Data article

Time-course investigation of *Phytophthora infestans* infection of potato leaf from three cultivars by quantitative proteomics

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**Abstract**

Potato late blight is one the most important crop diseases worldwide. Even though potato has been studied for many years, the potato disease late blight still has a vast negative effect on the potato production [1–3]. Late blight is caused by the pathogen *Phytophthora infestans* (*P. infestans*), which initiates infection through leaves. However, the biological activities during different stages of infection are poorly described, and could enable novel or improved ways of defeating late blight infection [4]. Therefore, we investigated the interactions between *P. infestans* (mixed strain culture) and potato (*Solanum tuberosum*). Three commercially available field potato cultivars of different resistance to late blight infection; Kuras (moderate), Sarpo Mira (highly resistant) and Bintje (very susceptible) were grown under controlled greenhouse conditions and inoculated with a diversity of *P. infestans* populations.

We used label-free quantitative proteomics to investigate the infection with *P. infestans* in a time-course study over 258 h. Several key issues limits proteome analysis of potato leaf tissue [5–7]. Firstly, the immense complexity of the plant proteome, which is further complicated by the presence of highly abundant proteins, such as ribulose bisphosphate carboxylase/oxygenase (RuBisCO). Secondly, plant leaf and potato, in particular, contain abundant levels amounts of phenols and polyphenols, which hinder or completely prevent a successful protein extraction. Hitherto, protein profiling of potato leaf...
tissues have been limited to few proteome studies and only 1484 proteins have been extracted and comprehensively described \[5,8,9\]. We here present the detailed methods and raw data by optimized gel-enhanced label free quantitative approach. The methodology enabled us to detect and quantify between 3248 and 3529 unique proteins from each cultivar, and up to 758 \textit{P. infestans} derived proteins. The complete dataset is available via ProteomeXchange, with the identifier PXD002767.

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### Specifications Table

| Subject area | Biology |
|--------------|---------|
| More specific subject area | Plant proteomics |
| Type of data | Raw files and text/excel files |
| How data was acquired | Mass Spectrometry Liquid Chromatography High-resolution/high-accuracy mass spectrometer system was used: Q Exactive (Thermo Scientific) |
| Data format | Raw and analyzed data. |
| Experimental factors | Proteome analysis of three potato cultivars following time-course infection with late blight (\textit{Phytophthora infestans}) |
| Experimental features | Three potato cultivars were infected with \textit{Phytophthora infestans} in a time-course over 258 h. Leaf proteins were extracted and analyzed by GelMS using 298 runs of 2.5 h UPLC-OrbitrapMS prior to label-free proteome analysis using MaxQuant. Aalborg, Denmark |
| Data source location | Aalborg, Denmark |
| Data accessibility | The MS proteomics data have been deposited to the ProteomeXchange Consortium via the PRIDE partner repository with the dataset identifier PXD002767. Direct download link: http://www.ebi.ac.uk/pride/archive/projects/PXD002767 |

### Value of the data

- Highly optimized protein extraction method for potato leaf significantly reducing effects of polyphenols and Rubisco.
- Most extensive proteomic analysis of potato leaf so far ( > 4000 unique proteins).
- Time-course infection of three cultivars of wide resistance towards late blight enabling study of biological response to infection.

### 1. Experimental design, materials and methods

#### 1.1. Collection of potato leaf samples

Potato plants of the three cultivars Kuras, Bintje, and Sarpo Mira were grown over the summer of 2008 in a big greenhouse at Landbrugets Kartoffelfond (LKF) Vandel, Denmark. The potato plants were divided into five groups of \textit{Phytophthora infestans} inoculated plants and four groups of control...
plants. Each group contained three plants per cultivar. The inoculation was obtained by bulk spraying a poly-strain inoculum (sporangial suspension) derived from late blight strains collected in Denmark in 2007 on the plants. In order to ensure infection, a high humidity was maintained by covering the plants with plastic overnight. The same procedure was executed for the control plants, which were however sprayed with water instead of *P. infestans* inoculum. For each cultivar, five different sampling time points were selected: 4, 16, 65, 120, and 258 h after spraying. One control and one inoculated group were sampled per time point by taking three replicates per group. One control group was used for two samplings. For each replicate five to six leaves with varying size were sampled and immediately snap-frozen in liquid nitrogen and later stored at −80 °C at Aalborg University, Denmark.

### 1.2. Extraction of from potato leaf

All solutions and samples were kept cold under the entire protein extraction procedure either by using ice or liquid nitrogen. Three to five leaves from one time point were added to a teflon shaking flask (Sartorius, Gottingen, DE) containing a grinding ball (B. Braun, Mannheim, DE). The container was afterwards strapped on to a micro-dismembrator (B. Braun, Mannheim, DE) and the leaf material was homogenized by shaking at 2000 rounds per minute (rpm) for 1 min. 200 mg leaf powder was weighted into a 1.5 mL centrifuge tube (Beckman, Palo Alto, US) containing 500 µL pre-cooled extraction buffer (50 mM HEPES-KOH (pH 7.5), 10 mM potassium acetate, 5 mM magnesium acetate, 1 mM EDTA, 0.5 M tris(2-carboxyethyl)phosphine (TCEP), plant protease inhibitor cocktail (Sigma-Aldrich, Steinheim, DE) containing AEBSF, 1,10-Phenanthroline, Pepstatin A, Leupeptin, Bestatin, and E-64 and lastly a phosphatase inhibitor cocktail (Roche PhosStop). In order to be completely mixed, the leaf sample solution was vortex and then grinded with a pestle. Afterwards, an extra 500 µL extraction buffer was added. The sample was centrifuged in a Sorvall Discovery 90 ultra-centrifuge (Kendro Laboratory Products) at 18.000 rpm (34.000g) at 4 °C for 10 min. The yellow/light green supernatant (soluble proteins) was transferred to a new tube and used further on, while the dark green pellet (insoluble proteins) was stored in −80 °C.

### 1.3. Sample preparation and fractionation

The protein concentration of the leaf supernatants was determined by a Bradford assay using ovalbumin (MP biomedicals, FR) as standard. All the samples and standards were assayed in triplicate and diluted using 50 mM phosphate buffer. Ten µL supernatant, diluted supernatant or standard was used for each quantification and mixed with 250 µL Bradford reagent (0.05% (w/v) Brilliant Blue G-250, 25% (v/v) ethanol (Kemtyl, Køge, DK), and 50% (v/v) phosphoric acid (Merck Millipore, Darmstadt, DE). After 5 min, the resulting color change of the samples was measured at 600 nm using an Infinite® M1000 PRO Tecan (Tecan, Salzburg, AT).

SDS-PAGE was used to separated and fractionate proteins found in the different leaf samples. Supernatant (24 µL) from the inoculated samples or a mixture of the supernatant from the three replicates (8 µL of each) of the corresponding control was mixed with 6 µL 5x reducing SDS sample buffer (0.25 M Tris–HCl (pH 6.8), 5% (w/v) SDS, 50% (v/v) glycerol (Saveen Werner AB), 0.25 M DTT and a couple of bromphenol blue corns) and boiled for 10 min at 100 °C. After boiling, the samples were centrifuged at 14.000 g for 2 min and used for gel electrophoresis with 12% Mini-Protean TGX gels (Biorad, US). The gel electrophoresis was performed at 70 V for 10 min and afterwards at 200 V for 15–20 min (until the sample line had reached ~2/3 of the gel). The gels were stained with Coomassie Brilliant Blue (0.2% solution in 50% ethanol, 7% acetic acid) and destained in 8% ethanol, 5% acetic acid.

In-gel digestion was performed as previously described with few modifications [9]. Each sample lane in the SDS-gels were parted into four big gel slices (top 1–4 bottom), giving 80 gel samples for each potato cultivar (5 time points*4 samples (three inoculated and one control)*4 gel pieces). Each of these four gel samples for one supernatant samples were further chopped into small cubes with a razor blade and transferred to 0.5 mL Protein LoBind tubes (Eppendorf, Hamburg, DE). The gel pieces were washed by incubation for 5 min in 100 µL 0.1 M NH₄HCO₃ and for 15 min in 0.1 M NH₄HCO₃/acetonitrile (1:1), afterwards both washing steps were repeated. The gel pieces were covered by an appropriate amount of
acetonitrile (Panreac, Barcelona, ES) in order to dehydrate the pieces. After the gel pieces had shrunk, the acetonitrile was removed and if residual staining was still observed the washing cycle was performed once more. The samples were reswelled in 125 μL 10 mM DTT in 0.1 M NH₄HCO₃ and incubated at 56 °C for 45 min. The samples were cooled to room temperature and the liquid was replaced with 100 μL 55 mM iodacetic acid in 0.1 M NH₄HCO₃, incubated in the dark for 30 min, and a washing procedure was carried out as before. After dehydration, the gel pieces were reswelled in 120 μL digestion buffer (50 mM NH₄HCO₃, pH 8 and 12.5 ng/μL of sequencing-grade modified porcine trypsin (Promega, Madison, US)). The samples were incubated on ice for 45 min and the supernatant was removed and replaced with 120 μL 50 mM NH₄HCO₃, pH 8, and incubated overnight at 37 °C. The following day the peptides were extracted from the gel pieces. The overnight supernatant was transferred to a new 0.5 mL Protein LoBind tube and saved. One hundred μL 5% formic acid was added to the gel pieces and after 15 min 100 μL acetonitrile was added and the samples were incubated an additionally 15 min. The supernatant was pooled with the overnight supernatant and the procedure was repeated. The pooled samples were dried in a Centrivap Concentrator until no solution was left. The samples were then re-dissolved in 20 μL 0.1% TFA with either 10 fmol/μL MassPREP Enolase digestion standard (Waters, SwissProt P00924) or 10 fmol/μL MassPREP Bovine Serum Albumin (BSA) digestion standard (Waters, SwissProt P02769). The enolase digestion standard was used for all Sarpo Mira samples, while the BSA digestion standard was used for both Bintje and Kuras samples.

1.4. LC–MS/MS analysis

UPLC was performed on a Dionex Thermo Scientific UPLC designed to operate at ultra-high pressures up to 1000 bars with a commercial pre-column set up also from Dionex Thermo Scientific. Peptides from an in gel digestion sample (5 μL (1/4 of the sample) ~ 12.5 μg) were first loaded on to a 2 cm long pre-column with 100 μm inner diameter packed with 5 μm C18 particles and further analyzed by a 50 cm separation-column with 75 μm inner diameter packed with 2 μm C18 particles. The columns were operated under a constant temperature of 40 °C. The reversed phase chromatography was executed with a binary buffer system consisting of buffer A (0.1% (v/v) formic acid and 0.005% (v/v) heptafluorobutyric acid in ultrapure water) and buffer B (90% (v/v) acetonitrile, 10% (v/v) Ultrapure water, 0.1% (v/v) formic acid and 0.005% (v/v) heptafluorobutyric acid). The peptide mixture was loaded onto the pre-column with 2% buffer B at a flow rate of 4 μL/min. After loading, the Sarpo mira peptide samples were separated and eluted by first a gradient of 4–15% buffer B over 6 min and afterwards a long gradient of 15–55% buffer B over 114 min. The peptide samples of both Bintje and Kuras were separated using an optimized LC method with first a gradient of 4–20% buffer B over 3 min and then a 114 min gradient of 20–45% buffer B. In both methods, the eluted peptides entered the Q Exactive with a flow rate of 0.3 μL/min. The LC was coupled to a Q Exactive mass spectrometer (Thermo Scientific) via an electrospray source (Proxeon, Thermo Scientific).

The MS data was obtained using a data-dependent method with full scans (MS) obtained from range 325–2000 m/z at a resolution of 70,000 at m/z 200 (transient time at 120 ms for the Sarpo mira samples and at 250 ms for Bintje and Kuras samples). Up to the top 12 most abundant precursor ions from each survey scan were in the Sarpo mira samples selected with an isolation window of 1.2 m/z and fragmented with higher energy collisional dissociation (HCD) fragmentation with normalized collision energies at 25. For both Bintje and Kuras samples, the precursor ions were selected with an isolation window of 2 m/z and fragmented with normalized collision energies at 30. The MS/MS scans were furthermore acquired with a resolution of 17,500 at m/z 200 (transient time at 60 ms for the Sarpo mira samples and at 80 ms for Bintje and Kuras samples). The automatic gain control (AGC) used for analyzing Sarpo mira samples in both full- and MS/MS scans was 1e⁶, while AGC for analyzing Bintje and Kuras samples was 3e⁶ for full scans and 2e⁵ for MS/MS scans. Repeated sequencing of peptides was kept at a minimum by dynamic exclusion of the sequenced peptides for 30 s (Sarpo mira samples) or 40 s (Bintje and Kuras samples). Furthermore, the fragmentation event was for the Bintje and Kuras samples triggered at highest intensity of the MS peak by using an Apex trigger from 1 to 20 s.
Table 1
Description of file-names in the ProteomeXchange repository PXD002767. MS files were searched using using MaxQuant [10]. Combining fractions of GelMS (1top-4bottom) gel lanes. The number of identified proteins after filtration of typical contaminants and Phytophthora infestans is reported.

| File              | Sample                                      | Cultivar | Digestion | MS system | ID proteins |
|-------------------|---------------------------------------------|----------|-----------|-----------|-------------|
| Bin_K4_1.raw      | Control, 4 h                                | Bintje   | GelMS (4 lanes) | Q Exactive | 2894        |
| Bin_K4_1b.raw     | (Pooled control leafs (n=3) at 4 h)         |          |           |           |             |
| Bin_K4_2.raw      |                                             |          |           |           |             |
| Bin_K4_2b.raw     |                                             |          |           |           |             |
| Bin_K4_3.raw      |                                             |          |           |           |             |
| Bin_K4_3b.raw     |                                             |          |           |           |             |
| Bin_K4_4.raw      |                                             |          |           |           |             |
| Bin_K4_4b.raw     |                                             |          |           |           |             |
| Bin_K16_1.raw     | Control, 16 h                               | Bintje   | GelMS (4 lanes) | Q Exactive | 2831        |
| Bin_K16_1b.raw    |                                             |          |           |           |             |
| Bin_K16_2.raw     |                                             |          |           |           |             |
| Bin_K16_2b.raw    |                                             |          |           |           |             |
| Bin_K16_3.raw     |                                             |          |           |           |             |
| Bin_K16_3b.raw    |                                             |          |           |           |             |
| Bin_K16_4.raw     |                                             |          |           |           |             |
| Bin_K16_4b.raw    |                                             |          |           |           |             |
| Bin_K65_1.raw     | Control, 65 h                               | Bintje   | GelMS (4 lanes) | Q Exactive | 2841        |
| Bin_K65_1b.raw    |                                             |          |           |           |             |
| Bin_K65_2.raw     |                                             |          |           |           |             |
| Bin_K65_2b.raw    |                                             |          |           |           |             |
| Bin_K65_3.raw     |                                             |          |           |           |             |
| Bin_K65_3b.raw    |                                             |          |           |           |             |
| Bin_K65_4.raw     |                                             |          |           |           |             |
| Bin_K65_4b.raw    |                                             |          |           |           |             |
| Bin_K120_1.raw    | Control, 120 h                              | Bintje   | GelMS (4 lanes) | Q Exactive | 2889        |
| Bin_K120_1b.raw   |                                             |          |           |           |             |
| Bin_K120_2.raw    |                                             |          |           |           |             |
| Bin_K120_2b.raw   |                                             |          |           |           |             |
| Bin_K120_3.raw    |                                             |          |           |           |             |
| Bin_K120_3b.raw   |                                             |          |           |           |             |
| Bin_K120_4.raw    |                                             |          |           |           |             |
| Bin_K120_4b.raw   |                                             |          |           |           |             |
| Bin_K258_1.raw    | Control, 258 h                              | Bintje   | GelMS (4 lanes) | Q Exactive | 2784        |
| Bin_K258_1b.raw   |                                             |          |           |           |             |
| Bin_K258_2.raw    |                                             |          |           |           |             |
| Bin_K258_2b.raw   |                                             |          |           |           |             |
| Bin_K258_3.raw    |                                             |          |           |           |             |
| Bin_K258_3b.raw   |                                             |          |           |           |             |
| Bin_K258_4.raw    |                                             |          |           |           |             |
| Bin_K258_4b.raw   |                                             |          |           |           |             |
| Bin_S4_2207_1.raw | P. infestans infect, 4 h (sampling leaf 2207,2208, 2209) | Bintje   | GelMS (4 lanes) | Q Exactive | 3057        |
| Bin_S4_2207_2.raw |                                             |          |           |           |             |
| Bin_S4_2207_3.raw |                                             |          |           |           |             |
| Bin_S4_2207_4.raw |                                             |          |           |           |             |
| Bin_S4_2208_1.raw |                                             |          |           |           |             |
| Bin_S4_2208_2.raw |                                             |          |           |           |             |
| Bin_S4_2208_3.raw |                                             |          |           |           |             |
| Bin_S4_2208_4.raw |                                             |          |           |           |             |
| Bin_S4_2209_1.raw |                                             |          |           |           |             |
| Bin_S4_2209_2.raw |                                             |          |           |           |             |
| Bin_S4_2209_3.raw |                                             |          |           |           |             |
| Bin_S4_2209_4.raw |                                             |          |           |           |             |
| Bin_S16_2258_1.raw| P. infestans infect, 16 h                    | Bintje   | GelMS (4 lanes) | Q Exactive | 3065        |
| Bin_S16_2258_2.raw|                                             |          |           |           |             |
| Bin_S16_2258_3.raw|                                             |          |           |           |             |
| Bin_S16_2258_4.raw|                                             |          |           |           |             |
| Bin_S16_2259_1.raw|                                             |          |           |           |             |
| Bin_S16_2259_2.raw|                                             |          |           |           |             |
| Sample | Description | Genotype | MS Type | Q Exactive | Result (h) |
|--------|-------------|----------|---------|------------|------------|
| Bin_S16_2259_3.raw | *P. Infestans* infect, 65 h | Bintje | GelMS (4 lanes) | Q Exactive | 3099 |
| Bin_S16_2259_4.raw | | | | | |
| Bin_S16_2260_1.raw | | | | | |
| Bin_S16_2260_2.raw | | | | | |
| Bin_S16_2260_3.raw | | | | | |
| Bin_S16_2260_4.raw | | | | | |
| Bin_S65_2309_1.raw | P. Infestans infect, 120 h | Bintje | GelMS (4 lanes) | Q Exactive | 3149 |
| Bin_S65_2309_2.raw | | | | | |
| Bin_S65_2309_3.raw | | | | | |
| Bin_S65_2309_4.raw | | | | | |
| Bin_S65_2310_1.raw | | | | | |
| Bin_S65_2310_2.raw | | | | | |
| Bin_S65_2310_3.raw | | | | | |
| Bin_S65_2310_4.raw | | | | | |
| Bin_S65_2311_1.raw | | | | | |
| Bin_S65_2311_2.raw | | | | | |
| Bin_S65_2311_3.raw | | | | | |
| Bin_S65_2311_4.raw | | | | | |
| Bin_S120_2360_1.raw | P. Infestans infect, 258 h | Bintje | GelMS (4 lanes) | Q Exactive | 3097 |
| Bin_S120_2360_2.raw | | | | | |
| Bin_S120_2360_3.raw | | | | | |
| Bin_S120_2360_4.raw | | | | | |
| Bin_S120_2361_1.raw | | | | | |
| Bin_S120_2361_2.raw | | | | | |
| Bin_S120_2361_3.raw | | | | | |
| Bin_S120_2361_4.raw | | | | | |
| Kur_K4_1.raw | Control, 4 h | Kuras | GelMS (4 lanes) | Q Exactive | 3006 |
| Kur_K4_1b.raw | | | | | |
| Kur_K4_2.raw | | | | | |
| Kur_K4_2b.raw | | | | | |
| Kur_K4_3.raw | | | | | |
| Kur_K4_3b.raw | | | | | |
| Kur_K4_4.raw | | | | | |
| Kur_K4_4b.raw | | | | | |
| Kur_K16_1.raw | Control, 16 h | Kuras | GelMS (4 lanes) | Q Exactive | 3004 |
| Kur_K16_1b.raw | | | | | |
| Kur_K16_2.raw | | | | | |
| Kur_K16_2b.raw | | | | | |
| Kur_K16_3.raw | | | | | |
| Kur_K16_3b.raw | | | | | |
| Kur_K16_4.raw | | | | | |
| Kur_K16_4b.raw | | | | | |
| Kur_K65_1.raw | Control, 65 h | Kuras | GelMS (4 lanes) | Q Exactive | 3177 |
| Kur_K65_1b.raw | | | | | |
| Kur_K65_2.raw | | | | | |
| Kur_K65_2b.raw | | | | | |
| Kur_K65_3.raw | | | | | |
| Kur_K65_3b.raw | | | | | |
| Kur_K65_4.raw | | | | | |
| Kur_K65_4b.raw | | | | | |
| Sample Code | Description | Treatment | Instrument | Duration |
|-------------|-------------|-----------|------------|----------|
| Kur_K120_1.raw | Control | Kuras | GelMS (4 lanes) | Q Exactive | 3144 |
| Kur_K120_1b.raw | 120 h | Kuras | GelMS (4 lanes) | Q Exactive | 3122 |
| Kur_K120_2.raw | | Kuras | GelMS (4 lanes) | Q Exactive | 3346 |
| Kur_S4_2231_1.raw | P. Infestans infect, 4 h | Kuras | GelMS (4 lanes) | Q Exactive | 3261 |
| Kur_S65_2333_1.raw | P. Infestans infect, 65 h | Kuras | GelMS (4 lanes) | Q Exactive | 3370 |
| Kur_K258_1.raw | Control, 258 h | Kuras | GelMS (4 lanes) | Q Exactive | 3406 |
| Kur_S120_2384_1.raw | P. Infestans infect, 120 h | Kuras | GelMS (4 lanes) | Q Exactive | 3406 |

*Table 1 (continued)*
| Sample Code          | Description                      | Time (h) | GelMS Type           | Q Exactive | Score |
|---------------------|----------------------------------|----------|----------------------|------------|-------|
| Kur_S120_2386_4.raw | P. Infestans infect, Kuras       | 258      | GelMS (4 lanes)      | Q Exactive | 3414  |
| Kur_S258_2435_1.raw | Kuras GelMS                      |          |                      |            |       |
| Kur_S258_2435_2.raw |                                  |          |                      |            |       |
| Kur_S258_2435_3.raw |                                  |          |                      |            |       |
| Kur_S258_2435_4.raw |                                  |          |                      |            |       |
| Kur_S258_2436_1.raw |                                  |          |                      |            |       |
| Kur_S258_2436_2.raw |                                  |          |                      |            |       |
| Kur_S258_2436_3.raw |                                  |          |                      |            |       |
| Kur_S258_2436_4.raw |                                  |          |                      |            |       |
| Kur_S258_2437_1.raw |                                  |          |                      |            |       |
| Kur_S258_2437_2.raw |                                  |          |                      |            |       |
| Kur_S258_2437_3.raw |                                  |          |                      |            |       |
| Kur_S258_2437_4.raw |                                  |          |                      |            |       |
| Sar_K4_1.raw        | Control, SarpoMira               | 4        | GelMS (4 lanes)      | Q Exactive | 3096  |
| Sar_K4_1b.raw       |                                  |          |                      |            |       |
| Sar_K4_2.raw        |                                  |          |                      |            |       |
| Sar_K4_2b.raw       |                                  |          |                      |            |       |
| Sar_K4_3.raw        |                                  |          |                      |            |       |
| Sar_K4_3b.raw       |                                  |          |                      |            |       |
| Sar_K4_4.raw        |                                  |          |                      |            |       |
| Sar_K4_4b.raw       |                                  |          |                      |            |       |
| Sar_K16_1.raw       | Control, SarpoMira               | 16       | GelMS (4 lanes)      | Q Exactive | 2880  |
| Sar_K16_1b.raw      |                                  |          |                      |            |       |
| Sar_K16_2.raw       |                                  |          |                      |            |       |
| Sar_K16_2b.raw      |                                  |          |                      |            |       |
| Sar_K16_3.raw       |                                  |          |                      |            |       |
| Sar_K16_3b.raw      |                                  |          |                      |            |       |
| Sar_K16_4.raw       |                                  |          |                      |            |       |
| Sar_K16_4b.raw      |                                  |          |                      |            |       |
| Sar_K65_1.raw       | Control, SarpoMira               | 65       | GelMS (4 lanes)      | Q Exactive | 3099  |
| Sar_K65_1b.raw      |                                  |          |                      |            |       |
| Sar_K65_2.raw       |                                  |          |                      |            |       |
| Sar_K65_2b.raw      |                                  |          |                      |            |       |
| Sar_K65_3.raw       |                                  |          |                      |            |       |
| Sar_K65_3b.raw      |                                  |          |                      |            |       |
| Sar_K65_4.raw       |                                  |          |                      |            |       |
| Sar_K65_4b.raw      |                                  |          |                      |            |       |
| Sar_K120_1.raw      | Control, SarpoMira               | 120      | GelMS (4 lanes)      | Q Exactive | 3092  |
| Sar_K120_1b.raw     |                                  |          |                      |            |       |
| Sar_K120_2.raw      |                                  |          |                      |            |       |
| Sar_K120_2b.raw     |                                  |          |                      |            |       |
| Sar_K120_3.raw      |                                  |          |                      |            |       |
| Sar_K120_3b.raw     |                                  |          |                      |            |       |
| Sar_K120_4.raw      |                                  |          |                      |            |       |
| Sar_K120_4b.raw     |                                  |          |                      |            |       |
| Sar_K258_1.raw      | Control, SarpoMira               | 258      | GelMS (4 lanes)      | Q Exactive | 3124  |
| Sar_K258_1b.raw     |                                  |          |                      |            |       |
| Sar_K258_2.raw      |                                  |          |                      |            |       |
| Sar_K258_2b.raw     |                                  |          |                      |            |       |
| Sar_K258_3.raw      |                                  |          |                      |            |       |
| Sar_K258_3b.raw     |                                  |          |                      |            |       |
| Sar_K258_4.raw      |                                  |          |                      |            |       |
| Sar_K258_4b.raw     |                                  |          |                      |            |       |
| Sar_S4_2240_1.raw   | P. Infestans infect, SarpoMira   | 4        | GelMS (4 lanes)      | Q Exactive | 3363  |
| Sar_S4_2240_2.raw   |                                  |          |                      |            |       |
| Sar_S4_2240_3.raw   |                                  |          |                      |            |       |
| Sar_S4_2240_4.raw   |                                  |          |                      |            |       |
| Sar_S4_2241_1.raw   |                                  |          |                      |            |       |
| Sar_S4_2241_2.raw   |                                  |          |                      |            |       |
| Sar_S4_2241_3.raw   |                                  |          |                      |            |       |
| Sar_S4_2241_4.raw   |                                  |          |                      |            |       |
| Sar_S4_2242_2.raw   |                                  |          |                      |            |       |
| Sar_S4_2242_3.raw   |                                  |          |                      |            |       |
### 1.5. Proteomic data analysis

The raw data-files from potato leaf analyzed on the Q Exactive were searched using MaxQuant 1.5.3.8 (separate cultivars) and 1.5.3.28 (all cultivars) against two databases containing UniProtKB Potato reference proteome AUP000011115 (53104 sequences, july 2015) and UniProtKB *P. infestans* T30-4 (17609 sequences, july 2015). All standard settings were employed with carbamidomethyl(C) as a static modification and deamidation (NQR), oxidation (M), and protein N-terminal acetylation were included as variable modifications. Label-free quantitation of all proteins was performed in MaxQuant based on integrated precursor intensities using default settings, experiments and fractions settings to

| Sample   | Treatment | Instrument | Type | Retention Time |
|----------|-----------|------------|------|----------------|
| Sar_S4_2242_4.raw | P. *Infestans* infect | SarpoMira GelMS (4 lanes) | Q Exactive | 16 h |
| Sar_S4_2242_1.raw |  |  |  |  |
| Sar_S16_2291_1.raw | P. *Infestans* infect | SarpoMira GelMS (4 lanes) | Q Exactive | 3361 |
| Sar_S16_2291_2.raw |  |  |  |  |
| Sar_S16_2291_3.raw |  |  |  |  |
| Sar_S16_2291_4.raw |  |  |  |  |
| Sar_S16_2292_1.raw |  |  |  |  |
| Sar_S16_2292_2.raw |  |  |  |  |
| Sar_S16_2292_3.raw |  |  |  |  |
| Sar_S16_2292_4.raw |  |  |  |  |
| Sar_S16_2293_1.raw |  |  |  |  |
| Sar_S16_2293_2.raw |  |  |  |  |
| Sar_S16_2293_3.raw |  |  |  |  |
| Sar_S16_2293_4.raw |  |  |  |  |
| Sar_S65_2342_1.raw | P. *Infestans* infect, | SarpoMira GelMS (4 lanes) | Q Exactive | 3326 |
| Sar_S65_2342_2.raw | 65 h |  |  |  |
| Sar_S65_2342_3.raw |  |  |  |  |
| Sar_S65_2342_4.raw |  |  |  |  |
| Sar_S65_2343_1.raw |  |  |  |  |
| Sar_S65_2343_2.raw |  |  |  |  |
| Sar_S65_2343_3.raw |  |  |  |  |
| Sar_S65_2343_4.raw |  |  |  |  |
| Sar_S65_2344_1.raw |  |  |  |  |
| Sar_S65_2344_2.raw |  |  |  |  |
| Sar_S65_2344_3.raw |  |  |  |  |
| Sar_S65_2344_4.raw |  |  |  |  |
| Sar_S120_2393_1.raw | P. *Infestans* infect, | SarpoMira GelMS (4 lanes) | Q Exactive | 3389 |
| Sar_S120_2393_2.raw | 120 h |  |  |  |
| Sar_S120_2393_3.raw |  |  |  |  |
| Sar_S120_2393_4.raw |  |  |  |  |
| Sar_S120_2394_1.raw |  |  |  |  |
| Sar_S120_2394_2.raw |  |  |  |  |
| Sar_S120_2394_3.raw |  |  |  |  |
| Sar_S120_2394_4.raw |  |  |  |  |
| Sar_S120_2395_1.raw |  |  |  |  |
| Sar_S120_2395_2.raw |  |  |  |  |
| Sar_S120_2395_3.raw |  |  |  |  |
| Sar_S120_2395_4.raw |  |  |  |  |
| Sar_S258_2444_1.raw | P. *Infestans* infect, | SarpoMira GelMS (4 lanes) | Q Exactive | 3348 |
| Sar_S258_2444_2.raw | 258 h |  |  |  |
| Sar_S258_2444_3.raw |  |  |  |  |
| Sar_S258_2444_4.raw |  |  |  |  |
| Sar_S258_2445_1.raw |  |  |  |  |
| Sar_S258_2445_2.raw |  |  |  |  |
| Sar_S258_2445_3.raw |  |  |  |  |
| Sar_S258_2445_4.raw |  |  |  |  |
| Sar_S258_2446_1.raw |  |  |  |  |
| Sar_S258_2446_2.raw |  |  |  |  |
| Sar_S258_2446_3.raw |  |  |  |  |
| Sar_S258_2446_4.raw |  |  |  |  |
combine the GelMS raw data [11]. Time-course control samples and time-course *P. infestans* infected samples were searched as independent replicates linked by option "experiment" and the Gel-MS fractions were linked using the option "fraction". This enables statistical analysis based on the search output files as provided in the ProteomXchange and described in detail in Table 2.

2. Data

The proteomics raw data and MaxQuant result-files from the analysis have been deposited to the ProteomeXchange Consortium via the PRIDE partner repository with the dataset identifier PXD0002767 [11–14], and can be downloaded directly (http://www.ebi.ac.uk/pride/archive/projects/PXD0002767). Table 1 contains the list of submitted proteomics raw-datafiles and information regarding sample types, species, digestion protocol, and MS system. Table 2 contains the list of submitted analysis result files and a short description of the content. In our protein extraction yield data, the GelMS protocol was the most efficient, yielding the highest number of identifiable proteins in potato leaf. Therefore, we conducted the comprehensive analysis of late blight infected potato leaf from three different cultivars (Bintje, Sarpo Mira and Kuras), analyzed in biological triplicates on a Q Exactive MS (Thermo Scientific).

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Appendix A. Supplementary material

Supplementary data associated with this article can be found in the online version at http://dx.doi.org/10.1016/j.dib.2015.11.069.

References

[1] S. Kamoun, C.D. Smart, Late blight of potato and tomato in the genomics era, Plant Dis. 89 (2005) 692–699.
[2] W. Fry, Phytophthora infestans: the plant (and R gene) destroyer, Mol. Plant Pathol. 9 (2008) 385–402.
[3] P.R.J. Birch, et al., Towards understanding the virulence functions of RXLR effectors of the oomycete plant pathogen Phytophthora infestans, J. Exp. Bot. 60 (2009) 1133–1140.
[4] A. Ali, et al., Quantitative proteomics and transcriptomics of potato in response to Phytophthora infestans in compatible and incompatible interactions, BMC Genom. 15 (2014) 497.
[5] J.F. Buyel, R.M. Twyman, R. Fischer, Extraction and downstream processing of plant-derived recombinant proteins, Biotechnol. Adv. 33 (2015) 902–913.
[6] S. Lim, et al., Protein profiling in potato (Solanum tuberosum L.) leaf tissues by differential centrifugation, J. Proteome Res. 11 (2012) 2594–2601.
[7] W. Pierpoint, The extraction of enzymes from plant tissues rich in phenolic compounds, Methods Mol. Biol. 244 (1996) 69–80.
[8] S. Lim, et al., Proteomics analysis suggests broad functional changes in potato leaves triggered by phosphites and a complex indirect mode of action against Phytophthora infestans, J. Proteom. 93 (2013) 207–223.
[9] A. Stensballe, S. Hald, G. Bauw, A. Blennow, K.G. Welinder, The amyloplast proteome of potato tuber, FEBS J. 275 (2008) 1723–1741.
[10] J. Cox, et al., Accurate proteome-wide label-free quantification by delayed normalization and maximal peptide ratio extraction, termed maxLFQ, Mol. Cell. Proteom. 13 (2014) 2513–2526.
[11] J.A. Vizcaíno, et al., ProteomeXchange provides globally coordinated proteomics data submission and dissemination, Nat. Biotechnol. 32 (2014) 223–226.
[12] J.A. Vizcaíno, et al., The PRoteomics IDEntifications (PRIDE) database and associated tools: status in 2013, Nucleic Acids Res. 41 (2013) D1063–D1069.
[13] R. Wang, et al., PRIDE Inspector: a tool to visualize and validate MS proteomics data, Nat. Biotechnol. 30 (2012) 135–137.
[14] R.G. Côté, et al., The PRoteomics IDEntification (PRIDE) Converter 2 framework: an improved suite of tools to facilitate data submission to the PRIDE database and the ProteomeXchange consortium, Mol. Cell. Proteom. 11 (2012) 1682–1689.
[15] J. Cox, et al., MaxLFQ allows accurate proteome-wide label-free quantification by delayed normalization and maximal peptide ratio extraction, Mol. Cell. Proteom. M113 (2014) 031591. http://dx.doi.org/10.1074/mcp.M113.031591.