# Expression of a hyperthermophilic endoglucanase in hybrid poplar modifies the plant cell wall and enhances digestibility

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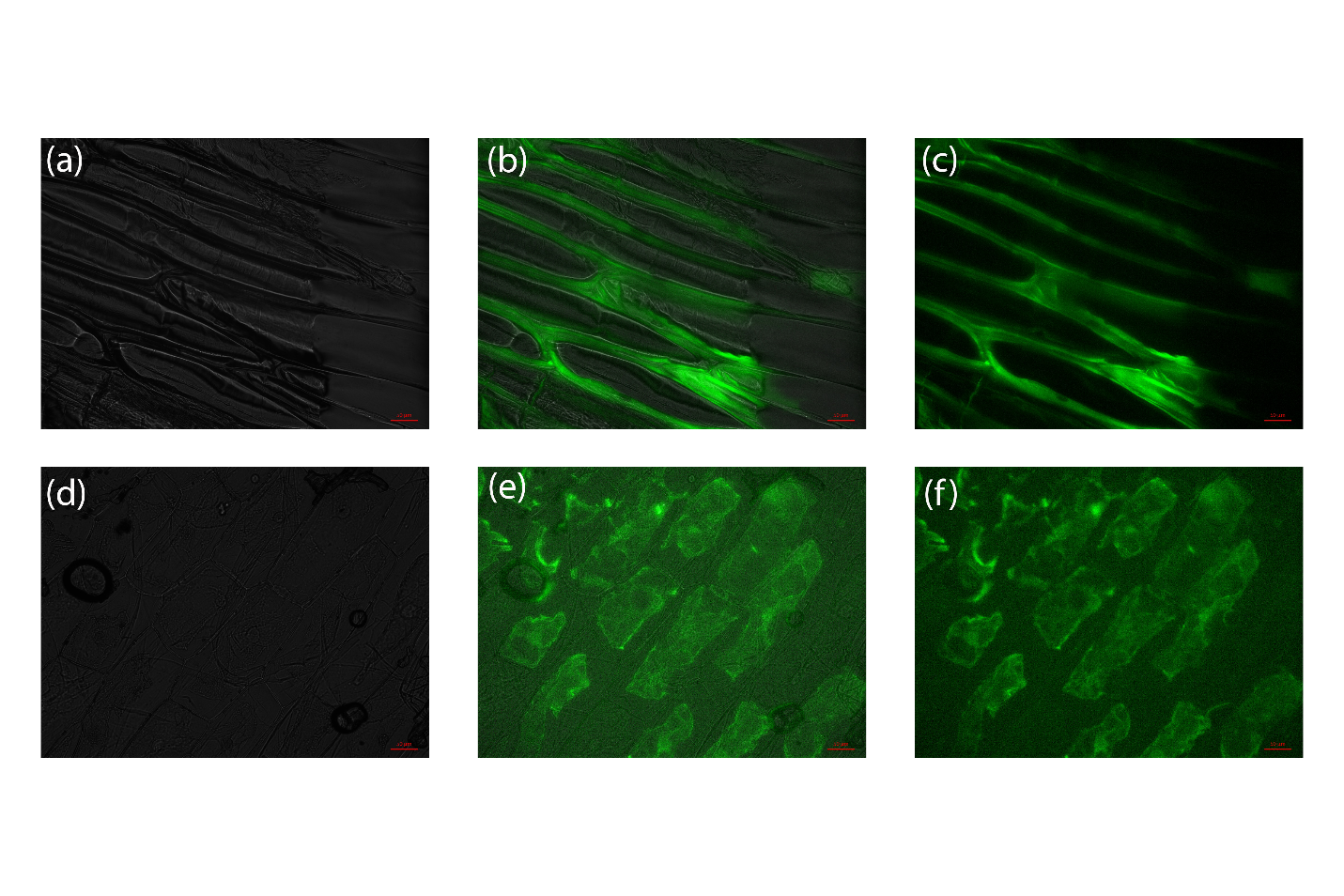
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ADDITIONAL FILE 1



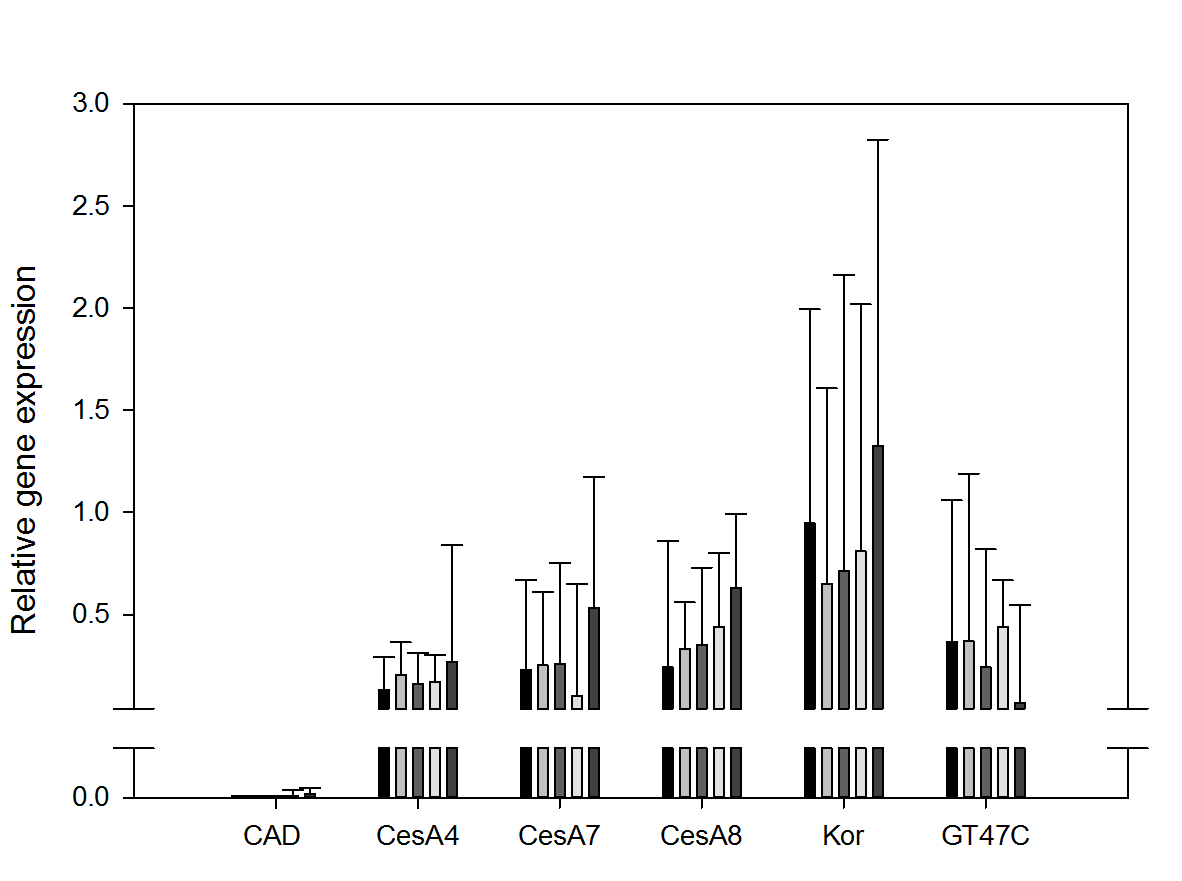
**Fig S1. Fig S1.** Synthesized *TnCelB* gene sequence and splicing confirmation. (a) Codon optimized sequence of TnCelB (b) Primer location to detect splicing of intron (c) PCR results detecting correct intron splicing



**Fig S2.** Mutated PR1a targeted GFP to the apoplast. Onion epidermal cells infiltrated with GFP fused with the mutated PR1a sequence (a,b,c) or with GFP without the targeting sequence (d,e,f); pictures were taken under white field (a,d); fluorescent light (e,f); merged (b,e)



**Fig S3.** Abnormal phenotypes of CaMV-4. (a) Wrinkled leaf surface of CaMV-4 line expressing *Tn*CelB compared with WT; (b) Dehydrated apical region of CaMV-4 line expressing *Tn*CelB compared to WT

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**Fig S4.** Expression level of genes involved in synthesis of monolignol (CAD), cellulose (CesA4, CesA7, CesA8, Kor), and hemicellulose (GT47C). Three individual plants per event were analyzed. Error bars are standard error.

**Table S1.** Primers used in the study

|  |  |  |  |
| --- | --- | --- | --- |
| **Primer name** | **Sequence** | **Note** | **Tm(°C)** |
| **PRSE-Fwd** | GCGGCCGCCATGGAGTTCCTCAAAAGCTTCC | Forward primer to amplify Pr1a sequence with Asn mutated to Glu, NotI site added | 65 |
| **TnCelB-Rvs-Not1** | GCGGCCGCTTATTCTCCAATCTCCACAGAA | Reverse primer for cloning TnCelB, NotI site added |
| **UBC11-Fwd** | GTTGATTTTTGCTGGGAAGC | Homologous locus in *Arabidopsis*: NM\_001125464 | 60 |
| **UBC11-Rvs** | GATCTTGGCCTTCACGTTGT |
| **EF1β-Fwd** | GGCATTAAGTTTTGTCGGTCTG | elongation factor 1-beta 2 (LOC7489318) | 60 |
| **EF1β-Rvs** | GCGGTTCATCATTTCATCTGG |
| **TnCelB-Fwd** | ATGCGTTTGGTGGTTTCTTTTC | Forward primer for qPCR | 60 |
| **TnCelB-Rvs** | AAAGTTGAGTTCCATTGTAACT | Reverse primer for qPCR |
| **CesA4-Fwd** | CACAGGTTATCCCACTTTTGCT | JCI accession number: eugene3.00002636 | 60 |
| **CesA4-Rvs** | CATACGCTTGCTTGCTAACAGA |
| **CesA7-Fwd** | CAAGCAATGTGGACTCAACTGTTA | JCI accession number: gw1.XVIII.3152.1 | 60 |
| **CesA7-Rvs** | AAGCAGGATGCACATGTATCTTCT |
| **CesA8-Fwd** | AAGCACATATCGCTGTCAGTATTTA | JCI accession number: eugene3.00040363 | 60 |
| **CesA8-Rvs** | TTCAACACAATCAAAACCTGTATTT |
| **Kor-Fwd** | GCAGCAAAATCATCTTACCAA | GenBank: AY535003.1 | 60 |
| **Kor-Rvs** | GGATTGACAAGAACACCATAT |
| **CAD1-Fwd** | GCAAGCTTATTCACTGAACAACAAT | JCI accession number: estExt\_Genewise1\_v1.C\_LG\_IX2359 | 60 |
| **CAD1-Rvs** | AAAGCAAAGACACACTGTCACATTT |
| **GT47C-Fwd** | TCCTCCAACTCCACTTTCATTC | Accession numbers: PoGT47C (DQ899955) | 60 |
| **GT47C-Rvs** | AGCCAGTCCGTGTTGTATTT |
| **CaMV-Fwd** | CTGCAGGGCTAGAGCAGCTTGCCAAC | Primer to check presence of expression cassette | 55 |
| **TnCelB-Rvs-Not1** | GCGGCCGCTTATTCTCCAATCTCCACAGAA | Reverse primer for checking presence of expression cassette |