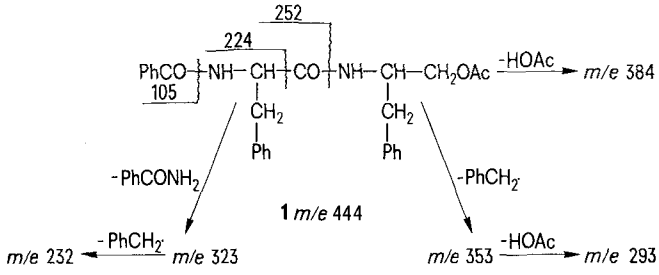
and to the NH proton at δ 6.88, while the second methine proton at δ 4.34 was coupled to the other 2 methylene groups and the other NH proton. Confirmatory evidence for all these carbon-containing groups was provided by the ^{13}C NMR-spectrum which also established the presence of 3 carbonyl groups (170.71, 170.41, and 167.17 ppm). These data suggested structure 1 which was supported by the mass spectral fragmentation as indicated.



Structure 1 was confirmed by synthesis from L-phenylalanyl-L-phenylalanine. The methyl ester hydrochloride, m.p. 197–198°, $[\alpha]_D$ = +4.55 (c, 0.91; MeOH), prepared in the usual way[5], was treated with benzoyl chloride in cold pyridine for 30 min. After removal of solvent in vacuo and trituration with ether, the residue crystallized from chloroform-ether to give the *N*-benzoyl methyl ester as needles, m.p. 177–178°, $[\alpha]_D$ = +11.5° (c, 1.08; $CHCl_3$), M^+ 430.1894 ($C_{26}H_{26}N_2O_4$ requires M, 430.1892); δ ($CDCl_3$) inter alia 6.56 and 6.86 (each 1H, d, exchanged with D_2O, 2NH). This ester was reduced with lithium borohydride[6] (2 mols) in dry THF for 30 min in the cold to form the alcohol 1 (OH in place of OAc) which crystallized from chloroform in plates, m.p. 174–175°, $[\alpha]_D$ = –51.1° (c, 1.42; $CHCl_3$), M,+ 402.1945 ($C_{25}H_{26}N_2O_3$ requires M, 402.1943); ν_{max} (KBr) 3400 (OH), 3292 (NH) cm^{-1}. A longer reduction time with $LiBH_4$ led to some degree of racemization. Final acetylation in cold pyridine yielded the acetate 1, m.p. 185–186° (from chloroform-petrol) identical (UV, IR, NMR, MS, CD, $[\alpha]_D$, and mmp) with the natural dipeptide.

Added in proof: Since this Note was submitted the same compound, named asperglaucide, has been reported in *Aspergillus glaucus*.[7]

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Oxygenation of a Dipyrromethene Model for Bilirubin: Formation of a Singlet Oxygen-like Product

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Summary. An oxodipyrromethene model compound for bilirubin is found to undergo oxidation to a blue tetrapyrrole and a water-propentdyopent on a silica gel thin layer chromatography plate. The reaction involves ground state oxygen and requires silica gel, although the propentdyopent is an expected product from reaction with singlet oxygen.

In connection with a widely employed phototherapy for neonatal jaundice due to unconjugated hyperbilirubinemia[2,3], we have been investigating the photooxygenation of bilirubin IXα (BR)[4,5] and various model compounds[6], especially 5′-oxo-3′, 4, 4′-triethyl-3, 5-dimethyl-1′, 5′-dihydro-(2·2′)-dipyrromethene (**1**)[7]. Most of the photochemical investigations of BR reported to date have been in vitro studies in solution[4,8,9], with the exception of some recent work on BR in micelles[10]. Thus environmental effects on the photochemistry have not been studied extensively, and there are only a few reported studies which mimic the environment of (aggregated) BR deposited in the skin[10–12], the presumed major photo-active site. We have studied the reactions of **1**, which serves as a convenient model for one half of BR, in the aggregated state deposited on thin layers of silica gel and alumina.

Materials and methods. Oxodipyrromethene (**1**) was prepared by the base-catalyzed condensation of krypto-

[1] The authors wish to thank the National Science Foundation for generous support of this work. We also thank Mr. Richard Gillespie and Ms. Elizabeth Irwin for running the high resolution mass spectra.

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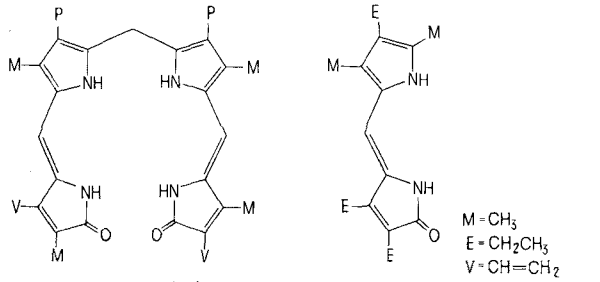
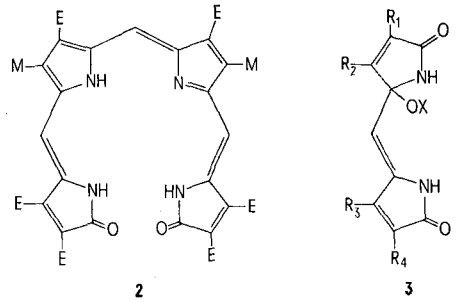
pyrrole aldehyde and 3,4-diethyl-3-pyrrolin-2-one[7,13], and gave satisfactory elemental analyses and spectroscopic properties[13]. The spectroscopic properties on all new substances were determined as follows: mass spectra on an AEI MS-9 mass spectrometer, NMR-spectra on a JEOL 4H-100 spectrometer, IR-spectra on a Beckman IR-8 instrument and UV-Visible spectra on a Cary 14 spectrophotometer. The silica gel used in this work was silica gel F (M. Woelm, Eschwege), and the alumina was from E. Merck, Darmstadt.

Thin layer chromatography (TLC) plates were prepared as follows: analytical (0.125 mm) – silica gel or alumina was coated as an aqueous slurry using distilled, deionized water, and plates were activated in an oven at 110° for 2–3 h; preparative (1.0 mm) – 95% ethanol slurries of adsorbent were coated, and the plates were activated at 110° for 2–3 h. The methanol and chloroform used in this work was distilled reagent grade Baker Analyzed. Ethanol (95%) was Gold Shield. DABCO, hydroquinone and 2,6-di-t-butylphenol were obtained from Aldrich Chemicals. The nitrogen used was ultra high purity Matheson (99.999%).

For these studies, **1** was either spotted or streaked as a $CHCl_3$ solution on a TLC plate in the dark and, in some cases, in the absence of air. When DABCO, hydroquinone or 2,6-*di-t*-butylphenol were admixed, this was done to the $CHCl_3$ solution of **1** before spotting. When spotted TLC plates were photo-irradiated, a Westinghouse 500 W Quartz-Iodine lamp, No. 500 Q/CL, 120 V, run at 80 V and placed in a water-cooled pyrex jacket was used. Samples were held at a distance of 5–10 cm from the lamp.

Results and discussion. When yellow **1** was spotted on a silica gel F TLC plate and allowed to stand in the dark and in the air, the spot turned green[14]. New compounds were formed as shown after solvent ($CHCl_3$:CH_3OH/95:5) development of the TLC plate: the green spot resolved principally into yellow, unchanged **1** [Rf 0.50], a blue substance (**2**) [Rf 0.64] and a colorless substance (**3**) [Rf 0.03]. The former (**2**) proved to be a mesobiliviolin type tetrapyrrole; whereas, the latter (**3**) was shown to be a propentdyopent by a combination of spectroscopic techniques.





The brilliant, deep-blue tetrapyrrole (**2**) isolated in 18% yield from a preparative plate, mp 252–3° after crystallization from CH_3OH, was characterized by its mass spectrum: *m/e* (relative intensity) 526.3313 [M^+, $C_{33}H_{42}N_4O_2$] (61%), 497 (13%), 271 (32%), 258 (100%), 243 (52%) atome mass units (AMU); NMR-spectrum: δ ($CDCl_3$) 1.08 (t, J=7.5Hz, 6H, CH_2CH_3), 1.24 (t, J=7.5Hz, 12H, CH_2CH_3), 2.07 (s, 6H, CH_3), 2.28 (q, J=7.5Hz, 4H, CH_2CH_3), 2.55 (q, J=7.5Hz, 8H, CH_2CH_3), 5.90 (s, 2H, =C–H), 6.64 (s, 1H, =C–H), 8.05 (bs, 3H, NH) ppm; and UV-Visible spectrum ($CHCl_3$): λ_{max} 630 nm (ε 12,000), 370 nm (ε 41,000), and in $CHCl_3$ with 30 mg/100 ml *p*-toluenesulfonic acid: λ_{max} 670 nm (ε 27,000), 366 nm (ε 46,000) and 293 nm (ε 14,000). Its striking deep-blue color in $CHCl_3$ solution is essentially the same as that of the closely related octaethylbilatriene-*abc*[15].

The dipyrrole **3** (9% isolated yield from a preparative TLC plate) was shown to be a very sensitive water-propentdyopent (λ_{max} 517 as pentdyopent)[16] and had λ_{max} 277 nm (ε 19000) in CH_3OH. Because of its instability, it did not give a clean mass spectrum, and the NMR-spectrum; δ ($CDCl_3$) 1.10 (m, 9H, CH_2CH_3), 1.84 (s, 1.70H, CH_3), 1.93 (s, 1.30H, CH_3), 2.28 (m, 6H, CH_2CH_3), 4.80 (s, 1H, =CH), 5.7 (bs, 1H, OH), 7.50 (bs, 1H, NH) and 8.75 (bs, 1H, NH) ppm indicated that **3** was probably a mixture of allylic isomers (**3**, X=H, R^1=R^3=R^4=E, R^2=M and **3**, X=H, R^1=R^2=R^4=E, R^3=M).

The water-propentdyopent (mixture) was converted to an inseparable mixture of the corresponding methanol-propentdyopents (**3**, X=M, R^1=R^3=R^4=E, R^2=M and **3**, X=M, R^1=R^2=R^4=E, R^3=M) through the action of CH_3OH and a trace of *p*-toluenesulfonic acid. This mixture of isomers was characterized by its mass spectrum: *m/e* (relative intensity) 304.1787 [M^+, $C_{17}H_{24}N_2O_3$] (7%), 289 (9%), 275 (100%), 273 (30%), 257 (18%), 243 (21%), 229 (13%), 215 (18%), 149 (19%) and 138 (17%) AMU; NMR-spectrum: δ ($CDCl_3$) 1.12 (t, J=7.5Hz, 9H, CH_2CH_3), 1.80 (s, 2H, CH_3), 1.96 (s, 1H, CH_3), 2.3 (q, J=7.5Hz, 6H, CH_2CH_3), 3.15 (s, 3H, OCH_3), 4.68 (s, 1H, =C–H), 6.1 (bs, 1H, NH) and 8.25 (bs, 1H, NH) ppm; and UV-spectrum: $\lambda_{max}^{CH_3OH}$ 277, ε 6100); pentdyopent: λ_{max} 517 nm.

It may be noted that both **2** and **3** are formed on a silica gel plate in the presence or absence of light, but not without O_2 (N_2 atmosphere) and not on an acidic or basic alumina TLC plate. Thus, a silica surface appears to exhibit a unique effect on the reactions of **1**, and O_2 is clearly necessary for the formation of **2** and **3**, but light is not. As nearly as we can tell, the formation of **2** is accelerated with light, but the formation of **3** is insensitive to it. The dark reactions leading to **2** and **3** are completely arrested when **1** is spotted on the plate as a solution with 2,6-*di-t*-butylphenol, hydroquinone or DABCO. The former are good radical traps or radical reaction suppressors. Under the 3 'protective' conditions, the formation of **2** is repressed even in the presence of light; however the formation of **3** is prevented only with hydroquinone during the light irradiation. Although the reaction **1** → **3** is reminiscent of singlet oxygen ($^1O_2$) reactions with bilirubin[4–10,17] and pyrroles[4], $^1O_2$ clearly

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cannot be involved in the dark reactions and is probably not involved in the photo reactions either. It is especially curious that the seemingly logical reaction product (**3**) of $^1O_2$ with **1** is at best only a very minor product, if it occurs at all, in the Rose Bengal-sensitized photooxygenation of **1**[7]. There are very few instances reported where ground state oxygen ($^3O_2$) is directly implicated in what appear to be (or are) $^1O_2$ reaction products. One such example is the formation of ergosterol acetate *endo*-peroxide with $^3O_2$ and $(p\text{-}BrC_6H_4)_3N^+$[18]. The reaction of **1** → **3** on silica gel may be another.

The mechanism of formation of **3** is unclear, but a radical pathway appears to be implicated. Silica gel is known to catalyze oxidations of catechol and pyrogallol[18] and induce esr signals in adsorbed aromatic hydrocarbons[20]. It is also thought to give O_2^- in adsorbed O_2[21, 22]. The mechanism of formation of **2** is also unclear. Tetrapyrrole **2** is not obtained during the self or dyesensitized photooxygenation of **1**. Further work is in progress to elucidate the role of silica gel in these reactions and to investigate the chemistry and photochemistry of adsorbed and aggregated bilirubin.

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The Chemical Structure of Capreomycin

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Summary. The chemical structure of capreomycin, antituberculous peptide antibiotic, was revised from the results of NMR-analysis in comparison with tuberactinomycins. Capreomycin IA and IB were concluded to possess the similar amino acid sequences in their cyclic peptide moieties to those of tuberactinomycins.

The antituberculous peptide, capreomycin, was first isolated from *Streptomyces capreolus* in 1960 by Herr[1]. This antibiotic has a similar structural, as well as biochemical character, to that of viomycin, which was found in 1951[2, 3] and is in practical use as chemotherapic agent. Subsequently, capreomycin was shown to be a mixture of 4 related compounds which are named capreomycin IA, IB, IIA, and IIB[4]. The only difference in composition between I and II is attributed to the absence of β-lysine in the latter; B differs from A in exchange of 1 amino acid component, serine with alanine, having otherwise the same composition in the rest of amino acid residues.

Inspite of somewhat earlier isolation of this antibiotic, its structural elucidation for the whole molecule had not proceeded until a proposal by Bycroft et al.[5]. They deduced the structure of β-Lys→Dpr→Cpd→Ala→Dpr→Uda—[6] for capreomycin IB from some chemical evidence and an analogy to the tentative structure of viomycin, β-Lys→Dpr→Tbd→Ser→Ser→Uda—[6], which was proposed by the same authors in 1971[7].

However, shortly after this proposal, the structure of viomycin was revised as β-Lys→Dpr→Ser→Ser→Uda→Tbd—[6] by our studies on tuberactinomycin[10, 12], which is a similar antibiotic group including viomycin as one congener. This conclusion was subsequently supported by X-ray analysis by Bycroft[13]. If the similarities between capreomycin and tuberactinomycin including viomycin are accounted for in chemical, physical, and biological features, the structure of capreomycin proposed formerly might involve the wrong amino acid sequence.

In our recent studies on tuberactinomycins, the NMR-spectra of natural compounds, as well as the cyclic peptide moiety of tuberactinomycin N and O, *i.e.*, tuberactinamine N, were successfully analyzed[14]. In all the spectra, two Ser residues in the positions 3 and 4 showed significant differences in the chemical shifts and coupling patterns at α-methine, β-methylene, and α-amide protons. The results could be summarized as follows: a) the chemical shift of α-methine proton of position 3 is remarkably lower than that of position 4; b) β-methylene protons of position 3 appear as a magnetically equivalent doublet, while those of position 4 are manifested as magnetically non-equivalent two quartets; c) α-amide proton of position 4 is observed in a lower field than that of position 3. Similar phenomena were also recognized in spectra of synthetic tuberactinamine analogs, *i.e.*,

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