

# A cDNA clone encoding HBP-1b homologue in *Arabidopsis thaliana*

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Our researches on the transcriptional regulation of wheat histone H3 gene have revealed the presence of several *cis*- and *trans*-acting factors (1). One of the best characterized regulatory sequences is the hexameric motif, ACGTCA (2), and we have identified two distinct hexamer-specific DNA-binding proteins, HBP-1a and HBP-1b, in wheat (3). HBP-1b can bind to the hexameric motifs found in the CaMV35S and NOS promoters as well as the hexameric motif of the H3 promoter, whereas the HBP-1a binds exclusively to the H3 promoter (3, 4). We have already isolated cDNA clones encoding HBP-1a and HBP-1b from wheat (5, 6). Structural analyses of the cDNAs revealed that both proteins are member of bZIP-type transcription factors (7). HBP-1b-like factors capable of binding to the hexameric (or related) motif of several T-DNA and plant viral gene promoters have been found as OCSTF in maize (8) and as ASF-1 in tobacco (9), and their cognate cDNAs, OCSBF-1, -2 (10), and TGA1a (11), have been isolated. All of these have bZIP domains and similar DNA-binding specificity. This prompted us to speculate the genes of HBP-1b homologues may widely be present in various plant species. Isolation of the HBP-1b homologues from various plants is necessary to clarify this issue. Furthermore, function of HBP-1b is not known yet. To understand the function of HBP-1b, it will be important to study the transcription factor by genetic approaches as well as biochemical ones. In this respect, we isolated a cDNA clone for HBP-1b counterpart from *Arabidopsis thaliana* which is a useful plant for genetic analyses.

We firstly screened a genomic library of *A. thaliana* (cv. Landsberg) and obtained one positive clone out of  $1 \times 10^5$  plaques by using wheat HBP-1b cDNA as a probe. The genomic clone was used as a probe to isolate a cDNA clone from a cDNA library in  $\lambda$ ZAPII which had been prepared with poly(A)<sup>+</sup>RNA from 20 days-old *A. thaliana* (cv. Landsberg) aerial part tissues. Under high stringent conditions ( $1 \times \text{SSC}$ , 60°C), five overlapping cDNA clones were obtained, and a cDNA clone (bA19) containing the longest insert was sequenced. DNA sequence analysis revealed that the bA19 clone had a 1654 bp insert that contains a single open reading frame (ORF) beginning at position 267. The ORF encodes a polypeptide consisting of 330 amino acids with calculated molecular weight of 36,684. A deduced amino acid sequence contains a bZIP domain in the N-terminal portion of the protein with 100% homology to the wheat HBP-1b (Fig. 1). Therefore, we conclude that the bA19 clone contains the DNA sequences encoding a HBP-1b counterpart in *A. thaliana*. A homology of the overall amino acid sequences

between the bA19-encoded protein and wheat HBP-1b (c-38) is 75%, whereas the bA19 protein shared 53% and 33% homology with TGA1a and OCSBF-1, respectively. Isolation of the HBP-1b counterpart from *Arabidopsis* would confer understanding of the real HBP-1b functions *in vivo* by using the T-DNA tagging and gene disruption techniques.

During the preparation of this manuscript, a cDNA clone (*PosF21*) encoding a bZIP-type DNA binding protein has been reported in *A. thaliana* (cv. Columbia) (12). There is no obvious homology in amino acid sequence between bA19 and *PosF21*, thus they are different transcription factors in *A. thaliana*.

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bA19  49  RRLAQNRREAAKRSRLRKKAYVQQLNSRLKLTQLQEQELQRRARQQGV  94
HBP-1b 49  RRLAQNRREAAKRSRLRKKAYVQQLNSRLKLTQLQEQELQRRARQQGi  94
TGA1a  77  RRLAQNRREAAKRSRLRKKAYVQQLNSRLKLTQLQEQELERARKQCGm  122
OCSBF-1 29  KRFLenREsARfSRLRKqghideLvqvearLqadnarVvaaraatar  74

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