Virulence and fungicide sensitivity of the most prevalent genotypes collected in Belgium in 2019-2021

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Summary
A total of 281 isolates of *Phytophthora infestans* were collected in the southern part of Belgium (Wallonia) in several potato fields, volunteers, dumps and field trials during the years 2019, 2020 and 2021. The isolates were analyzed using standardized 12 plex Euroblight SSR genotyping (Euroblight monitoring). Most of isolates were tested for several phenotypic characteristics, such as virulence and fungicide resistance.

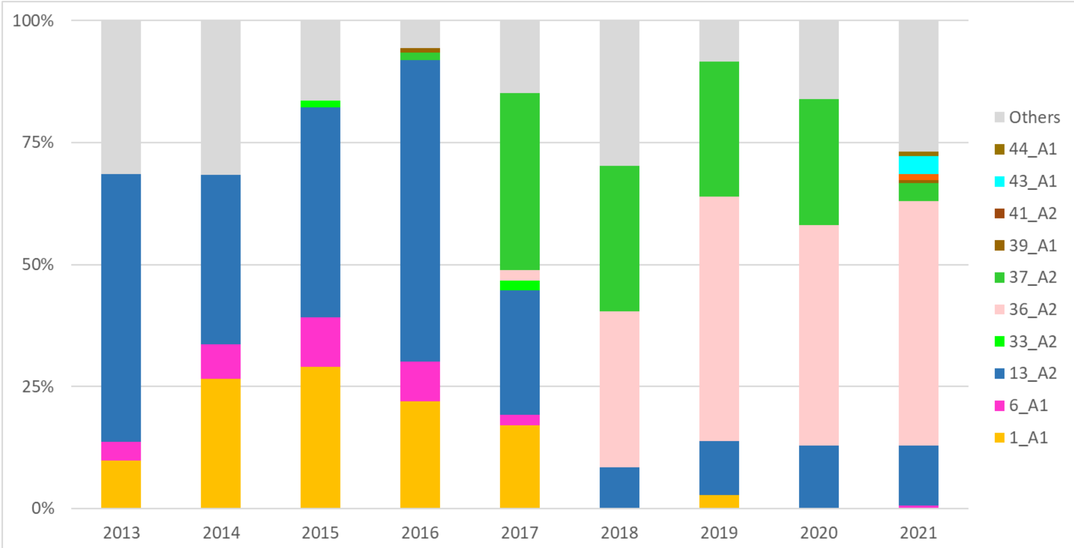
Materials and Methods

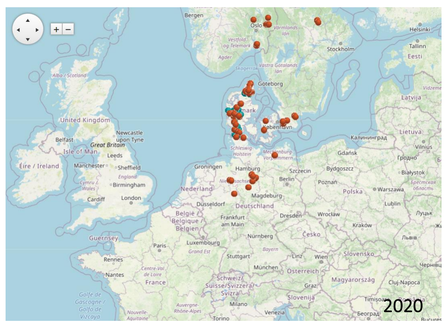
Phytophthora infestans isolates : Single-lesion isolates were obtained by placing pieces of infected tissue on tuber slices of a susceptible potato cultivar (Bintje). Pure cultures were obtained by transferring small pieces of mycelium growing on the upper side of the potato slice on pea agar medium.

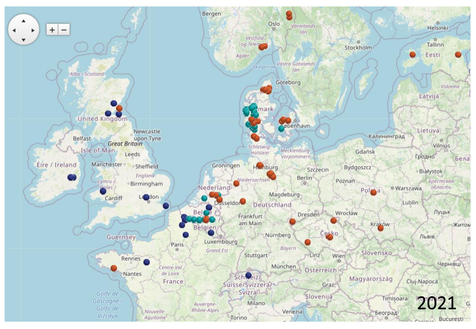
Genotyping: The isolates were analyzed using standardized 12 plex Euroblight SSR genotyping. Genotypes were determined by comparing fragment sizes with isolates previously genotyped (Euroblight monitoring).

Virulence: Virulence was determined using Black's differential set of potato clones, each having one of the R1-R11 resistance genes. Virulence was also determined by detached leaf assay on several varieties which have a good rating about resistance in field. Each leaflet was inoculated with 10µl droplet of sporangial suspension (50,000 sporangia/ml) and incubated in humid chambers under controlled conditions (18°C). After seven days of incubation, the sporulation was evaluated (high sporulation, low sporulation and no sporulation).

Fungicide resistance: Metalaxyl and Mandipropamid resistance was assessed using a floating leaf disk method (leaf of potato cv. Bintje). Leaf disk were floated abaxial side up in Petri dishes each containing water or fungicide at concentrations of 0 to 100 µg/ml. Each disk was inoculated with 10µl droplet of sporangial suspension (50,000 sporangia/ml). After seven days of incubation, isolates sporulating on the disks floating on water containing 100 µg/ml fungicide were rated resistant.
Fluazinam resistance was evaluated on potato leaf discs (cv. Bintje) by mixing fungicide at different concentrations (0,1 - 1 - 10 - 30 and 100 µg/ml) with the sporangial suspensions. After seven days of incubation, the sporulation was evaluated and isolates sporulating with 100 µg/ml fluazinam were rated resistant and isolates sporulating with 30 µg/ml were rated intermediate.







Results

Genotyping: Since 2018, EU_36_A2 has dominated in Belgium: in 2021, the clone made up 50% of the population. EU_13_A2 continues to decline in Europe but still represents 12% of the population in 2021. EU_37_A2 represents only 4% of the population in 2021 while it still represented 25% in 2019 and 2020.
In 2021, monitoring highlighted the emergence of three new clonal lineage, named EU_41_A2, EU_43_A1 and EU_44_A1. The northeast winds during the 2021 season could explain the appearance of these new clones.
The genetically diverse "Other" samples comprised 27% of the sampled population in 2021.

Virulence: There is a great diversity of virulence profiles. All known virulence genes were found in Wallonia isolates. EU_ 13_A2 had a more complex virulence profile than others genotypes. The new clones EU_41_A2 and EU_43_A1 were also more virulent than older (overcome 10 to 11 *R* genes).
Significant differences were observed between genotypes regarding their virulence on resistant varieties. EU_13_A2 was still the more virulent genotype. Varietal resistance were often overcame with EU-36_A2, EU_41_A2 and EU_43_A1.

Fungicide resistance: EU_13_A2 and EU_41_A2 were resistant to Metalaxyl whereas others genotypes were sensitive. EU_37_A2 were resistant to Fluazinam. All genotypes were sensitive to Mandipropamid.