

# ANESTHESIA: XXVI. THE DETERMINATION OF ETHERS IN BLOOD WITH SPECIAL REFERENCE TO METHYL n-PROPYL ETHER\* †

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## INTRODUCTION

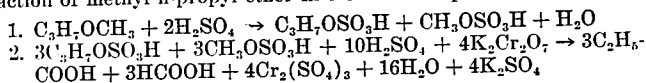
In studying the clinical usefulness of new inhalation anesthetic agents, it is desirable to correlate the degree of anesthesia with the concentration of the agent in the blood of the patient. When the compound being studied is volatile and capable of oxidation, the determination of blood concentrations has been carried out in the following manner.

1. Vaporize the compound.
2. Pass the vapors into a known quantity of suitable oxidizing reagent.
3. Determine how much of the oxidizing agent is consumed in the reaction.
4. From the stoichiometric relation in the reaction between anesthetic and oxidizing agent, calculate the amount of anesthetic.

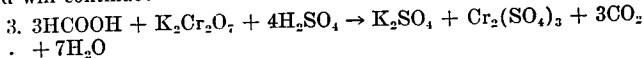
This general scheme was adapted to the determination of blood concentrations of a new anesthetic agent, methyl n-propyl ether (metopryl). The method used in this laboratory is based in part on the work of Andrews, Potter, Friedemann, and Livingstone (1).

## THEORETICAL

A potassium dichromate-sulfuric acid solution, 5 cc. 0.100 N  $K_2Cr_2O_7$  and 20 cc. of 75 per cent  $H_2SO_4$ , was used as the oxidizing agent. The reaction of methyl n-propyl ether in this medium proceeds as follows:



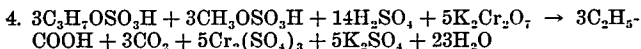
The propionic acid undergoes no further oxidation, but the formic acid will continue:



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Combining equations 2 and 3 we have:



Since the two alkyl sulfonic acids are derived from one original ether molecule, the total quantity of ether referred to by equation 4 is 3 moles. It will be seen that 5 moles of  $K_2Cr_2O_7$  are required to oxidize the sulfonic acids derived from this quantity of ether. This ratio of 3 moles of ether to 5 moles of  $K_2Cr_2O_7$  is the basis for the stoichiometric calculation of ether concentrations.

#### TITRATION

It was found that the titration of solutions containing such strong sulfuric acid was not reliable owing to the indefinite and variable end-point. Even test titrations of known quantities of  $K_2Cr_2O_7$  in the presence of 20 cc. of 75 per cent  $H_2SO_4$  gave erroneous results. In order to avoid this difficulty, an excess of solid  $BaCl_2 \cdot 2H_2O$  was added to aqueous sulfuric acid solutions containing a known amount of  $K_2Cr_2O_7$ . When the  $SO_4^{2-}$  ions had all been removed as solid  $BaSO_4$ , reproducible iodometric titrations with  $Na_2S_2O_3$  were made possible, and the results checked the known amount of  $K_2Cr_2O_7$  originally used. The end-point, in the presence of the precipitated  $BaSO_4$ , was the change from a light powder blue to chalky white. The equations for this reaction are as follows:

1.  $H_2SO_4 + BaCl_2 \rightarrow BaSO_4 + 2HCl$
2.  $K_2Cr_2O_7 + 6KI + 14HCl \rightarrow 8KCl + 2CrCl_3 + 7H_2O + 3I_2$
3.  $2Na_2S_2O_3 + I_2 \rightarrow 2NaI + Na_2S_4O_6$

#### CALCULATION

The titration with standardized  $Na_2S_2O_3$ , 0.01 N, shows the amount of 0.100 N  $K_2Cr_2O_7$  still present in the oxidizing solution, which is calculated as follows:

1. (ml. thiosulfate) (N thiosulfate) = (X)(0.1) where X = the number of ml. of 0.100 N  $K_2Cr_2O_7$  solution.  
(5 - X) therefore is the number of ml. of 0.100 N  $K_2Cr_2O_7$  solution used in the oxidation of the ether vapors.

The number of moles of  $K_2Cr_2O_7$  per ml. in a 0.100 N solution is  $1.67 \times 10^{-5}$ . Therefore, letting M represent number of moles of ether,

2.  $\frac{(5 - X)(1.67 \times 10^{-5})}{M} = \frac{5}{3}$  since the molar ratio between  $K_2Cr_2O_7$  and ether is 5 to 3.

Rearranging, and multiplying by the molecular weight of methyl *n*-propyl ether in milligrams, the final equation becomes:

$$3. \text{ Mg. ether} = (5 - X) \frac{(3)(1.67 \times 10^{-5})(7.408 \times 10^4)}{5}$$

$$4. \text{ Thus Mg. ether} = (5 - X)(0.742)$$

In the actual determinations on blood samples, a blank was always carried out on the patient's blood before anesthesia, and the amount of  $\text{K}_2\text{Cr}_2\text{O}_7$  solution unused by the blank was substituted for the 5 in equation 4. In this way a correction was introduced in case any volatile oxidizable substances were present in the blood of the un-anesthetized patient.

In adapting this method for the determination of other ethers, changes would have to be made in this calculation in order to allow for differences in molecular weight and in the degree of oxidation.

#### EXPERIMENTAL

*A. Apparatus:* The apparatus used for the determination is shown in figure 1. The  $\text{CaCl}_2$  tube and  $\text{H}_2\text{SO}_4$  scrubbing tower A were used to remove moisture and volatile oxidizable substances from the air, which was drawn through the apparatus by means of suction as indicated at the top of column E. The flask C held the sample through which the air current was passed, and the flask D contained the oxidizing medium. The tower E was filled with glass beads into which the oxidizing solution was drawn by the suction. The beads caused the breaking up of the air stream containing the ether vapor into small bubbles, thus insuring thorough contact with the oxidizing solution. Ground glass connections were used throughout except the tube connections indicated on the diagram. These connections were effected by small pieces of transparent Tygon\* tubing which were entirely airtight. The 2-way stopcock B allowed the air to be directed either through the sample flask or the bypass.

#### *B. Reagents:*

1. 0.100 N  $\text{K}_2\text{Cr}_2\text{O}_7$  solution—containing 4.902 g. powdered dried reagent grade  $\text{K}_2\text{Cr}_2\text{O}_7$ , per liter.
2. 0.01 N  $\text{Na}_2\text{S}_2\text{O}_3$  solution—containing about 2.5 g.  $\text{Na}_2\text{S}_2\text{O}_3$  and a small amount of  $\text{Na}_2\text{CO}_3$  per liter. This solution was made and standardized according to the directions of Kolthoff and Sandell (2).
3. 75 per cent by weight solution of  $\text{H}_2\text{SO}_4$ .
4. Reagent grade KI crystals.
5. Starch solutions (3).
6. Reagent grade  $\text{BaCl}_2 \cdot 2\text{H}_2\text{O}$  crystals.

\* Will Corporation.

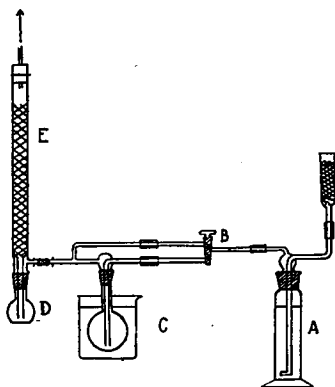


FIGURE 1.

In making these solutions care should be taken that the distilled water is free from oxidizable volatile material. Traces of  $H_2S$  found in some water are especially troublesome, and such water must be boiled in the open air in order to expel volatile impurities.

In addition to the foregoing reagents, a standard solution of the ether should be prepared for use in testing the apparatus and acquiring facility with the method. In the case of metopryl, one ml. of the ether at exactly 16 C. drawn into a chilled pipet and added rapidly to 999 ml. of distilled water at the same temperature will give a solution containing 0.726 mg. per ml. This solution should be kept cold and made fresh every two or three days.

*C. Drawing the blood:* Blood was drawn from the patient by the anesthetist in the operating room. A few crystals of sodium oxalate in the syringe will prevent clotting. A 5 ml. graduated syringe was used and 5 ml. of blood were drawn from the patient before the anesthetic had been administered. A second sample was taken at the point in the anesthesia which was judged clinically to be suitable for surgery (usually second or third plane, third stage). When the blood was drawn, the needle was removed at once and an airtight syringe cap was placed on the syringe. It was then promptly placed in an ice bath or the freezing compartment of an ice box and the blood was not removed from the syringe for analysis until it was thoroughly chilled.

*D. Procedure:* Exactly 5 ml. of the  $K_2Cr_2O_7$  solution and 20 ml. of 75 per cent  $H_2SO_4$  were placed in the reagent flask, heated with a Bunsen burner until boiling began (about 1 minute), and boiled for one half minute. The 2 ml. sample of oxalated blood was measured from the 5 ml. syringe into the chilled sample flask, containing 15 ml. of cold

distilled water, and the flask was immediately connected to the apparatus. With the stopcock turned so that the air stream went through the bypass, gentle suction was applied until the oxidizing solution had been drawn up into the column. The stopcock was then turned, directing the air through the sample. A beaker of water at 70 C. was placed under the sample flask, and the air was allowed to pass through the sample for five minutes.

At the end of this time the air was again directed through the bypass, the suction removed, and the reagent flask was lowered from the apparatus. One hundred ml. of distilled water was poured down the column into the flask in divided portions, and a small amount was run from a pipet over the outside of the tube extending below the glass joint of the column. The flask was then cooled in ice water.

The sample flask was removed and the tube rinsed off with distilled water and dried on the outside. One hundred ml. of 75 per cent sulfuric acid (which may be used repeatedly) was poured down the column of beads in divided portions. The apparatus was then ready for another determination.

The cooled reagent solution was transferred quantitatively into a 500 ml. Erlenmeyer flask using 100 ml. of distilled water in several portions. Approximately 50 g. of  $\text{BaCl}_2 \cdot 2\text{H}_2\text{O}$  crystals was added to the solution, and the flask was shaken until the precipitation of  $\text{BaSO}_4$  seemed to be complete. About 0.6 g. crystalline  $\text{KI}$  was then added, and 0.01 N sodium thiosulfate solution was run in from a 50 ml. buret until the brown color of iodine had nearly disappeared. Ten cubic centimeters of starch solution was then added, and the titration continued until the color of the mixture turned from light blue to white.

## RESULTS

One example selected from a number of experiments on aqueous solutions of known quantities of ether in blood will suffice to illustrate the results obtained using this method. In this one case the calculation is shown in detail.

Reagent blanks in duplicate showed a loss of 0.46 ml. of 0.100 N  $\text{K}_2\text{Cr}_2\text{O}_7$  out of a total of 5 ml. started with, leaving 4.54 ml. on which the calculation is based. The actual volumes of thiosulfate solution required in the titration of the ether samples were 34.2 and 34.4 ml., respectively, at normality 0.0104. Calculating from these titrations the quantities of  $\text{K}_2\text{Cr}_2\text{O}_7$  still present in the solution we have 3.56 and 3.58 ml. Subtraction of these from the blank value gave 0.98 and 0.96 ml. of 0.100 N  $\text{K}_2\text{Cr}_2\text{O}_7$  solution consumed by the oxidation of the ether. Multiplying each of these values by the calculated factor of 0.742 to determine the milligrams of ether in each case, we have 0.727 and 0.712. The known amount of ether used in each of these determinations was 0.726 mg. A short list of similar results on known quantities of ether is tabulated:

Mg. Ether Used	Analytical Results in Duplicate	
0.726	0.727	0.712
1.550	1.580	1.550
2.180	2.240	2.220

These data indicate that the method affords a satisfactory degree of accuracy.

A tabulation of results obtained on 10 surgical patients selected from a series of clinical cases will show the results obtained on blood.

A complete report on the clinical significance of blood concentrations of the anesthetic will appear in a subsequent publication. The findings on the entire series will be reported at that time. The results included here are intended only as a demonstration of the method in actual practice.

The lack of agreement in a few of the cases listed in the table is ascribed to the difficulty in measuring blood accurately. Syringes with tight fitting plungers and dark lines around the bottom of the plunger afford the highest degree of accuracy.

#### SUMMARY

A method has been described by which methyl n-propyl ether concentrations in the blood of anesthetized patients may be determined.

The stoichiometric relationships involved in the method have been discussed.

An improvement in the reproducibility of the iodometric titration has been described.

Blood levels of methyl n-propyl ether in 10 surgical patients have been reported.

No.	Blank (K <sub>2</sub> Cr <sub>2</sub> O <sub>7</sub> left)	Titration Ml. of Thiosulfate		Ml. Dichromate Consumed in Reaction		Mg. Per Cent Metopryl		Normality of Thiosulfate
1	4.60	30.5	29.2	1.48	1.42	54.8	52.7	1.040 × 10 <sup>-2</sup>
2	4.75	35.4	35.5	1.07	1.06	39.7	39.4	1.040 × 10 <sup>-2</sup>
3	4.70	25.8	20.1	2.02	2.60	75.0	96.5	1.040 × 10 <sup>-2</sup>
4	4.88	28.1	28.5	1.27	1.22	47.2	45.3	1.286 × 10 <sup>-2</sup>
5	4.92	29.4	29.4	1.14	1.14	42.3	42.3	1.286 × 10 <sup>-2</sup>
6	4.85	26.5	22.1	1.44	2.01	53.4	74.5	1.286 × 10 <sup>-2</sup>
7	4.80	31.3	29.0	0.78	1.07	29.4	39.6	1.286 × 10 <sup>-2</sup>
8	4.62	23.0	22.4	1.77	1.84	65.7	68.2	1.24 × 10 <sup>-2</sup>
9	4.54	23.3	23.1	1.65	1.68	61.2	62.2	1.24 × 10 <sup>-2</sup>
10	4.52	19.0	20.0	2.35	2.48	87.2	75.7	1.24 × 10 <sup>-2</sup>

#### REFERENCES

1. Andrews, E.; Potter, R. M.; Friedemann, T. E., and Livingstone, H. M.: Determination of Ethyl Ether in Blood, *J. Lab. & Clin. Med.* **25**: 966-970 (June) 1940.
2. Kolthoff and Sandell: *Textbook of Quantitative Inorganic Analysis*, New York, Macmillan, 1936, p. 594.
3. Kolthoff and Sandell: *Textbook of Quantitative Inorganic Analysis*, New York, Macmillan, 1936, p. 589.