THE COMBINED ACTION OF PENTOBARBITAL AND MEPERIDINE, AND OF PROCAINE AND MEPERIDINE, IN GUINEA PIGS [1]

MADELAINE O. MAYKUT, PH D.[2]

SEVERAL AUTHORS (1, 2, 3, 4, 5) have noted that meperidine reduces the amount of barbiturate required to produce adequate narcosis. It is known that morphine, to which meperidine is distantly related, prolongs the depressant action of barbiturates (6, 7, 8, 9). Recently a profound respiratory depression was reported with thiopental when premedication with morphine or meperidine was given (10) This respiratory depression had been reported earlier (11), but the degree of depression was not considered serious at the time since it was compared with that occurring with methadone.

On the other hand, reports have appeared (12,13) indicating that meperidine possesses local anaesthetic action preceded by preliminary irritation. It was therefore of interest to see whether meperidine would augment the action of procaine and whether the combined effect of meperidine and pentobarbital had both antagonistic and synergistic trends, as was previously found with procaine and pentobarbital (14).

MATERIALS AND METHODS

The substances used were Nembutal® (pentobarbital sodium), Demerol® (meperidine hydrochloride), and procaine hydrochloride (Novocaine®).

The experimental animals were white guinea pigs. Injections were made in areas where the hair was clipped and, when only one drug was injected, physiological saline replaced the injection of the second drug. The first of a series of experiments, during which each animal was closely observed, was designed to survey qualitatively the effects of the combination of two drugs. The most conspicuous and consistent symptoms were subsequently translated into symbols. From such a survey dosages of the two drugs could be chosen to give graded responses which could then be analysed.

The acute toxicities of the drugs, administered by intramuscular or subcutaneous injection, alone and in combination, were determined; all animals died within the 24-hour period. The LD50's (lethal dose) and their standard errors were estimated as described by Finney (15)

Pentobarbital and Meperidine

The average body weight of the guinea pigs was 624 gm. with a standard deviation of 144 gm.

Pentobarbital 2 per cent solution was injected in divided doses intramuscularly into the hind limbs of the animals, after which an injection of 4 per cent meperidine solution in divided doses was made intramuscularly into the fore limbs of

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[2]Department of Pharmacology, University of Toronto.

the animals. Sleeping times indicated by the absence of the righting reflex were measured using ten animals at each dose level The same animals were used only once a week since it had been reported (16, 17, 18, 19, 20, 21) that tolerance results in decreased sleeping time after the first few daily injections of pentobarbital.

Procaine and Meperidine

The guinea pigs used had an average body weight of 537 gm. with a standard deviation of 180 gm.

A 4 per cent solution of meperidine was injected in divided doses intramuscularly into the hind limbs of the animals following which 10 per cent procaine was injected subcutaneously on both sides of the back of the guinea pig.

Since convulsions produced by meperidine and procaine were intermittent and not continuous, it was decided that it would not be possible to use the duration of convulsive state as an index of action; instead it was decided to determine merely the CD50, that is, the dose required to produce convulsions. A total of 178 animals was used in these experiments, the number at each dose level ranging from 10 to 35 guinea pigs because the convulsive response obtained was irregular.

Results

Pentobarbital and Meperidine

Pentobarbital alone gave rise to sleep; meperidine alone produced convulsions.

The hypnotic action of the combined drugs was investigated, and the results are presented in Table I and in a three-dimensional diagram (Fig. 1a); this diagram is similar to that presented in a previous report on procaine and pentobarbital (14). The axes indicate concentrations of pentobarbital (N) and meperidine (D) expressed in logarithms, the height of the diagram representing the average duration of sleep in minutes. The lowest combination N_1D_1 represents 65 minutes and the highest N_3D_3 563 minutes. While the duration of sleep obtained with pentobarbital and meperidine increases with increasing concentrations of meperidine for all doses of pentobarbital, it should be noted that the concentration of meperidine must be increased considerably to bring about any change (Fig. 1a).

The data were subjected to statistical analysis using the logarithm of response. The χ^2 test for over-all homogeneity of variance, although slightly significant, did not vary significantly and the variance does not change with increasing doses

TABLE I

Duration of Sleeping Time in Minutes following Meperidine and Pentobarbital

Dose of Meperidine (mg /kg)	Dose of pentobarbital (mg /kg)		
	15	21 2	30
2	65 3	126 6	190 1
18	124 8	175 7	270 4
162	201 4	326 6	563 4

of either drug. It seemed reasonable, therefore, to calculate the results using the logarithm of the response since the data would be presented in a more straightforward fashion and the curved surface approaches a plane when the log response is used (Fig. 1b). The increase in sleeping time with increase in pentobarbital (N_1 to N_3) and meperidine (D_1 to D_3) is statistically significant. The slopes of the lines for increasing pentobarbital dose effects at each meperidine dose level are

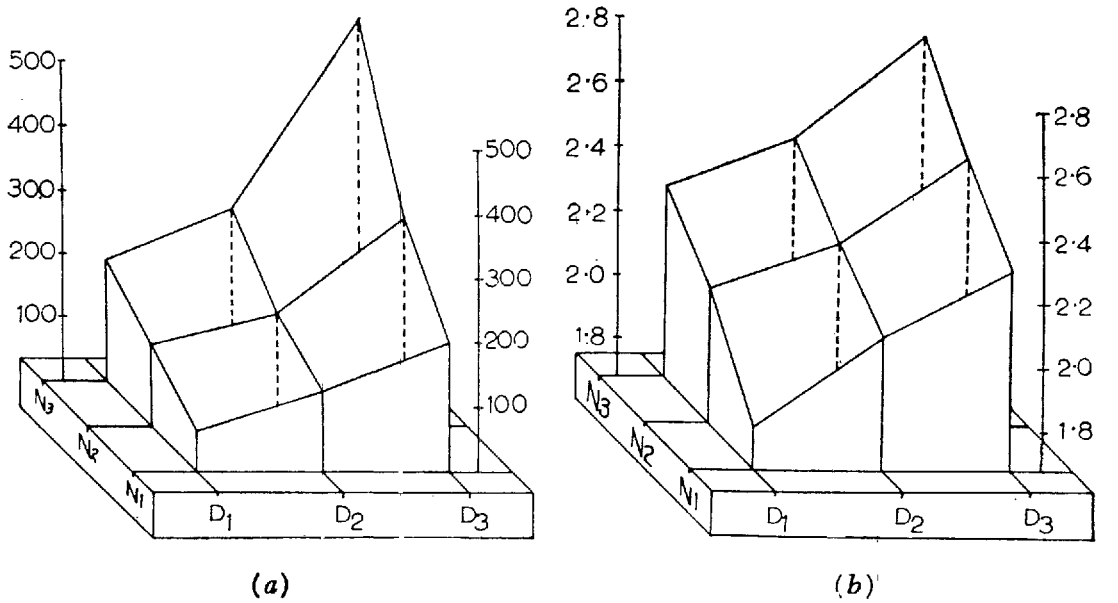

FIGURE 1. Hypnotic action of combinations of meperidine and pentobarbital in guinea pigs. The two horizontal axes represent increasing doses of the two drugs in logarithms; D stands for meperidine, N for pentobarbital: (*a*) the vertical axis represents sleeping time in min. (*b*) the vertical axis represents sleeping time in log min.

similar; the same is true for increasing meperidine dose effects at each pentobarbital dose level. Since the slopes are quite similar, interaction does not occur as is indicated in the analysis. On the basis of this statistical analysis the surface may be interpreted as a simple plane since no deviation from linearity occurs and the slopes are the same.

Table II lists the figures obtained for the acute toxicities of pentobarbital and meperidine, alone and in combination. The effect of meperidine on the LD50 of pentobarbital, as well as that of pentobarbital on the LD50 of meperidine is

TABLE II

ACUTE TOXICITIES OF PENTOBARBITAL AND MEPERIDINE(INTRAMUSCULAR) ALONE AND IN COMBINATION

	LD50 mg./kg.	±S.E.	Slope*	Number of guinea pigs
Pentobarbital	70.3	±4.7	7.9	35
Pentobarbital +35 mg./kg. meperidine	46.5	± 6.5	7.1	40
Meperidine	111.0	±14.0	9.2	30
Meperidine ±10 mg./kg. pentobarbital	315.0	±22.0	14.6	40

*Probit units per log dose.

significant. In the presence of meperidine (35 mg./kg. which is less than LD1) the LD50 of pentabarbital is decreased to 46.5 mg./kg. This amount of pentobarbital when used alone is equivalent to LD4. The toxicity of meperidene is decreased by pentobarbital, the LD50 being raised from 111 mg./kg. to 315 mg./kg. The latter amount of meperidine alone is equivalent to LD92.

Procaine and Meperidine

The symptoms noted, excitement, tenseness, unsteadiness, convulsions, resulting from the administration of various concentrations of meperidine and procaine, are shown in Figure 2. Attention is directed to the observations that the combination of meperidine and procaine is more toxic than either drug alone in similar dosage.

DEMEROL

PROCAINE mgm. per kgm.	2.5	5	10	20
0	N	N	N	E
100	N	E	T	T+E
200	G	X	X	X
400	X	X	X	X

N = normal T = tense

E = excited G = groggy

X = convulsions

FIGURE 2. Symptoms of individual guinea pigs given various dose combinations of meperidine (Demerol) and procaine.

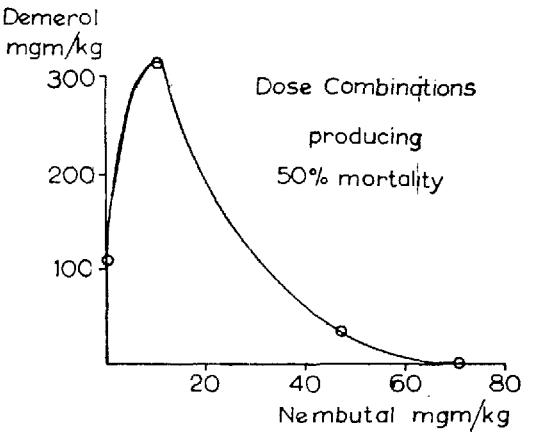

FIGURE 3. Lethal action of meperidine (Demerol) and pentobarbital (Nembutal), alone and in combination, in guinea pigs.

The values obtained for the CD50 of meperidine and of procaine are shown in Table III. When one-half the CD50 of both drugs was injected, the combined effect resembled potentiation; it was expected that 41.5 mg./kg. meperidine and 90.5 mg./kg. procaine injected into 40 animals would give a 50 per cent response but an 87.5 per cent response was observed.

The results obtained by combining procaine and meperidine in various doses are indicated in Table IV. This table shows that an increase in percentage of convulsions occurs with increasing procaine doses at all meperidine dose levels and a similar increase occurs with increasing meperidine doses at all procaine levels, but an erratic trend can be noted at the 9.0 mg./kg. meperidine level.

TABLE III

CONVULSIVE DOSE 50 OF MEPERIDINE AND PROCAINE ALONE

	CD50 (mg./kg.)	±S.E.	Slope	Number of guinea pigs
Meperidine (intramuscular)	83.1	±9.9	12.6	35
Procaine (subcutaneous)	180.9	±28.0	6.8	40

TABLE IV

PERCENTAGE OF CONVULSIONS IN GUINEA PIGS FOLLOWING MEPERIDINE AND PROCAINE

Dose of procaine (mg /kg)	Dose of meperidine (mg /kg)			
	4 5	6 5	9 0	12 5
104	20	40	36	40
125	35	50	50	72
150	70	80	61	100
CD50	134 5	119 0	126 6	110 1
±S E	17 5	17 9	21 5	8 8

In Table V are recorded the values obtained for the acute toxicities of procaine and meperidine, alone and in combination The effect of procaine on the LD50 of meperidine, as well as that of meperidine on the LD50 of procaine, is not significant, meperidine 90 mg./kg represents LD42 and procaine 402 mg /kg., LD45.

TABLE V

ACUTE TOXICITIES OF MEPERIDINE AND PROCAINE ALONE AND IN COMBINATION

	LD50 ±S E (mg /kg)		Slope*	Number of guinea pigs
Meperidine†	111	±14	9 2	30
Meperidine +300 mg /kg procaine	90	±5	11 7	30
Procaine†	438	±23	10 4	30
Procaine ±35 mg /kg meperidine	402	±76	5 4	40

*Probit units per log dose
†Meperidine given intramuscularly, procaine subcutaneously

DISCUSSION

Pentobarbital and Meperidine

The results obtained are similar to those previously reported for pentobarbital and procaine (14). That is, meperidine like procaine, increases the toxicity of pentobarbital. Further, pentobarbital appears to antagonize the effects of meperidine since the LD50 of meperidine is raised in the presence of pentobarbital. These double effects are represented graphically in Figure 3. In such a plot a strictly additive effect of the two drugs would be characterized by points lying on a straight line joining the LD50 of meperidine and the LD50 of pentobarbital (22). The hump in the curve represents the antagonistic effect of pentobarbital and meperidine; the lower part of the curve indicates that the central depressant action of meperidine is very nearly additive to that of pentobarbital. From this it could be argued that a patient under the toxic influence of meperidine might be saved by the administration of pentobarbital, whereas in a case of barbiturate poisoning an additional injection of meperidine might prove fatal. Furthermore, these investigations emphasize that meperidine has two independent actions on the central nervous system; one is the production of convulsions, and this effect

is counteracted by pentobarbital; the second action is a depressant one and augments the action of pentobarbital. Isonicotinic acid hydrazide (INA) and some related compounds appear to have similar actions to procaine and meperidine when combined with pentobarbital (23). That is, pentobarbital action is prolonged by these compounds, and it antagonizes the stimulant action of INA on the central nervous system. Other drugs which have been reported to enhance the action of barbiturates are benadryl (24), thiambutene (25), 5-hydroxy tryptamine (26, 27), sulfonamides and derivatives (28, 29, 30), disulfiram (31), antihistamine (32), chlorpromazine (33), SKF 525A (34, 35), reserpine (36), and serotonin (37).

Acute toxicity studies of meperidine have been conducted in many species such as the mouse, rat, rabbit, cat, and dog, using various routes of administration (38). The present data cannot be compared since results were obtained on guinea pigs using the intramuscular route. However, the symptoms of excitement and convulsions observed were similar (39).

Procaine and Meperidine

The toxicities for both drugs, procaine and meperidine, were increased by using them in combination, as may be seen from the graphical presentation of the results (Figs. 4 and 5). The contours of the curves indicate that the convulsive effects of the two drugs combined are more than additive, whereas the lethal effects of the drugs are less than additive.

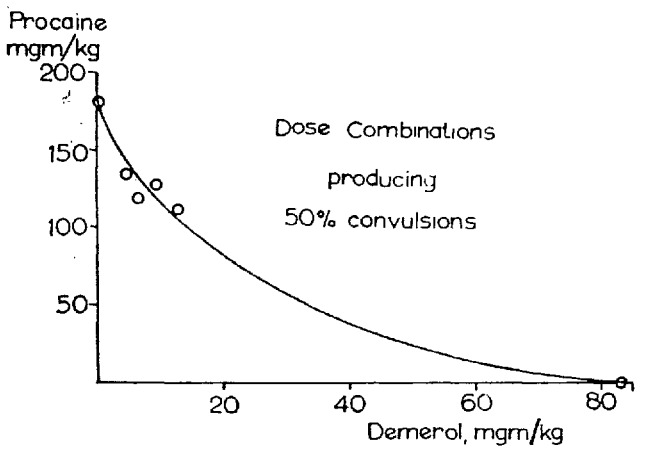

FIGURE 4. Convulsive action of meperidine (Demerol) and procaine, alone and in combination, in guinea pigs.

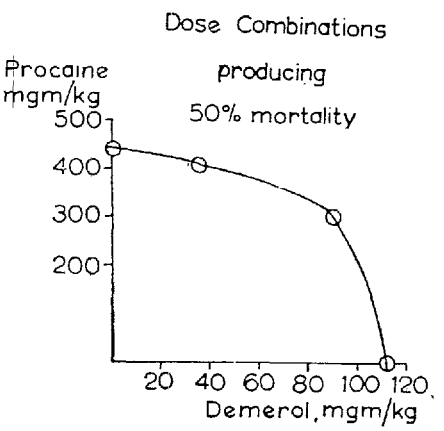

FIGURE 5. Lethal action of meperidine (Demerol) and procaine, alone and in combination, in guinea pigs.

Both drugs, meperidine and procaine, produce convulsions but the mechanisms responsible for this action may be different, since the slopes obtained for the CD50 of the drugs are different (Table III). However, the drugs combined produce additive effects. Also the slopes obtained for the CD50 (Table III) and the LD50 (Table V) of procaine as well as of meperidine are different, which may indicate that the mechanism of action giving rise to convulsions differs from that

causing death. Similar potentiation phenomena hve been reported (40) where the analgesic effect of some drugs increases with the simultaneous administration of local anaesthetics and a synergism of acute toxicity occurs when quinidine, meperidine, and procaine are used in combination (41).

Summary

Meperidine increases the sleeping time induced by pentobarbital in guinea pigs

The effect of pentobarbital on the toxicity of meperidine was antagonistic, whereas the effect of meperidine on the toxicity of pentobarbital was nearly additive.

The results of these experiments show that pentobarbital will protect experimental animals against the convulsive effects of meperidine and that the depressant action of pentobarbital is augmented by the presence of meperidine It has been pointed out that when meperidine and procaine are combined there is a potentiation of convulsive effects, but their combined lethal effects, while greater than that of either one alone, are not additive.

Acknowledgments

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Résumé

Des résultats expérimentaux ont laissé croire que la meperidine (Demerol®) avait un certain pouvoir comme anesthésique local et qu'elle pourrait diminuer également la quantité de barbiturique requise pour produire une narcose satisfaisante Il nous a donc paru intéressant de vérifier si la meperidine n'augmenterait pas l'action de la procaine (un anesthésique local) et de vérifier également si l'effet associé de la meperidine et du pentobarbital (un barbiturique) ne présenterait pas des tendances à la fois antagoniste et synergique comme on l'avait découvert au préalable pour la procaine et le pentobarbital.

Chez les cobayes, l'expérience a démontré que la meperidine augmente la durée du sommeil provoqué par le pentobarbital. Nous avons déterminé les doses toxiques (LD50) de ces médicaments employés seuls ou associés. Sur la toxicité de la meperidine, la pentobarbital s'est révélé un antagoniste et, sur la toxicité du pentobarbital, la meperidine a semblé avoir un effet presqu 'additionnel. Les résultats de l'expérience font croire que le pentobarbital peut protéger les animaux contre les effets convulsivants de la meperidine et que cette même meperidine augmente l'action déprimante du pentobarbital

Si l'on associe la meperidine et la procaine, il se produit une potentialisation de leurs effets convulsivants mais leurs doses léthales associées ne s'additionnent pas bien qu'elles soient plus grandes que l'une ou l'autre seule. Les mécanismes en jeu à l'occasion de convulsions peuvent être différents puisque les courbes obtenues pour les doses convulsivantes de ces médicaments 50 (CD50) sont différentes. Ainsi, les courbes obtenues pour le CD50 et le LD50 de l'un ou l'autre des médicaments, sont différentes, ce qui peut laisser croire que le mécanisme qui entre en jeu à l'occasion de convulsions est différent de celui qui cause la mort.

REFERENCES

1 Barlow, D W, Climenko, D R, & Homburger, E. Comparative Potentiating Effects of Certain Therapeutic Agents on Sodium Evipal Hypnosis. Proc Soc. Exper. Biol & Med *49*. 11 (1942)

2 Rovenstein, E A, & Batterman, R C The Utility of Demerol as a Substitute for the Opiates in Pre-anaesthetic Medication Anesthesiology *4*· 126 (1943).

3 Stephen, C. R, & Pasquet, A Anesthesia for Neurosurgical Procedures. Analysis of 1000 Cases Anesth & Analg *28*: 77 (1949).

4 Randall, H. S, Belton, M. K, & Leigh, M D Continuous Infusion of Demerol during Anaesthesia Canad M A. J *67*: 311 (1952).

5 Glassman, J M., & Seifter, J. The Effects of Analgesic Agents on Barbiturate Responses in Mice J. Pharmacol Exper Therap *115* 21 (1955).

6 Lundy, J S Intravenous Anesthesia Particularly Hypnotic Anesthesia and Toxic Effects of Certain New Derivatives of Barbituric Acid Anesth. & Analg. *9*. 210 (1930).

7. Rowbotham, S. Premedication Brit M J *2*. 693 (1931).

8. Seevers, M. H. Morphine Pentobarbital Anesthesia for Dog Surgery J Lab & Clin. Med. *19* 202 (1933)

9 Vogeler, K, & Kotzoglu, P Administration of Morphine before inducing Anesthesia by Sodium Evipan Internat Surg Digest *20*. 18 (1935).

10 Helrich, M, Eckenhoff, J E, Jones, R E, & Rolph, W D Influence of Opiates on the Respiratory Response of Man to Thiopental Anesthesiology *17*. 459 (1956)

11 Luduena, F P, Miller, L C., Ananenko, E, & Frick, J. D. Some Pharmacological Actions of Isomers of Methadon Federation Proc 7: 241 (1948).

12 Way, E L Studies on the Local Anesthetic Properties of Isonipecaine J Am Pharm A. *35*: 44 (1946).

13 Everett, F. G The Local Anesthetic Properties of Amidone (Dolophine) Anesthesiology 9: 115 (1948).

14 Maykut, M O, & Kalow, W Experiments with Animals on the Combined Action of Procaine and Barbiturates Canad Anaesth Soc. J. 2· 109 (1955).

15. Finney, D. J. In Burn, Biological Standardization, 2nd ed. London Oxford (1950)

16. Fitch, R H An Experimental Study of Tolerance to Barbiturates. J Pharmacol & Exper Therap. 39 266 (1930).

17. Carmichael, E. B Observations on Effect of Repeated Administration of Nembutal in Guinea Pigs Proc Soc Exper Biol & Med *30*. 1329 (1933).

18 Carmichael, E. B, & Posey, L C Studies on the Toxicity and Tolerance of Nembutal in Guinea Pigs. J. Pharmacol & Exper Therap *57* 116 (1936)

19. Ettinger, G. H The Duration of Anaesthesia Produced in the Dog by the Repeated Administration of Dial and Nembutal J. Pharmacol & Exper. Therap. *63* 82 (1938).

20. Masuda, M, Budde, R. N., & Dille, J. M. An Investigation of Acquired Tolerance to Certain Short-Acting Barbiturates. J. Am. Pharm. A. *27*: 830 (1938).

21. Kinsey, V E. The Use of Sodium Pentobarbital for Repeated Anesthesia in the Guinea Pig. J. Am Pharm A 29· 342 (1940)

22 GADDUM, J. H. Pharmacology, 4th ed. London: Oxford (1953).
23 GOLDIN, A., DENNIS, D.; VENDITTI, J. M.; & HUMPHREYS, S. R. Potentiation of Pentobarbital Anesthesia by Isonicotinic Acid Hydrazide and Related Compounds Science *121*. 364 (1955).
24 SHERMAN, J F. Enhancement of the Central Nervous System Effects of Strychnine and Pentobarbital by Diphenhydramine. Science *123*: 1170 (1956).
25. OWEN, L. N. Thiambutene and Barbiturate Anesthesia in the Dog J Pharm & Pharmacol 7. 533 (1955)
26. FORNAROLI, P, & KOLLER, M Interaction between 5-hydroxy-tryptamine and Narcotic Substances. Chem Abstr *50*. 6674a (1956).
27. PIERRE, R, & CAHN, J Essais d'anesthésie prolongée par la 5-hydroxytryptamine Compt rend Soc. de Biol. *149*: 1406 (1955).
28. ADRIANI, J. Effects of Anesthetic Drugs upon Rats Treated with Sulfanilamide. J Lab & Clin. Med. *24*: 1066 (1939).
29 BUTLER, T C., DICKINSON, H L, GOVIER, W M, GREER, C M, & LAMSON, P. D The Effect of Sulfanilamide and Some of Its Derivatives on the Reaction of Mice to Anesthetics. J Pharmacol & Exper. Therap *72* 298 (1941)
30 GOLDBAUM, L R & HUBBARD, T F Effects of Carinamide (4'carboxyphenylmethane sulfonanilide) on the Metabolism of Thiopental in Mice J Pharmacol. & Exper. Therap *99* 366 (1950)
31. GRAHAM, W D, CARMICHAEL, E J., & ALLMARK, M G The *in vivo* Potentiation of Barbiturates by Tetraethylthiuram Disulphide J. Pharm & Pharmacol *3*: 497 (1951).
32 LIGHTSTONE, H, & NELSON, J. W Antihistamine Potentiation of Pentobarbital Anesthesia J Am. Pharm. A *43* 263 (1954).
33 DUNDEE, J. W. A Review of Chlorpromazine Hydrochloride Brit J. Anaesth *26* 357 (1954).
34 MAXWELL, J W, COOK, L, DAVIES, G J, TONER, J J, & FELLOWS, E G Effects of Beta-diethylaminoethyl-diphenylpropylacetate Hydrochloride (SKF No 525A) on a Series of Hypnotics Federation Proc *12* 349 (1953)
35 ACHOR, L B, & GEILING, E M. K Effects of SKF Compound 525A on Excretion of Pentothal-S^{35} in Mice. Proc Soc. Exper Biol & Med *87*: 261 (1954).
36 BRODIE, B B, SHORE, P A, & SILVER, S. L Potentiating Action of Chlorpromazine and Reserpine Nature *175* 1133 (1955)
37 SHORE, P A, SILVER, S L, & BRODIE, B B Interaction of Serotonin and Lysergic Acid Diethylamide (LSD) in the Central Nervous System Experientia *11* 272 (1955)
38 TAINTER, M. L, & BUCHANAN, O H. A Comparison of Certain Actions of Demerol and Methadone California Med. *70*. 35 (1949)
39 BARLOW, O W, & LEWIS, J R Toxicity and Addiction Liability of Meperidine (Demerol) in Experimental Animals J Pharmacol & Exper. Therap. *103* 147 (1951).
40 UETSUKA, A, & OKUJIMA, Y. Increase of the Action of Analgesics by Simultaneous Administration of Other Drugs, particularly of Local Anesthetics Chem Abstr *47*: 7662b (1953).
41 CALESNICKI, B, SMITH, N H, & BEUTNER, R Combined Action of Cardiotoxic Drugs· A Study on the Acute Toxicity of Combined Quinidine, Meperidine, Pentobarbital, Procaine and Procaine Amide. J. Pharmacol & Exper Therap *102* 138 (1951)