The *Saccharomyces cerevisiae* RPB4 gene is tightly linked to the TIF2 gene

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A *S. cerevisiae* genomic expression library, constructed in λ gt11 (1), was screened using polyclonal antibodies raised against poly(A) polymerase isolated from rat hepatoma cells (2). The 3.8 kb insert from clone λ AS109 obtained by this procedure was sequenced in its entirety. One long open reading frame (ORF) and portions of two other ORFs were identified. The partial ORFs were found to correspond to the genes encoding translation initiation factor eIF-4A (TIF2) (3) and the fourth largest subunit of RNA polymerase II (RPB4) (4). The other ORF, designated YUR1, did not resemble any other sequences in the swiss-protein, EMBL, or genbank databases. Figure 1 shows the arrangement of these genes with respect to one another. The nucleotide sequence of the region between TIF2 and RPB4 has been assigned accession number X58099.

The RPB4 gene was fused to the β -galactosidase gene in the λ gt11 vector in λ AS109, suggesting that RPB4 encodes the antigen recognized by the anti-poly(A) polymerase antibodies. This result was confirmed on immunoblots by demonstrating that β -galactosidase protein fusion constructions with RPB4 but not YUR1 nor TIF2 were bound by the antibodies after expression in *E. coli* (A.B.S. data not shown). However in yeast strains in which the RPB4 gene is partially deleted by gene transplacement, the major 48 kDa antigen recognized by the anti-poly(A) polymerase antibodies remains intact (P.K.F. unpublished observations). These results suggest that RNA polymerase II and poly(A) polymerase may share common antigenic determinants.

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REFERENCES

1. Huynh,T.V., Young,R.A. and Davis,R.W.(1985) In Glover,D.M. (ed.) *DNA Cloning. A Practical Approach*. IRL Press, Oxford, Vol. I., pp. 49–78.
2. Stetler,D.A. and Jacob,S.T. (1984) *J. Biol. Chem.* **259**, 7239–7244.
3. Linder,P. and Slonimski,P. (1988) *Nuc. Acids Res.* **16**, 10359.
4. Woychik,N.A. and Young,R.A. (1989) *Mol. Cell. Biol.* **9**, 2854–2859.

Figure 1. Arrangement of the TIF2, YUR1 and RPB4 genes. Open reading frames for each of the genes are indicated by arrows.

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