Poly-tRNA Structure in the Bacilluis Subtilis rrnB Operon is a Relic of an Early Peptide-Synthesizing Ribozyme Co-Ancestral to the E. coli trpE Gene and the Exon 2 of the Chicken Triose-Phosphate Isomerase Gene

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Poly-tRNA theory proposed that the tandemly arranged 16 tRNAs (trrnD-type-poly-tRNA) in the RNA transcript from B.subtilis (BSu) trrnD operon is most likely a relic of an early peptidesynthesizing RNA molecule (Ohnishi, Endocytobiology V, 407-14, '93; Genome Informatics Workshop IV, 325-31, '93; Origins of Life, 24: 191-92, '94; Ohnishi & Yanagawa, Endocytobiology VI, in press). Another poly-tRNA structure (comprizing 21 tRNAs) in the BSu rrnB operon was analyzed from a viewpoint of poly-tRNA theory. A hypothetical 21-amino acid(aa)-long "rrnB-peptide" was considered, whose as sequence is the order of the as specificities in the rrnB-poly-tRNA. A 63-base "rrnB-mRNA" was hypothesized where the k-th triplet is complementary to the anticodon triplet of the k-th tRNA in the poly-tRNA structure (k = 1, 2, ..., 21) (See Fig.1). Homology searches from PIR and GenBank DNA Databases revealed that (i) rrnB-peptide is homologous to aa's 268-290 of yeast glyceraldehyde dehydrogenase (GAP DH) and aa's 27-46 of E.coli trpE gene product, (ii) rrnB-mRNA is homologous to tRNASer (54.2% match) in the rrnB poly-tRNA, and (iii) the tRNAAsn-tRNASertRNAGlu region of the rrnB operon is homologous to the aa sequence-encodingtrpE DNA region (base match = 48% in 252 bases, Pnuc = 1.0 X 10-15), to GAPDH-encoding gene segment, to adenylate kinase gene, and to the exon 2 (and its flanks) of the chicken triose-phosphate isomerase (TIM) gene, as shown in Fig. 1. Thus the trpE mRNA region evolved from a rrnB-type poly-tRNA, meaning that an ancient tRNASer or its close homologue helped the rrnB-poly-tRNA make a rrnBpeptide, by interacting with the anticodon triplets inrrnB-type poly-tRNA. This interaction selected complementarity-generating base-changes in both tRNASer-like presumptive rrnB-mRNA and the 21 presumptive anticodon triplets. By this selection, base sequence complementarity between mRNA and triple t anticodons had been acquired, resulting in the emergence of early mRNAs. The alignment in Fig. 1 strongly suggests that the protein module structure encoded in the TIM exon 2 had been built up throuout early evolution, depending on the tRNA-module structure of the early peptide-synthesizing poly-tRNA ribozyme.

1 *M Q T Q K P T L E L L T* C E G A Y R D N P T A L F H 26 /*ATG-CAAACACAAAAACCGACTCTCGAACTGCTAACC*TGCGAAGGC--GCATATCG--CGACAATCCCA---CC-GCGCTTTTTCAC trpE (EC) vs_rrnB (+) *rrnB* (BSu) <=== 5'-end of tRNA-Asn 3'-end of tRNA-Asn ===> VTKL G *rrnB* peptide *rrnB*-mRNA RPA MISMDFHG 21 T 1 GUAACAAAACUG-GGCUUAC----GUCCAGCAA-UGAUGUCAAUGGACUUCCACGGA-AUC---AACAGCGAA 63 27 Q L C G D R P A T L L L E S A D I ----cagttgtgtggggatc----gtccggcaa-cgctgctgctgga-atccgcagatatc-п 46 s *trpE* (EC) vs rrnB (+) *rrnB* (BSu) -GĂCAĞCAÂA <=== 5'-end of tRNA-Ser
L K G V L G Y</pre> D 37 L K G V L G Y T E D A V V S S D F L G X S N S S /gttgaag-ggt----gtcttg-gg-ttacactgaagacgctgttgtc-tcctctgacttcttgggtgantct-aactcttcc 267 290 GAPDH (yeast) vs rrnÉ (+) ++ ++ ++ + + + P S I Y L D F A t Q R 54 -CCTTCAA-T-CTA--CCTTGA-TTTTGCC---CGCCAG-AAG TIM (exon 2) vs GAPDH(o) 00 00 000 00 00 00 0 0 0 000 00 0 8/A P G I∕A P G A G K G – T Q A Q F I – M E K Y G I P Q I S / /gctccg-ggc-gcggggaaa-ggg----act-caggctcagttcatc---atggagaaatatggt-attccgcaaatctcc/ QAQFI-MEKYG S / 30 adk (FC) 92/Q A D A - M K E A G I N V D / /CAGGCAGAC--GCC---ATGAAAGAAGCGGGC-ATC---AATGTTGAT/ vs rrnB (+) N V D /105 adk (EC) vs rrnB (+) ++ ++ + + ++++ + 47 trpE (EC) vs rrnB (+) GAPDH vs rrnB (+) TIM (chk) vs GAPDH(o) ooo ooooo 3 L S G N G E A L L A L L D N A L P A 90 ACTITCCGGCAACGGCGA-AGCCC----TCCTGG-CAC-TACTGGATAACGCC--CTGCCT-GCG/ 48.4%(122/252) ++ ++ ++++++ ++ ++ ++ ++ ++ +++ +++ Pnuc = 1.00 X ACCGCCCTTTCACGGCGGTAACACGGGTTCGAAT-CCCGTACGGG-TCAtcccagaagccttgca/ 3'-end of tRNA-Glu ===> 73 trpE (EC) vs rrnB (+) Pnuc = 1.00 X 10-15

Fig. 1. Alignment of *E. coli* (EC) *trpE* gene, chicken (chk) triose-phosphate isomerase (TIM) gene segment (exon 2 and its flanks), yeast GAPDH gene, and EC adenylate kinase gene (*adk*) against the spacer-tRNAAsn-spacer-tRNASer-spacer-tRNAGIu region of *Bacillus subtilis* (BSu) *rrnB* operon. Bases in Spacers and intron regions are written in lower-case letters. Tentatively aligned regions are italisiz-ed. Base-matches to *rrnB*-mRNA and amino acid matches among aligned peptides are boxed (). Probabilities that m or more of the n aligned base-posisions are occupied by identical bases by chance, Pnuc(m,n), are given. Data are from; rrnB = Green,C.J. & al.,Gene 37:261-266,1985;trpE = Nicholas,B.P. & al.,J.mol.Biol.146:45-54;GAPDH = Halland,J.P. & Holland,M.,J.biol.Chem. 255:2596-2606,1989;TIM = Straus,D. & Gilbert,W.,Mol.cell.Biol. 5:3497-3506; adk = Brune,M. & al.,Nuc.Acids Res. 13:7139-51,1985.