EFFECT OF PREMEDICATION ON DURATION OF ANAESTHESIA WITH HALOGENATED VAPOURS: CHLOROFORM, TRICHLORETHYLENE, HALOTHANE, METHOXYFLURANE, ENFLURANE (ETHRANE*) AND ISOFLURANE (FORANE*)

DUCKSOOK KIM, M.D. AND ALLEN B. DOBKIN, M.D., F.R.C.P.(C)†

IN PREVIOUS STUDIES of the interaction of sedatives with thiopentone given intravenously, it was found that unstimulated trained dogs very often "slept" much longer than when thiopentone was administered alone. This response varied with the type of sedative given and was dose-related.¹⁻⁵

This study was undertaken to determine whether a similar interaction occurs between representative sedatives commonly used and nonflammable halogenated inhalation anaesthetics, and to determine whether the response is primarily due to the dose of the sedative or to the concentration of the anaesthetic.

METHODS

Female albino rats of the CFE strain, of uniform age and size (weight approx. 200 gm), were used to carry out dose-response tests with several sedatives given intravenously, and with several inhalation anaesthetics administered with oxygen in a closed jar.

Two problems had to be solved initially before the interactions could be studied. In order to reduce or eliminate as many gross physiological and pharmacological variables as possible, we had to determine the concentration of each anaesthetic which would produce a moderate depth of anaesthesia rapidly and the dose of sedative appropriate to cause light to moderate sedation within 10 minutes after intravenous injection.

First it was necessary to determine the appropriate range of concentration for chloroform, trichlorethylene, halothane, methoxyflurane, enflurane and isoflurane between the AC₅₀ and the LC₅₀ for this breed and size (age) of rat. This was accomplished by testing pairs of rats at various concentrations of each of the anaesthetics. Each pair of rats was placed in a jar of approximately 4 litres capacity filled with oxygen and a vapourised calculated dose of the liquid anaesthetic. After sealing, the jar was rotated manually 12 to 15 times per minute for 10 minutes. The speed with which unconsciousness and flaccidity supervened was observed and noted during this period. If the rats did not lose consciousness within three minutes, a higher concentration of the anaesthetic was tested with fresh rats until two appropriate doses (concentrations) were determined, at which

*Trademark of Ohio Medical Products, Division of Airco Inc., Madison, Wisconsin.

†From the Department of Anesthesiology, State University Hospital, State University of New York, Upstate Medical Center, Syracuse, New York, 13210, U.S.A.

TABLE I
CONCENTRATION AND PHYSICAL PROPERTIES OF ANAESTHETICS TESTED IN RATS

Anaesthetic	Molecular weight	Specific gravity	Partition coefficients		Est. MAC* (% conc.)	Conc. tested (%)	Liquid volume† (ml in 4 L jar)
			oil/gas	blood/gas			
Chloroform	119	1.49	400	10.3	0.55	1.5-2.0	.20-.27
Trichlorethylene	131	1.47	725	9.2	0.30	2.0-2.5	.28-.35
Methoxyflurane	164	1.42	900	13.0	0.25	1.0-1.5	.19-.29
Halothane	197	1.86	225	2.4	0.90	1.5-2.0	.26-.34
Enflurane	184	1.52	100	1.9	2.1	2.5-3.0	.50-.60
Isoflurane	184	1.50	95	1.4	1.4	1.5-2.0	.30-.40

*Analgesia is related to oil/gas partition coefficients.⁷

†Adjustments were made for elevated ambient temperature.

TABLE II
DOSES OF SEDATIVES TESTED IN RATS

Sedative	Dose administered intravenously (mg per 200-gm rat*)		
	Low	Median	High
Secobarbitone (Seconal®)	3.0	4.0	5.0
Chlorpromazine (Largactil®)	1.5	2.0	2.5
Methotrimeprazine (Nozinan®)	1.0	2.0	3.0
Hydroxyzine (Vistaril®)	1.5	2.0	2.5
Diazepam (Valium®)	1.0	2.0	3.0

*Adjustments in dosage were made for >10-gm variation in rat's weight.

several pairs *all* had satisfactory anaesthesia. Higher concentrations were tested at increments until the approximate LD₅₀ was found. After the ten minutes in the anaesthetic mixture the rats were transferred to a large basin, where they were observed. Notes were kept of the time and character of recovery to normal ambulation. The concentrations selected were on the basis that they produced anaesthesia within three minutes, that severe respiratory depression or cyanosis did not appear during maintenance and that recovery was not prolonged (> 5 minutes) after the ten-minute exposure.

The data from these tests were used in a graphic method for approximating the median effective dose (concentration) as recommended by Litchfield and Wilcoxon.⁶ The concentrations selected are listed in Table I. It is to be noted that with chloroform, trichlorethylene and methoxyflurane, the dose selected exceeded the estimated MAC on account of the high fat (oil) solubility and possibly the rapid rate of metabolism of these three agents, which may delay the onset of anaesthesia. On the other hand the doses selected for halothane and enflurane were in the approximate range of the estimated MAC and those for isoflurane were lower.⁷⁻¹⁰ It is interesting that isoflurane also appeared to be a more potent anaesthetic in man than the estimated MAC indicates.¹⁰

Secondly, graded doses of secobarbitone, chlorpromazine, methotrimeprazine, hydroxyzine and diazepam were injected into the dorsal tail-vein of similar rats. If no undesirable side-effects were observed, at least five rats were injected at each dose level. All animals were rechecked at the end of the day and followed up for 3 days to rule out delayed toxicity. After completing these tests, three appropriate doses of each premedicant were selected, doses at which the rats would be drowsy or quiescent without cyanosis and without more than momentary loss of the startle or righting reflex.

Finally, the interaction of the anaesthetics and the sedatives were tested in the following manner: five rats, each weighing approximately 200 gm, were given an intravenous injection of a selected dose of a sedative, as shown in Table II. Ten minutes after the injection of the sedative, the five rats together with one unmedicated control rat were placed in pairs in three capped glass jars of approximately 4-litre capacity, each filled with 100 per cent oxygen and a precisely measured amount of the liquid anaesthetic that would provide the desired vapour concentration (see Table I). The jars were rotated slowly (12 to

15 r.p.m.) while the rats were observed closely. The onset of unconsciousness, character of respiration, colour change (cyanosis) and degree of flaccidity were observed. After 10 minutes, the jars were opened to room air and the rats were transferred to a large plastic pan. A lucite pane was used as a cover so the rats could be observed and notes made of the rate and character of recovery up to steady ambulation without tactile stimulation. A similar set of tests was carried out with each dose of sedative and anaesthetic, but the rats were stimulated by manual prodding and tail-pressure (mild pain stimulus) during recovery to determine if recovery could be accelerated in this way.

The rats were acclimatized on a standard laboratory (high protein) diet in individual cages for several days before the tests. Fresh rats were used for every tested sedative-anaesthetic combination and at each dose level, in order to avoid spurious results due to cumulation, enzyme induction or other metabolic effects of the drugs. All tests were done in the morning in a temperature-regulated room where the rats were maintained at an ambient temperature of approximately 32° C, in order to facilitate rapid intravenous injections and to ensure rapid vapourization of the anaesthetics. Simultaneous written records were made of the observations on each rat during the tests.

RESULTS AND DISCUSSION

Preliminary Tests

The concentrations of the anaesthetics shown in Table I produced anaesthesia and flaccidity in the rats usually within 3 minutes without evidence of severe respiratory or cardiovascular depression. Recovery from a 10-minute exposure was invariably less than five minutes, even though a relatively high concentration of chloroform, trichlorethylene or methoxyflurane was used to accelerate the induction of anaesthesia. Induction and recovery from halothane, enflurane and isoflurane were always rapid.

As shown in Table II, the rats tolerated higher doses of secobarbitone (3 mg to 5 mg) than of the other sedatives. Chlorpromazine had a narrow, safe and effective dose range: at 3 mg or more, the rats died, while, at 1.0 mg, the rats were much too active. Methotrimeprazine caused convulsions at 4 mg, while sedation was satisfactory with 1 mg to 3 mg intravenously. Hydroxyzine was lethal at 3 mg, while the rats were too active when less than 1.5 mg was given. With diazepam, the safe range of dosage for satisfactory sedation was between 1 mg and 3 mg intravenously.

Sedative-Anaesthetic Interaction Tests

The two concentrations of each of the halogenated inhalation anaesthetics were found to be sufficient to produce smooth anaesthesia without mortality, whether or not a sedative was used. The mean duration of anaesthesia for each sedative-anaesthetic combination and control (anaesthetic alone) is shown in the 15 figures. The abscissa in each figure shows the concentration of the inhalation anaesthetic vapours used for the stimulated and the unstimulated groups of rats. The code marks on each graph represent the doses of the sedative injected

FIGURE 1. Rate of recovery from chloroform and trichlorethylene anaesthesia in stimulated and unstimulated rats sedated with secobarbitone (Seconal®).

intravenously using five rats at each dose level, one control rat for each dose tested and the standard error of the mean delay in recovery. The ordinate of each graph indicates the mean recovery time in minutes for each group of three control rats and the three groups of five rats that received progressive doses of the sedative.

Secobarbitone prolonged anaesthesia when combined with each of the inhalation agents. All control rats were active within 10 minutes while, in those which had received 5 mg of the sedative beforehand, recovery varied from more than three hours with methoxyflurane to approximately one hour with isoflurane. The smaller doses of secobarbitone showed a corresponding graded effect. It was interesting to observe that the stimulated rats did not recover faster than those which were not stimulated (see Figures 1, 2 and 3).

Chlorpromazine prolonged anaesthesia substantially with chloroform, trichlorethylene and methoxyflurane, but not with halothane, enflurane and isoflurane. The unstimulated rats usually recovered a little more slowly, but the difference in rate of recovery was not significant with any of the anaesthetics (see Figures 4, 5 and 6).

Methotrimeprazine acted quite similarly to chlorpromazine (see Figures 7, 8 and 9).

Hydroxyzine had the least influence on the rate of recovery from anaesthesia. Except following methoxyflurane, all rats recovered within 10 minutes. Stimulating the rats did not accelerate recovery. [Note that the time scale (ordinate) on

SECOBARBITONE

FIGURE 2. Rate of recovery from halothane and methoxyflurane anaesthesia in stimulated and unstimulated rats sedated with secobarbitone (Seconal®).

SECOBARBITONE

FIGURE 3. Rate of recovery from enflurane and isoflurane anaesthesia in stimulated and unstimulated rats sedated with secobarbitone (Seconal®).

CHLORPROMAZINE

FIGURE 4. Rate of recovery from chloroform and trichlorethylene anaesthesia in stimulated and unstimulated rats sedated with chlorpromazine (Largactil®).

CHLORPROMAZINE

FIGURE 5. Rate of recovery from halothane and methoxyflurane anaesthesia in stimulated and unstimulated rats sedated with chlorpromazine (Largactil®).

CHLORPROMAZINE

FIGURE 6. Rate of recovery from enflurane and isoflurane anaesthesia in stimulated and unstimulated rats sedated with chlorpromazine (Largactil®).

METHOTRIMEPAZINE

FIGURE 7. Rate of recovery from chloroform and trichlorethylene anaesthesia in stimulated and unstimulated rats sedated with methotrimepazine (Nozinan®).

METHOTRIMEPRAZINE

FIGURE 8. Rate of recovery from halothane and methoxyflurane anaesthesia in stimulated and unstimulated rats sedated with methotrimeprazine (Nozinan®).

METHOTRIMEPRAZINE

FIGURE 9. Rate of recovery from enflurane and isoflurane anaesthesia in stimulated and unstimulated rats sedated with methotrimeprazine (Nozinan®).

HYDROXYZINE

FIGURE 10. Rate of recovery from chloroform and trichlorethylene anaesthesia in stimulated and unstimulated rats sedated with hydroxyzine (Vistaril®).

HYDROXYZINE

FIGURE 11. Rate of recovery from halothane and methoxyflurane anaesthesia in stimulated and unstimulated rats sedated with hydroxyzine (Vistaril®).

HYDROXYZINE

FIGURE 12. Rate of recovery from enflurane and isoflurane anaesthesia in stimulated and unstimulated rats sedated with hydroxyzine (Vistaril®).

DIAZEPAM

FIGURE 13. Rate of recovery from chloroform and trichlorethylene anaesthesia in stimulated and unstimulated rats sedated with diazepam (Valium®).

DIAZEPAM

FIGURE 14. Rate of recovery from halothane and methoxyflurane anaesthesia in stimulated and unstimulated rats sedated with diazepam (Valium®).

DIAZEPAM

FIGURE 15. Rate of recovery from enflurane and isoflurane anaesthesia in stimulated and unstimulated rats sedated with diazepam (Valium®).

Figures 10, 11 and 12 with hydroxyzine has been spread by a factor of 10 as compared to the remaining figures.]

The response with *diazepam*^{11,12} was similar to that seen with chlorpromazine in combination with chloroform, trichlorethylene and methoxyflurane. Diazepam appeared to have a somewhat more pronounced depressant effect than chlorpromazine in the combinations with halothane, enflurane and isoflurane, but the differences were not great (see Figures 13, 14 and 15).

SUMMARY AND CONCLUSIONS

Tests were carried out on approximately 2,600 female CFE strain albino rats (mean weight 180 gm) to determine the safe range of concentration (median effective dose) of non-flammable halogenated inhalational anaesthetics for rapid induction to unconsciousness (< 3 minutes) for a brief period of anaesthesia (10 minutes); to identify the safe dose range of secobarbitone, chlorpromazine, methotrimeprazine, hydroxyzine and diazepam, to be administered intravenously and to determine the effect of the interaction of the sedatives and the anaesthetics – chloroform, trichlorethylene, halothane, methoxyflurane, enflurane and isoflurane – with particular reference to the duration of recovery to unimpaired ambulation. The rate of recovery was determined with and without stimulation by manual prodding and tail pressure (pain stimuli).

The concentrations of the anaesthetics were selected from the range of the median effective dose between the AC_{50} and the LC_{50} . On account of the requirement for a relatively rapid induction to unconsciousness, higher concentrations of chloroform (1.5 per cent and 2.0 per cent), trichlorethylene (2.0 per cent and 2.5 per cent) and methoxyflurane (1.0 per cent and 1.5 per cent) were used than if analgesia only had been the end-point. Although duration of recovery was always less than five minutes with the anaesthetics alone (control), there was a dose-dependent delay in recovery whenever a sedative was given, to which the above three anaesthetics contributed. These observations showed that secobarbitone, like thiopentone, clearly prolongs recovery from inhalation anaesthesia, while the effect of hydroxyzine in this respect appeared to be only slight. Diazepam, chlorpromazine and methotrimeprazine had an intermediate prolonging action on recovery time. The longer recovery period varied from approximately 15 minutes with hydroxyzine to more than 3 hours with secobarbitone.

Recovery was delayed appreciably less when the sedatives were combined with halothane (1.5 per cent and 2.0 per cent), enflurane (2.5 per cent and 3.0 per cent) and isoflurane (1.5 per cent and 2.0 per cent). When the latter three anaesthetics were used, the duration of recovery was often of the same order as in the controls (< 10 minutes) at the low dose of the sedatives, but was somewhat longer when the high doses of sedatives were used.

It is evident therefore that whenever sedatives are given, particularly if they have potent hypnotic activity, delayed recovery from anaesthesia should be expected. This delay is further augmented when more than an analgesic concentration of the inhalation anaesthetic is employed and when a large dose of a sedative is given.

The effect of physical stimulation of the rats appeared to be negligible in terms of accelerating recovery from anaesthesia probably because sensory deprivation outlasted the recovery of wakefulness and motor ability. It also appeared that the higher the oil/gas and blood/gas partition coefficients of the anaesthetic, the more likely is the recovery period to be prolonged by a sedative.

RÉSUMÉ

Nous avons fait des tests sur 2,600 rats blancs femelles (poids moyen 180 grammes) pour déterminer la moyenne de concentration en toute sécurité (dose efficace moyenne) de vapeurs d'un halogène anesthésique non-inflammable administré par inhalation pour faire une induction rapide jusqu'à l'inconscience (3 minutes) pour une anesthésie d'une brève période (10 minutes); nous songions également à préciser les doses moyennes de sécobarbital, de chlorpromazine, de methotrimeprazine, d'hydroxyzine et de diazepam qu'il faut donner par voie endo-veineuse et aussi pour déterminer l'effet de l'interaction des sédatifs et des anesthésiques - chloroforme, trichlorethylene, halothane, méthoxyflurane, enflurane et isoflurane - et cela en visant plus particulièrement la durée du réveil jusqu'à la démarche sans difficulté. La vitesse du réveil a été évaluée avec ou sans stimulation par attouchement manuel et pression de la queue (stimuli douloureux).

Les concentrations des anesthésiques ont été choisies à partir de l'ordre de la dose moyenne efficace entre l'AC₅₀ et l'LC₅₀. A cause de l'exigence d'une induction relativement rapide jusqu'à l'inconscience, nous avons employé des concentrations plus élevées de chloroforme (1.5 pour cent à 2 pour cent), de trichlorethylene (2.0 pour cent à 2.5 pour cent) et de méthoxyflurane (1.0 pour cent à 1.5 pour cent) qu'il n'aurait été nécessaire de donner si l'on avait voulu produire l'analgésie seulement. Bien que le délai pour le réveil a toujours été inférieur à cinq minutes avec les anesthésiques seuls (témoins), il y avait un délai dépendant de la dose dans le réveil à chaque fois qu'une sédation était donnée; les trois anesthésiques ci-haut mentionnés étaient un facteur dans ce délai.

Nos observations démontrent que le sécobarbitone tout comme le thiopentone prolonge clairement le délai du réveil des anesthésiques par inhalation, alors que l'effet d'hydroxyzine, sous cet angle, semble bien modeste. Le diazepam, la chlorpromazine et la methotrimeprazine n'exercent qu'une action modérée sur le retard du réveil. Le plus long délai du réveil a varié d'une durée d'environ 15 minutes avec l'hydroxyzine, à plus de trois heures avec le sécobarbitone.

Le délai pour le réveil a été plus court lorsque les sédatifs étaient associés à l'halothane (1.5 pour cent à 2.0 pour cent), à l'enflurance (2.5 pour cent à 3.0 pour cent), à l'isoflurane (1.5 pour cent à 2.0 pour cent). Lorsque les trois derniers anesthésiques étaient employés, le délai pour le réveil était souvent de l'ordre de celui des témoins (10 minutes) lors de l'usage de la faible dose de sédatif mais le délai était un peu plus long lors de l'usage de la dose forte de sédatif.

En conséquence, il a été évident que lorsque des sédatifs sont administrés, particulièrement s'ils possèdent une activité hypnotique puissante, il faut s'attendre à un réveil retardé de l'anesthésie. Le retard du réveil est plus long si l'on a donné une dose d'anesthésique par inhalation plus forte que la dose analgésique et si l'on

a donné une forte dose de sédatif. L'effet de la stimulation physique des rats nous a semblé négligeable pour accélérer le réveil de l'anesthésie probablement à cause du fait que le manque de sensibilité persiste plus longtemps que le retour à la conscience et à la locomotion. Selon toute apparence, plus les coefficients de partition huile/gaz et sang/gaz de l'agent anesthésique sont élevés, plus les sédatifs vont réussir à retarder le réveil.

ACKNOWLEDGMENTS

This study was carried out with grant support funds from Airco Inc. and Ayerst Laboratories. The authors are grateful for the invaluable technical assistance provided by Dr. Benjamin F. Africa, George D. Mann, R.T., Larry W. Eller, B.Sc., John W. White, B.Sc., and Judith D. Dudik, R.N., and for the statistical analyses of the data by Ashley A. Levy, Ph.D.

REFERENCES

1. DOBKIN, A.B. Potentiation of thiopental anesthesia by derivatives and analogues of phenothiazine. *Anesthesiology* 21: 292 (1960).
2. DOBKIN, A.B. Potentiation of thiopental anaesthesia with tigan, panectyl, benadryl, gravol, marzine, histadil, librium, and haloperidol. *Can. Anaes. Soc. J.* 8: 265 (1961).
3. DOBKIN, A.B. Prolongation of thiopental-induced sleep in dogs by narcotic analgesics. *Anesthesiology* 22: 291 (1961).
4. DOBKIN, A.B., ISRAEL, J.S., & CRISWICK, V.G. Prolongation of thiopental anaesthesia with hydroxyzine, SA 97, thiethylperazine and thioridazine. *Can. Anaes. Soc. J.* 9: 342 (1962).
5. DOBKIN, A.B. Sedatives, analgesics, antidotes and their interactions: A review. *Can. Anaes. Soc. J.* 11: 252 (1964).
6. LITCHFIELD, J.T., JR. & WILCOXON, F. A simplified method for evaluating dose-effect experiments. *J. Pharmacol. & Exper. Therap.* 96: 99 (1949).
7. EGER, E.I., II, LUNDGREN, C., MILLER, S.L., & STEVENS, W.C. Anesthetic potencies of sulfur hexafluoride, carbon tetrafluoride, chloroform and Ethrane in dogs: correlation with the hydrate and lipid theories of anesthetic action. *Anesthesiology* 30: 129 (1969).
8. DOBKIN, A.B., HEINRICH, R.G., ISRAEL, J.S., LEVY, A.A., NEVILLE, J.F., JR., & OUNKASEM, K. Clinical and laboratory evaluation of a new inhalation agent: compound 347 ($\text{CHF}_2\text{-O-CF}_2\text{CHF Cl}$). *Anesthesiology* 29: 275 (1968).
9. DOBKIN, A.B., NISHIOKA, K., GENGAJE, D.B., KIM, D.S., EVERS, W., & ISRAEL, J.S. Ethrane (compound 347) anesthesia: A clinical and laboratory review of 700 cases. *Anesth. & Analg.* 48: 477 (1969).
10. DOBKIN, A.B., BYLES, P.H., GHANOONI, S., & VALBUENA, D.A. Clinical and laboratory evaluation of a new inhalation anaesthetic: Forane (compound 469) $\text{CHF}_2\text{-O-CH Cl CF}_3$. *Can. Anaes. Soc. J.* 18: 264 (1971).
11. DOBKIN, A.B., ISRAEL, J.S., EVERS, W., & BISSET, C.M. Double-blind evaluation of diazepam for premedication. *Can. Anaes. Soc. J.* 17: 52 (1970).
12. PERISHO, J.A., BUECHEL, D.R., & MILLER, R.D. The effect of diazepam (Valium®) on minimum alveolar anaesthetic requirement (MAC) in man. *Can. Anaes. Soc. J.* 18: 536 (1971).