

Regio- and stereo-selectivity in the intramolecular quenching of the excited benzoylthiophene chromophore by tryptophan

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Laser flash photolysis studies on the photobehaviour of a series of bichromophoric derivatives bearing benzoylthiophene and tryptophan groups have shown that the efficiency of the intramolecular quenching process depends on both the stereochemistry of the chiral centers and the relative ketone *versus* tryptophan orientation.

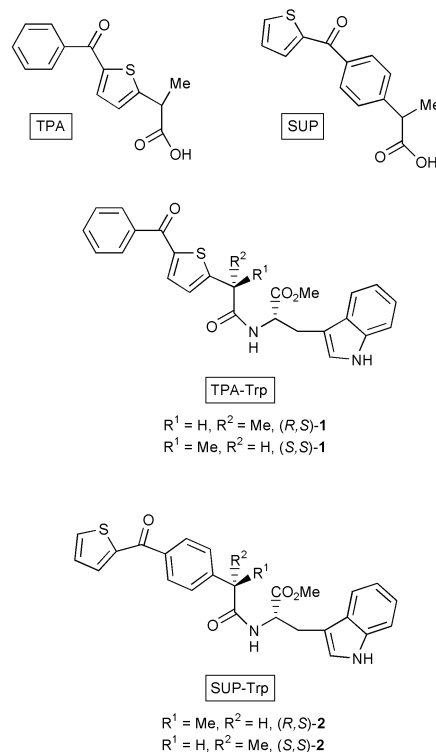
Drug photoallergy is a subject of increasing interest in medicine and involves the formation of drug–protein photoadducts (photoantigens) that may ultimately trigger an immunological response.¹ Tiaprofenic acid (TPA) and suprofen (SUP) are non-steroidal antiinflammatory 2-arylpropionic acids exhibiting high photoallergic activity. Previous studies have established that these drugs sensitize the photooxidation of proteins: His, Tyr and Trp are the reactive amino acid units.^{2,3} His undergoes TPA-photosensitized oxidation according to a Type II (singlet oxygen) mechanism, while in the case of Tyr a Type I (radical) mechanism predominates. Both reaction pathways seem to be operative in the photooxidation of Trp. Thus, only Tyr and Trp can be directly involved in the formation of photoantigens. TPA and SUP drugs share the same 2-benzoylthiophene chromophore, whose excited triplet state is quenched by Tyr and Trp.³ Furthermore, pre-association of these drugs to the protein must play a key role in their photoreactivity, as suggested by the dramatic decrease of the triplet state lifetimes of model bichromophoric compounds TPA-Tyr and SUP-Tyr compared to the parent drugs.⁴ Actually, although pre-association generally involves non-covalent interactions (hydrogen bonding, hydrophobic interactions, *etc.*), the covalently linked bichromophores can provide some insight into the type of effects that may occur upon collision of the two reaction centers. Besides, such bichromophores have the advantage of being well defined chemically. TPA and SUP differ only in the position of the propionic acid side chain linked to the thiophene or the benzene ring. Although their photoreactivity toward the different protein active sites could depend on the way the drug approaches the proteins, such dependence has not been observed in bichromophoric TPA-Tyr and SUP-Tyr compounds.⁴ We report now photophysical studies on the new bichromophoric compounds, TPA-Trp **1** and SUP-Trp **2**, models for the two types of association of the benzoylthiophene chromophore of TPA and SUP to the Trp units in the protein (Scheme 1). Interestingly, laser flash photolysis studies show a high degree of regio- and stereo-selectivity in the intramolecular electron transfer reaction between the excited triplet and tryptophan. To our knowledge, this is one of the few cases of such types of selectivities observed in an intramolecular interaction in the excited state.⁵

Compounds **1** and **2** differ in the relative aromatic ketone *versus* indole orientation. Condensation of racemic TPA or SUP with the methyl ester of the natural occurring (*S*)-tryptophan in the presence of a carbodiimide {(1-ethyl-3-[3-dimethylamino]-

propylcarbodiimide), EDC} led to (*R,S*)-TPA-Trp [(*R,S*)-**1**], (*S,S*)-TPA-Trp [(*S,S*)-**1**], (*R,S*)-SUP-Trp [(*R,S*)-**2**] and (*S,S*)-SUP-Trp [(*S,S*)-**2**]. These amides were purified by column and HPLC chromatography and were fully characterized. The stereochemistry assignments were confirmed by alternative synthesis, using small amounts of the enantiomerically pure TPA and SUP, prepared according to literature procedures.⁶

Leigh *et al.*⁷ have suggested that hydrogen abstraction from phenols by carbonyl π , π^* triplets involves electron transfer within a hydrogen-bonded triplet exciplex, followed by proton transfer. Benzoylthiophene is a heterocyclic diaryl ketone with a lowest lying π , π^* triplet with an energy⁸ of *ca.* 63 kcal mol⁻¹ and is able to react with phenols and indoles.³ Bichromophoric compounds **1** and **2** have the substructures that allow study of the intramolecular electron transfer process between the excited benzoylthiophene chromophore and tryptophan.

Dynamic studies were performed in deaerated methanol using a 355 nm laser (Nd:YAG). Transients with absorption maxima at *ca.* 370 and 580 nm were observed in all cases; they can be assigned to the expected benzoylthiophene triplets.⁸ The



Scheme 1 Chemical structures of tiaprofenic acid (TPA), suprofen (SUP) and the bichromophores TPA-Trp **1** and SUP-Trp **2**.

Table 1 Lifetimes and rate constants for quenching of the triplet states of bichromophoric compounds **1** and **2**^a

Compound	Lifetime τ /ns	$k_{ET}^b/10^5 \text{ s}^{-1}$
(<i>R,S</i>)- 1	460	18
(<i>S,S</i>)- 1	92	105
(<i>R,S</i>)- 2	440	19
(<i>S,S</i>)- 2	841	8
2-Benzoylthiophene	2800	

^a Concentrations: **1**: $8 \times 10^{-4} \text{ M}$, **2**: $7 \times 10^{-4} \text{ M}$, 2-benzoylthiophene: $5 \times 10^{-4} \text{ M}$. ^b Electron-transfer rate constants are obtained by using the equation $k_{ET} = (1/\tau_1) - (1/\tau_2)$, where τ_1 and τ_2 are the lifetimes of the ketone triplets in the bichromophoric compound and 2-benzoylthiophene, respectively.

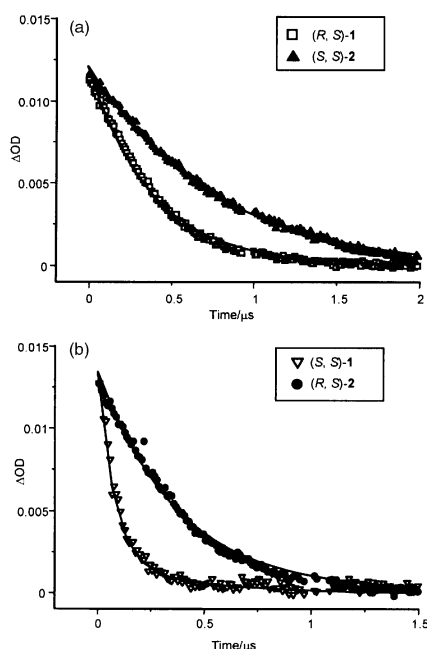


Fig. 1 Comparative decay of the triplets of regioisomers (*R,S*)-**1** and (*S,S*)-**2** (a) and (*S,S*)-**1** and (*R,S*)-**2** (b) monitored at 360 nm.

transient generated upon laser excitation of the TPA or SUP moieties was quenched differently by the (*S*)-tryptophan moiety. The regioselectivity, as shown by the electron-transfer rate constant ratios for pairs with the same relative configuration,⁹ was: 5.5 for (*S,S*)-**1**/*(R,S)*-**2** and 2.25 for (*R,S*)-**1**/*(S,S)*-**2** (Table 1, Fig. 1). On the other hand, the stereoselectivities were 5.8 for (*S,S*)-**1**/*(R,S)*-**1** and 2.4 for (*R,S*)-**2**/*(S,S)*-**2**. It is also noteworthy that the intramolecular electron-transfer process in compounds **1** and **2**, containing the indolic group, is slower than in the analogous bichromophores with a tyrosine unit as quencher; by contrast, the rate of intermolecular quenching of the triplet state of benzoylthiophene chromophore by Trp methyl ester is higher than by Tyr methyl ester.³

It has been suggested that the optimum geometry for hydrogen abstraction from phenols by carbonyl π , π^* triplets is a 'sandwich-like' hydrogen-bonded exciplex, involving an in-plane arrangement of the O–H bond and the carbonyl *n*-orbital.^{7,10} In compounds **1** and **2**, the N–H bond is held in the plane of the aromatic indolic group, whereby no hydrogen bond can favour the face-to-face geometry. The observed decrease of the ketone triplet state lifetime in these bichromophores can then be interpreted as due to the involvement of non-bonding electronic interactions between aromatic π -systems of the ketone and indole groups in the quenching process. Overall, the most remarkable results are the dramatic dependence of the photobehaviour on the absolute configuration of the drug and the relative orientation of the chromophores in compounds **1**

and **2**. The latter was not observed in the analogous Tyr derivatives, where the major factor determining the approach of the chromophores is the formation of a hydrogen bond between the C=O and the phenolic OH; as this arrangement can be attained for both TPA-Tyr and SUP-Tyr, no regioselectivity was observed. However, the data obtained for compounds **1** and **2** show the strong influence of the non-bonding electronic interactions even when no hydrogen bonded exciplex can be formed. It seems that the most favoured approach of the quencher (indole group) to the excited chromophore involves the thienyl side of the ketone.

It is known that the lowest lying (π , π^*) triplet state of 2-benzoylthiophene involves predominantly the thienyl ring⁸ and has a considerable charge transfer character. A plausible explanation for the results obtained in the present work can be based on a more efficient π -stacking interaction between the π -electron cloud of the indolic moiety and the polarized thienyl group of **1**, which favours the approach of both chromophores and accelerates the electron-transfer process.

In summary, the efficiency in the interchromophoric quenching processes suggests that pre-association of TPA and SUP to Trp units in the protein might be important. On the other hand, the different lifetimes of ³TPA-Trp and ³SUP-Trp compounds reveal a high degree of regio- and stereo-selectivity in the intermolecular quenching of the excited ketone. This behaviour suggests that the position of the propionic acid chain plays a major role in the approach of the drug to the Trp moiety in the protein.

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