# Laboratory Methods

## *Direct Injection Method for Gas Chromatographic Measurement of Inhalation Anesthetics in Whole Blood and Tissues*

*Terukazu Yokota, M.D.,* Yasumitsu Hitomi, M.D.,† Kosuke Ohta, M.D.,† Futami Kosaka, M.D.‡*

A direct injection method for gas chromatography has been developed for rapid measurement of the concentration of inhalation anesthetics in whole blood and tissue. By the use of a specially designed sample vaporizing apparatus attached to the gas chromatograph, a 5 to 10 μl. sample of blood or liquefied tissue is injected directly into the sample space and heated at 100° C. for a period of 60 to 90 seconds. Only the gas components vaporized from the sample are introduced into the column and detector. The residue of the sample can easily be removed from the sample space. An ultrasonic wave homogenizer is used for preparation of tissue samples. Gas chromatography is done by the use of a single column, Molecular Sieve Type 5-A coated with 0.3 per cent diethylene glycol succinate, combined with a hydrogen flame ionization detector for measurement of cyclopropane, halothane, methoxyflurane and diethyl ether. For analysis of nitrous oxide, however, Molecular Sieve Type 13-X was used with a thermal conductivity detector. Coefficient of variation of the reproducibility of peak height is less than 5 per cent for each anesthetic. The entire procedure is simple and practically applied to clinical determination of the depth of anesthesia.

VARIOUS methods have been introduced for quantitative analysis of inhalation anesthetics in blood and tissues; besides physical and chemical analyses,[1, 2] gas chromatographic methods proposed so far include procedures such as extraction with organic solvents,[3-8] distillation,[9, 10] vacuum-extraction,[11, 12] gas-bubbling,[13] trapping,[14] equilibration,[15, 16] and direct injection.[7, 17-20] These procedures, with the exception of direct injection, have proved to be time-consuming with complicated pretreatment of samples before gas chromatography, and they are not readily applicable for practical estimation of the depth of anesthesia in the management of anesthetized subjects. For this purpose it has always been necessary to place samples of fresh whole blood or tissues directly into a gas chromatograph.

A direct injection method has been devised and the procedures developed are applicable for the rapid assessment of concentrations of the inhalation anesthetics in blood and tissues.

### Method

*Principle.* Inhalation anesthetics physically dissolved in blood and tissues are vaporized by heat and the gas components from a known amount of a sample are introduced into a gas chromatograph. Quantitative analyses are made by comparing the response peaks from experimental samples with those from control samples containing a known amount of anesthetics analyzed under the same conditions.

*Apparatus.* (1) The gas chromatograph used in this study was a Shimadzu Model GC-1C,* double parallel column type, equipped with both a hydrogen flame ionization detector ($H_2$-F.I.D.) and a thermal conductivity detector (T.C.D.).

* Instructor in Anesthesiology, Okayama University Medical School, Okayama, Japan.
† Postgraduates, Okayama University Medical School, Okayama, Japan.
‡ Professor of Anesthesiology, Okayama University Medical School, Okayama, Japan.
Accepted for publication June 21, 1967.

* Shimadzu Seisakusho, Ltd., Kyoto, Japan.

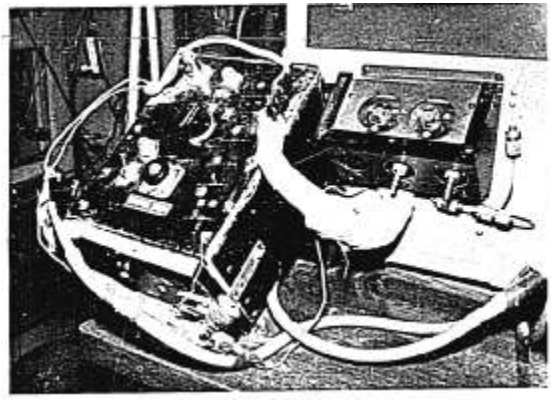

FIG. 1. The sample vaporizing apparatus attached to the gas chromatograph at the entrance to the column-bath.

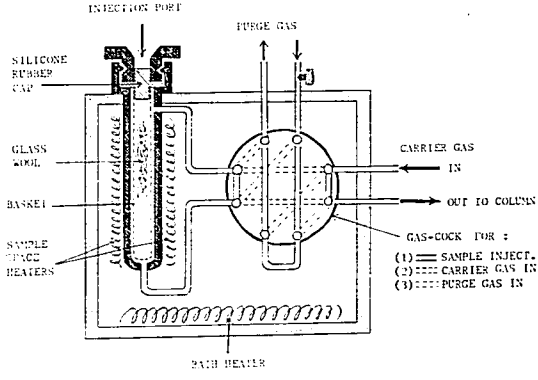


FIG. 2. The sample vaporizing apparatus. Sampling is done with the gas-cock set in turn at: (1) "sample injection"; the sample space is in a closed circuit in which vaporization takes place, while carrier gas flows separately. (2) "carrier gas in"; carrier gas stream is connected to the sample space. At this time gases vaporized from sample are introduced into the column. (3) "purge gas in"; purge gas lines are connected to the sample space, while carrier gas is bypassed at the same time. At this time the silicone rubber cap at the injection port is unscrewed and residue of sample removed. Thereafter, purge gas is flushed through the sample space with the cap screwed tightly.

(2) A sample vaporizing apparatus was devised and attached to the gas chromatograph at the entrance to the column. As shown in figures 1 and 2, the apparatus is a heat-tight box, consisting of a sample space, a gas-cock and a heater with a temperature controlling thermostat. The sample space is a tube of stainless steel of approximately 3.6 ml. with a tight silicone rubber cap at the top which can be unscrewed easily for cleaning the sample space. A small stainless steel basket in which a small amount of glass wool has been placed is attached to the rubber cap and inserted into the sample space (fig. 2).

The gas-cock diverts the gas stream into three different paths, namely, "sample injection," "carrier gas in" and "purge gas in."

The gas-cock is first set at "sample injection" in order to close off the heated sample space for injection and vaporization of a sample.

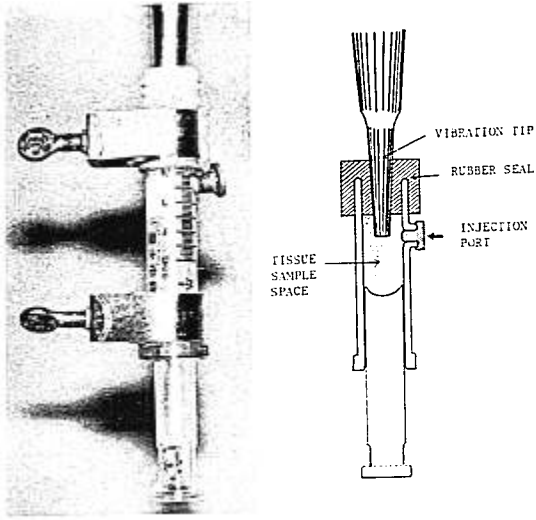


FIG. 3. Photograph (A) and diagram (B), of the tissue sample space, consisting of a 5 ml. syringe inserted into a rubber seal over the vibration tip of the ultrasonic wave homogenizer.

When vaporization of an anesthetic is complete the cock is turned to "carrier gas in" to connect the sample space to the carrier gas stream. At this time the gas components vaporized from the sample are introduced into the column. The cock is then turned to "purge gas in." This time the sample space is connected to the purge gas lines from which purge gas is flushed in the space, while carrier gas is bypassed with no disturbance of the flow. Clearing the sample space of the residue and flushing it with purge gas can be done at this time.

The heater with thermostatic control maintains the apparatus at a stable temperature. The conduits for carrier gas between the apparatus and the column bath are wrapped with ribbon-heaters to prevent condensation of the vaporized gases (fig. 1).

(3) An ultrasonic wave homogenizer, Model TA-4201 † equipped with a Type-4251 vibration tip was employed for liquefaction of tissue samples. Anaerobic management of a sample was insured by a special tissue sample space attached to the vibration tip (fig. 3).

† Kaijo Electric Co., Ltd., Tokyo, Japan.

The tissue sample space was made of a conventional 5 ml. syringe, the tip of which was cut off and tightly inserted into a rubber seal over the vibration tip (fig. 3). A small opening with a silicone rubber cap as an injection port was bored at the side of the syringe for the purpose of anaerobic introduction and withdrawal of the liquefied tissue through a needle. The bottom of the plunger was made concave to secure even mixing of the sample and to obtain maximum cavitation effect of the waves.

A small mass of brain tissue (0.3 g.), biopsied anaerobically in a special sampling syringe and weighed accurately, was injected through a 22-gauge needle into the "tissue sample space" previously filled with a known amount of distilled water. The sample was then exposed to ultrasonic waves, 19,500 cycles per second for a period of 20 to 30 seconds, concentrated via the vibration tip, resulting in complete conversion to a homogenized liquid.

*Sampling Procedure.* With a heparinized syringe and a needle of convenient size, a sample of blood was taken from an anesthetized

subject. The needle was removed and the syringe tip quickly sealed with a rubber cap. The needle of a Jintan Microsyringe,‡ 25 to 50 μl., was inserted through the rubber cap, and a 5 μl. sample of blood was withdrawn and injected directly into the heated sample space with the gas-cock set at "sample injection." A sample of liquefied tissue was also withdrawn in a microsyringe from the "tissue sample space" for direct injection, in the same way. The microsyringe was either rinsed with water or refilled with sample blood or tissue immediately after use, to prevent the needle and plunger from clogging. Rinsing with the subsequent sample is better to avoid dead space in the needle attached to the microsyringe.

After injection, the sample was heated at 100° C. for a period of 60 to 90 seconds. Then, the gas-cock was quickly switched to "carrier gas in" to introduce gases vaporized from the sample into the column and the detector.

The peak height of the anesthetic was recorded after a brief retention time, and when the recorder signal returned to the original base-line the gas-cock was turned to "purge gas in," so that water vapor from the sample could not further enter the gas chromatograph. Then, the cap of the sample space was unscrewed and the residue of blood or tissue removed. Removal of the residue is easy and complete because the glass wool in which the blood has diffused can be picked out and cleaned. Thereafter, the cap was screwed tightly and purge gas flushed through the sample space to replace the air inside with the carrier gas. The procedure for sampling took only a few minutes and sampling could be repeated every three minutes.

*Chromatographic Arrangements.* Nitrogen was used as carrier gas for the hydrogen flame ionization detector ($H_2$-F.I.D.) at a flow of 90 ml. per minute and hydrogen 250 ml. per minute for the thermal conductivity detector (T.C.D.).

A stainless steel U-tube column, 3 mm. i.d. and 75 cm. long, was packed with Molecular Sieve Type 5-A,§ 60/80 mesh, coated with 0.3 per cent diethylene glycol succinate for measurement of cyclopropane, halothane, methoxyflurane and diethyl ether, and Molecular Sieve Type 13-X,§ 60/80 mesh for nitrous oxide.

The $H_2$-F.I.D. was conditioned with hydrogen 45 ml. per minute and compressed air 0.5 liters per minute for an appropriate size of the paired jet flames.

Temperature settings were as follows: 100° C. for the sample space, 120° C. for the column bath and 130° C. for the detector.

The peak heights of anesthetics in blood were recorded with a sensitivity of 1,000 and the range 0.4 to 1.6 volts for the $H_2$-F.I.D., and the d.c. bridge current 170 mA. at 130° C. and the range 1 to 4 millivolts for the T.C.D. The chart speed was 10 mm. per minute for usual measurement. These specifications are summarized in table 1.

TABLE 1. Gas Chromatographic Arrangements

| | Cyclopropane, Halothane, Methoxyflurane and Diethyl Ether | Nitrous Oxide |
|---|---|---|
| Carrier gas | $N_2$, 90 ml./min. | $H_2$, 250 ml./min. |
| Column | Molecular Sieve 5-A coated with 0.3% diethylene glycol succinate, 75 cm. × 3 mm. (i.d.), 120° C. | Molecular Sieve 13-X 110 cm. × 3 mm. (i.d.), 130° C. |
| Detector | $H_2$-F.I.D. $H_2$, 45 ml./min. Air, 0.5 l./min., 130° C. | T.C.D. d.c. Bridge Current 170 mA., 130° C. |
| Sampling | | |
| Sample space | 100° C. | 80° C. |
| Heating time | 60–90 sec. | 60 sec. |
| Sample volume | 5 μl. | 10 μl. |
| Recording | | |
| Sensitivity | 1000 | d.c. 170 mA. at 130° C. |
| Range | 0.4–1.6 V. | 1–4 mV. |
| Chart speed | 10 mm./min. | 10 mm./min. |

‡ Jintan Thermo Co., Ltd., Tokyo, Japan.
§ Products of Shimadzu Seisakusho, Ltd., Kyoto, Japan.

*Calibration.* (1) LIQUID AGENTS (HALOTHANE, METHOXYFLURANE AND DIETHYL ETHER). Standard samples of known concentration in blood were prepared for calibation. This was accomplished by adding each weighed liquid agent to 100 ml. of bank blood in the range of clinical concentrations. The standard control samples thus prepared were subjected to gas chromatography and the recorded peak heights of the anesthetic plotted against their concentrations for a calibration curve.

(2) GASEOUS AGENTS (NITROUS OXIDE AND CYCLOPROPANE). A volume of pure gas was measured in an air-tight microsyringe and injected into the gas chromatograph. The volume of gas sample injected for calibration was converted to weight (W) by the following formula:

$$W\ (\text{mg. gas}) = N\ (\mu\text{l.}) \times \frac{M_w \times 10^3\ (\text{mg.})}{22.4 \times 10^6\ (\mu\text{l.})} \times \frac{P_b}{760} \times \frac{273}{273 + T_b}$$

where; N = microliters of gas sample injected for calibration.

$M_w$ = molecular weight (44.02 for $N_2O$, 42.08 for $C_3H_6$).
$P_b$ = barometric pressure, mm. of mercury.
$T_b$ = barometric temperature, degrees in centigrade.

With peak heights obtained from the gas sample and blood, the concentration in the subject blood sample was calculated by the following formula:

$$\text{Concentration (mg. \%)} = W\ (\text{mg. gas}) \times \frac{H_b}{H_G} \times \frac{100 \times 10^3\ (\mu\text{l.})}{V_B\ (\mu\text{l.})}$$

where; W = weight of gas sample injected for calibration, milligrams.

$H_B$ = peak height obtained from subject blood sample for measurement.
$H_G$ = peak height obtained from gas sample for calibration.
$V_B$ = volume of subject blood sample for measurement, microliters.

In practice, since W and $V_B$ were constant, concentration was obtained simply from two peak heights, $H_B$ and $H_G$, and their ratio.

## Evaluation of Methods and Results

*Sampling Assays.* The minimum temperature and time necessary for complete vaporization of volatile anesthetics from blood were first determined for halothane. The temperature of the sample space was set at 100° C. since the boiling point of halothane is 50.2° C., and only the length of heating was varied. Each peak height from the same sample was plotted against heating time (fig. 4A); time zero, or immediately after injection showed apparently incomplete vaporization, producing a lower peak with tailing. As the time was lengthened to 60 seconds or longer, peak heights increased to the maximum with complete vaporization.

Next, heating time being set for 60 seconds, the temperature was varied (fig. 4B); a low temperature also resulted in incomplete vaporization. More than 80° C. was necessary to obtain the maximum peak height owing to complete vaporization of halothane from blood. Peak height did not increase upon further heating; this was evidence that all of the anesthetic in a sample had been vaporized and entered the gas chromatograph.

Therefore, in the case of halothane, we decided that 5 $\mu$l. of blood should best be heated at 100° C. for 60 seconds. For other anesthetics with different physical properties, slightly modified sampling conditions were employed. For methoxyflurane, sampling was done at 120° C. for 90 seconds. Too high a temperature (150° C. or more) causes rapid clotting of blood that may interfere with complete vaporization of an anesthetic, resulting in a slightly lower peak (fig. 4B). A longer heating time would also minimize peak height (fig. 4A).

*Microsyringe Sampling.* Use of a microsyringe for sampling is simple but may introduce error. To minimize this error, a second scale was marked on the microsyringe on the opposite side of the original scale. Thus, the plunger of the microsyringe could be adjusted accurately each time on a straight line between the scales.

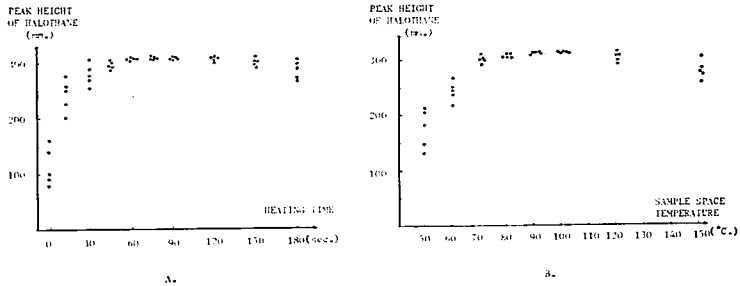


FIG. 4. Peak heights plotted against heating time (A) and temperature (B). The best sampling arrangement for complete vaporization of the anesthetic was sought; 5 μl. of control blood containing 17 mg.% halothane used as sample. (A) Temperature of the sample space set at 100° C. and heating time varied. (B) Heating time determined as 60 seconds and temperature varied.

The volumes of blood injected in this way were checked gravimetrically by means of a sensitive Direct Reading Analytical Balance §; the standard deviation of the mean was ±1.46 per cent. The accuracy was roughly equivalent to that of an internal standard technique [8, 17] for sampling. The use of a microsyringe, therefore, is justified for a practical measurement since simplicity is an important factor.

*Ultrasonic Homogenization of Tissue Samples.* Application of an ultrasonic wave homogenizer for rapid conversion of tissue to liquid had not been tried by previous investigators. The study was facilitated by our device of a tissue sample space for the homogenizer, as shown in figure 3.

Ultrasonic homogenization itself produced considerable heat and pressure that could cause leakage of volatile gases even though managed anaerobically. Thus, homogenization should be limited to the minimum time. We found that, when the tissue sample space of a syringe was immersed in ice water, 20 to 30 seconds was sufficient for complete liquefaction, with less than 2.5 per cent loss of anesthetic. The homogenization was particularly easy for cerebral tissue.

*Chromatographic Assay.* Anesthetic gases should be recovered completely and separated from other gaseous components such as water and organic vapors emerging from blood or tissue.

Various column-packings were tested. Silicone oil, polyethylene glycol, dioctyl phthalate and porapak type Q, specific for separation of organic anesthetics, produced varieties of baseline drifts owing to water and organic vapors. Absorbent materials such as activated charcoal, activated alumina and silica gel also combined some of anesthetic. Dual use of a pre-column packed with water absorbents, such as anhydrous calcium sulfate ($CaSO_4$), phosphorous pentoxide ($P_2O_5$) and anhydrous magnesium perchlorate ($Mg(ClO_4)_2$) as proposed by Jacobs,[7] Lowe [18] and Borgstedt and Gillies,[20] was not successful because these materials deliquesced upon absorption of water, and disturbed the flow of carrier gas.

A molecular sieve column was found to offer several advantages over these materials; it absorbed and retained a large amount of water.[20-22] We found that organic vapor from blood and tissue were also absorbed, but that elimination of volatile organic anesthetics did not occur. Hara [21] reported that molecular sieve type 5-A absorbed approximately 10 per cent of water by weight at a vapor pressure of 20 mm. of mercury at 120° C., and that this absorption capacity was nearly 5 times as great as that of silica gel and activated alumina. Deliquescence or swelling did not occur on absorption of water.

In our study, Molecular Sieve Type 5-A alone [22] or coated with 0.3 per cent diethylene glycol succinate not only separated anesthetic

gases from one another, producing sharp peaks with very minimum tailing (fig. 5), but also absorbed water and organic vapors from 5 $\mu$l. of blood or tissue. Molecular Sieve Type 13-X provided the best absorption and was utilized as a column for measurement of nitrous oxide which required the use of a thermal conductivity detector particularly sensitive to water vapor.

When a molecular sieve column was used in combination with a flame ionization detector for analysis of a single organic anesthetic in blood or tissue, there was only a single response peak recorded on a stable base-line, since inorganic gases ($N_2$, $O_2$, $CO_2$, $N_2O$, etc.) do not produce a response in a flame ionization detector.

Column temperature and carrier gas flow produced considerable effects on peak characteristics; too low a temperature and too little flow of carrier gas resulted in a low, slow peak with a long retention time. Temperature of the column bath had to be raised and kept at 120° C. with the carrier gas flow at 90 ml. per minute, in order to obtain a sharp peak.

The detectors ($H_2$-F.I.D. and T.C.D.) were kept at 130° C. and conditioned so as to maintain a sensitivity for recording sufficient anesthetic peaks on scale. The maximum sensitivity of the T.C.D. was gained by increasing the d.c. bridge current to 170 milliamperes at 130° C. for measurement of nitrous oxide in blood from a subject inhaling 50 per cent nitrous oxide in oxygen. Halothane or methoxyflurane inhaled with nitrous oxide did not appear as a response peak with this sensitivity of the T.C.D.

*Peaks.* Peak heights of anesthetics in blood and tissues were excellently recorded by this method, as shown in figure 5. The peaks were sharp, high and symmetrical with negligible tailing, and the height was considered to be more representative than the area under the curve.

Retention time for each anesthetic was different; cyclopropane 17, halothane 23, methoxyflurane 35, diethyl ether 73 seconds. A peak for nitrous oxide appeared following that of mixed inorganic gases ($N_2$, $O_2$, etc.). The retention time of nitrous oxide differed with water content absorbed in the column; it was 36 seconds when free of, and 20 seconds or less when saturated with water.

With the use of Molecular Sieve Type 13-X for measurement of nitrous oxide, the peak height from the same sample injected and measured under the same conditions gradually increased as sampling was repeated (fig. 6); the increase was minimal for the first 30 samplings and then became more and more

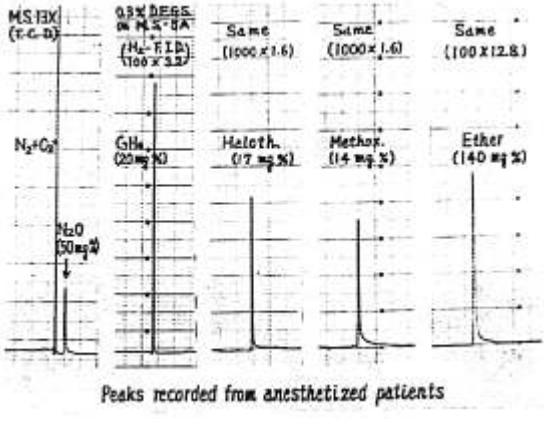


FIG. 5. Peaks of anesthetics in blood recorded from anesthetized patients.

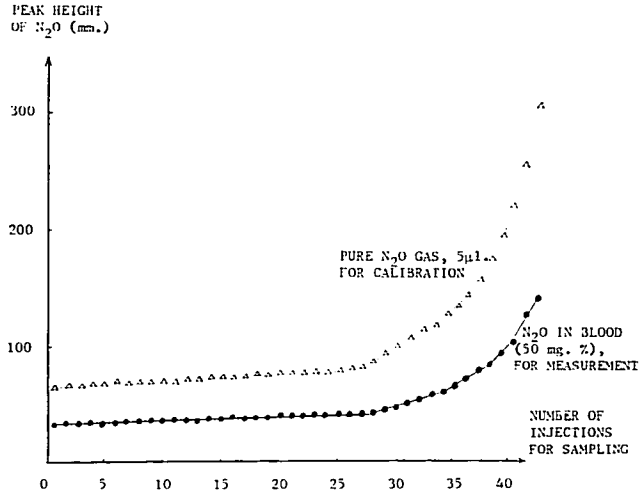


FIG. 6. Phenomenon of increase in peak height with Molecular Sieve Type 13-X column for measurement of nitrous oxide ($N_2O$) when the same sample was injected repeatedly under the same conditions. Peak height of pure $N_2O$ gas for calibration also increased in the same proportion.

prominent as sampling was repeated. At the same time the retention time of the peak became shorter.

On analysis, we found that this phenomenon was caused by water absorbed and retained in the molecular sieve column; that is, when the column became saturated with water vapor, both subject and control peaks (whether blood or gas) became higher in the same proportions. It seemed that the passage of carrier gas through the column was faster when saturated with water. Therefore for accurate calibration of nitrous oxide, a control sample of the gas was intermittently injected and measured for peak height so that the concentration of the subject sample could be calculated from the ratio of the two peak heights. Intermittent calibration required only a few minutes and did not greatly complicate the procedure. After about 30 samplings, the column was recharged or purged at higher temperature overnight in order to regain the original conditions. This was not necessary with the use of Molecular Sieve Type 5-A for organic volatile anesthetics.

The reproducibility of peak heights, representing the accuracy of measurement, was determined by ten successive injections of the same sample. The coefficient of variation (relative standard deviation of the mean) calculated for each anesthetic was; nitrous oxide and cyclopropane ±3.2, diethyl ether ±3.3, halothane ±4.2 and methoxyflurane ±4.8 per cent. Recovery rates were checked by comparing peak heights of halothane in blood with those of halothane vapor in air, the amount of which were theoretically equivalent. The peak height of halothane in blood was 97.96 (±2.1) per cent of that in air.

## Discussion

Our direct injection method for gas chromatography enables rapid and accurate measurement of the concentration of inhalation anesthetics in blood and tissues. In methods hitherto reported by Parker *et al.*,[17] Jacobs,[7]

Lowe,[19] Laasberg and Etsten [19] and Borgstedt and Gillies,[20] blood samples were injected into a heated injection port with carrier gas streaming constantly. However, injection of blood into a streaming carrier gas can cause problems, such as contamination of the instrument with residues of blood, the need for and frequent change of water absorbents,[7, 19] irregular base-line drifts [17, 18] and earlier decay of separation columns owing to more water and organic vapors entering the column and detector. The flow of carrier gas is disturbed by blood clots. An injection port has to be heated to 170° C. to 200° C. to accomplish rapid vaporization of samples. Microsyringe needles are also easily clogged with blood when the temperature is too high. In order to overcome these problems associated with direct injection into a streaming carrier gas, a sample space should be separated from the carrier gas stream so that a sample can be heated in a closed space. Inhalation anesthetics are relatively volatile and do not require much time for complete vaporization. Water and organic compounds, on the other hand, continue to evaporate even after carrier gas has swept the sample space. In our study the residues of blood and tissues were still slightly moist when picked out of the sample space. Such a large amount of vapor other than the anesthetic should be prevented from entering the gas chromatograph as much as possible. Our design of a "sample vaporizing apparatus" fulfilled this purpose.

Another major problem in the method is the selection of chromatographic columns. Conventional column-packings for separation of organic anesthetics do not absorb the water vapor which interferes with clear recording of anesthetic peaks. Lowe [18] reported that chromatographic columns were not necessary for measurement of a single volatile organic anesthetic with the use of a flame ionization detector. However, we found that the response of the detector had been produced by the mixture of the anesthetic and water vapor from blood, and that water must be absorbed and eliminated for stable gas chromatography. Combined use of a pre-column packed with water absorbents was not satisfactory, as noted above. In our study, molecular sieve packed in a single column sufficed.

A blood/gas coefficient, essential in an equilibration method,[15, 16] is not needed in our method since calibration is accomplished by directly comparing the peak heights either from whole blood (for liquid anesthetics) or from pure gas (for gaseous anesthetics) as controls.

The coefficient of variation of the reproducibility of peak height, is minimal and the method developed by us is applicable to accurate determination of anesthetic concentrations in blood.

Because the procedure is easy, blood anesthetic concentrations can be determined in the operating room.

## Summary and Conclusion

A practical method for quantitative analysis of inhalation anesthetics in whole blood and tissues has been developed. A 5 μl. sample of blood or homogenized tissue is injected directly into the heated sample space of a vaporizing apparatus specially designed for gas chromatography. Complete vaporization of the anesthetic from a sample requires 60 to 90 seconds at 100° C. A carrier gas is then passed through the sample space for introduction of the gases into a molecular sieve column. The sample residue can be removed easily without contamination of the instrument. A single sharp peak for an anesthetic is recorded on a stable base-line since water and organic vapors from the sample are completely absorbed and eliminated. The reproducibility of peak height was satisfactory with the coefficient of variation less than 5 per cent for each anesthetic. The sampling procedure is simple and the method has been utilized in clinical and experimental studies.

We wish to express our appreciation to Mr. Tsuyoshi Namba for his cooperation and assistance in technical arrangements for gas chromatography and construction of the sample vaporizing apparatus.

## References

1. Van Slyke, D. D., and Neill, J. M.: The determination of gases in blood and other solutions by vacuum extraction and manometric measurement, J. Biol. Chem. 61: 523, 1924.
2. Duncan, W. A. M.: The estimation of halothane in tissues, Brit. J. Anaesth. 31: 316, 1959.

3. Butler, R. A., and Hill, D. W.: Estimation of volatile anaesthetics in tissues by gas chromatography, Nature 189: 488, 1961.
4. Chenoweth, M. D., Robertson, O. N., Erley, D. S., and Gollike, R.: Blood and tissue levels of ether, chloroform, halothane and methoxyflurane in dogs, ANESTHESIOLOGY 23: 101, 1962.
5. Butler, R. A., and Freeman, J.: Gas chromatography as a method for estimating concentrations of volatile anesthetics in blood, Brit. J. Anaesth. 34: 440, 1962.
6. Rutledge, C. O., Seifen, E., Alper, M. H., and Flacke, W.: Analysis of halothane in gas and blood by gas chromatography, ANESTHESIOLOGY 24: 862, 1963.
7. Jacobs, E. S.: The gas chromatographic determination of halopropane in blood, Anesth. Analg. 43: 177, 1964.
8. Wolfson, B., Ciccarelli, H. E., and Siker, E. S.: Gas chromatography using an internal standard for the estimation of methoxyflurane levels in blood, Brit. J. Anaesth. 38: 29, 1966.
9. Dyfverman, A., and Sjoevall, J.: Estimation of fluothane by gas chromatography, Acta anaesth. Scand. 6: 171, 1962.
10. Gadsden, R. H., McCord, W. M., and Woods, E. F.: Gas chromatographic determination of methoxyflurane in blood, ANESTHESIOLOGY 23: 831, 1962.
11. Noehren, T. H., and Cudmore, J. W.: Ethyl ether content in blood as determined by gas chromatography, ANESTHESIOLOGY 22: 519, 1961.
12. Hamilton, L. H.: Gas chromatography for respiratory and blood gas analysis, Ann. N. Y. Acad. Sci. 102: 15, 1962.
13. Galla, S. J., and Ottenstein, D. M.: Measurement of inert gases in blood by gas chromatography, Ann. N. Y. Acad. Sci. 102: 4, 1962.
14. Fabian, L. W., Bourgeois-Gavardin, M., and Stephen, C. R.: A method for determining vapor concentrations of volatile anesthetic drugs, ANESTHESIOLOGY 19: 51, 1958.
15. Purchase, I. F. H.: Estimation of halothane tensions in blood by gas chromatography, Nature 198: 895, 1963.
16. Yamamura, H., Wakasugi, B., Sato, S., and Takebe, Y.: Gas chromatographic analysis of inhalation anesthetics in whole blood by an equilibration method, ANESTHESIOLOGY 27: 311, 1966.
17. Parker, K. D., Fontan, C. R., Yee, J. L., and Kirk, P. L.: Gas chromatographic determination of ethyl alcohol in blood for medicolegal purposes, Anal. Chem. 34: 1234, 1962.
18. Lowe, H. L.: Flame ionization detection of volatile organic anesthetics in blood, gases and tissues, ANESTHESIOLOGY 25: 808, 1964.
19. Laasberg, L. H., and Etsten, B. E.: Gas chromatographic analysis of cyclopropane in whole blood, ANESTHESIOLOGY 26: 216, 1965.
20. Borgstedt, H. H., and Gillies, A. J.: Determination of the solubility of $N_2O$ in water by gas chromatography, ANESTHESIOLOGY 26: 675, 1965.
21. Hara, N.: Molecular sieves—their properties and applications, Chem. Industr. 10: 63, 1957.
22. Yokota, T.: Gas chromatographic measurement of halothane in whole blood by "direct injection method," Jap. J. Anesth. 15: 1314, 1966.

## Anesthesia

**TRENDELENBERG POSITION** To ascertain if the use of Trendelenberg's position (defined as 20 degrees or greater head down) contributed to serious complication or death during gynecologic surgery, a questionnaire was sent to 266 chiefs of service in this country as to frequency of use of the position, incidence of serious complications or death, and any additional comments. Of 240 replies, very few reported serious difficulty because of the position. Those who used it often reported few complications, and those who used it infrequently reported the largest incidence of serious complications (50 per cent). Serious complications included respiratory embarrassment, brachial plexus palsy (11 patients), cardiac failure, and air embolism. The author concluded that since a majority of the respondents used the position, since a minority reported serious complication, and since all complications except brachial palsy were insignificant, that objections of anesthesiologists to the position are invalid. (*Menger, W. E.: Trendelenburg's Position: Current Usage, Obstet. Gynec. 29: 734 (May) 1967.*) ABSTRACTOR'S NOTE: The conclusions from this retrospective study, obtained from a group with undoubted bias, are of questionable validity.