Nucleotide sequence of rat elongation factor-1 α cDNA

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Elongation factor-1, which is composed of four subunits; α , β , γ and δ , participates in protein synthesis by binding aminoacyl-tRNA to ribosomes under hydrolysis of GTP. EF-1 α is the protein mainly responsible for the binding of GTP and aminoacyl-tRNA and is functionally equivalent to prokaryotic EF-Tu.

In this study, full length rat EF-1 α cDNA clone was isolated from embryonic brain λ Zap cDNA library as one of ubiquitously expressed clones by differential hybridization screening between adult brain cDNA probe and embryonic brain cDNA probe. The pBluescript phagemid was prepared from phage clone by in vivo excision and nucleotide sequences were determined by the dideoxy method. Rat EF-1 α cDNA clone was consisted of 1714 bases, which contained single open reading frame encoding 462 amino acids with a calculated molecular weight of 50 kDa. The nucleotide sequence around the ATG initiator codon is compatible with the Kozak's consensus sequence (ACCATGG) including A at position -3 and G at position +4 (1). Computer-assisted comparison with known nucleotide and amino acid sequences from other species revealed that the nucleotide sequence of rat EF-1 α had 92% similarity to human EF-1 α (2), 85% to Xenopus *laevis* EF-1 α (3), 80% to rat statin (4), and 74% to Artemia EF-1 α (5). The amino acid sequence showed 99% similarity to human EF-1 α (2), 95% to rat statin (4), 83% to Artemia EF-1 α (5), 80% to yeast EF-1 α (6), and 76% to tomato EF-1 α (7). Recently, Ann et al. reported the cloning of rat statin (S1) gene that is highly homologous to EF-1 α but shows distinct pattern of expression; i.e., G₀ phase specific expression, which is comparable to cell cycle independent expression of EF-1 α (4). Amino acid alignment between rat EF-1 α and S1 is shown in Fig. 1. Consensus sequences of GTP-binding proteins (Fig. 1, underlined) including EF-2, EF-G, EF-Tu, bacterial initiation factor IF-2 α , and c-Ha-ras (8) are all conserved in two proteins. Moreover, Asp-306, anchoring site of EF-1 α by phosphatidylinositol (9), is also conserved between two proteins (Fig. 1). The similarity especially in N-terminal domain strongly suggested that statin is the cytoplasmic GTP binding protein with different transcriptional regulation. Ann et al. indicated that S1 is unique copy of the EF-1 α /S1 gene family by genomic Southern analysis probing 3' specific probe of S1 (4). EF-1 α is multiple copy gene in mammalian genomes including human, mouse and pig (2). Madsen et al. have isolated 9 EF-1 α cDNA clones from liver cDNA library and they were all identical to that from a fibroblast cell line and from a lymphoid cell line, arguing that there is only one EF-1 α gene active in human (2). In rat, on the other hand, two active genes of EF-1 α /S1 genomic loci have been reported including this report. Comparison of 3' untranslated regions from these two active genes revealed that rat EF-1 α is clearly evolved from the same ancestral gene of Xenopus laevis EF-1 α (EMBL accession no. X55324) but that of S1 lacks significant homology with any EF-1 α from other species, suggesting that S1 has acquired the distinct transcriptional specificity by recombination of the promoter sequence as well as 3' untranslated regions during the evolution of EF-1 α .

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REFERENCES

- 1. Kozak, M. (1986) Cell 44, 283-292.
- Madsen, H.O., Poulsen, K., Dahl, O., Clark, B.F.C. and Hjorth, J.P. (1990) Nucleic Acids Res. 18, 1513-1516.
- Poting, A., Danker, K., Hartmann, L., Koster, M., Wedlich, D. and Knochel, W. (1990) Differentiation 44, 103-110.
- Ann,D.K., Moutsatsos,I.K., Nakamura,T., Lin,H.H., Mao,P.L., Lee,M.J., Chin,S., Liem,R.K.H. and Wang,E. (1991) J. Biol. Chem. 266, 10429-10437.
- Van Hemert, F.T., Amons, R., Pluijms, W.J., Van Ormondt, H. and Moener, W. (1984) EMBO J. 3, 1109-1113.
- 6. Schirmaier, F. and Philippsen, P. (1984) EMBO J. 3, 3311-3315.
- Pokalsky,A.R., Hiatt,W.R., Ridge,N., Rasmussen,R., Houck,C.M. and Shewmaker,C.K. (1989) Nucleic Acids Res. 17, 4661-4673.
- Kohno, K., Uchida, T., Ohkubo, H., Nakanishi, S., Nakanishi, T., Fukui, T., Ohtsuka, E., Idehara, M. and Okada, Y. (1986) Proc. Natl. Acad. Sci. USA 83, 4978-4982.
- Hayashi, Y., Urade, R., Utsumi, S. and Kito, M. (1989) J. Biochem. (Tokyo) 106, 560-563.

Figure 1. Comparison of amino acid sequence of EF-1 α with S1. Dashes denote the identical amino acid residues. Five underlined sequences are consensus sequences of GTP-binding protein (8). Boxed aspartic acid is phosphatidylinositol anchoring site (9).