14.76; N, 16.26; C, 27.86; H, 2.81. Found: Na, 13.20, 13.62; Cu, 14.35, 14.85; N, 15.47, 15.55; C, 27.14, 27.30; H, 3.73, 3.69.

The salt was dehydrated completely at 78° over phosphorus pentoxide. A sample of the salt weighing 25.270 mg. dried at room temperature over phosphorus pentoxide when completely dehydrated at 78° lost 0.378 mg. weight, or 1.50%. This dehydrated salt was analyzed for nitrogen.

Anal. Calcd. for $Na_3CuC_{10}H_{12}N_5O_6$: N, 16.26. Found: N, 15.81, 15.98.

The analytical data of the product dried at room temperature were corrected for the loss of weight on complete dehydration at 78°.

Anal. Calcd. for $Na_3CuC_{10}H_{12}N_5O_6$: Na, 16.02; Cu, 14.76; N, 16.26; C, 27.86; H, 2.81. Found (corr.): Na, 13.41, 13.83; Cu, 14.57, 15.07; N, 15.71, 15.79; C, 27.56, 27.72; H, 3.62, 3.58.

Sodium copper tetraglycylglycine is a hygroscopic pink powder. When heated in a capillary-size melting point tube it begins to decompose with a blackening of the sample at 268°, shrinks to a considerable extent at 272°, and evolves gas at 279–281° (all temp. corr.).

Summary

The sodium biuret salt of the pentapeptide tetraglycylglycine has been isolated and analyzed. The character of this salt is discussed in terms of the theory of the biuret reaction so far developed.

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[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY OF STANFORD UNIVERSITY]

The Composition of Grignard Reagents as Determined by Precipitation with Dioxane

BY C. R. NOLLER AND W. R. WHITE

Schlenk and Schlenk, Jr.,[1] who first investigated the action of dioxane on Grignard solutions explained their results by postulating that Grignard solutions are equilibrium mixtures represented by equation I or II. They favored equation I

$$2RMgX \rightleftarrows R_2Mg + MgX_2 \qquad (I)$$

$$R_2Mg_2X_2 \rightleftarrows R_2Mg + MgX_2 \qquad (II)$$

since eight-fold dilution of a solution of ethylmagnesium iodide did not affect the position of equilibrium as determined by precipitation of the halogen-containing compounds with dioxane. This result has been confirmed for Grignard solutions from normal, secondary and isobutyl bromides.[2] One would expect from this to be able to calculate equilibrium constants for Grignard solutions which would hold not only for variations in concentration but also for different ratios of halogen to alkyl radical. Miss Probert[2] determined the equilibrium compositions of solutions in which the ratio of halogen to alkyl radical varied widely for the normal, secondary and isobutyl compounds but no equilibrium constants could be obtained satisfying either I or II. In this work the equilibrium composition was determined by precipitation of the halogen compounds with dioxane and immediately removing the precipitate by filtration.

Although duplicate determinations by the same worker usually check fairly well, the values for dialkylmagnesium content obtained by different workers frequently vary considerably and it was thought that this and possibly also the inability to obtain satisfactory equilibrium constants might be due to occlusion of varying amounts of dialkylmagnesium under different conditions of manipulation and concentrations. It was thought also that occlusion might explain the results of Cope[3] who reported that the amount of diphenylmagnesium left in solution after precipitation of the halogen-containing compounds from a solution of phenylmagnesium bromide increased from 67 to 75% on standing in contact with the precipitate. Accordingly solutions of Grignard reagents from ethyl bromide containing varying concentrations and ratios of halogen to ethyl radical were precipitated with dioxane and shaken with the precipitate for eighteen to twenty hours before centrifuging and analyzing the supernatant liquid. The reproducibility of determinations under these conditions was increased but the data, given in Table I, again do not give satisfactory equilibrium constants on the basis of either I or II. It can be seen, however, that in general the more halogen present in the solution, the greater the amount of alkyl radical precipitated. This is illustrated better in Fig. 1 where the equivalents of ethyl radical precipitated are plotted against the equivalents of halogen in the orginal solution. While it is highly questionable whether one is justified in attempt-

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(1) Schlenk and Schlenk, Jr., *Ber.*, **62**, 920 (1929).

(2) Probert, Master's Thesis, Stanford University, 1933.

(3) Cope, THIS JOURNAL, **57**, 2238 (1935).

ing to draw a curve through these points, it appears that after shaking the precipitate has roughly a constant composition regardless of the composition of the original solution.

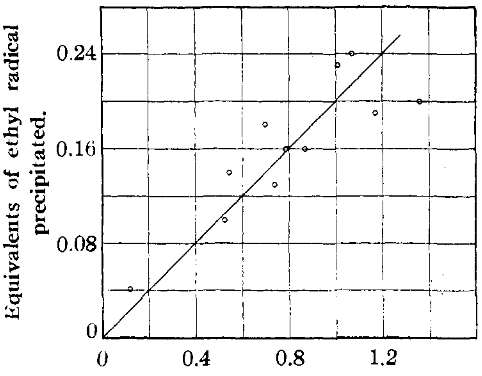

Fig. 1.

TABLE I

DIOXANE PRECIPITATION OF GRIGNARD SOLUTIONS CONTAINING VARIOUS RATIOS OF BROMIDE TO ETHYL RADICAL

Original solution		After dioxane precipitation	
Normality of ethyl radical	Normality of bromide	Equivalents of ethyl radical in solution	Equivalents of ethyl radical precipitated
0.803	0.120	0.761	0.042
.256	.523	.156	.100
1.102	.540	.962	.140
0.550	.690	.374	.176
.472	.732	.343	.129
.626	.781	.468	.158
2.010	.856	1.850	.160
1.457	1.001	1.225	.232
1.061	1.063	0.818	.243
1.083	1.163	.893	.190
1.200	1.354	.995	.205

After shaking an ordinary Grignard solution from ethyl bromide for twenty hours, about 76% of the ethyl radical was left in solution as diethylmagnesium, which is much higher than the values of 42–60% obtained when the precipitate is removed shortly after precipitation.[4] Moreover, the increase was greater than one would expect to be caused by simple occlusion and an investigation of the variation of the amount of dialkylmagnesium with time of shaking was undertaken. The results for three solutions containing different amounts of ethyl radical and bromide ion are shown graphically in Fig. 2. That this behavior is general is shown in Fig. 3 where the results for several ordinary Grignard reagents are plotted against time of shaking.[5] These curves show that it is impossible to obtain reliable data on the composition of Grignard solutions by means of dioxane precipitations because of the rapid change in composition immediately after precipitation. It is hoped that a careful study of solubility equilibria will give more information concerning this subject.

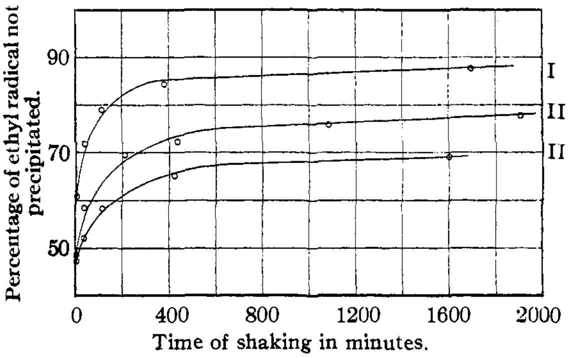

Fig. 2.—I, ethyl radical 1.10 *N*, bromide 0.54 N; II, ethyl radical 1.06 *N*, bromide 1.06 N; III, ethyl radical 0.55 *N*, bromide 0.69 N.

From a practical standpoint it should be emphasized that greatly improved yields of dialkylmagnesium solutions can be obtained from ordinary Grignard reagents by shaking for four to ten hours before removing the halogen compounds precipitated by dioxane.

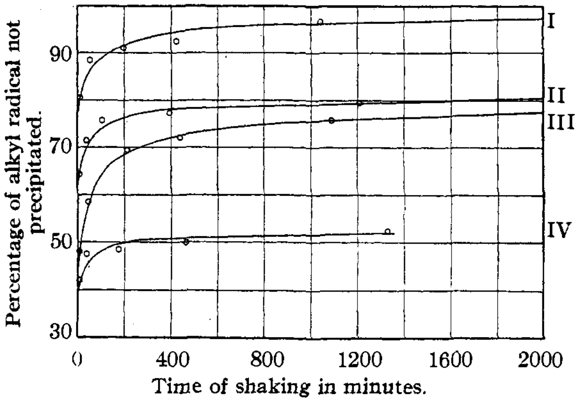

Fig. 3.—I, isobutyl 1.123 *N*, bromide 1.377 N; II, phenyl 1.077 *N*, bromide 1.159 N; III, ethyl 1.061 *N*, bromide 1.063 N; IV, *t*-butyl 0.548 *N*, bromide 1.677 N.

Experimental

The Grignard solutions were prepared in the usual manner in an atmosphere of purified nitrogen, allowed to settle for twenty-four hours and transferred to storage cylinders where further settling produced perfectly clear solutions. To vary

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(4) Schlenk, Jr., *Ber.*, **64**, 734 (1931); Johnson and Adkins, THIS JOURNAL, **54**, 1944 (1932); Noller and Hilmer, *ibid.*, **54**, 2503 (1932); Bartlett and Berry, *ibid.*, **56**, 2684 (1934).

(5) The Grignard solution from methyl bromide gave erratic results but there appeared to be a small decrease in the amount of dimethylmagnesium with time of shaking.

the ratio of bromide ion to ethyl radical, ether solutions of anhydrous magnesium bromide prepared by fusion of magnesium bromide with ammonium bromide, or of diethylmagnesium from diethylmercury and magnesium, were added in the desired amounts. The solutions were analyzed by titration with standard sulfuric acid followed by titration with standard silver nitrate.

The precipitation with dioxane was carried out in Babcock milk-testing bottles with a volume of about 40 cc. All glassware was dried at 150° and swept out with nitrogen before use. To 20 cc. of absolute ether and 2 cc. of purified dioxane[6] was added 10 cc. of the Grignard solution. The flasks were tightly stoppered with dried No. 00 rubber stoppers that fitted well down into the neck, and fastened to a shaking machine. After shaking for the desired length of time at room temperature, the flasks were centrifuged, the stoppers removed, and a 20-cc. aliquot taken for analysis. In all cases only traces of halogen were present in the solution. For all analyses duplicate determinations checked within 0.1 to 1.0%.

Summary

1. The rapid change in composition after precipitation of the halogen compounds in a Grignard reagent by dioxane makes the method useless for determining the composition of the original Grignard solution.

2. In most cases greatly improved yields of solutions of dialkylmagnesium can be obtained from ordinary Grignard reagents by shaking for four to ten hours before removing the precipitate formed on the addition of dioxane.

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[6] Dioxane is hygroscopic and it, as well as the ether, was kept over clean sodium.

[CONTRIBUTION FROM THE CHEMICAL LABORATORY OF STANFORD UNIVERSITY]

The Behavior of Inulin in Liquid Ammonia[1]

BY F. W. BERGSTROM AND A. E. GILMORE

Haworth[2a] and Irvine[2b] have shown by chemical methods that the molecular weight of inulin in water is about 5000, while Berner[3] has found values in water lying between 4600 and 5100 by cryoscopic means. In marked contrast are the results of a number of workers who have repeatedly obtained low results for the molecular weight of inulin in solvents such as liquid ammonia, acetamide, formamide and water. Thus, Schmid and co-workers[4,5] as a result of cryoscopic determinations in liquid ammonia conclude that inulin dissolves as a difructose anhydride, in agreement with the earlier work of Pringsheim, Reilly and Donovan[6] with solutions in acetamide. Pringsheim[6] and Reilly,[7] however, report that inulin has a molecular weight of around 1200 in water. Bergmann,[8] in an attempt to explain the low values given by these cryoscopic measurements, advanced the theory that inulin and certain of its derivatives were depolymerized when dissolved in liquid ammonia, formamide or acetamide. Berner,[3,9] on the other hand, has contended that the low results obtained by other investigators were due to the use of specimens of inulin which may have contained alcohol or other impurities.

The present investigation has as its purpose an examination of solutions of inulin in liquid ammonia to determine whether any reaction occurs between solute and solvent, and whether the physical properties of these solutions are best to be explained by assuming a solute of high or of low molecular weight.[10]

Behavior of Inulin in Liquid Ammonia.—If inulin is stirred into liquid ammonia contained in an open Dewar vessel, it dissolves readily, forming a water clear solution. If added faster than it will dissolve, it forms a clear very sticky gummy mass, which slowly passes into solution when broken up and agitated with the solvent. If this gummy mass is allowed to stand overnight in liquid ammonia, it changes to a non-adhesive translucent gel, which breaks up easily underneath the solvent, but does not again dissolve. If ammonia is allowed to evaporate rapidly from a freshly prepared solution of inulin, a thick sirup is formed, which hardens to a brittle crystal clear glass when all of the solvent has disappeared. Specimens of this glass

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[1] Original manuscript received April 27, 1936.

[2] (a) Haworth, *J. Chem. Soc.*, 2384 (1932); (b) Irvine, THIS JOURNAL, **55**, 1988–1994 (1933).

[3] Berner, *Ber.*, **66**, 397–400 (1933).

[4] Schmid and Becker, *ibid.*, **58**, 1968–1971 (1925).

[5] Schmid and Haschek, *Monatsh.*, **59**, 328–334 (1932).

[6] Pringsheim, Reilly and Donovan, *Ber.*, **62**, 2378–2381 (1929).

[7] Reilly, *Proc. Roy. Soc. Dublin*, **19**, 409–414 (1930).

[8] Bergmann, *Ann.*, **449**, 302–312 (1926).

[9] Berner, *Ber.*, **63**, 1356–1362 (1930).

[10] Taft, *J. Phys. Chem.*, **34**, 2792–2800 (1930), finds that inulin disperses well in liquid ammonia at its boiling point.