The ps transient absorption spectra of the ethyletioporphyrin (EEP)–toluqunone (TQ) system were measured with a microcomputer-controlled Nd^{3+}:YAG laser photolysis system.[5] In acetone (Figure 1), where EEP fluorescence is almost completely quenched by TQ, the transient spectra are very similar to the $S_n \leftarrow S_1$ spectra of EEP itself. We obtained the decay time of this transient absorbance as $\tau_{obsd} \approx 70$ ps, which was in approximate agreement with the value (~80 ps) estimated from the relation $\tau_{calcd}S_1 = \tau_0/(1 + k_q\tau_0[TQ])$ with τ_0 and k_q values determined in the present work. These results indicate that the excited EEP in acetone is quenched by encounter collision with TQ but the produced ion pair is immediately deactivated without producing separated ion radicals. Moreover, an examination of the ground-state absorption spectra and the relationship between fluorescence yield and [TQ] reveals that about 66% of EEP forms a ground-state loose complex with TQ in acetone.[6] In the light of the above results of ps transient absorption studies, this fact leads to the conclusion that the loose complex undergoes ultrafast deactivation via a solvated ET state or ion-pair state immediately after excitation.

Contrary to the above results, the ps transient absorption spectra in benzene (Figure 2) are quite different from the $S_n \leftarrow S_1$ spectra of EEP. The absorption band shows a maximum aroung 650–700 nm, and its intensity drops strongly in the longer wavelength side (in contrast to the EEP $S_n \leftarrow S_1$ spectra, which show a flat band in this region) and is similar to that of porphyrin cation.[7] Moreover, it has been confirmed that about 90% of EEP forms a loose complex in benzene solution.[6] Since the TQ anion does not show an absorption band in this wavelength region, the transient absorption spectra in Figure 2 can be assigned to the exciplex ($EEP^+ \cdot TQ^-$) formed by the excitation of the loose complex. We obtained the decay time of this exciplex as $\tau_{obsd} \approx 40$ ps. This τ_{obsd} is much shorter than the $\tau_{calcd}S_1$ (~130 ps) obtained by assuming encounter collisional quenching.

The above results provide a direct connection between the porphyrin–quinone system and the typical exciplexes. Although it is rather short-lived, the porphyrin–quinone exciplex can be observed in nonpolar solvents, while the photoinduced ET state undergoes ultrafast deactivation to the ground state in polar solvents. Solvation in the ET state lowers its energy but lifts up the energy of the neutral ground state compared to that relaxed with respect to solvation,[3e] which results in a very small energy gap between two states leading to the ultrafast deactivation in the porphyrin–quinone system.[6] We have confirmed the same result also in the excited EDA complex of pyromellitic dianhydride–pyrene.[8] However, the energy gap in the case of typical exciplexes such as pyrene–DMA or –DCNB is not so small, according to our estimate of the solvation energy.[6,8]

We have examined also the exciplex systems of the cyclophane type face to face porphyrin dimer (FTFP) (etioporphyrins combined by two $(CH_2)_2$–CO–NR–$(CH_2)_2$ chains) and TQ. The ps transient absorption spectra of the FTFP–TQ system in benzene can be assigned to the exciplex ($FTFP^+ \cdot TQ^-$) since they are similar to that of the FTFP cation,[7c] the decay time of which was shorter than 25 ps according to our measurement. The shorter τ_{obsd} compared to the EEP–TQ system can be ascribed to the faster nonradiative deactivation owing to the smaller energy gap between the ET and ground states for the FTFP–TQ system, since the oxidation potential of FTFP is a little lower than that of the EEP, according to our measurement. The lower oxidation potential indicates some delocalization interaction between two porphyrin rings. Therefore, the delocalization of positive charge over two porphyrin rings has little effect for lengthening the ET state lifetime. In acetone, no ET state was observed due to the ultrafast deactivation.

The above results clearly demonstrate the crucial role of a polar environment that causes ultrafast deactivation of porphyrin–quinone ET state. This finding is very important for designing biomimetic artificial photosynthetic systems.

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trans-6-Amino-5-[(1-carboxyethenyl)oxy]-1,3-cyclohexadiene-1-carboxylic Acid: An Intermediate in the Biosynthesis of Anthranilate from Chorismate

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Microorganisms utilize chorismate (**1**) for the biosynthesis of tryptophan.[1] The initial enzymatic reaction, the conversion of **1** to anthranilate (**2**), is catalyzed by anthranilate synthase, and the amide nitrogen of glutamine serves as the nitrogen source.[2,3] The enzyme from *Serratia marcescens* and other enteric bacteria has two subunits. One subunit (AS I) catalyzes the conversion of **1** and NH_3 to **2** and pyruvate. The other subunit (AS II) serves as the glutamine amidotransferase. It has been established that the nitrogen atom becomes attached at C-2 of **1**,[4] and the C-2 hydrogen atom of **1** is not incorporated into the pyruvate formed in the reaction.[5,6]

Amino acid **3** (Scheme I) has been postulated as the intermediate in the biosynthesis of **2** from **1**,[4,7] but attempts to isolate an intermediate have not been successful.[8] It has been suggested that the stereochemistry of **3** ought to be cis rather than trans.[1a] The isolation of *trans*-2,3-dihydro-3-hydroxyanthranilic acid from a strain of *Streptomyces aureofaciens*,[9] however, suggests that **3** is the metabolic intermediate. Described below are our synthesis of (±)-**3** and the enzyme-catalyzed transformation of **3** to **2** with *S. marcescens* AS I from a plasmid-containing *E. coli* strain.[10]

Carbamate **5** (Scheme II), prepared in 10% yield from the Diels–Alder reaction of methyl propiolate and *tert*-butyl *trans*-1,3-butadiene-1-carbamate,[11] was epoxidized (*m*-chloroperoxybenzoic acid, CH_2Cl_2), and the epoxide was isomerized to **6**[12] with 1,3-diazabicyclo[5.4.0]undec-7-ene in THF (40% from **5**). Re-

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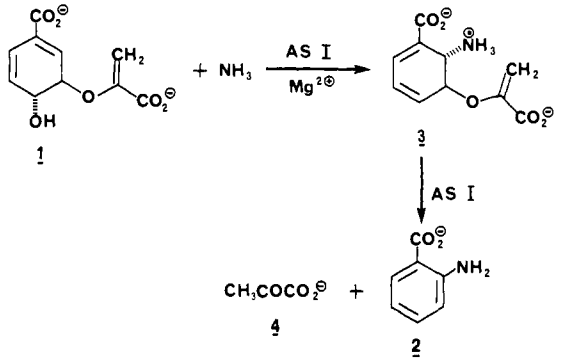
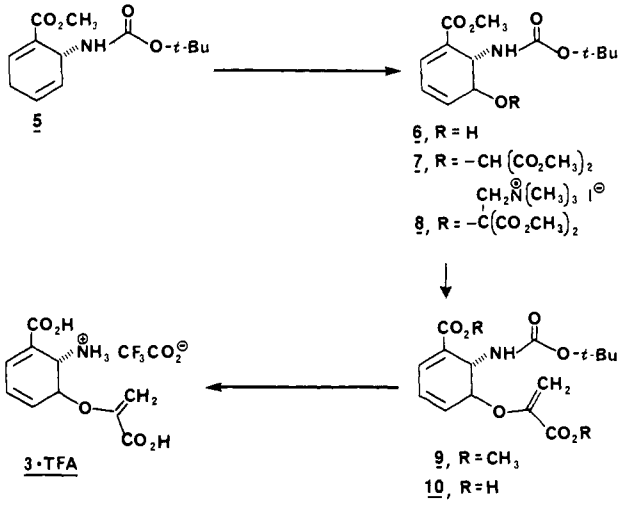
Scheme I



Scheme II



action of **6** with dimethyl diazomalonate and 1 mol % $Rh_2(OAc)_4$ in benzene at 65 °C gave **7** (56%).[13] Reaction of **7** with Eschenmoser's salt [$CH_2{=}N(CH_3)_2{}^+I^-$, $(C_2H_5)_3N$, CH_2Cl_2] and quaternization of the Mannich base (CH_3I, CH_2Cl_2) provided **8** (100%). Base-induced fragmentation (1.5 equiv of NaOH, THF/H_2O, 0 °C, 45 min) gave **9**[14] (46%). Saponification of **9** (2.2 equiv of NaOH, THF/H_2O, 4 °C, 40 h) followed by acidification with Amberlite IR-120 resin afforded **10** (94%). Treatment of **10** with dry, freshly distilled CF_3CO_2H (TFA) at 0 °C for 15 min followed by workup gave salt **3**·TFA (43%).[15,16]

To test compound **3** as a potential intermediate in the enzymic biosynthesis of anthranilate from chorismate and ammonia, samples of the trifluoroacetate salt, **3**·TFA, were incubated with pure *S. marcescens* AS I enzyme.[17] For comparison chorismate,[18] with or without ammonia, was used as control substrate. Compound **3** was an excellent substrate, undergoing enzymic conversion to anthranilate[19] in the absence of NH_4^+ with a K_m of 0.2 mM and V_{max} of 300 (nmol/min)/mg enzyme compared to a K_m of 0.11 mM and V_{max} of 500 (nmol/min)/mg for the natural substrate chorismate[17,20] in the presence of ammonia. In the absence of NH_4^+ ions, chorismate gave no anthranilate. Addition of 50 mM NH_4^+ to enzymic incubations of **3**·TFA did increase V_{max} values ca. 2-fold such that under these conditions **3**·TFA was processed to anthranilate at higher V_{max} than chorismate so **3**·TFA is both a kinetically and chemically competent candidate for a reaction intermediate.

It was anticipated that anthranilate synthase would act stereospecifically on only one of the enantiomers of (±)-**3**·TFA, presumably the 5*S*,6*S* isomer. In incubations containing 0.2–3.2 mM **3**·TFA[15] with varying enzyme levels, we routinely observed 24–27% conversion in the absence of NH_4^+ and 35% in the presence of NH_4^+ by fluorescence assay.[17] In parallel incubations where coproduct pyruvate (**4**) was monitored by coupled in situ reduction by L-lactate dehydrogenase and NADH, 34–35% conversions were detected, with or without added NH_4^+. This is substantial conversion but less than 50% for reasons as yet unclear.[21]

In sum, compound **3** is processed enzymically to anthranilate by the *S. marcescens* synthase at rates that support its role as reaction intermediate and thereby substantiate the mechanism of Scheme I for this enzyme, with trans geometry in the amino enol pyruvyl intermediate.

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Supplementary Material Available: Physical data for **6**, **7**, **9**, and **3**·TFA (1 page). Ordering information is given on any current masthead page.

(13) Procedure of Ganem, B.; Ikota, N.; Muralidharan, V. B.; Wade, W. S.; Young, S. D.; Yukimoto, Y. *J. Am. Chem. Soc.* **1982**, *104*, 6787–6788.

(14) Trans stereochemistry for the substituents at C_5 and C_6 of **9** was established from the ^1H NMR spectrum of the products from reaction of **9** with 4-phenyl-1,2,4-triazoline-3,5-dione. Two Diels–Alder adducts were formed. For one adduct J = 2.9 Hz for the two H's derived from H_5 and H_6 of **9**. The corresponding J for the other adduct was 1.8 Hz.

(15) **3**·TFA: mp 93–95 °C; IR (KBr) 3600–3250, 1685, 1630 cm^{-1}; UV (H_2O), 280 nm (ϵ 5900); ^1H NMR (CD_3OD) δ 7.37 (1 H, d, J = 5 Hz), 6.47 (2 H, m), 5.63 (1 H, d, J = 3 Hz), 5.07 (1 H, d, J = 3 Hz), 4.53 (1 H, d, J = 6 Hz), the remaining absorption is obscured by the DOH peak.

(16) Salt **3**·TFA is a white, nonhygroscopic powder. The neutral amino acid (hygroscopic) can be obtained by eluting a cold aqueous solution of **3**·TFA through a column of ion-retardation resin AG11A8 (Bio-Rad Corp). The material is most conveniently handled in salt form.

(17) Assay conditions were adapted from Zalkin and Kling (Zalkin, H.; Kling, D. *Biochemistry* **1968**, *7*, 3566–3573) and were performed at 26 °C with a Perkin-Elmer LS-3 Fluorimeter. Buffers were adjusted to be pH 8.6 with or without NH_4^+.

(18) Chorismate was isolated from culture growth of *K. pneumoniae* 62-1 (formerly *A. aerogenes* 62-1) according to: Gibson, F. *Methods Enzymol.* **1970**, *17A*, 362–364. We thank Professor F. Gibson for a generous gift of *K. pneumoniae* 62-1.

(19) Monitored as in ref 17 and also by TLC on silica plates, developed in 80:18:2 ether:hexane:acetic acid and 93:5:2 $CHCl_3$:CH_3OH:CH_3COOH.

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^{57}Fe NMR: Relaxation Mechanisms and Chemical Shifts

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Despite the importance of iron in biological, organometallic, and coordination chemistry, only limited studies of ^{57}Fe NMR have been reported.[2-7] ^{57}Fe, the only isotope of iron suitable for

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