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The prokaryotic thermophilic TF_1-ATPase is functionally compatible with the eukaryotic CF_o-part of the chloroplast ATP-synthase

Jean Michel Galmiche[b,**], Stephane Pezennec[b], Rongbao Zhao[a,***], Guy Girault[b], Edmund Baeuerlein[a,*]

[a]*Max-Planck-Institut für Biochemie, D-82152 Martinsried, Germany*

[b]*Centre d'Etudes de Saclay, Département de Biologie Cellulaire et Moléculaire, Section Bioénergétique, F-91191 Gif-sur-Yvette, France*

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Abstract

The ATP synthase from chloroplasts, $CF_o \cdot F_1$, was reconstituted into liposomes, from which most of CF_1 was removed by a short treatment with guanidinium chloride. ATP-dependent proton uptake was restored with these CF_o-liposomes even better by the addition of the bacterial TF_1- than of the related CF_1-part. This proton uptake was prevented by tentoxin, a specific inhibitor of the CF_1-ATPase, in these $CF_o \cdot F_1$-liposomes, but not in the hybrid $CF_o \cdot TF_1$-liposomes. Venturicidin, a specific inhibitor of proton flow through CF_o, was able to block it in both the hybrid $CF_o \cdot TF_1$-liposomes and reconstituted $CF_o \cdot F_1$-liposomes. These results indicate that the bacterial TF_1-part binds to the eukaryotic CF_o-part of four subunits forming a functional $CF_o \cdot TF_1$-ATPase.

Key words: Thermophilic bacterium PS3; Spinach chloroplast; $CF_o \cdot TF_1$-ATP synthase; Reconstruction (in vitro); Functional compatibility

1. Introduction

The ATP synthase of chloroplasts, bacteria and mitochondria consists of two sectors, the F_1-part, which bears the catalytic sites for ATP synthesis and the F_o-part, that functions to transfer protons [1] or in some bacteria sodium ions [2] across the correlated membran es. Only Na^+ ions allowed to show that the F_o-part of a bacterial $F_1 \cdot F_o$ ATPase from *Propionigenium modestum* binds ions [3] involved in energy transduction. This ion specificity of the F_o-part could be supported by a hybrid constructed in vitro with F_1 from *E. coli* and F_o from *P. modestum* [4]. Thus first energy conservation occurs in the F_o-part, probably by binding Na^+ or H_3O^+ ions [5]. The crucial question is now, in which way this energy is transferred from F_o to F_1 to release tightly bound ATP, i.e. which subunits are involved in the interaction between F_o and F_1.

We report here the formation of a hybrid constructed in vitro with prokaryotic TF_1 from the thermophilic bacteria PS3 and eukaryotic CF_o from spinach chloroplast on $CF_o \cdot CF_1$ liposomes, which were largely depleted of CF_1. This $CF_o \cdot TF_1$ hybrid is more active in ATP-driven ACMA quenching than the $CF_o \cdot F_1$ ATPase, reconstituted with isolated CF_1.

2. Materials and methods

$CF_o \cdot F_1$ was prepared according to the method of Fromme et al. [6], CF_1 according to Berger et al. [7], and TF_1 according to Kagawa and Yoshida [8]. Protein concentration in CF_1 and TF_1 was measured by the method of Lowry and by optical absorbance at 280 nm using an extinction coefficient of 0.48 cm^2/mg.

500 mg of phosphatidyl choline type II S from Sigma were solubilised in 10 ml buffer of 10 mM Tricine, pH 8, 0.2 mM EDTA, 5 mM DTT containing 100 mg of desoxycholate and 200 mg of cholate. The suspension was stirred at room temperature until homogeneity, then cooled to 0°C and sonicated for 1 min with the standard tip at full energy (Labsonic U, Braun Biotech Int.; 350 W/min). The lipid solution was centrifugated in a Beckman rotor SW 60 for 10 min at 27,000 rpm. 2 mg $CF_o \cdot F_1$ in 0.32 ml were mixed with 3.68 ml of the phospholipid solution and submitted to 10 times 0.5 s bursts (with 0.5 s cooling between each) with the needle tip of the sonicator at half energy at 0°C. Liposomes were kept in ice for 1 h. This sample of 4 ml was dialysed against 2 liters of buffer containing 10 mM Tricine pH 8, 0.2 mM EDTA, 2.5 mM $MgSO_4$, 130 mM K_2SO_4 and 0.25 mM DTT preequil-

* Corresponding author. Fax: (49) (89) 8578 3777.

** *Present address:* Max-Planck-Institut fuer Biochemie D-82152 Martinsried, Germany.

*** *Present address:* A. Einstein College of Medicine, Dept. Physiology and Biophysics, Bronx, N.Y. 10461, USA

Abbreviations: ACMA, 9-amino-6-chloro-2-methoxy-acridine; CHAPS, 3-[(cholamidopropyl)-dimethyl-ammonio]-1-propane sulfate; DCCD, dicyclohexylcarbodiimide; DTT, D,L-dithio-threitol; EDTA, ethylenediaminetetraacetic acid; FCCP, carbonyl cyamide-*p*-trifuorohydroxyphenyl hydrazone; GCL, guanidinium chloride; [^{125}I]ASA-βala-OH, 3[^{125}Iodo]-4-azido-2-hydroxybenzoyl-β-alanine; Tricine, *N*-[2-hydroxy-1,1-bis(hydroxymethyl)ethyl] glycine.

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ibrated at 30°C. Dialysis was run for 20 h at 30°C with one change of the dialysis buffer. The $CF_o \cdot F_1$ liposomes suspension contained 0.5 mg of $CF_o \cdot F_1$ per 46 mg phospholipid in 1 ml.

CF_1 was depleted from the $CF_o \cdot F_1$ liposomes by guanidinium chloride treatment according to McEnery et al. [9]. 0.5 ml of the $CF_o \cdot F_1$ liposomes were mixed with 3 ml buffer containing 10 mM Tricine pH 8, 0.2 mM EDTA and centrifugated in a Beckman rotor SW 60 for 1 h at 50,000 rpm. Pellets were resuspended in 1 ml buffer containing 10 mM Tricine, pH 8, 5 mM EDTA and 315 mg guanidinium chloride (GCl). The suspension was kept in ice for 5 min, then mixed with 6 ml of the same buffer but without GCl and cooled at 0°C. After centrifugation as described above, the pellets were resuspended in 3.5 ml of the same buffer without GCl at 0°C and centrifugated again as above. The pellet of CF_1 depleted $CF_o \cdot F_1$ liposomes was resuspended in 0.5 ml buffer containing 10 mM Tricine pH 8, 0.2 mM EDTA, 2.5 mM $MgSO_4$ and 130 mM K_2SO_4, and stored in ice. In photolabeling experiments the same procedure was used after incubation with 0.5 μM [^{125}I]ASA-βala-OH (10 Ci/mmol) and irridation by wavelengths longer than 300 nm.

The CF_1 depleted liposomes were reconstituted with CF_1 or TF_1, as follows: 20 μl of CF_1 depleted liposomes were mixed with 0 to 20 μg of CF_1 or TF_1 in a buffer containing 10 mM Tricine pH 8, 0.2 mM EDTA, 2.5 mM $MgSO_4$ and 130 mM K_2SO_4, and kept in ice for at least 2 h. Not incorporated protein was separated from the $CF_o \cdot F_1$ liposomes by flotation in a discontinuous sucrose gradient according to Rigaud [10].

By trypsin treatment of the $CF_o \cdot F_1$ liposomes ATPase molecules facing the outside medium were degraded. The remaining ATPase activity in the liposomes after their solubilization in 1.5% Triton X-100 allowed to determine the amount of ATPase molecules facing the inside of the liposomes.

ATPase activity of soluble F_1-ATPase as well as $F_o \cdot F_1$ liposomes (each containing 35–70 μg protein/ml) were measured. They were incubated for 10 min at 37°C in a buffer containing 50 mM Tricine pH 8.0, 37.5 mM $NaHCO_3$, 6.8 mM $MgSO_4$, 15 mM ATP and 1.5% Triton X-100 with various amounts of trypsin. Reactions were quenched by perchloric acid and inorganic phosphate was determined colorimetrically as molybdeneblue.

3. Results

3.1. Reconstitution of $CF_o \cdot F_1$ into liposomes

The chloroplasts ATP synthase $CF_o \cdot F_1$ was reconstituted into liposomes, which were separated by a discontinuous sucrose gradient according to Rigaud [10]. The proteoliposomes were found between 10–20% sucrose. The protein, which was not incorporated into the liposomes, was detected on the bottom of the tube. After the proteoliposome fraction was dissolved by Triton X-100, its ATPase activity represented 85%, and that of the protein about 15% of the total activity measured.

When the proteoliposomes were incubated with trypsin for two hours and their ATPase activity was determined, after they were dissolved by Triton X-100, only 8% of the original 85% were found. Thus apparently 90% CF_1 of $CF_o \cdot F_1$ reconstituted into the proteoliposomes, is located on the outer surface.

3.2. Removal of CF_1 from $CF_o \cdot F_1$-liposomes

CF_1 depleted liposomes were obtained, when $CF_o \cdot F_1$ liposomes were incubated with 1,4 M guanidinium chloride. ATP-dependent proton uptake was restored best by the addition of TF1-ATPase, when only a short incubation time of 5 min with guanidinium chloride in diluted liposomes samples was used before (Table 1). These CF_1 depleted liposomes remain active at least for one week, when stored at 0°C. After photolabeling experiments with [^{125}I]ASA-βala-OH incubation with guanidinum chloride yielded depletion of CF_1 subunits, the efficiency of which was placed between the incubation with EDTA and NaBr. (Fig. 1).

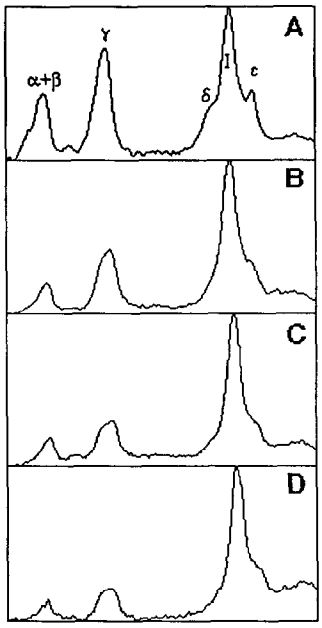

Fig. 1. Solubilisation of the CF_1 part from CF_oF_1 liposomes after treatment with (A) 10 mM Tricine, 0.2 mM EDTA, pH 8.0 (reconstitution buffer), (B) 0.7 mM EDTA, 1 mM Tricine, pH 8.0, (C) 1.4 M guanidinium chloride, 5 mM EDTA, 10 mM Tricine, pH 8.0, (D) 2 M NaBr, 10 mM Tricine, pH 8.0[19]. A concentration of 0.5 μM [^{125}I]ASA-βala-OH (10 Ci/mmol) was used for photolabeling before solubilisation procedures. The peak of subunit I was used as internal standard.

Table 1
ATP-dependent proton uptake after addition of various amounts of CF_1 and TF_1

TF1 added μg per 5 μg of CFOF1 initially present	CFOF1 liposomes treated with 1.4 M guanidinium hydrochlorid for different periods		
	5 min	10 min	20 min
0	0	0	0
1.56	1.12	0.65	0.28
3.9	0.87	0.67	–
7.8	0.81	0.63	0.35
15.6	0.79	0.57	0.33

CF_1 was depleted from $CF_o \cdot F_1$ liposomes by treatment with 1.4 M quanidinium chloride in 5, 10 and 15 min. Reconstitution with TF_1 to the hybrid $CF_o \cdot TF_1$ ATPase was determined by measuring the rate of the initial ATP-dependent ACMA fluorescence quenching. CF_1 depleted liposomes were incubated in the dialysis medium with increasing amounts of TF_1 (μg of TF_1 per 5 μg of $CF_o \cdot F_1$ initially present in the liposomes). Rate was ACMA fluorescence quenching per second expressed as percent of the fluorescence observed before ATP addition.

3.3. Reconstitution of ATP-dependent proton uptake by addition of CF_1 or TF_1 to CF_o-liposomes

CF_1 was treated with 50 mM DTT before addition to CFo liposomes, what is not necessary for TF_1. ATP-dependent proton uptake had to be measured by quenching of the fluorescent dye ACMA in the presence of valinomycin, when CF_1 was added to CF_o-liposomes. Without valinomycin no ACMA quenching could be observed. With TF_1 added, valinomycin was not necessary, but in its presence this proton uptake was faster and greater. After addition of increasing amounts of CF_1 and TF_1 resp. the initial rate of fluorescence quenching, R%/s, was determined as a measure for proton uptake (Fig. 2). Maximal initial rates of ACMA quenching were obtained by addition of 0.8 μg CF_1 and 0.44 μg TF_1 resp. to CF_o liposomes, prepared from 5 μg $CF_o \cdot F_1$. This corresponds to a maximal reconstitution of 16% active $CF_o \cdot F_1$-ATPase and 8.8% hybrid $CF_o \cdot TF_1$-ATPase resp. The maximal initial rate of reconstituted $CF_o \cdot F_1$-ATPase with 0.8 μg CF_1 was found to be 36% of that which was obtained by the hybrid $CF_o \cdot TF_1$-ATPase, using 0.44 μg TF_1. The maximal final fluorescence quenching, Q%, (measured in% of the fluorescence observed just after ATP addition) was 44% with 0.16 μg CF_1 and 57% with 0.44 μg TF_1.

3.4. Action of the CF_1-specific inhibitor, tentoxin and of the CF_o-specific inhibitor venturicidin on $CF_o \cdot F_1$- and the hybrid $CF_o \cdot TF_1$-liposomes

A final concentration of 12 μM tentoxin inhibited completely quenching of ACMA, which is obtained by

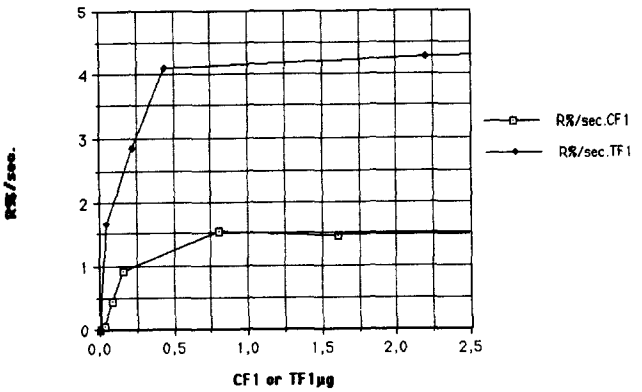

Fig. 2. ATP-dependent proton uptake of CF_o-liposomes after addition of various amounts of CF_1 and TF_1. ATP induced fluorescence quenching was measured in 10 mM tricine pH 8, 0.2 mM EDTA, 2.5 mM $MgSO_4$, and 0.25 μM ACMA. 1.25 mM ATP was added, then 0.9 nM valinomycin. R% was the initial rate of fluorescence quenching per second. R was measured in % of the fluorescence observed just after ATP addition to take into account the direct fluorescence quenching induce by ATP on the ACMA dye. For reconstitution CF_1 depleted proteoliposomes (prepared from $CF_1 \cdot F_o$ liposomes, containing 5 μg $CF_1 \cdot F_o$) were mixed with various amounts of CF_1 (50 mM DTE treated) or TF_1 in dialysis medium.

CF_1, —⊡—; TF_1, —◆—;

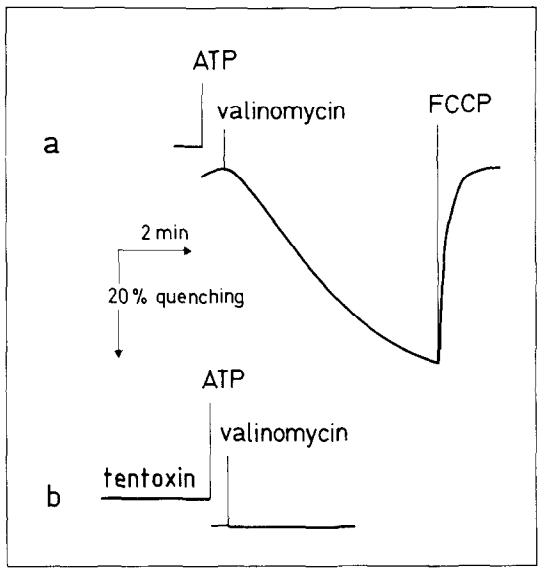

Fig. 3. Quenching of ACMA fluorescence by CF_o-liposomes, reconstituted with CF_1, in the presence or absence of tentoxin. CF_1 depleted $CF_o \cdot F_1$ liposomes were incubated with 6 μg CF_1 for 10 μg $CF_o \cdot F_1$ initially present before guanidinium chloride treatment. 10 μl of these liposomes, (corresponding to 500 μg of lipids) were added to 0.4 μl of the medium: 10 mM tricine pH 8, 0.2 mM EDTA, 2.5 mM $MgSO_4$ and 0.25 μM ACMA. Valinomycin was added to a final concentration of 1 nM, ATP to 1.25 mM and FCCP to 250 nM. When added tentoxin ATP dependent fluorescence quenching of reconstituted $CF_1 \cdot F_o$ liposomes in the absence (curve a) or in the presence (curve b) of tentoxin.

ATP-dependent proton uptake in liposomes, into which $CF_o \cdot F_1$-ATPase was reconstituted as a whole (Fig. 3) and CF_o-liposomes, with which $CF_o \cdot F_1$ was reconstituted by the addition of CF_1 (Fig. 4). This inhibition of ACMA quenching could not be found, after the hybrid $CF_o \cdot TF_1$- ATPase was formed by adding TF_1 to CF_o-liposomes (Fig. 5). In all liposomes, studied above, 13 μM venturicidin, a specific inhibitor of the CF_o-part, as well as 63 μM DCCD, a specific inhibitor of all F_o-parts known, prevented quenching of ACMA, driven by ATP-dependent proton uptake (not shown here). The addition of TF_1 to CF_o-liposomes created obviously an hybrid $CF_o \cdot TF_1$- ATPase, which transported protons into the liposomes by hydrolysis of ATP.

4. Discussion

Hybrid $F_o \cdot F_1$-ATPases which are reconstituted with F_1- and F_o-parts from different species, are useful in studying function and interaction of these two parts. Recently it was demonstrated by F_1-stripped everted membrane vesicles that F_1 and F_o-parts from *E. coli* and the thermophylic bacterium PS3 [11] as well as those from the closely related bacteria *E. coli* and *Salmonella typhymurium* [12] are functionally compatible. Furthermore a hybrid ATPase, composed of F_1 from *E. coli* and

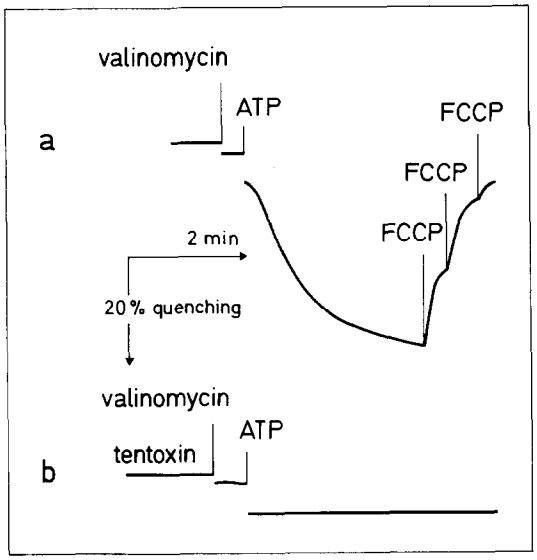

Fig. 4. ATP-dependent quenching of ACMA fluorescence by $CF_o \cdot F_1$ liposomes in the presence or absence of tentoxin. 10 μl of the $CF_o \cdot F_1$ liposomes (corresponding to 250 μg of lipids) were added to 0.4 μl of the medium containing 10 mM EDTA, 2.5 m $MgSO_4$ and 0.25 μM ACMA Valinomycin was added to a final concentration of 0.36 nM, ATP to 2 mM, FCCP to 125 nM. When added tentoxin was 12 μM and liposomes were preincubated for 5 min with tentoxin ATP-dependent fluorescence quenching by $CF_1 \cdot F_o$ liposomes in the absence (curve a) or in the presence (curve b) of tentoxin was recorded.

F_o from *Propionigenium modestum* in a reconstituted system, was shown to be a pump of sodium ion as the original $F_o \cdot F_1$-ATPase from *P. modestum*. It was concluded that the F_o-part determines ion specificity [4]. This essential finding was supported by extended experiments with F_1 and F_o parts of *P. modestum* and the thermophilic bacterium PS3, demonstrating that the hybrid $PF_o \cdot TF_1$-ATPase is a sodium pump and the hybrid $TF_o \cdot PF_1$-ATPase a proton pump. Both transports could be inhibited by DCCD (E.B., unpublished). A first hybrid ATPase from prokaryotic and eukaryotic F_1 and Fo parts was formed by binding of *E. coli* F_1 to membranes of rat liver motochondria and the related mitochondrial F_1 to *E. coli* everted membranes [14]. In a second approach the thermophilic TF_1 was incubated with lettuce chloroplasts, which were partially depleted of CF_1 by EDTA. Because the photophosphorylation activities after reconstitution with CF_1 as well as with TF_1 were inhibited by tentoxin in both cases, it was concluded that TF_1 sealed only the membrane, and endogenous CF_1 promoted tentoxin-sensitive ATP synthesis [15].

The hybrid CFo · TF1-ATPase, however, reconstituted here in liposomes, was completely insensitive to tentoxin (Fig. 5c). Furthermore rebinding of TF_1 to CF_o-liposomes was better reproducible than that of CF_1, reflecting the extraordinary stability of TF_1. This parallels our experience, mentioned above, in the reconstitution of a sodium ion pump by adding TF_1 to liposomes with F_o of *P. modestum*, where the hybrid $PF_o \cdot TF_1$-ATPase was ten times better coupled (ratio: Na^+/ATP) (E.B., unpublished) than the related hybrid with F_1 from *E. coli* [4]. It was, therefore, surprising that only about 1 of 10 CF_o had bound TF_1 forming an active ATPase. This was the case, though about 90% CF_1 was on the outside of $CF_o \cdot F_1$ liposomes and most of CF_1 was removed by guanidinium chloride (Table 1). This removal could be evaluated semiquantitatively by a new water soluble, radioactive photolabel [^{125}I]ASA-βAla-OH, which was introduced for bulk phase modification of membrane proteins (Fig. 1). Apparently most of the subunits α, β and δ were removed, whereas some ε and predominantly remained on the liposomes. The treatment with NaBr appeared in our hands the most effective one for the removal of CF_1 subunits, but not in reconstitution experiments with CF_1 or TF_1 resp. After treatment with guanidinium chloride for 5 min none of the CF_o-parts appeared to be leaky under the conditions used for ACMA quenching (Fig. 5a).

One possible explanation may be that these CF_o moieties do not function as open channels, but by a carrier-type mechanism, as found in studies with the F_o-part of

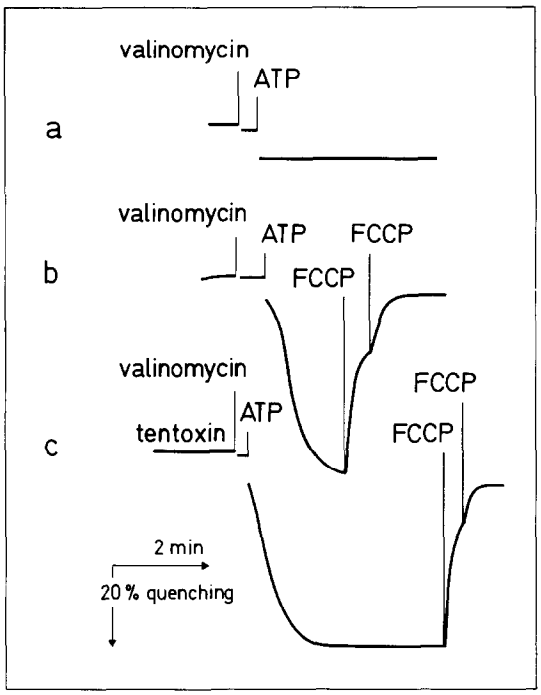

Fig. 5. ATP-dependent quenching of ACMA fluorescence by CF_o liposomes and $CF_o \cdot TF_1$ liposomes in presence and absence of tentoxin. CF_1 depleted $CF_o \cdot CF_1$ liposomes were incubated or not with 25.2 μg TF_1 for 10 μg $CF_o \cdot F_1$, initially present before guanidinium chloride treatment. 10 μl of the liposomes (corresponding to 250 μg of lipids) were added to 0.4 μl of the medium containing 10 mM tricine pH 8, 0.2 mM EDTA, 2.5 mM $MgSO_4$ and 0.25 μM ACMA. Valinomycin was added to a final concentration of 0.36 nM, ATP to 2 mM and FCCP to 125 nM. When added tentoxin was 12 μM and liposomes were preincubated for 5 min with tentoxin, ATP-dependent fluorescence quenching was prevented. Fluorescence quenching of CF_1 depleted liposomes (curve a), of $TF_1 \cdot CF_o$ liposomes in the absence (curve b) or in the presence (curve c) of tentoxin was recorded.

P.modestum[3]. On the other hand even short treatment of $CF_o \cdot F_1$ liposomes with guanidinium chloride may have changed composition and structure of many CF_o parts, because longer treatment decreased more and more proton induced ACMA quenching after addition of TF_1 (not shown here).

Obviously is the membrane part CF_o of spinach chloroplast ATP synthase functionally compatible with the TF_1 moiety of the bacterial ATP synthase $TF_o \cdot F_1$, though CF_o has two different b subunits, b(CF_oI) and b'(CF_oII), whereas all bacterial F_o posess two identical subunits b. In an *E. coli* mutant carrying a chain termination in the unc F gene, subunit b may be replaced by spinach subunit b(CF_oI) yielding a strain of *E. coli*, which was now able to grow on succinate-minimal medium [16]. This result reflects the possibility that spinach subunit b may interact functionally with EF_1 without any subunit b'(CF_oII) present. On the other hand evidence was presented that subunit b'(CF_oII) is an essential subunit of the photosynthetic ATP synthase $CF_o \cdot F_1$ [17,18] and is necessary for rebinding of CF_1 to EDTA-treated thylakoids [17]. In addition remarkable similarity in secondary structure predictions allows to suggest that subunit b(CF_oI) and subunit b'(CF_oII) possess a similar membrane topography. Because subunit b'(CF_oII) has been found in all photosynthetic bacterial and chloroplast enzymes (for review see [18]) and appears to be necessary for their structure and function, it is an open question, if the bacterial TF_1 interacts with subunit b'(CF_oII) or not in the above described $CF_o \cdot TF_1$ hybrid.

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References

[1] Fillingame, R.H. (1990) in: The Bacteria, Vol. XII, Bacterial Energetics (T.A. Krulwich, Ed.) Academic Press, New York, pp. 345–391.
[2] Laubinger, W. and Dimroth, P. (1989) Biochemistry 28, 7194–7198.
[3] Kluge, C. and Dimroth, P. (1992) Biochemistry 31, 12665–12672.
[4] Laubinger, W., Deckers-Hebestreit, G., Altendorf, K. and Dimroth, P. (1990) Biochemistry 29, 5458–5463.
[5] Boyer, P.D. (1988) Trends Biochem. Sci. 13, 65–7.
[6] Fromme, P., Boekema, E.J. and Gräber, P. (1987) Z. Naturforsch. 42c, 1239–1245.
[7] Berger, G., Girault, G., Andre, F. and Galmiche, J.M. (1987) J. Liquid. Chrom. 10, 1507–1517.
[8] Kagawa, Y. and Yoshida, M. (1979) Methods Enzymol. 55, 781–787.
[9] McEnery, M.W., Hulliken, J. and Pedersen, P.L. (1989) J. Biol. Chem. 264, 12029–12036.
[10] Rigaud, J.L., Paternostre, M.-T. and Bluzat, A. (1988) Biochemistry 27, 2677–2668.
[11] Steffens, K., Di Gioia, A., Deckers-Hebestreit, G. and Altendorf, K. (1987) J. Biol. Chem. 262, 6334–6338.
[12] Hsu, S.Y., Scuda, M., Kanazawa, H., Tschuchiya, T. and Futai, M. (1984) Biochemistry 23, 988–993.
[13] Laubinger, W., Deckers-Hebestreit, G., Altendorf, K. and Dimroth, P. (1990) Biochemistry 29, 5458–5463.
[14] Liu, S.-S., Gao, F.H., Tsai, H.L. and Ding, Y.Z. (1982) Biophys. J. 37, 88–91.
[15] Bar-Zvi, D., Yoshida, M. and Shavit, N. (1985) Biochim. Biophys. Acta, 806, 341–347.
[16] Schmidt, G., A.J. W. Rodgers, S.M. Howitt, A.L. Munn, G.S. Hudson, T.A. Holten, P.R. Whitfeld, W. Bottomley, F. Gibson and G.B. Cox (1990) Biochim. Biophys. Acta, 1015, 195–199.
[17] Berzborn, R.J., L. Klein-Hitpaß, J. Otto, S. Schünemann, R. Oworah-Nkruma and H.E. Meyer (1990) Z. Naturforsch. 45c, 772–784.
[18] Hermann, R.G., J. Stepphun, G.S. Herrmann and N. Nelson (1993) FEBS Lett. 326, 192–198.
[19] Grotjohann, I. and P. Graeber (1990) Biochim. Biophys. Acta 1017, 177–180.