

TABLE II
HALOQUINOLINE DERIVATIVES

Salt from 6-chloroquinoline and	Empirical formula	M.p., °C.	Analyses, % Ionic halogen	
			Calcd.	Found
Decyl iodide	C ₁₉ H ₂₇ ClIN	113	29.39	29.33
Glycerol- α,γ -dibromohydrin	C ₁₂ H ₁₂ Br ₂ CINO	234	20.95	21.22
β -Cyclohexylethyl bromide	C ₁₇ H ₂₁ BrClN	102	22.52	22.33
β -Phenylethyl bromide	C ₁₇ H ₁₈ BrClN	108-111	22.92	22.83
β -Phenylethyl iodide	C ₁₇ H ₁₈ ClIN	164	32.09	31.90
Phenacyl bromide	C ₁₇ H ₁₃ BrClNO	215	22.04	21.78
<i>p</i> - <i>t</i> -Butylphenacyl bromide	C ₂₁ H ₂₁ BrClNO	232	19.08	18.98
<i>p</i> -Chlorophenacyl bromide	C ₁₇ H ₁₂ BrCl ₂ NO	205	20.12	20.12
<i>p</i> -Bromophenacyl bromide	C ₁₇ H ₁₂ Br ₂ CINO	207	18.09	18.11
<i>m</i> -Nitrophenacyl bromide	C ₁₇ H ₁₂ BrClNO ₂	215	19.60	19.52
<i>p</i> -Methoxyphenacyl bromide	C ₁₈ H ₁₆ BrClNO	211	20.34	30.33
β -Naphthacyl bromide	C ₂₁ H ₁₆ BrClNO	236.5	19.37	19.43
5,6,7,8-Tetrahydro- β -naphthacyl bromide	C ₂₁ H ₁₉ BrClNO	252	19.17	18.95
3-Bromoquinoline and				
<i>p</i> -Fluorophenacyl bromide	C ₁₇ H ₁₂ Br ₂ FNO	258	18.80	18.84

samples of 3-fluoro, 3-chloro and 3-iodopyridine, and to Miss Emogene Stephen, Miss Carolyn Cate, Mr. Tom Fuller and Mr. Lynn Easley for assistance in the purification of the products.

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Color Reactions of Human Antibody and Normal Human Gamma Globulin¹

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As a criterion of purity of the blood group A substances^{2,3,4,5} determinations were carried out of the proportions of two characteristic constituents of these antigens, hexosamine and methylpentose, specifically precipitated by an excess of antibodies to A substance. Since such specific precipitates consist of both antigen and antibody, total color values for hexosamine⁶ and methylpentose⁷ in specific precipitates must be corrected for any color given in these reactions by the antibody. The equivalent color values of normal human γ -globulin were used for this purpose although the possibility was recognized² and commented upon⁸ that human antibody and normal human gamma globulin may not give identical color values. This report shows that human antipneumococcal antibodies give color values identical with those for human gamma globulin in the reactions for hexosamine and methylpentose as well as with Folin-Ciocalteu tyrosine reagent.⁹

(1) The work reported in this paper was carried out under a research grant from the Division of Research Grants and Fellowships of the National Institutes of Health, United States Public Health Service and in part under the William J. Matheson Commission.

(2) A. Bendich, E. A. Kabat and A. E. Bezer, *J. Exp. Med.*, **83**, 485 (1946).

(3) E. A. Kabat, A. Bendich, A. E. Bezer and S. M. Beiser, *ibid.*, **85**, 685 (1947).

(4) E. A. Kabat, H. Baer and V. Knaub, *ibid.*, **89**, 1 (1949).

(5) E. A. Kabat, *Bact. Revs.*, **13**, 189 (1949).

(6) L. A. Elson and W. T. J. Morgan, *Biochem. J.*, **27**, 1824 (1933).

(7) Z. Dische and L. B. Shettles, *J. Biol. Chem.*, **175**, 595 (1948).

(8) G. Holzman and C. Niemann, *This Journal*, **72**, 2048 (1950).

(9) M. Heidelberger and C. F. C. MacPherson, *Science*, **97**, 405 (1943); **98**, 63 (1943).

Experimental

Antipneumococcal antibodies were produced by injection into a human being of a mixture of pneumococcal polysaccharides.¹⁰ A large sample of serum was obtained and found to contain 38 μ g. anti-C¹¹, 23 μ g. anti-SII¹¹ and 9 μ g. anti-SIII¹¹ N per ml. Specific precipitates of C-anti-C, SIII-anti-SIII and SII-anti-SII were obtained from about 100-ml. portions of serum, which had been in the refrigerator until the complement was destroyed, washed free from excess serum protein^{9,10,12} dissolved in water with 0.5 ml. of 0.1 M NaOH, and made up to a known volume. Four samples of normal human gamma globulin were available for comparison with the human antibodies.¹³

Hexosamine/Total N Ratio.—Aliquots of the dissolved SII-anti-SII and SIII-anti-SIII specific precipitates were analyzed for nitrogen by the Markham micro-Kjeldahl method^{14,12} and for hexosamine by a modification¹⁵ of the Elson-Morgan procedure.⁶ The hexosamine values were corrected for the color given in this reaction by the SII and SIII in the dissolved precipitates; these samples of polysaccharide gave color values equivalent to 3.3 and 0.9% hexosamine, respectively. Two lots of normal human gamma globulin were analyzed for nitrogen and hexosamine. C-anti-C precipitates were not suitable for determining the hexosamine/total N ratio since the C substance has a high (22%) hexosamine content.

Methylpentose/Total N Ratio.—SIII-anti-SIII and C-anti-C specific precipitates and three gamma globulin samples were analyzed for methylpentose and nitrogen,¹⁴ and the values for the specific precipitates corrected for the methylpentose color given by these polysaccharides; SIII and C gave color values equivalent to 1.4 and 0.8% methylpentose, respectively. SII-anti-SII specific precipitates were not used in determining the methylpentose/N ratios since the SII sample contained 40% of methylpentose.

Folin-Ciocalteu Color Equivalent.—SII-anti-SII and SIII-anti-SIII specific precipitates and two gamma globulin samples were used. SII and SIII contain no nitrogen and give no color with the Folin-Ciocalteu tyrosine reagent and no correction for their presence in the precipitates was necessary. Appropriate dilutions of known nitrogen content were analyzed as described by Heidelberger and MacPherson.^{9,12} Color development at 7500 Å. was proportional to N up to about 25 μ g. N.

The values of hexosamine/N, methylpentose/N and mean

(10) M. Heidelberger, C. M. MacLeod, S. J. Kaiser and B. Robinson, *J. Exp. Med.*, **83**, 303 (1946).

(11) C denotes the group specific polysaccharide of pneumococcus and SII and SIII the type-specific capsular polysaccharides of types II and III pneumococci.

(12) E. A. Kabat and M. M. Mayer, "Experimental Immunochimistry," C. C. Thomas, Springfield, Ill., 1948.

(13) E. A. Kabat and J. P. Murray, *J. Biol. Chem.*, **182**, 251 (1950).

(14) R. Markham, *Biochem. J.*, **36**, 790 (1942).

(15) K. Meyer, E. M. Smyth and J. W. Palmer, *J. Biol. Chem.*, **119**, 491 (1937).

values for Folin color per 10 μg . N are given in Table I. Within experimental error antipneumococcal antibodies and human gamma globulin give identical results.

TABLE I
COLOR EQUIVALENTS OF HUMAN ANTIBODY AND NORMAL HUMAN GAMMA GLOBULIN

	Hexos- amine ^{a,b/} Total N	Methyl- pentose ^{b/} Total N	Folin- Ciocalteu color D 7500 10 μg . N
Anti-C		0.032 ^e	
Anti-SII	0.07 ^c		0.155
Anti-SIII	.07 ^d	.030 ^f	.150
Gamma globulin			
Fraction II-1, 2 ¹³	.07		.149
Fraction II-3 ¹³	.07	.034	.146
Sample B ¹³		.032	
Sample S ¹³		.026	

^a Hexosamine was determined after hydrolysis with 2 *N* HCl at 100° for 2 hours. ^b The color values, while reproducible, are not assumed to be specific for either hexosamine or methylpentose.^{15,16} ^c Correction for hexosamine color value of SII-14%. ^d Correction for hexosamine color value of SIII-7%. ^e Correction for methylpentose color value of "C" substance-8%. ^f Correction for methylpentose color value of SIII-14%.

(15) E. Vasseur and J. Immers, *Arkiv Kemi*, **1**, 253 (1949).

(16) H. N. Horowitz, M. Ikawa and M. Fling, *Arch. Biochem.*, **25**, 226 (1950).

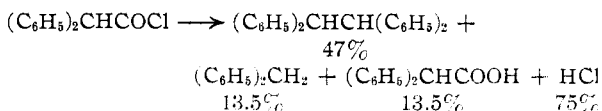
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The Attempted Rosenmund Reduction of Diphenylacetyl Chloride¹

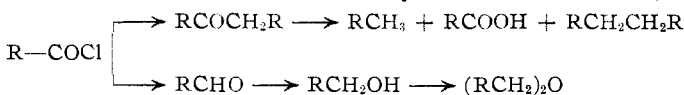
By JOHN G. BURR, JR.

Recently a supply of diphenylacetaldehyde was desired in this Laboratory, and since diphenylacetyl chloride was available, the Rosenmund reduction of this compound was investigated. The products which were obtained are



The occurrence of a product of the nature of tetraphenylethane as a product of a Rosenmund reduction has not been previously reported. Triphenylacetyl chloride also loses carbon monoxide and hydrogen chloride under Rosenmund conditions; the product in this case is triphenylmethane.²

The more usual products and by-products of this reaction were accounted for by Rosenmund³ as



This scheme does not predict products of the nature of tetraphenylethane or triphenylmethane.

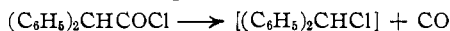
A possible explanation of these two products may be found in the temperature at which these

(1) This document is based upon work performed under Contract Number W-7405, eng. 26 for the Atomic Energy Commission at the Oak Ridge National Laboratory.

(2) S. Daniloff and E. Venus-Danilova, *Ber.*, **59**, 377 (1926).

(3) K. W. Rosenmund and F. Zetzche, *ibid.*, **54**, 642, 2038 (1921).

reactions were carried out. It is known, for example, that diphenylacetyl chloride decomposes at temperatures of around 200–250° into carbon monoxide, hydrogen chloride, tetraphenylethylene and diphenylketene.^{4,5} Staudinger⁴ explains the formation of tetraphenylethylene through an intermediate diphenylchloromethane which is known to decompose upon heating to tetraphenylethylene^{6,7} and tetraphenylethane.⁷ The moderate



temperature of the reaction observed here (boiling xylene solution) with its long duration might well be sufficient for a smooth production of diphenylchloromethane (but little diphenylketene), and the thermal conversion of this to tetraphenylethylene and tetraphenylethane. The tetraphenylethylene, in the reductive atmosphere, might be converted to tetraphenylethane.

Similarly, triphenylacetylchloride is known⁵ to be converted at 170–180° quantitatively to triphenylchloromethane and carbon monoxide. Under the conditions of the Rosenmund reduction, the triphenylchloromethane would probably be converted to triphenylmethane.

Experimental⁸

Attempted Rosenmund Reduction of Diphenylacetyl Chloride.—A suspension of 4 g. of 5% palladium-barium sulfate catalyst, poisoned with 0.6 ml. of quinoline-S solution, in 200 ml. of toluene was prepared, heated to boiling, and a small amount of solvent distilled to dry the remainder. To the cooled solution was added 44.6 g. of diphenylacetyl chloride. A moderately fast stream of hydrogen was passed through the stirred, refluxing mixture. The effluent gases were passed into water, and the absorbed hydrogen chloride titrated with 5 *N* sodium hydroxide. After overnight reaction, 28 ml. of NaOH had been consumed (75% of theory). The cooled solution was filtered from the catalyst, and the solvent was removed under vacuum. The residual pasty solid was heated with aqueous sodium bicarbonate. The undissolved material was filtered off, and the filtrate, after extraction with ether, acidified to give 5.5 g. (13.5%) of diphenylacetic acid (m.p. 145°). The neutral organic substances were stirred with ether. The ether-soluble material was obtained by filtration. It was evaporated, and the residue distilled at high vacuum. The distillate, 4.4 g. (13.5%), b.p. 87–89° (0.1 mm.), n_D^{20} 1.5788, was diphenylmethane. The residue was about 5 g. of a semisolid.

The ether-insoluble material weighed 15 g. (47% yield), and after several crystallizations from benzene formed a white microcrystalline powder melting at 211–212°. This material conforms in analysis, melting point, and general solubilities to tetraphenylethane. A mixture melting point with authentic tetraphenylethane (prepared from diphenylchloromethane and zinc, and melting at 211–212°) showed no depression.

(4) H. Staudinger, *ibid.*, **44**, 1619 (1911).

(5) A. Bistrzycki and A. Landtwing, *ibid.*, **41**, 686 (1908).

(6) R. Anschütz, *Ann.*, **235**, 220 (1886).

(7) C. Engler and H. Bethge, *Ber.*, **7**, 1128 (1886).

(8) All melting points were taken on a Fisher-Johns block and are uncorrected. Microanalyses are by Dr. H. W. Galbraith, Knoxville, Tenn.

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Phenylalanine Analogs

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The following new compounds related to phenyl-