

Plant Gene Register

Sequence of a Poplar Bark Storage Protein Gene¹

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Temperate deciduous tree species retranslocate nitrogen from leaves to storage sites during the fall and remobilize stored nitrogen during the spring to support new shoot growth (Taylor and May, 1967; Ryan and Bormann, 1982). The majority of reserve nitrogen is stored as protein (Kang and Titus, 1980; Chapin and Kedrowski, 1983). In poplar, a 32-kD BSP that accumulates in the protein storage vacuoles of the inner bark parenchyma during autumn and winter has been described (Wetzel et al., 1989). The accumulation of poplar BSP is associated with short days (Coleman et al., 1991). SD accumulation of poplar BSP is also correlated with a large increase in the steady-state levels of BSP mRNA (Coleman et al., 1992). DNA gel blot analysis suggests that poplar BSP is encoded by a small multigene family.

As part of our effort to understand nitrogen recycling and photoperiod control of BSP gene expression, we have isolated and sequenced a gene encoding a poplar BSP (Table I). A genomic library was prepared in the vector λ Gem-11 (Promega, Madison, WI) and screened at low stringency (approximately 25°C below T_m) using a poplar BSP cDNA (Coleman et al., 1992). Twelve positive clones were identified and rescreened with different regions of the BSP cDNA at high stringency (approximately 5°C below T_m). One positive clone (designated *bspA*) was isolated and subcloned into pBluescriptII SK (Stratagene, La Jolla, CA), and the nucleic acid sequence was determined.

The poplar BSP gene was contained within a 3.9-kb *EcoRV*/*AflIII* fragment, which included a 1646-bp coding region, 1246 bp upstream of the coding region and 1022 bp downstream of the coding region. The open reading frame was interrupted by four introns 152, 262, 175, and 144 nucleotides in length. A putative TATA sequence is located at nucleotides 1145 to 1151 and the transcriptional start site (as determined by primer extension) is located at nucleotide 1175. The DNA sequence is 99% identical with the BSP cDNA, and the decoded amino acid sequence is 97% identical with the amino acid sequence of the poplar BSP cDNA.

Table I. Characteristics of *bspA*

Organism:	<i>Populus deltoides</i> Bartr. ex Marsh, clone 172-2.
Localization of Chromosome:	Unknown: according to Southern data, gene is a member of a small gene family.
Gene function:	32-kD poplar BSP.
Clone Type:	Genomic.
Sources:	Genomic library in λ Gem-11 constructed from leaf genomic DNA.
Sequencing Strategy:	Chain termination sequencing of single-stranded DNA from nested exonuclease III deletions of both strands.
Method of Identification:	Library was screened with a 1.2-kb BSP cDNA and identity confirmed by nucleotide sequence comparison of exons with the cDNA clone of poplar BSP (GenBank accession no. M77504).
Regulation and Expression:	Transcript accumulates to high levels in bark of SD-exposed plants.
(G+C) Content:	32.3%; within reading frame, 43.7%.
Structural Features of Gene:	TATA box, transcriptional start site, translation stop codon, coding region interrupted by four introns.
Antibodies:	Polyclonal rabbit.
Subcellular Location:	Protein storage vacuole of bark phloem parenchyma.

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The EMBL accession number for the sequence reported in this article is X70064.

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Abbreviation: BSP, bark storage protein.

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