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MicroCorrespondence

An Archaebacterial eIF-1A: new grist for the mill

Sir,

The archaebacteria (or Archaea) were shown, by molecular phylogenetecists, to be the closest prokaryotic relatives of the nuclear component of eukaryotic cells (Iwabe *et al.*, 1989, *Proc Natl Acad Sci USA* **86:** 9355–9359). Many aspects of their molecular biology have also been found to support this conclusion, by demonstrating close similarity to the corresponding eukaryotic systems. There are several good examples of this, including the proteasome, small subunit ribosomal RNA processing, and, most convincingly, the transcription complex (Pühler *et al.*, 1994, *Syst App Microbiol* **16:** 734–741; Duravic and Dennis, 1994, *Mol Microbiol* **13:** 229–242; reviewed in Keeling *et al.*, 1994, *Curr Op Genet Dev* **4:** 816–822).

In other characteristics, the archaebacteria bear a closer resemblance to their eubacterial cousins: genome and gene structure are shared, as well as many features of the translational process. It is well known, for instance, that archaebacterial mRNAs are often polycistronic, uncapped and do not include a long poly-A tail as do eukaryotic transcripts. Moreover, ribosome binding to mRNA is thought to be effected by a Shine–Dalgarno-like interaction between the 16S ribosomal RNA and mRNA (Brown and Daniels, 1989, *CRC Crit Rev Microbiol* **16:** 287–335) — all features which suggest that archaebacterial translational initiation is possibly similar to that in eubacteria and unlike that in eukaryotes.

However, what little direct evidence there is bearing on the nature of the archaebacterial translation-initiation complex is somewhat contradictory. The single putative initiation factor that has been identified in the archaebacteria is actually homologous to the eukaryotic initiation factor eIF-5A (Bartig *et al.*, 1992, *Eur J Biochem* **204:** 751–758), for which no eubacterial equivalent is known. Moreover, the role of eIF-5A in eukaryotic translation initiation has recently been seriously questioned (Kang and Hershey, 1994, *J Biol Chem* **269:** 3934–3940), and no functional analysis has been performed on the archaebacterial homologue. Nevertheless, the possibility that archaebacterial translation involves both eukaryotic and eubacterial elements is a tempting speculation, and has prompted a search for other, as yet unidentified, archaebacterial translation-initiation proteins.

Using the BLASTP search strategy (Altschul *et al.*, 1990, *J Mol Biol 215:* 403–410) we compared a representative sequence of each eubacterial and eukaryotic initiation factor, for which a sequence is available, to the existing sequence database. From this search, two archaebacterial reading frames were unambiguously identified: the aforementioned *Sulfolobus acidocaldarious* eIF-5A homologue, and a previously unidentified open reading frame (ORF), ORF125 (Fig. 1), located downstream of the *Thermoplasma acidophilum* RNA polymerase operon (Klenk *et al.*, 1992, *Nucl Acids Res* **20:** 5226). The inferred polypeptide sequence of ORF125 is approximately 30% identical and 50% similar over the aligned region to eIF-1A (also called eIF-4C), which is a highly conserved initiation factor recently sequenced from animals, plants (Dever *et al.*, 1994, *J Biol Chem* **269:** 3212–3218), and fungi (GenBank accession number U11585). The physical nature of the *Thermoplasma* eIF-1A is somewhat different to that of its eukaryotic counterparts, which have a dipole nature characterized by an acidic C-terminal tail, and a highly basic N-terminus. The archaebacterial sequence has neither of these features, and it is actually missing the 22–31 amino acids corresponding to the C-terminal

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        1                                                          60
Human   .MPKNKGKGG KNRRRGKNEN ESEKRE.LVF KEDGQEYAQV IKMLGNGRLE AMCFDGVKRL
Wheat   ..PKNKGKGG KNRKRGKNEA DDDKRE.LVF KEDGQEYAQV TRMLGNGRCE AICVDGTKRL
Yeast   .MGKKNTKGG KKGRRGKNDS DGPKRE.LIY KEEGQEYAQI TKMLGNGRVE ASCFDGNKRM
           :    : •  : •:    :   •  •   •: •: :  •  •• •  •:  ::• ••  •
Therm   MSEDDVDNSV KDFESGEENE ESIGRVILPN KKKGEMFGIV EKMEGASRLS VMCEDGYTRN

        61                                                        120
Human   CHIRGKLRKK VWINTSDIIL VGLRDYQDNK ADVILKYNAD EARSLKAYGE LPEHAKI.NE
Wheat   CHIRGKMHKK VWIAAGDIVL VGLRDYQDDK ADVILKYMND EARLLKAYGE LPDTVRL.NE
Yeast   AHIRGKLRKK VWMGQGDIIL VSLRDFQDDQ CDVVHKYNLD EARTLKNQGE LPENAKI.NE
        • • •::••: :••   •::: •   ::• :• •••: :    :•  •    ••• : • ••
Therm   ARIPGRMRKR MWIREKDLVI VKPWEFQPEK ADVVYRNTKT QASYLSRNHM LPEVIDISNE

        121                                 156
Human   TDTFGPGDDD EIQF..... ...DDIGDDD EDIDDI
Wheat   GVDVDGPEEG EGDS..... ...DYIQFED EDIDKI
Yeast   TDNFGFESDE DVNFEFGNAD EDDEEGEDEE LDIDDI

Therm   VMKDR..... ..... ..... .....
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Fig. 1. Multiple alignment of eIF-1A and *Thermoplasma* ORF125. Amino acid identity and similarity between the *Thermoplasma* sequence and the consensus of eukaryotic sequences are specified by dots and colons, respectively. The similarity extends the entire length of the polypeptide except that the archaebacterial eIF-1a lacks the acidic C-terminal tail.

tail; while these dissimilarities may be of some significance to the function of the protein *in vivo*, the divergence time between the sequences and the thermoacidophilic nature of *Thermoplasma* may also account for some differences in its properties.

In eukaryotes, eIF-1A is thought to drive the dissociation of the 80S ribosome by holding the 40S subunit in its monomeric form, and in doing so increases the efficiency of the interaction between the 40S subunit, the mRNA and the ternary complex (Thomas *et al.*, 1980, *Eur J Biochem* **107:** 39–45). This role is a general requirement of translation initiation, and is fulfilled in eubacteria by a non-homologous factor, IF-1. If this *Thermoplasma* homologue of eIF-1A does play a role in translation initiation (in which case we suggest calling it aIF-1A), its presence in the archaebacteria raises the interesting possibility that the initiation complex may be more like that of eukaryotes despite the apparent similarities to that of eubacteria

Unfortunately, there is insufficient information at present to even guess what factors are important in archaebacterial translation initiation. Hopefully, when more is known, it will hold as many interesting surprises as other, better studied, archaebacterial molecular processes.

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