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## Data Article

# Cell wall proteomic datasets of stems and leaves of *Brachypodium distachyon*



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## ABSTRACT

This article provides experimental data describing the cell wall protein profiles of stems and leaves of *Brachypodium distachyon* at two different stages of development. The cell wall proteomics data have been obtained from (i) stem internodes at young and mature stages of development, and (ii) leaves at young and mature stages of development. The proteins have been extracted from purified cell walls using buffers containing calcium chloride (0.2 M) or lithium chloride (2 M). They have been identified by LC-MS/MS and bioinformatics. These data allow deepening our knowledge of these cell wall proteomes. They are a valuable resource for people interested in plant cell wall biology to understand the roles of cell wall proteins during the growth of vegetative organs.

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**Specifications Table**

Subject	Omics: Proteomics
Specific subject area	Plant cell wall proteomics
Type of data	Figure 1: Short separation by 1D-E of proteins extracted from the cell walls of internodes and leaves of <i>B. distachyon</i>
How data were acquired	The data have been acquired by mass spectrometry, using a LTQ XL ion trap instrument (Thermo Electron, Thermo Fisher Scientific Inc, Courtaboeuf, France). The data have been managed with the X!Tandem Piledriver software (2015.04.01.1, <a href="http://www.thegpm.org/">http://www.thegpm.org/</a> )
Data format	Raw data
Parameters for data collection	Stem internodes and leaves of <i>Brachypodium distachyon</i> have been collected during active growth and at mature stage of development. Cell walls have been purified prior to protein extraction with salt solutions containing either calcium chloride 0.2 M or lithium chloride 2 M.
Description of data collection	The four conditions have been analyzed in triplicate. Proteins have been identified by LC-MS/MS and bioinformatics.
Data accessibility	The data have been deposited in two public repositories. Repository name: <i>WallProtDB</i> (cell wall proteomics data) Data identification: <i>Brachypodium distachyon</i> , leaves, stems Direct URL to data: <a href="http://www.polebio.lrsv.ups-tlse.fr/WallProtDB/">http://www.polebio.lrsv.ups-tlse.fr/WallProtDB/</a> Repository name: PROTIcDb (LC-MS/MS data) Data identification: <i>Brachypodium</i> cell wall proteome Direct URL to data: <a href="http://moulon.inra.fr/protic/brachypodium_cell_wall_proteome">http://moulon.inra.fr/protic/brachypodium_cell_wall_proteome</a>

**Value of the Data**

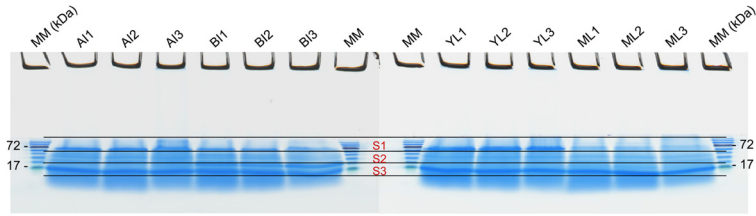
- The datasets provide an enlargement of the known cell wall proteomes of stems and leaves of *B. distachyon* at two different developmental stages.
- People interested in cell wall biology will benefit from these datasets.
- The data can be used to study the impact of the developmental stage on the *B. distachyon* cell wall proteome and in turn on the role of cell wall proteins in development.

**1. Data Description**

The data provide a cell wall proteomic approach to contribute to the understanding of the role of the cell wall proteins during growth in a monocot model plant, *Brachypodium distachyon*. They complement a previous study on the same material, but using a different strategy for protein separation and data analysis [1]. The raw data include information about protein identification in stem internodes and leaves, at two developmental stages, i.e. during active growth and in mature organs. All the LC-MS/MS proteomics data have been deposited at PROTIcDb (<http://moulon.inra.fr/protic/wallomics>). The data regarding the proteins predicted to be secreted have been deposited at *WallProtDB* (<http://www.polebio.lrsv.ups-tlse.fr/WallProtDB/>).

**2. Experimental Design, Materials and Methods****2.1. Plant material**

To favor a homogeneous germination rate, the seeds of *B. distachyon* line Bd21 were maintained in the dark in a wet chamber at 4 °C during 4 days. They were sowed in a Tref H4 compost (Jiffy France SARL, Trévoux, France). They were grown in a growth chamber with the following day/night conditions: 16 h/8 h–25 °C/22 °C and 80% humidity. Pots were covered with Saran® wrap until the first leaves reached a size of about 3 cm. Two month-old plants were used for the experiments. Four samples have been analyzed: apical internodes (AI, first internodes at the top of the culm); basal internodes (BI, first lignified internodes at the basis of the culm); young



**Fig. 1.** Short separation by 1D-E of proteins extracted from the cell walls of apical (AI) and basal (BI) internodes and of young (YL) and mature (ML) leaves of *B. distachyon*. The proteins have been quickly separated by SDS-PAGE in a 12.50%/0.33% of acrylamide/bisacrylamide gel. Then, for each biological replicate (AI 1-3, BI 1-3, ML 1-3 and YL 1-3), the gel has been cut in three slices (S1, S2 and S3) which have been analyzed separately. MM stands for molecular mass markers.

leaves (YL, less than 6 cm in length); and mature leaves (ML, fully expanded). Three biological replicates were analyzed for each type of organ.

## 2.2. Sequential extraction of proteins from purified cell walls

The cell walls were purified as described [2]. The proteins were extracted from purified cell walls as described [3]. The final protein extracts were lyophilized. Proteins were quantified with the CooAssay Protein Assay kit (Interchim, Montluçon, France). Typically, 250 µg proteins/g fresh material were obtained for AI and YL samples whereas 60 and 70 µg proteins/g fresh material were collected for BI and ML samples, respectively.

## 2.3. Identification of proteins by LC-MS/MS

The LC-MS/MS analyses have been done at the PAPPISO proteomics platform ([pappiso.inra.fr/](http://pappiso.inra.fr/)) according to [4]. Short 1D-E were performed and each lane was cut in three gel fragments as shown in Fig. 1. In-gel digestion was performed with the Progest system following a standard trypsin protocol (Genomic Solution, Digilab, Cambridgeshire, UK). HPLC was performed on a NanoLC-Ultra system (Eksigent, Dublin, CA). MS/MS analyses were achieved using a LTQ XL ion trap (Thermo Electron, Thermo Fisher Scientific Inc, Courtaboeuf, France) using a nano electrospray interface as described [4]. One run took 75 min including the regeneration step at 95% B and the equilibration step at 95% A. Two technical replicates have been performed. However, the second technical repeat of one of the ML samples had to be discarded due to technical problems. The parameters used for protein identification are described at PROTIcDb ([http://moulon.inra.fr/protic/brachypodium\\_cell\\_wall\\_proteome](http://moulon.inra.fr/protic/brachypodium_cell_wall_proteome)). Briefly, peptide ions were analyzed using Xcalibur 2.5.0 (Thermo Fisher Scientific Inc) with the following data-dependent acquisition steps: full MS scan (mass-to-charge ratio (m/z) 350 to 1300, centroid mode); and MS/MS ( $q_z = 0.25$ , activation time = 30 ms, collision energy = 35%; centroid mode). The second step was repeated for the three major ions detected in the first one. Dynamic exclusion was set to 30 s. The *B. distachyon* genomic data were downloaded from (i) the Phytozome version V13 ([https://phytozome.jgi.doe.gov/pz/portal.html#!info?alias=Org\\_Bdistachyon](https://phytozome.jgi.doe.gov/pz/portal.html#!info?alias=Org_Bdistachyon)) and (ii) the version 3.2 of the *B. distachyon* genome (<https://genome.jgi.doe.gov/>). Raw data were converted using ReAdW 4.3.1 before the database search. The X!Tandem software (Release 2015.04.01.1) was used for protein identification [5] and the X!Tandem Pipeline (Release 0.4.2) [6] was used for MS data processing. Only proteins identified with at least two different peptides in the same biological replicate and found in at least two biological replicates were validated.

## 2.4. Bioinformatics annotation of proteins

The sub-cellular localization as well as the function of the identified proteins were predicted using the ProtAnnDB tool (<http://www.polebio.lrsrv.ups-tlse.fr/ProtAnnDB/>) [7]. A protein

was considered as a cell wall protein if no more than one trans-membrane domain was found, there was no predicted intracellular retention signal and if it was predicted as secreted by two bioinformatics programs, as described [8]. Whenever possible, a literature survey was performed to look for experimental data to check the predictions. Functional annotation was done by combining available experimental data to bioinformatics predictions and is available at PROTiCdb ([http://moulon.inra.fr/protic/brachypodium\\_cell\\_wall\\_proteome](http://moulon.inra.fr/protic/brachypodium_cell_wall_proteome)) and at WallProtDB for cell wall proteins (<http://www.polebio.lrsv.ups-tlse.fr/WallProtDB/>).

## Ethics Statement

This is no ethical issue for this study.

## CRediT Author Statement

**Thibaut Douché:** Investigation, Data curation; **Benoît Valot and Thierry Balliau:** Mass spectrometry analyses; **Hélène San Clemente:** Data management, Database implementation; **Michel Zivy:** Supervision of mass spectrometry analyses; **Elisabeth Jamet:** Supervision, Original draft preparation. All the authors have contributed to the writing of the manuscript and approved the final version.

## Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships which have or could be perceived to have influenced the work reported in this article.

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