Table IV—Activity of *N*-Substituted 1,2,3,4-Tetrahydro-2-naphthylamines in Rabbit Corneal Reflex Test[a]

| Compound | Mean Duration (Range) of Anesthesia, min |
|---|---|
| VIII | 21.8 (15–27) |
| IX | 9.4 (5–15) |
| X | 20.5 (15–27) |
| Lidocaine hydrochloride | 13.3 (10–15) |

[a] All compounds were tested at 1%.

In contrast to these highly active derivatives, IV, which has a nine-carbon chain on the nitrogen, was active in only one of three eyes but was relatively insoluble. The compound with a cyclohexyl ring (V) rather than the straight alkyl chain was active at 1% but had a relatively short duration of action; it also was only partially soluble at 1%. The introduction of hetero chains on the alkylamino group resulted in two compounds, VI and VII, which were inactive in the rabbit corneal reflex test.

Derivatives VIII–X were tested at a concentration of 1% and were all active in this test (Table IV). The potency and duration of anesthesia with these compounds were less than those observed with I–III but were generally comparable to those of lidocaine.

Derivatives I–III, which were active in the rabbit corneal reflex test, also were tested in the mouse sciatic nerve block test (Table III). Changing the position of the methoxy group from six to five (I and II) appeared to decrease the duration of action; increasing the length of the alkylamino group by one carbon (I and III) had no effect on the local anesthetic activity in this test.

The estimated intraperitoneal $LD_{50}$ values are presented in Table V. The $LD_{50}$ values of I and III, which exhibited the best local anesthetic activity in the rabbit corneal reflex and mouse sciatic nerve block tests, were similar. These $LD_{50}$ values were lower than the value of lidocaine but comparable to that of tetracaine.

These preliminary pharmacological evaluations suggested that the potency, duration of action, and relative safety of I and III were as great or greater than those of tetracaine. Compound I, *N*-heptyl-1,2,3,4-tetrahydro-6-methoxy-1-naphthylamine methanesulfonate, was identified as the most promising local anesthetic compound in these series and has been examined in more detail (5).

Table V—Estimated $LD_{50}$ Values of *N*-Substituted 1,2,3,4-Tetrahydro-1- and 2-naphthylamines, Tetracaine, and Lidocaine in Mice

| Compound | Estimated $LD_{50}$, mg/kg ip |
|---|---|
| I | 50 |
| II | 100 |
| III | 50 |
| IV | 200 |
| V | 200 |
| VI | 500 |
| VII | 300 |
| VIII | 80 |
| IX | 175 |
| X | 40 |
| Tetracaine hydrochloride | 75 |
| Lidocaine hydrochloride | 175 |

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# Local Anesthetic Activity and Acute Toxicity of *N*-Heptyl-1,2,3,4-tetrahydro-6-methoxy-1-naphthylamine Methanesulfonate


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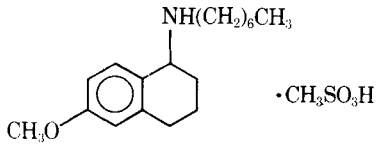
**Abstract** □ *N*-Heptyl -1,2,3,4- tetrahydro-6-methoxy-1-naphthylamine methanesulfonate (I) is a potent, long lasting local anesthetic. It was as potent as tetracaine and at least 10 times more potent than lidocaine in the rabbit corneal reflex, guinea pig wheal, and mouse sciatic nerve block tests. The threshold anesthetic concentration (TAC), defined as the concentration required to produce anesthesia lasting 5 min, was calculated from each linear regression line fitted to the log dose–duration data, and these values were used to compare the potencies of the local anesthetics. In the rabbit corneal reflex test, the TAC values were 0.04% for I, 0.04% for tetracaine, and 0.66% for lidocaine. In the guinea pig wheal test, I had a TAC of 0.02%, which was equipotent to tetracaine and 11 times more potent than lidocaine; epinephrine (1:100,000) prolonged the duration of action of all three local anesthetics but had the least effect with I. In the mouse sciatic nerve block test, the TAC values were 0.06% for I, 0.10% for tetracaine, and 0.86% for lidocaine. The acute $LD_{50}$ values of I in mice were 138 mg/kg sc and 26 mg/kg iv. By either route, I was less toxic than tetracaine and more toxic than lidocaine. Comparison of the $LD_{50}$ and TAC values in the mouse sciatic nerve block test indicated that I had a greater therapeutic index than either reference standard.



**Keyphrases** □ *N*-Heptyl -1,2,3,4- tetrahydro-6-methoxy-1-naphthylamine methanesulfonate—local anesthetic activity and acute toxicity evaluated □ Local anesthetic activity—*N*-heptyl-1,2,3,4-tetrahydro-6-methoxy-1-naphthylamine methanesulfonate evaluated □ Toxicity—*N*-heptyl-1,2,3,4-tetrahydro-6-methoxy-1-naphthylamine methanesulfonate evaluated


Several *N*-substituted 1,2,3,4-tetrahydro-1-naphthylamines, originally synthesized as antiparkinsonian agents (1) and monoamine oxidase inhibitors (2), showed potent local anesthetic activity in the rabbit corneal reflex test. Consequently, other 1- and 2-alkylamino derivatives were prepared and tested for local anesthetic activity (3). This




I

paper describes the pharmacological studies on the local anesthetic activity and acute toxicity of the most potent member of these series, *N*-heptyl-1,2,3,4-tetrahydro-6-methoxy-1-naphthylamine methanesulfonate (I).

## EXPERIMENTAL

**Local Anesthetic Activity**—Three test procedures, the rabbit corneal reflex test, the guinea pig wheal test, and the mouse sciatic nerve block test, were used to evaluate the local anesthetic activity of I. For comparison, lidocaine hydrochloride and tetracaine hydrochloride were tested concurrently; all concentrations are expressed as salts.

Simultaneous linear regression analysis was used to fit the best line to the log dose–duration data in each test. The threshold anesthetic concentration (TAC), defined as the concentration required to produce anesthesia lasting 5 min, was calculated from each regression line. This parameter has frequently been used to compare the potencies of local anesthetics (4).

**Rabbit Corneal Reflex Test**—Topical anesthetic activity was evaluated in the rabbit corneal reflex test originally described by Sollmann (5). A volume of 0.1 ml of the local anesthetic solution was instilled into the conjunctival sac of each eye; the cornea was not rinsed following drug application. The presence or absence of the corneal reflex (blinking) was determined by touching the cornea with a stiff hair at 5-min intervals. Each concentration (three or four per compound) was tested in six rabbit eyes.

**Guinea Pig Wheal Test**—Infiltration anesthetic activity was evaluated in the guinea pig wheal test using a modification of the method of Bulbring and Wajda (6). Each wheal was made by injecting 0.1 ml of the local anesthetic solution intradermally on the shaved back of a male albino guinea pig. The presence or absence of anesthesia was evaluated at 5-min intervals by the skin twitch response to pricking of the wheal area with a hypodermic needle. Six to eight wheals were made with each of the three concentrations tested for each drug. The wheals were examined 24 hr later to evaluate any tissue irritation. This test was also used to determine the effects of the addition of epinephrine (1:100,000) to the local anesthetic solutions.

**Mouse Sciatic Nerve Block Test**—Conduction anesthesia was evaluated using the mouse sciatic nerve block test, a modification of the method originally developed in rats (7). A volume of 0.05 ml of the local anesthetic solution was injected into the midthigh region of female ICR mice so that the drug was localized around the sciatic nerve. Following injection, the mice were observed to determine the frequency and duration of the motor nerve blocks as well as any systemic effects. For each drug, 10 mice were tested at each concentration (four or five per compound).

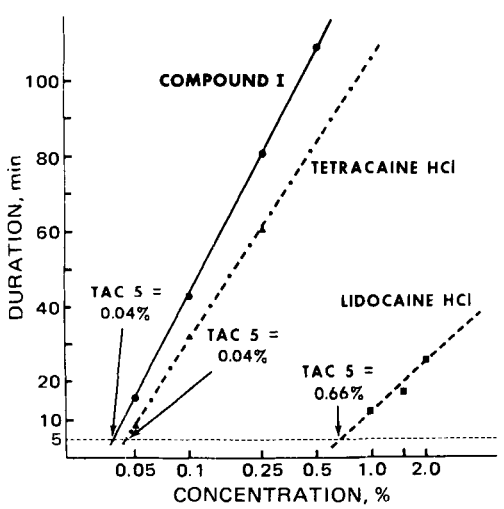



**Figure 1**—*Activities of local anesthetics in the rabbit corneal reflex test.*

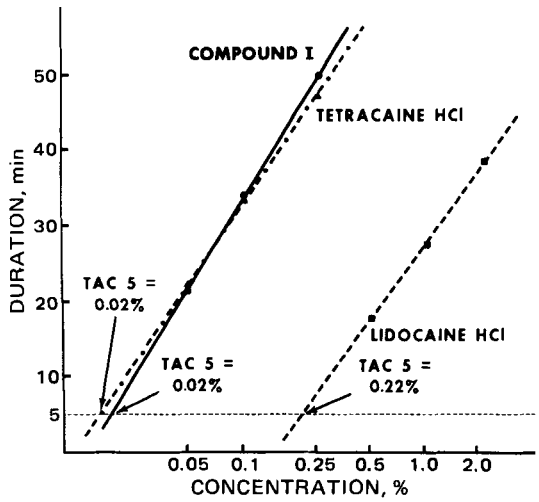



**Figure 2**—*Activities of local anesthetics in the guinea pig wheal test.*

**Acute Toxicity**—Acute toxicity was determined in female ICR mice following either intravenous or subcutaneous administration. Ten mice were tested at each dose level, and five or six doses were used for each $LD_{50}$ determination. The survivors were observed for 1 week. The $LD_{50}$ values were calculated by the method of Litchfield and Wilcoxon (8).

## RESULTS

**Rabbit Corneal Reflex Test**—The dose–response curves in the rabbit corneal reflex test are presented in Fig. 1. Compound I and tetracaine were active in the same concentration range, but lidocaine was at least 10 times less potent. The TAC values were 0.04% for I, 0.04% for tetracaine, and 0.66% for lidocaine. The slopes of the three lines were significantly different, with I having the steepest dose–response line.

**Guinea Pig Wheal Test**—The dose–duration curves in the guinea pig wheal test are shown in Fig. 2. The slopes of the three lines were not significantly different. The potencies of I and tetracaine did not differ significantly, and both compounds were approximately 11 times as potent as lidocaine. Examination of the wheals 24 hr after injection indicated that I caused more tissue irritation than either tetracaine or lidocaine.

The addition of the vasoconstrictor epinephrine increased the duration of the anesthesia produced by I approximately twofold while that of tetracaine was increased 2.5 times (Fig. 3). Epinephrine exerted the most pronounced effect when added to the lidocaine solutions. The duration of anesthesia was increased 2.8 times with the 0.5% solution and 5.3 times with the 2% solution. The addition of epinephrine also increased the amount of tissue irritation caused by all three local anesthetics. A similar incidence of irritation was seen with I and tetracaine when epinephrine was present in the test solutions.

**Mouse Sciatic Nerve Block Test**—The results of the sciatic nerve block test are shown in Fig. 4. At the lowest concentration tested, 0.05%, I was effective in 80% of the mice and tetracaine was effective in 90% of the mice. Lidocaine was ineffective at 0.05 and 0.1% concentrations and

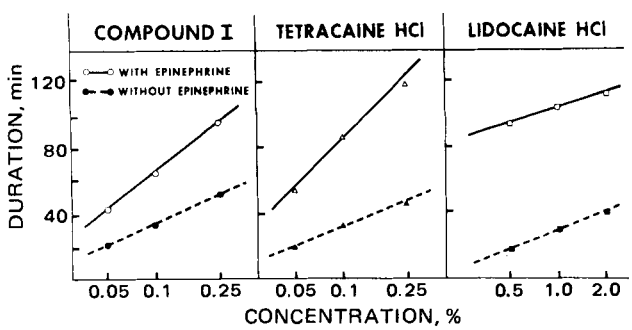



**Figure 3**—*Epinephrine (1:100,000) potentiation of local anesthetic activity using guinea pig wheal test.*

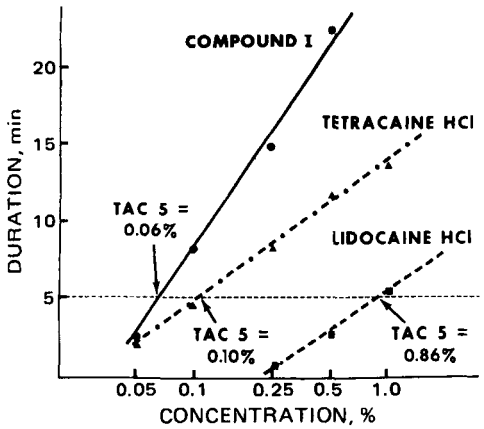



**Figure 4**—*Activities of local anesthetics in the mouse sciatic nerve block test.*

effective in 50% of the mice at the 0.25% concentration. The TAC values calculated from the regression lines were 0.06% for I, 0.10% for tetracaine, and 0.86% for lidocaine. For an average body weight of 20 g, the TAC values were 1.7, 2.5, and 21.4 mg/kg for I, tetracaine and lidocaine, respectively.

The slopes of the three lines were significantly different. As indicated by the slopes, the duration of anesthesia caused by the higher concentrations of I (0.25 and 0.5%) was longer than that produced by the same concentration of tetracaine. No overt symptoms were observed in the mice receiving I or lidocaine. However, at the 0.5 and 1.0% concentrations of tetracaine, representing approximately 12.5 and 25 mg/kg, jerks, tremors, and mild clonic convulsions were noted.

Therapeutic ratios were calculated for the three local anesthetics based on the TAC values, expressed in milligrams per kilogram, from the mouse sciatic nerve block test and the intravenous or subcutaneous $LD_{50}$ values (Table I). The intravenous therapeutic ratio of I was three times higher than tetracaine and 15 times higher than lidocaine. The subcutaneous therapeutic ratio of I was 5.5 times higher than tetracaine and 9.5 times higher than lidocaine.

**Table I—Comparison of I, Tetracaine, and Lidocaine**

| Parameter | I | Tetracaine Hydrochloride | Lidocaine Hydrochloride |
|---|---|---|---|
| Local anesthetic activity (TAC values) | | | |
| Rabbit corneal reflex, % | 0.04 | 0.04 | 0.66 |
| Guinea pig wheal, % | 0.02 | 0.02 | 0.22 |
| Mouse sciatic nerve, % | 0.06 | 0.10 | 0.86 |
| | 1.7[a] | 2.5[a] | 21.4[a] |
| Acute toxicity in mice | | | |
| Intravenous $LD_{50}$, mg/kg | 26 | 13 | 36 |
| Subcutaneous $LD_{50}$, mg/kg | 138 | 35 | 190 |
| Therapeutic ratios in mice | | | |
| Intravenous $LD_{50}$/sciatic nerve TAC | 15.3 | 5.0 | 1.7 |
| Subcutaneous $LD_{50}$/sciatic nerve TAC | 81.2 | 14.0 | 8.9 |

[a] Mouse sciatic nerve TAC percentages converted to milligrams per kilogram assuming an average body weight of 20 g. Values were used in therapeutic ratio calculations.

**Table II—Acute Intravenous and Subcutaneous Toxicity in Mice**

| Compound | Subcutaneous $LD_{50}$, mg/kg | Intravenous $LD_{50}$, mg/kg |
|---|---|---|
| I | 138 (118–162)[a] | 26 (23.4–28.8) |
| Tetracaine hydrochloride | 35 (28.7–42.7) | 13 (11.6–13.7) |
| Lidocaine hydrochloride | 190 (169.5–213) | 36 (31.8–40.7) |

[a] The 95% confidence limits.

**Acute Toxicity**—The $LD_{50}$ values for the three local anesthetics are found in Table II. The $LD_{50}$ values of I were 138 mg/kg sc and 26 mg/kg iv. Tetracaine was four times more toxic than I when administered subcutaneously and twice as toxic intravenously. By either route of administration, I was 1.4 times as toxic as lidocaine. Overt effects observed in the mice receiving I included decreased activity, jerks, tremors, opisthotonus, and clonic convulsions; similar effects were seen in the mice receiving either tetracaine or lidocaine.

## DISCUSSION

These studies have indicated that I is a potent local anesthetic with a long duration of action and relatively low acute toxicity. As shown by the threshold anesthetic concentrations summarized in Table I, the potency of I in the rabbit corneal reflex, guinea pig wheal, and mouse sciatic nerve block tests was comparable to that of tetracaine and at least 10 times greater than that of lidocaine. By either the subcutaneous or intravenous route of administration, I was less toxic than tetracaine but more toxic than lidocaine in mice.

Compound I represents a chemically novel type of local anesthetic since most currently marketed agents are either *p*-aminobenzoic acid derivatives such as tetracaine or amide derivatives related to lidocaine. This difference in structure might lessen the likelihood of allergic reactions among individuals known to be sensitive to the existing classes of local anesthetics.

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