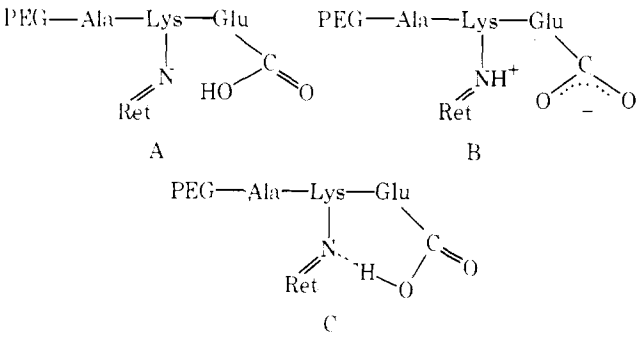
trum becomes more intense in its long-wavelength region (550–700 nm) where the formally protonated RSB has its emission maximum. The excitation spectrum monitored at the observed emission maximum (550–590 nm) of RSB's of peptides III and IV is composed of two distinct bands, one with a maximum at 340–440 nm (strong) and the other at 440–500 nm (weak), Figure 1.

Both the absorption and emission spectral data described above clearly indicate that, in the case of the RSB's of the peptides III and IV, more than one well-defined species is present in solution. The two bands observed in the excitation spectra are identifiable in position with the main absorption band of the nonprotonated and formally protonated forms. The latter data plus that of absorption and emission regarding the long-wavelength regions, vide supra, strongly indicate that the RSB's of the peptides III and IV contain at least two distinct species which we shall formally call nonprotonated (A) and protonated (B). A third viable possibility is an intramolecular



hydrogen-bonded form (C) which is expected to have spectral properties similar to those of B. Protonation could occur by dissociation of the H of the carboxyl group leading to the formation of a zwitterion involving the basic imino nitrogen of the Schiff base. H-bond formation between the different molecules is expected to be much less significant because of the expected large steric hindrance involved in the approach of the macromolecules to one another.

The question of whether a completely protonated form (B) or internally H-bonded form (C) (or possibly an intermolecularly H-bonded form) is predominantly involved remains to be settled by other means. However, in a relatively nonionizing solvent such as dichloromethane and at low temperature, the dissociation of the carboxyl group is expected to be small and the formation of an internally H-bonded species such as C is most likely. *To the best of our knowledge, this work represents the first study and observation of a molecular perturbation of known origin involving a peptide moiety of known sequencing that results in a significant alteration of the wavelength maximum of an RSB.* The presence of internal H-bonding protonation indicated in the present work may be exploited to resolve some of the issues involved in the mechanism of the visual processes including protonation–deprotonation.[8-10,12,13] In addition to the protein-like microenvironment offered by the polymeric PEG portion, it is possible for further study to have peptides containing suitable polar, polarizable, or aromatic groups which may take part in internal electrostatic or charge-transfer interactions with the polyene chain.[4,14,15]

It should be noted that, although the emission spectral maxima of the RSB's of peptides I and II show a *slight* dependence on excitation wavelengths, this behavior is much less pronounced compared with that of III (and IV) and is not explainable in terms of absorption of or emission from protonated–nonprotonated species.

The preparation of the RSB complexes was done by mixing dry methanol solutions of a PEG peptide and *all-trans*-retinal (Sigma, five–six times molar excess) over 3A molecular sieves and standing for 12 h at 0 ° under nitrogen in the dark. After the molecular sieves were removed by filtration, the RSB complex was precipitated from the methanol solution by adding ether (until the solution was opalescent) and cooling to 0 °C. This was filtered off, washed three times with cold, dry ether, and dried under high vacuum ($\sim 10^{-5}$ Torr). For spectral measurements, the RSB was dissolved in methanol followed by precipitation with ether, washing, and drying and this procedure was repeated twice to ensure the complete removal of unreacted retinal.

Acknowledgment. This research was supported by a grant from the Robert A. Welch Foundation and the North Atlantic Treaty Organization, No. 1076.

References and Notes

(1) R. S. Becker, G. Hug, P. K. Das, A. M. Schaffer, T. Takemura, N. Yamamoto, and W. Waddell, *J. Phys. Chem.*, **80**, 2265 (1976).
(2) B. Honig and T. G. Ebrey, *Annu. Rev. Biophys. Bioeng.*, **3**, 151 (1974).
(3) B. Honig, A. Warshel, and M. Karplus, *Acc. Chem. Res.*, **8**, 92 (1975).
(4) W. H. Waddell, A. M. Schaffer, and R. S. Becker, *J. Am. Chem. Soc.*, **99**, 8456 (1977), and references therein.
(5) B. Honig, A. D. Greenberg, U. Dinur, and T. G. Ebrey, *Biochemistry*, **15**, 4593 (1976).
(6) P. E. Blatz and J. H. Mohler, *Biochemistry*, **11**, 3240 (1972); P. E. Blatz, J. H. Mohler, and H. V. Navangul, *ibid.*, **11**, 848 (1972).
(7) H. J. A. Dartnall, *Vision Res.*, **8**, 339 (1968); A. Alchalel, B. Honig, M. Ottolenghi, and T. Rosenfeld, *J. Am. Chem. Soc.*, **97**, 2161 (1975).
(8) C. R. Goldschmidt, M. Ottolenghi, and T. Rosenfeld, *Nature (London)*, **263**, 169 (1976); T. Rosenfeld, B. Honig, M. Ottolenghi, J. Hurley, and T. G. Ebrey, *Pure Appl. Chem.*, **49**, 341 (1977).
(9) K. van der Meer, J. J. C. Mulder, and J. Lugtenburg, *Photochem. Photobiol.*, **24**, 363 (1976); A. Kropf, *Nature (London)*, **264**, 92 (1976).
(10) Y. Shichida, T. Kobayashi, H. Ohtani, T. Yoshizawa, and S. Nagakura, *Photochem. Photobiol.*, **27**, 335 (1978).
(11) E. Bayer and M. Mutter, *Nature (London)*, **237**, 512 (1972); *Angew. Chem., Int. Ed. Engl.*, **13**, 88 (1974). M. Mutter, R. Uhmann, and E. Bayer, *Justus Liebigs Ann. Chem.*, 901 (1975).
(12) A. M. Schaffer, T. Yamaoka, and R. S. Becker, *Photochem. Photobiol.*, **21**, 297 (1975).
(13) J. Favrot, J. M. Leclercq, R. Roberge, C. Sandorfy, and D. Vocelle, *Chem. Phys. Lett.*, **53**, 433 (1978).
(14) M. Muthukumar and L. J. Weimann, *Chem. Phys. Lett.*, **53**, 436 (1978).
(15) D. S. Kliger, S. J. Milder, and E. A. Dratz, *Photochem. Photobiol.*, **25**, 277 (1977).

P. K. Das, Ralph S. Becker*
Department of Chemistry, University of Houston
Houston, Texas 77004

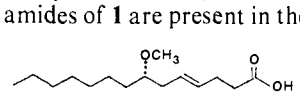
Dieter Hannak, Ernst Bayer
Institut fur Organische Chemie, Universitat Tubingen
7400 Tübingen 1, West Germany

Received May 19, 1978

Malyngamide A, a Novel Chlorinated Metabolite of the Marine Cyanophyte *Lyngbya majuscula*

Sir:

(−)-7(*S*)-Methoxytetradec-4(*E*)-enoic acid (**1**) and chlorine-containing amides of **1** are present in the extracts of sev-



1

eral shallow-water varieties of the marine blue-green alga *Lyngbya majuscula* Gomont.[1-3] Herein we report the structure of one of these amides, malyngamide A.

Malyngamide A (**2**) was obtained as a neutral, colorless oil, $[\alpha]^{25}_D$ −6.5° (CH_2Cl_2, *c* 0.8) by gel filtration (Sephadex LH-20) and HPLC (μ-Bondapak-CN) of the dichloromethane extract of freeze-dried *L. majuscula* collected at Kahala Beach, Oahu. High resolution mass spectrometry established the molecular formula of **2** as $C_{29}H_{45}ClN_2O_6$. Negative re-

0002-7863/79/1501-0240$01.00/0

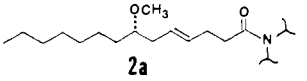
Table I. Spectral Data for Malyngamide A (**2**), Dysidin (**6**), and Hydrolysis Products

	2	**5**[a]	**6**	**7**
^{1}H NMR (δ, ppm)				
C-2	6.85	3.99	6.84	4.12, 3.87[b]
C-4	3.52	3.33	3.15, 3.47[b]	
OMe on C-3	3.74		3.76	
UV (λ_{max}, nm (ϵ))				
in EtOH	265 (17 600)	240 (13 000)[c]	264 (26 900)	
	213 (15 700)	214 (16 200)[c]	225 (15 900)	
in basic EtOH		310 (21 000)		
		219 (37 000)		

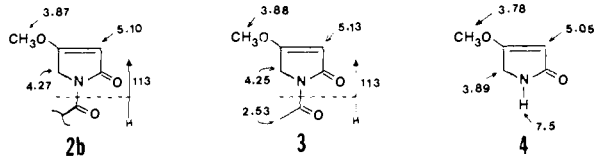
[a] High resolution mass spectrum *m/e* 538.2801 (M^+; calcd for $C_{28}H_{43}N_2O_6{}^{35}Cl$, 538.2809). [b] AB quartet; all other signals in this table are singlets. [c] Similar to UV of **3** in EtOH: λ_{max} 238 nm (ϵ 11 000), 218 (10 000).

actions with $AgNO_3$ in refluxing EtOH and with NaI in acetone indicated that **2** was an alkenyl chloride.

Partial structure **2a** was inferred from ^{1}H NMR spectral data[4] and was confirmed by alkaline hydrolysis of **2** to **1**, $[\alpha]^{26}_D$ −10.0° ($CHCl_3$, *c* 0.5). Similarly partial structure **2b** was concluded by comparison of the ^{1}H NMR and mass spectral data of malyngamide A and **3**, mp 89–90 °C, a minor constituent of one or more polar fractions of *L. majuscula*. Verification of this moiety was secured by alkaline hydrolysis of **2** to **4**.[5] Compound **4**, mp 133–134 °C, was also a constituent of the cyanophyte.

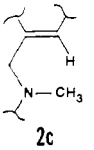


2a



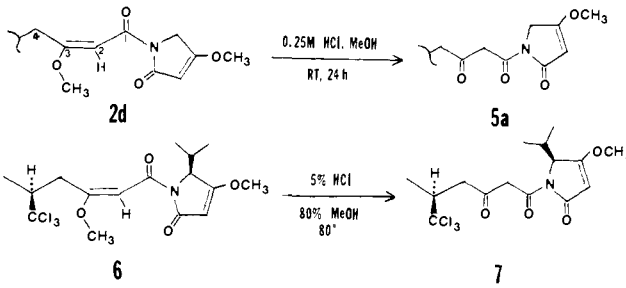
2b **3** **4**

Three absorptions in the ^{1}H NMR spectrum of malyngamide A were doubled in a 2:1 ratio (δ 2.90/2.84, 4.33/4.18, 6.09/6.18) owing to two slowly interconverting conformers. At 100 °C (Me_2SO-d_6) these three pairs of signals coalesced to 3 H, 2 H, and 1 H singlets at δ 2.83, 4.19, and 6.19, respectively. Irradiation of the 3 H signal, assigned to an *N*-methyl group, produced a 9% positive NOE in the 1 H olefinic signal and a 5% negative NOE in the 2 H methylene signal. These data implied that malyngamide A had partial structure **2c.**



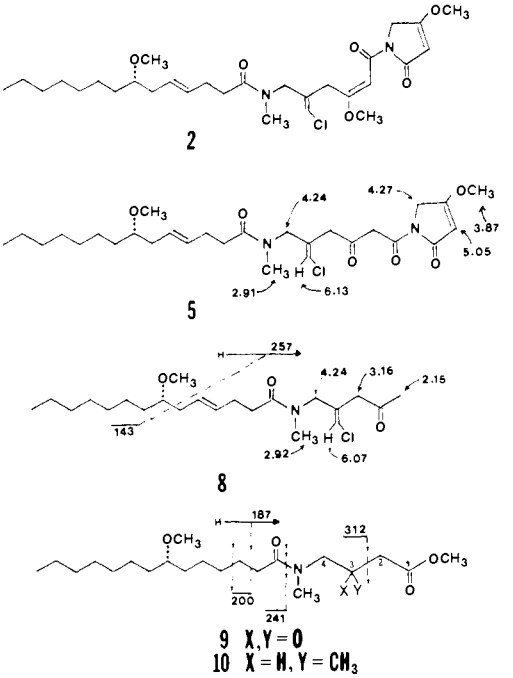
2c

Partial structure **2d** was suggested by comparison (Table I) of the UV and ^{1}H NMR spectra of malyngamide A, the sponge metabolite dysidin[6,7] (**6**), and the β-keto amides **5** and **7** from mild acid hydrolysis of **2** and **6**. An NOE experiment showed that the OMe on C-3 and the olefinic proton of C-2 in **2d** were cis to each other, since irradiation of the methoxyl signal at δ 3.74 gave an 18% increase in intensity for the olefinic proton signal at δ 6.85.



2d **5a** **6** **7**

The spectral and chemical data above were consistent with structure **2** for malyngamide A. Proof of this structure was disclosed from the following chemical degradations.[8] Treatment of **2** with acid (2 N HCl, 75% MeOH, reflux 6 h) produced a β-keto acid which decarboxylated to the methyl ketone **8**.[9] Interestingly the alkenyl chloride functionality of **8** was not



2 **5** **8** **9** X,Y = O; **10** X = H, Y = CH_3

altered during the acid hydrolysis.[9] Selective catalytic hydrogenation of the tetradecenoyl unit in **2** (Pd/C, EtOAc) and subsequent ozonolysis ((1) O_3, CH_2Cl_2, −5 °C; (2) Ph_3P) led to the β-keto methyl ester **9**.[10] A small amount of methyl ester **10**[11] was also formed, obviously from concomitant hydrogenation of the alkenyl chloride functionality prior to ozonolysis.

Acknowledgment. This work was supported by Grant No. CHE76-82517 from the National Science Foundation.

References and Notes

(1) J. H. Cardellina II, D. Dalietos, F.-J. Marner, J. S. Mynderse, and R. E. Moore, *Phytochemistry*, in press.
(2) Stylocheilamide and deacetylstylocheilamide are two nontoxic chlorine-containing 7-methoxytetradec-4(*E*)-enoic amides (absolute configurations unknown) that have been isolated from the sea hare *Stylocheilus longicauda*: A. F. Rose, P. J. Scheuer, J. P. Springer, and J. Clardy, *J. Am. Chem. Soc.*, **100**, 7665–7670 (1978).
(3) None of these compounds are responsible for the antibiotic activity of Hawaiian *Lyngbya majuscula*: T. J. Starr, E. F. Dieg, K. K. Church, and M. B. Allen, *Texas Rep. Biol. Med.*, **20**, 271–278 (1962).

(4) ^{1}H NMR spectra were determined in chloroform-*d* unless noted otherwise.
(5) D. J. Cram, O. Theander, H. Jager, and M. K. Stanfield, *J. Am. Chem. Soc.*, **85**, 1430–1437 (1963).
(6) W. Hofheinz and W. E. Oberhänsli, *Helv. Chim. Acta*, **60**, 660–669 (1977).
(7) Dysidin was isolated from the sponge *Dysidea herbacea*. Interestingly blue-green algae are associated with this sponge.
(8) Using the program CONGEN 24 gross structures were assembled from the following fragments: R_1 = **2b**, R_2 = **2a** with one Me group on the nitrogen, two XCH_2X groups with no protons on the X atoms, one >C=CH– group, one MeO—C=CH– group, and a chlorine atom. Only one of these 24 structures satisfactorily explains the degradation of malyngamide A to **8**, **9**, and **10**. We thank T. Varkony and C. Djerassi for this determination.
(9) Oil; mass spectrum *m/e* (rel intensity) 401 (1), 399 (4, M^+), 384 (3), 364 (11), 332 (8), 315 (7), 259 (26), 257 (75), 222 (22), 205 (20), 203 (57), 158 (35), 146 (53), 143 (100), 111 (41); high resolution mass spectrum *m/e* 399.2503 (calcd for $C_{22}H_{38}{}^{35}ClNO_3$, 399.2540); UV (MeOH) λ_{max} 212 nm (ϵ 3900) assigned to the $\pi \rightarrow \pi^*$ transition for the β,γ-unsaturated ketone carbonyl; IR (neat) 1718, 1655, 980 cm^{-1}; ^{1}H NMR δ 6.07 (1 H, br s), 5.50 (2 H, br t), 4.24 (2 H, br s), 3.33 (3 H, s), 3.16 (2 H, s, on 1 H, br quintet), 2.92 (3 H, s), 2.34 (4 H, br m), 2.15 (3 H, s, on 2 H, br m), 1.26 (12 H, br s with low-field sh), 0.88 (3 H, br t, J = 7 Hz). Two signals in the ^{1}H NMR spectrum are doubled in a 6:1 ratio (δ 6.07/6.11, assigned to =CHCl, and 2.92/2.83, assigned to the NCH_3 for the two conformers); irradiation at δ 2.92 produces a 19% positive NOE in the signal at δ 6.07 and a 5% negative NOE in the methylene signal at δ 4.24.
(10) Oil; mass spectrum *m/e* (rel intensity) 385 (>1), 312 (2), 241 (12), 200 (23), 187 (19), 143 (25); high resolution mass spectrum *m/e* 385.2838 (M^+; calcd for $C_{21}H_{39}NO_5$, 385.2828), 312.2511 (calcd for $C_{18}H_{34}NO_3$, 312.2539), 241.2173 (calcd for $C_{15}H_{29}O_2$, 241.2168), 200.0931 (calcd for $C_9H_{14}NO_4$, 200.0923), 187.0847 (calcd for $C_8H_{13}NO_4$, 187.0845); UV (MeOH) λ_{max} 213 nm (ϵ 5400) → 271 (11 000), 214 (7900) in methanolic NaOH; IR 1735, 1720 (sh), 1650 cm^{-1}; ^{1}H NMR δ 4.24 (2 H on C-4, s), 3.71 (3 H, s, ester *OMe*), 3.46 (2 H on C-2, *s*), 3.28 (3 H, s), 3.05 (1 H, br quintet), 3.02 (3 H, s, *N*-Me), 2.30 (2 H, br t, J = 7 Hz), 1.65–1.20 (20 H, br m), 0.83 (3 H, br t, J = 7 Hz).
(11) Identified by high resolution mass spectrum: *m/e* 385.3169 (M^+, calcd for $C_{22}H_{43}NO_4$, 385.3192), 312.2874 (calcd for $C_{19}H_{38}NO_2$, 312.2903), 241.2173 (calcd for $C_{15}H_{29}O_2$, 241.2168), 200.1296 (calcd for $C_{10}H_{18}NO_3$, 200.1287), 187.1198 (calcd for $C_9H_{17}NO_3$, 187.1208).

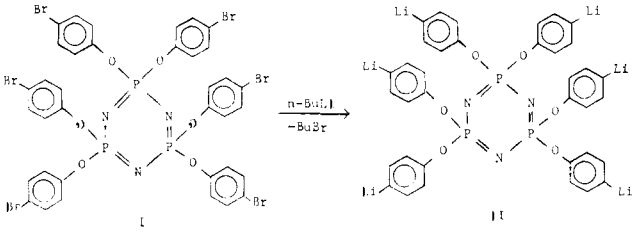
John H. Cardellina II
Franz-Josef Marner, Richard E. Moore*
Department of Chemistry, University of Hawaii
Honolulu, Hawaii 96822
Received June 19, 1978

Preparation of $[NP(p\text{-}OC_6H_4Li)_2]_3$ by Metal–Halogen Exchange, and Its Reactions with Electrophiles

Sir:

The reactions of cyclic and polymeric *halo*phosphazenes with organolithium reagents have been studied extensively,[1-3] but the reactions of organometallic reagents with cyclic and polymeric *organo*-functional phosphazenes have not been explored in detail. Of particular interest to us were reactions that could yield carbanionic species bound directly to phosphazene cyclic and polymeric compounds. Such reactive intermediates could be used to synthesize a wide range of new cyclic and high polymeric phosphazenes not accessible by other synthetic routes, including those that might form unusual ligands for transiton metals.

We have found that hexa(*p*-bromophenoxy)cyclotriphosphazene (I) undergoes a high yield metal–halogen exchange reaction with *n*-butyllithium to yield the hexalithio derivative



I II

(II). The reaction conditions employed involved a rapid addition of *n*-butyllithium (1.6 M in hexane) in a 15% excess to a tetrahydrofuran solution of I at −40 °C.

The presence of II was confirmed by its reactions at −40 °C with electrophiles, such as deuterium oxide, carbon dioxide, chlorodiphenylphosphine, and triphenyltin chloride to yield the following derivatives: $[NP(p\text{-}OC_6H_4D)_2]_3$ (III), $[NP(p\text{-}OC_6H_4COOH)_2]_3$ (IV), $\{NP[p\text{-}OC_6H_4P(C_6H_5)_2]_2\}_3$ (V), and $\{NP[p\text{-}OC_6H_4Sn(C_6H_5)_3]_2\}_3$ (VI). All of these compounds were identified by ^{31}P NMR spectra, infrared spectra, and chemical analysis. The position of lithium incorporation on the aromatic ring was confirmed by the ^{13}C NMR spectrum of compound III which revealed both the presence of a triplet structure and a decrease in the resonance signal for the carbon at the para position of the aromatic unit when compared with the ^{13}C NMR spectrum of $[NP(OC_6H_5)_2]_3$.[4] The absence of significant skeletal cleavage during metalation is a considerable advantage for the use of such processes in phosphazene high polymer syntheses.

The binding of metal complexes to phosphazene compounds is of structural, catalytic, and potential biomedical importance.[5,6] This reaction system possesses a capacity for the binding of metals both through reactions of II with metal halides, as demonstrated by the synthesis of compound VI, and through the reactions of compound V with metal complexes. To illustrate this second reaction pathway, V was allowed to react with $H_2Os_3(CO)_{10}$ (VII), a compound which has been demonstrated previously to react with tertiary phosphines to yield monosubstituted phosphine osmium cluster compounds, $H_2Os_3(CO)_{10}(PR_3)$.[7] The high reactivity of this osmium cluster (VII) was ascribed to a metal–metal double bond.[8] When compound V was allowed to react with a deficiency of VII at 25 °C in methylene chloride solvent, the expected color change from violet to yellow was observed. Furthermore infrared spectral comparisons of the carbonyl stretching regions for the osmium complex derived from triphenylphosphine and that derived from V confirmed the existence of metal binding through the phosphine residues of V rather than through the skeletal nitrogen atoms.

Experiments are now underway in our laboratory to extend these small molecule cyclic model reactions to high polymeric phosphazenes.

Acknowledgment. This work was supported by grants from the Office of Naval Research and the Army Research Office.

References and Notes

(1) N. L. Paddock, T. N. Ranganathan, and S. M. Todd, *Can. J. Chem.*, **49**, 164 (1971).
(2) M. Biddlestone and R. A. Shaw, *Phosphorus*, **3**, 95 (1973).
(3) H. R. Allcock, D. B. Patterson, and T. L. Evans, *J. Am. Chem. Soc.*, **99**, 6095 (1977).
(4) The para ^{13}C NMR resonance of $[NP(OC_6H_5)_2]_3$ was identified on the basis of its chemical shift by comparison with phenol (G. C. Levy and G. L. Nelson, "Carbon-13 Nuclear Magnetic Resonance for Organic Chemists", Wiley-Interscience, New York, p 10, 1972), by the signal intensity based on the ratio of the number of carbon atoms (two ortho carbons, two meta carbons, and one para carbon per phenolic residue), and by comparing ^{13}C NMR spectra that were obtained by ^{1}H decoupled and partially ^{1}H decoupled modes.
(5) H. P. Calhoun, N. L. Paddock, and J. Trotter, *J. Chem. Soc., Dalton Trans.*, 2708 (1973).
(6) H. R. Allcock, R. W. Allen, and J. P. O'Brien, *J. Am. Chem. Soc.*, **99**, 3987 (1977).
(7) A. J. Deeming and S. Haaso, *J. Organomet. Chem.*, **88**, C21–23 (1975).
(8) A. J. Deeming, S. Hasso, and M. Underhill, *J. Organomet. Chem.*, **80**, C53 (1974).

T. L. Evans, T. J. Fuller, H. R. Allcock*
Department of Chemistry
The Pennsylvania State University
University Park, Pennsylvania 16802
Received August 15, 1978

0002-7863/79/1501-0242$01.00/0