Plant Gene Register

Nucleotide Sequence of a cDNA Encoding the Small Subunit of Ribulose-1,5-Bisphosphate Carboxylase/Oxygenase from *Aegilops squarrosa*

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Rubisco (EC 4.1.1.39), the most abundant protein in the world, is a bifunctional enzyme that catalyzes the carboxylation and the oxygenation of ribulose-1,5-bisphosphate (Mizioro and Lorimer, 1983). The holoenzyme is composed of eight LSUs encoded by chloroplast DNA and synthesized in the chloroplast and eight SSUs encoded by nuclear DNA, synthesized on free cytoplasmic polysomes, transported into chloroplasts, and assembled with the LSUs to form the active enzyme. The SSUs are attached to the top and the bottom of the large octamer (Chapman et al., 1988; Anderson et al., 1989), whereas the binding sites for ribulose-1,5-bisphosphate activator CO₂ and the divalent metal cofactor are all located on the LSUs. The SSU may not be essential for catalysis, but it is evident that it exerts an important role in maximizing the activity of the enzyme (Andrews, 1988; Gutteridge, 1991). Here we cloned a full-length cDNA for the SSU of Rubisco from the C₃ monocot *Aegilops squarrosa*, which belongs to one of the genera nearest to wheat (Table I). An *A. squarrosa* cDNA library was prepared and screened with a *rbcS* DNA fragment of rice (a generous gift from Prof. Ray Wu, Cornell University) (Xie et al., 1987).

An 815-bp nucleotide sequence was obtained that includes 525 bp of coding region (from 39–563), 38 bp of 5' untranslated region, and 252 bp of 3' untranslated region. The transit and mature peptide have, respectively, 47 and 128 amino acid residues. Comparison with the coding region of wheat Rubisco (Broglie et al., 1983) revealed 87.5% homology for the mature peptide and 80.8% homology for the transit peptide at the amino acid level.

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**Table I. Characteristics of a cDNA encoding the SSUs of Rubisco from *Aegilops squarrosa***

| Organism: | *Aegilops squarrosa*. |
| Clone Type, Designation: | cDNA, full length; pRAS1. |
| Gene Location: | Nuclear genome; chromosome location unknown. |
| Gene Copy Number: | Multiple copy. |
| Gene Product: | Encoding the SSU of Rubisco. |

**Source:**

A λ gt10 cDNA library was constructed from *A. squarrosa* seedling leaf poly(A) RNA (S.-d. Guo, G.-y. Wu, X.-y. Wu, unpublished data) and screened with a [α-³²P]dCTP-radiolabeled rice rbcS DNA probe. The λ clone 1–1 was subcloned into BluescriptSK plasmid vector and sequenced by the dideoxy sequencing method on double-stranded DNA.

**Sequence Identification:**

Sequence comparison with the rbcS gene from other eight plant species in the EMBL and SwissProt sequence data bases at the Life Science Center, Peking University, revealed high conservation of the amino acid sequence.

**Features of Gene Sequence:**

- The *A. squarrosa* rbcS gene consists of 38 bp of 5' noncoding region, 252 bp 3' noncoding region, and triplet codes for 47 amino acid residues of transport peptide and 128 amino acid residues of mature peptide of the SSU.

**Expression:**

- The *A. squarrosa* rbcS gene was expressed in *Escherichia coli*.

**Antibodies:**

Not prepared.

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**Abbreviations:** LSU, large subunit; SSU, small subunit.
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