proximately four times greater than that of the enzyme before dialysis.

Attempts to further purify the enzyme by fractional precipitation from water solution with ethanol were unsuccessful. The fractions were far less active than the starting enzyme, indicating that the enzyme was partially inactivated by the ethanol treatment.

Optimum *p*H.—Based on measurement of reducing power the optimum *p*H of the dialyzed and lyophilized enzyme at 40° on a 0.5% solution of guaran was 5.0 for a citric acid–disodium phosphate buffer and was *p*H 4.4 for a sodium acetate–acetic acid buffer.

Optimum Temperature.—For these measurements 1 mg. of enzyme was added for each 10 ml. of a 0.5% solution of guaran which was buffered at *p*H 5.0 with citric acid–disodium phosphate. The optimum temperature as indicated by measurement of reducing power was 45° for periods of time up to one hour, 40° for periods of about one to two and one-half hours and 35° for longer periods. At 35° no appreciable inactivation of the enzyme occurred.

Kinetics and Substrate Concentration.—At 35°, guaran concentration of 0.5% and an enzyme concentration of 1 mg. per 10 ml. of solution, a constant number of reducing sugar equivalents are liberated per unit of time through the test period of four hours. Thus at this substrate concentration the reaction rate is of zero order. At guaran concentrations in the neighborhood of 0.1% or less the reaction rate is no longer of zero order.

Inhibitory Effect of Ammonium Sulfate.—To 100 ml. portions of guaran solutions containing 0.00, 0.01, 0.10% and 1.0% ammonium sulfate were added equal amounts (1 ml.) of dialyzed and lyophilized enzyme (1 mg. per ml.). By following the increase in reducing power it was observed that ammonium sulfate gave no inhibitory effect until a concentration of greater than 0.1% was reached.

Extent of Guaran Hydrolysis.—A 0.5% solution of guaran at 35° and *p*H 5 was hydrolyzed by enzyme during a period of thirty-six hours to a reducing sugar value of 80% of that for complete hydrolysis. This extent of hydrolysis was not increased by further addition of enzyme. Partial analysis of the hydrolysate showed that 65% of the guaran had been converted to monosaccharides while the remainder consisted principally of di- and tri-saccharides.

Discussion

Although the enzyme preparations from guar seeds increase the reducing value and decrease the viscosity of guaran solutions, it is observed that the maxima of these two effects do not coincide for all enzyme preparations. Thus, it is likely that two or more guaran-hydrolyzing enzymes are present. The occurrence of at least two enzymes is probable since the enzyme preparation brings about not only hydrolysis of mannosido bonds in the main chain but also the hydrolysis of the galactosido bonds.

Regardless of whether the enzyme preparation is a mixture it is valuable as a tool for hydrolyzing guaran to produce oligosaccharide fragments, the identification and characterization of which will lead to further information regarding the structure and properties of guaran. The enzyme may also be of value for the commercial modification of industrial galactomannans.

Summary

An enzyme preparation separated from germinated guar seeds has been found to cause a guaran solution to undergo both a rapid decrease in the viscosity and a rapid increase in reducing power. As characterized by its ability to increase the reducing power of a guaran solution the enzyme preparation shows greatest activity at *p*H 5 in a citric acid–disodium phosphate buffer and at *p*H 4.4 in an acetic acid–sodium acetate buffer. The effect of temperature on the enzyme action is also examined. It is likely that more than one enzyme is present. While malt diastase causes a decrease in viscosity of a guaran solution the results suggest that the active enzyme is neither alpha- nor beta-amylase but an enzyme impurity similar in properties to beta-amylase.

LAFAYETTE, INDIANA RECEIVED APRIL 29, 1950

[CONTRIBUTION FROM ROHM AND HAAS COMPANY]

Structures of the OXO Aldehydes from Styrene

BY LEWIS A. WETZEL, CHARLES H. MCKEEVER AND CHARLES L. LEVESQUE

In two recent publications, Adkins and Krsek[1] have reported the preparation of 2-phenylpropionaldehyde in 30% yield by the application of the OXO reaction to styrene. The aldehyde was identified by its semicarbazone. There was no evidence of the simultaneous formation of 3-phenylpropionaldehyde, which, from the results obtained with alkenes,[2] would be expected to be a major product. Adkins and Krsek noted this discrepancy and mentioned that the 3-phenylpropionaldehyde was probably formed, but not isolated. When this work was repeated in these Laboratories, again the semicarbazone of 2-phenylpropionaldehyde was the only one readily isolated. There was, however, evidence that another semicarbazone was present.

We have obtained additional evidence that the primary product of the OXO reaction on styrene is actually a mixture of the 2- and 3-phenylpropionaldehydes. The data indicate that at least 23% is the 2-isomer, at least 29% is the 3-isomer, while the remaining 47% cannot be assigned to one or the other at present. Following Adkins' procedure,[1] but limiting the time of distillation, styrene gives a 46% yield of phenylpropionaldehydes. Oxidation of this aldehyde mixture with gaseous oxygen gives 3-phenylpropionic acid and acetophenone in approximately equal amounts. The former must come from the oxidation product of 2-phenylpropionaldehyde, via the following steps

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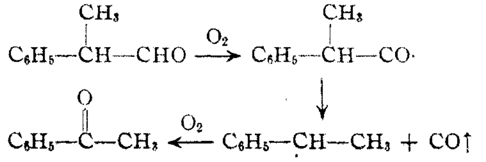
[1] Adkins and Krsek, (a) THIS JOURNAL, **70**, 383 (1948); (b) **71**, 3051 (1949).

[2] *E. g.*, Keulemans, Kwantes and van Bavel, *Rec. trav. chim.*, **67**, 298–308 (1948).



Carbon monoxide was detected in the effluent gases. In several trials, no 2-phenylpropionic acid could be isolated.

Loss of CO from an acyl radical is a common phenomenon.[3] Work in these Laboratories has indicated that branching at the alpha carbon atom facilitates this reaction, at the expense of the normal formation of the acid, in oxidation of aldehydes with gaseous oxygen.[4]

Experimental

OXO.—To a 500-cc. rocking bomb was charged 165 cc. of commercial grade styrene, 50 cc. benzene and 15–18 cc. of cobalt carbonyl solution (10% cobalt carbonyl content).[5] The bomb was pressurized to 2700 p. s. i. g. with a 50-50 carbon monoxide, hydrogen mixture and heated. At 110° gas absorption started. The pressure was maintained at 3000–3400 p. s. i. g. by recharging the bomb when necessary. The temperature was maintained at 115°. After 55 minutes, gas absorption became quite slow. The bomb was cooled to room temperature and the contents discharged. The crude aldehyde analyzed 9% $-\overset{\displaystyle O}{\overset{/\!\!/}{C}}-H$ or a conversion of 50%. The aldehyde was removed from the catalyst by rapid distillation at about 0.5 mm. from a Claisen flask. The distillate was then redistilled and the material boiling from 70–100° at 1.0 mm. taken as product. The yield was 46% of distilled material which on the basis of carbonyl content[6] contained 95% phenylpropionaldehydes.

Oxidation.—Two hundred and thirty grams of the aldehyde mixture was poured into a Pyrex column wrapped with heating wire and equipped with a water-jacketed side-arm for circulation and cooling, a sintered glass oxygen inlet at the bottom, and a thermometer and reflux condenser at the top. The flow of oxygen and the flow of cooling water were adjusted to bring the temperature up to 50° in one hour. The reaction was exothermic for about the first two hours, and then small amounts of heat were applied to maintain the temperature at 50°. The % COOH was checked every hour, and leveled off at about 14.5 after six hours. The calculated % COOH for phenylpropionic acid is 30.0. Carbon monoxide[7] was detected in the effluent gases.

The column was drained and the product distilled through an 8-inch helix-packed column, giving the following fractions

Fraction	G.	B. p., °C.	CHO, %	COOH %
Trap	9.0	(Acetophenone)		
I	37.9	55–64/2 mm.	16.1	..
II	17.6	64–76/2	12.1	..
III	3.0	76–103/1.5	..	23.3
IV	50.4	103–13/1.5	..	28.0
V	22.8	113–17/1.5	..	26.7
Residue	41.6	....	..	3.1
Total	182.3			

The carbonyl component was identified as acetophenone through the semicarbazone (m. p. 193–195.5°) and 2,4-dinitrophenylhydrazone (m. p. 243–246°). The melting point of a mixture of this latter derivative with an authentic sample of acetophenone 2,4-dinitrophenylhydrazone was 244–247°. The acid was identified as 3-phenylpropionic acid by neutral equivalent and m. p. (47–48.5°). From these data, it may be calculated that the product contained 44.4 g. of acetophenone and 72.5 g. of 3-phenylpropionic acid, equivalent to 49.5 g. and 64.8 g. of the 2- and 3-phenylpropionaldehydes, respectively.

Summary

Oxidation of the phenylpropionaldehyde produced by the OXO reaction with styrene gives a mixture of acetophenone and 3-phenylpropionic acid. This indicates that both 2- and 3-phenylpropionaldehydes are formed in the OXO reaction.

PHILADELPHIA, PA. RECEIVED MARCH 9, 1950

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(3) *E. g.*, Winstein and Seubold, THIS JOURNAL, **69**, 2916 (1947).

(4) McKeever, unpublished results.

(5) McKeever, U. S. Patent No. 2,477,553; 2,476,263.

(6) Ref. 1b, p. 3053.

(7) Carbon monoxide was detected by an immediate blue coloration when a tube packed with a molybdate compound was placed in the gas stream. The tube was obtained from the late Professor Adkins, and we have been unable to ascertain the details of its preparation.

[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY, CARNEGIE INSTITUTE OF TECHNOLOGY]

Reactions of 1,4-Diphenyl-1-bromo-3-butene-2-one. I. A Synthesis of 1-Aryl-2,5-diphenyl-3-pyrrolidones

BY PHILIP L. SOUTHWICK, DAVID I. SAPPER AND L. A. PURSGLOVE[1]

The preparation of 1,4-diphenyl-1-bromo-3-butene-2-one (I) by the action of N-bromosuccinimide on benzyl styryl ketone was described in a recent communication from this Laboratory.[2] Since this compound appears to offer unusual possibilities as a synthetic intermediate, an investigation of its reactions has been undertaken. The reactions to be described here, those with primary aromatic amines, have afforded a means of preparing an interesting new type of unsaturated arylamino ketone and have led to a new synthesis of 3-pyrrolidones. Only a few derivatives of 3-pyrrolidone have previously been described, and all of these were made by the use of the Dieckmann condensation or related methods.[3]

Contrary to the original expectation, 1,2,5-

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(1) Institute Graduate Fellow in Organic Chemistry, 1948–1949.

(2) Southwick, Pursglove and Numerof, THIS JOURNAL, **72**, 1604 (1950).

(3) (a) Prill and McElvain, *ibid.*, **55**, 1233 (1933); (b) De-Mouilpied, *J. Chem. Soc.*, **87**, 435 (1905); (c) Ruzicka and Seidel, *Helv. Chim. Acta*, **5**, 715 (1922); (d) Cook and Reed, *J. Chem. Soc.*, 399 (1945).