RO-1-7780, a Potent Antagonist of Alphaprodine.* (22528)

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The narcotic antagonists have recently been reviewed(1). The hope that agents of this type would control undesirable side actions when given simultaneously with the narcotic has not been fully realized both because the time courses of action of the drugs used together have differed and because the antagonists have affected not only the undesirable actions of the opiates but the analgesia as well(2-12). Ro-7780‡ has received limited study previously(13-14); the purpose of the present work was to study its efficacy as an opiate antagonist and to determine its characteristics when used with alphaprodine (Nisentil) or morphine.

Methods. Ro-7780 was tested in conscious dogs and in dogs anesthetized with pentobarbital, 25 to 30 mg/kg. Male or female dogs weighing 8 to 12 kg were chosen. In addition a number of normal human volunteers and post-operative patients were given alphaprodine either alone or in combination with Ro-7780. All drug injections were made intravenously. Alphaprodine was administered in control studies on 6 conscious dogs; in later experiments in the same animals Ro-7780, 0.25 or 0.5 mg/kg, was given at the time of maximum apparent depression following administration of alphaprodine. Respiration, general activity and the effects of gross stimuli were observed. Two dogs also received 2 mg morphine per kg in similar studies. Experiments were next carried out to determine the effect of Ro-7780 in established alphaprodine depression in anesthetized dogs. The 9 animals used in this phase of the work were given pentobarbital and an endotracheal tube inserted. Expired gas volumes were measured using a wet test gas meter, arterial oxygen was determined according to Van Slyke and Neil using a manometric gas apparatus, and respiration and pulse were counted directly. Fifteen to 20′ after tracheal intubation in each animal alphaprodine (5-8 mg/kg) was injected. Artificial respiration was required for a period of 1 to 5 minutes in order to prevent death. On the resumption of spontaneous respiration, observations were made. Approximately 5 minutes later Ro-7780 (0.25 or 0.35 mg/kg) was injected and the measurements repeated immediately. In control experiments, which were carried out 7 to 10 days before or after the tests, Ro-7780 or no medication was given, and readings were made at the corresponding time intervals after anesthesia was induced. The same experimental design was used in 8 anesthetized dogs that received alphaprodine alone (4 or 5 mg/kg) in control experiments, while this dose of narcotic was given simultaneously with 1/75th or 1/100th the amount of Ro-7780 in test studies; arterial blood was drawn 5 to 8 minutes and 30 to 40 minutes following administration of the narcotic or of the narcotic-antagonist combination. In human subjects minute ventilation determined by an Emerson meter, respiratory and pulse rates, and blood pressure were recorded after alphaprodine alone (20-40 mg) in 4 cases and following combined administration of the same dose of alphaprodine with Ro-7780 at 1/75th or 1/100th the amount of narcotic in 6 cases. A period of 10 minutes with a variation in minute ventilation of less than 10% immediately prior to administration of the drugs was taken as the control and results were calculated as percent of this volume.

Results. In the 6 conscious dogs responsiveness was affected consistently while respiratory rate and pulse were variable and unpredictable. Alphaprodine by itself (2-10 mg/kg) always diminished the general ac-

* Supported by Hoffmann-LaRoche, Inc.

‡ This material was supplied by Hoffmann-LaRoche, Inc., under code number Ro-1-7780; and it is 1-3-hydroxy-N-propargyl morphinan tartrate.

TABLE I. Influence of Ro-7780 following Alphaprodine in 9 Dogs under Pentobarbital.

Pentobarbital alone*			Alphaprodine			Alphaprodine and Ro-7780		
Minute ventilation (L)	Respiratory rate	Arterial oxygen vol (%)	Minute ventilation (L)	Respiratory rate	Arterial oxygen vol (%)	Minute ventilation (L)	Respiratory rate	Arterial oxygen vol (%)
1.85	21	18.60	1.54	16	16.25	4.80	30	21.35
2.95	19	16.48	2.00	16	18.34	2.72	16	20.13
1.46	6	17.34	.94	7	14.40	4.70	23	20.04
1.49	16	16.97	.92	7	4.50	2.82	21	19.53
1.26	8	17.26	———	†	———	1.35	5	19.90
1.46	10	14.64	1.20	7	12.59	5.93	28	20.40
1.46	15	15.06	1.20	14	11.26	3.87	36	19.74
1.41	7	19.30	1.57	14	14.80	3.14	26	20.50
1.18	8	14.53	.93	6	10.15	2.27	17	16.56

Statistical comparisons by paired data:
A: Ventilation vol columns 1 cf. 4, $p < .02$; 1 cf. 7, $p < .01$; 4 cf. 7, $p < .01$.
B: Respiratory rate " 2 cf. 5, $p > .4$; 2 cf. 8, $p < .01$; 5 cf. 8, $p < .01$.
C: Arterial oxygen " 3 cf. 6, $p > .05$; 3 cf. 9, $p < .01$; 6 cf. 9, $p < .01$.

* Ro-7780 alone after pentobarbital had no effect on these measurements.
† Too severely depressed for measurements.

tivity and response to stimuli. Ro-7780 administered at the time of maximum depression after the narcotic produced a sharp increase in responsiveness in every animal. The animals that received morphine responded similarly to the antagonist.

In the 9 dogs under pentobarbital Ro-7780 (0.25 or 0.35 mg/kg) injected during a period of severe respiratory depression incident to the action of alphaprodine (5 to 8 mg/kg) produced a significant improvement in minute ventilation, respiratory rate and arterial oxygenation. The changes produced by alphaprodine alone were significant with respect to depression of minute ventilation and arterial oxygenation; but the spontaneous respiratory rate, after breathing was resumed, was not changed. Results from the individual animals are presented in Table I; paired data analysis was used for interpretation.† Ro-7780 had substantially no effect unless alphaprodine had previously been given. (On the basis of these experiments a somewhat lower dose of alphaprodine (4 or 5 mg/kg) was chosen as suitable in the subsequent animal work.)

Fig. 1 shows the mean results from studies of the combined administration of Ro-7780 and alphaprodine in dogs. Preliminary experiments had indicated that the antagonist should be given in a dose of about 1/75th to 1/100th the narcotic in order to prevent respiratory depression. The administration of alphaprodine by itself (4 or 5 mg/kg) produced a severe respiratory depression which lasted for about 12-15 minutes; 3 of the 8 animals, in fact, required artificial respiration for 1 to 7 minutes. When Ro-7780 was given contemporaneously with alphaprodine in the same animals, respiratory rate was significantly higher during this critical period (20 cf. 5), as were minute ventilation (1.45

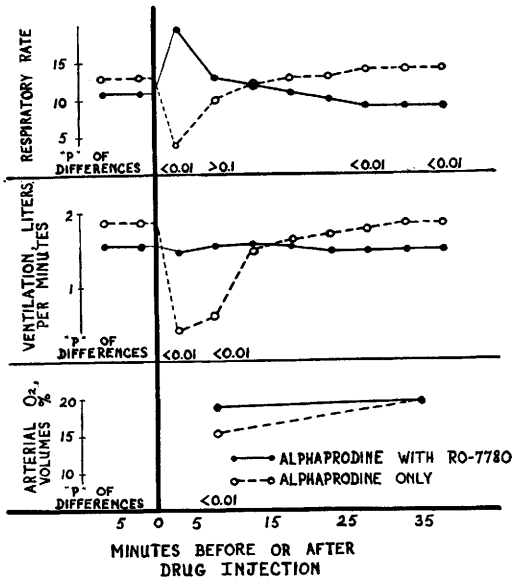

FIG. 1. Mean effects of alphaprodine alone and given simultaneously with Ro-7780 on 9 dogs (18 experiments).

† This and subsequent "p" values were calculated by Student's "t" test.

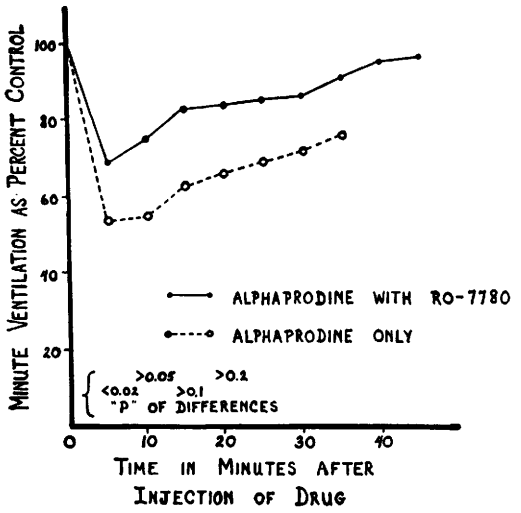

FIG. 2. Mean effects on minute ventilation of alphaprodine alone (4 subjects) and given simultaneously with Ro-7780 (16 subjects).

cf. 0.45 L/min.) and arterial oxygenation (19.8 cf. 15.2 vol. %). Statistical analysis of these data showed that Ro-7780 was effective in controlling the respiratory depression of alphaprodine. Although the figures for arterial oxygenation include the 3 dogs that were artificially ventilated after alphaprodine alone, nevertheless the mean oxygenation was significantly better in the group receiving the combined medication.

The mean effects of alphaprodine alone and combined with Ro-7780 in man are shown in Fig. 2. The use of the combined medication was associated with a significantly higher minute ventilation at the beginning of the test period. Although ventilation was improved, the patients who received antagonist were all either intermittently or constantly asleep during 30 or 40 minutes after the drug was given and had to be awakened for the observations.

Discussion. Since the original studies of Pohl on N-allylnorcodeine(15), several allyl narcotic derivatives have been investigated. These have included N-allylnormorphine (1-4), N-allylnorheroin (cited by Lasagna, 1), 1 - allyl-4-phenyl-4-carbethoxypiperidine (11), and levallorphan(5-12). Ro-7780 is closely related to levallorphan, differing only by having a propargyl instead of an allyl substituent on the nitrogen.

A comparison of the potency of levallorphan and N-allylnormorphine, the two most widely studied antagonists, indicated that the former is at least twice as active as the latter. Costa and Bonnycastle(11) found both capable of preventing respiratory depression in rats given alphaprodine; as to relative potency, ratios of antagonist to alphaprodine of 1 to 20 for levallorphan and 1 to 7.5 for N-allylnormorphine were comparable. Swerdlow, Foldes and Siker(10) studied the combined effect of alphaprodine and levallorphan in humans and found ratios of levallorphan to alphaprodine of 1 to 50 or 1 to 100 adequate in correcting respiratory depression.

In our experiments on dogs a ratio of 1 to 75 or 1 to 100 of Ro-7780 to alphaprodine prevented respiratory depression. These ratios, however, did not completely restore normal respiratory exchange in man. They might have if larger doses of alphaprodine had been used, since antagonists appear more effective in more severely depressed subjects (9). Ro-7780 given either contemporaneously with or after alphaprodine produced an initial increase in ventilation rate and/or volume above control values in spite of the fact that Ro-7780 alone had no significant effect. Such action of opiate antagonists is in keeping with the reports of other workers(2,12).

It appears that Ro-7780 is a more potent opiate antagonist than N-allylnormorphine and at least as potent as levallorphan. Further studies would be necessary comparing the actions of the three agents under identical conditions in order to establish their relative potencies.

Conclusions. (1) 1-3-hydroxy-N-propargyl morphinan has been found a potent antagonist to alphaprodine or morphine in the dog and to alphaprodine in man. (2) Ro-7780 given contemporaneously with alphaprodine at 1/75th or 1/100th the dose of the latter appeared to counteract completely the respiratory depression produced in the dog by the narcotic alone. (3) At the same dose ratios of Ro-7780 to alphaprodine in man, the antagonist diminished the severity of the respiratory depression produced by alphaprodine,

but otherwise the narcotic effect was judged unaltered.

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Further Observations on Distribution of Radioactivity Following Parenteral Administration of Co^{60} Vitamin B_{12}.* (22529)

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It is now well established that following oral or parenteral administration of Co^{60} vit. B_{12} radioactivity may be detected over the liver, spleen and left kidney for periods up to 315 days(1-4). It has been further demonstrated that following the intramuscular injection of $Co^{60}Cl_2$ the pattern is entirely different, suggesting that uptake studies with the radiovitamin probably represent the B_{12} molecule in the organs surveyed(4).

The present report is concerned with further investigations on the distribution of Co^{60} vit. B_{12} under a variety of conditions: (1) following intravenous injection of the radiovitamin; (2) in association with large doses of "cold" B_{12} before, simultaneous with, or after intramuscular injection of the radiovitamin; and (3) administration of amethopterin prior to intramuscular injection of the radiovitamin.

Material and methods. The Co^{60} vit. B_{12} had a specific activity of 0.750 $\mu c/\mu g$. Each dose was diluted with sterile distilled water so that one ml contained one μg of radiovitamin. All administered doses were one ml in volume. Intramuscular injections were given in the right deltoid muscle, using a 1-inch 22 gauge needle. "Cold" vit. B_{12} was administered in the deltoid muscle of the opposite arm. A well-shielded sodium iodide crystal scintillation counter having an aperture of 2 cm was placed over (1) the site of injection (right deltoid muscle), (2) precordium, (3) lower right chest (liver), (4) lower left chest (spleen), (5) left lumbar region (left kidney), (6) lower end of sacrum, (7) lateral aspect of thigh. Radioactivity was recorded for 5-minute periods over each site within ½, 1, 2, 3, 4, 8, 12 and 24 hours after injection. Thereafter, measurements were made at 2, 3, 5 and 7-day intervals up to 168 days. Background counts were subtracted from observed values. No correction was made for Co^{60} decay. All patients were inmates of a chronic disease hospital and received essentially the same diets. Their ages varied from 43 to 80 years. The diagnoses included hypertensive heart disease (1), bronchial asth-

* Co^{60} vit. B_{12} and "cold" B_{12} were supplied by Merck and Co., Rahway, N. J. Amethopterin was supplied by Lederle Laboratories, American Cyanamid Co., Pearl River, N. Y.