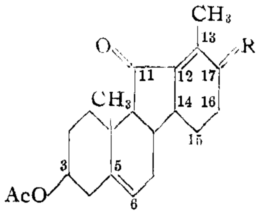
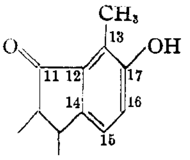
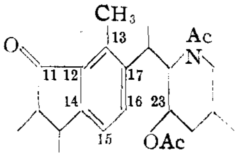
teristics of which ($\lambda^{\text{alc.}}_{\text{max.}}$ 255 mμ (3.98), 330 mμ (3.51)) were identical with those of 6-keto-β-estradiol,[4] inclusive of the shifts produced by alkali and by acetylation of the phenolic group. Other compounds derived from II are the 12,13-dihydro derivative (m.p. 171–173°, $[\alpha]^{24}\mathrm{D}$ $-208°$), the free diketo alcohol (m.p. 170–171°, $[\alpha]^{22}\mathrm{D}$ $-220°$), and the Δ^4-3-ketone (m.p. 196–199°; $[\alpha]^{22}\mathrm{D}$ $+8°$), all giving correct analyses.



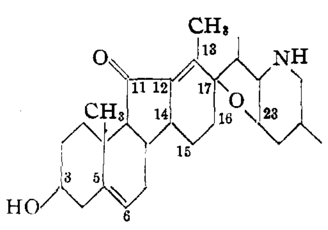
I, R = CHCH$_3$
II, R = O



III



IV



V

O,N-Diacetyljervine with acetic anhydride, acetic acid and sulfuric acid at 24° yielded besides a sulfonic acid the indanone IV (m.p. 239–240°; $[\alpha]^{24}\mathrm{D}$ $-29°$; calcd. for $C_{33}H_{43}O_6N$: C, 72.10; H, 7.89; found: C, 71.82; H, 7.98), identified as such by the ultraviolet spectrum ($\lambda^{\text{alc.}}_{\text{max.}}$ 251 mμ (4.08), 300 mμ (3.30)), and the fact that the amorphous product obtained on hydrogenation (PtO_2, acetic acid, uptake 2 moles) exhibited a spectrum similar to that of neoergosterol ($\lambda^{\text{alc.}}_{\text{max.}}$ 268 mμ (2.63)) which reverted to that of IV on chromic acid oxidation. The acetoxy group formed by acetolysis of the oxidic ring is placed at C_{23} on the strength of the evidence adduced by Jacobs and Sato[2b] and additional facts ascertained by us.

The position of the methyl group (C_{18}) in I–IV is arbitrarily assigned. In all the compounds described the 11-keto group is inert toward keto reagents, as it is in jervine. Such hindrance would not obtain in the analogous perhydrochrysene structures carrying the keto group at C_{12}. The presence in jervine itself of the perhydrobenzfluorene nucleus receives support from the nature of the hydrocarbons $C_{20}H_{16}$ and $C_{22}H_{20}$, evidently homologs of 1,2-benzfluorene, found among the selenium dehydrogenation products.[5] Barring rearrangements in the acetolysis reactions jervine would then appear to be V, or perhaps the $\Delta^{12,14}$-double bond isomer of V. We prefer C_{17} to C_{16} as the other point of attachment of the oxide bridge on account of the ease which which double bond formation takes place at that site not only in the reaction leading to IV but also, as will be reported later, in the sulfuric acid-catalyzed acetolysis of tetrahydrojervine derivatives.

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RECEIVED APRIL 16, 1951

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[4] B. Longwell and O. Wintersteiner, *J. Biol. Chem.*, **133**, 219 (1939).

[5] W. A. Jacobs, L. C. Craig and G. L. Lavine, *ibid.*, **141**, 51 (1941).

IDENTIFICATION OF RIBULOSE IN $C^{14}O_2$ PHOTOSYNTHESIS PRODUCTS[1]

Sir:

The intermediates involved in carbon dioxide fixation by plants are largely phosphorylated hydroxy acids and sugars. A compound observed during the first few seconds of $C^{14}O_2$ photosynthesis in all the plants investigated in this laboratory has now been identified as ribulose (adonose) diphosphate.

The diphosphate ester occupies a paper chromatographic position near that of fructose and glucose 1,6-diphosphates[2] and 2,3-diphosphoglyceric acid. A monophosphate[3] ester which gives the same labeled sugar upon phosphatase ("Polidase") hydrolysis occupies a chromatographic position intermediate between triose phosphates[2] and the hexose monophosphates. In young cultures of *Scenedesmus* the concentration of the diphosphate approaches that of phosphoglycerate.

Independent evidence of the phosphorus content of ribulose diphosphate was obtained from measurements of C^{14}/P^{32} ratios in chromatographically separated compounds derived from *Scenedesmus* saturated with P^{32} (12 hours equilibration in radiophosphate) and C^{14} (35 minutes photosynthesis in $C^{14}O_2$). The measured ratios (samples were counted when the ratios were near unity for optimum accuracy) were all multiplied by an appropriate factor to give 3.0 for phosphoglycerate, 5.1 for glucose monophosphate, 5.8 for fructose plus sedoheptulose monophosphate, and 2.0 for ribulose diphosphate. The calculated value for ribulose diphosphate is 2.5.

The chromatographic position of the radioactive sugar R_f(phenol) = 0.60; R_f(butanol–propionic acid–water[2]) = 0.27, corresponds exactly to that of ribulose prepared by epimerization of ribose or arabinose in pyridine. No common hexoses or tetroses have such a position.

The radioactive sugar resists bromine but is cleaved by oxygen, particularly under basic conditions such as in diethylamine solutions or on anion exchange resins. Radioactive glycolic, glyceric and a polyhydroxy acid (presumably erythronic[4]) are obtained upon air oxidation. These products are those expected from ribulose oxidation. The labeled diphosphate was found to be oxidized by air in diethylamine solutions to give phosphoglyceric and phosphoglycolic acids as major products. These were identified by chromatography of the hydroly-

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[1] This work was sponsored by the United States Atomic Energy Commission.

[2] A. A. Benson, J. A. Bassham, M. Calvin, T. Goodale, V. Haas and W. Stepka, THIS JOURNAL, **72**, 1710 (1950).

[3] B. L. Horecker and P. Z. Smyrniotis, *Arch. Biochem.*, **29**, 232 (1950).

[4] J. U. Nef, O. F. Hedenberg and J. W. E. Glattfeld, THIS JOURNAL, **39**, 1638 (1917).

sis products which were found in the expected positions of glyceric acid and glycolic acid.[5]

The radioactive sugar was epimerized in pyridine.[6] Co-chromatography of the resultant mixture[7] with ribose and arabinose showed identity of the two major radioactive products with the added sugars. The radioactive 2,4-dinitrophenylosazone[8] of D-arabinose was prepared with a tracer quantity of the labeled ribulose. It was found to have the calculated specific activity and this was undiminished by repeated recrystallizations from methyl cellosolve.

The radioactive sugar was catalytically hydrogenated with Adams catalyst and the product was found to co-chromatograph with added ribitol but not with arabitol.

The foregoing observations lead to the conclusion that the radioactive compounds isolated from plants are ribulose 1,5-diphosphate and monophosphate. An examination of the kinetics of formation of this compound from $C^{14}O_2$ during steady state photosynthesis and a discussion of its importance as a C_2 donor in the cycle for regeneration of the CO_2-acceptors will be published.

(5) Phosphoglycolic acid observed as a minor radioactive product of $C^{14}O_2$ photosynthesis has been separated and identified. Oxidation during chromatography may provide one source of this phosphoglycolic acid.

(6) O. T. Schmidt and R. Treiber, *Ber.*, **66B**, 1765 (1933).

(7) The equilibrium mixture of epimers is known to be largely ribose and arabinose (P. A. Levene and R. S. Tipson, *J. Biol. Chem.*, **102**, 563 (1933)). Small amounts of radioactive ribose observed in isolations of ribulose may indicate that it is also present in the original phosphate esters.

(8) C. Nenberg and E. Strauss, *Arch. Biochem.*, **11**, 457 (1946).

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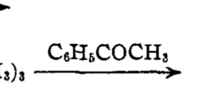
RECEIVED MAY 9, 1951

β-HYDROXY ESTERS FROM KETONES AND ESTERS

Sir:

We have simulated the Reformatsky type of reaction employing *t*-butyl acetate instead of an α-halo ester in accordance with the following equation

$$CH_3COOC(CH_3)_3 \xrightarrow[2.\ ZnCl_2]{1.\ NaNH_2} ClZnCH_2COO(CH_3)_3 \xrightarrow{C_6H_5COCH_3}$$



$$\underset{CH_3}{C_6H_5C}(OH)CH_2COOC(CH_3)_3 \quad I$$

t-Butyl acetate was converted to its sodio derivative by means of sodium amide in liquid ammonia, the liquid ammonia replaced by ether, and an ether solution of anhydrous zinc chloride added at −70°. After stirring forty minutes at this temperature, acetophenone was added and the mixture refluxed two hours. There was obtained a 31% yield of *t*-butyl β-hydroxy-β-phenylbutyrate (I), b.p. 124–127° at 4.5 mm. *Anal.*[1] Calcd. for $C_{14}H_{20}O_3$: C, 71.16; H, 8.53. Found: C, 70.77; H, 8.22. Acid hydrolysis and dehydration yielded β-methylcinnamic acid, m.p. and mixed m.p. with an authentic sample, 97–98°.[2]

In a similar manner, *t*-butyl acetate condensed with cyclohexanone and benzaldehyde to form *t*-butyl 1-hydroxycyclohexylacetate (34% yield, b.p. 132–136° at 18 mm. *Anal.*[1] Calcd. for $C_{12}H_{22}O_3$: C, 67.35; H, 10.36. Found: C, 67.24; H, 10.02), and *t*-butyl β-hydroxy-β-phenylpropionate (58% yield, b.p. 154–158° at 10 mm. *Anal.*[1] Calcd. for $C_{13}H_{18}O_3$: C, 70.33; H, 8.17. Found: C, 69.98; H, 7.83), respectively.

We have further found that this type of aldol condensation may be effected more conveniently and in higher yield by means of lithium amide (without the use of zinc chloride).

$$CH_3COOC(CH_3)_3 \xrightarrow{LiNH_2} LiCH_2COOC(CH_3)_3 \xrightarrow{C_6H_5COCH_3} I$$

The lithium derivative of *t*-butyl acetate was prepared with lithium amide in liquid ammonia, the ammonia replaced by ether, and the acetophenone added immediately. After refluxing two hours, the hydroxy ester (I), b.p. 111–112.5° at 2 mm. was isolated in 76% yield.

This aldol condensation using lithium amide is particularly striking since use of sodium amide under similar conditions failed to give hydroxy ester.

These important types of aldol condensations are being investigated more thoroughly, employing various lithium and other metallic reagents. The results promise to be of both theoretical interest and of practical value.

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RECEIVED APRIL 24, 1951

(1) Analyses by Clark Microanalytical Laboratory, Urbana, Illinois.

(2) Rupe and Busolt, *Ber.*, **40**, 4538 (1907).

(3) Carbide and Carbon Chemicals Fellow.

THE HEAT OF DISSOCIATION OF NITROGEN*

Sir:

In recent publications[1,2,3,4] each of the four spectroscopically acceptable nitrogen dissociation energies, 7.383, 8.573, 9.764 and 11.8 ev. has been again recommended as the correct one. The study of stationary detonation waves provides information on equilibria at high temperatures because Zeldovitch[5] and v. Neumann[6] conclusively show on solid hydro-thermodynamic basis that the properties of an infinite plane wave without rarefaction are determined by the state of complete thermodynamic equilibrium of explosion products. The experimental problem consists largely in adducing the proof that a finite wave has the same velocity as the ideal one. We had been thus engaged to redetermine the heats of sublimation of carbon and of the dissociation of nitrogen when a paper by Döring and Schön appeared.[7] They compare the calculated

* This research was made possible by funds extended Harvard University under ONR Contracts M5ori-76 to XIX NR-053-094.

(1) L. Pauling, *Proc. Nat. Acad. Sci.*, **35**, 359 (1949).

(2) L. Brewer, L. K. Templeton and F. A. Jenkins, THIS JOURNAL, **73**, 1462 (1951).

(3) A. G. Gaydon, "Dissociation Energies," Chapman and Hall, London, 1947.

(4) G. Glockler, *J. Chem. Phys.*, **19**, 124 (1951).

(5) Ya. B. Zeldovich, *J. Exp. Theor. Phys. (USSR)*, **10**, 542 (1950).

(6) J. v. Neumann, OSRD Report No. 549, May, 1942.

(7) W. Döring and G. Schön, *Z. Elektrochem.*, **54**, 231 (1950).