Cytosolic aminopeptidase P (APP-1): a possible role in meiotic progression in Caenorhabditis elegans.

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Cytosolic Aminopeptidase P (APP-1): a possible role in meiotic progression in Caenorhabditis elegans. **Richard Elwyn Isaac**[1], Hannah Craig[1], Enrique Martinez-Perez[2], Darren Brooks[3]. 1) Faculty of Biological Sciences, Miall Building, University of Leeds, Leeds LS2 9JT, UK; 2) Department of Molecular Biology and Biotechnology, University of Sheffield, Firth Court, Western Bank, Sheffield S10 2TN, UK; 3) Biomedical Sciences Research Institute, School of Environment and Life Sciences, University of Salford, Salford M5 4WT.

Aminopeptidase P (APP) is a metallopeptidase that specifically removes amino acids from the N-terminus of peptides with a penultimate N-terminal proline residue. The cyclic structure of the proline side chain places a conformational restraint on peptide bonds in its vicinity, which therefore tend to be resistant to hydrolysis by non-specialist peptidases. APP is one of the few peptidases capable of cleaving peptide bonds involving a proline residue and therefore has an important role in protein/ peptide processing and catabolism. In humans, at least two APP genes exist, coding for a soluble cytosolic enzyme (APP-1) and a secreted membrane-bound form (APP-2). APP-2 has been the subject of much research due to its role in the regulation of blood pressure and potential as a drug target in the treatment of hypertension, but there is little information available on the biological role of APP-1. We have previously shown that a C. elegans APP-1::GFP fusion protein is strongly expressed as a cytosolic protein in the intestinal cells of late embryos, larvae and adult worms. Immunocytochemistry using antibodies specific for C. elegans APP-1 has confirmed the initial promoter-GFP expression pattern and has revealed additional strong expression of APP-1 in the female germline. We have obtained a C. elegans app-1 mutant (tm1715), generously provided by the Japanese National Bioresource Project, that has a 452 bp deletion spanning most of exon 2 and part of exon 3. We have shown that tm1715 is a null mutant by using a specific assay for APP aminopeptidase activity and by western blot analysis. Phenotypic analysis for tm1715 revealed a higher (18-fold) than normal number of males (XO) generated on self-fertilisation of the adult hermaphrodite (XX) and a 50 percent reduction in brood size. These phenotypes are similar to those seen in a sub-set of him mutants (high incidence of males) that are defective in segregation of autosomes as well as the X chromosome during meiosis. The APP-1 antibody gives diffuse staining inside meiotic nuclei of the female gonad and staining with anti-RAD-51 antibodies of app-1 mutants suggests an altered pattern in the repair of DNA double-strand breaks (DSBs) generated during meiotic prophase. These preliminary results suggest an important biological role for APP-1 in meiotic progression in C. elegans.