

Plant Gene Register

Complementary DNA Sequence of a Low Temperature-Induced *Brassica napus* Gene with Homology to the *Arabidopsis thaliana* *kin1* Gene¹

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Low temperature triggers the acquisition of frost tolerance in plant species that are capable of developing cold hardiness. Recently, the transcription of specific genes was observed to be induced, at least in part, by low temperature in alfalfa (9), *Arabidopsis* (5, 7, 10), barley (2, 3), and wheat (6, 8) during cold acclimation. In wheat and *Arabidopsis*, the cold-induced mRNAs were shown to direct the translation of heat-stable polypeptides (6, 8); in rice, exposure to low temperatures resulted in the accumulation of *rab* (responsive to ABA) gene products (4). The mRNAs encoding these gene products appear rapidly when the plants are exposed to low temperature and, in the majority of cases, are also inducible by water stress or by the direct application of ABA (5, 7, 10). Although the exact function of the products of these cold-regulated genes has not been elucidated, evidence of similarity in deduced amino acid motifs and similar physicochemical properties with *rab* or *lea* (late embryogenic abundant) proteins suggests that they may play a role in protecting the cell from the cellular dehydration that accompanies extracellular freezing.

Differential screening of a λ gt10 library, constructed from mRNA isolated from leaves of the winter *Brassica napus*, cv Jet neuf, grown at 2°C, was used to obtain the cDNA of a cold-induced transcript. We report the cDNA and deduced amino acid sequences of BN28, a low-temperature-regulated mRNA (Fig. 1, Table I). The coding region nucleotide sequence is homologous (77%) to the cold and ABA-inducible *kin1* gene of *Arabidopsis thaliana* cloned and characterized by Kurkela and Franck (7) and also by Artus *et al.* (1). At the protein level, divergence of BN28 from ATkin1 resulted in mostly conservative substitutions in amino acid residues within the coding region. The function of BN28 is not yet

known. However, based on regions of deduced amino acid homology to flounder antifreeze protein, Kurkela and Franck (7) suggested that ATkin1 may possess antinucleation properties. This has not been confirmed experimentally.

LITERATURE CITED

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1      AAAAAACACAACAAAACCTCAATAAATAAACAAATGGCAGACAACAAGCAGAGCTTCCAA
BN28                                     M A D N K Q S F Q      9
Kin1                                     M S E T N K N* A F Q

61     GCCGGTCAAGCCTCTGGTCGTGCTGAGGAGAAGGGTAATGTGCTGATGGACAAGGTCAAG
BN28  A G Q A S G R A E E K G N V L M D K V K      29
Kin1  A G Q T A G K* A E E K S N V L L D K A* K

121    GATGCTGCTACCGCAGCTGGAGCGTCTGCGCAAACCGCGGGACAGAAGATAACGGAGGCG
BN28  D A A T A A G A S A Q T A G Q K I T E A      49
Kin1  D A A A G* A G G* G* A Q Q A G K* S V S* D* A

181    GCAGGGGGAGCCGTTAATCTCGTGAAGGAGAAGACCGGCATGAACAAGTAGCCCCATTGG
BN28  A G G A V N L V K E K T G M N K      65
Kin1  A A* G G* V N F V K D* K T G L N K

241    AAATAAAATTGGGAGTTATAGTTTCCCTTTTTAATGTTAATCGTTGTGGTTTTAAATAAA
301    AATTGGGTGTTCTAATTGATCTTCACCGTAGTTGTTGTTGTTGTTGTTTTAGTGTTTGA
361    AGTAATGTTTTCAACCTTTAGGTGTATGTCTTGATGTTTATGTACTTAGTTCCCCCTTAA
421    TGAACATCTGATTTAAGCTTCAAAAAAAAAAAAAAAAAAAAAA

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Figure 1. Nucleotide and deduced amino acid sequences of cDNA (BN28) from *B. napus*. The deduced amino acid sequence is compared to the coding region of the *kin1* gene isolated from *Arabidopsis* (7). Underlined amino acid residues and asterisks indicate homology between BN28 and *kin1* and conservative substitutions, respectively.

Table 1. Characteristics of the BN28 cDNA from Winter *B. napus*

Organism:

Winter *Brassica napus* cv Jet neuf leaves.

Gene:

cDNA with homology to coding region of *Arabidopsis thaliana kin1* gene (7).

Techniques:

cDNA library constructed from mRNA isolated from leaves of *B. napus* grown for 4 d at 2°C, 16/8 h light/dark at 250 $\mu\text{E m}^{-2} \text{s}^{-1}$. Isolation by differential screening of unamplified library with single-strand cDNA probes of mRNA isolated from leaves grown at 2 and 24°C. Complete sequence of forward and reverse strands.

Chromosome Location:

Unknown.

Method of Identification:

Sequence comparison to GenBank EMBL database; sequence identity to coding region of *A. thaliana kin1* gene (7).

Expression Characteristics:

Abundant mRNA of approximately 0.45 kilobases, induced by the exposure of whole plants to 2°C, with the transcript appearing within 6 h of exposure. Transcript disappears within 20 h of return of plants to 22°C. Inducible by foliar application of ABA to plants grown at 22°C and water stress at room temperature. The levels of expression, however, were much lower than those induced by cold treatment. Not induced by heat shock.

Codon Usage:

Strong bias for AAG(K) and GAG(E) in protein-coding region.

(G + C) Content:

40.7%.

Structural Features of Protein:

Deduced amino acid sequence of 65 residues with 34% (alanine + glycine) and absence of tyrosine, tryptophan, cysteine, and proline. Highly basic with a calculated isoelectric point of 10. Hydropathy plot is identical with that of *Arabidopsis kin1* (7).

Antibodies:

Not prepared in this laboratory.

GenBank/EMBL Accession No:

M81224.
