Plant Physiol. (1993) 101: 697–698

Plant Gene Register

Cotton (*Gossypium hirsutum*) *MatP6* and *MatP7* Oleosin Genes[1]

D. Wayne Hughes, Helen Y.-C. Wang, and Glenn A. Galau*

Department of Botany, University of Georgia, Athens, Georgia 30602

Cotton (*Gossypium hirsutum*) *MatP* genes are expressed during both the maturation and postabscission stages of embryogenesis (Hughes and Galau, 1989). *MatP* genes are also environmentally induced, along with the *Lea* and *LeaA* components of the postabscission program, in embryos by desiccation or during culture in ABA or high osmoticum (Hughes and Galau, 1991; M. Swain, G.A. Galau, unpublished observations). The various inducers of *Lea* and *LeaA* genes are consistent with their probable role in water stress and desiccation protection (Hughes and Galau, 1989, 1991), but it is unclear what functions *MatP* genes have, or if, as predicted (Hughes and Galau, 1989, 1991), their regulation is by the same mechanisms that separately regulate maturation-specific and postabscission/water stress-specific genes. We report here that two *MatP* genes encode oleosins, which comprise most of the protein in oil body half-unit membranes (reviewed by Huang, 1992; Keddie et al., 1992). Oleosins have been suggested to stabilize oil bodies, especially during embryo desiccation (Murphy et al., 1989). The *MatP6* gene contains sequences found separately in maturation-specific and postabscission/water stress-specific cotton genes, suggesting that its regulation is in fact by separate program-specific and stress-specific mechanisms.

MatP6-A cDNA D129, *MatP7-D* cDNA D103, the A genome alloallele *MatP6-A*, and a small part of *MatP7-A* were sequenced (Table I). *MatP6-A* encodes an 18.1-kD oleosin and *MatP7-D* cDNA D103 encodes a 16.4-kD oleosin. In colinear regions, the two cotton oleosins are about 77% identical, and they are about 55% identical to a carrot oleosin (Hatzopoulos et al., 1990). They are 50 to 75% identical to all sequenced oleosins in the central hydrophobic region, and they share the conserved features identified by others (Huang, 1992; Keddie et al., 1992).

Possible maturation-specific and stress-responsive sequences are both present in *MatP6-A*. Beginning at −102 nucleotides relative to the transcription start, there is a 26-nucleotide sequence that has similarity to both the legumin box and the 2S albumin element present in the cotton maturation-specific genes *Mat2-D Legumin A* (Galau et al., 1991)

Table I. *Characteristics of oleosin gene and cDNAs from Gossypium hirsutum*

Organism:
Gossypium hirsutum L. cv Coker 201 (Upland cotton), Malvaceae.

Function:
Encode oleosin oil body membrane proteins.

Expression:
During the maturation and postabscission stages of embryo development and during water-related stress in embryo culture (Hughes and Galau, 1989, 1991).

Source:
Nuclear DNA from embryo cotyledons 20 to 23 d postanthesis (preendoreduplication). A partial *Sau*3AI digest was cloned in LambdaGEM-12 (Promega), and the phage were identified by hybridization with *MatP7-D* cDNA clone D103. The cDNA clones have been described (Hughes and Galau, 1989).

MatP6-A:
Phage isolate 41 contains the A genome alloallele *MatP6-A*, based on the size of the terminal *Eco*RI fragment that hybridizes to *MatP6* cDNA D129 (Hughes and Galau, 1989). Cloning of this 6.5-kb *Eco*RI fragment into Bluescript (Stratagene) was followed by subcloning of restriction fragments and deletions and complete dideoxy sequencing of both strands with *Taq* polymerase using double-stranded templates.

MatP6-A cDNA D129 and *MatP7-D* cDNA D103:
Subcloning of the cDNA insert into Bluescript was followed by complete sequencing of both strands as described above. The D129 clone contains a portion of its 3′ end in reverse orientation on its 5′ end (not reported). The cDNA sequence is identical to the gene sequence except that it contains (one fewer residue in the poly(dG) sequence beginning at *MatP6-A* nucleotide 4011, which is believed to be due to an error made during cDNA synthesis. In a comparison of a partially sequenced *MatP7-A* gene (clone GD103-27R) and cDNA D103, there are two substitutions in the 3′ nontranslated region, suggesting that cDNA D103 is encoded by the other alloallele, *MatP7-D*.

Transcription Start:
A leftward primer (*MatP6-A* nucleotide 3362-3343) was used in primer extension to determine the transcription start of *MatP6-A* mRNAs present in maturation-stage and postabscission stage embryos.

Abbreviations: *MatP*, gene expressed during maturation and postabscission stages of embryogenesis; *Lea*, late embryogenesis abundant.

[1] Supported by grants from the National Institutes of Health and the United States Department of Agriculture Competitive Grants Program.

* Corresponding author; fax 1–706–542–1805.

and *Mat5-A* 2S albumin (Galau et al., 1992a), respectively. An ACGTG-containing element has been shown to be involved in the ABA induction of a wheat *Lea* gene (Guiltinan et al., 1990). As has also been found in some other oleosin genes (Huang, 1992; Keddie et al., 1992) and most cotton *Lea* and *LeaA* genes (Galau et al., 1992b), the cotton oleosin gene contains such a sequence at −65 nucleotides from the transcription start. Putative postabscission-responsive elements have not yet been identified by comparative sequence or functional analysis.

Received September 1, 1992; accepted September 14, 1992.
Copyright Clearance Center: 0032-0889/93/101/0697/02.
The GenBank/EMBL accession numbers for the sequences reported in this article are *MatP6-A* GD129-41RX, L00936; *MatP6-A* cDNA D129, L00935; *MatP7-D* cDNA D103, L00934.

LITERATURE CITED

Galau GA, Wang HY-C, Hughes DW (1991) Sequence of the *Gossypium hirsutum* D-genome alloallele of *Legumin A* and its mRNA. Plant Physiol **97:** 1268–1270

Galau GA, Wang HY-C, Hughes DW (1992a) Cotton *Mat5-A* (C164) gene and *Mat5-D* cDNAs encoding methionine-rich 2S albumin storage proteins. Plant Physiol **99:** 779–782

Galau GA, Wang HY-C, Hughes DW (1992b) Cotton *Lea4* (D19) and *LeaA2* (D132) Group 1 *Lea* genes encoding water stress-related proteins containing a 20-amino acid motif. Plant Physiol **99:** 783–788

Guiltinan MJ, Marcotte WR Jr, Quatrano RS (1990) A plant leucine zipper protein that recognizes an abscisic acid response element. Science **250:** 267–271

Hatzopoulos P, Franz G, Choy L, Sung RZ (1990) Interaction of nuclear factors with upstream sequences of a lipid body membrane protein gene from carrot. Plant Cell **2:** 457–467

Huang AHC (1992) Oil bodies and oleosins in seeds. Annu Rev Plant Physiol Plant Mol Biol **43:** 177–200

Hughes DW, Galau GA (1989) Temporally modular gene expression during cotyledon development. Genes Dev **3:** 358–369

Hughes DW, Galau GA (1991) Developmental and environmental induction of *Lea* and *LeaA* mRNAs and the postabscission program during embryo culture. Plant Cell **3:** 605–618

Keddie JS, Hübner G, Slocombe SP, Jarvis RP, Cummins I, Edwards E-w, Shaw CH, Murphy DJ (1992) Cloning and characterisation of an oleosin gene from *Brassica napus*. Plant Mol Biol **19:** 443–453

Murphy DJ, Cummins I, Kang AS (1989) Synthesis of the major oil body membrane protein in developing rape embryos. Biochem J **65:** 285–293