batches cannot be processed into French fries or potato chips, or only to a limited extent. The quality defects have a very negative economic impact on the entire value chain, from production and trade to processing. The losses were particularly high in 2022 and 2023. Possible causes include the endophytic bacterium *Candidatus Arsenophonus phytopathogenicus (*Arseph*)*, the phytoplasma *Candidatus Phytoplasma solani* (Stolbur) or the fungus *Verticillium dahliae* (Vert). Weather-related stresses (heat, drought) seem to enhance the disease symptoms. Arseph causes the “Basses richesses” syndrome (SBR) in sugar beets and was detected in potatoes for the first time in Germany in 2022. Stolbur is also frequently found in potatoes or sugar beets in certain regions of Germany. Both pathogens can occur individually or as a double infection, causing various symptoms (e.g. wilting, air pockets or gummy roots/tubers) and, overall, significant crop losses. The bacteria are mainly transmitted by the planthopper species *Pentastiridus leporinus.*
In a joint project involving the entire potato industry, BFH-HAFL, Agroscope and Bioreba AG are investigating the causes of the observed quality defects and looking for solutions to be implemented in collaboration with the industry. The project (2024-2027) is funded by the Federal Office for Agriculture and the potato industry. This summary discusses the results of the monitoring carried out by BFH-HAFL and Bioreba AG 2024.

MATERIALS AND METHODS

In autumn 2024, harvest samples were collected from 142 potato fields across the entire potato-growing region and tested for Arseph, Stolbur and Vert using PCR analysis. The number of samples collected per region was a function of the proportion of potatoes in the crop rotation. The coordinates of each field and the crops within a 500 m radius were recorded. The quality of the samples was assessed by the potato industry using the baking test (Backtestemethode 86). Tubers (N=165) were also chosen randomly from potato batches which had a bad baking test score and subjected individually to the baking test. Based on the results, the tubers were graded on a scale of 1 to 9 (9 = very good chips with optimal baking quality, 1 = poor chips with severe browning of the vascular bundles and multiple spots). The same 165 tubers were individually tested for Arseph, Stolbur and Vert using PCR.
To investigate the influence of location and variety, variety trials with important processing varieties (Agria, Fontane, Innovator, Pirol, SHC 1010) were conducted at two locations. One location (Oberwil, BE) was within the infestation area of the “Basses richesses” syndrome in sugar beet, the second was outside (Dägerlen, ZH). To monitor the flight activity of *P. leporinus* in potatoes fields, sticky traps were installed at 17 locations from calendar week 21 (mid-May). Additional sticky traps were placed in vegetable and sugar beet plots. The traps were checked weekly and the planthopper species identified visually. For verification purposes, 70 individuals were identified using qPCR.

RESULTS AND DISCUSSION

Arseph was detected in 43% of potato samples, while 11% of samples showed poor baking quality. Samples infected with Arseph were limited to the western part of the potato-growing region, within the distribution area of SBR in sugar beet. Only 2.9% of samples tested positive for Stolbur (Fig. 1). The results of the individual tuber analyses show that infection with Arseph increases significantly with the deterioration of baking quality. 11% of tubers with baking grades 9 and 8 were infected with Arseph, the figure rising to 75% for grades 6 and 5 and reached 100% for grades 4 and 3. No similar correlation could be established for Vert.
The first planthopper were caught in calendar week 21. Insects were caught at all locations with sticky traps in all fields within the distribution area of SBR in sugar beet. The highest number of catches occurred in areas with high proportions of sugar beet, potatoes and vegetables.
Of the 70 planthoppers analyzed by qPCR, 64 were identified as *P. leporinus*, the other six to other species. 75% of the 64 *P. leporinus* tested using qPCR were positive for Arseph, and only one for Stolbur. No planthoppers were caught in potato fields at sites outside the SBR distribution area. But a monitoring in sugar beet fields by the Swiss Sugar Beet Research Centre (SFZ) showed that planthoppers occur at least sporadically in sugar beet outside the SBR distribution area. No planthoppers were caught in the Dägerlen variety trial, and all the harvest samples tested negative for Arseph and Stolbur. The situation was different at the Oberwil site, where planthoppers were caught in the sticky traps. 20 % of harvest samples were positive for Arseph, but no Stolbur was found. Harvest samples with evidence of Arseph exhibited a significantly poorer baking quality than those with no Arseph. In addition, differences between varieties in the severity of vascular bundle browning (baking test) were observed. These must be confirmed in further trials.

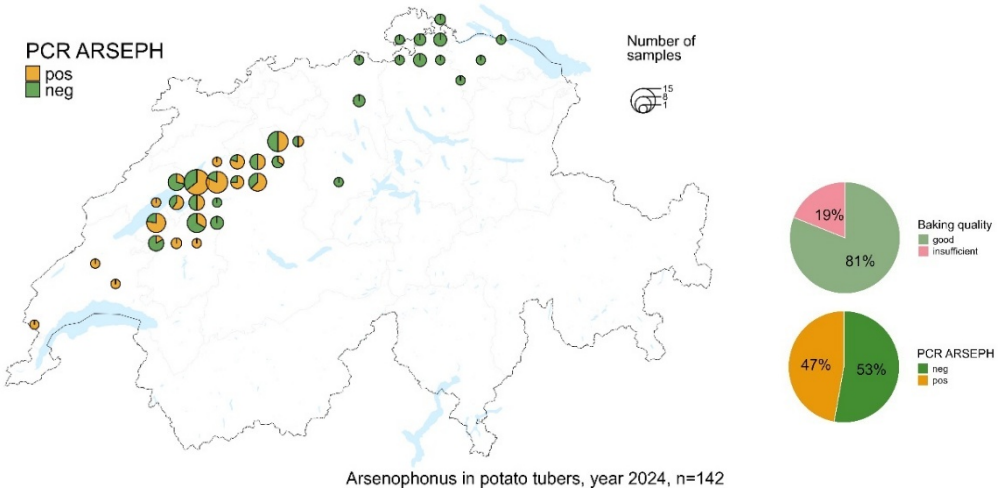

Fig. 1: Regional distribution of harvest samples (n= 142) collected from potato fields in 2024 with percentages (%) of positive detection of *Candidatus Arsenophonus phytopathogenicus* and percentages (%) of insufficient baking quality.

The results suggest that the bacterium Arseph is the main cause of the poor baking tests. This hypothesis is supported by the fact that vascular bundle browning is still limited to western Switzerland, with concomitant occurrence of SBR in sugar beet. The phytoplasma Stolbur was only found rarely in potato samples. Yet, the SBR vector *P. leporinus* is also present in eastern Switzerland and individual sugar beet samples have tested positive for Arseph. Therefore, urgent and effective measures are required to prevent the further spread of the pathogen and the disease.

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12. Arsenophonus and Stolbur in Potato Crop in Switzerland: A Threat for the Processing Industry?

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ABSTRACT
Two phloem-restricted pathogens are associated with the disease known as “Syndrome Basses richesses” (SBR): the Stolbur (16SrXII group) phytoplasma ‘*Candidatus* Phytoplasma solani’ and the γ-proteobacterium ‘*Candidatus* Arsenophonus phytopathogenicus’. Both are transmitted by planthoppers, *Pentastiridius leporinus* (Hemiptera: Cixiidae) being the main vector in sugar beet.
Here, we report the first identification of ‘*Candidatus* Arsenophonus phytopathogenicus’ and its planthopper vector *Pentastiridius leporinus* in potato fields in Switzerland in 2023. The bacterium was detected in potato plants and tubers exhibiting phytoplasma-like symptoms and collected from cantons currently experiencing SBR outbreaks. In infected tubers, we show that Arsenophonus can be detected after dormancy from the stem end to the emerging sprouts with decreasing titers. Importantly, Arsenophonus might induce threadlike sprouts and the browning of the flesh upon frying, raising strong concerns for varieties marketed for chips production. Altogether, our results align with recent studies performed in Germany, highlighting the host shift of Arsenophonus and its vector from sugar beet to potato crops.

13. Screening of potato varieties against Candidatus Arsenophonus phytopathogenicus and Stolbur Phytoplasma group pathogens

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INTRODUCTION

Since 2022 is has been reported that the bacterial pathogens *γ-Proteobacterium Candidatus Arsenophonus phytopathogenicus* (ARSEPH) and pathogens from the Stolbur Phytoplasma group (STPHYP), both transmitted by the reed glass-wing cicada (*Pentastiridius leporinus L.*), causing infections with bacterial potato tuber wilt (BPTW) (Behrmann et al. 2023, Therhaag et al., 2024). The outbreak of this disease is strongly favored by rising temperatures and mild winters and is therefore, among other factors, an indirect consequence of climate change (Behrmann et al. 2023). The disease first broke out in south-west Germany and is now spreading rapidly to other growing regions, and affecting other crops such as sugar beet, carrots, beetroot and other vegetable crops. In potatoes, the pathogens cause considerable qualitative and quantitative losses and jeopardizes profitable cultivation in the affected regions (Lang et al., 2025). While ARSEPH and STPHYP reduce crop viability and processing quality, their combined infections exacerbate the damage, introducing secondary pathogens and compounding challenges for farmers.
Control measures remain limited and the knowledge about varietal resistance or tolerance is limited. Symptoms of infected potato plants are diverse and unspecific, hence can easily misinterpreted. Field symptoms can be yellowing or reddening of shoots and leaves, wilting and or pre-mature plant death and development of aerial tubers. Tuber symptoms are rubber-like tubers, reduced storability and early sprouting which are all associated with increased sugar contents. Increasing infestation produces smaller and softer tubers (Behrmann et al., 2024). Tubers also show increased defects, such as browning of the vascular ring, necrosis at the junction between the stolon and the tuber, or an adversely affected sugar content (Lang et al., 2025).

MATERIALS AND METHODS

All field trials were carried out in Bingen, Germany (49°58′ N 7°54′ E). During the trial period from 2022 to 2024, the average annual temperature was around 12 - 12.2 °C. 2022 was the driest year with only 496 l/m² and 2024 the wettest with 600 l/m², with a negative climatic water balance in all years. Seed potatoes of in total 104 varieties were provided by 16 potato breeding European companies.
All field trials used a completely randomized block design, which included three replications for each variety. Field symptoms were assessed visually starting from the occurrence of first symptoms on a weekly. In order to quantify disease progress we calculated rAUDPC values for leaf area loss and number of wilted plants and the number of plants with aerial tubers. Tuber assessments compromised besides the yield parameters the percentage of rubber-like tubers and with a subset the discoloration of the vascular ring among other defects. To confirm the presence of ARSEPH and STPHYSO in the varieties DNA extracts were prepared from 0.1 g tuber heel end samples, using a modified CTAB method followed by qRT-PCR diagnostics (Behrmann et al. 2023, Toth et al 2025).

RESULTS AND DISCUSSION

Hyalestes obsoletus and *P. leporinus* have been detected in Bingen since 2022, with a steady increase in *P. leporinus*. This resulted a change in infection patterns over the trial years. From a dominance of '*Candidatus Stolbur Phytoplasma solani* in 2022 to a dominance of infections with *Candidatus arsenophonous* in 2023 and 2024. In 2024 ARSEPH was detected in more than 90 % of

samples and STPHYSO in 35%, but the latter mostly in combination with ARSEPH. High genetic diversity of both pathogens has been observed (Witczak, unpublished, 2024).
In total 38 varieties expressed extreme wilting symptoms starting from 75 days after planting causing early plant death less than 50% of the respective average trial yields. Most of those varieties have been dropped after one season. The expression of symptoms differed largely among the tested varieties with no variety being without symptoms. About 20% of the varieties showed little above ground symptoms, but with a high proportion of rubber-like tubers, while another 10% it has been vice versa. Most of the varieties expressed both above ground and tuber symptoms. Across the three years only eight varieties performed at commercially viable production level with acceptable tuber qualities. Two more promising varieties were only tested in 2024. It can be concluded that the exsistence of an R-gene resistance is highly unlikely, but some varieties some tolerance mechanisms, which need to be assed in further studies.

To determine the correlation between infection type, infection titer, disease symptoms and yield we tested tubers from two symptomless plants of five popular varieties at 90 days after planting (July 18th, 2024). While more susceptible varieties started showing symptoms already at the beginning of July, all sampled varieties (remaining plants) expressed above ground field symptoms a month later. However, at different severities wit two of them wilting quickly and one having 22% of plant with aerial tubers (Tab.1). All varieties were infected with ARSEPH and two out of five having a double infection. A low titer with ARSEPH led to a small number of rubber-like tubers, whereas double infections significantly increased their amounts. Yields weren't affected by the infections as all varieties were able generate enough yield before symptoms set in (Tab.1).

Table 1. Infections with bacterial potato tuber wilt rand related symptoms of five potato varieties at the Bingen University trial site in 2024

	ARSEHP		PHYPSO		Field symptoms			Tubers	
Variety		Titer		Titer	rAUDPC	rAUDPC wilted plants	% plants with aerial tubers	Rubber-like %	t*ha^{-1}
A	+	+++	0	0	0,28	0,01	0,0	12,2	51,2
B	+	+++	+	+++	0,30	0,03	3,7	27,8	57,8
C	+	++	0	0	0,52	0,07	0,9	3,3	43,5
D	+	++	+	+	0,34	0,01	0,9	31,1	62,1
E	+	+	0	0	0,53	0,10	22,2	1,1	50,1

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14. Exploring resistance to Candidatus Phytoplasma solani and Candidatus Liberibacter solanacearum in potato

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Abstract
Potato (*Solanum tuberosum*) faces emerging threats from the bacterial vector-borne diseases Zebra Chip and Stolbur, caused by the invasive *Candidatus* Liberibacter solanacearum and *Candidatus* Phytoplasma solani, respectively. Identifying resistance sources against these phloem-limited pathogens is particularly challenging, because these bacteria are not culturable, quarantine regulations heavily restrict disease testing, and in potentially resistant plants, it is often unclear whether the defense response targets the bacteria or the insect vector. To address these challenges, we take one puzzle piece at the time. First, we apply an effectoromics approach in wild *Solanum* genotypes. Subsequently, genotypes that respond to effectors are tested for bacterial resistance by grafting. Ultimately, genotypes that exhibit resistance in the grafting assays undergo disease testing with their insect vectors. So far, we have identified various *Solanum* genotypes that mount defense responses to effectors of Liberibacter and Phytoplasma. We have successfully established grafting experiments with Phytoplasma in wild *Solanum* accessions, and screening efforts are ongoing. Our goal is to identify immune receptors against these bacteria that can be introduced into potato cultivars to enhance the resistance against this emerging disease.

15. Current results and insights from interlaboratory comparison studies on the detection of PHYPSO and ARSEPH in potato and sugar beet

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INTRODUCTION

Candidatus Phytoplasma solani (PHYPSO) and *Candidatus* Arsenophonus phytopathogenicus (ARSEPH) can induce diseases in several crop species and are commonly transmitted via insect vectors in the family *Cixiidae*. In potato, PHYPSO has long been known as the causal agent of potato stolbur, which is characterized by yellowing, wilting and rubbery tubers (Lindner *et al.* 2011). It belongs to the genus *Candidatus* Phytoplasma, which describes non-cultivatable, bacteria-like, cell wall-free organisms in the class Mollicutes. Various members have been reported as plant pathogens, causing disease symptoms such as witches' broom, dwarfism and phyllody. ARSEPH belongs to the proteobacteria, which is the largest and most diverse class within the domain of bacteria. The subgroup *γ*-proteobacteria colonizes the intercellular space or cytoplasm of their hosts and can modify the host metabolism in various ways. Interestingly, in contrast to phytoplasma, only a few plant pathogenic members are known, whereas several others are endosymbionts of arthropods. ARSEPH alone and in co-infection with PHYPSO can cause SBR (Syndrome Basses Richesses) in sugar beet (Bressan *et al.* 2008) and wilting in potato (Behrmann *et al.* 2023). In sugar beet, the disease is characterized by a low sugar content and reduced biomass in taproots, whereas in potato symptoms are similar to stolbur. Both pathogens cause losses in quantity and quality of yields, especially for potato and sugar beet. Their distribution is attributed to the presence of vectors, which varies strongly between regions (Duduk *et al.* 2024). In most cases, where samples from Germany were tested positive for both pathogens, *Pentastiridius leporinus* has been identified as the main vector (Duduk *et al.* 2023; Therhaag *et al.* 2024).
Several molecular methods are available for the detection of PHYPSO and ARSEPH in plants and vectors. In order to support official diagnostic laboratories in the implementation and evaluation of methods for DNA-based detection, the JKI organizes interlaboratory comparison studies, one already conducted in 2024 and a second in 2025. These investigations are targeting the process of disease detection in crops and planting material in the context of official testing and routine diagnostics.

MATERIALS AND METHODS

As part of the interlaboratory comparison study in 2024, the bacterial pathogens PHYPSO and ARSEPH were detected qualitatively, through molecular methods, in official diagnostic laboratories. Potato and sugar beet samples, infected with either one or both pathogens simultaneously, were provided and distributed by the Bavarian State Research Centre for Agriculture (Freising) and the Plant Protection Service Hesse (Regional Council Gießen, Wetzlar), respectively. Briefly, navel ends of potato were homogenized in Bioreba maceration bags. For sugar beet samples, tissue from the

vascular bundle area was used for maceration. Following two centrifugation steps, the pellet was resuspended and used for extraction of total genomic DNA (Kingfisher Flex, MagMAX Plant DNA Kit, Thermofisher). All extracts were then tested for the presence of PHYPSO and ARSEPH in conventional end-point PCR and real-time PCR. After aliquoting and randomization, samples were sent to the participating laboratories where detection was carried out according to established procedures. Various PCR-based approaches, including conventional PCR and nested PCR with sequencing of the products or real-time PCR, were performed.

RESULTS AND DISCUSSION

The results of the interlaboratory comparison study can be used by the participating laboratories as proof of the suitability of the established methods for detection of PHYPSO and ARSEPH. The conformity of results in the first interlaboratory comparison study in 2024 varied between participating laboratories. False negative results, due to ct-values above the individual limits of detection, were reported mostly from potato samples. This could be partially explained by a low concentration of the target DNA of PHYPSO and ARSEPH in the original samples. Only naturally-infected, pre-tested samples were used for extraction of DNA.

A second interlaboratory comparison study, which will be carried out between May and July 2025, will also focus on the process of DNA extraction. In order to increase the target DNA content and allow proper validation and verification of methods in the participating laboratories, approaches for artificial spiking of DNA extracts from other hosts and synthesized DNA fragments are currently being tested.

Different approaches are to be compared and suitable combinations of extraction method and PCR/real-time PCR protocols will be identified. Again, the DNA concentrations in the provided samples will be checked directly before dispatch, in order to supply sufficient amounts of DNA for several molecular tests. Furthermore, samples will be sent to the participating laboratories at 4 - 7 ◦C, in order to maintain the stability of the target DNA. Finally, all results from 2025 will be evaluated and insights from both tests, in 2024 and 2025, will be made available.

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Phytopathogenicus' and 'Candidatus Phytoplasma Solani'. Insects 15 (3). DOI: 10.3390/insects15030189.

Keywords: Stolbur, Arsenophonus, Detection methods, Potato, Sugar beet

16. qPCR-based quantification of Candidatus Arsenophonus phytopathogenicus and Candidatus Phytoplasma solani in potato (Solanum tuberosum L.) and their effect on yield parameters in southwest Germany

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INTRODUCTION

Pentastiridius leporinus is a known vector of two phloem-limited bacterial pathogens, Candidatus Arsenophonus phytopathogenicus (ARSEPH) and Candidatus Phytoplasma solani (PHYPSO) (Bressan et al., 2008). This planthopper species is rapidly spreading across large distances (Gatineau et al., 2002) and expanding its host range by adapting to new plant species for both transmission and reproduction (Behrmann et al., 2022). In 2022, the regions of Rhineland-Palatinate and Southern Hesse experienced the most severe potato crop damage on record. Shortly thereafter, P. leporinus was confirmed to transmit these pathogens to potatoes, resulting in the emergence of bacterial potato tuber wilt (BPW) disease (Behrmann et al., 2023; Therhaag et al., 2024). Currently, no effective practical strategies exist to control either the disease or its insect vector. As a result, ongoing research is focused on identifying high-performing potato cultivars that can maintain stable yields and desirable quality traits under pathogenic pressure.

MATERIALS AND METHODS

Cultivars and test sites

A total of 40 different potato cultivars were evaluated in two different test sites in Bürstadt (south Hesse, Bergstraße county) and in Ibersheim (south-east Rhineland-Palatia). The cultivars were categorized based on their primary purpose: table potatoes, French fries, or crisps. Maturity groups were defined as very early, early, mid-early, and medium-late.

Scorings in the field:

The assessment in the field was based on 1. Scroing of symptoms. 2. Measurement of key parameters including number of tubers, size, firmness, and fresh weight. 3.Vector Monitoring: Tracking the abundance and flight patterns of the vector P.leporinus using sticky traps.

Molecular analysis

DNA was extracted from pooled samples using a modified CTAB protocol. Quantitative real-time PCR (qRT-PCR) was performed according to the Behrmann protocol (2022). A standard curve was created using plasmid DNA standards to quantify the bacterial DNA in each sample. The copy number for each pathogen per cultivar and site was determined based on the Ct values and the standard curve parameters.

RESULTS AND DISCUSSION

Co-infection with both pathogens was predominant in 53.9% of samples from Bu¨rstadt and 70.8% from Ibersheim. A single infection with ARSEPH was found in 40% of Bu¨rstadt and 26.7% of Ibersheim samples, while PHYPSO was detected in only 1.7% and 2.5% of the respective samples. In Bu¨rstadt, 4.3% of the samples showed no presence of either pathogen. The threshold for a positive infection was set at 10 copies, and cultivars were categorized based on the average pathogen copy number. The environmental conditions in Ibersheim appeared to favor higher yields.

ANOVA showed that the site factor was the primary influence on yield variability, more so than the cultivar or pathogen. The pathogen type had a greater effect on infection levels than the cultivar, and the site did not significantly affect infection variability. While no correlation was found between pathogen copy numbers and the presence of rubbery tubers, however a significant negative impact on various yield parameters was observed.

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Keywords: Arsenophonus, Phytoplasma solani, qPCR, Yield, Potato, Germany

17. Identification and characterization of potential resistance genes in potato to control potato virus Y infection: interest of the N hypersensitive genes

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INTRODUCTION

In a constrained regulatory context for the use of phytosanitary products, genetic resistance is one of the important strategies of fighting against *Potato virus Y* (PVY) in potato crops. This potyvirus, transmitted by aphids, is the main problematic virus on potato, and one of the most damaging virus on crops (Scholthof et al, 2011).

In potatoes, two types of dominant monogenic resistance genes have been described: extreme resistance genes (*R*), which provide resistance to all PVY strains (Valkonen et al, 1996), and hypersensitivity genes (*N*), which provide PVY strain-specific resistance (Jones, 1990). Due to the absence of reliable molecular markers for the detection of the vast majority of these resistance genes, notably the *N* genes, little is known about the diversity of resistance genes present in the cultivated potato varieties, and their effectiveness in PVY resistance. The objectives of this study were to identify and define the resistance spectrum of a selected range of potato cultivars thanks to phenotyping.

MATERIALS AND METHODS

37 potato genotypes were phenotyped in a greenhouse by mechanical inoculation with 10 PVY isolates (4 PVYNTN, 2 PVYN-W, 4 PVYO). For each modality, 6 plants were inoculated. The dynamic of the disease symptoms expression was monitored, and serological tests were carried out on the mother plants and on the plantlets obtained after replanting the daughter tubers.

RESULTS AND DISCUSSION

Out of the 37 genotypes, no symptom was observed on 13 of them with all PVY isolates, 4 showed mosaic or necrotic symptoms in upper non-inoculated leaves and 20 expressed a reaction of hypersensitivity, with necrotic symptoms of different intensity on inoculated leaves and in upper parts of the plants according to the potato genotype/PVY isolate combination (Fig.1). Indeed, 6 to 7 days after the inoculation, first symptoms of necrotic local lesions or necrotic rings (called marita symptom) were noticed on the inoculated leaves of these potato genotypes. In most cases, these leaves dried and fell. Almost 10 days later, the necrosis extended to

Fig.1: Typical symptoms of hypersensitivity reaction. (a): local necrotic lesions; (b): necrotic rings on inoculated leaves; (c): systemic top necrosis

the stem and to the lower and upper leaves leading to the drying out of the whole stem and its death. This phenotyping highlighted that the HR reaction was expressed with PVY isolates of one or two strain groups, but not always with all PVY isolates of each of these groups.

Based on the expression of the HR reaction in relation to the inoculated PVY isolate, and according to the detection of virus into the plants, 6 phenotypic groups were obtained (Table 1).

Within 4 of these groups, for which *N* genes seem to be present, a wide diversity of sensitivity to PVY infection was observed depending on the plant genotype and the viral isolate. Our data showed that hypersensitive resistance is rarely associated with the absence of PVY infection in the plant. This resistance would be isolate-specific, not strain-specific, which may limit the interest in deploying such resistance genes in potato genotypes.

: % of plants expressing an HR reaction; : % of PVY-positive plants

Genotype	NTN				W		O				
	V1	V4	BP40	CCS-3	V2	V6	V3	BP28	Y^{O}-Holl702	CCS-12	
G-60	100 / 67	100 / 100	100 / 83	100 / 83	33 / 83	0 / 50	100 / 0	100 / 17	100 / 0	100 / 0	Group I (3 genotypes) *Ny-1*
G-10	100 / 0	100 / 50	100 / 0	100 / 0	67 / 67	0 / 0	83 / 0	83 / 17	100 / 0	100 / 17	
G-445	100 / 50	100 / 100	100 / 100	0 / 83	0 / 100	0 / 83	100	100	100	100	Group II (11 genotypes) *Ny-1 or Ny-2*
95T.118.2	83 / 0	67 / 17	100 / 0	100 / 0	0 / 83	0 / 0	50 / 17	50 / 33	100 / 0	83 / 33	
Nazca	33	67 / 83	100 / 0	50 / 17	0 / 100	0 / 67	0 / 0	0 / 100	0 / 17	0 / 100	Group III (1 genotype) *Nn*
Betty	0 / 100	0 / 100	0 / 100	0 / 100	0 / 100	0 / 67	0 / 17	67 / 17	17 / 0	67 / 50	Group IV (5 genotypes) *Nytbr*
G-63	0 / 100	0 / 100	0 / 100	0 / 100	0 / 100	0 / 50	100 / 17	67 / 100	100 / 0	67 / 100	
Acoustic	0 / 0	0 / 0	0 / 0	0 / 0	0 / 0	0 / 0	0 / 0	0 / 0	0 / 0	0 / 0	Group V (13 genotypes) *R*
Bintje	0 / 83	0 / 100	0 / 100	0 / 67	0 / 100	0 / 67	0 / 50	0 / 83	0 / 0	0 / 83	Group VI (4 genotypes) *Susceptible*
Fontane	0 / 100	0 / 100	0 / 100	0 / 100	0 / 100	0 / 83	0 / 100	0 / 100	0 / 17	0 / 100	

Table 1: Presentation of the 6 different phenotyping groups obtained after mechanical inoculation of 37 potato genotypes with 10 PVY isolates (4 PVY^{NTN}, 2 PVY^{N-W}, 4 PVY^{O}).

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Keywords: PVY, potato, resistance, phenotyping

18. Saikai 35, A Potato Cultivar Believed to Carry Rychc, Exhibits the Classical Characteristics of N Gene-Mediated Temperature-Dependent Responses upon Potato Virus Y Infection

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INTRODUCTION

Potato virus Y (PVY, *Potyvirus yituberosi*) is one of the most economically important viruses affecting potato production in the world. The virus is mainly managed through phytosanitary measures in combination with practices such as mineral oil application. Clearly, using PVY-resistant cultivars is the most economical and effective means for PVY management, especially for countries where the seed certification program is not established or implemented. Two types of resistance, namely hypersensitive resistance (HR) and extreme resistance (ER), have been recognized in potato. The former is characterized by localized or systemic necrosis upon PVY infection and is conferred by *N* gene; whereas the latter is featured by factual immunity, which is manifested by the lack of symptoms, and limited or lack of pathogen replication and spread (Karasev and Gray 2013), and is mediated by *R* gene. A number of the resistance genes including *Rysto*, *Ryadg* and *Rychc* as well as *Ny, Nz, Nc, Ny 1, Ny 2,* and others (Singh et al. 2008; Szajko et al. 2008; 2014) have been introgressed into potato varieties, thus providing a certain level of protection against PVY (Nie et al. 2016). Although *Rychc* has been grouped into the *R* gene category, the cultivars Konafubuki and Sakurafubuki, which are believed to carry the gene, developed local lesions upon mechanical inoculation with isolates belonging to O, N or NTN strains (Ohki et al. 2018), especially at higher temperature (28oC). This raises a question on whether other cultivars bearing the same gene exhibit similar responses to PVY and, moreover, on whether resistance in these cultivars are truly controlled by an *R* gene or an *N* gene. Bearing this in mind, cultivar Saikai 35, which was bred for its resistance to PVY and to cyst nematodes (Mori et al. 2012), was investigated for its responses to different strains of PVY under various conditions.

MATERIALS AND METHODS

Virus-free (VF) *in vitro* plantlets of potato cultivars Saikai 35 (Mori et al. 2012) and Shepody (Young et al. 1983) were planted in Pro-Mix potting soil in 6-inch (15.2 cm) pots in the greenhouse under 18-24oC with a photoperiod of 16/8 (L/D) and humidity of 75%. The plants were inoculated mechanically with various isolates of PVY (specifically, PVYO-FL, PVYO-RB, PVYN:O-MB58, PVYNTN-SI, and PVYN-Jg) or with 10 mM phosphorate buffer (pH 7.5; mock) at 5-leaf stage as described previously (Nie et al. 2012) and maintained in the same greenhouse or transferred to growth chambers with different temperature regimes for various periods of time. The plants were tested for PVY using double-antibody sandwich-ELISA as described previously (Nie et al. 2012) at various time points. In addition, one-step quantitative RT-PCR (RT-qPCR) with SYBR® Green (Biorad Canada) and a newly designed primer pair (PVY qPCR F2, 5'-GAACACAGAGAGGCACACCA-3' and PVY qPCR R2, 5'-GGAAAAGCCAAAATACTTACTGC-3'; fragment size, 136 bp) was employed in some samples following the manufacturer's guideline. All experiments were repeated at least two times.

RESULTS AND DISCUSSION

Saikai 35 is reported to possess *Rychc* and therefore considered to be extremely resistant to PVY (Mori et al. 2012). Diagnostic testing for *Rychc* using PCR markers MG64-17 (Li et al. 2022) and 1648F24/1648R22 (Akai et al. 2023) yielded positive results. To validate the resistance, a serial inoculation experiments with different PVY strains including PVYO, PVYN, PVYN:O and PVYNTN were carried out initially in the greenhouse in different seasons. In the greenhouse in the winter season (November-February), local lesions started to emerge on PVYO-inoculated plants at 5 days post-inoculation (dpi), and systemic lesions in new branches started to occur at 28 dpi. PVYN:O and PVYN did not incite any visible local or systemic symptoms. PVYNTN, on the other hand, induced local lesions but failed to induce systemic necrosis. Postharvest examination of tubers from all of the inoculated Saikai 35 plants, mock included, found one progeny tuber (out of 10) of the PVYO-inoculated plants exhibiting internal necrosis. The daughter plant resulting from this tuber started to develop systemic leaf necrosis soon after its emergence, tested PVY positive by ELISA, and died 14 days post-emergence. In the spring season (Apr-July), all PVY strains induced local lesions, but only one out of four of each of PVYO and PVYNTN inoculated plants developed systemic necrosis. ELISA testing on the inoculated leaves at 12 dpi yielded a positive result, though at relative low absorbance values. ELISA testing on the upper uninoculated leaves produced negative results for all inoculated plants at 12 and 35 dpi on all plants regardless of the strains. Nevertheless, one of each of PVYO and PVYNTN inoculated plants developed systemic necrosis on lately emerged leaves and flower petals, which tested positive for PVY by qPCR. These results suggest the cultivar may possess a HR-like response to PVY infection. As the temperature fluctuates considerably in the greenhouse, especially in the April-July experiment, further investigation with different temperatures was thus warranted.

In the growth cabin at 22oC, local lesions were induced by all PVY strains, but no systemic symptoms were incited, and the plants tested negative for PVY by ELISA. At 30oC, on the other hand, no local lesion nor systemic necrosis was induced by any PVY strains, but the plants tested high-positive for PVY by ELISA. A swap of the plants between 22oC and 30oC led to a rapid development of systemic necrosis and tissue death in plants that were pre-grown at 30oC and transferred to 22oC. The responses of the plants to PVY at different temperatures ranging between 22oC and 30oC varied, ranging from local lesions only (22oC) to local and systemic lesions (26oC, 27oC and 28oC) and to mild leaf mosaic with completely systemic infection (29oC and 30oC). It is also noteworthy that the HR at 22oC is manifested by local lesions and an ELISA-undetectable level of PVY in the plants; but at 26oC and 27oC, the HR is comprised of both local and systemic necrosis along with a high PVY titre.

Taken together, the results demonstrate that Saikai 35 shows a hypersensitive resistance phenotype at 22oC - 28oC and a complete susceptible phenotype at 29oC and 30oC, similar to that in tobacco carrying the N gene against tobacco mosaic virus (TMV) (Whitham et al. 1996), indicating that the resistance in the cultivar against PVY is mediated by an N gene rather than an R gene.

ACKNOWLEDGEMENTS

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Keywords: Potato virus Y, resistance, cultivar Saikai 35, temperature, dependent response

19. Study of Variety-Specific Responses to the Necrotic Strain of PVYNTN

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INTRODUCTION

Tuber necrotic ringspot disease (PTNRD) is responsible for severe quality degradation of potato tubers on a global scale and is induced by the PVY recombinant PVY^{NTN}. The resulting brown lesions significantly diminish the marketability of affected tubers, posing a persistent challenge to potato production and trade.

MATERIALS AND METHODS

Over the past decade, we evaluated symptom development in approximately 80 potato cultivars through biennial field trials. Cultivars were evaluated in biennial field trials under controlled inoculation conditions or in naturally infected environments. Symptom expression was recorded during storage.

RESULTS AND DISCUSSION

This long-term dataset allowed us to assess both cultivar-specific susceptibility and year-to-year variability. Results show significant differences in PTNRD expression among cultivars and across seasons, indicating strong genotype and environmental effects.

20. Long-Term Assessment of Variety-Specific Yield Losses in Potatoes Caused by PVY (and PLRV)

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Potato virus Y (PVY) transmitted by aphids, remains one of the most impactful viral pathogens affecting potato production globally, including in Switzerland. It causes substantial yield losses and plays a central role in the legal framework for seed potato certification.
Since 1991, Agroscope has conducted a long-term field trial near Zurich to monitor the impact of PVY—and to a lesser extent, Potato Leafroll Virus (PLRV)—on a wide range of potato varieties. For each variety, two types of seed material are compared annually: farm-saved seed produced on-site and certified class A seed potatoes. Trials are deliberately run without mineral oil treatments or premature haulm killing to allow natural virus transmission and symptom development via aphids.
Each year, visual assessments of virus symptoms (percentage of infected plants) and yield measurements are recorded. This setup enables the evaluation of virus susceptibility (based on symptom expression) and associated yield losses for officially listed Swiss varieties. To facilitate year-to-year comparisons, the estimated yield reduction per 1% virus infection is calculated, providing a standardized indicator of virus impact.
Preliminary results illustrate the value of long-term monitoring to track trends in virus susceptibility and related yield effects at the variety level. This trial contributes to the development of national variety recommendation lists, by supplying robust field-based data under natural infection conditions.

21. The role of endosymbionts in mediating plant virus transmission by insect vectors

Keynote 3

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Plant viruses frequently alter traits of both their host plants and insect vectors to enhance their own transmission. Recent studies indicate that these virus-induced effects can be modulated by other microbes, including mutualistic plant symbionts. Insect vectors themselves also harbor microbial endosymbionts, which profoundly influence insect biology and interactions with host plants. These endosymbiont effects are likely to influence plant virus dynamics; however, such interactions remain largely unexplored. In this talk, I will present our recent work showing how aphid endosymbionts influence virus–vector–host interactions in ways relevant to disease spread. I will discuss our findings on how these endosymbionts affect aphid vector biology, behavior, and virus transmission efficiency, as well as more recent results on the underlying mechanisms and on aphid vector biocontrol using natural enemies. I will also place these findings in a wider context, outlining research priorities to better understand the impacts of vector endosymbionts on the ecology and evolution of plant viruses, including in potato pathosystems.

Key words: Insect symbionts, Disease ecology, Microbial ecology, Vector–microbe interactions, Symbiosis, Aphid-borne diseases

Plant–aphid interactions

22. Gut content analysis of Myzus persicae reveals insights into viral reservoirs of sugar beet viruses

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In Switzerland, sugar beet (Beta vulgaris subsp. vulgaris) is a significant agricultural crop, cultivated across 16,000–18,000 hectares primarily along the Central Plateau region. Following the ban on neonicotinoid seed coatings in 2019, a marked decline in sugar yields was observed in 2020. Massive outbreaks of virus yellows and the syndrome “basses richesses” (SBR) were thought to be responsible for the major loss of sugar beet yields in the western cantons of Switzerland. The primary viral threats to sugar beet are vectored by aphids, including beet western yellows virus (BWYV), beet mild yellowing virus (BMYV), beet chlorosis virus (BChV), beet yellows virus (BYV), and beet mosaic virus (BtMV). A nationwide survey conducted in 2020 revealed high incidences of BYV and BChV, while BMYV was only detected at two sites in the western region. Notably, no instances of BWYV or BtMV were found. Subsequent surveys in 2021, 2022, and 2023 indicated a decrease in yellowing symptoms compared to 2020, suggesting a potential shift in viral dynamics.

Among the viruses detected, the aphid *Myzus persicae* can transmit all three: the poleroviruses BChV and BMYV are transmitted through a persistent, circulative mechanism, while the closterovirus BYV is transmitted through a non-persistent mechanism. In addition, *Aphis fabae* can also act as a vector for BYV.

In order to investigate the viral reservoirs and host interactions, alate aphids captured by suction traps early in the season were analysed. This approach demonstrated the presence of viruses exclusively in *M. persicae*, underscoring its role as a primary vector. Additionally, molecular gut content analysis was conducted to identify alternative host plants where these viruses may overwinter. DNA was extracted from *M. persicae*, and the ITS2 region of the plant was barcoded and sequenced using a MinION Flongle cell. A total of 114 aphids were tested in 2021, with 96 sequenced, and 349 in 2023, with 206 sequenced. The results revealed a diverse range of 50 different plant species across 19 families, predominantly including *Brassicaceae*, *Solanaceae*, and *Rosaceae*. However, the genetic proximity of some plants limited the ability to distinguish them at the species level, particularly among *Brassica*, due to their evolutionary relationships. Additionally, the sensitivity and potential contamination associated with the nested PCR method unfortunately compelled the removal of all plant samples that were handled by the laboratory from the results to ensure accuracy.

Overall, this study provides valuable insights into the complex interactions between *M. persicae* and its variable hosts, contributing to a better understanding of viral dynamics in sugar beet cultivation. The innovative methodology employed here not only enhances the detection of viral reservoirs but also holds significant potential for broader application across various pathosystems, paving the way for improved management strategies in agricultural practices.

23. Mitigating Potato Virus Y Spread Through Sunflower Intercropping in Seed Potato Fields

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INTRODUCTION

Potato virus Y (PVY) is one of the main causes of economic losses in the seed potato sector in Continental Europe. According to estimates by Brice Dupuis *et al*. (2024), these losses amount to approximately €96 million annually across the European Union. Currently, PVY management relies primarily on the regular application of mineral oil to the foliage. Although this method provides a certain level of control, its effectiveness is limited, particularly at the beginning of the growing season when vegetative growth is rapid. Therefore, there is a need to complement this approach with additional protective strategies, especially during the early stages of plant development.

Among the alternative methods explored, the use of intercrops such as oats or vetch has shown promising results in previous studies (Dupuis *et al*., 2017). In the present study, we evaluated the potential of sunflower as an intercrop, used either alone or in combination with mineral oil, to reduce the spread of PVY in potato fields.

MATERIALS AND METHODS

Two field trials were conducted: one in Nyon, Switzerland, and the other in Achicourt, northern France. Sunflower was sown at the same time as the potatoes was planted and was destroyed shortly before potato canopy closure to avoid interfering with potato growth. Mineral oil was applied at regular intervals following standard agronomic practices during the virus-sensitive period.

The experimental design allowed for comparisons between the following treatments: sunflower alone, mineral oil alone, their combination, mineral oil combined with straw mulching, and an untreated control. One progeny tuber per plant was analyzed by DAS-ELISA after harvest to detect the presence of the virus.

RESULTS AND DISCUSSION

The results from both field trials showed that using sunflower as an intercrop, without mineral oil, did not significantly reduce PVY incidence. Furthermore, the combination of sunflower and mineral oil did not offer improved efficacy compared to mineral oil alone. These findings suggest that, under the conditions of this study, sunflower is not an effective intercrop for enhancing PVY control.
The role of aphid flights, the primary vectors of the virus, is also discussed as a potential factor influencing the observed results.
In conclusion, although sunflower did not prove effective in this context, the study highlights the importance of continued research into other plant species as intercrops. Such approaches may ultimately help improve protection of potato crops against PVY and complement existing control strategies.

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24. Increasing Rejected Areas in Swiss Seed Potatoes Production from 2016 to 2024 are Influenced by Viral Infections and Climate Change

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INTRODUCTION

Seed potatoes certification is a control process aimed at ensuring the production of high-quality seed potatoes and to limit the spread of diseases. Among these pathogens, viruses can easily transmit through the vascular system, from one generation to the next and are subject to post-harvest testing and in particular, Potato virus Y (PVY) and Potato leafroll virus (PLRV). Dupuis *et al.* (2024) estimated the average yield loss of 223 kg/ha for 1% PVY infection that comes with economic impacts of about an estimated at 187 M EUR in the European Union for the whole potato sector. Numerous species of aphids transmit these viruses respectively in a nonpersistent (PVY) and persistent manner (PLRV).

MATERIALS AND METHODS

In Switzerland, to rigorously monitor the sanitary status of the lots, each multiplication plot is inspected two to three times during the growing season. Since 2016, the prevalence of PVY and PLRV has been monitored annually in the laboratory through molecular analysis as part of the seed potato certification process. 220,000 tubers were collected per year out of 1,000 lots, with an average of 200 tubers per lot. Then, 10,000 RT-qPCR were performed on tuber samples to detect PVY and PLRV (Schumpp *et al.*, 2021). These data allowed us to assess the evolution of the prevalence of these viruses in Switzerland over the last nine years.

RESULTS AND DISCUSSION

Each year, the seed potato lots are mostly infected by PVY, but the infection rate of PLRV and of infections by both PVY and PLRV tend to increase. We note that from 2019 to 2023, 12 to 21% of the lots were infected by both PVY and PLRV, although 2% to 5% of the lots were infected by both PVY and PLRV from 2016 to 2018 and in 2024 (Figure 1). Some varieties are more susceptible to these viral infections, as reported by Dupuis *et al.* (2019). The data indicate that, the rejected areas tend to increase in Switzerland since 2016, not only due to virus infection but also because of blackleg (*Pectobacterium* spp. and *Dickeya* spp.) and late blight (*Phytophthora infestans*), depending on the weather conditions. Given the increase of rejected areas, which could rise further in the context of climate change, improved protection and new control strategies must be developed to limit pathogen infections.

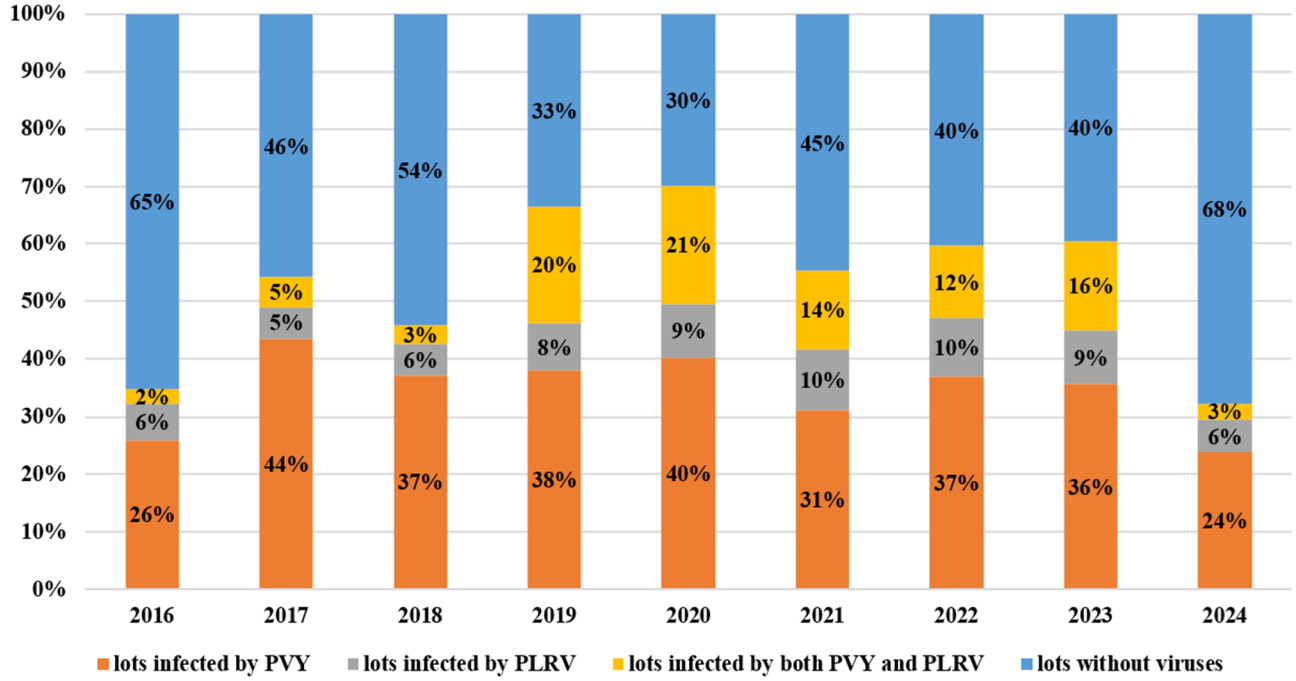

Figure 1: Prevalence of PVY and PLRV in Switzerland from 2016 to 2024

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25. Long-Term Monitoring of Potato Virus Y In Estonian Seed Potatoes and an Evaluation of Susceptibility of Potato Varieties

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INTRODUCTION

The potato (*Solanum tuberosum*) is one of the most important agronomic crops worldwide, and the leading vegetable crop in Estonia. Seed potatoes are consistently monitored in certification process to ensure virus free or nearly virus free propagation material. Potato virus Y is the most significant virus affecting potato crops. The current research report describes the incidence of potato virus Y and the susceptibility of potato varieties grown during the regular seed potato monitoring from 2007 to 2022 in Estonia.

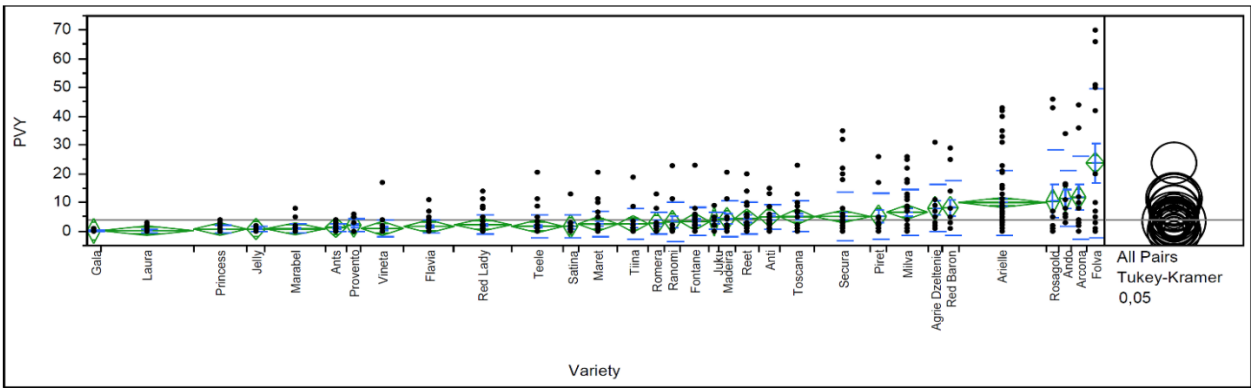
MATERIALS AND METHODS

The seed potato samples were collected for virus testing as part of the potato seed certification system by inspectors from the Estonian Agriculture and Food Board (EAFB). In total, 1023 seed lots were tested for PVY over the period from 2007 to 2022. At the beginning of the surveys, ELISA-based virus detection was used, and from 2015 onward, Real-Time RT-PCR was implemented for PVY detection (van der Sman et al 2025a). 1041 PVY positive samples were used for molecular typing of PVY strains using the primer set from Lorenzen et al (2006) in multiplex RT-PCR. Statistical analyses were performed using JMP10.0 and Microsoft Excel software, the datasets were tested using the least squares multiple linear regression model. The resistance/susceptibility to PVY was evaluated for the 32 most frequently grown foreign and Estonian varieties. Total RNAs of 6 leaf samples originating from individual seed tubers grown out for further studies during the first survey period, and nine total RNAs of tuber samples collected after 2015 were sequenced using Illumina MiSeq at The Food and Environment Research Agency (FERA Ltd, UK). Library preparation and sequencing followed the protocol described by Fowkes et al. (2021). Bioinformatic analysis was done with in-house pipeline (van der Sman et al 2025b).

RESULTS AND DISCUSSION

The overall average level of PVY in potato grown for Super Elite certification 2007 to 2022 was 2.43%, with the highest level recorded at 6.56% in 2011, while no PVY was detected in 2020. The average PVY incidence in lower seed classes was 3.92%, with the lowest percentages (under 1%) recorded in 2013, 2014, 2017, and 2022; the highest PVY prevalence (8.29%) was observed in 2021. Multiple linear regression analysis showed that weather conditions were statistically significant ($p= 0.0005^*$; $r= 0.1675$). Temperatures in July ($p< 0.0001^*$) and August ($p= 0.0299^*$), and precipitation in July ($p= 0.0266^*$) were significant for PVY incidences. One-way ANOVA analysis on the most common 32 varieties confirmed that the variety played a crucial role in PVY infection rates ($p< 0.0001^*$;$r= 0.5193$). There was a clear difference between resistant and susceptible varieties (Fig. 1). PVY infection rates enabled to divide potato varieties into five resistance/susceptibility classes. Notably, all varieties within the very high resistance group and those in the very susceptible group had statistically significant p-values. Varieties with very high PVY resistance had negative t -ratios, and very susceptible varieties had positive t-ratios in the linear regression analysis. Neither temperature in July and August nor precipitation in July had a statistically important effect on overall PVY rates in very resistant and resistant varieties. However, these environmental variables did affect some susceptible and very susceptible varieties.

Fig. 1. One-way ANOVA analysis of PVY incidences (%, shown in y-axis) in tested individual seed lots by potato variety (shown in x-axis). The width of green diamonds represents the group size of the variety. The grey line shows a grand mean. Black dots indicate individual seed lots. Blue lines are at ± one or two STDEV; dots exceeding the blue lines show the outliers



The strain genotyping was applied to 1041 PVY-positive samples that were identified in pre-screening during the three survey periods in the frame of long-term monitoring: 2011 to 2013, 2016 to 2018, and 2020 to 2022. It was confirmed that the prevalent PVY strains in Estonia are $PVY^{N\text{-}Wi}$ and PVY^{NTN}. Statistical analysis showed that there was statistically significant ($p<0.05$) negative trend for PVY^{NTN}. The results showed that variety plays statistically significant role on PVY strain composition (the p- values were 0.01* for PVY^{NTN} and 0.0021* for $PVY^{N\text{-}Wi}$). RT-PCR genotyping revealed that more than 10% of isolates showed inconclusive results. 12 of such samples and, in addition, one typical PVY^{NTN} and two typical $PVY^{N\text{-}Wi}$ genotype samples were sequenced to clarify the results. HTS resulted in assembly of 7 near full genomes of Estonian PVY isolates, and it was strongly dependent on initial PVY concentration in the sample. Surprisingly, all assembled genome sequences had typical genomes of PVY^{NTNa} or $PVY^{N\text{-}Wib}$. In silico primer mapping revealed that all used primers mapped correctly to the assembled genome sequences, indicating that the extra bands seen in RT-PCR were most probably originating from potato genome.

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26. Historical Perspective of Challenges in Potato Breeding and Seed Potato Production Related to Virus Testing in Slovenia

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Potato production has been and still is an important part of agricultural production in Slovenia. The first planned intervarietal crossings in Slovenia began at Agricultural Institute of Slovenia in 1949 and led to the release of 14 Slovenian cultivars of potato by 1990. A major challenge for Slovenian potato production was the emergence of NTN strain of potato virus Y (PVYNTN) in the late 1980s. The use of Slovenian variety 'Igor', which was released in 1965 and very popular in Slovenia, was completely abandoned within a few years, mainly due to its high sensitivity to PVYNTN and significant tuber symptoms. Persistent disease pressure, particularly from viruses, forced breeders and scientists to produce more resistant cultivars. Therefore, one of the main goals of the potato breeding program at Agricultural Institute of Slovenia from 1993, was breeding for extreme resistance to PVY. From 2004 15 new Slovenian potato cultivars ('KIS Tamar', 'KIS Mangart', 'KIS Blegoˇs', 'KIS Razor', 'KIS Savinja', 'KIS Slavnik', 'KIS Vipava', 'KIS Krka', 'KIS Mura', 'KIS Kokra', 'KIS Sotla', 'KIS Mirna', 'KIS Sora', 'Pˇsata', 'Bistra') were released, all extremely resistant to PVY (Dolniˇcar et al., 2023). Moreover, 'Bistra' variety is resistant to potato leaf roll virus (PLRV) as well. Throughout this period, virologists supported the breeding and seed production programs by monitoring the health status of potato material both *in vitro* and *in vivo*. Routine virus screening was performed using DAS-ELISA, targeting six major potato viruses: PVY (*Potyvirus yituberosi*), potato virus A (PVA, *Potyvirus atuberosi*), potato virus S (PVS, *Carlavirus sigmasolani*), potato virus X (PVX, *Potexvirus ecspotati*), potato virus M (PVM, *Carlavirus misolani*) and PLRV (*Polerovirus PLRV*).

After the introduction of varieties extremely resistant to PVY, infections with PVS and PVM started to increase in breeding material and seed potato as well. These viruses, however depending on the strain virulence, have no or low influence on potato yield and quality. Some new variety candidates were subjected to meristem tip culture and chemotherapy for virus elimination. The need for detection of low virus concentrations in plants after virus elimination demanded the introduction of more sensitive detection methods, particularly quantitative PCR (qPCR). The qPCR for PVY based on Kogovsek et al. (2008) was introduced for research purposes followed by qPCR for PVS and PVM. Consistent and vigorous testing of all breeding and seed production material enabled effective management of PVS and PVM infections in Slovenian potato production.

During all this period, PVA and PVX were not detected in the material under evaluation, and PLRV incidence remained minimal. However, in 2020 the infections with PLRV increased significantly in variety 'KIS Razor'. In order to better control the PLRV infection in seed stocks, and prevent the infection of other cultivars, cultivar 'KIS Razor' was terminated from the seed and ware production in 2021. After that the incidence of PLRV dropped, but still remained higher than in years before 2020. Due to the persistence of PLRV, a qPCR for detection of this virus was introduced in 2024 (Lacomme et al., 2015). All qPCR tests were used for detection of potato viruses in potato leaves and potato *in vitro* plants.

However, nowadays needs for faster and more sensitive virus diagnostic is posing a need for implementation of new methods also for postharvest control. From grow-out test and DAS-ELISA

testing, we are currently in the process of transitioning to qPCR detection of viruses in tubers. The paper by Schumpp et al. (2021) is used as the basis of this method. These improvements will enable us to prevent virus spread and maintain high yield in potato production.

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Keywords: potato breeding, seed certification, potato viruses, epidemiology

27. Current status of Potato virus Y (PVY) in the Belgian seed potato sector

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Potato virus Y (PVY) is a major viral pathogen contributing to significant economic losses in potato production globally. Transmitted primarily by aphid vectors in a non-persistent manner, PVY encompasses a complex of genetically and biologically diverse strains, each associated with varying symptomatology and degrees of severity across different potato cultivars.

In Belgium, the last comprehensive study of PVY isolates dates back to 2010. Since then, high infection levels have occasionally been detected in seed potato lots despite roguing based on visible symptoms. This discrepancy is likely due to asymptomatic infections escaping detection in the field. Symptom expression is influenced by a complex interplay between PVY strain, cultivar, and environmental conditions, complicating virus management in seed production.

To update the understanding of PVY strain dynamics within Belgium, molecular strain typing analyses were conducted by RT-PCR on PVY-positive samples (determined by ELISA) collected from certified seed potato fields during the 2023-2024 growing season. The resulting data provide critical insights into the evolving epidemiology of PVY in Belgium and underscore the need for adaptive management strategies in seed potato certification and production.

Keynote 4

28. The Detection and Diagnosis of Potato Viruses – Are We Making Progress?

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Plant virus diagnostics has been revolutionized since the advent of molecular diagnostic methods. Methods such as real-time PCR, offered enhanced sensitivity and speed of turnaround by comparison to serological tests, and have now become ”routine” in their application. More recently the development of high throughput sequencing (HTS) has revolutionized plant virology, with applications such as virus discovery, resolving diseases of unknown aetiolgy, and post-entry quarantine compliance testing. However, with a few notable exceptions, the uptake of these methods in potato virology has been limited. The use of HTS in plant virology for ”front line” diagnostics will be discussed with specific examples from Fera used to highlight the developments of this technology for plant health applications. Drawing from case studies both within potato virology and from broader plant health, examples will be discussed on the use of the technology to determine the cause of an unknown disease (Adams et al., 2014), and to influence plant health regulations on both specific cases (Hammond et al., 2021) and those with broader implications for policy makers (Fuentes et al., 2022; Fuentes, Gibbs, Adams, et al., 2021; Fuentes, Gibbs, Hajizadeh, et al., 2021). The challenges to broader application of HTS will also be discussed, with an emphasis on the changes needed in philosophical as well as practical approaches in potato virus diagnostics, where there is a shift needed from the researcher, the plant health risk assessor, and the policy maker.

Keywords: High Throughput Sequencing, Potato, Virology

Diagnostic

29. Development of a Duplex IC-RT-qPCR for Large-Scale Detection of Potato Mop-Top Virus in Dormant Potato Tubers

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INTRODUCTION

Potato mop-top virus (PMTV) is a member of the genus *Pomovirus*, family *Virgaviridae* with a genome comprises three single-stranded positive-sense RNAs. PMTV affects the quality of potato tubers by causing internal and external tuber necrosis (spraing) in sensitive potato cultivars, which appear as necrotic flecks or concentric rings and arcs. The reliable and large-scale detection of PMTV, especially in asymptomatic tubers, is a crucial step toward the effective control of PMTV. In the current research, we developed a duplex immunocapture reverse-transcription quantitative polymerase chain reaction (IC-RT-qPCR) assay for the large-scale detection of PMTV.

MATERIALS AND METHODS

Potato plants of three cultivars, Yukon Gold, Castle Russet, Soraya, and four breeding lines were infected with PMTV in a greenhouse to obtain infected tubers. The immunocapture step was performed using different dilutions of PMTV-specific IgG antibody from Bioreba AG. The cDNA was prepared using two PMTV-specific reverse primers. The qPCR was performed using two primer-probe sets targeting the CP-RT (RNA-CP) and TGB3 (RNA-TGB).

RESULTS AND DISCUSSION

The IC step eliminated the need for RNA extraction kits, making this assay appropriate for large scale tuber testing. To enhance the reliability of the current assay and reduce the chance of false negatives, a duplex format was used by deploying two primer-probe sets, including a previously reported primer-probe set targeting the RNA-CP, and a newly designed primer-probe set targeting a conserved region of RNA-TGB of PMTV genome. We also determined that peels from the stem end of the tubers were more likely to test positive for PMTV than bud end peels or lateral tuber cores. The duplex IC-RT-qPCR will provide a reliable and sensitive tool for the large-scale detection of PMTV in dormant tubers and will help safeguard potato movement.

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30. Simple and efficient PCR-based detection of phytoplasmas: validation and application in potato production systems

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Abstract

Phytoplasmas (*Candidatus* Phytoplasma spp.) are membrane-less bacteria affecting a wide range of crops, including potatoes (*Ca.* P. solani aka stolbur) and fruit trees (*Ca.* P. mali on Malus spp.). Transmitted by insect vectors such as leafhoppers and psyllids, they cause symptoms ranging from witches' broom in apples to aerial tubers in potatoes, leading to significant yield losses. Several generic diagnostic methods have been developed for phytoplasma detection, with PCR-based approaches allowing both detection and identification in a single test. However, many methods require nested PCR, which, while sensitive, is labor-intensive and prone to cross-contamination.

In this study, we present the development and validation (following EPPO PM7/98 guidelines) of a simple, conventional PCR targeting the well-characterized 18S rRNA gene. This method offers a robust, sensitive, and cost-effective solution for diagnosing phytoplasmas in plant samples. The method was tested against various phytoplasma species and showed sensitivity comparable to a reference qPCR method. Specificity was confirmed across a range of phytoplasmas, including regulated species, with no amplification observed in uninfected plants from different genera. Although the 1 kb amplicon generated may not always distinguish closely related species, we validated an alternative PCR method targeting the SecA locus, useful for distinguishing such species.

While *Ca.* P. solani has been reported only once in Belgium, recent detections in northeastern France might indicate a rising prevalence. With increasing temperatures favoring leafhopper populations and the expansion of vine cultivation-another host for stolbur-this pathogen could pose a growing threat to potato production in Belgium.

31. Multiplex RT-qPCR for Strain-Specific Detection of Potato Virus Y

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Potato virus Y (PVY) is one of the most damaging viruses in potato cultivation, significantly impacting yield and seed certification. Since 2016, Swiss seed certification has relied on RT-qPCR for PVY detection due to its superior sensitivity compared to ELISA. However, current assays only confirm the presence of PVY without identifying the infecting strain, omitting critical epidemiological information.

PVY exists in several biologically distinct strains (PVY-O, PVY-N, PVY-NTN, PVY-N-Wilga), and tracking their prevalence is essential for disease management. This project aimed to develop a multiplex RT-qPCR assay capable of distinguishing these four main PVY strains in a single reaction. The assay uses a universal primer pair flanking a key recombination region of the PVY genome (nt 9000–9200) combined with strain-specific TaqMan probes, each labeled with a distinct fluorophore. Probes were designed to target unique nucleotide polymorphisms specific to each strain.

Initial testing with purified reference isolates revealed specificity issues in some probes. The final configuration included:

- PVY-O-specific probe (FAM)
- PVY-N-specific probe (Cy5.5)
- A dual PVY-NTN/N probe (ROX)
- A dual PVY-Wi/O probe (Cy5)

This design allows unique Cq profiles per strain and the detection of co-infections. Specificity was confirmed across PVY isolates and against a panel of other potato viruses (PVA, PVX, PLRV, etc.). No cross-reactions were observed.

The assay maintained high sensitivity (LOD \~10-5 dilutions, Cq \~33–35) and showed comparable performance to the universal PVY RT-qPCR. Multiplexing had minimal impact on Cq values. The internal control ensured reliable reaction performance.

The assay was applied to field samples from the 2018 Swiss certification program. It confirmed the predominance of recombinant strains PVY-N-Wilga and PVY-NTN. Classic PVY-O was less frequent. Several co-infections were identified an advantage over conventional assays.

This tool offers both qualitative (strain identity) and quantitative (viral load) insights and is suitable for certification and epidemiological monitoring. It allows better understanding of PVY dynamics, supports targeted management strategies, and fills a major gap in molecular diagnostic capabilities for potato viruses.

Beyond technical success, the development journey highlighted critical lessons: challenges in probe design, balancing specificity and sensitivity, and integrating genomic knowledge into applied virology. This multiplex RT-qPCR lays the groundwork for more nuanced and actionable virus diagnostics in seed certification and beyond.

32. Development of a Luminex xTAG assay for the determination of aphid species and simultaneously detection of plant viruses

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INTRODUCTION

Aphids are plant-infesting, phloem-feeding insects. A limited number cause damage for agricultural crops by feeding or indirectly by transmitting certain plant viruses. In the case of seed potato growing, 15 species are identified as important vectors for potato virus Y (PVY). For the monitoring of aphid flights and determination of the vector pressure, migrating aphids are caught in yellow water traps or Rothamsted suction traps, and subsequently determined to species visually. As an alternative for visual species determination, a molecular test was developed for these 15 aphid species in a multiplex Luminex platform, including simultaneously detection of a number of plant infecting viruses.

MATERIALS AND METHODS

For this project, the Luminex xTAG technology was used to develop an assay for 32 targets. The targets were chosen on the COX1 gene of aphids, or on the genomes of PVY and potato leaf roll virus (PLRV). After DNA and RNA extraction, generic amplification of the COX1 gene with outer primers, and specific cDNA amplification for PVY and PLRV was carried out with an RT-PCR. Subsequently, target specific primers for extension (TSPE) were used to detect specific parts of the amplicons in a isothermal elongation reaction. These biotinylated TSPE primers are tagged with a specific sequence which can anneal to a complementary sequence attached to a Luminex bead. These beads have specific colours which can be identified in the Luminex MagPix, with simultaneous detection of fluorescence for a positive detection of the target after adding a fluorescent reporter which binds to the in the TSPE-primers incorporated biotin.

RESULTS AND DISCUSSION

Samples of the 15 aphid species which were visually determined to species by the Dutch Inspection Service and were used as reference material. The Luminex system was able to detect all 15 species and matched the visual determination. Samples were only considered positive when the generic aphid TSPE had a Luminex value above the calculated threshold, using Luminex software.
Field samples of aphids caught in an Ashby funnel trap and control samples of *Myzus persicae* from a lab colony were analysed using the designed outer primers and TSPEs. As the specific binding sites for the TSPE primers sometimes overlap in other aphid species than the 15 of interest, it is until now only possible to analyse single aphids per sample.

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