Cloning and characterisation of a yeast homolog of the mammalian ribosomal protein L9

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We have cloned and sequenced a Saccharomyces cerevisiae homolog of the mammalian ribosomal protein L9. The corresponding ORF, which is uninterrupted by introns, is 573 bp, encoding a protein of 191 amino acids with a calculated MW of 21.6 kDa. The gene, which we have named YL9A (Yeast L9A), is located on chromosome VII, immediately downstream of the ARO2 gene, encoding chorismate synthase (1). We have named this gene YL9A in order to distinguish it from a transcribed and presumably functional homolog, YL9B, which we have located by OFAGE and blot hybridisation on chromosome XVI. Gene disruption experiments demonstrate that the YL9A gene, although expressed as a 0.8 kb transcript, is non-essential, probably due to the presence of the homolog.

In common with many yeast ribosomal protein genes the YL9A upstream region contains large poly (dAT) stretches and a putative RAP1 binding site centered at position −305 with respect to the translation start (2). In conjunction with two putative TATA elements located at positions −85 and −93, these elements may be at least partially responsible for the production of the single mapped YL9A transcript species, which initiates at position −19.

The sequence analysis and the use of directional probes in Northern analysis demonstrate that the YL9A and ARO2 gene are convergently transcribed with only 224 bp between the stop codons of the two genes (Figure 1). Transcript end point analyses reveal a single 3' end of YL9A mRNA at position +671, showing that the 3' ends of the ARO2 (1) and YL9A mRNAs virtually abut, resulting in a mere 11 bp of intergenic space between the two transcribed regions.

The YL9A ORF exhibits a typically marked codon bias as for other yeast ribosomal protein genes (3). The predicted amino acid sequence of YL9 shows a 49% identity to the rat ribosomal protein L9 (RL9; 4) and the N-terminus is virtually identical (allowing for peptide sequencing ambiguities) to a partial 40 amino acid N-terminal peptide sequence of the yeast ribosomal protein YL11 (5). Both these proteins are components of the large ribosomal subunits of their respective organisms. Furthermore, there exists significant identity to equivalent prokaryotic ribosomal proteins, namely the L6 proteins of Methanococcus vannielii and Escherchia coli (33% and 26% identity, respectively; 6, 7; Figure 2).

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REFERENCES


Figure 1. Position of YL9A ORF relative to ARO2 ORF. A 2.0 kbp region, ranging from the rightmost EcoRV site to the rightmost SalI site, containing the YL9A gene was sequenced previously (1). An adjoining 1.3 kbp region, ranging from the SalI site to the rightmost EcoRV site, containing the YL9A gene was sequenced in this work. The boxes represent the YL9A and ARO2 ORFs as determined by the sequence analysis with the YL9A translation start codon at the indicated position +1. The transcripts of both genes are indicated by black arrows. Transcript end points of the YL9A mRNA are relative to the translation start codon.

Figure 2. Alignment of the amino acid sequences of yeast L9 (YL9), rat L9 (RL9; 3), M. vannielii L6 (ML6; 5) and E. coli L6 (EL6; 6) ribosomal proteins. A consensus sequence for the L9-L6 protein moiety is shown, representing residues conserved throughout at least 3 of the examined proteins. Residues conserved throughout all 4 proteins are in bold type. Positions where conservative amino acid changes occur are indicated by asterisks. The alignment was performed using the GCG program LINEUP.