

SUPPLEMENTARY DATA

Table S1. Sequences of the oligonucleotide primers used to get the PAC-V5-6xHis construct allowing the expression of the PAC-V5-6xHis recombinant protein.

Primer	Orientation	Nucleotide sequence (5' → 3')
SP	forward	ATGGGTTTCATTGGTAAGAGTG
SP^a	reverse	TAGACTACGGTTGAA AGTGAAGACAGAGGA
PAC1^a	forward	TCCTCTGTCTTCAC TTCAACCGTAGTCTA
PAC2	reverse	TTTGGGGCAAGACGGGTAAAG
internal^b	forward	GGAGATAGAACCATG GGTTCATTGGTAAG
internal^c	reverse	CAAGAAAGCTGGGTCT CAATGGTGATGGTGATG
external^b	forward	GGGGACAAGTTTGTACAAAAAAGCAGGCTTCGA AGGAGATAGAACCATG
external^c	reverse	GGGGACCACTTTGTAC CAAGAAAGCTGGGTC

- Blue and black sequences are reverse complementary sequences spanning the SP and the PAC encoding sequences of AGP31 respectively, thus allowing the fusion of the two PCR fragments encoding the SP and the PAC domain respectively.
- Sequences common to internal and external forward oligonucleotide primers are in bold.
- Sequences common to internal and external reverse oligonucleotide primers are in bold.

Table S2. Commercial polysaccharides used for in vitro protein/polysaccharide interaction assays. These polysaccharides were purchased either from Sigma-Aldrich (Saint-Quentin Fallavier, France) or from Megazyme (Wicklow, Ireland).

molecule	name	origin	reference	properties	purity	carbohydrate content
pectins						
polygalacturonic acid, sodium salt	citrus PGA	citrus fruit	Sigma-Aldrich P3850	nd	≥75%	nd
polygalacturonic acid	orange PGA	orange	Sigma-Aldrich P3889	nd	≥85%	nd
polygalacturonic acid methyl ester	apple m.e.PGA	apple	Sigma-Aldrich 76282 (Fluka)	MW 30,000-100,000 70-75% esterification	>90%	nd
polygalacturonic acid methyl ester	citrus m.e.PGA	citrus peel	Sigma-Aldrich 76280* (Fluka), replaced by P9135	MW 30,000-100,000 ~60% esterification	>90%	nd
galactan	galactan	potato	Megazyme P-GALPOT	nd	nd	galactose 88%, arabinose 3%, rhamnose 3%, galacturonic acid 6%
rhamnogalacturonan I	RGI	potato pectic fiber	Megazyme P-RHAM1	nd	> 97%	galacturonic acid 62%, rhamnose 20%, arabinose 3.3%, xylose 1%, galactose 12%, other sugars <1.7%
arabinogalactan	AG	larch	Megazyme P-ARGAL	nd	> 95%	galactose 81%, arabinose 14%, other sugars 5%
hemicelluloses						
xyloglucan	XG	tamarind seed	Megazyme P-WYGLN	MW 202,000	> 97%	xylose 38%, glucose 42%, galactose 16%, arabinose 4%
β-glucan	β-glucan	barley	Megazyme P-BGBL	MW 165,000	~95%	nd
xylan	birch wood xylan	birch wood	Sigma-Aldrich X0502*	nd	≥90%	xylose
xylan	beech wood xylan	beechwood	Sigma-Aldrich X4252	nd	>90%	xylose
arabinoxylan	AX	wheat	Megazyme P-WAXYL	nd	~ 90%	arabinose 37%, xylose 61%, other sugars 2%
cellulose						
cellulose	cellulose	cotton	Sigma-Aldrich C6413*	nd		nd

* No longer available.

nd, not determined.

Table S3. Features common to proteins homologous to AGP31, in addition to the presence of a predicted signal peptide

Protein name	His stretch ^a	AGP motifs ^b	β -Yariv ^c	X(P) _{n≥2} X motifs	PAC domain ^d
AtAGP31	+	+	+	+	+
AtAGP30	-	+	+	+	+
DcAGP1	+	++	nd	+	+
GhAGP31	+	+	nd	+	+
TTS-1/TTS-2	+	++	+	+	+
NaTTS	nd	nd	+	nd	nd
PELPs	-	+	nd	+	+
NaPRP4	+ (4 His)	+++	weak	+	+
NaPRP5	+ (3 His)	- (ext)	-	+	+
PvPRP1	+	+	nd	+	+ (4)
CaPRP1	+ (3 His)	+	nd	+	+
PhPRP1	+	+	nd	+	+

In the whole table, nd means not determined, + means presence and – absence.

a. The number of His residues is mentioned when the His stretch is very short.

b. Presence of at least one series of non-contiguous Pro residues in (XP)_n motifs where X can be Ala, Ser or Thr. ++ means that such motifs are present both on the N- and C-sides of the His stretch. +++ means that a third AGP motif is found at the C-terminus of the Pro-rich domain. ext means that canonical extensin motifs (Ser-Pro_n) are present at the N-terminus of the Pro-rich domain.

c. Precipitation or staining with the β -glucosyl Yariv reagent.

d. (4) means that only 4 instead of 6 conserved Cys residues are present.

References are given in the main text.

Fig. S1. Test of the polysaccharide array with specific monoclonal antibodies.

One μL (0.5 μL for cellulose) of each polysaccharide solution was spotted on a nitrocellulose membrane: cell wall polysaccharide enriched-fractions from *A. thaliana* etiolated hypocotyls (pectins, hemicelluloses and cellulose) and commercial polysaccharides (citrus and orange PGA; apple and citrus m.e.PGA; XG; β -glucan; galactan; all at 1 mg/mL and RGI at 10 mg/mL). Specific monoclonal antibodies against PGA (JIM5), m.e.PGA (JIM7), XG (LM15) and galactan (LM5) were used. Secondary antibody coupled to alkaline phosphatase allowed signal detection. Interaction assays were carried out in duplicates.

As expected, JIM5 and JIM7 only gave a signal with the pectin-enriched cell wall fraction. LM15 gave the same signal with the hemicellulose- and cellulose-enriched cell wall fractions. This can be explained by the tight association between XG and cellulose microfibrils [1]. Finally, LM5 gave a weak signal with the pectin-enriched cell wall fraction and a stronger signal with the hemicellulose- than with the cellulose-enriched cell wall fraction. This was probably due to the fact that CDTA mainly extracted PGA. The same results were obtained with *B. distachyon* leaf cell wall fractions (not shown). Besides, JIM5 gave a signal with the commercial PGA and m.e.PGA. JIM7 only gave a signal with m.e.PGA. LM15 only gave a signal with XG and LM5 with galactan and RGI.

All these experiments allowed the characterization of the cell wall polysaccharide fractions and the validation of the polysaccharide array.

