

We gladly acknowledge the generous support of this work by The Rockefeller Foundation and Standard Brands, Inc., of New York.

Summary

A sensitive and accurate assay for biotin is described which involves the growth of a strain of *Saccharomyces cerevisiae* from a small seeding. Under the test conditions the response of this organism to biotin is quantitative within a concentration range from 0.00002 to about 0.001 γ of

biotin. Assay results on crude extracts agree well at different levels within this range, and the curves obtained are very similar to those obtained with pure biotin. Assay results on a number of natural materials are given.

The effectiveness of biotin under these conditions depends to a large extent on the presence of vitamin B₆ (and of β -alanine) in the medium. Biotin in liver concentrates is destroyed by nitrous acid at a rate which indicates that it may be an α -amino acid.

AUSTIN, TEXAS

RECEIVED NOVEMBER 28, 1939

[CONTRIBUTION FROM THE RESEARCH LABORATORIES OF THE UNIVERSAL OIL PRODUCTS COMPANY]

Catalytic Hydration of Acetylene and of Some Alkylacetylenes¹

BY RAYMOND E. SCHAAD AND VLADIMIR N. IPATIEFF

Previously sulfuric acid, sulfuric acid with mercury salts, and the latter composite in the presence of organic solvents have been used as catalysts in the hydration of acetylene and of alkylacetylenes to acetaldehyde and ketones, respectively, as reviewed and described by Thomas, Campbell and Hennion.² For the hydration of acetylene several patents claim a similar use of phosphoric acid and mercury salts,³ and of phosphoric acid on charcoal with⁴ and without^{5,6} added metals.

In the present work solid phosphoric acid polymerization catalyst^{7,8,9} was found to catalyze hydration of butyne-1 rather than its polymerization, the water required for the hydration being removed from the catalyst which was thereby decreased in activity. Continuous hydrations of this and of other alkynes, including acetylene, propyne, pentyne-1, pentyne-2, hexyne-1, and heptyne-1, were effected without rapid decrease in catalyst activity by simultaneous passage of

the hydrocarbon and steam through a heated tube containing the granular solid catalyst.

Acetaldehyde formed when steam and acetylene or mixtures of steam with acetylene and ethylene or nitrogen were passed through a tube containing solid phosphoric acid catalyst at 260–300° and atmospheric pressure. Some higher boiling products also formed apparently by condensation of acetaldehyde in the presence of the catalyst.

The different alkylacetylenes tried, both mono-alkyl and dialkyl derivatives, underwent hydration to ketones in the presence of solid phosphoric acid catalyst at 204° and atmospheric pressure. Butyne-1 was hydrated also at 150°. Condensation of some of the ketone to oils of relatively high boiling point and refractive index accompanied its formation. By this hydration reaction propyne yielded acetone, butyne-1 formed 2-butanone, pentyne-1 and pentyne-2 each gave 2-pentanone, hexyne-1 produced 2-hexanone and heptyne-1 yielded 4-heptanone. The latter result indicates isomerization possibly involving migration of the triple bond along the carbon chain of heptyne-1 before hydration occurred producing 4-heptanone.

Experimental Part

Sources of Acetylene Hydrocarbons.—Acetylene from a commercial cylinder was purified from acetone vapors by passing through three scrubbers containing 96% sulfuric acid and then through a soda lime tower before use in the hydration experiments. Propyne was prepared following Hennion¹⁰ and butyne-1 was produced according to the

(1) Presented before the Division of Organic Chemistry at the 98th meeting of the American Chemical Society, Boston, Mass., Sept. 12, 1939.

(2) Thomas, Campbell and Hennion, *THIS JOURNAL*, **60**, 718 (1938).

(3) I. G. Farbenindustrie A.-G., British Patent 460,862 (Feb. 5, 1937).

(4) Walter, U. S. Patent 2,098,842 (Nov. 9, 1937).

(5) Eberhardt, U. S. Patent 2,093,146 (Sept. 14, 1937).

(6) I. G. Farbenindustrie A.-G., French Patent 46,616 (July 11, 1936).

(7) Ipatieff, U. S. Patents 1,993,512–13 (Mar. 5, 1935); 2,018,065–6 (Oct. 22, 1935); 2,020,649 (Nov. 12, 1935); 2,057,433 (Oct. 13, 1936); 2,060,871 (Nov. 17, 1936).

(8) Ipatieff and Schaad, U. S. Patents 2,120,702 (June 14, 1938); 2,157,208 (May 9, 1939).

(9) Universal Oil Products Co., British Patents 437,188 (Oct. 14, 1935); 463,272 (Mar. 25, 1937); 463,864 (Apr. 7, 1937); 464,671–2 (Apr. 19, 1937); French Patent 797,584 (April 29, 1936).

(10) Hennion, Meeting of the Indiana Academy of Science, Manchester, Indiana, November 5, 1937.

TABLE I
HYDRATION OF ALKYLACETYLENES IN THE PRESENCE OF SOLID PHOSPHORIC ACID CATALYST AT 204°

Hydrocarbon	B. p., °C.	Ketone	B. p., °C.	d_{4}^{20}	n_{D}^{20}	Semicarbazone, m. p., °C.	2,4-Dinitro- phenylhydrazone, m. p., °C.
Propyne	-23 to -22	Acetone	56- 60	0.789	...	183	...
Butyne-1	+8	2-Butanone	72- 82	.806	1.380	138-9	114-6
Pentyne-1	39-40	2-Pentanone	96-104	.818	1.392	103-4	142-4
Pentyne-2	55	2-Pentanone	99-107	.830	1.395	106-8	142-4
Hexyne-1	71	2-Hexanone	125-131	.814	1.401	116-7	107-8
Heptyne-1	98.5-99.5	4-Heptanone	150-153	.818	1.410	117-8	74-5

method published by Hurd and Meinert.¹¹ The pentyne-1, pentyne-2, hexyne-1, and heptyne-1 were purchased from the University of Notre Dame.

Apparatus and Procedure.—When hydrating a liquid alkylacetylene, water and the acetylene hydrocarbon were dropped simultaneously at constant rates from burets into the upper end of a vertical Pyrex tube (10 mm. inside diam. \times 90 cm.) containing the solid phosphoric acid catalyst (7.0 or 14.0 g. of 6-10 mesh particles) and surrounded by an insulated, electrically heated, aluminum bronze block furnace (60 cm. long) provided with a thermostat which maintained the temperature constant to within $\pm 2^\circ$. The exit end of the catalyst tube was attached by a ground glass joint to an ice-cooled Pyrex receiver connected through a calcium chloride drying tube to a trap cooled by solid carbon dioxide and acetone.

When the alkylacetylene used was gaseous, it was passed from a weighed aluminum bomb through a calibrated flowmeter and thence to the top of the catalyst tube, the rest of the apparatus and procedure being the same as described. In the experiments on acetylene purified by sulfuric acid the amount of this gas entering and leaving the catalyst chamber (of 100 g. catalyst capacity) was measured by means of wet-test meters through which acetylene had been passed previously to saturate the water contained in them.

The trap cooled by solid carbon dioxide remained empty during the experiments on liquid alkylacetylenes, it collected small amounts of unreacted hydrocarbon in the experiments with gaseous alkylacetylenes and collected the main part of the acetaldehyde as an aqueous solution in the runs on the hydration of acetylene. In experiments on the alkylacetylene, the reaction products and water were collected mainly in the ice-cooled receiver from which the aqueous layer was removed by a pipet, and the main ketone layer and that obtained by salting-out the aqueous layer with potassium carbonate were combined, dried over anhydrous sodium sulfate or potassium carbonate and later distilled. These ketone fractions were characterized by their physical properties and by preparation of semicarbazones and 2,4-dinitrophenylhydrazones.

Distillation of the aqueous aldehyde solution, produced from acetylene, separated acetaldehyde with 20-22° boiling point from the excess of water used in the experiment, the higher boiling oily condensation products having been separated previously from the heavier lower layer of aqueous acetaldehyde solution.

Reaction between Acetylene and Water.—Formation of acetaldehyde from mixtures of approximately equal molec-

ular proportions of acetylene and water began at about 260° under atmospheric pressure in the presence of solid phosphoric acid catalyst. At 300° hydration was more rapid in the equimolecular mixture and at the beginning of the experiment as much as 70% of the entering acetylene reacted. Of the acetylene consumed at the beginning of the reaction, about one-third yielded acetaldehyde and one-half produced higher boiling materials which apparently consisted of acetaldehyde condensation products containing crotonaldehyde and relatively high boiling aromatic hydrocarbons.

After the catalyst was in use for twenty-one hours and the absorption of acetylene had decreased to approximately 30% per pass, the catalyst was reactivated by heating in dry air at 500° for sixteen hours, followed by steaming at 260° for sixteen hours. Following this treatment the test was continued with 70% initial absorption of acetylene which dropped to 30% at the end of twenty-three hours and continued at that value for twenty-two hours more. The acetaldehyde yield reached 56% of the theoretical on the basis of the acetylene consumed with 9% accompanying condensation during this twenty-two hour period in which a reaction mixture consisting of two moles of water to one mole of acetylene was contacted with the reactivated catalyst. In all these acetylene hydration experiments the reaction mixture was in contact with the catalyst for an average time of 4.3 to 5.5 seconds.

Acetaldehyde from Acetylene-Ethylene and Acetylene-Nitrogen Mixtures.—Acetylene mixed with approximately an equal molecular proportion each of steam and ethylene, and with one proportion of steam and one-half proportion of nitrogen, also underwent hydration to acetaldehyde in the presence of solid phosphoric acid catalyst. Ethylene was not consumed, but apparently acted only as a diluent as was the case with nitrogen. These tests were made at an average contact time in the range of 5.0 to 5.7 seconds on both fresh, reactivated, and used catalysts. On the basis of the acetylene consumed, fresh catalyst gave the lowest yield of acetaldehyde and the highest production of oily condensation products; while used catalyst gave more acetaldehyde and less condensation products.

Hydration of Alkylacetylenes.—Propyne, butyne-1, pentyne-1 and -2, hexyne-1, and heptyne-1 underwent hydration in the presence of solid phosphoric acid catalyst and steam at 204° under atmospheric pressure and yielded acetone, 2-butanone, 2-pentanone, 2-hexanone, and 4-heptanone, respectively, as shown in Table I.

In addition to ketone formation by hydration of the different alkylacetylenes, there was also a production of small amounts of oils with relatively

(11) Hurd and Meinert, *This Journal*, **52**, 4544 (1930); **53**, 289 (1931).

high boiling points and refractive indices. These were probably ketone condensation products but no attempt was made to identify them because of the small quantities available.

There were also indications that isomerization of alkylacetylenes involving migration of the triple bond occurred in contact with the catalyst. Thus the liquid products recovered from an experiment on butyne-1 (b. p. $+8^{\circ}$) contained a small quantity of material which may have been butyne-2 as it boiled at $25-30^{\circ}$, was insoluble in water, gave no reaction with semicarbazide hydrochloride, and turned red on contact with sulfuric acid, as is characteristic of butyne-2.

The materials available were insufficient to show whether pentyne-1 and pentyne-2 isomerized into each other. It is probable that each of these

may hydrate to the same ketone without isomerization.

The fact that heptyne-1 hydrated to 4-heptanone is evidence that isomerization of this alkylacetylene occurred before hydration.

The authors are indebted to Mr. G. L. Hervert for assistance in conducting the hydration experiments.

Summary

Catalytic hydration of acetylene and of six alkylacetylenes has been accomplished readily by passing a mixture of the hydrocarbon and steam over a solid phosphoric acid-containing catalyst under atmospheric pressure at $260-300^{\circ}$ for acetylene and at $150-204^{\circ}$ for the alkylacetylenes.

CHICAGO, ILLINOIS

RECEIVED NOVEMBER 10, 1939

[CONTRIBUTION FROM THE LABORATORY OF THE NORTHERN COKE RESEARCH COMMITTEE, NEWCASTLE-UPON-TYNE, ENGLAND]

Crystallite Growth during Carbonization

BY H. E. BLAYDEN, H. L. RILEY AND A. TAYLOR

The following is a description and discussion of further results obtained in a detailed X-ray and chemical study of the changes which occur in the solid phase during the carbonization of various carbonaceous materials. We already have described¹ the results of our study of the carbonization in the temperature range $400-1300^{\circ}$ of cellulose and of a Northumberland coal. This work now has been extended in the case of cellulose to 2000° and also to the carbonization of two South Wales anthracites, an American anthracite, lignin and an Irish peat. Although the investigations are still incomplete and because of this their presentation must in some ways be tentative, the results already obtained appear to be of sufficient interest to justify publication at this stage. The demand for new knowledge of this kind need not be emphasized, for the fuller understanding of the molecular mechanism of the combustion and the reactivity of solid carbonaceous fuels demands more complete and fundamental information of their ultimate structure and of the effect of temperature, in the presence and absence of catalysts, upon this structure.

The Hull-Debye-Scherrer method of X-ray analysis has provided a means of obtaining this

more detailed information. From the broadening of the diffraction lines in the X-ray powder photographs of the so-called "amorphous" carbons compared with the lines in the X-ray spectrum of macrocrystalline graphite, it is possible to calculate the average dimensions of the crystallites present (assumed to be cylindrical). This method already has been employed for this purpose by Koch-Holm,² Lowry and Bozorth,³ U. Hofmann and co-workers,⁴ Berl,⁵ Corriez⁶ and others. The magnitude of the results reported is affected by the particular method of computation used and to some extent also by the X-ray technique employed. This should be borne in mind when comparing the results of different investigators. The method and technique employed by us, which have already been described in detail,¹ are believed to be in closest keeping with the most recent developments of X-ray crystallographic analysis. Although, in the present state of our knowledge, it is not possible to claim any high order of fundamental accuracy, it is probable

(2) Koch-Holm, *Wiss. Veroff. Siemens Konzern*, **6**, 188 (1927).

(3) Lowry and Bozorth, *J. Phys. Chem.*, **32**, 1524 (1928).

(4) Hofmann and Wilm, *Z. Elektrochem.*, **42**, 504 (1938).

(5) Berl, Adress, Reinhardt and Herbert, *Z. physik. Chem.*, **A158**, 273 (1932).

(6) Corriez, "Contribution à l'étude des variétés noires du carbone," *Les Presses Universitaires de France, Paris, 1937*.

(1) Blayden, Riley and Taylor, *J. Chem. Soc.*, 67 (1939).