Plant Gene Register

Two cDNAs for Tomato Heat Stress Transcription Factors¹

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The remarkable conservation of the heat-stress response from bacterial to plant and animal cells includes the structure and function of the heat-stress proteins as well as the control of their stress-dependent expression (for summaries, see Lindquist and Craig, 1988; Nover, 1991). The definition of a universal heat-stress-responsive promoter element shown to function in practically all types of eukaryotic cells (Nover, 1991) led recently to the characterization of the corresponding regulatory genes coding for the HSF (Sorger and Pelham, 1988; Wiederrecht et al., 1988; Clos et al., 1990; Scharf et al., 1990; Sarge et al., 1991; Schuetz et al., 1991). As expected from the conservation of the promoter element, all HSFs are characterized by a similar DNA-binding domain of 93 to 100 amino acid residues and two to three hydrophobic heptad repeats of the Leu-zipper type (for a summary, see Scharf et al., 1993).

Southwestern screening of a λ gt11 cDNA expression library of tomato (*Lycopersicon peruvianum*) resulted in the isolation of three different HSF clones. In correspondence with *hsf* genes of other organisms, one of the tomato genes (*hsf8*) is constitutively expressed. But expression of the two others (*hsf24, hsf30*) is induced by heat stress. The *hsf24* cDNA sequence was published previously (Scharf et al., 1990). This report concerns the cDNA sequences of *hsf8* and *hsf30* and some structural features of the corresponding proteins (Table I; Scharf et al., 1993).

In contrast to HSF24, the C-terminal activation domains of HSF30 and HSF8 are characterized by a third hydrophobic repeat and extended, negatively charged regions dominated by Asp and Glu. A peculiarity of the HSF30 is the isolation of a family of different clones with varying length of the 3' trailor sequence. They evidently result from aberrant splicing events during the heat-stress period. The genomic clones of all three *hsf* genes contain introns at the corresponding positions (K.-D. Scharf, unpublished data).

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 Table I. Characteristics of cDNA clones coding for HSFs HSF8 and

 HSF30

Organism:

- Lycopersicon peruvianum (Peruvian tomato) cell cultures. Location on Chromosome:
- Both represent single-copy genes localized by restriction fragment length polymorphism-mapping on short arm of chromosome 8 of the culture tomato (*L. esculentum*). The *hsf8* gene is close to the end, whereas *hsf30* maps close to the centromere with a 17-centimorgan distance between the genes (Tanksley et al., 1993).

Gene Function:

Encode two transcription factors activating heat-stress-inducible genes (Treuter et al., 1993).

Source:

cDNA library in λgt11 constructed from poly(A)⁺ RNA of cultured *L. peruvianum* cells; Southwestern screening with labeled concatemers of (-nGAAnnTTCn-) oligonucleotide (Scharf et al., 1990).

Sequencing Techniques:

Exonuclease III deletion subcloning and dideoxy sequencing of both strands.

Method of Identification:

Sequence homology of the N-terminal DNA-binding motif with known HSF from tomato and other organisms; gel retardation assays with recombinant HSF8 and HSF30 proteins; transactivation assays with heat-stress-inducible β -glucuronidase reporter constructs using a transient expression assay in tobacco protoplasts (Treuter et al., 1993).

Expression Characteristics:

- HSF8 mRNA (2.0 kb) is constitutively expressed at low levels in cell cultures and all tissues of tomato plants. The mRNA levels are transiently reduced under heat-stress conditions. HSF30 mRNA (1.4 kb) is only observed after heat-stress induction (Scharf et al., 1990, 1993).
- Structural Features of Proteins and Similarity to Other HSFs: HSF8: Open reading frame 527 amino acids; 57.5 kD. HSF30: Open reading frame 351 amino acids; 40.2 kD. Similar to other HSFs, both proteins contain a highly conserved DNAbinding domain near the N terminus and three heptad hydrophobic repeats of the Leu-zipper type in the C-terminal activation domain (Scharf et al., 1993).

The GenBank accession numbers for the sequences reported in this article are X67600 (*hsf8*) and X67601 (*hsf8*).

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Abbreviation: HSF, heat-stress transcription factor.

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