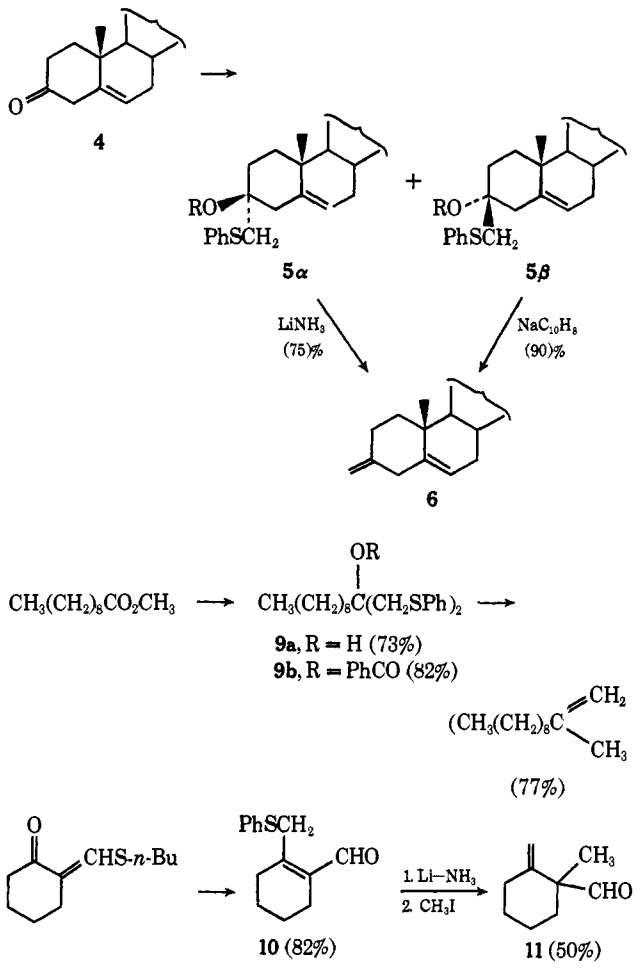
Table I. Ketone Methylenation *via* Phenylthiomethyllithium Addition, Acylation, and Reductive Elimination

Ketone	Acylating agent	Yield of ester,[a] %	Olefin (% yield)[b]
Cyclohexanone	$(CH_3CO)_2O$	82	Methylenecyclohexane (60)[c]
	$(C_6H_5CO)_2O$	77 (96)	
Cyclohexanone[d]	$(C_6H_5CO)_2O$	49	Methylenecyclohexane-d_2 (60)
2-Methylcyclohexanone	$(C_6H_5CO)_2O$	73	2-Methylmethylenecyclohexane (70)[c]
	C_6H_5COCl	72 (91)	
cis-1-Decalone[e]	$(C_6H_5CO_2)O$	73[e]	1-Methylenedecalin (50)[e]
	$(C_6H_5CO)_2O$	73 (81)[e]	
(±)-Norzizanone (**2**)	C_6H_5COCl	49 (71)	(±)-Zizaene (**3**, 64)
Δ^5-Cholesten-3-one (**4**)	$(C_6H_5CO)_2O$	18 α (27)	3-Methylene-Δ^5-cholestene (**6**, 75, 90[f])
		22 β (28)	3-Methylene-Δ^5-cholestene (**6**, 76)

[a] From ketone. The yield of the alcohol, if isolated, follows in parentheses. Conversion to the ester was achieved by lithiation (1 equiv of n-C_4H_9Li/THF), then acylation. [b] Lithium–ammonia reduction and isolated yield from ester unless indicated otherwise. [c] Yield by glc analysis. [d] Reaction with **1**-d_2. [e] Contains ~15% of trans-fused isomer. [f] Reduction with sodium–naphthalene in tetrahydrofuran.



Reaction of α-n-butylthiomethylenecyclohexanone[15] with **1** followed by dehydration of the rather unstable adduct with 10% hydrochloric acid and mercuric chloride in ethanol affords phenylthiomethylene aldehyde **10**. When **10** was subjected to reduction and methylation, the exocyclic β,γ-unsaturated aldehyde **11** was obtained.[16]

A typical procedure for ketone methylenation is as follows: *cis*-1-decalone (1.00 g, 6.57 mmol)[17] in 4 ml of tetrahydrofuran (THF) was added to a solution of **1** (7.3 mmol)[4] in THF–hexane (metallation of thioanisole with n-butyllithium–Dabco[4]) at 0°. After 3 hr at room temperature, the solution was recooled to 0° and benzoic anhydride (3.39 g, 15.0 mmol) in 5 ml of THF was added. After an additional 1.5 hr at room temperature, the mixture was diluted with pentane, filtered, and evaporated. Purification by silica gel chromatography afforded the benzoate (2.00 g, 78%)[17] as a pale yellow oil.

The benzoate (3.23 g, 8.50 mmol) in ether (60 ml) was added to a stirred, refluxing solution of lithium (0.35 g, 51.0 mmol) in liquid ammonia (165 ml) over a 30-min period. After another 30 min, pentane (60 ml) and then solid ammonium chloride (in small portions) were added prior to slow evaporation of the ammonia and addition of water. The pentane layer was combined with a second pentane extraction, washed with 1 *N* sodium hydroxide and water, dried ($MgSO_4$), and evaporated. Purification by chromatography on silica gel gave 0.65 g (~50%) of *cis*-1-methylenedecalin (>95% pure by glc analysis).[17]

Acknowledgment. We are grateful to the National Institutes of Health, the National Science Foundation, and Eli Lily and Co. for financial assistance.

(15) R. E. Ireland and J. A. Marshall, *J. Org. Chem.*, 27, 1615 (1962); R. M. Coates and R. L. Sowerby, *J. Amer. Chem. Soc.*, 93, 1027 (1971).

(16) In addition, 2-methyl-1-cyclohexenecarboxaldehyde was formed in ~20% yield.

(17) Containing ~15% of the trans fused isomer.

(18) University of Illinois Fellow, 1970–1971; Johnson and Johnson Fellow, 1971–1972.

(19) A. P. Sloan Foundation Fellow, 1971–1973.

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Syntheses of Actinomycin and Analogs. VIII. A Synthesis of Actinomycin D Lactam[1,2]

Sir:

Actinomycin D lactam (Chart I) has been synthesized in efforts to obtain analogs of the natural, clinically used antitumor agent which might possess improved therapeutic properties.[3,4] In this peptide analog both threonine residues of actinomycin D[5]

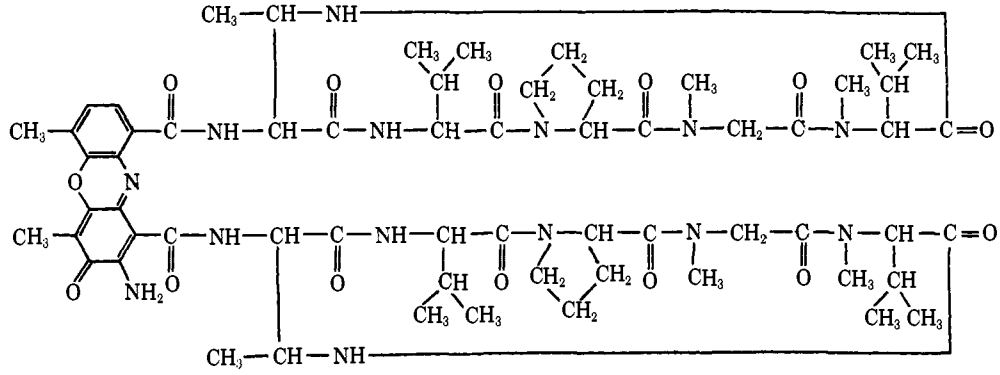
(1) Part VII: J. Meienhofer, R. Cotton, and E. Atherton, *J. Org. Chem.*, 36, 3746 (1971). Supported, in part, by Public Health Service Research Grants C-6516 from the National Cancer Institute and FR-05526 from the Division of Research Facilities and Resources, National Institutes of Health.

(2) Abbreviations follow the rules of the IUPAC-IUB Commission on Biochemical Nomenclature in *Biochemistry*, 5, 1445, 2485 (1966); 6, 362 (1967); *J. Biol. Chem.*, 241, 2491 (1966).

(3) S. Farber, *J. Amer. Med. Ass.*, 198, 826 (1966).

(4) J. Meienhofer, *J. Amer. Chem. Soc.*, 92, 3771 (1970).

Chart I. Structure of Actinomycin D Lactam: 2-Amino-4,6-dimethylphenoxazinone(3)-1,9-bis[carbonyl-L-*threo*-α,β-diaminobutyryl-D-valyl-L-prolylsarcosyl-L-*N*-methylvaline(*N*$^{\beta}$-diaminobutyric acid) Lactam]



[Dbu-D-Val-Pro-Sar-MeVal]

have been replaced by L-*threo*-α,β-diaminobutyric acid. The synthetic pathway is characterized by the cyclization reaction between the proline and sarcosine residues[4] and the subsequent introduction of the *N*-2-nitro-*m*-cresotyl substituent serving as precursor of the phenoxazinone chromophore.

L-*threo*-α,β-Diaminobutyric acid was prepared from L-threonine.[6] Treatment with tosyl chloride afforded *N*-tosyl-L-threonine[7] which was esterified with diazomethane and then O-tosylated to give *N*,*O*-ditosyl-L-threonine methyl ester. Treatment with ammonia-saturated methanol for 48 hr followed by hydrolysis in 6 *N* HCl gave *N*$^{\alpha}$-tosyl-L-*threo*-α,β-diaminobutyric acid (I): mp 233–235°; $[\alpha]^{20}D$ +28.2° (*c* 1, 6 *N* HCl). *Anal.* Calcd for $C_{11}H_{16}N_2O_4S_1 \cdot 0.5H_2O$ (281.24): C, 47.0; H, 6.05; N, 9.96. Found: C, 47.4; H, 6.54; N, 9.88.

Acylation of the β-amino group of I with *tert*-butyloxycarbonyl azide[8] was followed by treatment with sodium in liquid ammonia for removal of the *N*$^{\alpha}$-tosyl group.[9] Successive *N*$^{\alpha}$-carbobenzoxylation, esterification, and acidolytic cleavage of the *N*$^{\beta}$-*tert*-butyloxycarbonyl group gave the required *N*$^{\alpha}$-benzyloxycarbonyl-L-*threo*-α,β-diaminobutyric acid methyl ester hydrochloride (II): mp 197–198°; $[\alpha]^{20}D$ −14.3° (*c* 1, dimethylformamide). *Anal.* Calcd for $C_{13}H_{19}N_2O_4Cl$ (302.8): C, 51.6; H, 6.33; N, 9.25. Found: C, 51.7; H, 6.75; N, 9.41.[6]

Condensation of II with *tert*-butyloxycarbonyl-L-*N*-methylvaline[10] by the mixed anhydride procedure using isobutyl chloroformate[11,12] gave *N*$^{\alpha}$-benzyloxycarbonyl-*N*$^{\beta}$-(*tert*-butyloxycarbonyl-L-*N*-methylvalyl)-L-*threo*-α,β-diaminobutyric acid methyl ester (III) in crystalline form: 64%; mp 122–123°; $[\alpha]^{20}D$ −6.3° (*c* 1, methanol). *Anal.* Calcd for $C_{24}H_{37}N_3O_7$ (479.6): C, 60.1; H, 7.78; N, 8.76. Found: C, 60.1; H, 7.92; N, 8.51.

The *tert*-butyloxycarbonyl group was removed by anhydrous trifluoroacetic acid[13] and the product converted to the hydrochloride salt IIIa. *tert*-Butyloxycarbonylsarcosine[14] was coupled with IIIa utilizing the mixed anhydride method to give crystalline *N*$^{\alpha}$-benzyloxycarbonyl-*N*$^{\beta}$-(*tert*-butyloxycarbonylsarcosyl-L-*N*-methylvalyl)-L-*threo*-α,β-diaminobutyric acid methyl ester (IV): 84%; mp 109°; $[\alpha]^{20}D$ −26.0° (*c* 1, methanol). *Anal.* Calcd for $C_{27}H_{42}N_4O_8$ (550.7): C, 58.9; H, 7.69; N, 10.2. Found: C, 58.5; H, 7.92; N, 10.0.

Saponification by 1 *N* NaOH in aqueous acetone at 0° gave the corresponding acid V as a homogeneous oil, 84.5%. Condensation of V with D-valyl-L-proline *p*-nitrophenyl ester hydrobromide[10] by the mixed anhydride method in the presence of triethylamine gave *N*$^{\alpha}$-benzyloxycarbonyl-*N*$^{\beta}$-(*tert*-butyloxycarbonylsarcosyl-L-*N*-methylvalyl)-L-*threo*-α,β-diaminobutyryl-D-valyl-L-proline *p*-nitrophenyl ester (VI): 72.5%; mp 72–76°; $[\alpha]^{20}D$ −31.2° (c 1, methanol). *Anal.* Calcd for $C_{42}H_{59}N_7O_{12}$ (854.0): C, 59.1; H, 6.96; N, 11.5. Found: C, 59.2; H, 7.18; N, 11.4.

The *tert*-butyloxycarbonyl group was cleaved by anhydrous trifluoroacetic acid and the product converted to the hydrochloride salt and dissolved in a minimum amount of dimethylformamide–acetic acid (9:1). Cyclization was carried out in a dilute pyridine solution (*c* ~0.05) in the presence of triethylamine for 5–6 hr at 60°.[4,10,15] The pyridine was replaced by ethyl acetate. The solution was washed with 1 *M* $NaHCO_3$, water, 1 *N* H_2SO_4, and water, dried over $MgSO_4$, and evaporated. The residual oil was purified by column chromatography on Sephadex LH-20 in ethanol. Evaporation of the fastest eluting fraction gave *N*$^{\alpha}$-benzyloxycarbonyl-L-*threo*-α,β-diaminobutyryl-D-valyl-L-prolylsarcosyl-L-*N*-methylvalyl(*N*$^{\beta}$-

(5) H. Brockmann, G. Bohnsack, B. Franck, H. Gröne, H. Muxfeldt, and C. Süling, *Angew. Chem.*, **68**, 70 (1956); E. Bullock and A. W. Johnson, *J. Chem. Soc.*, 3280 (1957).

(6) Crystalline compounds were obtained at each reaction stage and were fully characterized. The authenticity of L-*threo*-α,β-diaminobutyric acid was established by nmr spectroscopy in 2 *N* DCl; a H_{α}–H_{β} coupling constant of 3.6 Hz was observed. Also, other physical data were in agreement with the naturally isolated compound [W. K. Hausmann, D. B. Borders, and J. E. Lancaster, *J. Antibiot.*, 22, 207 (1969); A. A. Bodanszky and M. Bodanszky, *ibid.*, 23, 149 (1970)]. Details of the synthesis of all four isomers of α,β-diaminobutyic acid will be published elsewhere.

(7) M. Brenner, K. Rüfenacht, and E. Sailer, *Helv. Chim. Acta*, 34, 2102 (1951).

(8) L. A. Carpino, A. Giza, and B. A. Carpino, *J. Amer. Chem, Soc.*, **81**, 955 (1959); L. A. Carpino, *ibid.*, **82**, 2725 (1960); R. Schwyzer, P. Sieber, and H. Kappeler, *Helv. Chim. Acta*, 43, 2622 (1959).

(9) V. du Vigneaud and O. K. Behrens, *J. Biol. Chem.*, **117**, 27 (1937).

(10) J. Meienhofer and R. P. Patel, *Int. J. Protein Res.*, 3, 347 (1971).

(11) T. Wieland and H. Bernhard, *Justus Liebigs Ann. Chem.*, 572, 190 (1951); R. A. Boissonnas, *Helv. Chim. Acta*, 34, 874 (1951); J. R. Vaughan, Jr., *J. Amer. Chem. Soc.*, 73, 3547 (1951).

(12) G. W. Anderson, J. E. Zimmerman, and F. M. Callahan, *ibid.*, **89**, 5012 (1967).

(13) H. Kappeler and R. Schwyzer, *Helv. Chim. Acta*, 43, 1453 (1960).

(14) E. Schnabel, *Justus Liebigs Ann. Chem.*, 702, 188 (1967).

(15) R. Schwyzer and P. Sieber, *Helv. Chim. Acta*, 40, 624 (1957).

diaminobutyric acid) lactam (VII): 38%; mp 145–149°; $[\alpha]^{20}D$ −25.3° (*c* 0.94, methanol). *Anal.* Calcd for $C_{31}H_{46}N_6O_7$ (614.7): C, 60.6; H, 7.54; N, 13.6. Found: C, 60.4; H, 7.82; N, 13.6.

The benzyloxycarbonyl group was removed by catalytic hydrogenolysis[16] and condensed with the symmetrical anhydride of 2-nitro-3-benzyloxy-4-methylbenzoic acid[17] giving, after purification (as described for VII), N^{α}-(2-nitro-3-benzyloxy-4-methylbenzoyl)-L-*threo*-α,β-diaminobutyryl-D-valyl-L-prolylsarcosyl-L-*N*-methylvalyl(N^{β}-diaminobutyric acid) lactam (VIII) in crystalline form: 74%; mp 164–168°; $[\alpha]^{20}D$ −35.3° (*c* 1, methanol). *Anal.* Calcd for $C_{38}H_{51}N_7O_9$ (749.9): C, 60.9; H, 6.86; N, 13.1. Found: C, 60.7; H, 7.18; N, 12.91.

Catalytic hydrogenation of VIII followed by oxidation in the presence of potassium ferricyanide[18] in a 1:1 mixture of methanol and 0.066 *M* phosphate buffer, pH 7.1, gave actinomycin D lactam which was purified by column chromatography on Sephadex LH-20 in ethanol. Crystallization was achieved from ethyl acetate–methanol–hexane giving actinomycin D lactam (XI) (Chart I) as orange red needles: 18%; mp 260–267°; $[\alpha]^{20}D$ −206.5 ± 3° (*c* 0.23, methanol). *Anal.* Calcd for $C_{62}H_{87}N_{13}O_{15}\cdot H_2O$ (1272.6): C, 58.5; H, 7.14; N, 15.4. Found: C, 58.7; H, 6.98; N, 15.1.

Microbiological assays using *Lactobacillus arabinosus* (ATCC 8014) and *L. casei* in pantothenate- and thiamine-dependent systems, respectively,[19] showed IX to possess high antibacterial activity [ID_{50} 0.5 μg/ml and 0.25–0.5 μg/ml, respectively].[20] Preliminary toxicity studies, in AKD_2F_1 male mice, indicate an LD_{50} of approximately 1.5 mg/kg. Suppression of ROS mouse tumor growth[21] was observed at daily doses of 0.6–1.2 mg/kg.

Acknowledgments. We wish to thank Dr. G. F. Foley for microbiological assay, Miss B. L. Brown for toxicity and antitumor studies, and Dr. S. Sengupta for a generous supply of 2-nitro-3-benzyloxy-*p*-toluic acid.

(16) M. Bergmann and L. Zervas, *Ber. Deut. Chem. Ges. B*, **65**, 1192 (1932).
(17) J. Meienhofer, R. Cotton, and E. Atherton, *J. Org. Chem.*, **36**, 3746 (1971).
(18) W. G. Hanger, W. C. Howell, and A. W. Johnson, *J. Chem. Soc.*, 496 (1958).
(19) G. E. Foley, *Antibiot. Annu.*, 432 (1955–1956); G. E. Foley, R. E. McCarthy, and V. M. Binns, *Ann. N. Y. Acad. Sci.*, **76**, 413 (1958).
(20) ID of actinomycin D in these systems is 0.05 μg/ml and 0.055–0.07 μg/ml, respectively.
(21) G. J. D'Angio, C. L. Maddock, S. Farber, and B. L. Brown, *Antibiot. Annu.*, **25**, 1002 (1965).

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Phenylphosphinidene Oxide. Thermal Decomposition of 2,3-Benzo-1,4,5,6,7-pentaphenyl-7-phosphabicyclo-[2.2.1]hept-5-ene Oxide

Sir:

The generation of phosphinidene and phosphinidene oxide, analogs of carbene, is of interest because of their possible intermediacy and synthetic value in the formation of phosphorus heterocycles. In particular, the addition of phosphinidene to acetylene could afford phosphacyclopropene, a heterocycle π isoelectronic with cyclobutadiene. Although much attention has been given to the generation and intermediacy of phosphinidenes,[1–5] there is little information on phosphinidene oxides. A possible route to these species is the thermal decomposition of bicyclic compounds containing a phosphorus bridge. One such compound with a trivalent phosphorus atom, 9-phenyl-9-phosphabicyclo[4.2.1]nonatriene, has been prepared,[4] but no phosphinidene was detected on its thermolysis. Although the Diels–Alder adduct of pentaphenylphosphacyclopentadiene and maleic anhydride has been reported,[6] other phosphine-bridged compounds in the bicyclo [2.2.1] series could not be prepared.[7]

Bicyclic compounds with a bridged phosphine oxide group are more stable, however, and can be isolated.[8,9] Thus, 2,3-benzo-1,4,5,6,7-pentaphenyl-7-phosphabicyclo[2.2.1]hept-5-ene oxide (**2**) was prepared in 92% yield by the addition of benzyne,[10] generated *in situ* at 40°, to pentaphenylphosphacyclopentadiene oxide.[11] All attempts to reduce **2** to **1** afforded only tetraphenylnaphthalene (**3**). Decomposition of **2** at 155° also gave a quantitative yield of tetraphenylnaphthalene and a polymer which analyzed for $(C_6H_5PO)_n$, poly(phenylphosphinidene oxide) (**4**), mol wt 1770. The mass spectrum of **2** (70 eV) showed *m/e* 432 (tetraphenylnaphthalene) and *m/e* 124 (C_6H_5PO), indicating that the bridge portion of **2** cleaves completely and exists independently for a finite period before polymerization occurs.

The thermal decomposition of **2** in a sealed tube in the presence of diethyl disulfide afforded *S,S*-diethyl phenyldithiophosphonate (**5**) which was identified by comparison with an authentic sample obtained from the oxidation of *S,S*-diethyl phenyldithiophosphonite:[12] nmr δ 7.6 (5 H, aromatic), 2.92 (s, 4 H, J = 7 Hz), and 1.35 (t, 6 H, J = 7 Hz). When **2** was thermally decomposed in the presence of methanol, methyl phenylphosphinate (**6**) was obtained; nmr δ 7.1 (5 H, aromatic), 3.4 (d, 3 H, J_{H-P} = 11 Hz), and 7.0 (d, 1 H, J_{H-P} = 520 Hz). Thermal decomposition of **2** in a sealed tube in the presence of ketene diethylacetal (**7**), an electron-

$$\underset{\mathbf{7}}{CH_2{=}C(OCH_2CH_3)_2} + \underset{\mathbf{9}}{(C_6H_5P)_n} \xrightarrow[2.\ O_2]{1.\ \Delta} \underset{\mathbf{8}}{C_6H_5\overset{\overset{O}{\uparrow}}{P}(OCH_2CH_3)_2}$$

(1) See U. Schmidt, I. Boie, C. Osterroht, R. Schröer, and H. F. Grützmacher, *Chem. Ber.*, **101**, 1381 (1968), and references cited therein.
(2) W. Mahler, *J. Amer. Chem. Soc.*, **86**, 2306 (1964).
(3) F. Johnson, R. S. Gohlke, and W. A. Nasutavicus, *J. Organometal. Chem.*, **3**, 233 (1965).
(4) T. J. Katz, C. R. Nicholson, and C. A. Reilly, *J. Amer. Chem. Soc.*, **88**, 3832 (1966).
(5) B. Bloch and Y. Gounelle, *C. R. Acad. Sci., Ser. C.*, **266**, 220 (1968).
(6) E. H. Braye, W. Hübel, and I. Caplier, *J. Amer. Chem. Soc.*, **83**, 4406 (1961).
(7) A. N. Hughes and S. Uaboonkul, *Tetrahedron*, **24**, 3437 (1968).
(8) R. Kluger, F. Kerst, D. G. Lee, and F. H. Westheimer, *J. Amer. Chem. Soc.*, **89**, 3919 (1967).
(9) G. Märkl and R. Potthast, *Tetrahedron Lett.*, 1755 (1968).
(10) T. F. Mich, E. J. Nienhouse, T. E. Farina, and J. J. Tufariello, *J. Chem. Educ.*, **45**, 272 (1968).
(11) All compounds reported gave satisfactory carbon and hydrogen analyses.
(12) M. J. Gallagher and I. D. Jenkins, *J. Chem. Soc. C*, 2176 (1966).