

# An Expressed Sequence Tag Analysis for the Fast-Growing Shoots of *Bambusa edulis* Murno

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**Abstract** Bamboo is one of the fastest growing plants in the world and is an economically important crop species in Asia. To identify the genes involved in fast shoot growth, an expressed sequence tag analysis was performed on *Bambusa edulis* Murno fast-growing shoots. Sequencing of the cDNA clones generated 1,402 5'-end high-quality expressed sequence tags (JG296384-JG297785, average length 655 bp), of which 1,101 clusters (143 consensus and 958 singletons)

were revealed by sequence comparison to be unique and 597 (54% of total clusters) of them have a putative ATG start codon. A Basic Local Alignment Search Tool X analysis showed that 995 of these genes were similar to genes present in the National Center for Biotechnology database. A total of 868 genes were most similar to rice genes. The most abundant genes were three thionin-coding genes, which have 27, 17, or 10 clones, respectively, followed by aminocyclopropanecarboxylate oxidase and cysteine protease. Thionin and putative cell elongation-associated genes, xyloglucan endotransglycosylase/hydrolase, expansin, cellulose synthase, and pectin esterase were analyzed by real-time reverse transcription polymerase chain reaction using gene-specific primers. These results suggest that this high-quality library could be a good resource for understanding molecular events of bamboo shoot elongation, and the full-length clones could be used for crop improvement studies in the future.

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## Abbreviations

ACC Aminocyclopropanecarboxylate  
EST Expressed sequence tag  
NCBI National Center for Biotechnology  
XTH Xyloglucan endotransglycosylase/hydrolase  
UDPG Uridine diphosphate glucose  
GAP Glyceraldehyde 3-phosphate

## Introduction

Bamboo is an economically important plant resource in the Asian culture. Its products are widely used for construction

materials, food source, and musical instruments. Bamboo is one of the tallest species in the Poaceae and is one of the fastest growing plants in the world (Liu et al. 2008).

Shoot growth of bamboo plants is very important for economic reasons. The shoot is the major part of plant biomass which is used for manufacturing and is essentially determining plant height and crop efficiency (Lawson and Poethig 1995). Shoot growth consists of three steps: cell division, cell expansion, and cell wall hardening (Lee et al. 2001; Choi et al. 2008a). Cell division is tightly regulated by the close interactions among various plant hormones; cell expansion is closely related to cell wall loosening, cell expansion by turgor pressure, and cellulose synthesis; cell wall hardening is accompanied by secondary cell wall deposition. Shoot growth is controlled by various endogenous plant hormones and environmental signals (Zhang et al. 2009).

Monocot plants are good subject in which to study fast shoot growth because all stages of growth are merged together in a growing internode. Bamboo has a specific feature of shoot growth: growing rapidly in a short period of time (Gritsch and Murphy 2005). Genes encoding sucrose synthase, invertase, and cellulose synthase have been isolated and characterized from various bamboo species (Chiu et al. 2006; Hsieh et al. 2006; Chen et al. 2010). Expressed sequence tag (EST) analysis was performed with in vitro-grown *Bambusa edulis* Murno shoots to identify the genes involved in photosynthesis (Liu et al. 2008) and with samples of various tissues to construct a full-length cDNA database (Peng et al. 2010). Microarray analysis was conducted of an albino mutant of bamboo (Lin et al. 2009). However, the growth of bamboo is still lacking in gene analysis for genes related to fast shoot elongation.

In this research, we constructed a cDNA library from fast-growing bamboo shoots and established an EST database. We compared the database with ESTs acquired from plants grown under different conditions and analyzed the expression of highly expressed genes and the genes related to cell wall growth.

## Materials and Methods

### Plant Materials

*B. edulis* Murno shoots were collected from the Korea Forest Research Institute in Jinju, Republic of Korea, on May 23, 2008. After collection, bamboo stems were rapidly frozen in liquid nitrogen and stored at  $-70^{\circ}\text{C}$  until needed.

### cDNA Library Construction

Total RNA of the collected bamboo shoots was extracted using TRIzol (Invitrogen) according to the manufacturer's

instructions. Poly(A)+mRNA was isolated using the Poly-ATtract mRNA Isolation System (Promega), and a cDNA library was constructed using a cDNA Synthesis Kit (Uni-ZAP XR vector, Stratagene). Mass excision was performed using XL1-Blue-MRF (Stratagene) as host cells and ExAssist (Stratagene) as helper phage.

### Sequencing and Sequence Analysis

A total of 1,511 cDNA clones were sequenced for analysis of the 5' region. We performed sequencing reactions in a MJ Research PTC-225 Peltier Thermal Cycler using an ABI PRISM BigDye Terminator Cycle Sequencing Kit with AmpliTaq DNA polymerase (FS enzyme; Applied Biosystems) following the protocols supplied by the manufacturer. Single-pass sequencing was performed on each template using T3 primer. The fluorescent-labeled fragments were purified from the unincorporated terminators with an ethanol precipitation protocol. The samples were resuspended in distilled water and subjected to electrophoresis in an ABI 3730xl sequencer (Applied Biosystems).

Sequence analysis was conducted using GeneMaster Ver 3.0 (Ensoltech, Korea). Each EST was first individually processed using a multimodule custom pipeline that linked sequence backup, base calling, the elimination of sequences shorter than 50 bp (and low-quality sequences), vector trimming, and sequence assembly. The Basic Local Alignment Search Tool X (BLASTX) algorithm (Altschul et al. 1990) was employed for the assignment of predicted functions to assembled consensus sequences on the basis of homology, as previously described (Lee et al. 2008). Finally, functional categories were defined using the Gene Ontology (GO) Database ([www.geneontology.org](http://www.geneontology.org)) and the BLASTX algorithm. To compare our data and the previous ESTs from *B. edulis* Murno in vitro-grown multiple shoots (Liu et al. 2008), we downloaded a dataset from the National Center for Biotechnology (NCBI) database (FG551848-FG552834) to our local system. Afterwards, sequence assembly was carried out using the combined dataset of previous ESTs and our ESTs to find library-specific or common unigenes.

### Real-Time RT-PCR Analysis

Total RNA (1  $\mu\text{g}$ ) was extracted from each part of internodes of wild-grown shoots using a Plant RNeasy Mini Kit (Qiagen, Germany) and subjected to real-time reverse transcription polymerase chain reaction (RT-PCR). cDNA was synthesized using the Superscript III First-Strand Synthesis System (Invitrogen, Carlsbad, CA, USA). SYBR Green Master Mix (Bio-rad, USA) was used for quantification with iQ5 optical system (Bio-Rad, USA).

The data were analyzed using the delta-delta-Ct method (Livak and Schmittgen 2001), and bamboo alpha-tubulin (accession: JG297090) and ubiquitin (accession: JG296613) were used as internal controls. All data points were calculated from biologically triplicated and technically duplicated measurements.

## Results and Discussion

### EST Analysis

An EST analysis approach was undertaken to identify the genes involved in cell wall elongation of *B. edulis* Murno. A cDNA library was constructed from fast-growing *B. edulis* shoots. Sequencing of 1,511 of the cDNA clones generated 1,402 5'-end high-quality ESTs of which 1,101 were revealed as unigenes by sequence comparison (Table 1). We identified 143 unigenes as consensus sequences and 958 as singleton sequences (Fig. 1).

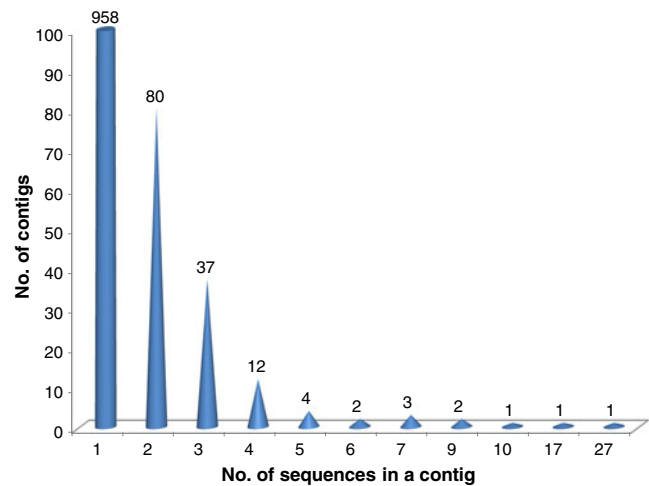
The average length of the EST was  $655 \pm 144$  bp. From the 1,101 clusters, 597 (54% of the clusters) were found to have an “ATG” start codon and a full-length open reading frame. The clones collected from this study could be a good resource for further analysis of *Bambusa* genes or for studies of transformation of other crop plants, including rice.

### Comparison with Other EST Databases from Bamboo

Clustering analysis of combined EST datasets of this study and that of *B. edulis* in vitro cultured shoots (Liu et al. 2008) has shown that 93.2% (1,029 from 1,104 clusters) of our EST was specific to fast-growing tissues (Fig. 2). This result means that a significant portion of the bamboo

**Table 1** Statistics of expressed sequence tags of *B. edulis* fast-growing shoots

Statistics	Number
Base calling sequences	1,511
Low-quality sequences	106
Ambiguous sequences ( $N > 5\%$ )	0
<100 bp	52
All vector regions	21
All low-quality regions	33
All repeat regions	0
High-quality sequences	1,402
Clusters	1,101
Singleton sequences	958
Consensus sequences	143



**Fig. 1** The distribution of copy numbers in the *B. edulis* cDNA library

transcriptome is expressed differentially from the rapidly growing and mature tissues.

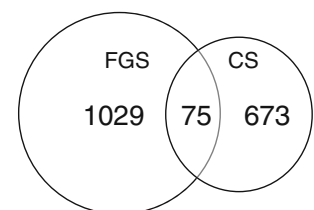
### BLAST Analysis

A BLASTX analysis showed that 995 of these genes were similar to genes present in the NCBI database (cutoff  $E$  value was 0.01). A total of 868 genes were most similar to rice genes and 32, 15, and 14 genes were most similar to those of *Zea mays*, *Hordium vulgare*, and *Triticum aestivum*, respectively (Fig. 3). Only 106 genes showed no significant similarity with any record in the NCBI nr database.

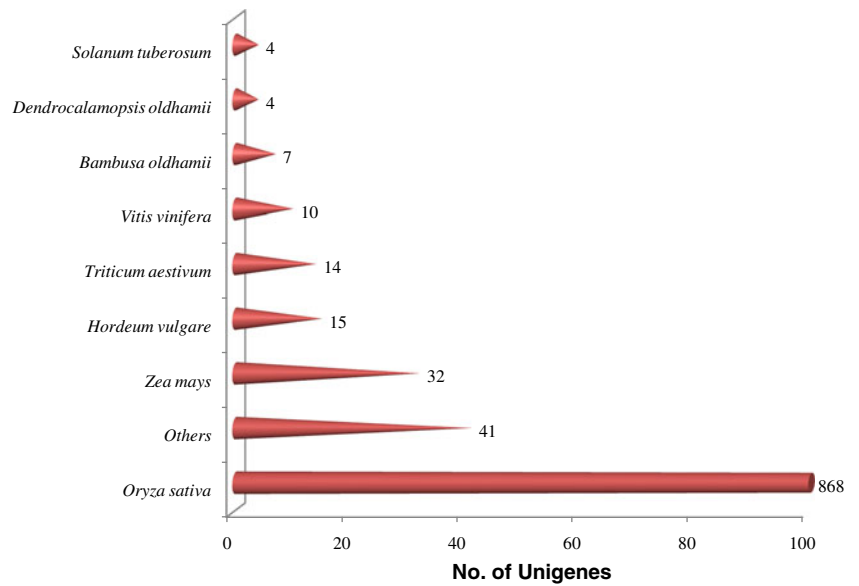
### GO Analysis

Comparing the unigenes with annotations obtained through the Gene Ontology Consortium (Ashburner et al. 2000), we were able to obtain GO terms for 881 unigenes. GO graphs using second-level GO terms are presented under the categories of biological process, molecular function, and cellular components (Fig. 4). Of the cellular component GO terms, 37.7% and 35.4% were directly associated with intracellular organelle components and cytoplasm, respectively. The combined nucleotide binding (17.9%) and hydrolase activity (17.4%) made up more than a third of the molecular functions measured. Under the biological processes category, 18.4% were involved in gene expression and 14.9% were involved in transport.

**Fig. 2** Comparison of EST databases from fast-growing shoots (FGS) and tissue-cultured shoots (CS) of *B. edulis*



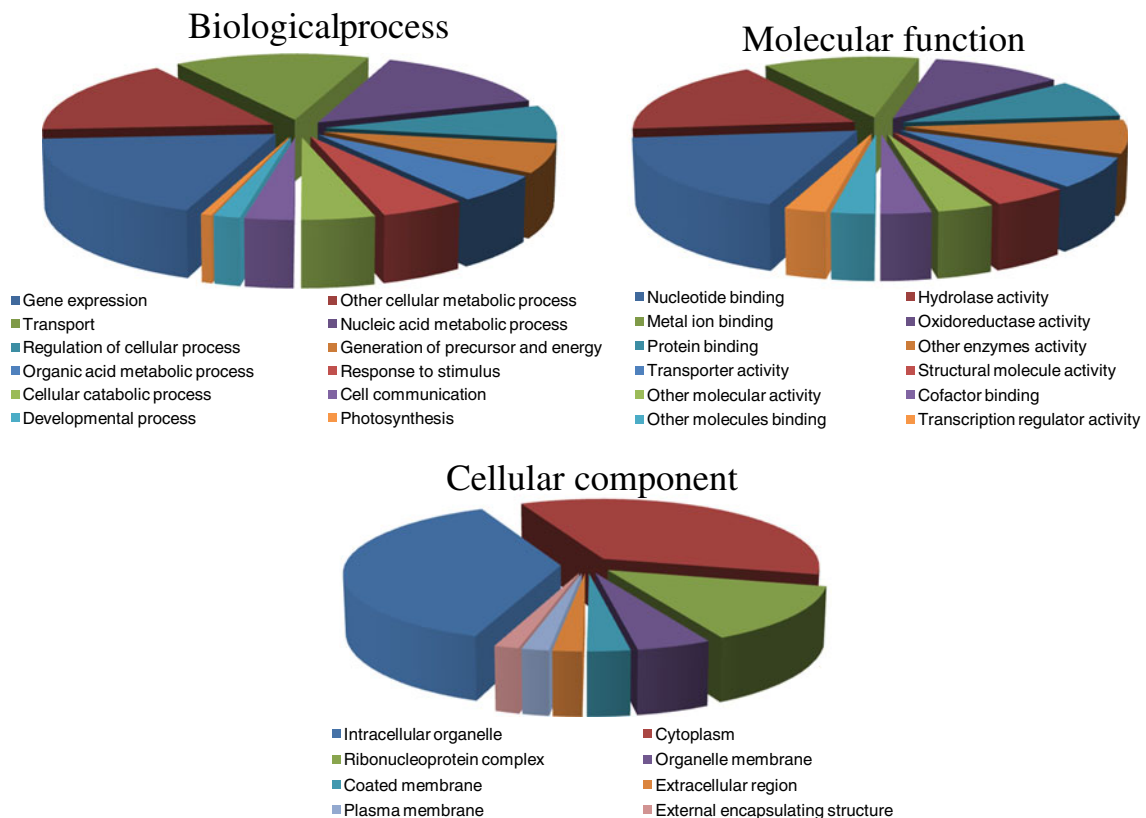
**Fig. 3** The distribution of unique sequences of the *B. edulis* cDNA library having the highest levels of identity with those from other plant species



**Abundant Genes from Bamboo Growing Shoots**

Thionin was the most abundant gene from fast-growing shoots of bamboo (Table 2). The top three most abundant genes were thionin related. The number of clones for clusters 12B08, 08G06, and 13D04 was 27, 17, and 10,

respectively. Thionins belong to a rapidly growing biologically active peptide family in plants (Stec 2006). These peptides are small, cysteine-rich, with toxic and antimicrobial activities, and contributing to plant defense molecules (Epple et al. 1997; Choi et al. 2008b; Hammami et al. 2009). This protein synthesized in a precursor and



**Fig. 4** Pie charts of the second-level GO terms. Overall, 881 unigenes were annotated with GeneMaster software and included in the graphs. Each of three GO categories is presented: biological process, molecular function, and cellular component

**Table 2** Annotations and BLAST scores for *B. edulis* shoot unigene sequences representing genes with a copy number of more than four

Gene ID	Annotation	Most similar gene species	Locus number	<i>E</i> value	Copy number
Contig12B08	Putative thionin	<i>O. sativa</i>	BAD62228.1	5.00E-41	27
Contig08G06	Putative thionin	<i>O. sativa</i>	BAD62228.1	2.00E-42	17
Contig13D04	Thionin	<i>O. sativa</i>	NP_001057737.1	4.00E-31	10
Contig14E01	ACC oxidase	<i>Triticum monococcum</i>	ABJ15735.1	6.00E-97	9
Contig01E02	Cysteine proteinase	<i>O. sativa</i>	NP_001052019.1	3.00E-45	9
Contig07H11	40S ribosomal protein s3a	<i>O. sativa</i>	EAZ00522.1	4.00E-08	7
Contig12A02	UDPG 6-dehydrogenase	<i>O. sativa</i>	NP_001066706.1	1.00E-116	7
Contig04G02	<i>S</i> -adenosylmethionine synthase 2	<i>Triticum monococum</i>	ABJ15731	3.00E-67	7
Contig15F02	Peroxidase	<i>O. sativa</i>	EAZ12255.1	6.00E-99	6
Contig04E04	B22EL8	<i>Hordeum vulgare</i>	AAB19699.1	2.7	6
Contig10A04	Beta-glucosidase	<i>Triticum aestivum</i>	AAR20919.1	1.00E-86	5
Contig014T3	Alpha tubulin	<i>Setaria viridis</i>	CAE52515.1	1.00E-124	5
Contig10E08	Fructokinase-2	<i>O. sativa</i>	EAZ05375.1	1.00E-101	5
Contig06A12	GAP dehydrogenase	<i>O. sativa</i>	NP_001053139.1	1.00E-132	5

*GAP* glyceraldehyde 3-phosphate

processed by vacuolar protease to a peptide of about 5 kD (Romero et al. 1997). Gene expression of thionin is induced by methyl jasmonate treatment in barley (Andresen et al. 1992). Interestingly, the most abundant EST from fast-growing bamboo shoots was not reported from the previous studies on bamboo genes. This means that thionin must be expressed only in a specific period of time, e.g., during shoot growth. It is supposed that thionin could have a function as a protective peptide in soft regions against predators or pests, as viscotoxins or thionins found from *Viscum album* (Holtorf et al. 1998; Giudici et al. 2006).

Ethylene is responsible for seed germination, root initiation, growth, floral differentiation, initiation of fruit ripening, senescence, dormancy, sex differentiation, and responding to environment stresses. One of the rate-limiting steps for ethylene biosynthesis is catalyzed by 1-aminocyclopropane-1-carboxylate (ACC) oxidase (Chang and Bleecker 2004). ACC oxidase has nine clones in our study (Table 2). It was reported that the gene was highly expressed from the elongating cotton fiber (Shi et al. 2006). ACC oxidase must be related with the rapid shoot elongation of bamboo shoots, and the scar surrounding the growing shoot might prevent the diffusion of the produced ethylene from the elongating shoot.

Cysteine protease showed nine clones in this study (Table 2). Cysteine protease could be related to pathogen defense (van der Hoorn 2008). There is some disagreement as to whether cysteine protease activity is related to cell wall growth (Grobe et al. 1999; Grobe et al. 2002). Uridine diphosphate glucose (UDPG)-dehydrogenase participates in four metabolic pathways: pentose and glucuronate interconversions, ascorbate and aldarate metabolism, starch and sucrose metabolism, and nucleotide sugar metabolism. *S*-

adenosylmethionine synthase functions in the synthesis of *S*-adenosylmethionine. This molecule also serves as a methyl group donor in many transmethylation reactions and plays a role in ethylene biosynthesis (Peleman et al. 1989a, b)

In addition to defense-related genes and ethylene biosynthesis-related genes, housekeeping genes, ribosomal protein, UDPG 6-hydrogenase, peroxidase, tubulin, and glyceraldehyde 3-phosphate dehydrogenase are highly expressed and contribute to growth in the fast-growing region of bamboo shoots. It is reasonable that these genes are considered housekeeping genes and that the fast-growing shoot needs high levels of metabolites and cell-forming materials.

#### Quantitative RT-PCR of Thionin and Cell Growth-Related Genes

The most abundant gene, thionin, and putative cell wall elongation-related genes, expansin, cellulose synthase, pectin esterase, and xyloglucan endotransglucosylase/hydrolase (XTH), were analyzed using real time RT-PCR with gene-specific primers from the elongating internodes of shoots (Table 3). The genes were differentially expressed from elongating internodes of bamboo shoots. Thionins were most highly expressed in the basal region of the internode, the softest part, and dramatically decreased with increasing distance from the node; we could not detect expression from regions further than 4 cm from the node (Fig. 5). If the thionin is not involved in cell wall elongation or cell wall formation, it may protect the soft region of the shoot from pathogen or insect damage (Florack and Stiekema 1994).

**Table 3** Primer sets used for the quantitative reverse-transcriptase-PCR

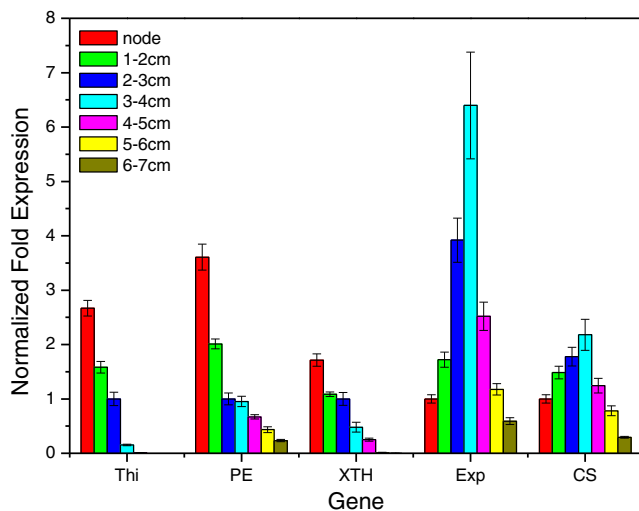
Gene name	Sequence name	Accession number	Primer name	Primer sequence(5' to 3')
Thionin	07B12	JG296801	YL959	TCCCACGGTTGTTGCTTAAGAAAG
			YL960	ACACGCACATAAGATGGAACACTG
Pectin esterase	07G05	JG296431	YL905	GAGGTGCCTTCTTCTTGACAATC
			YL906	AGCACTGCATTCCGATTACATCAC
Xyloglucan endotransglucosylase/hydrolase 3	14A05	JG296597	YL917	TACCCGTGTGATTCTGTTCAGATTATG
			YL918	GTGAGACGACCATACAGGCATAC
Expansin	15E04	JG296918	YL919	GACTTCTTCTCCGCAAGCATTGG
			YL920	ATTAGCAGCCTCAGCGTAGCC
Cellulose synthase	13B11	JG297342	YL913	CCTCCCTTGCCATGAAACCTG
			YL914	AACAGCATCGCCATAGTATGTATAAC
$\alpha$ -Tubulin	12A10	JG297090	YL957	TGAGGAAGGTGATGAGGGAGAC
			YL958	AGATCATAGGATAGCAGTAGTAGGC
Ubiquitin	13D07	JG296613	YL915	CCATAGCTGAAGGACATTAGGAGAG
			YL916	GCAGAAGCAGGTGGATATACTAGG

The expression pattern of pectin esterase and XTH showed similar expression patterns with those of thionin. Pectin esterase could have two functions, one related to modulation of cell wall mechanical stability and the other to cell wall loosening (Ridley et al. 2001; Di Matteo et al. 2005). Loosening of cell wall structure to allow for cell expansion is regulated both by expansins (Cosgrove 2005) and XTHs (Fry et al. 1992; Lee et al. 2010).

Expansin and cellulose synthase expression levels were highest from the middle of the growing internodes. The expansin expression level was similar to that of the EXPA expression pattern from rice leaves. The expression level is

highest from the middle region of the elongating leaves (Lee and Kende 2002) and gradually decreases as the cell wall hardens. Cellulose synthase is differentially expressed for fine regulation of cell wall synthesis. A similar result was reported from research in other bamboo, *Bambusa oldhamii* (Chen et al. 2010).

In conclusion, the expression pattern in fast-growing bamboo shoots is very different from that in normal bamboo shoots. Ethylene production genes, ACC oxidase and *S*-adenosylmethionine synthase, and protective genes for soft regions, thionin and cysteine protease, are highly expressed and are precisely regulated during shoot growth.



**Fig. 5** Quantitative reverse-transcriptase-PCR analysis of thionin and cell wall elongation related to gene expression. *Thi* thionin, *PE* pectin esterase, *XTH* xyloglucan endotransglucosylase/hydrolase, *Exp* expansin, *CS* cellulose synthase

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## References

- Altschul SF, Gish W, Miller W, Myers EW, Lipman DJ (1990) Basic local alignment search tool. *J Mol Biol* 215:403–410
- Andresen I, Becker W, Schluter K, Burges J, Parthier B, Apel K (1992) The identification of leaf thionin as one of the main jasmonate-induced proteins of barley (*Hordeum vulgare*). *Plant Mol Biol* 19:193–204
- Ashburner M, Ball CA, Blake JA, Botstein D, Butler H, Cherry JM, Davis AP, Dolinski K, Dwight SS, Eppig JT, Harris MA, Hill DP, Issel-Tarver L, Kasarskis A, Lewis S, Matese JC, Richardson JE, Ringwald M, Rubin GM, Sherlock G (2000) Gene ontology: tool for the unification of biology. The Gene Ontology Consortium. *Nat Genet* 25:25–29
- Chang C, Bleecker AB (2004) Ethylene biology. More than a gas. *Plant Physiol* 136:2895–2899

- Chen CY, Hsieh MH, Yang CC, Lin CS, Wang AY (2010) Analysis of the cellulose synthase genes associated with primary cell wall synthesis in *Bambusa oldhamii*. *Phytochemistry* 71:1270–1279
- Chiu WB, Lin CH, Chang CJ, Hsieh MH, Wang AY (2006) Molecular characterization and expression of four cDNAs encoding sucrose synthase from green bamboo *Bambusa oldhamii*. *New Phytol* 170:53–63
- Choi D, Kim JH, Lee Y (2008a) Expansins in plant development. *Adv Bot Res* 47:51p
- Choi Y, Choi YD, Lee JS (2008b) Antimicrobial activity of gamma-thionin-like soybean SE60 in *E. coli* and tobacco plants. *Biochem Biophys Res Commun* 375:230–234
- Cosgrove DJ (2005) Growth of the plant cell wall. *Nat Rev Mol Cell Biol* 6:850–861
- Di Matteo A, Giovane A, Raiola A, Camardella L, Bonivento D, De Lorenzo G, Cervone F, Bellincampi D, Tsemoglou D (2005) Structural basis for the interaction between pectin methylesterase and a specific inhibitor protein. *Plant Cell* 17:849–858
- Epple P, Apel K, Bohlmann H (1997) Overexpression of an endogenous thionin enhances resistance of *Arabidopsis* against *Fusarium oxysporum*. *Plant Cell* 9:509–520
- Florack DEA, Stiekema WJ (1994) Thionins—properties, possible biological roles and mechanisms of action. *Plant Mol Biol* 26:25–37
- Fry SC, Smith RC, Renwick KF, Martin DJ, Hodge SK, Matthews KJ (1992) Xyloglucan endotransglycosylase, a new wall-loosening enzyme activity from plants. *Biochem J* 282(Pt 3):821–828
- Giudici M, Poveda JA, Molina ML, de la Canal L, Gonzalez-Ros JM, Pfuller K, Pfuller U, Villalain J (2006) Antifungal effects and mechanism of action of viscotoxin A(3). *FEBS J* 273:72–83
- Gritsch CS, Murphy RJ (2005) Ultrastructure of fibre and parenchyma cell walls during early stages of culm development in *Dendrocalamus asper*. *Ann Bot* 95:619–629
- Grobe K, Becker WM, Schlaak M, Petersen A (1999) Grass group I allergens (beta-expansins) are novel, papain-related proteinases. *Eur J Biochem* 263:33–40
- Grobe K, Poppelmann M, Becker WM, Petersen A (2002) Properties of group I allergens from grass pollen and their relation to cathepsin B, a member of the C1 family of cysteine proteinases. *Eur J Biochem* 269:2083–2092
- Hammami R, Ben Hamida J, Vergoten G, Fliss I (2009) PhytAMP: a database dedicated to antimicrobial plant peptides. *Nucleic Acids Res* 37:D963–968
- Holtorf S, Ludwig-Muller J, Apel K, Bohlmann H (1998) High-level expression of a viscotoxin in *Arabidopsis thaliana* gives enhanced resistance against *Plasmodiophora brassicae*. *Plant Mol Biol* 36:673–680
- Hsieh CW, Liu LK, Yeh SH, Chen CF, Lin HI, Sung HY, Wang AY (2006) Molecular cloning and functional identification of invertase isozymes from green bamboo *Bambusa oldhamii*. *J Agric Food Chem* 54:3101–3107
- Lawson EJ, Poethig RS (1995) Shoot development in plants: time for a change. *Trends Genet* 11:263–268
- Lee H, Cho HH, Kim IC, Yim JH, Lee HK, Lee YK (2008) Expressed sequence tag analysis of Antarctic hairgrass *Deschampsia antarctica* from King George Island, Antarctica. *Mol Cells* 25:258–264
- Lee J, Burns TH, Light G, Sun Y, Fokar M, Kasukabe Y, Fujisawa K, Maekawa Y, Allen RD (2010) Xyloglucan endotransglycosylase/hydrolase genes in cotton and their role in fiber elongation. *Planta* 232:1191–1205
- Lee Y, Choi D, Kende H (2001) Expansins: ever-expanding numbers and functions. *Curr Opin Plant Biol* 4:527–532
- Lee Y, Kende H (2002) Expression of alpha-expansin and expansin-like genes in deepwater rice. *Plant Physiol* 130:1396–1405
- Lin CS, Wu FH, Liu NT, Chou SJ, Shen SC, Chang BCH, Pan CY (2009) Identification of repressed gene transcript accumulation in three albino mutants of *Bambusa edulis* Munro by cDNA microarray analysis. *J Sci Food Agr* 89:2308–2316
- Liu NT, Wu FH, Tsay HS, Chang WC, Lin CS (2008) Establishment of a cDNA library from *Bambusa edulis* Munro in vitro-grown shoots. *Plant Cell Tissue Organ Cult* 95:21–27
- Livak KJ, Schmittgen TD (2001) Analysis of relative gene expression data using real-time quantitative PCR and the 2<sup>-(delta delta C<sub>T</sub>)</sup> method. *Methods* 25:402–408
- Peleman J, Boerjan W, Engler G, Seurinck J, Botterman J, Alliotte T, Van Montagu M, Inze D (1989a) Strong cellular preference in the expression of a housekeeping gene of *Arabidopsis thaliana* encoding S-adenosylmethionine synthetase. *Plant Cell* 1:81–93
- Peleman J, Saito K, Cottyn B, Engler G, Seurinck J, Van Montagu M, Inze D (1989b) Structure and expression analyses of the S-adenosylmethionine synthetase gene family in *Arabidopsis thaliana*. *Gene* 84:359–369
- Peng ZH, Lu TT, Li LB, Liu XH, Gao ZM, Hu T, Yang XW, Feng Q, Guan JP, Weng QJ, Fan DL, Zhu CR, Lu Y, Han B, Jiang ZH (2010) Genome-wide characterization of the biggest grass, bamboo, based on 10,608 putative full-length cDNA sequences. *BMC Plant Biol* 10:116
- Ridley BL, O'Neill MA, Mohnen D (2001) Pectins: structure, biosynthesis, and oligogalacturonide-related signaling. *Phytochemistry* 57:929–967
- Romero A, Alamillo JM, Garcia-Olmedo F (1997) Processing of thionin precursors in barley leaves by a vacuolar proteinase. *Eur J Biochem* 243:202–208
- Shi YH, Zhu SW, Mao XZ, Feng JX, Qin YM, Zhang L, Cheng J, Wei LP, Wang ZY, Zhu YX (2006) Transcriptome profiling, molecular biological, and physiological studies reveal a major role for ethylene in cotton fiber cell elongation. *Plant Cell* 18:651–664
- Stec B (2006) Plant thionins—the structural perspective. *Cell Mol Life Sci* 63:1370–1385
- van der Hooft RA (2008) Plant proteases: from phenotypes to molecular mechanisms. *Annu Rev Plant Biol* 59:191–223
- Zhang LY, Bai MY, Wu J, Zhu JY, Wang H, Zhang Z, Wang W, Sun Y, Zhao J, Sun X, Yang H, Xu Y, Kim SH, Fujioka S, Lin WH, Chong K, Lu T, Wang ZY (2009) Antagonistic HLH/bHLH transcription factors mediate brassinosteroid regulation of cell elongation and plant development in rice and *Arabidopsis*. *Plant Cell* 21:3767–3780