plastics were used as a matrix, (*b*) the amount of drug in matrix was varied, (*c*) drug solubility was changed, (*d*) additives were used, (*e*) different solvents were used.

Analysis of results shows that the above factors not only altered release rates directly as predicted by theory, but also indirectly by altering apparent porosities and tortuosities. Because of the latter effect, quantitative correlations were difficult to obtain. It is apparent that the porosity and tortuosity parameters must be accurately determined if the equation is to be applied quantitatively.

This aspect of the study will be discussed in another paper (5).

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# Gas Chromatographic Method of Moisture Determination

## By J. H. MARTIN and A. M. KNEVEL

The acid-catalyzed reaction between 2,2-dimethoxypropane (DMP) and water to form acetone and methanol was investigated to determine the applicability of the reaction in the quantitative determination of water by gas chromatography. An 8-ft. column packed with 30 per cent Theed (tetrahydroxyethyl ethylenediamine) was found to give a satisfactory separation of the DMP and the reaction products, acetone and methanol. An equation relating the heights of the peaks observed on the chromatogram to the moisture content of the sample was derived. The accuracy of the method was determined using samples of known water content, and a comparative study using the Karl Fischer titrimetric method was performed. The gas chromatographic method gave better accuracy and precision than the Karl Fischer method. The method was applied to two common solvents, and a general procedure for moisture determination was developed.

SINCE its introduction, gas chromatography has found applications in a wide variety of separations and analytical techniques. Until recently, however, gas chromatographic methods have found very limited use in the field of moisture determinations, and few were reported prior to 1959. The principal reason for this is that water exhibits a very long, flat, tailing peak upon elution from a gas chromatographic column. Most of the studies involving moisture prior to 1959 were simply attempts to eliminate the tailing effects of water so that other substances could be assayed without interference (1–4). The first attempt at using gas chromatography as a means of determining moisture quantitatively was reported by Smith (5) in 1959. In the same year, Elvidge and Proctor (6) reported the use of gas chromatography in the determination of water in some pharmaceutical formulations, while more recently Bennett (7) used a Teflon support to eliminate the tailing of water in a quantitative method.

The utilization of the reaction between 2,2-dimethoxypropane (DMP) and water was first reported by Erley (8) in 1957. He used the reaction as a convenient means of rendering samples intended for infrared analysis moisture free. The reaction was also utilized by Bousquet *et al.* (9) for the removal of water from biological samples and extracts. Critchfield and Bishop (10) reported the use of DMP as a reagent suitable for moisture determination. In their procedure the acetone formed in the reaction is determined by infrared analysis.

During the course of this study a paper was presented by Hager and Baker (11) in which the use of DMP in a gas chromatographic method of moisture determination was suggested. Their paper was concerned primarily with the feasibility of such a method. They presented no data and drew no conclusions.

## THEORETICAL CONSIDERATIONS

A derivation of the equation relating peak heights to molar concentration of water is presented.

In the following derivation, let *a* = number of moles of acetone in sample, *d* = number of moles

of DMP in sample, $A$ = height of acetone peak, and $D$ = height of DMP peak.

Assume a nonideal, but linear, response of the thermal conductivity detector over a limited range of molar concentrations of acetone and DMP. The peak height of each component then will be proportional to the molar concentration of substance present and may be expressed mathematically as

$$A = k'a \qquad \text{(Eq. 1)}$$

and

$$D = k''d \qquad \text{(Eq. 2)}$$

Dividing Eq. 1 by Eq. 2 gives

$$k(a/d) = A/D \qquad \text{(Eq. 3)}$$

Rearranging and solving for $k$ gives

$$k = (A/D) \times (d/a) \qquad \text{(Eq. 4)}$$

The constant $k$ constitutes a correction factor for column conditions as well as detector response.

When an excess of DMP is reacted with a sample of unknown water content, the resulting mixture will contain the excess DMP and an amount of acetone formed in the reaction. The number of moles of acetone present will be equal to the number of moles of water present originally. When a portion of this mixture is injected into a gas chromatographic column, an acetone peak and a DMP peak will be recorded, and the same relationship as stated in Eq. 3 will hold. That is,

$$k(a'/d') = (A'/D') \qquad \text{(Eq. 5)}$$

where the prime denotes values relating to the unknown sample, $a'$ is equal to the number of moles of acetone formed in the reaction or the number of moles of water reacted, and $d'$ is equal to the number of moles of DMP in excess. $A'$ and $D'$ can be measured directly in any units since the units will cancel. $a'$ is unknown, and $d'$ is equal to the number of moles of DMP added to the sample minus the number of moles which reacted with the water. Since the reaction between DMP and water is mole for mole, the number of moles of DMP which reacted is equal to the number of moles of water reacted which, in turn, is equal to the number of moles of acetone formed. Therefore

$$d' = d_0 - a'$$

where $d_0$ is equal to the number of moles of DMP originally present in the reaction vessel. Substituting the above expression in Eq. 5 gives

$$ka'/(d_0 - a') = A'/D'$$

Rearranging this gives

$$a'kD' = A' (d_0 - a')$$

$$a'kD' = A'd_0 - A'a'$$

and

$$a'kD' + A'a' = A'd_0$$

Solving for $a'$ gives

$$a' = A'd_0/(kD' + A') \qquad \text{(Eq. 6)}$$

In Eq. 6, $a'$ represents the number of moles of

acetone formed and is equal to the number of moles of water reacted. Therefore,

$$a' = W$$

where $W$ equals the number of moles of water present in the sample. Substituting this in Eq. 6 gives

$$W = A'd_0/(kD' + A') \qquad \text{(Eq. 7)}$$

where

$d_0$ = number of moles of DMP originally added to the sample
$A'$ = height of acetone peak produced by water in the sample
$D'$ = height of DMP peak
$k$ = $(A/D) \times (d/a)$ determined from a standard

The $k$ value above can be determined using a standard containing known amounts of DMP and acetone. There is no need to add an internal standard to the sample. In this way, the DMP acts not only as a reagent used to react with water, but also as an internal standard.

## EXPERIMENTAL

**Apparatus.**—The apparatus used in this study comprised an Aerograph gas chromatographic instrument (model A-90-S), a Minneapolis-Honeywell chart recorder [model Y143X(12)-V-II-III(30)], a microliter syringe, 10- and 20-ml. serum bottles with self-sealing stoppers, two 2-ml. tuberculin syringes, and a hot water bath.

**Reagents.**—2,2 - Dimethoxypropane, practical grade DMP, was obtained from Eastman Organic Chemicals and was triple distilled before use. Commercial grade acetone was obtained and also was triple distilled. Methanesulfonic acid was obtained from Eastman Organic Chemicals and was used without further treatment. Karl Fischer reagent was obtained as the single stabilized solution from Fisher Scientific Co.

**Selection of Column.**—A Theed (tetrahydroxyethylethylenediamine) column gave satisfactory separation of the DMP, acetone, and methanol. The column was constructed of an 8-ft. section of 0.25-in. O.D. copper tubing and was packed with 30% Theed on Chromosorb-W (Wilkens). A sample chromatogram is shown in Fig. 1.

**Operating Parameters.**—Oven temperature, 115–120°; injector temperature, 150°; collector temperature, 150°; filament current, 175 ma.; and flow rate of helium, 60–75 ml./min.

**General Procedure.**—All gas chromatographic sample preparations were made in 10- or 20-ml. serum bottles with self-sealing stoppers. The ethanol and dioxane samples were added to the serum bottles by means of a buret immediately
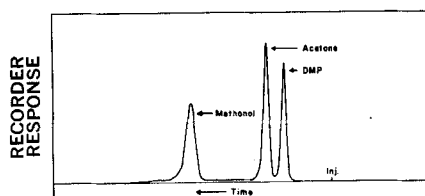


Fig. 1.—Separation obtained with 8-ft. Theed column.

prior to sealing. All other samples and reagents were added to the sealed bottles by means of a 2-ml. tuberculin syringe or a 50-$\mu$l. syringe. The ethanol and dioxane samples were determined on a volume basis, while all other samples were done on a weight basis.

After the sample had been weighed accurately, or measured in the case of ethanol and dioxane, in the tared serum bottle, a quantity of DMP was added to the bottle by means of a 2-ml. tuberculin syringe, and the weight of the DMP was accurately determined. Approximately 10 $\mu$l. of 0.1 $N$ methanesulfonic acid (MSA) in anhydrous methanol was added to this mixture. The bottle then was placed, after shaking, on a boiling water bath for about 1 min. This brief heating period was used to ensure completeness of reaction. The sample was then ready for injection into the gas chromatographic column.

Each injection of the sample mixture was preceded by an injection of a standard mixture containing accurately weighed quantities of approximately equal volumes of acetone and DMP. This standard solution was prepared in advance and was stored in a serum bottle with the usual self-sealing stopper.

The heights of the DMP and acetone peaks of the standard and the sample were read directly from the chart paper of the recorder, and the number of moles of water ($W$) in the sample was calculated using Eq. 7.

Three injections of each sample were made, and the average of the three injections then was taken as the water content of each sample.

A "dead-stop" method was used for all samples by the Karl Fischer titrimetric method. The titrations were performed using the back-titration method with a standard methanol–water solution. The system was arranged so that the titrant was automatically filled into and delivered from the buret in a closed system to minimize the entrance of atmospheric moisture. A magnetic stirrer was used during the titration.

**Statistical Consideration of the Gas Chromatographic Method.**—Using the general procedure for the gas chromatographic method described previously, twenty-five 0.5-ml. samples of distilled water were assayed and the mean, relative standard deviation and relative error were calculated (12). The samples were assumed to be 100% pure. The results of the statistical evaluation of the gas chromatographic method are summarized as follows: mean = 100.09%, relative standard deviation = 0.746%, and relative error = 0.09%.

**Comparison of the Gas Chromatographic Method with the Karl Fischer Titration.**—*Ethanol Samples.*—Anhydrous ethanol, prepared by distillation with benzene, was obtained and was assayed for water content by the Karl Fischer method and the gas chromatographic method. The ethanol was found to contain a negligible amount of water. A number of samples of known water content were carefully prepared, and those solutions then were assayed for water using the Karl Fischer method and the gas chromatographic method according to the general procedure described previously.

A similar study was carried out on samples of *p*-dioxane containing known amounts of water. The results obtained for these solvents and methods of moisture determination are shown in Table I.

### Stepwise Procedure to be Used in Gas Chromatographic Moisture Determinations

A step-by-step procedure for general moisture determinations is presented.

A.—Estimate the approximate moisture content of the sample.

B.—Weigh or measure into a tared serum bottle, a sample of such size as to contain not less than 10 mg. of water. Seal bottle.

C.—Calculate the approximate number of grams of water present in the sample bottle and multiply by 11.3. This yields the approximate number of ml. of DMP to be added to the sample.

D.—Add the above amount of DMP directly to the sample bottle by means of a syringe and needle, and accurately determine the weight of the DMP added.

E.—Add approximately 10 $\mu$l. of 0.1 $N$ methanesulfonic acid in methanol to the sample and place in a hot water bath for about 1 min.

F.—While the sample is in the hot water bath, the standard can be injected into the gas chromatographic column.

G.—Remove the sample from the hot water bath, dry the outside of the bottle, rinse the syringe 3 or 4 times with the sample, and inject.

H.—Inject the standard and sample 3 times each, preferably alternately, attempting to keep all peak heights roughly the same.

I.—Measure the peak heights and record $A$, $D$, $A'$, and $D'$ for each set of injections. A set consists of one injection each of the standard and the sample, and $A$ and $D$ denote peak heights of acetone and DMP, respectively, in the standard, and $A'$ and $D'$ denote the peak heights of acetone and DMP in the unknown sample.

J.—Calculate the value of $k$ using the formula

TABLE I.—RESULTS OF A COMPARATIVE STUDY OF MOISTURE DETERMINATION IN ETHANOL AND *p*-DIOXANE USING KARL FISCHER (KF) AND GAS CHROMATOGRAPHIC (GC) METHODS

| Sample | W/w Water in Sample, % | KF Found[a] | KF Relative Error, % | GC Found[a] | GC Relative Error, % |
|--------|------------------------|-------------|----------------------|-------------|----------------------|
| *Ethanol* | | | | | |
| 1 | 0.625 | 0.666 | 6.40 | 0.624 | 0.16 |
| 2 | 2.500 | 2.598 | 3.92 | 2.504 | 0.16 |
| 3 | 5.000 | 5.103 | 2.06 | 4.984 | 0.32 |
| *p-Dioxane* | | | | | |
| 1 | 0.664 | 0.689 | 0.765 | 0.666 | 0.301 |
| 2 | 2.539 | 2.521 | 0.708 | 2.357 | 0.078 |
| 3 | 5.039 | 5.040 | 0.019 | 5.031 | 0.158 |

[a] Average of three determinations.

$$k = (A/D)(d/a)$$

where $d$ is the number of moles of DMP in the standard, and $a$ is the number of moles of acetone in the standard; calculate the number of moles of water, $W$, in the sample using the formula

$$W = A'd_0/(kD' + A')$$

where $d_0$ is the number of moles of DMP added to the sample. If the per cent water of the sample is desired, the formula

$$\% \ W = \frac{(1802)A'd_0}{s(kD' + A')}$$

may be used, where $s$ is equal to the sample size.

A value for $W$ will be obtained for each set of injections, and the average of the three calculated $W$'s can be taken as the moisture content of the sample.

Although most of the steps in the above procedure are self-explanatory, step $C$ requires clarification. Step $C$ involves multiplication of the approximate number of grams of water in the sample by 11.3 to determine the number of ml. of DMP to be added to the sample. One gram of water will react with 6.3 ml. of DMP. Therefore, in order to get a DMP peak which is roughly the same size as the acetone peak which is produced, approximately twice as much DMP as that required to react with all of the water in the sample will be needed. However, since DMP evokes a greater response in the detector per mole than acetone, slightly less than 100% excess is required to produce acetone and DMP peaks of approximately the same size. In this investigation, multiplication of the estimated water content of the sample by 11.3 was found to give satisfactory results. This will, of course, vary slightly from instrument to instrument depending on column conditions and detector sensitivity. The amount of DMP to be added to the sample to give best relative heights of acetone and DMP peaks can also be determined graphically as shown in Fig. 2. The amount of DMP to be
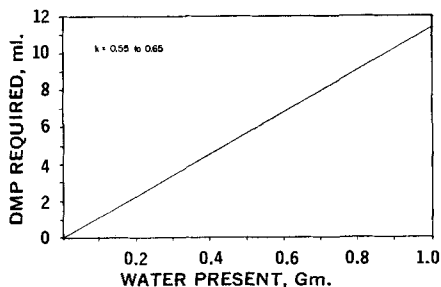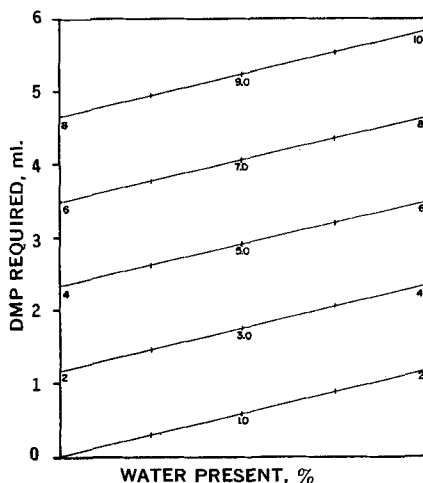


Fig. 3.—Amount of DMP to be added to a five-unit sample to yield the best relative peak heights of acetone and DMP.

added to 5-unit (Gm. or ml.) samples of different water content is presented in Fig. 3.

## DISCUSSION

A gas chromatographic method of moisture determination has been developed. In the samples tested, the method gave greater accuracy than the Karl Fischer method. Although the method was not applied to the determination of water of crystallization in this study, its utility for this purpose is apparent.

An inherent advantage in the use of the reaction between DMP and water is that the unreacted DMP acts as an internal standard. This eliminates the need for addition and weighing of a separate internal standard.

The method is relatively simple in operation and requires much less time than the manual Karl Fischer titrimetric method.

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Fig. 2.—Amount of DMP to be added to sample to yield best relative peak heights of acetone and DMP.