

## Plant Gene Register

# Nucleotide Sequence of a Tomato *psbS* Gene

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Examination of O<sub>2</sub>-evolving PSII preparations from spinach has revealed the presence of a 22-kD intrinsic protein component (Berthold et al., 1981), which appears to be associated with the PSII polypeptides constituting the reaction center (polypeptides obligately required for charge separation) and also with the light-harvesting antennae proteins CP29 and CP26 (Ghanotakis et al., 1987; Green and Camm, 1990). Removal of the 22-kD polypeptides has little effect on the kinetics of water oxidation, but the fact that the 22-kD polypeptide is found in PSII of all plants examined (Ghanotakis et al., 1987) and also is reported to be present in *Synechocystis* 6803 PSII (Nilsson et al., 1990) suggests that it must have a role in PSII function or assembly. Although several studies have attempted to define the exact location and function of the polypeptide, the role of the 22-kD PSII protein remains unclear (Ljungberg et al., 1984, 1986; Bowlby and Yocum, 1993; Mishra and Ghanotakis, 1993). However, it has recently been reported to bind Chls, and thus suggested to be the apoprotein of a minor Chl *a/b* complex, CP22, which may play a light-harvesting role (Funk et al., 1994).

The nucleotide sequence of a spinach cDNA clone of the *psbS* gene, encoding the 22-kD protein, has recently been reported (Kim et al., 1992; Wedel et al., 1992). The amino acid sequence derived from the *psbS* cDNA sequence indicates that the protein is highly hydrophobic, with four potential membrane-spanning regions predicted from hydropathy plotting analyses (Kim et al., 1992; Wedel et al., 1992). The protein consists of tandem domains in which there is appreciable amino acid sequence similarity, suggesting that the *psbS* gene arose from an internal gene duplication (Kim et al., 1992; Wedel et al., 1992; Green and Pichersky, 1994). In addition, the transmembrane helices of the 22-kD protein have sequence similarity to the three transmembrane helices of CABs. These data suggest that the CABs and the 22-kD polypeptide arose from a common ancestor that contained four transmembrane helices.

Here we report the isolation of tomato (*Lycopersicon esculentum*) *psbS* genomic and cDNA clones (Table I). The tomato *psbS* gene encodes a precursor polypeptide of 276 amino acids. The gene has three introns: 136 nucleotides between codons 72 and 73, 558 nucleotides between codons 112 and 113, and 247 nucleotides between codons 195 and 196. Comparison with CAB genes shows that none of these introns occur at positions where any of the CAB introns occur. The

**Table I.** Characteristics of *psbS* gene from *L. esculentum*

|                                  |                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                |
|----------------------------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| Organism:                        | <i>Lycopersicon esculentum</i> .                                                                                                                                                                                                                                                                                                                                                                                                                                                                                               |
| Location:                        | Nuclear gene, found on a 4.5-kb <i>Xba</i> I fragment.                                                                                                                                                                                                                                                                                                                                                                                                                                                                         |
| Techniques:                      | The cDNA clone was isolated from a Charon 16 leaf cDNA library (Hoffman et al., 1987) by screening with a spinach <i>psbS</i> cDNA clone. The gene was isolated from a Charon 35 genomic library by screening with the tomato <i>psbS</i> cDNA clone. The sequence of the cDNA clone is identical to the corresponding region in the genomic clone. The isolates were subcloned into the plasmid Bluescript SK+ and sequenced on both strands by the single-stranded dideoxy method. Specific and universal primers were used. |
| Method of Identification:        | The introns in the genomic <i>psbS</i> clone were identified by comparison with the tomato <i>psbS</i> cDNA sequence.                                                                                                                                                                                                                                                                                                                                                                                                          |
| Structural Features of the Gene: | TATA box, G box core consensus, translation initiation and termination sites, common light-responsive element, three introns.                                                                                                                                                                                                                                                                                                                                                                                                  |
| Amino Acid Sequence:             | The gene encodes a protein of 276 amino acids, including a transit peptide of undetermined length.                                                                                                                                                                                                                                                                                                                                                                                                                             |
| Antibodies:                      | No specific antibodies for the <i>L. esculentum</i> protein are available.                                                                                                                                                                                                                                                                                                                                                                                                                                                     |
| Subcellular Location:            | The protein is found associated with PSII in the thylakoid membranes of chloroplasts.                                                                                                                                                                                                                                                                                                                                                                                                                                          |

deduced amino acid sequence of the tomato 22-kD protein is 86% identical (92% with conservative substitutions) with that of the spinach protein. Although the N terminus of the 22-kD polypeptide is blocked (Kim et al., 1992), the transit sequence cleavage site is probably after residue 67 by homology with the spinach protein (Wedel et al., 1992).

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Abbreviation: CAB, Chl *a/b*-binding protein.

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