Nucleotide sequence of the cDNA encoding silk gland elongation factor 1α

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Received December 23, 1992; Accepted January 9, 1993 DDBJ accession no. D13338

Most of eukaryotic elongation factor 1 (EF-1) consists of four subunits, EF-1α, -β, -β', and -γ, as shown in silk gland, wheat germ (1, 2), *Artemia salina* (3), and *Xenopus laevis* (4). EF-1α catalyzes the binding of aminoacyl-tRNAs to ribosomes concomitant with the hydrolysis of GTP, while EF-1ββ'γ catalyzes the exchange of GTP for GDP bound to EF-1α subsequent to its release from the ribosome and stimulates the binding of aminoacyl-tRNA to ribosomes. Recently, it was shown that two independent EF-1α genes in *Drosophila melanogaster* expressed individually during development (5). In *Xenopus laevis*, two EF-1α genes encoding the somatic and the oocyte form were also reported (6). In this report, we describe isolation of silk gland EF-1α cDNA clones and nucleotide sequence of a clone.

Poly(A)$^+$ RNA was isolated from the posterior silk gland of *Bombyx mori* at the 3rd day of the 5th instar and the cDNA was synthesized using the cDNA synthesis kit (Amersham Corp.). The silk gland cDNA library was constructed in λgt11 according to the manufacturer's instructions (Amersham Corp.). Several positive clones were obtained out of 1.0×10^4 recombinants by screening with human EF-1α cDNA as a probe (7). DNA fragments from these clones were subcloned into the pUC19 vector and one clone named KA71 was sequenced using the Sequenase Version 2.0 Kit applied to double stranded DNA (USB Corp.) (8).

The cDNA insert of KA71 contains 1392 bp of coding region encoding 464 amino acids, 47 bp of 5' untranslated region, and 248 bp of 3' untranslated region. The amino acids sequence had 94% similarity to *Drosophila melanogaster* EF-1α (5), 89% to *Artemia salina* EF-1α (9), 86% to human EF-1α (10), and 85% to *Xenopus laevis* EF-1α (11). These results suggest that EF-1α genes are well conserved among these species.

The consensus sequences GHVDHGKT (18–25), DCPG (80–83), and NKCD (135–138) involved in three GDP binding domains of *E.coli* EF-Tu (12) are conserved in silk gland EF-1α at residues 14–22, 91–94, and 153–156, respectively (Figure 1). Moreover, an ethanolamine–phosphoglycerol moiety is linked to the murine EF-1α via an amide bond to Glu-301 and Glu-374 (13) and Chinese hamster fibroblast EF-1α is modified by the covalent attachment of phosphatidylinositol at Asp-306 (14). These amino acids residues are also conserved in silk gland EF-1α at the same sites. Experiments are underway to isolate another clone of silk gland EF-1α.

ACKNOWLEDGEMENTS

We thank Drs T.Uetsuki and Y.Nabeshima (National Institute of Neuroscience, National Center of Neurology and Psychiatry) for the gift of plasmid pAN7 containing human EF-1α cDNA.

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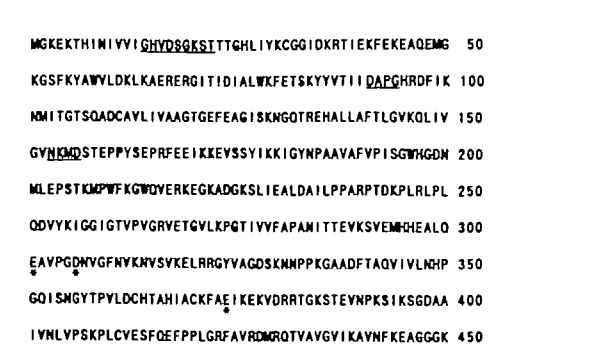

Figure 1. Amino acid sequence of silk gland EF-1α. Three underlined sequences are consensus sequence of GDP binding domains of *E.coli* EF-Tu (12). Two glutamic acids residues modified by ethanolamine-phosphoglycerol (13) and aspartic acid residue modified by phosphatidylinositol (14) are indicated by asterisks.

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