acid-catalyzed conditions which have proved effective with 17α-hydroxy-20-keto steroids,[31] but only unreacted starting material was recovered.

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[31] *Cf.* Huang-Minlon, E. Wilson, N. L. Wendler and M. Tishler, THIS JOURNAL, **74**, 5394 (1952); R. B. Turner, *ibid.*, **75**, 3489 (1953).

Acknowledgment.—We should like to acknowledge the benefit of a stimulating discussion with Prof. Gilbert Stork of Columbia University.

DETROIT, MICHIGAN

COMMUNICATIONS TO THE EDITOR

THE RESOLUTION OF O-ETHYL ETHYLPHOSPHONOTHIOLIC ACID

Sir:

We wish to record the successful resolution of O-ethyl ethylphosphonothiolic acid ($C_2H_5(C_2H_5O)P(O)SH$).[1] The compound was resolved by fractional recrystallization of its quinine salt (I) from acetone–ether. The more insoluble diastereoisomeric salt (Ia) crystallized as a monohydrate: prisms, m.p. 151–153° (with loss of its water of hydration), $[\alpha]^{26}\text{D}\ -96.6 \pm 0.8°$ ($\alpha_{\text{obs}}\ -1.990 \pm 0.015°$, acetone, 2-dcm., $c = 1.130$), equiv. wt., 492 (calcd. 497 for the monohydrate). When vacuum dried over phosphorus pentoxide for three hours at 100°, Ia gave rise to anhydrous product: m.p. 158–160°, $[\alpha]^{33}\text{D}\ -97.6 \pm 0.6°$ ($\alpha_{\text{obs}}\ -1.070 \pm 0.007°$, acetone, 1-dcm., $c = 1.096$), equiv. wt., 469 (calcd. 479).

The acid was separated from Ia as its sodium salt in an essentially aqueous solution by treating Ia in methanol with an equivalent amount of aqueous base. That the phosphorus atom maintains its tetrahedral configuration in the anion is demonstrated by the optical activity of the product recovered from the sodium salt. The acid was recovered by the addition of an equivalent amount of dilute hydrochloric acid to the sodium salt solution and extraction of the product from the resulting solution with ether. The acid was characterized as its dicyclohexylamine salt: m.p. 159–160.5° $[\alpha]^{25}\text{D}\ -7.11 \pm 0.23°$ ($\alpha_{\text{obs}}\ -0.153 \pm 0.005°$, methanol, 1-dcm., $c = 2.150$), found: C, 57.38; H, 10.00 (calcd. for $C_{16}H_{34}O_3NP$: C, 57.28; H, 10.22).

After the removal of a mixed middle crop of I, the more soluble diastereoisomeric salt (Ib) crystallized as soft anhydrous needles: m.p. 166–168°, $[\alpha]^{25}\text{D}\ -81.7 \pm 0.6°$ ($\alpha_{\text{obs}}\ -1.613 \pm 0.012$, acetone, 2-dcm., $c = 0.9868$); equiv. wt., 475 (calcd. 479). The dicyclohexylamine salt of this enantiomorph of the acid gave m.p. 158–160°, $(\alpha)^{25}\text{D}\ +6.85 \pm 0.25°$ ($\alpha_{\text{obs}} + 0.221 \pm 0.008°$, methanol, 1-dcm., $c = 3.230$), found: C, 57.30, H 10.02, mixed melting point with the enantiomorphic dicyclohexylamine salt, above: 163–165°. Racemic O-ethyl ethylphosphonothiolic acid forms a dicyclohexylamine salt, m.p. 166–168°.

This communication represents the first reported resolution of a phosphorus acid, the optical activity of which is due solely to the presence of an asymmetric phosphorus atom. Moreover, the presence of a reactive group directly attached to phosphorus in a resolved compound of this type provides one with a convenient tool applicable to a study of the reactions and stereochemistry of the asymmetric phosphorus atom. Detailed reports on the resolution, reactions and stereochemistry of this and similar compounds will be published at a later date.

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RECEIVED MAY 21, 1956

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[1] The preparation of alkylphosphonothiolic acids will be described in a forthcoming paper by F. W. Hoffmann and co-workers.

ELECTROPHORETIC DEMONSTRATION OF THE ISOMERIZATION OF BOVINE PLASMA ALBUMIN AT LOW *p*H

Sir:

Recently much interest has been exhibited in a pronounced conformational change which takes place in bovine plasma albumin at *p*H values acid to the isoelectric point. It was first suggested by Tanford[1] that expansion of the protein molecule results upon titration with acid. Gutfreund and Sturtevant[2] demonstrated a slow thermal effect upon adding acid to this protein. Yang and Foster[3] demonstrated a parallel and reversible enhancement of the optical rotation and intrinsic viscosity acid to *p*H 4, and suggested that there exists an all-or-none equilibrium between two forms of the protein molecule. Tanford[4] has recently shown evidence for an intermediate which he terms the "expandable" form.

We have recently been successful in attaining excellent resolution of two boundaries in the electrophoretic patterns of this protein over the *p*H range 4.6 to 3.5. Heterogeneity of plasma albumins in this *p*H range has been reported previously.[5-8] However, we can now demonstrate that this heterogeneity is due in the main to a *p*H dependent transition of the normal form of the protein into a faster migrating form, presumably of higher positive charge. Results summarized in

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[1] C. Tanford, *Proc. Iowa Acad. Sci.*, **59**, 206 (1952).

[2] H. Gutfreund and J. Sturtevant, THIS JOURNAL, **75**, 5447 (1953).

[3] J. T. Yang and J. F. Foster, *ibid.*, **76**, 1588 (1954); **77**, 2374, 3895 (1955).

[4] C. Tanford, J. Buzzell, D. Rands and S. Swanson, *ibid.*, **77**, 6421 (1955).

[5] J. Luetscher, *ibid.*, **61**, 2888 (1939).

[6] D. Sharp, G. Cooper, J. Erickson and H. Neurath, *J. Biol. Chem.*, **144**, 139 (1942). In this paper it was also shown that heterogeneity disappears below *p*H 3.5, in accord with our own results.

[7] R. Alberty, *J. Phys. Chem.*, **53**, 114 (1949).

[8] L. Longsworth and C. Jacobsen, *ibid.*, **53**, 126 (1949).