compounds were 2-hydroxy-3-nitro-, 2-hydroxy-5-nitro, 2-hydroxy-3-nitro-6-methyl- and 2-hydroxy-5-nitro-6-methylpyridine.

When 2-hydroxy-3-nitropyridine was treated with molecular bromine and phosphorus tribromide according to the procedure of Berrie, Newbold and Spring,[3] we obtained, in addition to the expected 2-bromo-3-nitropyridine, a small quantity of 2,5-dibromo-3-nitropyridine.

The position of the substituents in the dibromo compound was determined by first treating the 2-hydroxy-3-nitropyridine with molecular bromine to obtain 2-hydroxy-3-nitro-5-bromopyridine which on treatment with phosphorus tribromide gave a dibromo compound identical with the original compound.

The structure of the 2-hydroxy-3-nitro-5-bromopyridine was proved by first brominating 2-aminopyridine using the method of Case,[4] nitrating the resulting 2-amino-5-bromopyridine, and finally diazotizing the resulting 2-amino-3-nitro-5-bromopyridine. Replacement of the hydroxyl group was performed using phosphorus tribromide and phosphorus oxybromide.

Our experiments with 2-hydroxy-3-nitropyridine were extended to other hydroxy compounds. The procedure for the proof of structure was the same as outline above except that when the nitro group was in the five position the original amino compound was first nitrated and then brominated. This sequence was necessary since otherwise bromine entered the five position.

For the bromination and nitration of 2-amino-6-methylpyridine we used the procedure of Adams and Schrecker.[5] For the bromination of 2-amino-5-nitro-6-methylpyridine we followed the procedure of Parker and Shive.[6]

Of the nine possible bromination products which can be obtained from 2-hydroxy-5-nitro-, 2-hydroxy-3-nitro-6-methyl- and 2-hydroxy-5-nitro-6-methylpyridine only two have been reported previously: 2-bromo-5-nitropyridine by Yamamoto,[7] and 2-hydroxy-3-bromo-5-nitro-6-methylpyridine by Parker and Shive.[6]

Acknowledgment.—We are indebted to the Nepera Chemical Co., Inc., for financial aid during this investigation.

Experimental

2-Bromo-3-nitropyridine.—A mixture of 5 g. of 2-hydroxy-3-nitropyridine, 4 ml. of phosphorus tribromide and 0.5 g. of phosphorus oxybromide, was heated on an oil-bath so that the temperature increased to 180° over the course of one hour. It was maintained at that temperature for 0.5 hour, cooled slightly, and the reaction quenched in ice and water. The precipitate was filtered and steam distilled. The distillate was filtered and air-dried to give 4.3 g. (54%) of product, m.p. 123–125°. Berrie, *et al.*,[3] reported 125°.

2-Bromo-5-nitropyridine: colorless needles, m.p. 137–138°, yield 45%. Yamamoto[7] reported 70% yield, m.p. 137° from phosphorus and bromine.

2-Bromo-3-nitro-6-methylpyridine: colorless needles, m.p. 71–72°, yield 24%. *Anal.* Calcd. for $C_6H_5O_2N_2Br_2$: C, 33.19; H, 2.30; Br, 36.85; N, 12.95. Found: C, 33.25; H, 2.39; Br, 36.89; N, 13.1.

2-Bromo-5-nitro-6-methylpyridine: colorless needles, m.p. 69–70°, yield 51%. *Anal.* Calcd. for $C_6H_5O_2N_2Br_2$: C, 33.19; H, 2.30; Br, 36.85; N, 12.95. Found: C, 33.13; H, 2.30; Br, 36.80; N, 13.1.

2,5-Dibromo-3-nitropyridine[8]: colorless needles, m.p. 94–95°, yield 51%. *Anal.* Calcd. for $C_5H_2O_2N_2Br_2$: C, 21.28; H, 0.71; Br, 57.01; N, 9.73. Found: C, 21.35; H, 0.74; Br, 56.93; N, 9.75.

2,3-Dibromo-5-nitropyridine: colorless needles, m.p. 75–76°, yield 90%. *Anal.* Calcd. for $C_5H_2O_2N_2Br_2$: C, 21.28; H, 0.71; Br, 57.01; N, 9.73. Found: C, 21.30; H, 0.76; Br, 57.03; N, 9.83.

2,5-Dibromo-3-nitro-6-methylpyridine: colorless needles, m.p. 87–88°, yield 90%. *Anal.* Calcd. for $C_6H_4O_2N_2Br_2$: C, 24.25; H, 1.35; Br, 54.49; N, 9.46. Found: C, 24.25 H, 1.39; Br, 54.53; N, 9.87.

2,3-Dibromo-5-nitro-6-methylpyridine: colorless needles, m.p. 111–112.5°, yield 66%. *Anal.* Calcd. for $C_6H_4O_2N_2Br_2$: C, 24.25; H, 1.35; Br, 54.49; N, 9.46. Found: C, 23.98; H, 1.39; Br, 54.76; N, 9.91.

2-Hydroxy-3-nitro-5-bromopyridine.—Bromine (1.8 ml.) was slowly added with rapid stirring to 5 g. of 2-hydroxy-3-nitropyridine. The reaction mixture at first became quite warm and when it started to cool it was heated on a steam-bath for two hours. It was then poured on a mixture of ice and water. The oil which separated crystallized overnight and the excess bromine was removed with bisulfite. The solid was filtered and recrystallized from 500 ml. of water using charcoal. The product was obtained in the form of yellow needles, m.p. 245–247°, yield 2.4 g. (33%). The product was also obtained by diazotization of 2-amino-3-nitro-5-bromopyridine using the precedure outlined by Berrie, *et al.*[3]

2-Hydroxy-3-bromo-5-nitropyridine: colorless needles, m.p. 221–223°, yield 85%; also obtained by diazotization of 2-amino-3-bromo-5-nitropyridine.[9] *Anal.* Calcd. for $C_5H_3O_3N_2Br$: C, 27.39; H, 1.37; Br, 36.58; N, 12.29. Found: C, 27.41; H, 1.35; Br, 36.63; N, 12.61.

2-Hydroxy-3-nitro-5-bromo-6-methylpyridine: recrystallized from ethanol as yellow needles, m.p. 221–223°, yield 35%. Diazotization in concentrated sulfuric acid gave only a small amount of product, but it was identical with the product prepared by direct bromination. *Anal.* Calcd. for $C_6H_5O_3N_2Br$: C, 30.91; H, 2.14; Br, 34.41; N, 11.99. Found: C, 30.51; H, 2.16; Br, 34.50; N, 12.58.

2-Hydroxy-3-bromo-5-nitro-6-methylpyridine: recrystallized from ethanol as colorless needles, m.p. 262–264°, yield 60%; also obtained by diazotization of the amino compound but in poor yield. Parker and Shive[6] reported 261°.

(3) A. H. Berrie, G. T. Newbold and F. S. Spring, *J. Chem. Soc.*, 2042 (1952).

(4) F. H. Case, THIS JOURNAL, **68**, 2574 (1946).

(5) R. Adams and A. W. Schrecker, *ibid.*, **71**, 1186 (1949).

(6) E. D. Parker and W. Shive, *ibid.*, **69**, 63 (1947).

(7) Y. Yamamoto, *J. Pharm. Soc. Japan*, **71**, 662 (1951).

(8) This compound is mentioned by Berrie, *et al.*, but it is not characterized, nor have we been able to find it elsewhere in the literature.

(9) This compound has not been reported previously. It was obtained by bromination of 2-hydroxy-5-nitropyridine in acetic acid and melted at 216–218°.

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Carbohydrates in Hazel (*Corylus sp.*). I. Major Sugar Component in Turkish Hazelnut Kernel

BY J. CERBULIS[1]

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Corylus Avellana is often abundant in Britain and Northern Europe, either as a bush or as a small tree, in woods and thickets. It is cultivated in many countries. Many hybrids of *C. Avellana* and *C. colurna* are cultivated in Turkey. The nut kernel is widely used for food and for the manufacture of nut chocolate and other types of confectionery.[2]

(1) 2848 N. Park Ave., Philadelphia 32, Pa.

(2) F. N. Howes, "Nuts," Faber and Faber Limited, London, 1948, p. 142.

Very little is known about the chemical composition of the nut kernel, especially about its carbohydrates. Ritthausen[3] has found that nut kernels contain a very easily crystallized sugar, which possibly could be sucrose, and some others, which could not be crystallized. Fang and Bullis[4] have found reducing sugars and sucrose. The data of carbohydrate content in nut kernel are as follows: carbohydrates 10.4–17.7%,[5,6] pentoses 2.62%,[7] reducing sugars 0.12–0.18%,[4] sucrose 4.79–5.57%[4] and starch 3.54–11.1%.[4,8]

The work reported below was undertaken to clarify the facts concerning the composition of the carbohydrate fraction of hazelnut kernels imported from Turkey. The examination of the 80% methanol-soluble constituents of the non-fat residue of hazelnut kernel led to the detection and isolation of numerous mono- and oligosaccharides. The analyzed nut kernels contained 66.5% fat and 4.5% sugars, most of which crystallized very rapidly. According to the paper and column chromatography findings, the sugars were divided in two groups: major and minor sugar components. The major sugar components were detected by paper chromatography utilizing the original sugar fraction, which then was separated by column chromatography, and the minor sugar components were detected. The major components were: D-fructose, D-glucose, sucrose, raffinose and stachyose. At first sugars were separated on a charcoal–Celite[9] column with aqueous ethanol in 10 fractions (Table I); the concentration of the ethanol being continuously increased from 1 to 30%.[10] Then each fraction was chromatographed once more or several times on a column and in cases, where it was necessary, the large scale paper chromatography was used. At least 14 minor sugar components were detected; most of them were ketose-containing.

TABLE I

SUGAR COMPOSITION AND YIELDS OF ELUATES FROM A CHARCOAL COLUMN

Fraction no.	Eluate, ml.	Dry sugar yield, g.	Sugars present
I	0–700	2.1	Sugar alcohols
II	700–1000	20.0	D-Fructose, D-glucose, sucrose, traces of several unidentified substances
III	1000–1500	21.6	Sucrose, traces of D-fructose, D-glucose, and several unidentified substances
IV	1500–2800	21.2	Sucrose
V	2800–3000	2.0	Sucrose, traces of melibiose and manninotriose, several unidentified substances
VI	3000–3700	2.4	Sucrose, traces of melibiose, manninotriose and unidentified substances
VII	3700–4500	7.2	Raffinose, stachyose, traces of unidentified substances
VIII	4500–5100	2.1	Raffinose, stachyose, traces of several unidentified substances
IX	5100–6000	0.8	Stachyose, traces of raffinose and unidentified substances
X	6000–7400	1.0	Unidentified; on paper chromatogram slower moving than stachyose
		80.4	

Experimental

Preparation of 80% Methanol-soluble Carbohydrate Extract.—Hazelnut kernels, imported from Turkey (1783 g.) were ground very fine in a mill. Hundred-gram portions of meal were extracted with petroleum ether by stirring in a glass beaker for 24 hr. Fat solution was decanted and filtered. Extraction was repeated seven times. After this preliminary extraction in a beaker, the meal was extracted with petroleum ether in a Soxhlet for two days. Then the petroleum ether was evaporated from the defatted residue; yield 598 g. of non-fat residue (33.5%) and 1185 g. of fats (66.5%).

Fifty-gram portions of the non-fat residue were extracted with four 500-ml. portions of 80% (v./v.) methanol by continuous stirring for 24 hr. Following the filtration, the extracts were evaporated to remove alcohol to a small volume. The portions were combined and diluted by a small amount of water. The neutral lead acetate solution was added in a small excess. After 2 hr., the solution was filtered and the precipitate was washed with water on the filter. Filtrate and washings were combined, treated with hydrogen sulfide, and filtered again; the excess of hydrogen sulfide was then boiled out. Then the solution was deionized with Amberlite IR-200 and IR-4B resins,[11] and evaporated to dryness; yield, approximately 90 g. of light-yellow sirup, which had the tendency to crystallize rapidly. This sirup was taken up with a small volume of water and analyzed by paper chromatography. Then the sugars were fractionated on a column.

Paper Chromatography of Sugar Fraction.—Paper chromatography was carried out on Whatman No. 1 filter paper by the ascending and descending methods at 20°. After development, the strips were air-dried, dipped in the reagent solution, and then gently blotted to remove excess reagent. Sugars were identified by authentic specimens.

A. **Solvents.**—(a) *n*-Propyl alcohol–ethyl acetate–water (7:1:2; v./v.)[12]; (b) *n*-butyl alcohol–pyridine–water (3:2:1.5; v./v.).[13]

B. **Color Reagents.**—Sugars were located by the *p*-anisidine phosphate in methanol,[14] orcinol,[15] triphenyltetrazolium chloride,[16] and ammoniacal silver nitrate.[17]

Before the separation on a column, the sugar fraction showed D-fructose, D-glucose, sucrose, raffinose and stachyose. These sugars were classified as "major components." All the sugars which were detected after the separation on the column were classified as "minor components."

Carbon Column Chromatography.—Four carbon columns were prepared. Each of them contained 220 g. of a mixture of equal parts, by weight, of Darco G-60 and Celite 535 held in a 4.5 × 50 cm. Pyrex pipe.[9] The carbon mixture of each column was washed with *ca.* 4 liters of water and then charged with 22.5 g. of the original sample of

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(4) S. C. Fang and D. E. Bullis, *J. Am. Oil. Chemists' Soc.*, **26**, 512 (1949).
(5) Research Department of H. J. Heinz Co., "Nutritional Charts," 8th edition, Pittsburgh, 1939.
(6) C. Chatfield and G. Adams, U. S. Dept. of Agriculture, Circ. No. 549, "Proximate Composition of American Food Materials," June, 1940.
(7) M. Rubner, *Arch. Anat. Physiol. Abt.*, 240 (1915); *Chem. Zentr.*, **87**, II, 939 (1916).
(8) Anon., *Bull. Imp. Inst.*, **14**, 261 (1916).
(9) R. L. Whistler and D. F. Durso, THIS JOURNAL, **72**, 677 (1950).
(10) R. S. Alm, R. J. P. Williams and A. Tiselius, *Acta Chem. Scand.*, **6**, 826 (1952).
(11) Rohm & Haas Co., Philadelphia, Pa.
(12) N. Albon and D. Gross, *Analyst*, **77**, 410 (1952).
(13) A. Jeanes, C. S. Wise and R. J. Dimler, *Anal. Chem.*, **23**, 415 (1951).
(14) J. Cerbulis, *ibid.*, **27**, 1400 (1955).
(15) R. Klevstrand and A. Nordal, *Acta Chem. Scand.*, **4**, 1320 (1950).
(16) O. Lüderitz and O. Westphal, *Z. Naturforsch.*, **7**, 548 (1952).
(17) K. T. Williams and A. Bevenue, *J. Assoc. Offic. Agr. Chemists* **36**, 969 (1953).

sugars dissolved in water. Then the columns were eluted with aqueous ethanol, the concentration of the ethanol being continuously increased from 1 to 30%.[10] The effluent from the columns was collected in 100-ml. portions. Each portion was analyzed by paper chromatography. Similar fractions were combined and concentrated to dryness. The results of the separation are summarized in Table I.

Identification of D-Fructose and D-Glucose.—The fraction no. II (Table I) was treated on a 4.5 × 50 cm. charcoal column using the original Whistler and Durso technique.[9] The sugars were separated in two fractions: monosaccharides and sucrose. D-Fructose and D-glucose were identified using authentic specimen by paper chromatography. Monosaccharide fraction was fermented with baker's yeast[18] for 2 days. D-Fructose and D-glucose were fermented completely. Only some sugar alcohols and pentoses were detected in the residue.

Identification of Sucrose.—Fraction no. IV (Table I) crystallized in large crystals. They were the characteristic sucrose crystals. Paper chromatography showed only sucrose. **A. Hydrolysis.**—Sucrose (*ca.* 0.1 g.) was heated with sulfuric acid (4%; 2 ml.) in a test-tube in a boiling-water bath for 2 hr. After neutralization with barium carbonate and filtration, the filtrate was evaporated to dryness and the sugars were estimated by paper chromatography; two equally large spots of D-fructose and D-glucose appeared. The hydrolysis by invertase[19] gave the same result. **B. Fermentation.**—A portion of sucrose was fermented with baker's yeast.[18] Chromatograms showed that sucrose was completely fermented, and only some unidentified substances appeared in traces. **C. Diazouracil Reaction (Raybin Test).**[20]—Twenty mg. of sucrose was dissolved in 2 ml. of 0.05 *N* NaOH and shaken with 5 mg. of diazouracil at 8°; it gave a deep yellow-green solution. After adding some drops of 0.1 *M* $MgCl_2$, this gave a blue precipitate, which was identical with the authentic specimen of sucrose. **D. Properties.**—M.p. 164°; $[\alpha]^{20}D$ +66.5° (H_2O; *c* 4).

Isolation of Raffinose and Stachyose.—Fraction no. VII (Table I) was treated on a 4.5 × 50 cm. charcoal column using aqueous ethanol, the concentration of the ethanol being continuously increased from 5 to 20%.[10] The effluent was collected in 100-ml. portions. The results are summarized in Table II. Portions 32–67, containing only raffinose and stachyose, were combined and the sugars separated on a 2.5 × 30 cm. charcoal column using the original Whistler and Durso technique.[9]

TABLE II

SUGAR COMPOSITION OF ELUATES FROM A CHARCOAL COLUMN OF FRACTION NO. VII (TABLE I)

Eluate in 100-ml. portions	Sugars present
1–9	Nil
10–12	D-Fructose, D-glucose, sucrose, several unidentified substances
13–19	Sucrose, melibiose
20–24	Manninotriose, several unidentified substances
25–31	Manninotriose, raffinose
32–67	Raffinose, stachyose

Identification of Raffinose. A. Hydrolysis.—Raffinose (*ca.* 10 mg.) was hydrolyzed in the case of sucrose; a paper chromatogram showed three equally large spots of D-fructose, D-glucose and D-galactose. The hydrolysis by invertase[19] gave D-fructose and melibiose. **B. Fermentation.**—The portion of raffinose was fermented with bakers' yeast.[18] After fermentation, the chromatogram showed melibiose. **C. Diazouracil Reaction.**[20]—This reaction gave a definite bluish precipitate, which was identical with the authentic specimen. **D. Properties.**—After several reprecipitations from 90% ethanol, the m.p. was 79°; $[\alpha]^{20}D$ +105.2° (H_2O; *c* 2).

Identification of Stachyose. A. Hydrolysis.—Stachyose was hydrolyzed with sulfuric acid as in the case of sucrose. Paper chromatograms showed the presence of D-fructose, D-glucose and D-galactose. The spot of D-galactose was approximately twice as large as the spots of D-glucose or D-fructose. Hydrolysis by invertase[19] gave D-fructose and manninotriose. **B. Fermentation.**—The portion of stachyose was fermented with baker's yeast.[18] After fermentation, the chromatogram showed manninotriose. **C. Diazouracil Reaction.**[20]—A definite bluish precipitate was identical with the authentic specimen from stachyose. **D. Properties.**—The m.p. was 167°; $[\alpha]^{20}D$ +148° (H_2O; *c* 2).

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(18) Fleischmann's yeast (Standard Brands, Inc.) was purchased from a local distributor.

(19) Difco Laboratories, Inc., Detroit, Michigan.

(20) D. French, G. U. Wild, B. Young and W. S. James, THIS JOURNAL, **75**, 709 (1953).

Synthesis of β-Cyclocitral

BY J. D. CHANLEY, E. CHOW AND H. SOBOTKA

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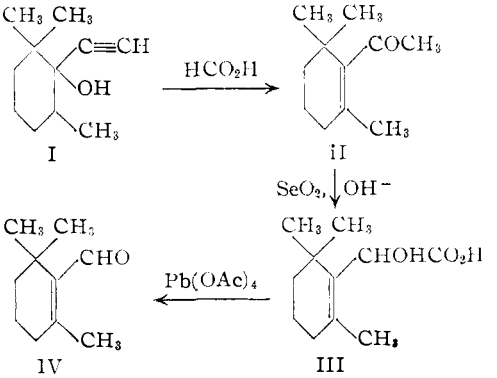
We have investigated the action of formic acid on various cyclohexanol acetylenes and have reported[1] the cases of 1-ethynylcyclohexanol and its 2,2-dimethyl homolog. At that time, we had also prepared 1-acetyl-2,2,6-trimethylcyclohexene (II) from 1-ethynyl-2,2,6-trimethylcyclohexanol (I), but were puzzled by its low molecular extinction coefficient ϵ_{max} 1300 at λ 242 mμ, which suggested that the reaction product did not consist exclusively of an α,β unsaturated ketone; in fact, it did not yield any keto derivatives. Recently, Henbest and Woods[2] have independently prepared this compound by the same method and also by hydration of 1-ethynyl-2,2,6-trimethylcyclohexene, employing a mercuric oxide–boron trifluoride catalyst. They also observed a low extinction coefficient at 243 mμ and likewise failed to obtain keto derivatives. Nevertheless, they deduced that the compound is an α,β-unsaturated ketone.

We have now obtained conclusive proof for the postulated structure of this compound. We converted it by the following sequence of reactions into β-cyclocitral: Oxidation of the compound with selenium dioxide yielded trimethylcyclohexenyl glyoxal, which on treatment with sodium hydroxide rearranged to the glycolic acid of m.p. 173° (III). This was oxidized with $Pb(OAc)_4$ in acetic acid to β-cyclocitral (IV). The latter compound and its semicarbazone were found to be identical with authentic samples.



1-Acetyl-2,2,6-trimethylcyclohexene (II).—One hundred grams of 1-ethynyl-2,2,6-trimethylcyclohexanol[3] was refluxed with 530 ml. of 90% formic acid for one hour. After

(1) J. D. Chanley, THIS JOURNAL, **70**, 244 (1948).

(2) H. B. Henbest and G. Woods, *J. Chem. Soc.*, 1150 (1952).

(3) H. Sobotka and J. D. Chanley, THIS JOURNAL, **71**, 4136 (1949).