Anal. Calcd. for $C_{24}H_{18}O_3$: C, 81.32; H, 5.12. Found: C, 81.11; H, 5.15.

The *p*-nitrobenzoate separates from alcohol as rosets of colorless needles which melt at 109.5–109.8°.

Anal. Calcd. for $C_{24}H_{17}O_5N$: N, 3.51. Found: N, 3.49.

Conversion to 2-Phenylfuran.—The oxime of 2-phenyl-4-benzoylfuran was prepared in absolute alcohol solution in the presence of pyridine. It is readily soluble in the common organic solvents and separates from dilute alcohol as colorless needles which melt at 149.0–149.4°.

Anal. Calcd. for $C_{17}H_{13}O_2N$: C, 77.55; H, 4.98; N. 5.32. Found: C, 77.70; H. 5.13; N, 5.46, 5.40.

The Beckmann rearrangement was carried out by adding phosphorus pentachloride to a solution of the oxime in absolute ether. After crystallization from alcohol with the aid of Norite the anilide of 2-phenyl-4-furoic acid was obtained as colorless plates which melted at 192.0–193.0°.

Anal. Calcd. for $C_{17}H_{13}O_2N$: N, 5.32. Found: N, 5.44.

The anilide was hydrolyzed with alcoholic potassium hydroxide; it was necessary to heat the solution under reflux for twenty-four hours to cause complete reaction. The aniline was separated by steam distillation and identified as 1-phenylazo-2-naphthol, m. p. 128–129°. The residue was treated with Norite and acidified. Crystallization from alcohol produced colorless needles of 2-phenyl-4-furoic acid which melted at 208.0–209.0°.

Anal. Calcd. for $C_{11}H_8O_3$: C, 70.25; H, 4.25; neut. equiv.. 188. Found: C, 70.20: H, 4.49; neut. equiv.. 191, 190.

The acid is readily soluble in acetone, moderately soluble in alcohol, sparingly soluble in benzene, xylene, ether and chloroform. It forms a yellow solution in concentrated sulfuric acid.

The acid was not converted to the corresponding pyrrole when treated with ammonium hydroxide in a bomb at 180°. Decarboxylation did not occur when it was subjected to the following treatments:

1. A mixture of the acid and copper bronze was heated to 250°.
2. A mixture of the acid and copper bronze in quinoline was heated at 225° in a stream of nitrogen.
3. A mixture of the acid and copper bronze in benzyl-methylaniline was heated at 280° for one hour.
4. A mixture of the sodium salt of the acid and soda-lime was heated for two hours at 375° under a pressure of 10 mm.

The reaction was finally carried out by heating the acid overnight at 275° in a high-pressure bomb. The **2-phenylfuran** obtained in this way had the following properties: b. p. 107–108° (18 mm.); $n^{20}D$ 1.5968; d^{20}_4 1.083. These data are in agreement with those found for a sample prepared by the method described by Kondo and Suzuki.[6]

Summary

1,2-Dibenzoyl-1-propene is transformed into 2-phenyl-4-benzoylfuran by treatment with selenium dioxide. The furan is readily hydrolyzed by aqueous alkalies to give 1,2-dibenzoylethane and formic acid. Ammonia and aniline react to give the corresponding pyrroles. By a series of reactions the benzoylfuran was degraded to the known 2-phenylfuran.

(6) Kondo and Suzuki. *J. Pharm. Soc. Japan.* No. **544**, 501 (1927) ' *C. A.*, **21**, 3362 (1927).

NOTES

The Structure of Cholesteryl Chloride

By Ernst Bergmann

In work with cholesteryl chloride, as carried out in various laboratories and also in this Institute, certain conflicting evidence has accumulated with regard to its usual formula (I). The main point is the following one: while the two epimeric cholestyl chlorides react with sodium acetate under Walden inversion,[1] cholesteryl chloride has been found in our laboratory to give the same cholesteryl acetate which is obtained by direct acetylation of cholesterol.[2] Recently,

Marker and co-workers[3] have reported that 7-oxocholesteryl chloride, the constitution of which follows from the characteristic absorption band at 270 mμ of α,β-unsaturated ketones[4] behaves "normally" toward sodium acetate, giving an acetate under configurational inversion. Besides that, Marker and his co-workers have pointed out that cholesteryl chloride reacts under much less drastic conditions than the other chlorides mentioned.[5]

Reactions of halogenides with acetate ions are

(1) Marker and co-workers. THIS JOURNAL. **57**, 1755. 2358 (1935).
(2) That cholesterol and cholesteryl chloride have identical configuration. has been discussed in *Helv. Chim. Acta.* **20**. 590 (1937).

(3) Marker and co-workers. THIS JOURNAL. **59**. 619 (1937).
(4) Unpublished results from our laboratory (Miss F. Goldschmidt): compare for 7-oxocholesteryl acetate. Eckhardt. *Ber.*. **71**. 461 (1938).
(5) Compare Mauthner. *Chem. Central.*. **80**. II. 1537 (1909).

now generally accepted to occur with Walden inversion,[5,6] with one exception, if the halogenide belongs to the allyl derivatives. In this case the possibility of allylic rearrangement prevents any prediction of the steric course taken by a substitution reaction. Therefore, the above difficulty would be overcome if cholesteryl chloride were able to react in the form (II),[7] deriving from *allo*-cholesterol.[8] The reaction of form (II) with sodium acetate would be accompanied by a shift of the double bond.[9] In the case of 7-oxocholesteryl chloride the conjugation with the carbonyl group would stabilize the $C=C$ double bond in the "classical" 5,6-position.

It is interesting to note that similar conditions have been observed by Linstead and Rydon[10] in the case of buten-(1)-ol-(4), which is converted by a mixture of sulfuric and hydrobromic acids partly into 4-bromobutene-(1), and partly into 4-bromobutene-(2). On the basis of Juvala's[11] results, the high substitution velocity, too, of cholesteryl chloride would be understood more easily.
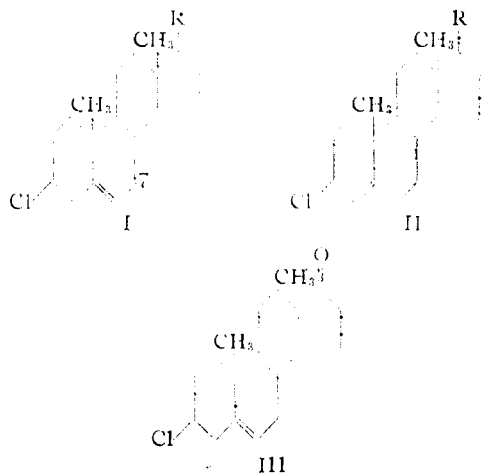
Analogous considerations apply to the case of chloroandrostenone (III), which in contrast with

(6) *Cf.* also Cowdrey, Hughes, Ingold, Masterman and Scott, *J. Chem. Soc.*, 1252 (1937). Fieser, in "The Chemistry of Natural Products Related to Phenanthrene," Reinhold Publishing Corp., New York, 1937, p. 392, has not yet taken these facts into account.

(7) The allylic nature of certain reactions of cholesteryl chloride has been discussed by Wagner-Jauregg and Werner [*Z. physiol. Chem.*, **213**, 119 (1932)] on the basis of the old allylic formula for cholesterol.

(8) The preparation of the chloride corresponding with *allo*-cholesterol is prevented by the easy dehydration of this substance: Schoenheimer and Evans, THIS JOURNAL, **58**, 182 (1936); *J. Biol. Chem.*, **114**, 567 (1936).

(9) The ability of cholesteryl chloride to react as (I) will account for the hydrogenative formation of a cholestane and not a coprostane derivative.

(10) Linstead and Rydon, *J. Chem. Soc.*, 1995 (1934).

(11) Juvala, *Ber.*, **63**, 1989 (1930).

the corresponding saturated chloroandrosterone is able to react with sodium benzoate without configurational inversion. This behavior may again be due to the possibility of III reacting in an allylic form.

Finally, it may be mentioned that this possibility is not necessarily limited to (I) and (III), but may apply to corresponding derivatives of other strong acids as, *e. g.*, toluenesulfonic acid.[12]

(12) Compare Stoll, *Z. physiol. Chem.*, **246**, 6 (1937).

THE DANIEL SIEFF RESEARCH INSTITUTE
REHOVOTH, PALESTINE   RECEIVED MARCH 4, 1938

Racemization During Esterification by Diazomethane

BY ERNST BERGMANN AND YAÏR SPRINZAK

In the course of experiments on Walden inversion, we had to prepare optically active methyl bromosuccinate. While esterification with methyl alcohol in the presence of concentrated sulfuric acid[1] gave satisfactory results, a series of experiments carried out with diazomethane gave invariably a racemic ester.

(–)-Bromosuccinic acid (1.5 g., rotation in acetone: c 3.000; l 9.9 cm.; αD $-2.18°$, hence $[\alpha]$ D $-72.7°$), finely pulverized, was added at zero temperature to a solution of diazomethane, prepared from ethyl nitrosomethylcarbamate (5 g.) and 25% methyl-alcoholic potash solution (7 cc.) in ether (30 cc.). The acid disappeared quickly; after thirty minutes of standing, the yellow solution was evaporated and the remaining racemic methyl bromosuccinate distilled *in vacuo*; b. p. 87° (2.5 mm.); yield 1.4 g. (Calcd. for $C_6H_9O_4Br$: Br, 35.5. Found: Br, 35.4).

As check experiments, a sample of the same (–)-bromosuccinic acid was kept for thirty minutes with a mixture of ether and methyl alcohol (6:1) and the optically active methyl ester in contact with diazomethane solution, prepared as above, for two hours. In both cases, no changes in optical rotation were observed.

Therefore we are inclined to assume that the racemization is connected in some way with the esterification mechanism. An analogous observation has been made incidentally by Stoermer and Starck[2] in the case of the optically active acid

(1) Holmberg, *Ber.*, **59**, 125 (1926).

(2) Stoermer and Starck, *ibid.*, **70**, 479 (1937).
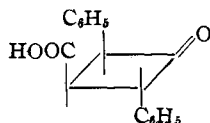
which is racemized too in the course of esterification by diazomethane.

Furthermore, the observation reported by Schlenk and Bergmann[3] may be recalled, that fluorene-9-carboxylic acid behaves—at least to a certain extent—abnormally toward diazomethane, giving dimethyl dibiphenylenesuccinate.

(3) Schlenk and Bergmann. *Ann.*, **463**. 194 (1928).

THE DANIEL SIEFF RESEARCH INSTITUTE
REHOVOTH. PALESTINE   RECEIVED MARCH 4. 1938

Remark on the Mechanism of the Fittig Reaction

BY O. BLUM-BERGMANN

(1) Bachmann and Clarke[1] have shown, some years ago, that the by-products formed in the biphenyl synthesis from boiling chlorobenzene and sodium metal indicate the intermediary formation of free phenyl radicals. Analogous results are obtained at room temperature, working in benzene solution.

To bromobenzene (50 g.), dissolved in benzene (50 g.) in a Schlenk tube,[2] sodium slices (10 g.) were added in nitrogen atmosphere, reaction starting quickly and causing a marked rise in temperature. When this first reaction ceased. the mass was shaken for two days, diluted with benzene (100 cc.) and separated by decantation from the excess sodium metal. Water was added, the benzene solution washed several times with water, dried and evaporated *in vacuo* and the residue fractionated at 13–15 mm. (a) B. p. 123–129°. This fraction crystallized completely; it consisted of pure biphenyl; m. p. 69–71°; yield 6.8 g. (b) B. p. 165–200°. The yellowish oil solidified almost quantitatively, on trituration with methyl alcohol; yield 2.1 g. From methyl alcohol, stout prisms, m. p. 56–57°, which according to their physical properties and the analysis proved to be *o*-phenyl-biphenyl, described before by Bachmann and Clarke.[1] (Calcd. for $C_{18}H_{14}$: C, 93.9: H, 6.1; mol. wt., 230. Found: C, 93.5; H, 6.0; mol. wt. (camphor), 241, 236.) (c) B. p. 200–250°, yellowish oil, which crystallized on trituration with light petroleum (b. p. 80–100°) (yield 0.26 g.) and was purified from the same solvent. Long needles of triphenylene; m. p. and mixed m. p. with an authentic sample. 195° (Calcd. for $C_{18}H_{12}$: C, 94.7: H, 5.3: mol. wt., 228. Found: C, 94.6: H, 5.3; mol. wt. (camphor), 257, 255).

(2) Theoretically free phenyl radicals may occur either in the formation from sodium and bromobenzene of

(1) Bachmann and Clarke. THIS JOURNAL. **49**. 2089 (1927). *Cf.* J. v. Braun and Kurtz. *Ber.*. **70**, 1224 (1937); Oldham and Ubbelohde. *J. Chem. Soc.*. 201 (1938).
(2) Compare Houben-Weyl. "Die Methoden der organischen Chemie." Vol. IV. Georg Thieme Leipzig. 1924. p. 959.

phenylsodium[3] or in the interaction between the latter compound and a second bromobenzene molecule or in both processes. Horn and Polanyi[4] showed that, on interaction with dilute sodium vapor, bromobenzene forms biphenyl, undoubtedly via phenyl radicals. On the surface of metallic sodium, on the other hand, it will be expected that a second sodium atom will always be near enough for phenylsodium formation when the first metal atom has removed the halogen from the C-Hal bond,[5] although, obviously, temporary radical formation cannot be excluded. Therefore it is more likely that the reaction of the alkylsodium compound with bromobenzene involves the intermediary production of the radicals. In favor of this, the observation may be reported that phenylsodium, on interaction with bromobenzene. gives triphenylene $(C_6H_4)_3$ too, indicating formation of phenyl radicals and their disproportionation into benzene and phenylene radicals C_6H_4.[6]

Diphenylmercury (6 g.) was shaken in benzene solution with sodium slices (10 g.) for two days;[7] the phenylsodium containing mass was then separated by decantation from the excess metal, and bromobenzene (7 cc.) (twice the theoretical amount) added. The mixture was kept at 70° for forty-eight hours and poured out into water. Treatment as above gave (besides some bromobenzene) (a) biphenyl. b. p. 120–160° (22 mm.), m. p. 65°, yield 1.45 g.: (b) triphenylene, b. p. 200–220° (13 mm.) after trituration of the crude product (1 g.) with light petroleum and recrystallization from the same solvent. m. p. 194–195.5°.

(3) Schlubach and Goes. *Ber.*. **55**. 2889 (1922).
(4) Horn and Polanyi. *Z. physik. Chem.*. **25B**, 151 (1934).
(5) Ziegler and Schaefer. *Ann.*. **479**. 150 (1930).
(6) In the famous experiments of Wieland. Popper and Seefried [*Ber.*. **55**. 1816 (1922)] on the dissociation of benzene-azotriphenylmethane. the occurrence of benzene instead of free phenyl may also be due to incidental formation of triphenylene which so far has not been isolated from the reaction mixture.
(7) Compare Schlenk and Holtz. *ibid.*. **50**, 268 (1917).

THE DANIEL SIEFF RESEARCH INSTITUTE
REHOVOTH, PALESTINE   RECEIVED MARCH 4, 1938

Hydrogenation of Ethylene and Partially Deuterized Ethylene on Catalytic Metal Surfaces

BY G. G. JORIS AND J. C. JUNGERS

The hydrogenation on catalytic copper of ethylene, ethylene-d_4 and partially deuterized ethylene (50%D) has been measured and found to be faster for the heavy compound.[1] To gain further information on this reaction and establish that this was not due to accidental circumstances, the hydrogenation of ethylene and partially deuterized ethylene was carried out on nickel, cobalt and platinum surfaces. The rates measured for mixtures of 5 cm. of ethylene and 7.5 cm. of hydrogen are given in the table and show

(1) G. G. Joris. H. S. Taylor and J. C. Jungers. THIS JOURNAL 1982 (1938).

quite definitely that in all cases studied the reaction is faster for the heavy compound.

TIMES OF ONE-HALF REACTION (IN MIN.)

Catalyst	Temp., °C.	$C_2H_2D_2$	C_2H_4
Nickel	+64	13	28
Platinum	−21	12	18
Cobalt	0	2.8	3.5

The columns give the nature of the catalyst, the temperature of the experiment, the time of half reaction for the heavy and light compound.

DEPARTMENT OF CHEMISTRY RECEIVED JULY 21. 1938
UNIVERSITY OF LOUVAIN
LOUVAIN, BELGIUM

Some Phenacyl and p-Substituted Phenacyl Esters

BY R. V. LUNDQUIST

Some phenacyl and p-substituted phenacyl esters of organic acids have been prepared for the identification of those acids. They were prepared by the method of Judefind and Reid.[1]

Heptylic Acid.—The phenacyl ester was a liquid.

Dichloroacetic Acid.—The phenacyl ester was a liquid; the p-bromophenacyl ester, observed m. p. 98.2 to 99.3°; the p-chlorophenacyl ester, observed m. p. 93.0 to 93.8°.

α-Bromo-n-butyric Acid.—The phenacyl ester was a liquid; the p-phenylphenacyl ester, observed m. p. 103.5 to 104.0°.

Acetylsalicylic Acid.—The phenacyl ester, observed m. p. 105.0 to 105.5°.

The esters were obtained in excellent yield and the solid esters were purified easily to a constant melting point.

(1) Judefind and Reid. THIS JOURNAL. **42**, 1043 (1920).

CONTRIBUTION FROM THE RECEIVED MAY 25. 1938
CHEMISTRY LABORATORY
UNIVERSITY OF IDAHO
MOSCOW, IDAHO

Acid Dissociation Constants in Dioxane–Water Mixtures. A Correction of the Dissociation Constant of Benzoic Acid

BY CECIL C. LYNCH AND VICTOR K. LA MER

Professor Martin Kilpatrick and Dr. L. John Minnick of the University of Pennsylvania kindly called our attention to the fact that the dissociation constants of benzoic acid as given in the "International Critical Tables," Vol. VI, p. 279. are in error by a factor of ten and that this had led to an unnecessary restriction of statement in our paper of the above title.[1] When the most recent value for 25°[2] is plotted in our Fig. 8 instead of the erroneous "I. C. T." value (6.15×10^{-4}), the anomalous behavior of benzoic acid as compared with acetic, propionic and butyric acids disappears. Log $K_{benzoic}$ becomes a linear function of the reciprocal of the dielectric constant between $D = 78$ and 21, and now conforms closely to the behavior of the three aliphatic acids. In Table III, p. 1257, K_{acetic} ($D = 34.3$) should read 5.62×10^{-7} instead of 5.01×10^{-7} and the second datum for propionic acid ($D = 21.0$) should read 5.33×10^{-9} instead of 5.30×10^{-9}.

(1) Lynch and La Mer. THIS JOURNAL. **60**, 1252 (1938).
(2) 6.31×10^{-5}, Brockman and Kilpatrick. *ibid.*, **56**, 1483 (1934).

DEPARTMENT OF CHEMISTRY
COLUMBIA UNIVERSITY
NEW YORK. N. Y. RECEIVED JUNE 23, 1938

The Hydrolysis of Methyl Iodide

BY RICHARD A. OGG, JR.

The hydrolysis of methyl iodide in pure water[1] has been shown to be kinetically first order with respect to methyl iodide, and unretarded in rate by the resulting hydriodic acid. Essentially similar results have been found by the author in a brief study of methyl iodide hydrolysis in a mixture of equal volumes of anhydrous methanol and water. The solutions (some 0.02–0.08 molar in methyl iodide) were heated in sealed nitrogen-filled tubes. Reaction was followed by titration of the resulting hydriodic acid. The last stages of the reaction were complicated by formation of considerable iodine (Moelwyn-Hughes found only traces)—undoubtedly as a result of the side reaction $CH_3I + H_3O^+ + I^- \longrightarrow CH_4 + I_2 + H_2O$. (The solutions containing most iodine gave off inflammable gas on opening the tubes.) Displacement of the equilibrium

$$CH_3OH + H_3O^+ + I^- \rightleftharpoons CH_3I + 2H_2O$$
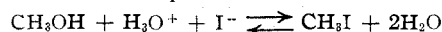
to the right by the large concentration of methanol in the solvent explains the importance of the side reaction—as contrasted to the results in pure aqueous solution.

Very good first order rate constants were obtained up to some 60% completion of the reaction (iodine formation then became troublesome). At 100 and 55° the rate constants were found to be, respectively, some 3×10^{-4} (average from

(1) E. A. Moelwyn-Hughes, *Proc. Roy. Soc.* (London). **A164**. 295 (1938).

three runs, with following respective sets of constants corresponding to successive time intervals— 3.1, 2.9, 3.0, 3.1, 3.2, 3.1, 3.0, 3.1; 2.9, 3.0, 2.8, 3.0) and 3.2×10^{-6} sec.$^{-1}$ (average from two runs, with following respective sets of constants—3.3, 3.4, 3.2, 3.1; 3.1, 3.0, 3.2, 3.1). The corresponding values found by Moelwyn-Hughes for the reaction in aqueous solution are 4.47×10^{-4} and 4.9×10^{-6} sec.$^{-1}$. The effect of replacing a portion of the solvent water by methanol is thus to diminish the rate of methyl iodide hydrolysis. Comparison of the figures at the respective temperatures indicates that the activation energy is but slightly affected, if at all. These results are entirely similar to the effects of solvent change on the rates of hydrolysis of secondary and tertiary alkyl halides in mixtures of ethanol and water.[2] It would appear that the hydrolyses by *neutral water* of methyl halides are kinetically similar to the corresponding reactions of secondary and tertiary halides, and that an identical intimate mechanism obtains for all of these reactions.

(2) See E. D. Hughes. *Trans. Faraday Soc.*, **34**, 185 (1938), for a review of this field.

CHEMICAL LABORATORY   RECEIVED JUNE 13, 1938
STANFORD UNIVERSITY
STANFORD UNIVERSITY. CALIF.

A New and Direct Method for the Determination of Creatine. I. Preliminary Report

BY CHARLES F. SCHAFFER

Investigations on the origin and significance of the excretion of creatine in the urine have received no general acceptance. The reason for this uncertainty is that creatine is not determined directly, but by difference. The preformed creatinine is determined by Folin's colorimetric method, and then the urine is heated with acid which converts the creatine to creatinine. The creatinine is then redetermined. The difference between this total creatinine value and that of the preformed creatinine is called creatine. The fallacies of such an analytical procedure are obvious; especially when considering that the quantitative determination of creatinine itself is made by measuring its reducing action. In fact it has been pointed out independently by Arnold,[1] Emden,[2] Hurtley,[3] Greenwald[4] and confirmed by

(1) Arnold. *Zentr. inn. Med.*, **21**, 417 (1900).
(2) Emden. *Zentr. Stoffenechael. Verdauungs-Krankheiten*, N. F., **2**, 250–289 (1907).
(3) Hurtley. *Lancet*, **184**, 1160 (1913).
(4) Greenwald. *J. Biol. Chem.*, **14**, 87 (1913).

Graham and Poulter,[5] that acetoacetic acid is capable of showing a false presence of creatine.

In attempting to formulate a copper reagent that would eliminate entirely the effect of reducing nitrogenous substances, but still retain its maximum ability to be reduced by sugars, Folin and Svedberg[6] developed one which is reduced by nitrogenous substances such as uric acid, creatine, creatinine and allantoin, but is not affected by sugar.

It was the use of this reagent which enabled Larson[7] to promulgate his colorimetric method for the determination of allantoin.

The subsequent use of this procedure for the determination of added allantoin in urine of man resulted in the recovery of excessive amounts. On investigation it was found that the substance responsible for this greatly increased reducing action was creatine.

Aware of the negligible amounts of allantoin present in human urine, it was decided that an earnest endeavor to utilize this procedure for the determination of creatine warranted investigation. It was soon discovered that by increasing the alkalinity of the copper solution, creatine demonstrated reducing properties which varied directly as its concentration and could be measured quantitatively.

Larson's procedure, slightly modified for its adaptation to creatine, is used. The determination is made by treating a portion of the filtrate of the test sample in a Folin–Wu[8] sugar tube with the ammoniacal copper solution; the alkalinity is increased by the addition of sodium hydroxide solution, and immediately it is placed in a rapidly boiling water-bath for ten minutes, cooled and the blue color, developed after the addition of acid molybdate, compared with a 1-mg. creatine standard similarly treated.

Reagents Required

Phospho-24-tungstic Acid.—It is essential that the phosphotungstic acid used be pure. Larson obtained inconsistent results in the precipitating power of eight different lots of C. P. phosphotungstic acid procured from four leading manufacturers and the results have been substantiated in this Laboratory. Basic lead acetate:[9] 5% (by weight) sulfuric acid solution: Folin ammoniacal cop-

(5) Graham and Poulter. *Proc. Roy. Soc.* (London), **B87**, 205 (1913).
(6) Folin and Svedberg. *J. Biol. Chem.*, **70**, 418 (1926).
(7) Larson. *ibid.*, **94**, 3 (1932).
(8) Wu. *ibid.*, **43**, 197 (1920).
(9) Hawk and Bergeim. "Physiological Chemistry." Blakiston. Philadelphia. Pa., 11th ed., 1937.

per reagent;[10] 20% sodium hydroxide solution; acid molybdate reagent;[11] creatine standard: dissolve 100 mg. of creatine in water and dilute to 100 cc.

Technique.—Place 1.5 g. of phosphotungstic acid in a large Pyrex tube, 30- to 50-cc. capacity, and dissolve in 6 cc. of water. With the aid of a pipet add 4 cc. of urine sample and centrifuge until clear. Add 4 cc. of the basic lead acetate solution and centrifuge again.[12] When clear, add 6 cc. of 5% sulfuric acid and recentrifuge. Transfer 2 cc. of the resultant, clear filtrate to a Folin–Wu sugar tube. Add 2 cc. of ammoniacal copper solution and 1 cc. of 20% sodium hydroxide. Place in a vigorously boiling water-bath for ten minutes, cool, add 2 cc. of acid molybdate reagent, dilute to mark and compare immediately with a 1-mg. creatine standard similarly treated.

Observations

The phosphotungstic acid precipitates the proteins and some of the other interfering substances while the addition of lead acetate removes the excess phosphotungstic acid and the remaining interfering substances. The subsequent use of sulfuric acid precipitates the excess lead completely.

The optimum alkalinity of the medium required for the creatine to exert its most efficient reducing action necessarily detracts from the non-sugar reducing properties of the copper solution.

Normally, urine contains traces of glucose which are so minute that the error introduced by such amounts is negligible. However, when sugar is present in any appreciable amounts it must either be removed by the brief fermentation process described by Folin and Svedberg[6] and designed for use on filtrates, or the quantitative sugar determinations of Folin–Wu[11] run in conjunction with the determination of creatine. This latter method is preferred as the fermentation procedure must be regulated rather too exactly, otherwise the reducing powers are increased rather than decreased.

Recovery of Creatine Added to Urine.— Urine that had no reducing action on the copper solution or at best produced only a slight coloration which was impossible to read colorimetrically was selected and varying amounts of a standard creatine solution added.

Filtrates of these various specimens were then prepared as described above, and the determination proper performed on the filtrates.

(10) Folin, *J. Biol. Chem.*, **82**, 88 (1929).

(11) Folin and Wu, *ibid.*, **41**, 367 (1920).

(12) Prior to the addition of the basic lead acetate, Larson recommends refrigerator incubation of the tube for one-half hour to minimize the time required for complete phosphotungstate precipitation. This procedure may be omitted as the results obtained, on final analysis of the filtrate, show very little variation.

From a series of twenty-eight different determinations recoveries ranging from 90 to 110% were obtained in contrast to recoveries of 82 to 92% employing the method of Folin.

The accompanying protocol shows a comparison of the values for creatine recovery, obtained by both the Folin and the suggested colorimetric method, when added to urine.

PROTOCOL
COMPARISON OF CREATINE RECOVERIES AFTER THE ADDITION OF A DEFINITE AMOUNT TO URINE

Urine no.	Creatine present, mg. per cc.	Recovery Folin method, mg. per cc.	Percentage error	Recovery Colorimetric method, mg. per cc.	Percentage error
1	2.6	2.5	− 3.9	2.6	0
2	3.6	3.2	−11.2	3.4	−5
3	4.6	3.8	−17.4	4.45	−3
4	1.0	0.92	− 8.0	0.97	−3
5	2.0	1.83	− 8.4	2.1	+5
6	3.0	2.57	−14.3	3.12	+4
7	1.0	0.88	−12.0	0.97	−3
8	2.0	1.74	−13.1	1.92	−4.2
9	3.0	2.38	−20.7	3.13	+4.5
10	0.6	0.49	−17.7	0.6	0

Conclusion

A preliminary report on a direct method for the determination of creatine in urine is presented. The full report is to be submitted on completion of the work.

BIOCHEMICAL AND RESEARCH LABORATORIES
THE NATIONAL DRUG COMPANY
PHILADELPHIA, PENNA. RECEIVED MAY 13, 1938

The Common Basis of Intramolecular Rearrangements. IV.[1] A Correction: The Benzilic Acid Rearrangement

BY FRANK C. WHITMORE

Since the formulation of the working hypothesis used in this Laboratory, no facts have been found contrary to the assumption of the basis of molecular rearrangement as due to a carbon with an open sextet of electrons. However, the extreme extension of the hypothesis to include the benzilic acid rearrangement involving the addition of a proton to benzil to form a positive ion, cannot be supported. Ingold[2] has proposed the existence of an intermediate negative ion produced by the addition of hydroxyl ion to benzil and indi-

(1) Whitmore, THIS JOURNAL, **54**, 3274 (1932); Whitmore and Stahly, *ibid.*, **55**, 4153 (1933); Whitmore and Fleming, *J. Chem. Soc.*, 1269 (1934); Wallis and Whitmore, THIS JOURNAL, **56**, 1427 (1934).

(2) Ingold, "Ann. Repts. Chem. Soc. (London)," Vol. XXV, 1928, pp 124–134.

cated the importance of an alkaline medium in the transformation.[3] The reaction rate study conducted by Westheimer[4] indicated the existence of a bimolecular reaction between benzil and hydroxyl ion which can best be explained by the formation of the same negative ion.   Recent investigation by

(3) Ingold. "Ann. Repts. Chem. Soc.. (London)" Vol. XXX. 1933. p. 177.

(4) Westheimer. THIS JOURNAL. **58**, 2209 (1936).

Roberts and Urey[5] on the oxygen interchange of benzil with water of a higher concentration of H_2O^{18} indicates a more rapid exchange in alkaline than in neutral solution which is explained by the rapid, reversible addition of hydroxyl ion to form a negative ion with benzil, followed by rearrangement.

(5) Roberts and Urey. *ibid.*. **60**, 880 (1938).

STATE COLLEGE, PENNA.   RECEIVED JUNE 4, 1938

COMMUNICATIONS TO THE EDITOR

THE SALT EFFECT IN THE PARAMAGNETIC CONVERSION OF p-HYDROGEN

Sir:

In 1934 Sachsse [*Z. physik. Chem.*, **B34**, 429 (1934); *Z. Elektrochem.*, **40**, 531 (1934)] reported that the rate constants for the conversion of p-hydrogen by solutions of paramagnetic ions were independent of the concentrations of these ions if the change in solubility of hydrogen with changing ionic strength were taken into account.   To this is now added the fact that these rate constants are also independent of the concentration of added diamagnetic salts.   The measurements were made by shaking the solutions with p-H_2 at a shaking speed in the range at which the conversion rate was independent of the shaker speed.   The ratio of gas volume to solution volume was known and kept constant.   The apparent rate was independent of hydrogen pressure.

Some representative data are given in the following table for which the solution volume is always 100 cc., the gas pressure *ca.* 100 mm., the concentration of manganous chloride 0.02 M, at room temperature.

Expt.	Molality of diamagnetic salt	$k'_{0.02} \times 10^3$	l(Ostwald)	$k(l. m^{-1}. min.^{-1})$
1	0	3.03	0.0198	7.9
2	0.3 M CaCl$_2$	2.75	.0173	8.2
3	0	2.15	.0198	8.0
4	1 M NaNO$_3$	1.65	.0158	7.7
5	2 M NaNO$_4$	1.41	.0127	8.2
6	0.6 M BaCl$_2$	1.56	.0151	7.7

In experiments 1 and 2 the gas volume is 45.1 cc.; in expts. 3 to 6 the gas volume is 64 cc. k' is the observed first order rate constant calculated by $k'_{0.02} = \frac{1}{t} \log \frac{(p\text{-}H_2)_0}{(p\text{-}H_2)_t} - k'_w$ where t is the time in minutes, $(p\text{-}H_2)_0/(p\text{-}H_2)_t$ is the rate of the initial

p-H_2 concentration to that at the time t and k'_w is the correction for the conversion by pure water under the same conditions and calculated in the same way.   For expts. 1 and 2 $k'_w = 0.07 \times 10^{-3}$ and for expts. 3 to 6 $k'_w = 0.05 \times 10^{-3}$. k is the velocity constant reduced to unit Mn^{++} ion concentration and corrected for the gas not in solution by the relation $k = \frac{V_g \times 2.303}{V_s \times l \times M_{MnCl_2}} \times k'$ where V_g is the gas volume, V_s the solution volume, l the Ostwald solubility of H_2 (from Seidell's "Solubilities") and M_{MnCl_2} the molality of the manganous chloride.

Thus the rate of conversion of p-$H_2 \longrightarrow o$-H_2 by Mn^{++} can be expressed by

$$\frac{-d(p\text{-}H_2)}{dt} = k(Mn^{++})(p\text{-}H_2)$$

where (Mn^{++}) and (p-H_2) are the concentrations of Mn^{++} ion and p-H_2, respectively.   There is no need to introduce any activity factor of Brönsted $f_{Mn^{++}}f_{H_2}/f_{Mn^{++}.H_2}$, although the concentration of the diamagnetic salts was carried as high as 4 M NaNO$_3$ and 2.4 M CaCl$_2$ in 0.02 M manganous chloride.   If it is incorporated it must be a constant and this is not unreasonable since the two reactants completely retain their independent identities throughout the reaction.

The effectiveness of the inhomogeneous magnetic field of the ion is independent of the ionic environment of the paramagnetic ion.   This is in agreement with the observation of Sachsse that solutions of manganous sulfate and manganous chloride give the same rate constant.

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