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(1) R. A. Moss, C. J. Talkowski, D. W. Reger, and C. E. Powell, *J. Am. Chem. Soc.*, **95**, 5215 (1973).

(2) Product ratios are given in the Ph.D. Theses of H. J. Ratajczak and G. Rauleder, Ruhr-Universität Bochum, 1975. The extent of hydride shift (and of other rearrangements) increased with concentration (F_m) at the expense of direct displacement. E.g., with **1d** the extreme values (percent of alcohol fraction) were 52.4 → 36.8% **2d** and 38.7 → 52.5% **3d** (F_m 0 → 1).

(3) The configuration of (−)-**5** is *R*: B. W. Christensen and A. Kjaer, *Acta Chem. Scand.*, **16**, 2466 (1962).

(4) T. D. Inch, G. J. Lewis, G. L. Sainsbury, and D. J. Sellers, *Tetrahedron Lett.*, 3657 (1969).

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(6) The deamination of **1d** has been studied previously: W. Kirmse and H. Arold, *Chem. Ber.*, **104**, 1800 (1971). Although the results are in qualitative agreement, the previously reported rotations have not been reproduced in the present work and may have been due to small impurities of high specific rotation. $[\alpha]^{20}D$ of **3d** is only 0.78°. In the present work, optical purities have been estimated both by polarimetry and by NMR in the presence of tris(3-(trifluoromethylhydromethylene)-*d*-camphorato)europium(III) = Optishift II. Satisfactory agreement was reached with all samples, but only after multiple GLPC in the case of **3d**.

(7) The cmc's given refer to deamination conditions;[1] without added sodium salts the cmc of **1a** is 0.72 *M*, and that of **1d** 0.25 *M*.

(8) Hydride shifts induced by deamination proceed with predominant inversion at the migration terminus: W. Kirmse, H. Arold, and B. Kornrumpf, *Chem. Ber.*, **104**, 1783 (1971); W. Kirmse and D. Krause, *ibid.*, in press.

(9) Counterion effects are also in accord with the model of micellar control. The stereochemistry of **7** was independent of concentration (49 ± 1% retention) when chloride was the counterion in the deamination of **6a,b**.

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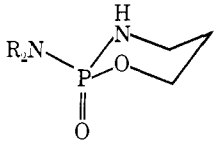
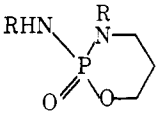
Molecular Structure of the Carcinostat Isophosphamide

Sir:

Among the diverse properties of Cyclophosphamide (**1**) (Cytoxan, Endoxan) are its potent carcinostatic action in the treatment of human cancers and its strong immunosuppressive properties in tissue transplants.[1]



1



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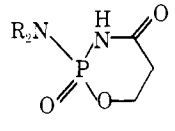

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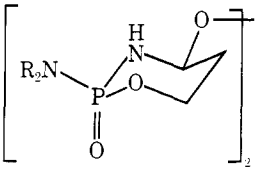


4

R = CH_2CH_2Cl

Isophosphamide (**2**) (Ifosfamide) an isomer of **1** also shows considerable promise in these respects,[2] and similar metabolic pathways have been found.[3]

Recent X-ray studies on **1**[4] and two of its metabolites, **3**[5] and **4**,[6] have shown that the P=O group is axial in each case although the ring is substantially buckled away from a chair form in **3** owing to the presence of the sp^2 carbonyl carbon. Numerous experimental results have been summarized which show that in solution some phosphorus substituents prefer the axial position in 2-oxo-1,3,2-dioxaphosphorinanes (**5a**) while others tend to be equatorial (**5b**). Although the R group in trivalent analogs of **5** prefers the equatorial position in solution when R = Me_2N (**6a**),[7] it has come to our attention that the phosphorus substituent displays a greater tendency to be axial (**6b**) when it is Me,[8] PhNH,[9a] or *t*-BuNH.[9b] This observation has been ascribed

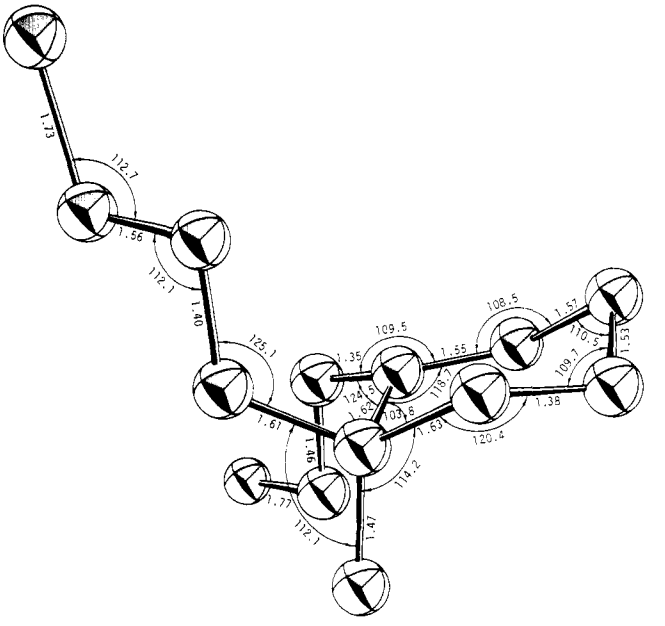
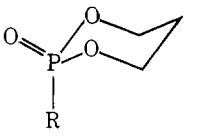
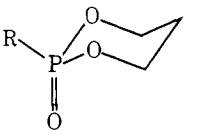


Figure 1. Isophosphamide.

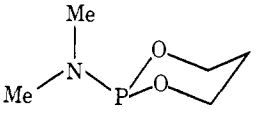


5a

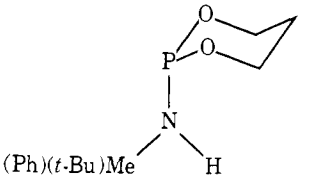


5b

R = H, OMe, halogen R = alkyl, Ph, dialkylamino



6a



6b

to the small steric requirements of an NH hydrogen under the ring coupled with the strong proclivity of the phosphorus lone pair to be equatorial.[7-10] Because the P=O group and the phosphorus lone pair in these ring systems prefer the equatorial position in the absence of steric effects of the R group,[7-10] it was of considerable interest to determine the orientation of the $ClCH_2CH_2NH$ group in Isophosphamide (**2**) since stereochemical inversion at phosphorus has been shown to be accompanied by distinct changes in chemical properties.[7]

Crystals of **2** were grown by slowly cooling a saturated boiling ether solution. A single crystal was selected for X-ray diffraction study and assigned to the orthorhombic space group P_{bca} after analysis of the Laue symmetry and the observed systematic absences. The unit cell parameters are a = 13.29 (1), b = 21.16 (1), and c = 8.78 (1) Å. Intensity data were collected on a four-circle diffractometer equipped with a scintillation counter. Positions of all nonhydrogen atoms were determined by a combination of Patterson analysis and subsequent electron density map calculation. Figure 1 shows a computer drawing of the molecular

structure (excluding hydrogens) resulting from preliminary refinement to a conventional crystallographic discrepancy factor of $R = 16.2\%$, based on 535 intensities with $I_N > 2\sigma(I)$.

Despite the less bulky nature of the $ClCH_2CH_2NH$ moiety compared to the $(Cl_2CH_2CH_2)_2N$ group, the conformation of **2** in the solid state is the same as that of **1.** At this stage of refinement, detailed comparisons of the bond lengths of **2** to those of **1** are not warranted; however, no unusual deviations in these parameters are apparent at this time. As is the case with virtually all $P-NR_2$ systems including **1** and **3,** the angles around both nitrogens in **2** suggest trigonal planar nitrogen geometries. With but one exception, solution studies on phosphorinane systems of type **5a** and **5b** indicated that the dominant solution conformer is also that found in the solid state as revealed in nine X-ray studies.[7] Solution spectroscopic and dipole moment measurements are underway in an effort to clarify the conformational character of **1–3** in solution.

Acknowledgments. J.G.V. thanks the National Cancer Institute and the National Science Foundation for generous grant support of this work. The authors thank the Drug Development Branch, Drug Research and Development, Division of Cancer Treatment, NCI for a sample of Isophosphamide, and Dr. S. J. Dykstra of the Mead Johnson Research Center for valuable discussions. We thank Professor Bentrude for sending us a preprint of ref 8.

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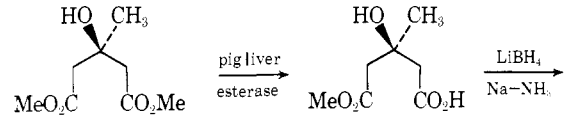
Preparation of (*R*)- and (*S*)-Mevalonic Acids

Sir:

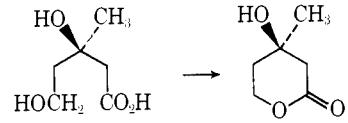
We report new, simple syntheses[1] of (*R*)- and (*S*)-mevalonic acids by a combination of enzymatic and chemical procedures. The methods offer the particular advantages of utilizing readily available starting materials and enzyme preparations, and further deomonstrate the usefulness of biochemical systems as *organic* chiral reagents in asymmetric synthesis.

It has been shown that α-chymotrypsin hydrolyzes the pro-*S* ester group of dimethyl β-hydroxy glutarate.[2] We have treated β-hydroxy-β-methyl dimethyl glutarate (**2**) with α-chymotrypsin, and obtained the (3*S*)-half ester **3a** of high optical purity (see below). However, the rate of hydrolysis was slow and virtually a stoichiometric amount of the enzyme was required. In contrast, pig liver esterase (purchased from Sigma Co.) hydrolyzed **2** more efficiently and the same half ester (**3a**) of high optical purity was formed. In a typical experiment, **2** (400 mg) in 0.1 *M* phosphate buffer (pH 8.0) (7 ml), was incubated with the esterase (432 units) at 25° for 3 hr. Following the usual work-up **3a** (218 mg; 62%), $[\alpha]^{26}D$ +0.72 (CH_3OH), was obtained, and its absolute configuration and optical purity were determined by conversion to mevalonolactone.

Reduction of **3a** with either $LiBH_4$[3] or Na in liquid ammonia–ethanol[4] afforded (*R*)-mevalonolactone (**4**) in yields of 81 and 73%, respectively. Both samples of **4** were converted to their benzhydrylamide derivatives which had identical physical constants:[5] mp 98–99°, $[\alpha]^{26}D$ −2.79° (EtOH). Since $LiBH_4$ and $Na-NH_3$ are known to reduce selectively carboxylic esters but not carboxylic acids,[3,4] it follows that the pig liver esterase cleaved the pro-*R* methyl ester group of **2** giving **3a.**

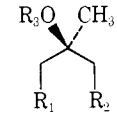


2 3a



4

It is worthy of note that when **3a** was treated with diborane at 0° for 1 hr, it also gave (*R*)-mevalonolactone (41%) (benzhydrylamide, mp 97–98°, $[\alpha]^{26}D$ −1.98 (EtOH)) (71% optical purity). Since it is generally accepted that diborane reduces preferentially carboxylic acids,[6,7] this could indicate that the pig liver esterase has hydrolyzed the pro-*S* methyl ester group of **2.** If correct, the finding would contradict the conclusions derived from the above $LiBH_4$ and $Na-NH_3$ experiments. The anomaly was clarified when the 3-acetoxy derivative (**3c**), $[\alpha]^{26}D$ −0.42 (CH_3OH) (obtained from **3a**), was reduced with diborane. In this instance, (*S*)-mevalonolactone (benzhydrylamide, mp 98–99°, $[\alpha]^{26}D$ +2.78 (EtOH)) was obtained. This confirmed the (**3a**) structure and hence the stereospecific hydrolysis of the pro-*R* methyl ester group of **2** by pig liver esterase. The reason for this abnormal behavior is not clear, but the results do clearly show that the presence of the free hydroxyl function at C-3 interfered with the normal course of borane reduction.



1, $R_1 = R_2 = CH_2OH$; $R_3 = H$
2, $R_1 = R_2 = CO_2Me$; $R_3 = H$
3a, $R_1 = CO_2Me$; $R_2 = CO_2H$; $R_3 = H$
b, $R_1 = CO_2H$; $R_2 = CO_2Me$; $R_3 = H$
c, $R_1 = CO_2Me$; $R_2 = CO_2H$; $R_3 = Ac$

An alternative approach to the synthesis of mevalonolactone was based on the ability of *Flavobacterium oxydans* to oxidize a wide variety of gem hydroxymethyl compounds to