

# Nucleotide sequence of a cDNA encoding ribosomal acidic phosphoprotein P1 from *Dictyostelium discoideum*: identification of a novel carboxy-terminal sequence in 'A' proteins

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The eukaryotic 'A' ribosomal proteins are acidic proteins of about 13 kDa which are related to *Escherichia coli* L7/L12 (1), and have been found in the large subunit of all the ribosomes examined to date. They constitute a family of conserved proteins, having features which are unique among the ribosomal proteins. They are acidic, and contain a large number of alanine residues (2), as well as highly conserved carboxyl-ends. Interestingly, the sequence of the eight C-terminal amino acid residues is identical in all the species studied (3). The eukaryotic ribosomal acidic proteins are derived from independent genes, and were also designated P1 and P2 because they are phosphorylated. Like L7/L12, P1 and P2 exist as dimers at the large ribosomal subunit, and play an important role in the elongation step of protein synthesis (4).

We report the nucleotide sequence of a cDNA clone encoding the complete amino acid sequence of *Dictyostelium discoideum* ribosomal acidic protein P1. The cDNA was isolated from a lambda gt11 expression library, constructed in this vector by insertion of cDNAs obtained from *Dictyostelium discoideum* (strain AX- 3) 4 hours-starved cells. The screening of this library was performed using polyclonal antibodies, raised against *Dictyostelium* ribosomal proteins P1 and P2 which can be selectively released from 80S ribosomal particles with salt/ethanol washes. The insert found in a lambda positive clone was subcloned into pUC plasmid and both strands were sequenced by the chain termination method. The cDNA was 442 nucleotides in length and contained the translation start and stop codons. It also has a 5' noncoding sequence of 12 bases, an open reading frame of 339, and a 3' noncoding sequence of 91 which includes a polyadenylation recognition sequence- AATAAA- located at position 418-423, as well as a poly(A) stretch. The open reading frame encoded a protein of 113 amino acid residues.

The primary structure inferred from the cDNA sequence corresponds to a negatively charged protein that includes sequential stretches of hydrophobic and hydrophilic residues, which was identified as ribosomal protein P1 by its homology with other equivalent proteins reported previously. *Dictyostelium* P1 has 53% homology with human P1 (3) and 37% with yeast ribosomal acidic protein L44' (5), as determined in a pairwise comparison of identical residues. Furthermore, the molecular weight of *Dictyostelium* P1 (11,694) is very similar to these same proteins.

The amino acid composition reveals that *Dictyostelium* P1, like the other P proteins, contains a large number of alanyl residues,

22, which represents about 20% of the amino acid contents. 14 of these alanines are clustered between positions 70 and 89. P1 has an excess of acidic residues (7 aspartyl and 11 glutamyl) over basic ones (10 lysyl, 1 arginyl, and 1 histidyl), although this excess has been found to be much greater in P1 proteins from other origins (3, 5). The decrease in negative charge might respond mainly to a loss of a portion of a cluster of glutamyl residues which is usually located near the C-terminus of P1 proteins (3, 5). Noteworthy, *Dictyostelium* P1 contains 62 hydrophobic residues, most of them located at a central domain of the protein.

The most interesting feature found in the *Dictyostelium* P1 sequence resides in the carboxy-terminal region. It has a methionine residue (position 109) in a location where all other P1 proteins examined contain a phenylalanine (see the alignment of C-terminal sequences shown below). This is the first amino acid change reported to date at this position of the conserved carboxyl-end sequence of A-proteins. Interestingly, we have also found the same amino acid change in the sequence of *Dictyostelium* protein P1 and in P0, (see accompanying papers). Therefore the sequence DMGMGLFD constitutes a novel 'A' carboxyl-terminal region which is a specific feature of A-type ribosomal proteins from *Dictyostelium*. This finding might be of interest in studies on the evolution of ribosomal 'A'-protein sequences.

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*Dictyostelium* P1  
P1 from other eukaryotes

SDDDMGMGLFD  
SDDDMGFGLFD

Alignment of P1 carboxy-terminal sequences