Drosophila AP3, a presumptive DNA repair protein, is homologous to human ribosomal associated protein PO

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A *Drosophila* gene (AP3), previously isolated using antibody to a human apurinic/apyrimidinic (AP) endonuclease (1, 8) was used to screen a HeLa cDNA library for its human homolog. One human gene was isolated that encodes a protein that is 66% identical and 79% similar at the amino acid level to the *Drosophila* AP3 gene, and is identical to the human protein PO (2). The PO gene was cloned using antibodies to systemic lupus erythematosus, and encodes a presumptive ribosomal associated protein (2). Both the *Drosophila* and human proteins are the same length, 317 amino acids, and have the characteristic acidic carboxy tail seen in PO proteins from yeast (AO;3), rat (4) mouse (5) and human (2) (Figure 1). The *Drosophila* protein also contains a 23 amino acid region similar to a region in the *E.coli* L10 protein that has been implicated in the binding of 28S rRNA molecules (3) (Figure 2). AP3 shares the greatest homology at the amino acid level to human PO (66%—no gaps), rat PO (66%—no gaps) and mouse PO (66%—no gaps), but is less homologous to the yeast AO (52%—two gaps). The *Drosophila* protein is approximately 34,000 Daltons and has a large number of alanines (37 or 12% of the 317 amino acids) which are located in the carboxy terminus of the protein, similar to PO proteins (2, 3, 4, 5). Although the eukaryotic PO protein is referred to as a member of the acidic protein family, presumably due to its highly acidic tail (Figure 1), the *Drosophila* protein, on the other hand, has three more basic than acidic amino acids for a net positive charge. The *Drosophila* protein also contains well-defined Casein Kinase I (CKI) and II (CKII) phosphorylation sites (CKI = XEXXSX and CKII= XSXXEX, 6) which may be involved in regulating the protein activity (Figure 3). Potential phosphorylation sites are also found in the yeast protein (CKI), but a consensus site for either CKI or CKII is absent from the human, rat and mouse PO proteins (Figure 3). Although not presented here, antibodies produced to fusion proteins of the *Drosophila* AP3 gene detected a single protein in the nuclear matrix, as well as one associated with ribosomes in the cytoplasm of *Drosophila* extracts. Further evidence pointing to the unusual nature of these genes is that PO is inducible in human cell lines by bifunctional alykylating agents used in clinical cancer treatments that also cause DNA intra- and interstrand cross-links (manuscript in preparation).

The cloning of a *Drosophila* homolog to the human PO gene using antibody to a purified human AP endonuclease DNA repair gene (1, 8) is intriguing. Whether this protein has a dual function as a DNA repair protein and also as a ribosomal associated protein remains to be clarified. The highly acidic carboxy tail seen in the human PO and *Drosophila* AP3 proteins has also been seen in the yeast *Rad* genes, particularly *Rad3* and *Rad6*, which are involved in DNA repair in yeast (9).

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Figure 1. Acidic carboxy-terminal amino acids.

Drosophila	N(302)–SESEEEDDDMGFGLFD–COOH
Human PO	N(302)–EESEESDEDMGFGLFD–COOH
Rat PO	N(302)–EESEESDEDMGFGLFD–COOH
Mouse PO	N(302)–EESEESDEDMGFGLFD–COOH
Yeast AO	N(297)–EEEEESDDDMGFGLFD–COOH

Figure 2. Potential 28S rRNA interaction region.

Drosophila	N(44)–RTSLRGLAVVLMGKNTMMRKAIR
Human PO	N(44)–RMSLRGKAVVLMGKNTMMRKAIR
Rat PO	N(44)–RMSLRGKAVVLMGKNTMMRKAIR
Mouse PO	N(44)–RMSLRGKAVVLMGKNTMMRKAIR
Yeast PO	N(42)–RKELRGRAVVLMGKNTMVRRAIR

Figure 3. Casein Kinase I (XESSSX) or II (XSXXEX) sites; the potential phosphorylated S is underlined.

Drosophila	ESESEEEDDD (Casein kinase I and II site)
Human PO	EESEESDED (no CK I or II consensus site)
Rat PO	EESEESDED (no CK I or II consensus site)
Mouse PO	EESEESDED (no CK I or II consensus site)
Yeast AO	EEEEESDDD (CK I site)