soluble in 5% trichloroacetic acid solution after being kept in boiling water for thirty minutes: the solubility increased from 5 to 10%, indicating that some degradation occurred during the acid-heat treatment. The mechanism of the enhancement of activity is being studied. We are also investigating the chemical changes in the ACTH peptides after activation with acid-heat treatment.

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PREPARATION AND PROPERTIES OF AN ACID GLYCOPROTEIN PREPARED FROM HUMAN PLASMA

Sir:

An acid glycoprotein has been isolated and crystallized from Fraction VI[1] of pooled normal human plasma. Its preparation and some properties are described herewith.

The solution (Fractions VI + VII) from which the major human plasma proteins[1] have been precipitated from an ethanol–water mixture of mole fraction 0.066, at −5°, at *p*H 5.8 was adjusted to *p*H 7.5 using an ammonium hydroxide–ammonium chloride buffer of *p*H 10. The proteins in solution were adsorbed and carried down by the zinc hydroxide formed. After centrifuging, the precipitate was resuspended in an equal volume of 0.066 mole fraction ethanol at −5° and the *p*H readjusted to 5.8. Insoluble material was removed and barium acetate added to the solution to a concentration of 0.02 *M*, and the *p*H brought to 6.1. The precipitate which formed was separated. An α_1-globulin constituted 93% of the protein remaining in the solution. It was precipitated by increasing the ethanol to mole fraction 0.136 and decreasing the temperature to −18°.

This protein was further purified by precipitation of other proteins concentrated by the above procedure in 0.066 mole fraction ethanol at *p*H 5.8, 0.02 *M* zinc and 0.02 *M* barium at −5°. The protein remaining in the concentrated solution was homogeneous by electrophoresis between *p*H 2.3 and 8.6, and in the ultracentrifuge at *p*H 2.3 and 6.1. Certain properties of this protein are given in Table I.

TABLE I
CONSTANTS FOR THE ACID GLYCOPROTEIN

Nitrogen, %	10.7
Hexose, %	17
Hexosamine, %	12
Phosphoric acid, %	1.2
$E^{1\%}_{1\,\text{cm.}}$ at 278 mμ	8.93
Isoelectric point, *p*H	2.9–3.0
Electrophoretic mobility, sq. cm./volt sec. × 10^{-5}	
*p*H 8.6, Γ/2 0.1, barbiturate	−5.2
*p*H 4.0, Γ/2 0.1, acetate	−2.0
Sedimentation constant, $S_{20,w}$, at infinite dilution	3.5

Hexuronic acid, fatty acid, cholesterol, phospholipid, free SH-groups and esters of sulfuric acid were not found. The acid glycoprotein could be precipitated from aqueous solution by saturation with ammonium sulfate or monosodium phosphate or by addition of 5% phosphotungstic acid in 2 *N* hydrochloric acid. It was not precipitated by addition of 1.8 *M* perchloric acid, 0.06 *M* sulfosalicylic acid, 20% trichloroacetic acid, or by boiling.

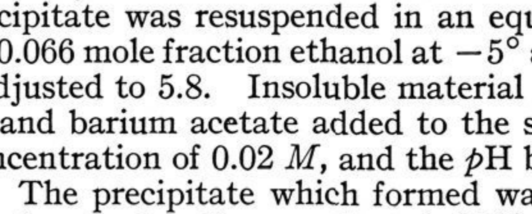

Fig. 1.—Crystals of acid glycoprotein (magnification × 660).

Crystals of this acid glycoprotein (Fig. 1) were obtained under the following conditions: protein 6%, 0.0072 *M* lead acetate, *p*H 5.4, Γ/2 0.02, methanol 10% and acetone 10% at 0°.

Further details of these investigations will be reported subsequently.

I am indebted to Professor E. J. Cohn for his generous advice throughout these studies.

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[1] E. J. Cohn, *et al.*, THIS JOURNAL, **72**, 465 (1950).

CONDENSATION OF METHYLGLYOXAL WITH β-OXO ACIDS

Sir:

The paper of M. S. Schechter, N. Green and F. B. LaForge on "Constituents of Pyrethrum Flowers. XXIII. Cinerolone and the Synthesis of Related Cyclopentenolones,"[1] describes condensations of methylglyoxal with β-oxo acids in aqueous solution at *p*H 4.9–8.0 and room temperature, *i.e.*, under so-called "physiological conditions," whereby 2-hydroxy-1,4-diketones are formed under decarboxylation. The authors say: "We have found that the decarboxylation proceeds spontaneously under the conditions of the reaction, the final product being the hydroxydiketone,..."

The fact that spontaneous decarboxylation occurs when condensing aldehydes with β-oxo acids within *p*H range 5–11 was reported first by us in 1932 when we condensed *o*-aminobenzaldehyde with acetoacetic acid, β-oxo-caprylic acid and benzoylacetic acid.[2] We also showed at that time that in a more alkaline solution at

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[1] Schechter, Green and LaForge, THIS JOURNAL, **71**, 3165 (1949).

[2] Schöpf and Lehmann, *Ann.*, **497**, 11 (1932).