

Homology between the α and β subunits of chloroplast and bacterial proton-translocating ATPases

Hiroshi Deno* and Masahiro Sugiura⁺

Department of Biology, Faculty of Science, Nagoya University, Chikusa, Nagoya 464, Japan

Received 5 April 1984

The α and β subunits of tobacco chloroplast proton-translocating ATPase show 25% sequence homology. When these subunits from tobacco chloroplast and *Escherichia coli* are compared, 66 amino acid residues are identical and the majority of them are localized in 4 regions. Some nucleotide-binding enzymes contain sequences homologous to the 4 regions, suggesting that these regions have common functions in catalysis.

Tobacco chloroplast H⁺-ATPase Amino acid sequence

1. INTRODUCTION

Chloroplast proton-translocating ATPase (H⁺-ATPase) is an essential component for light-driven ATP synthesis in plants. The H⁺-ATPase complex consists of two parts, CF₁ and CF₀. The CF₁ is composed of 5 distinct subunits (α , β , γ , δ and ϵ) and possesses ATPase activity. The CF₀ is composed of 3 distinct subunits (I, II and III) and functions as an H⁺ channel. The 5 subunits (α , β , ϵ , I and III) are encoded by chloroplast DNA and the 3 subunits (γ , δ and II) by nuclear DNA [1,2]. We recently cloned the genes for the α , β , ϵ and III subunits from tobacco chloroplast DNA and determined their nucleotide sequences [3–5]. The deduced amino acid sequences of the tobacco chloroplast α and β subunits show moderate homology (54 and 62%) with those of the corresponding *E. coli* subunits, while the ϵ and III subunits show low homology (28 and 27%).

Partial homology between the amino acid sequences of the *E. coli* α and β subunits has been reported [6–8]. We compare here the amino acid sequence of tobacco chloroplast α subunit with

that of the β subunit and show that there are 4 homologous regions not only between the two subunits from tobacco chloroplasts but also among the 4 subunits from tobacco chloroplasts and *E. coli*.

2. RESULTS AND DISCUSSION

The amino acid sequences of H⁺-ATPase α and β subunits from tobacco chloroplasts [3,4] and *E. coli* [6,9–11] have been deduced from their DNA sequences. The number of their amino acid residues are similar to each other (507, tobacco α ; 498, tobacco β ; 513, *E. coli* α ; 459, *E. coli* β). Fig. 1 shows an optimal alignment of these 4 amino acid sequences. There is 25% homology between the tobacco chloroplast α and β subunits. Homologies (20–22%) were also found between the tobacco chloroplast α subunit and *E. coli* β subunit, and between the tobacco chloroplast β subunit and *E. coli* α subunit. Sixty-six amino acid residues are identical among the α and β subunits from tobacco chloroplast and *E. coli*, the majority being localized in 4 regions (C,D,G,I; fig.1).

Authors in [6] and in [8] have reported that there are some homologous sequences between the *E. coli* α or β subunit and other nucleotide-binding enzymes and these sequences are suggested to par-

* Present address: Bioscience Research Center, Waki-cho, Kuga-gun, Yamaguchi-ken 740, Japan

⁺ To whom correspondence should be addressed

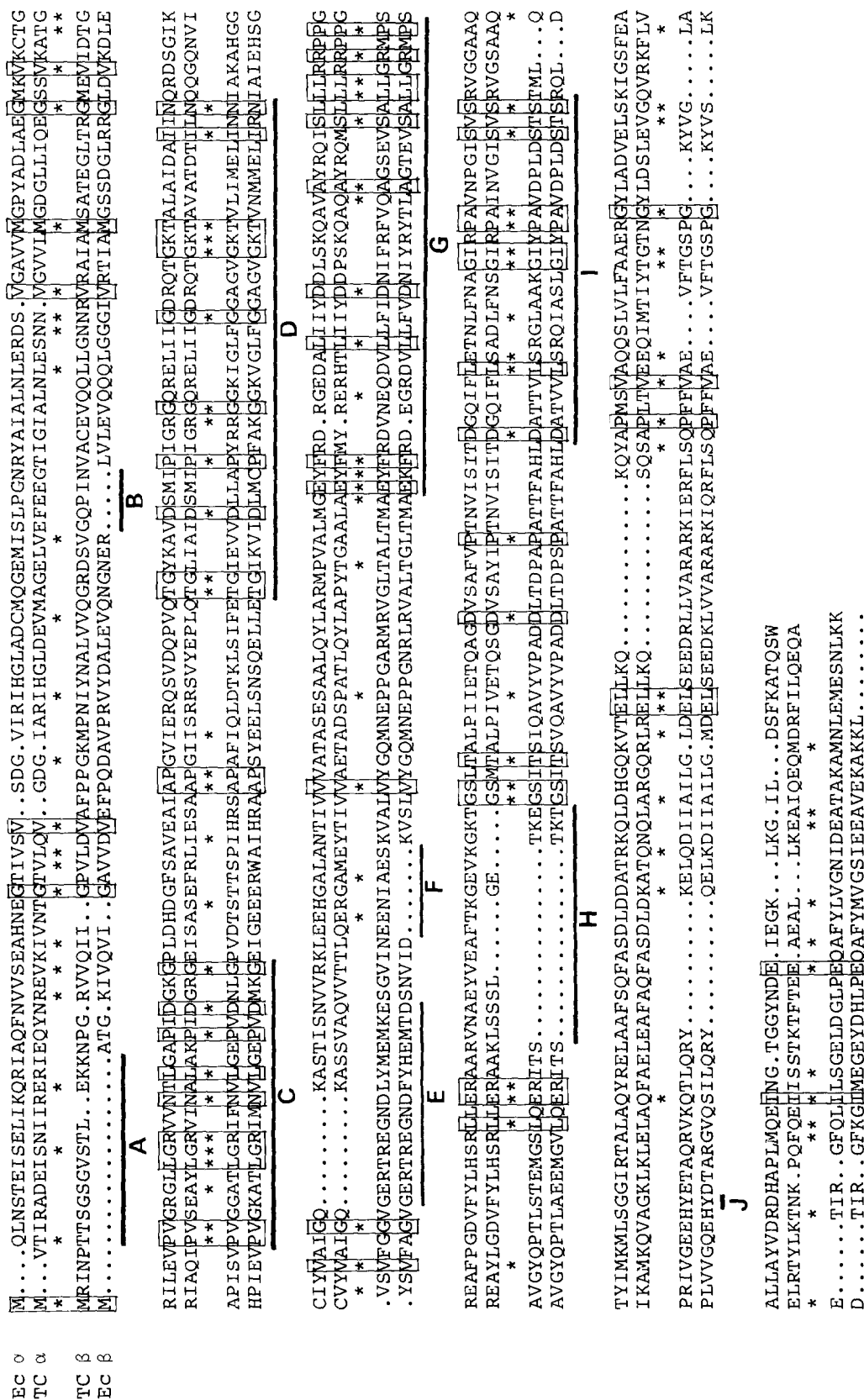


Fig.1. Comparison of amino acid sequences of the α and β subunits of H^+ -ATPase from tobacco chloroplasts (TC) and *E. coli* (Ec). Asterisks indicate homologous residues between the tobacco chloroplast α and β subunits. Boxes indicate homologous residues among the 4 subunits. A,B and F, regions deleted in the *E. coli* β subunit; E, Mg^{2+} -binding site in the β subunit; H, region inserted in the *E. coli* α subunit; J, tyrosine residue essential for ATPase activity in the β subunit; C,D,G and I, homologous regions in the 4 subunits.

(a) C-region	
TC α (103-122)	AYLGRVINALAKPIDGR.GEI
RecA (13-33)	<u>AALGQIEKNFGKGSIMRLGED</u>
(b) D-region	
TC α (165-188)	RELIIGDRQTGKTAVATDTILNQ
RecA (62-85)	IVEIYGPESSGKTTTLQVIAAAQ
EF-Tu (13-36)	NVGTIGHVDSGKTTLAITTVLAK
Harvey MSV (5-28)	KLVVVGARGVGKSALTIQLIQNH
Adenylate kinase (10-33)	IIFVVGPGSGKGTQCEKIVQKVG
(c) G-region	
TC α (251-274)	MYRERHTLIYDDPSKQAQAYRQM
Adenylate kinase (107-130)	RKIGOPTLLLYVDAGPETMTKRL
RecA (87-110)	NLKQNTLLIFINQIRMKIGVMFG
(d) I-region	
TC α (349-369)	ADLFNSGIRPAINVGISVSRVGS
Adenylate kinase (162-184)	<u>AFYEKRGIVRKVNAEGSVDDVFS</u>

Fig.2. Comparison of amino acid sequences of the 4 regions (C,D,G,I) in the tobacco chloroplast α subunit and of some nucleotide-binding enzymes. Homologous residues are doubly underlined and similar amino acid residues singly underlined. Numbers indicate residues in polypeptides from the amino terminal.

ticipate in binding adenine nucleotides. These homologous sequences reside in the D,E and G regions shown in fig.1. Besides those indicated by these authors, we found that some nucleotide-binding enzymes contain sequences homologous to the D and G regions of the tobacco chloroplast α subunit as shown in fig.2. We also found that *E. coli* recA protein and porcine adenylate kinase have sequences homologous to the C and I regions (fig.2). These observations suggest that the 5 regions (C,D,E,G,I) are important for binding adenine nucleotides.

The β subunit has been reported to contain an Mg^{2+} -binding site (or dicyclohexylcarbodiimide-binding site) in which two glutamic acid residues in the E region are involved [12]. Part of the E region

of the β subunits is deleted in the α subunits. The tyrosine residue in the β subunits (J, fig.1) has been shown to be essential for ATPase activity [13]. The α subunits do not have the tyrosine residue in corresponding regions and further no homology is observed between the sequences around the J regions of the α and β subunits. Therefore, the E and J regions may have different functions between the α and β subunits.

When the α or β subunits from tobacco chloroplasts and *E. coli* are compared, there are some deleted or inserted sequences (fig.1). The A,B and F regions are deleted only in the *E. coli* β subunit and the H region is present only in the *E. coli* α subunit (fig.1), suggesting that these sequences are not essential.

REFERENCES

- [1] Nelson, N., Nelson, H. and Schatz, G. (1980) Proc. Natl. Acad. Sci. USA 77, 1361-1364.
- [2] Westhoff, P., Nelson, N., Bünemann, H. and Herrmann, R.G. (1981) Curr. Genet. 4, 109-120.
- [3] Deno, H., Shinozaki, K. and Sugiura, M. (1983) Nucleic Acids Res. 11, 2185-2191.
- [4] Shinozaki, K., Deno, H., Kato, A. and Sugiura, M. (1983) Gene 24, 147-155.
- [5] Deno, H., Shinozaki, K. and Sugiura, M. (1984) Nucleic Acids Res., submitted.
- [6] Kanazawa, H., Kayano, T., Kiyasu, T. and Futai, M. (1982) Biochem. Biophys. Res. Commun. 105, 1257-1264.
- [7] Futai, M. and Kanazawa, H. (1983) Microbiol. Rev. 47, 285-312.
- [8] Walker, J.E., Saraste, M., Runswick, M.J. and Gay, N.J. (1982) EMBO J. 1, 945-951.
- [9] Kanazawa, H., Kayano, T., Mabuchi, K. and Futai, M. (1981) Biochem. Biophys. Res. Commun. 103, 604-612.
- [10] Gay, N.J. and Walker, J.E. (1981) Nucleic Acids Res. 9, 2187-2194.
- [11] Saraste, M., Gay, N.J., Eberle, A., Runswick, M.J. and Walker, J.E. (1981) Nucleic Acids Res. 9, 5287-5296.
- [12] Yoshida, M., Allison, W.S., Esch, F.S. and Futai, M. (1982) J. Biol. Chem. 257, 10033-10035.
- [13] Esch, F.S. and Allison, W.S. (1978) J. Biol. Chem. 253, 6100-6106.