group, are fully conserved in all of the non-LTR retrotransposons. Further support for this similarity has been determined from crystallographic analyses, which show that the fold of exo III is similar to that of DNase I, despite having less than 20% overall sequence similarity[2].

The presence of the exo III active residues and the conserved amino acids involved in DNase I acid–base catalysis, in similar regions of the non-site-specific non-LTR retrotransposons, provides an even stronger argument in favor of the potential nuclease activity of these elements. Within this framework, the potential endonuclease activity could be responsible for generating the 3′-OH sites necessary as primers for its reverse transcription (first step in the integration mechanism). This would render the existence of nicks in the DNA necessary for the integration of the non-LTR elements. As the non-LTR retrotransposons share DNase I conserved domains, it is also attractive to think that these elements might recognize sequence-dependent structural variations similar to those recognized by DNase I (Ref. 8) and generate the 3′-OH needed for transposition.

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Methionine aminopeptidase-1: the *MAP* of the mitochondrion?

Methionine aminopeptidases (MetAPs or proteins encoded by *MAP* genes) are ubiquitous enzymes that cleave the amino-terminal methionine from many newly translated polypeptides. In eubacteria, a single MetAP is generally sufficient to fulfil this role, but eukaryotic genomes contain two distantly related enzymes, which, at least in yeast, are both required for normal growth[1,2]. One of these proteins, MetAP-1, closely resembles the enzymes known from eubacteria, but the other, MetAP-2, is only distantly related to other MetAPs (Refs 1, 2).

Recently, putative *MAP* genes have been identified from two archaebacteria, one a fragment from *Methanothermus fervidus*[3], and the other a full length gene from the distantly related thermophile, *Sulfolobus solfataricus* (C. W. Sensen *et al.*, unpublished). The predicted protein products of these archaebacterial genes have all the hallmarks of other MetAPs, including the conservation of six residues implicated in the coordination of a cobalt ion co-factor, and the adjacent motifs (Fig. 1). Of all other sequences, the

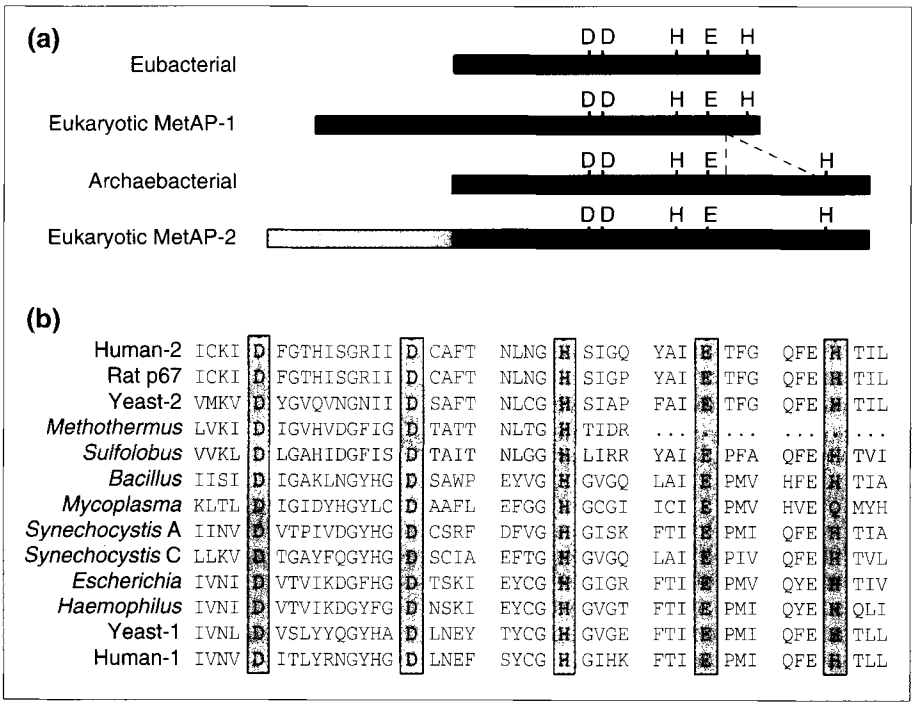

Figure 1

(a) Schematic of methionine aminopeptidase proteins. The core enzyme is shown in blue, the acidic amino-terminal domain of eukaryotic MetAP-2 is in yellow, the MetAP-1 amino-terminal extension is in red and the 64-amino acid insertion shared between archaebacteria and eukaryotic MetAP-2 is in green. The identity between the insertions of *Sulfolobus* and eukaryotes is 58–60%, as compared with a 65.5–70% identity over the rest of the alignable core enzyme (based on 145 positions used in the phylogenetic analysis). Five highly conserved cobalt co-ordinating residues (D, H and E) are also indicated. **(b)** Amino acid sequences surrounding the five highly conserved cobalt co-ordinating residues (boxed), which appear in the core enzyme at positions 97, 108, 171, 204 and 236, respectively, in the *E. coli* protein[1]. We include the rat p67 translation factor[5] in the MetAP-2 class because this enzyme is also likely to be an aminopeptidase: it shares 95% identity with human MetAP-2.

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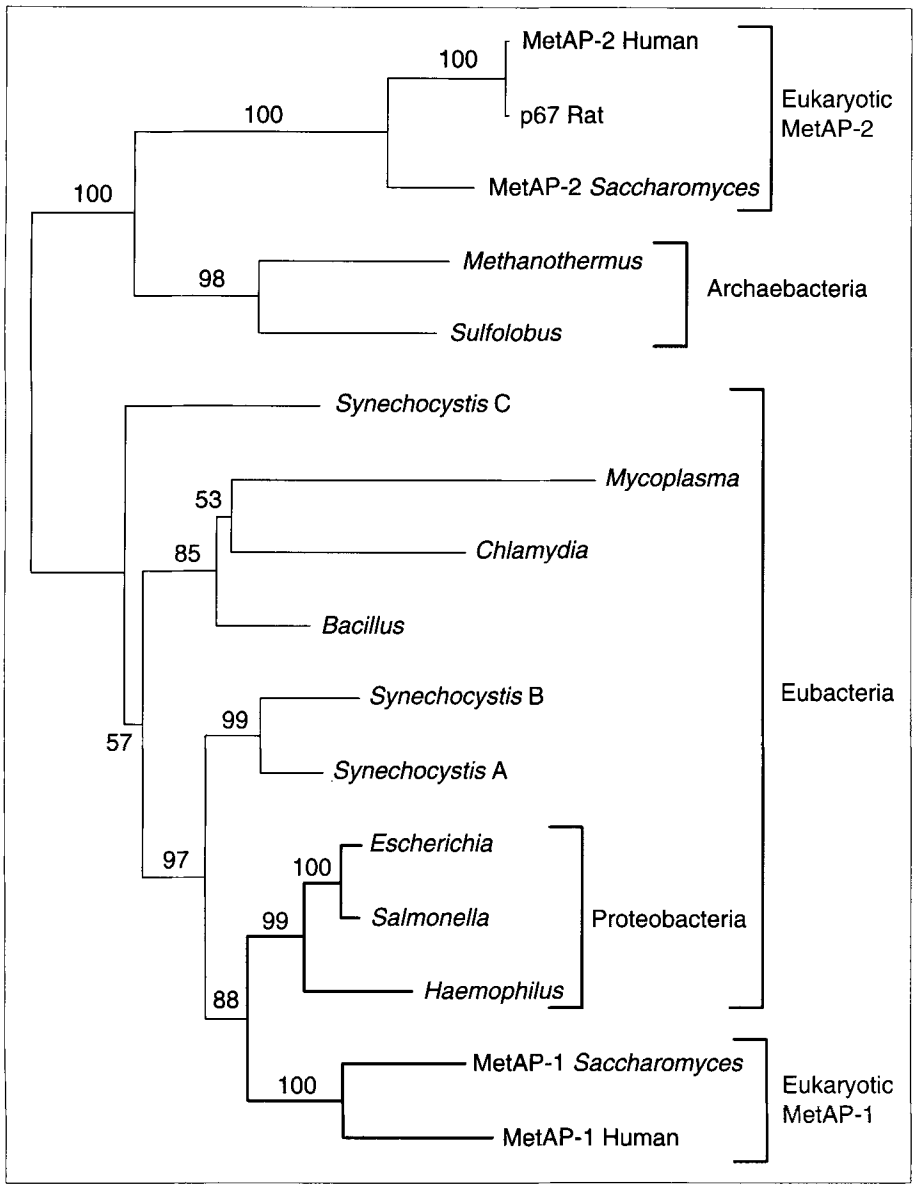

Figure 2

Phylogenetic tree of methionine aminopeptidase amino acid sequences. Corrected distances were calculated[6] based on the PAM 250 substitution matrix for 145 alignable sites. Neighbour-joining analysis of these distances resulted in the tree shown. Statistical support for individual nodes is shown as a fraction of 100 bootstrap replicates. Unweighted parsimony analyses[7] on the same dataset yielded an identical topology. Branches in bold represent the clade (supported in 88% of bootstrap replicates) containing the proteobacteria and eukaryotic MetAP-1.

archaebacterial genes bear a particularly close resemblance to eukaryotic *MAP2*, with which *Sulfolobus* also shares a 64-codon insertion (the determined *Methanothermus* sequence is truncated before this point). The similarity between archaebacterial *MAP* and eukaryotic *MAP2* genes suggests that the eukaryotic MetAP-1 and MetAP-2 enzymes must have diverged before the divergence of archaebacteria and eukaryotes and could not share a common ancestral sequence in the eukaryotic genome.

Indeed, phylogenetic analyses of all known *MAP* genes bears this out and reveals much more. Figure 2 shows the tree of methionine aminopeptidases, where it can be seen that the eukaryotic MetAP-2 and archaebacterial sequences form a clade, while the remainder branch together (eubacterial and eukaryotic MetAP-1 sequences). With the exception of the eukaryotic MetAP-1 genes, this tree is not extraordinary in that it is composed of three distinct domains, with the archaebacterial sequence bearing a greater resemblance to those of eukaryotes than eubacteria. However, the evolutionary history of eukaryotic MetAP-1 sequences appears to differ from conventional cytosolic proteins; they are, instead, specifically related to homologues from proteobacteria. While this might be an unusual relationship for a cytosolic protein, it is precisely the pattern that is expected for a gene derived from the mitochondrion, because this organelle is thought to have arisen from an α-proteobacterial endosymbiont[4].

MAP1 genes reside in the nucleus, as do most mitochondrion-specific genes, but the most straightforward explanation for their strongly supported relationship with proteobacteria is that they were ultimately derived from the mitochondrial symbiont. This naturally suggests that MetAP-1 also currently functions in the mitochondrion, but aside from the fact that both known MetAP-1 sequences have amino-terminal extensions, which could be mitochondrial-targeting peptides (the red box in Fig. 1), there is no direct evidence that MetAP-1 proteins are targeted to the organelle. Moreover, although knocking out both genes is lethal in yeast, eliminating each enzyme individually leads only to slow growth[1]. That one enzyme partly mitigates the effects of the other's loss argues that if there is a partitioning of enzymes to the mitochondrion and cytosol, it is not very strict, and might even suggest that both proteins function in the cytosol despite the clearly symbiotic provenance of *MAP1*.

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