Utility of the Wittig Reaction for the Construction of Side Chains of Steroids Starting from Pregnenolone

Steven R. Schow and Trevor C. McMorris*

Department of Chemistry, University of California, San Diego, La Jolla, California 92093

Received February 16, 1979

The Wittig reaction in which the ylide is generated with potassium *tert*-amylate in refluxing benzene or toluene has been found to be generally applicable to the preparation from pregnenolone of steroids having a variety of side chains. Catalytic hydrogenation of the 20(22)-unsaturated products thus formed is stereoselective and gives mainly the epimer with the natural stereochemistry (20*R*). The synthesis of a number of cholestene and stigmastene derivatives is described.

In the past 15 years a large number of new naturally occurring steroids have been identified which possess functionalized side chains of eight or more carbon atoms. Many of these steroids are of special interest because of their biological properties. Among them are the ecdysones (insect and crustacean moulting hormones), hydroxylated derivatives of vitamin D, and sex hormones of the aquatic fungus *Achlya*.

The chemistry and, in particular, the synthesis of these steroids have been extensively reviewed, and in a recent article special emphasis has been given to a consideration of side-chain synthesis.[1] We have had a direct interest in this subject because of our work on antheridiol, 23-deoxyantheridiol, and the oogoniols, steroids which have been isolated from culture liquids of *Achlya*. Structures for the oogoniols were proposed in 1975 and they have since been defined more completely.[2] Also, new oogoniols possessing a side-chain double bond at C-24(28) have been found.

In seeking ways to synthesize oogoniol (**1**) we noted that 11,15-dihydroxy derivatives of progesterone have been obtained by microbiological hydroxylation of progesterone.[3] It seemed to us that such derivatives could be transformed into oogoniol by modifying the functions in the ring system and attaching an appropriate side chain at C-20. Thus, we were prompted to examine the reactivity of 20-keto steroids in the Wittig reaction.

A survey of the literature revealed several examples of the Wittig reaction with 20-keto steroids leading to products with one or two more carbons. It has been reported that the Wittig reaction goes poorly, even with relatively small ylides.[4,5] However, good yields were obtained with the ylide from methoxymethyltriphenylphosphonium chloride when the reaction was carried out in refluxing diethylene glycol dimethyl ether.[6] The ylide from diethyl (cyanomethyl)phosphonate has been found to react with pregnenolone acetate in tetrahydrofuran at room temperature to give a high yield of the expected unsaturated nitrile.[7]

For our purposes, an article reporting several successful Wittig reactions with pregnenolone (**2**) and relatively bulky phosphoranes has proved to be important.[8] The phosphoranes were prepared from the corresponding phosphonium salts with sodium *tert*-amylate in refluxing benzene. A solution of pregnenolone in benzene was then added and the mixture refluxed for 2 h. A single product, the *E*-20(22) isomer, was obtained in high yield. Longer reaction times led to the formation of an aryl ether at C-3.

The configuration of the olefin was indicated by the chemical shift of the C-21 methyl protons, which appear as a singlet at ~1.62 ppm. This is within the range expected for the *E* isomer (1.60–1.65 ppm). The C-21 methyl protons in the *Z* isomer would be expected to occur in the range 1.68–1.71 ppm.[9] The signal for the C-18 methyl protons likewise occurs at higher field (0.55 ppm) than would be expected for a *Z*-20(22) configuration (0.65–0.78 ppm).[8]

We have applied the Wittig reaction to the synthesis of a number of steroid derivatives, some of which are inaccessible by more conventional procedures. It is necessary, however, to increase the reaction time to 3 h or more and to increase the ratio of phosphorane to steroid in order to ensure high yields. Potassium *tert*-amylate, conveniently prepared from *tert*-amyl alcohol and potassium hydride, can be used instead of sodium *tert*-amylate. Longer reaction times do give more aryl ether. This product probably results from steroidal alkoxide addition to the positive phosphorus (both formed in an acid–base reaction from pregnenolone and the phosphorane), followed by internal collapse of the pentacovalent phosphorus intermediate.[10] By protecting the hydroxyl group in the steroid, this side reaction can be avoided. The results of several experiments are summarized in Table I.

Both (*E*)-20(22)-dehydrocholesterol (**3**) and (*E*)-20-(22)-dehydro-24-norcholesterol (**4**) were obtained in high yield as reported earlier.[8] When pregnenolone was first converted to 6β-methoxy-3,5-cyclo-5α-pregnan-20-one and the latter reacted with isohexyltriphenylphosphorane, a quantitative yield of the expected product (**5**) was obtained. The phosphorane from methoxymethyltriphenylphosphonium chloride reacted with the tetrahydropyranyl (THP) ether of pregnenolone to give the expected 20(22) vinyl ether (**6**) in almost quantitative yield after 3 h at room temperature (cf. ref 6). The 20(22) vinyl ether (mixture of isomers, E/Z = 4:1) was readily cleaved with acid to give 3β-hydroxydinorchol-5-en-24-al and its C-20 epimer (2:1). This was the only case in which we

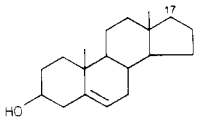
(1) D. M. Piatak and J. Wicha, *Chem. Rev.*, **78**, 199 (1978).
(2) M. W. Preus and T. C. McMorris, *J. Am. Chem. Soc.*, **101**, 3066 (1979).
(3) W. Charney and H. L. Herzog, "Microbial Transformation of Steroids", Academic Press, New York, 1967, Chapter 3.
(4) Y. M. Sheikh and C. Djerassi, *J. Org. Chem.*, **38**, 3545 (1973).
(5) N. K. Chaudhuri, R. Nickolson, J. G. Williams, and M. Gut, *J. Org. Chem.*, **34**, 3767 (1969).
(6) G. R. Pettit, B. Green, G. L. Dunn, and P. Sunder-Plassmann, *J. Org. Chem.*, **35**, 1385 (1970).
(7) A. K. Bose and R. T. Dahill, Jr., *J. Org. Chem.*, **30**, 505 (1965).
(8) J. P. Schmit, M. Piraux, and J. F. Pilette. *J. Org. Chem.*, **40**, 1586 (1975).
(9) R. B. Bates, R. H. Carnighan, R. O. Rakutis, and J. H. Schauble, *Chem. Ind.*, 1020 (1962); B. M. Trost, *Acc. Chem. Res.*, **3**, 120 (1970).
(10) C. T. Eyles and S. Trippett, *J. Chem. Soc. C*, 67 (1966).

0022-3263/79/1944-3760$01.00/0

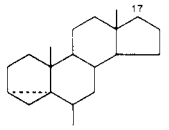
Table I. Alkenes from Reaction of Pregnenolone (and Derivatives) with Phosphoranes in Refluxing Benzene

steroid	phosphorane	product struct	no.	yield, %	mp, °C
pregnenolone	Ph_3P	R	3[a]	78	133-136
pregnenolone	Ph_3P	R	4	64	143-145[b]
6β-methoxy-3,5-cyclo-5α-pregnan-20-one	Ph_3P	R'	5[c]	100	*d*
pregnenolone tetrahydropyranyl ether	$Ph_3P{=}CHOCH_3$	R'' OCH_3	6[e]	96	*f*
pregnenolone tetrahydropyranyl ether	Ph_3P OTHP	R''' OAc	7[g,h]	59	117-119
pregnenolone tetrahydropyranyl ether	Ph_3P O O	R'' O	8[g,h]	70	115-118
pregnenolone tetrahydropyranyl ether	Ph_3P O O 11	R''' O	12[h]	85	110-112
pregnenolone tetrahydropyranyl ether	Ph_3P OTHP 13	OH R	16[h]	82	138-141

[a] The structure of R (to which the side chain is attached at C-17) is A. [b] Lit.[24] mp 148-151 °C. [c] The structure of R' (to which the side chain is attached at C-17) is B. [d] Lit.[23] amorphous. [e] R'' = tetrahydropyranyl ether of R. [f] Amorphous. [g] R''' = acetate of R. [h] Not the immediate product.



A



B

observed formation of *E* and *Z* isomers in the Wittig reaction at the 20-carbonyl.

Three-carbon homologation of pregnenolone to give a C-24 oxygenated steroid was also investigated. The phosphorane which we selected was prepared from the tetrahydropyranyl derivative of 3-bromo-1-propanol. It reacted with pregnenolone THP ether to give a moderate yield of the desired product which was converted to the corresponding diacetate (**7**) for further characterization. Catalytic hydrogenation of the 20(22) double bond resulted in partial loss (~50%) of the side chain acetate group. This finding, which has implications in the steric course of the catalytic hydrogenation of steroidal 20(22) olefins, is examined further below.

The Wittig reaction has been applied to the preparation of (*E*)-3β-acetoxy-27-norcholesta-5,20(22)-dien-25-one (**8**),[11] (24ξ,25ξ)-3β,26-dihydroxystigmast-5-ene (**9**), and (24ξ)-3β,29-dihydroxystigmast-5-ene (**10**). The latter steroids were required as models for spectral studies to determine the location of the primary hydroxyl group in the oogoniols. A preliminary communication of this work has been published.[12] Details of the preparation of the phosphoranes **11** and **13** for use in the Wittig reactions leading to **9** and **10** are given in the Experimental Section of this paper.

In order that the method of steroid side chain elaboration be of general applicability, it is necessary for the 20(22) double bond to be selectively reduced so as to yield the product having the natural stereochemistry (20*R*). Several examples of stereospecific catalytic hydrogenation of the double bond to give the 20*R* epimer have been reported.[8,13,14] However, these findings have been challenged by other workers.[15,16] In our experiments the dehydro steroid was dissolved in dioxane–acetic acid (50:1) and hydrogenated in the presence of platinum oxide. These conditions were used originally by Hershberg and co-workers who found that the 5(6) double bond was not affected.[17] Bergmann and co-workers reported that this method gave a high yield of cholesteryl acetate and no trace of the C-20 epimer was found.[18] The same method has been used by Piraux and co-workers who obtained an 80.5% yield of cholesteryl acetate.[8]

When we reduced 20(22)-dehydrocholesteryl acetate by this method, a mixture of epimers (20*R*- and 20*S*-cholesteryl acetate) was obtained in the ratio 4:1 as indicated by gas chromatography. Crystallization of the crude product from methanol–chloroform gave a 66% yield of pure cholesteryl acetate.

Similar catalytic hydrogenation of (*E*)-3β-acetoxy-27-norcholesta-5,20(22)-dien-25-one (**8**) gave an excellent yield

(11) T. C. McMorris and S. R. Schow, *J. Org. Chem.*, **41**, 3759 (1976).
(12) T. C. McMorris, S. R. Schow, and G. R. Weihe, *Tetrahedron Lett.*, 335 (1978).
(13) R. Ikan, A. Markus, and E. D. Bergmann, *J. Org. Chem.*, **36**, 3945 (1971).
(14) A. M. Porto and E. G. Gros, *J. Labelled Compd.*, **6**, 369 (1970).
(15) T. A. Narwid, K. E. Cooney, and M. R. Uskokovic, *Helv. Chim. Acta*, **57**, 771 (1974).
(16) W. R. Nes, T. E. Varkey, and K. Krevitz, *J. Am. Chem. Soc.*, **99**, 260 (1977).
(17) E. B. Hershberg, E. P. Oliveto, C. Gerold, and L. Johnson, *J. Am. Chem. Soc.*, **73**, 5073 (1951).
(18) E. D. Bergmann, M. Rabinovitz, and Z. H. Levinson, *J. Am. Chem. Soc.*, **81**, 1239 (1959).

(90%) of the 20*R* dihydro derivative, indicating better stereoselectivity than in the previous case. Both 20*R* and 20*S* derivatives had previously been well characterized by Uskokovic and co-workers,[15] and Dr. Uskokovic kindly supplied us with samples of the two epimers and also the 20(22) dehydro precursors. Thus, an accurate comparison of the compounds could be made which fully confirmed the assignments.[11]

The stereoselectivity was not as good in the catalytic hydrogenation of (*E*)-(24ξ)-3β-acetoxy-27-norstigmasta-5,20(22)-dien-25-one (**12**). Gas chromatography indicated a 3:1 mixture of 20*R* to 20*S* isomers but pure 20*R* isomer was obtained in 58% yield on recrystallization. This isomer (**14**) was treated with the phosphorane from methoxymethyltriphenylphosphonium chloride to give the expected vinyl ether in high yield. The latter was converted to the aldehyde by treatment with perchloric acid in ether and the aldehyde reduced with sodium borohydride to give (24ξ,25ξ)-3β,26-dihydroxystigmast-5-ene (**9**).

It is interesting to note that the intermediate (**14**) had properties identical with those reported for the compound obtained by cleavage of the 25(26) double bond in the acetate of the marine sterol clerosterol (**15**).[19] The intermediate (**14**) was treated with the ylide from methyltriphenylphosphonium iodide to give a product having the same spectral properties as clerosterol. The melting point was considerably lower than that of clerosterol, however, probably because the product was an epimeric mixture (at C-24).

Catalytic hydrogenation of the diacetate of (*E*)-(24ξ)-3β,29-dihydroxystigmasta-5,20(22)-diene (**16**) furnished the dihydro derivative as an epimeric mixture (ratio of 20R/20S = 3:1). In this experiment, complete hydrogenation of the 20(22) double bond required several days.

As mentioned earlier, an interesting result was observed on catalytic hydrogenation of the cholene derivative (**7**). A mixture of epimers was obtained as in other cases though the ratio of 20*R* to 20*S* epimers was only 65:35. In addition, extensive hydrogenolysis of the homoallylic C-24 acetate group occurred. This result implies that rearrangement of the double bond from C-20(22) to C-22(23) takes place to give the allylic acetate which then undergoes hydrogenolysis. Thus hydrogenation of the 20(22) double bond is not simply a cis addition of two hydrogen atoms but appears to be a stepwise process which involves π-allyl intermediates. Consistent with such intermediates is the finding of Uskokovic and co-workers that hydrogenation of the *E*-olefin **8** gave the same mixture of C-20 epimers as hydrogenation of its *Z* isomer.[15]

The stereoselectivity in the formation of the chiral center at C-20 thus depends on the nature of the side chain and, presumably, on the nature of the catalyst (type of metal and method of preparation) and the solvent. In all compounds we examined there was a clear preference for the production of the 20*R* epimer, ranging from about 90:10 to 65:35. It should be noted, however, that Nes and co-workers have reported that they obtained no stereoselectivity in this reaction.[16,20]

In contrast to catalytic hydrogenation, which evidently proceeds in a complex manner, we and others have observed that hydroxylation of the 20(22) double bond in **5** with osmium tetroxide is highly stereoselective and gives the 20*S*,22*S*-dihydroxy product almost exclusively.[21,22,23] This indicates that there is a preferred conformation of the side chain in which one side of the double bond is more exposed to attack by the cis hydroxylating reagent than the other. The stereochemistry at C-20 and C-22 in the product is opposite to that in ecdysterone and related hormones so construction of the ecdysterone side chain via a 20(22) dehydro precursor cannot be carried out directly.

Experimental Section

Melting points were determined on a Kofler hot stage apparatus and are uncorrected. Spectra were obtained on the following instruments: Varian T60, EM 390, HR220 (^{1}H NMR), Perkin-Elmer spectrophotometer 550 (UV), Beckman IR 18A-X (IR), LKB 9000 (mass spectra). The ionizing voltage for mass spectra was 70 eV. Infrared spectra were taken of KBr pellets. Elemental analyses were performed by Pascher Laboratories, Bonn, West Germany.

Column chromatography was done with Merck silica gel 60 (60–230 mesh, or finer than 200 mesh ASTM). Thin-layer chromatography was done on Merck 60 F-254 silica gel plates. Gas chromatographic (GC) analysis was carried out on a Varian Aerograph 2100 instrument with a flame ionization detector and glass columns (6 ft) packed with 3% OV-17 on Gas-Chrom Q or 10% SE-30 on firebrick. The column temperature was 280 °C and helium flow rate was 35 mL/min.

General Procedure for Preparation of Potassium *tert*-Amylate in Aromatic Hydrocarbon Solution. In a 1-L flask, with a drying tube and an inlet for admitting argon, was placed 144 g of 23% potassium hydride oil dispersion. The oil was washed away (2 × 300 mL of petroleum ether and 2 × 300 mL of benzene or toluene) and enough benzene (or toluene) was added to give a total volume of 400 mL. *tert*-Amyl alcohol (80 g, freshly distilled from sodium) was added dropwise to the stirred suspension. After addition was complete (~1 h), the flask was put in the refrigerator until the suspension had settled. An aliquot of the amber supernatant was titrated with standard 1 N hydrochloric acid with phenolphthalein as indicator. The molarity of the base was found to be in the range 1.1 to 2.1. Moisture should be excluded from the amylate solution and it should be stored in the refrigerator. Under these conditions, the base did not deteriorate during 1 year.

General Procedure for Wittig Reaction: Preparation of (*E*)-3β-Hydroxycholesta-5,20(22)-diene (3) and Subsequent Hydrogenation of Its Acetylated Derivative. A mixture of 10 g (0.023 mol) of isohexyltriphenylphosphonium bromide and 12.6 mL of 1.86 M potassium *tert*-amylate in benzene (0.023 mol) was refluxed under argon for 25 min. A solution of 1.07 g (0.0034 mol) of pregnenolone in 15 mL of hot benzene was added rapidly by syringe. The mixture was refluxed for 2.25 h and then cooled and poured into water (100 mL), and the resulting mixture was extracted with ether (3 × 75 mL). The ether extracts were combined and washed with 10% hydrochloric acid, saturated sodium bicarbonate solution, and water and dried ($MgSO_4$). Removal of the ether and chromatography of the residue (petroleum ether (bp 30–60 °C)–ethyl acetate, 4:1) yielded 1.01 g (78%) of **3**: mp 133–136 °C (lit. 135–138 °C);[5] NMR δ 0.55 (3, s), 0.88 (b, d, *J* = 6.5 Hz), 1.01 (3, s), 1.62 (3, s), 3.52 (1, m), 5.15 (1, t, *J* = 7 Hz), 5.33 (1, m).

A solution of 0.05 g of the acetylated derivative of **3** in 2.5 mL of dioxane–acetic acid (50:1) was stirred with 0.01 g of PtO_2 in an atmosphere of hydrogen at room temperature and pressure. After 4 h, more PtO_2 (0.01 g) was added and the hydrogenation continued for 4 h. The mixture was filtered and the solvent removed from the filtrate, leaving the product, which was recrystallized from methanol–chloroform, to give pure cholesteryl acetate (0.03 g, 66%), mp 114–115 °C (lit. mp 115–116 °C).[24,25]

(19) M. Manzoor-L-Khuda, *Tetrahedron*, **22**, 2377 (1966); see also, P. J. Pettler, W. J. S. Lockley, H. H. Rees, and T. W. Goodwin, *J. Chem. Soc., Chem. Commun.*, 844 (1974).
(20) W. R. Nes, *J. Am. Chem. Soc.*, **100**, 999 (1978).
(21) K. Bannai, M. Morisaki, and N. Ikekawa, *J. Chem. Soc., Perkin Trans. 1*, 2116 (1976).
(22) M. Koreeda, N. Koizumi, and B. A. Teicher, *Tetrahedron Lett.*, 4565 (1976).
(23) C. Byon and M. Gut, *J. Org. Chem.*, **41**, 3716 (1976).
(24) K. Shimizu, M. Gut, and R. I. Dorfman, *J. Biol. Chem.*, **237**, 699 (1962).
(25) R. Weast, Ed., "Handbook of Chemistry and Physics", Chemical Rubber Co., Cleveland, OH, pp 51, C 415.

Analysis of the crude product by GC indicated a mixture of 20*R* and 20*S* epimers in the ratio 4:1.

3-(2′-Tetrahydropyranyloxy)-20-methoxymethylene-5-pregnene (6). Methoxymethyltriphenylphosphonium chloride (8.6 g, 0.025 mol) was treated at room temperature with potassium *tert*-amylate (0.025 mol) in toluene. The mixture was stirred for 5 min and then 1.0 g (0.0025 mol) of pregnenolone tetrahydropyranyl ether in 5 mL of toluene was added. The mixture was stirred for 3 h and then the product (6) was isolated (1.03 g, 96%). The NMR spectrum and GC analysis indicated that 6 was a mixture of *E* and *Z* isomers (ratio of *E*/*Z* = 4:1): NMR δ 0.57 (2.4, s), 0.60 (0.6, s), 1.00 (3, s), 1.58 (2.4, s), 1.62 (0.6, s), 3.43, 3.55 (OCH_3), 4.68 (1, m), 5.75 (0.8, broad s), 5.83 (0.2, broad s). A portion of 6 (0.4 g) was dissolved in ether (10 mL) and 0.7 mL of 70% perchloric acid added. The mixture was stirred for 45 min and the ether layer was then separated and dried ($MgSO_4$), and the ether was removed in a stream of N_2. Filtration of the residue through a column of silica gel gave the hydroxy aldehyde (0.173 g, 45%). The NMR spectrum indicated that it was a mixture of 3β-hydroxydinorchol-5-en-24-al and its C-20 epimer in a ratio of 2:1; NMR δ 0.68 (0.67, s), 0.72 (0.33, s), 0.99, (3, s), 1.10, 1.11 (two doublets for H-21 in two epimers), 3.51 (1, m), 5.32 (1, m), 9.56 (0.67, d, *J* = 5 Hz), 9.58 (0.33, d, *J* = 3 Hz).

(*E*)-3β,24-Diacetoxychola-5,20(22)-diene (7). A mixture of 20.0 g of 3-bromo-1-propanol, 50.0 g of dihydropyran, and 200 mg of *p*-toluenesulfonic acid in 300 mL of ether was stirred for 5 h at room temperature. Methanolic ammonia solution was added, and the solvents were then evaporated in vacuo. The residue was filtered through a column of silica gel with petroleum ether–dichloromethane as eluant. Removal of the solvent from the eluate yielded 31.9 g of the bromo ether; NMR δ 1.68 (6, m), 2.22 (2, quintet, *J* = 7 Hz), 3.55 (6, m), 4.60 (1, m); MS *m*/*e* 143 (M^+ – HBr).

The bromo ether (15.0 g), triphenylphosphine (17.0 g), and potassium carbonate (10 g) in 75 mL of acetonitrile were heated under reflux for 24 h. The potassium carbonate was filtered off and ether added to the filtrate, thus precipitating the phosphonium salt. It was redissolved in chloroform and reprecipitated with ether, giving 13.7 g (43%) of the white amorphous product, mp 192–195 °C.

A portion of the salt (3.0 g) was converted to the phosphorane with potassium *tert*-amylate and this was refluxed with pregnenolone tetrahydropyranyl ether (0.35 g) for 4.5 h. The crude product was dissolved in tetrahydrofuran and treated with dilute hydrochloric acid to give the diol which was then acetylated (acetic anhydride–pyridine). Purification of the product was effected by chromatography (petroleum ether–ethyl acetate, 5:1) and recrystallization (methanol–chloroform) which gave 0.23 g (59%) of pure 7: mp 117–119 °C; IR 2860, 1740, 1252 cm^{-1}; NMR δ 0.54 (3, s), 1.02 (3, s), 1.65 (3, s), 2.02 (3, s), 2.03 (3, s), 4.05 (2, t, *J* = 7 Hz), 4.59 (1, m), 5.16 (1, t, *J* = 6.6 Hz), 5.38 (1, m); MS *m*/*e* 382 (M^+ – AcOH), 367 (M^+ – AcOH – CH_3), 322 (M^+ – 2 × AcOH).

A solution of 0.04 g of 7 in 2.0 mL of dioxane–acetic acid (50:1) was hydrogenated with 0.01 g of PtO_2. The reaction was complete within 4 h as indicated by TLC which showed the presence of two products. They were separated by chromatography (petroleum ether–ethyl acetate, 7:1). The more polar component was recrystallized (methanol–dichloromethane) to give the expected 20*R*-3β,24-diacetoxychol-5-ene (0.01 g, 24%). The mother liquor contained almost pure 20*S* epimer as indicated by GC (ratio of 20*R* to 20*S* epimers in crude product, 65:35). The 20*R* epimer had the following properties: mp 124–127 °C (lit. mp 126–127 °C);[26] NMR δ 0.68 (3, s), 0.94 (3, d, *J* = 6.3 Hz), 1.02 (3, s), 2.03 (3, s), 2.04 (3, s), 4.02 (2, t, *J* = 6.4 Hz), 4.60 (1, m), 5.37 (1, m); MS *m*/*e* 384 (M^+ – AcOH), 369.

NMR for 20*S* epimer: δ 0.68 (3, s), 0.84 (3, d, *J* = 6.7 Hz), 1.02 (3, s), 2.04 (3, s), 2.08 (3, s), 4.04 (2, t, *J* = 6 Hz), 4.63 (1, m), 5.39 (1, m).

The other component from chromatography of the hydrogenation product (48% of total product as indicated by GC) had the following spectral properties: NMR δ 0.68 (3, s), 0.85 (d, *J* = 7.9 Hz), 0.91 (d, *J* = 6.4 Hz), 1.02 (3, s), 2.03 (3, s), 4.61 (1, m), 5.38 (1, m); MS *m*/*e* 326 (M^+ – AcOH), 311.

1-Bromo-3-ethylpentan-4-one. A solution of 50 g (0.39 mol) of 2-acetylbutyrolactone in 50 mL of dry benzene was added dropwise to a stirred suspension of 9.9 g (0.41 mol) of sodium hydride (prepared by washing 17.4 g of a 57% oil dispersion with 3 × 150 mL of benzene) in 100 mL of dry benzene at 65 °C. The addition was made in 1 h in an argon atmosphere. Ethyl iodide (130 g, 0.83 mol) in 25 mL of dimethylformamide was next added and the mixture refluxed for 5 h, cooled, and poured into water. The product was extracted with benzene (2 × 150 mL) and the extract washed with 10% hydrochloric acid, 10% sodium bicarbonate solution, and water and dried, and the solvent was removed in vacuo. The residue was further purified by filtration of its ether solution through a column of silica gel (yield 57 g, 94%). GC analysis showed it to be free of starting material. Spectral properties included NMR δ 0.87 (3, t, *J* = 7 Hz), 2.03 (3, m), 2.30 (3, s), 2.87 (1, m), 4.13 (1, t, *J* = 8 Hz), 4.17 (1, dd, J_1 = 8 Hz, J_2 = 2 Hz); MS *m*/*e* 114 (M^+ – CH_2CO).

The lactone (25 g, 0.16 mol) was added dropwise to refluxing hydrobromic acid (48%, 70 mL) and water (30 mL) during 3 h. The product was steam distilled from the reaction vessel and collected in a Dean-Stark trap as it formed. It was dried ($MgSO_4$) and distilled in vacuo: yield 19.6 g (63%); bp 64–70 °C (2.2 Torr); IR 1710 cm^{-1}; NMR δ 0.90 (3, t, *J* = 7 Hz), 1.59 (2, m), 2.18 (3, s), 2.18 (2, m), 2.72 (1, quintet, *J* = 7 Hz), 3.39 (2, ABXY multiplet); MS *m*/*e* 112 (M^+ – HBr).

[3-Ethyl-3-(2-methyl-1,3-dioxolan-2-yl)propyl]triphenylphosphonium Bromide. A solution of 30 g (0.16 mol) of 1-bromo-3-ethylpentan-5-one and 44 g (0.17 mol) of triphenylphosphine in 150 mL of toluene was refluxed for 48 h. The precipitate which formed was filtered off, washed with ether, and dried in vacuo. This amorphous phosphonium salt (52 g, 72%) was slightly hygroscopic: mp 161–165 °C. Anal. Calcd for $C_{25}H_{28}BrOP$: C, 65.94; H, 6.20; Br, 17.55; O, 3.51; P, 6.80. Found: C, 65.87; H, 6.19; O, 3.69; P, 6.97.

A mixture of the salt (10.0 g), 10 mL of ethylene glycol, 400 mg of *p*-toluenesulfonic acid, and 1200 mL of benzene was rapidly stirred and heated so as to allow slow distillation of the benzene. The excess glycol was next removed in vacuo (5 h at 120 °C, 2 Torr), leaving the product as a glass. It was dissolved in dichloromethane, and potassium carbonate (3 g) was added. The mixture was stirred for 2 h and filtered, and the solvent was removed. Chromatography of the residue (dichloromethane–methanol, 100:3) gave 11.1 g of pure product. The compound was unstable and hygroscopic and was therefore used immediately. All traces of dichloromethane must be removed before reaction with potassium *tert*-amylate. This was done by dissolving the salt in methanol and distilling off the methanol in vacuo. NMR δ 0.88 (3, m), 1.13 (3, s), 1.78 (5, m), 3.75 (2, m), 3.92 (4, d, *J* = 1 Hz), 7.73 (15, m). Attempts at forming the phosphonium salt from the ketal derivative of the above ketone were unsuccessful.

(*E*)-(24ξ)-3β-Acetoxy-25-norstigmasta-5,20(22)-dien-25-one (12). The phosphonium bromide (25 g) was heated with potassium *tert*-amylate in toluene at reflux temperature (argon atmosphere) for 30 min. Pregnenolone tetrahydropyranyl ether (4 g) in toluene was added rapidly and the mixture refluxed until all the steroid had reacted (11 h). The product was isolated by chromatography (petroleum ether–ethyl acetate). It was dissolved in absolute ethanol (60 mL) and 200 mg of *p*-toluenesulfonic acid added. After stirring overnight at room temperature, the solvent was removed and the residue chromatographed (petroleum ether–ethyl acetate) to give 3.5 g (85%) of the keto alcohol: mp 110–112 °C; NMR δ 0.53 (3, s), 0.83 (3, t, *J* = 7 Hz), 0.98 (3, s), 1.63 (3, s), 2.10 (3, s), 3.47 (1, m), 5.12 (1, t, *J* = 7 Hz), 5.33 (1, m).

Acetylation of the keto alcohol with acetic anhydride–pyridine gave the keto acetate 12: mp 88–89 °C; IR 1732, 1714 cm^{-1}; NMR δ 0.53 (3, s), 0.87 (3, t, *J* = 7.5 Hz), 1.02 (3, s), 1.63 (3, s), 2.03 (3, s), 2.11 (3, s), 4.61 (1, m), 5.10 (1, broad t, *J* = 7 Hz), 5.37 (1, m); MS *m*/*e* 394 (M^+ – AcOH).

(24ξ,25ξ)-3β,26-Dihydroxystigmast-5-ene (9). A solution of the keto acetate (12) in dioxane–acetic acid (50:1) was hydrogenated in the presence of PtO_2 for 14 h. Analysis of the product indicated a mixture of 20*R* and 20*S* epimers in the ratio 74:26. Recrystallization of the product from methanol–dichloromethane yielded the pure 20*R* epimer (14, 58%): mp 130–133 °C (lit. mp 130–132, 131–133 °C);[19] IR 1713, 1710, 1250 cm^{-1}; NMR δ 0.67

(26) A. McIntosh, E. Meinzer, and R. Levin, *J. Am. Chem. Soc.*, 70, 2955 (1948).

(3, s), 0.86 (3, broad t, J = 7.4 Hz), 0.92 (3, d, J = 6.5 Hz), 1.02 (3, s), 2.03 (3, s), 2.11 (3, s), 4.64 (1, m), 5.40 (1, m); MS m/e 456 (M^+), 396 (M^+ - AcOH).

The 20R epimer (**14**, 0.58 g) was dissolved in 6 mL of toluene and added to the phosphorane prepared from 4.4 g of methoxymethyltriphenylphosphonium chloride. The mixture was kept for 3 h at room temperature, then the product was isolated by chromatography. The acetate group was cleaved in the reaction and the product was a mixture of E and Z isomers (65:35). The yield was 0.54 g, 92%.

The product was dissolved in ether (20 mL) and 30 drops of 70% perchloric acid added. The mixture was stirred for 3 h, then poured into water, and the ether layer was separated and washed with sodium bicarbonate solution and water. The ether layer was dried ($MgSO_4$) and the solvent removed, leaving the aldehyde (0.49 g). Because of its instability, the aldehyde was dissolved immediately in tetrahydrofuran–2-propanol (1:1, 20 mL) and treated with sodium borohydride (0.4 g). After stirring overnight at room temperature, the reaction mixture was added to water. Dilute hydrochloric acid was next added and the mixture extracted with ether. The ether extract yielded 0.48 g of the diol **9** which was recrystallized from methanol–chloroform. The pure diol (0.45 g, 81% from **14**) had mp 172–174 °C; IR 3300, 2940, 1463, 1389, 1056 cm^{-1}; NMR δ 0.67 (3, s), ~0.9 (H-21, H-27, and H-29, m), 0.98 (3, s), 1.57 (2, OH which disappeared on adding D_2O), 3.48 (3, m, ABX pattern), 5.33 (1, m); MS m/e 430 (M^+), 412 (M^+ - H_2O).

5-(2′-Tetrahydropyranyloxy)-3-isopropylpentyltriphenylphosphonium Bromide. To a stirred solution of 50.4 g (0.6 mol) of cyanoacetamide in 300 mL of water at 0 °C was added dropwise 21.6 g (0.3 mol) of 2-methylpropanal, followed by 1.5 mL of piperidine. The mixture was stirred at room temperature for 8 h and the resulting precipitate filtered off and dried (69.9 g, 100%). The product (62 g) was dissolved in 300 mL of concentrated hydrochloric acid and 300 mL of water added. The mixture was refluxed for 10 h and cooled and sodium chloride added so as to saturate the solution. It was extracted with ether (6 × 200 mL), the extracts were combined and dried, and the ether was removed, leaving the crude 3-isopropyl-1,5-pentanedioic acid (38.5 g, 79%), mp 90–96 °C.

A solution of the diacid (104.5 g) in 400 mL of ether–tetrahydrofuran (1:1) was added dropwise to a slurry of 60 g of lithium aluminum hydride in 800 mL of ether at room temperature. The mixture was refluxed for 1 h and cooled, and water (60 mL) was added slowly, followed by 60 mL of 15% sodium hydroxide solution and 180 mL of water. The resulting precipitate was filtered off and the filtrate distilled to remove the ether. The residue was distilled (125–130 °C, 3 Torr) to give 76.4 g (87%) of the diol: NMR δ 0.86 (6, d, J = 7 Hz), 1.49 (6, m), 3.54 (2, hydroxyl), 3.67 (4, t, J = 6 Hz).

To a solution of the diol (1.8 g) in 25 mL of benzene, 2-mL portions of 48% hydrobromic acid were added while the mixture was refluxed. Water was removed by azeotropic distillation after each addition. TLC analysis showed no starting diol remained after 8 mL of the acid had been added. The benzene was distilled off, 0.5 g of potassium carbonate added, and the mixture distilled at atmospheric pressure to give 4-isopropyltetrahydropyran (0.83 g, 54%), bp 161 °C.

A mixture of 10 g of the tetrahydropyran, 11.5 g of acetyl bromide, and 2 g of zinc bromide was stirred for 5 h with occasional warming on a steam bath.[27] The mixture was filtered through a column of silica gel with petroleum ether–ethyl acetate (5:1) and 1-acetoxy-5-bromo-3-isopropylpentane was isolated (19.2 g, 98%): bp 113 °C (2 Torr); NMR δ 0.85 (6, d, J = 6 Hz), 1.67 (6, m), 2.03 (3, s), 3.40 (2, t, J = 7 Hz), 4.07 (2, t, J = 7 Hz).

A stirred mixture of 5 g (0.019 mol) of triphenylphosphine, 7.5 g (0.03 mol) of bromoacetate, and 2 g of calcium carbonate was heated at 120 °C for 1.75 h. The mixture was cooled and chloroform added and the calcium carbonate filtered off. Addition of ether to the filtrate precipitated the phosphonium salt. The chloroform (and ether) was decanted and the residue redissolved in fresh chloroform and precipitated once more. The crude product (glass, 9.5 g) was hydrolyzed by dissolving it in methanol (125 mL) and 48% hydrobromic acid (1 mL) and letting it stand overnight. TLC indicated the presence of one major component. Removal of the solvent yielded 8.7 g (97%) of the crude alcohol (glass): NMR δ 0.75 (6, d, J = 6 Hz), 1.72 (6, m), 3.70 (4, m), 4.63 (1, hydroxyl), 7.85 (15, m).

The alcohol (25.6 g), dihydropyran (20 g), and p-toluenesulfonic acid (400 mg) were dissolved in 250 mL of dichloromethane. The solution was kept at room temperature for 48 h and the solvent then removed in vacuo. The viscous residue was washed with ether (6 × 100 mL), then it was dissolved in dichloromethane and chromatographed (methanol (3–14%)–ethyl acetate). The crude salt **13** was obtained as a glass (10.3 g, 35%). Because of instability it was used directly in the Wittig reaction; NMR δ 0.73 (3, d, J = 7 Hz), 0.82 (3, d, J = 7 Hz), 1.65 (12, m), 3.72 (6, m), 4.57 (1, m), 7.88 (15, m).

(24ξ)-3β,29-Dihydroxystigmast-5-ene (10). A mixture of the phosphonium salt (7.2 g) and potassium *tert*-amylate in toluene was refluxed for 10 min, then 0.55 g of pregnenolone tetrahydropyranyl ether in 1.5 mL of toluene was added. The mixture was refluxed for 29 h (no more starting steroid remaining) and the product isolated in the usual way. It was dissolved in tetrahydrofuran–methanol–water (4:4:1) containing a trace of 6 N hydrochloric acid and kept for 24 h. Removal of the solvent and chromatography of the residue yielded 0.49 g (82%) of the diol **16**: mp 138–141 °C; NMR δ 0.57 (3, s), 0.86 (6, d, J = 7 Hz), 1.01 (3, s), 1.63 (3, s), 3.68 (2, t, J = 7 Hz), 5.19 (1, t, J = 7 Hz), 5.37 (1, m); MS m/e 428 (M^+), 413, 410, 395, 299.

A solution of 0.64 g of the diacetate (prepared with acetic anhydride–pyridine) in 10 mL of dioxane–acetic acid (50:1) was hydrogenated with 0.1 g of PtO_2. More PtO_2 was added twice daily (0.05 g). After 4 days the catalyst was filtered off and the product hydrolyzed with dilute potassium hydroxide in tetrahydrofuran–methanol–water. The product (0.46 g) was a mixture of 20R and 20S epimers (74:26) as indicated by GC. On repeated recrystallization from dichloromethane–petroleum ether the pure 20R diol **10** was obtained: mp 169–171 °C; NMR δ 0.68 (3, s), 0.86 (6, d, J = 7 Hz), 0.92 (3, d, J = 6 Hz), 1.00 (3, s), 1.51 (2, hydroxyl), 3.64 (2, t, J = 6 Hz), 5.34 (1, m); MS m/e 430 (M^+), 415, 397, 345. Anal. Calcd for $C_{29}H_{50}O_2$: C, 80.87; H, 11.70; O, 7.43. Found: C, 80.63; H, 11.83; O, 7.29.

(24ξ)-3β-Hydroxystigmasta-5,25-diene (Clerosterol) (15). The 20R steroid (**14**, 0.1 g), dissolved in 2 mL of dry toluene, was added to a solution of the phosphorane prepared from 0.62 g of methyltriphenylphosphonium iodide and potassium *tert*-amylate in toluene. The mixture was refluxed for 2 h and the product isolated by chromatography. A yield of 0.076 g (84%) was obtained. The steroid **15** was recrystallized from dichloromethane–methanol. The C-24 epimers could not be separated by GC: mp 127–131 °C (lit. 24S epimer sinters at 130 °C, mp 147 °C);[19] NMR δ 0.68 (3, s), 0.80 (3, t, J = 7 Hz), 0.91 (1.5, d, J = 6 Hz), 0.92 (1.5, d, J = 6 Hz), 1.01 (3, s), 1.57 (3, s), 4.64 (1, broad s), 4.72 (1, broad s), 5.34 (1, m); MS m/e 412 (M^+), 397, 394, 379, 328 (M - C_6H_{12}).

Acknowledgment. This work was supported by Grant GM 21350 from the National Institutes of Health. We thank Professor T. W. Goodwin (University of Liverpool) for a sample of clerosterol.

Registry No. 3, 59905-87-2; **3** acetate, 54548-85-5; **4**, 71436-71-0; **5**, 60084-34-6; (*E*)-**6**, 71484-75-8; (*Z*)-**6**, 71484-76-9; **7**, 71436-72-1; **7** diol, 71436-73-2; **8**, 53139-44-9; **9**, 66760-37-0; **9** aldehyde, 71436-74-3; **10**, 66760-36-9; (20*S*)-**10**, 66791-72-8; **11**, 66760-33-6; **12**, 71436-75-4; **12** keto alcohol, 71484-77-0; **13**, 66760-34-7; **14**, 38863-94-4; (20*S*)-**14**, 66791-73-9; **15**, 62776-91-4; **16**, 71484-78-1; **16** diacetate, 71500-49-7; pregnenolone tetrahydropyronyl ether, 35961-41-2; 6β-methoxy-3,5-cyclo-5α-pregnan-20-one, 32249-55-1; 3β-hydroxydinorchol-5-en-24-al, 53906-49-3; (20*R*)-3β-hydroxydinorchol-5-en-24-al, 71484-79-2; (20*R*)-cholesteryl acetate, 604-35-3; (20*S*)-cholesteryl acetate, 38774-63-9; (20*R*)-3β,24-diacetoxychol-5-ene, 71129-66-3; (20*S*)-3β,24-diacetoxychol-5-ene, 71563-55-8; (*E*)-(24ξ)-3β-hydroxy-5,25-diene-26-methoxystigmastane, 71436-76-5; (*Z*)-(24ξ)-3β-hydroxy-5,25-diene-26-methoxystigmastane, 71436-77-6; [3-ethyl-3-(2-methyl-1,3-dioxolan-2-yl)propyl]triphenylphosphonium bromide, 71436-78-7; [5-(2′-tetrahydropyranyloxy)-3-isopropylpentyl]triphenylphosphonium bromide, 71436-79-8; (5-acetoxy-3-isopropylpentyl)triphenylphosphonium bromide, 71436-80-1; (5-hydroxy-3-

(27) D. E. Ames and P. J. Islip, *J. Chem. Soc.*, 4363 (1963).

isopropylpentyl)triphenylphosphonium bromide, 71436-81-2; (3-tetrahydropyranyloxypropyl)triphenylphosphonium bromide, 70665-02-0; isohexyltriphenylphosphonium bromide, 70240-41-4; (methoxymethyl)triphenylphosphonium chloride, 4009-98-7; (3-tetrahydropyranyloxypropylidene)triphenylphosphorane, 71436-82-3; [3-(2-methyl-1,3-dioxolan-2-yl)propylidene]triphenylphosphorane, 3054-93-1; (4-methylpentylidene)triphenylphosphorane, 54517-55-4; 3-methylbutylidenetriphenylphosphorane, 39110-24-2; methoxymethylidenetriphenylphosphorane, 20763-19-3; 4-isopropyltetrahydropyran, 66760-31-4; 3-isopropyl-1,5-pentanedioic acid, 4165-99-5; 3-isopropyl-1,5-pentanediol, 61898-54-2; 1-acetoxy-5-bromo-3-isopropylpentane, 66760-27-8; 1-bromo-3-ethylpentan-4-one, 66760-26-7; 3-bromo-1-propanol tetrahydropyranyl ether, 33821-94-2; pregnenolone, 145-13-1; 3-bromo-1-propanol, 627-18-9; dihydropyran, 110-87-2; triphenylphosphine, 603-35-0; 2-acetylbutyrolactone, 517-23-7; 2-acetyl-2-ethylbutyrolactone, 31770-00-0; cyanoacetamide, 107-91-5; 2-methylpropanal, 78-84-2; methyltriphenylphosphonium iodide, 2065-66-9.

A New Class of Antitumor Compounds: 5′-Nor and 5′,6′-Seco Derivatives of Vinblastine-Type Alkaloids

Pierre Mangeney, R. Zo Andriamialisoa, Nicole Langlois, Yves Langlois, and Pierre Potier*

Institut de Chimie des Substances Naturelles du Centre National de la Recherche Scientifique, 91190-Gif-sur-Yvette, France

Received March 16, 1979

The reaction medium obtained after applying the modified Polonovski reaction to the N_b-oxide of anhydrovinblastine (3) led mainly to seco C_5–C_6 derivatives after treatment with various nucleophiles. In the presence of cyanide ion, a minor product formed by nucleophilic attack onto the conjugated immonium salt 7 was also isolated. On the other hand, 20′-deoxyleurosidine N_b-oxide (20) led only to seco C_5–C_6 derivatives.

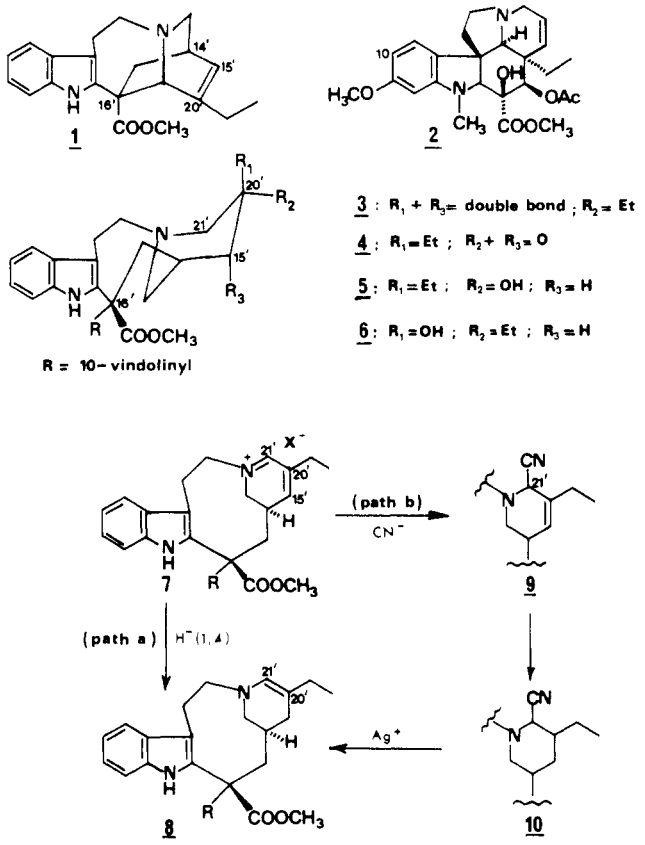
The discovery in our laboratory[1] of a coupling reaction, based on the modified Polonovski reaction[2] and leading to $\Delta^{15'(20')}$-20′-deoxyvinblastine (anhydrovinblastine, 3) from catharanthine (1) and vindoline (2), has stimulated a considerable amount of work aimed at the partial synthesis of leurosine (4),[3] leurosidine (5),[4] and vinblastine (6),[5] the main antitumor alkaloids extracted from various *Catharanthus* species.

The most fruitful approach for the preparation of these complex alkaloids took advantage of several stereoelectronic factors for the direct functionalization of carbon $C_{20'}$ and/or $C_{15'}$ of anhydrovinblastine (3) or other intermediates formed during the coupling reaction.

The conjugated immonium salt 7[6] appeared to be an interesting intermediate which could lead to $\Delta^{20'}$-20′-deoxyvinblastine (8)[7] either by a 1,4 reduction (path a, Scheme I) or by addition of cyanide ion at $C_{21'}$, followed by reduction of the remaining $C_{15'}$–$C_{20'}$ double bond and treatment with silver ion (path b, Scheme I).

The modified Polonovski reaction also appeared to be well suited for the preparation of the conjugated immonium salt 7 using anhydrovinblastine N_b-oxide (11) as the starting material. One of the two allylic protons at $C_{21'}$ would be regioselectively eliminated, giving rise to the desired conjugated immonium salt 7 via the trifluoroacetoxyammonium salt 12 (Scheme II).

Scheme I



Anhydrovinblastine N_b-oxide 11 was exposed to modified Polonovski reaction conditions (TFAA–CH_2Cl_2) and treated after evaporation with several reagents known to allow 1,4 reduction, such as formic acid in pyridine[8] or potassium formate in the presence of 18-crown-6.[9] All of

(1) (a) P. Potier, N. Langlois, Y. Langlois, and F. Gueritte, *J. Chem. Soc., Chem. Commun.*, 670 (1975); (b) N. Langlois, F. Gueritte, Y. Langlois, and P. Potier, *J. Am. Chem. Soc.*, **98**, 7017 (1976).

(2) P. Potier, *Rev. Latinoamer. Quim.*, **9**, 47 (1978), and references therein.

(3) (a) Y. Langlois, N. Langlois, P. Mangeney, and P. Potier, *Tetrahedron Lett.*, 3945 (1976); (b) N. Langlois and P. Potier, *J. Chem. Soc., Chem. Commun.*, 102 (1978); (c) J. P. Kutney, J. Balsevitch, G. H. Bokelman, T. Hibino, I. Itoh, and A. H. Ratcliffe, *Heterocycles*, **4**, 997 (1976).

(4) N. Langlois and P. Potier, *Tetrahedron Lett.*, 1099 (1976).

(5) (a) A. U. Rahman, A. Basha, and M. Ghazala, *Tetrahedron Lett.*, 2351 (1976); (b) P. Mangeney, R. Zo Andriamialisoa, N. Langlois, Y. Langlois, and P. Potier, *J. Am. Chem. Soc.*, in press.

(6) Obviously, this conjugated immonium salt 7 can be obtained directly from the coupling reaction between vindoline (2) and catharanthine *N*-oxide, but in this case it is always contaminated with unreacted vindoline and other byproducts.

(7) A procedure for the preparation of the enamine 8 has been described (see ref 5b).

(8) L. R. Gizzi and M. M. Joullié, *Tetrahedron Lett.*, 3117 (1969).

(9) Y. Ohnishi and S. Tamimoto, *Tetrahedron Lett.*, 1909 (1977).

0022-3263/79/1944-3765$01.00/0