

Antinociceptive Interaction Between Opioids and Medetomidine: Systemic Additivity and Spinal Synergy

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The antinociceptive interaction on the tail flick (TF) and hot plate (HP) tests between opioid analgesics and medetomidine after intravenous (iv) or intrathecal administration were examined by isobolographic analysis. Male Sprague-Dawley rats received fixed ratios of medetomidine to morphine, fentanyl, and meperidine of 1:10 and 1:30, 10:1, and 1:3, respectively, by iv administration or 10:1, 3:1 and 10:1, and 1:3 by intrathecal administration, respectively. Data were expressed as the percentage maximal possible effect (%MPE). The A_{50} (dose producing 50% MPE) for each drug or drug combination was determined from the dose-response curve. Isobolographic analysis revealed that the effect of medetomidine combined with fentanyl, morphine, or meperidine was additive after iv administration. The intrathecal administration of combinations of medetomidine with the opioids produced a synergistic antinociceptive effect in the TF but not HP test. These data confirmed that the interaction between medetomidine and opioids in producing antinociception may be additive or synergistic, depending on the route of administration, drug ratio administered, and level of processing of the nociceptive input (*i.e.*, spinal vs. supraspinal). Moreover, these results were consistent with a spinal role for alpha-2 adrenoceptors in mediating antinociception. The authors suggest that the interaction between the opioid and alpha-2 adrenergic receptors occurs within the spinal cord. (Key words: Analgesics, opioid: fentanyl; meperidine; morphine. Drug interactions: isobolographic analysis. Sympathetic nervous system, alpha-2 agonists: medetomidine.)

NUMEROUS STUDIES have demonstrated that clonidine promoted analgesia and anesthesia in the clinical setting.¹⁻⁵ Ghignone *et al.*¹ showed that clonidine promoted hemodynamic stability and reduced the need for fentanyl during induction. In other studies, the requirement for isoflurane² or fentanyl³ in the maintenance of anesthetic depth was reduced by clonidine.

In addition to a direct action on the spinal cord, opioid analgesics modulate responses to nociceptive stimuli by also activating descending serotonergic and noradrenergic inhibitory pathways coursing from the midbrain periaqueductal gray and through ventral medullary sites and descending along the dorsolateral funiculus.⁶ These spi-

notal pathways constitute a significant part of noradrenergic involvement in the expression of opioid-induced antinociception,⁷ which is expressed by spinal alpha-2 adrenoceptors.^{8,9} Opioid-produced antinociception results in part from the activation by descending noradrenergic neurons¹⁰ of spinal alpha-2 adrenergic receptors regulating the responses of primary afferent neurons to nociceptive stimuli.¹¹ Morphine-produced antinociception was augmented by the spinal administration of noradrenergic agonists and attenuated by spinally administered alpha-2 adrenergic antagonists, whereas the antinociceptive effect of clonidine was not attenuated by naloxone.¹² It has been suggested that the spinal alpha-2 adrenoceptors act "downstream" from the opioid receptor.¹³

Intrathecal administration of clonidine produced antinociception in mice,¹⁴ rats,^{14,15} and sheep¹⁶ and was used in humans for treatment of postoperative⁴ and cancer⁵ pain.

Despite its utility in augmenting anesthesia and analgesia, the advantage of clonidine may be limited by the fact that it is considered a partial agonist with action at the alpha-1-adrenergic receptor at higher doses.¹⁷ Medetomidine is a newer alpha-2 adrenergic agonist selective for the alpha-2 receptor and acts as a full agonist.¹⁸ Like clonidine, medetomidine possesses antinociceptive activity and also greatly reduces the halothane MAC in dogs.¹⁸ The effect of medetomidine appears to be stereospecific because the d-enantiomer of medetomidine (dexmedetomidine) is the active component of the racemic mixture.^{18,19}

The antinociceptive interaction between different opioid analgesic agents and medetomidine has not been rigorously studied. Because of the apparent superiority of medetomidine over clonidine, we examined the effect of combinations of medetomidine with fentanyl, morphine, and meperidine administered systemically (intravenously) and intrathecally to determine whether the antinociceptive interaction is additive or truly synergistic and to examine differences in the nature of this interaction after systemic and spinal routes of administration.

Materials and Methods

The protocols used for experiments described in this article were approved by our Animal Care and Use Com-

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mittee. Rats were randomly assigned to treatment groups. Blinding of observers to the treatments given was not part of the protocols.

ANALGESIOMETRIC TESTS

Tail Flick Test

The tail flick (TF) test of D'Amour and Smith²⁰ was performed by placing the tail of male Sprague-Dawley rats (175–225 g) under a focused radiant heat source and over a photocell. The light was activated with a timer, and, when the animal flicked its tail aside, the photocell was uncovered, stopping the timer. TF latencies were recorded (to 1/10th of a second) twice before and at several intervals after injection. A cutoff TF latency of either 10 s in intact rats or 7 s in rats with spinal catheters was used to prevent tissue damage.

Hot Plate Test

Hot plate (HP) latencies were determined by placing each rat on an HP kept at $55 \pm 0.5^\circ$ C and observing the occurrence of one of the following nociceptive responses: 1) licking of the paws; 2) stomping of the hind paws; or 3) jumping out of the enclosure.

A timer was started when the animal was placed on the HP surface and stopped when the first nociceptive response was observed. HP latencies were determined twice before and at several time periods after injection. A cutoff HP latency of 30 s was used to prevent tissue damage.

INJECTION PROTOCOLS

Intravenous Injections

Medetomidine, fentanyl, meperidine, and morphine were dissolved in 0.9% (w/v) saline and injected in a volume of 1 ml/kg into the lateral tail vein. The TF and HP latencies were determined at several intervals after injection to establish the time of peak effect. Dose-response curves were determined from the data obtained at the time of peak effect. Injections were timed so that the peak effects of medetomidine and the opioid coincided. Doses were administered to maintain fixed ratios of medetomidine to morphine, fentanyl, and meperidine. The ratios of medetomidine to morphine (1:10 and 1:30), fentanyl (10:1), and meperidine (1:3) were based on earlier studies and represent what was found to be the optimal combination of an alpha-2 agonist (clonidine) with an opioid (unpublished observations).

Intrathecal Injections

Rats were randomized to treatment groups and prepared for intrathecal drug injection according to the method originally described by Yaksh and Rudy.²¹ During

anesthesia with isoflurane, a PE10 catheter was inserted through the atlantooccipital membrane to the level of the lumbar enlargement of the spinal cord. The catheter was exteriorized at the back of the head and plugged and the wound was closed. The rats were allowed to recover for a week before undergoing any testing; those showing any neurologic deficit were excluded from the studies. Drugs were dissolved in 0.9% (w/v) saline and administered in a volume of 5 μ l, and the catheter was flushed with 10 μ l of saline. The drugs were coadministered in the same solution to minimize addition of fluid to the CSF. The HP and TF latencies were measured at several intervals after injection, and dose-response curves were constructed from data gathered at the time of peak effect. The administered ratios of medetomidine to morphine (10:1), fentanyl (3:1 and 10:1), and meperidine (1:3) were derived from earlier studies and represented the optimal ratios found when combinations of clonidine with opioids were examined (unpublished observations).

DATA ANALYSIS

The data obtained were converted to %MPE (maximal percent effect) by the following equation:

$$\%MPE = 100 \times \frac{\text{test latency} - \text{control latency}}{\text{cutoff} - \text{control latency}}$$

The log-dose was plotted *versus* %MPE, and the A_{50} (dose producing 50% MPE) along with confidence limits was calculated by linear regression.

The interactions between the alpha-2 agonist and the opioids were examined by isobolographic analysis. For each drug combination, the dose was presented as total drug injected. For example, a total dose of 11 mg/kg of a 10:1 medetomidine-morphine ratio meant that the animal received 10 mg/kg of medetomidine plus 1 mg/kg of morphine. With the use of the total dose in the dose-response curve, the regression line and the A_{50} with its associated variances were determined. The A_{50} was then resolved into its component parts according to the ratio of drug administered and plotted with medetomidine on the abscissa and opioid on the ordinate. A line of additive interaction was estimated by connecting the A_{50} for the opioid with that of medetomidine. For each ratio of drugs used, a theoretic additive A_{50} existed on the additivity line such that

$$A_{50\text{add}} = A_{50\text{opioid}} / (p_1 + R p_2)$$

where R was the potency ratio of opioid to medetomidine and p_1 was the proportion of opioid in the total dose and p_2 that of medetomidine. The confidence intervals for the opioid and medetomidine components of the theoretic additive A_{50} were obtained from the variances about the A_{50} for each drug administered alone. This theoretic additive point was then compared with the experimentally

TABLE 1. Potency of iv Medetomidine and Opioids Alone and in Combinations in the Tail Flick Test

	A ₅₀ (mg/kg, 95% confidence limits)	Slope	R	n
Fentanyl	0.0031 (0.0017–0.0056)	109	0.969	29
Med:fen 10:1	0.0187 (0.0014–0.0019)	170	0.977	30
Morphine	1.54 (0.91–2.62)	126	0.952	24
Med:mor 1:10	0.201 (0.11–0.36)	84	0.971	24
Med:mor 1:30	0.454 (0.31–0.67)	114	0.944	30
Meperidine	1.66 (1.23–2.22)	97	0.982	30
Med:mep 1:3	0.124 (0.089–0.172)	93	0.990	23
Medetomidine	0.034 (0.016–0.072)	87	0.971	30

Dose-response curves were constructed for fixed ratios of medetomidine and fentanyl, morphine, and meperidine in the rat tail flick test. The drug ratios are identified as medetomidine to opioid. The A₅₀ values, representing the dose producing one half the maximal possible effect in each test, were calculated from the logdose-response curve by linear regression using the opioid dose. The medetomidine dose at each combination may be derived from the ratio (as described in the text). Each dose was administered by iv injection, and the animals were tested at the time of peak effect.

R = regression coefficient of the dose-response curve; n = total animals per dose-response curve, minimum of six per dose.

Fen = fentanyl; mor = morphine; mep = meperidine; med = medetomidine.

derived A₅₀ for the mixture by a *t* test. A significant potency ratio with the experimental A₅₀ significantly less than the theoretic additive A₅₀ indicated a synergistic interaction. The statistical derivation and validation of this method of analysis were described by Tallarida *et al.*²²

Results

INTERACTION OF INTRAVENOUS MEDETOMIDINE AND OPIOIDS

Medetomidine produced a peak effect at 15 min after injection, fentanyl at 1 min, and meperidine and morphine at 5 min after injection. Thus, all testing of combinations of opioids with medetomidine was done 15 min after medetomidine injection, with the opioid injections timed so that the peak effects of the opioid and medetomidine coincided.

All agents tested produced dose-dependent antinociception in both the TF and HP tests. The rank-order of potency in both tests was fentanyl ≫ medetomidine ≫ meperidine ≧ morphine. The A₅₀ doses and the confidence intervals for the TF test were summarized in table 1, and those for the HP test were summarized in table 2. In each set of medetomidine-opioid combinations, the dose-response curves for the combinations fell between the dose-response curve for medetomidine and that for the opioid alone, indicating a positive interaction that could be either additive or synergistic (fig. 1). A similar pattern of interaction was seen after intravenous administration in the HP test.

ISOBOLOGRAPHIC ANALYSIS OF INTRAVENOUS MEDETOMIDINE-OPIOID INTERACTIONS

The isobolographic analysis of the medetomidine-opioid combinations in the TF test indicated that experimentally derived A₅₀ values were not significantly different (*P* > 0.05) from the theoretic additive A₅₀ values; thus, the 50% effective doses for each combination of opioid with medetomidine fell along the additive line, indicating an additive interaction between the opioids and medetomidine.

Likewise in the HP test, the experimentally derived A₅₀ values for each drug combination did not differ significantly (*P* > 0.05) from the theoretic additive A₅₀ values, indicating an additive effect between each of the opioids and medetomidine.

Isobols were constructed for several levels of effect—25%, 50%, 75%, and 100% MPE—to determine the nature of the interaction throughout the dose-response curve. For each level of effect, the morphine-medetomidine isobols (fig. 2a) did not deviate from linearity, indicating that the antinociceptive interaction between medetomidine and morphine was additive throughout the dose-response curve. Likewise for medetomidine-fentanyl (fig. 2b) and medetomidine-meperidine (fig. 2c), the interaction between the alpha-2 agonist and opioid agonist was additive. Similar results were seen with the HP test.

ANTINOCICEPTIVE EFFECTS OF INTRATHECAL MEDETOMIDINE AND OPIOIDS

Medetomidine, morphine, fentanyl, and meperidine produced dose-dependent antinociception in the TF (fig.

TABLE 2. Potency of iv Medetomidine and Opioids Alone and in Combinations in the Hot Plate Test

	A ₅₀ (mg/kg, 95% Con. lim)	Slope	R	n
Fentanyl	0.0072 (0.0029–0.018)	135	0.883	30
Med:fen 10:1	0.0297 (0.0176–0.0506)	171	0.947	24
Morphine	4.73 (2.98–7.50)	190	0.996	24
Med:mor 1:10	0.462 (0.057–3.76)	163	0.935	30
Med:mor 1:30	0.748 (0.51–1.10)	168	0.973	24
Meperidine	3.23 (1.91–5.44)	148	0.949	29
Med:mep 1:3	0.239 (0.20–0.28)	124	0.996	23
Medetomidine	0.042 (0.019–0.091)	104	0.948	30

Dose-response curves were constructed for fixed ratios of medetomidine and fentanyl, morphine, and meperidine and in the rat hot plate test. The drug ratios are identified as medetomidine to opioid. The A₅₀ values, representing the dose producing one half the maximal possible effect in each test, were calculated from the logdose-response curve by linear regression using the opioid dose. The medetomidine dose at each combination may be derived from the ratio (as described in the text). Each dose was administered by iv injection and the animals were tested at the time of peak effect.

R = regression coefficient of the dose-response curve; n = total animals per dose-response curve, minimum of six per dose.

Fen = fentanyl; mor = morphine; mep = meperidine; med = medetomidine.

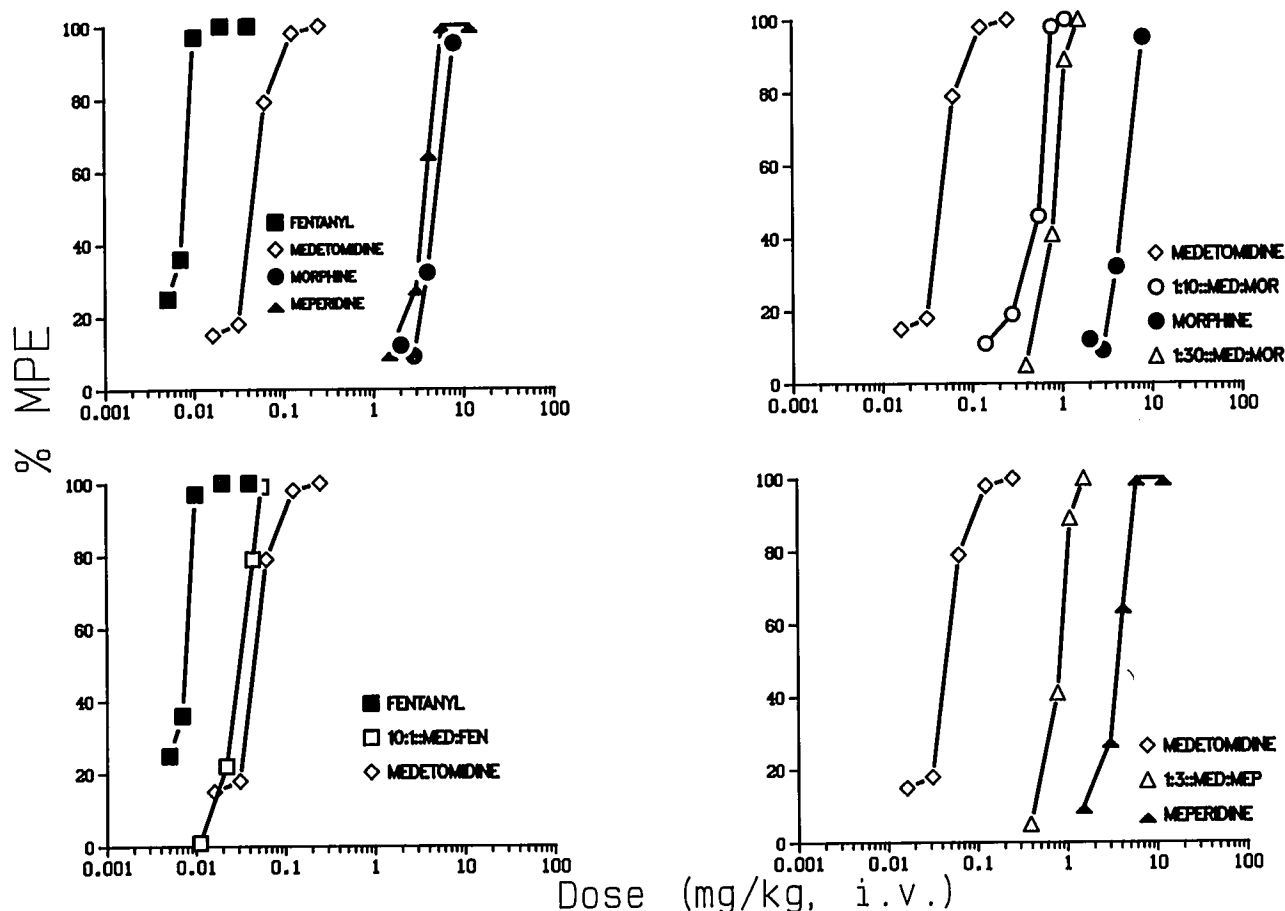


FIG. 1. Dose-response curves (DRC) for the opioids, medetomidine, and combinations of opioid and medetomidine in the tail flick test after iv administration, with response presented as % MPE. (Top left) The DRCs for each agonist administered alone. (Top right) The DRC for morphine alone and in the presence of ratios of medetomidine to morphine of 1:10 to 1:30. (Bottom left) The DRC for fentanyl administered alone and in a constant ratio of medetomidine to fentanyl of 10:1. (Bottom right) The DRC for meperidine administered alone and in the presence of a constant ratio of medetomidine to meperidine of 1:3. $N = 6$ animals per dose; error bars were omitted for the sake of clarity.

3) and HP tests after intrathecal administration. The A_{50} values for each dose-response curve were summarized for the TF (table 3) and HP (table 4) tests. The rank-order potency was fentanyl = morphine > medetomidine >> meperidine in both of the tests.

The dose-response curve for each medetomidine-opioid combination fell between those for medetomidine or the respective opioid alone in the TF (fig. 3) and HP tests (data not shown), indicating a positive interaction between medetomidine and the opioid.

ISOBOLOGRAPHIC ANALYSIS OF INTRATHECAL MEDETOMIDINE-OPIOID INTERACTIONS

The isobols at the 50% effect for the medetomidine-morphine and the medetomidine-meperidine combinations in the TF test deviated below the additive line. The experimentally derived A_{50} s for the combinations of medetomidine with morphine or with meperidine were

significantly ($P \leq 0.05$) less than their theoretic additive A_{50} , clearly indicating a synergistic effect. The isobols maintained a nonlinear shape throughout the dose-response curve, suggesting synergy at each level of effect examined (fig. 4). The experimentally derived A_{50} for the 10:1 ratio of medetomidine to fentanyl was not significantly different from the theoretic additive A_{50} for that ratio, indicating an additive interaction. However, the actual A_{50} for the 3:1 mixture of medetomidine with fentanyl was significantly ($P \leq 0.05$) less than that predicted for an additive interaction, demonstrating synergy between medetomidine and fentanyl at that ratio. As with the interaction with morphine and meperidine, the isobols for the 3:1 ratio of medetomidine and fentanyl were nonlinear throughout each level of effect examined.

The antinociceptive interaction between medetomidine and morphine or fentanyl in the HP test was not synergistic because the experimentally derived A_{50} values did not differ significantly from the theoretic additive A_{50}

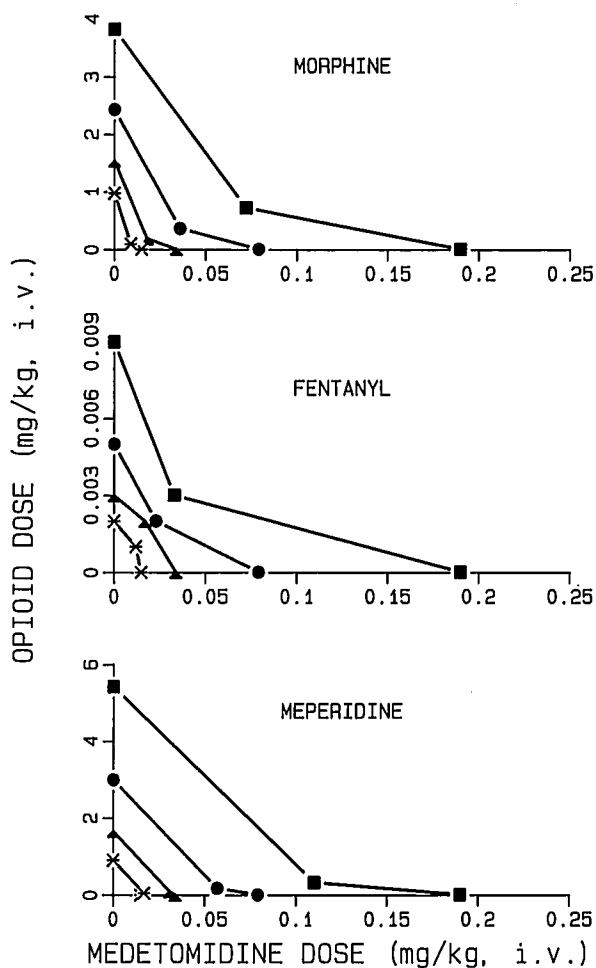


FIG. 2. Isobolograms for the 25 (*), 50 (▲), 75 (●), and 100% (■) effective doses for medetomidine plotted against morphine (*top*), fentanyl, (*middle*), and meperidine (*bottom*) after iv injection in the rat tail flick test. The isobol points are shown for ratios of medetomidine to morphine (1:10), fentanyl (10:1), and meperidine (1:3).

values for each respective drug combination. Because we could not determine an A_{50} dose for meperidine in the HP test after intrathecal administration, it was not possible to calculate a theoretic additive A_{50} value for this drug combination; however, a synergistic effect was inferred because an A_{50} of 67 μg was calculated when a ratio of 1:3 of medetomidine to meperidine was injected (table 4).

Discussion

The fact that observers were not blinded to the treatments given the rats may be viewed as a limitation placed on the interpretation of the results. However, preliminary experiments showed that blindedness of the observers did not significantly affect the outcome of the experiments,

at least for the analgesimetric procedures described herein (unpublished observations). The opioid analgesics and medetomidine all produced dose-dependent antinociception. Moreover, the dose-response curves for medetomidine and the opioids were all parallel in the TF and HP tests after intravenous or intrathecal administration. Meperidine was equipotent with morphine after intravenous administration in the TF and HP tests, which does not reflect relative clinical potency of the two opioids. This unexpected result may be an artifact of the experimental conditions, because the tests used and methods of analysis used may influence drug potencies.²³ The rest of the data do reflect the expected clinical rank-order of potency.

The dose-response curves for the mixtures of medetomidine and opioid fell between those for each drug alone, indicating either additive or synergistic interactions. Synergy was observed only after intrathecal administration and only in the TF test, which is a spinal reflex modulated at the spinal level. The results of this study showed that opioid and alpha-2 adrenergic receptor-mediated mechanisms may produce antinociception by acting through a final common pathway and strongly suggest a spinal site of interaction between the opioid and alpha-2 adrenergic receptor. A spinal site of action for alpha-2-adrenoceptor-mediated antinociception was suggested because spinal transection of mice reduced the antinociceptive potency of morphine but not of clonidine²⁴ and that ablation of descending noradrenergic neurons resulted in a spinal supersensitivity to the antinociceptive effects of clonidine.²⁵ Thus, it is likely that the site of interaction between opioid and alpha-2 adrenoceptors with regard to the modulation of nociception is in the spinal cord.

Opioid and alpha-2 receptors share a common mechanism: both elicit a hyperpolarization of neurons by altering K^+ channel conductance by means of G-protein-coupled receptor mechanisms, resulting in an inhibition of neurotransmitter release.²⁶ Like morphine, medetomidine produces membrane hyperpolarization through a pertussis-sensitive G protein.²⁷ Opioid and alpha-2 adrenergic receptors are present in the same superficial layers of the spinal cord and both inhibit C-fiber activity.²⁸ Should these receptors reside on the same spinal neuron, then it is possible that an allosteric interaction could occur. Alternatively, activation of the alpha-2 adrenergic and the opioid receptors may elicit an enhanced effect by independently altering intracellular mechanisms coupled to G-protein activation, yielding a net effect greater than the sum of each independent effect. These suggested mechanisms do not take into consideration other factors that may inhibit neurotransmitter release (*e.g.*, Ca^{++} influx); clearly, additional studies into the subcellular mechanisms of alpha-2 adrenergic and opioid interactions

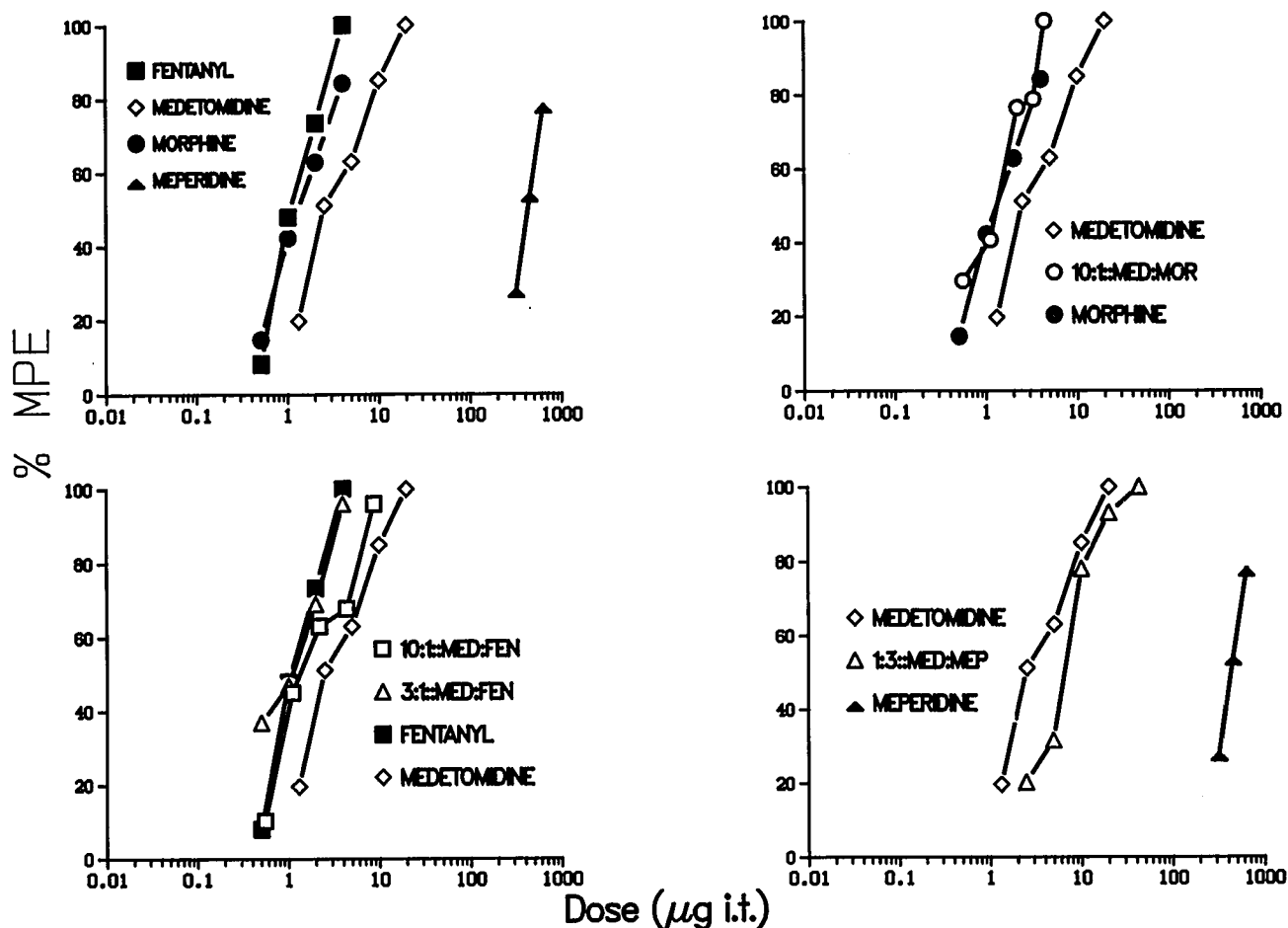


FIG. 3. Dose-response curves (DRC) for the opioids, medetomidine, and combinations of opioid and medetomidine in the tail flick test after i.t. administration, with response presented as % MPE. (Top left) The DRCs for each agonist administered alone. (Top right) The DRC for morphine alone and in the presence of ratios of medetomidine to morphine of 10:1. (Bottom left) The DRC for fentanyl administered alone and in a constant ratio of medetomidine to fentanyl of 3:1 and 10:1. (Bottom right) The DRC for meperidine administered alone and in the presence of a constant ratio of medetomidine to meperidine of 1:3. $N =$ minimum of 5 rats per dose; error bars were omitted for the sake of clarity.

with regard to neurotransmitter release and antinociception must be performed.

Several studies suggested a synergistic interaction between α -2 and opioid agonists to produce antinociception. Electrophysiologic studies in the cat¹¹ and rat^{28,29} demonstrated a spinal synergy between clonidine and morphine in the inhibition of the neuronal activity of neurons responsive to nociception. Synergy between systemic morphine and clonidine³⁰ and between systemic³¹ or intrathecal¹⁴ morphine and intrathecal clonidine has been described. In these studies, however, either a single dose of each drug was administered and the result was greater than the sum of effects produced by each dose alone or a fixed dose of one drug caused a significant shift to the left in the dose-response curve of the other drug. Moreover, combining a fixed dose of a drug with varying doses of another changed the drug ratio at each point of

the dose-response curve and may not reflect the true nature of the interaction. Although results from these and similarly conducted studies may reveal positive (*i.e.*, additive or synergistic) interactions between two drugs or treatments, the analysis of the interactions was not very rigorous.

The isobolographic analysis of dose-response curves of fixed ratios of drugs provided a more rigorous means of examination of drug-drug interactions. Gessner and Cabana³² showed that the ratio of drugs used could influence whether a synergistic or additive effect is produced. This point was well illustrated by our own observation that the 10:1 medetomidine to fentanyl ratio, representing a relative high α -2 to opioid ratio, produced an additive effect after intrathecal administration, whereas the 3:1 medetomidine to fentanyl ratio produced a synergistic interaction.

TABLE 3. Potency of it Medetomidine and Opioids Alone and in Combinations in the Tail Flick Test

	A ₅₀ (μg, 95% Con. lim.)	Slope	R	n
Fentanyl	1.192 (0.91–1.56)	100	0.994	22
Med:fen 3:1	0.93 (0.56–1.55)	66	0.985	15
Med:fen 10:1	1.74 (1.10–2.75)	65	0.971	25
Morphine	1.37 (1.16–1.62)	76	0.997	30
Med:mor 10:1	1.14 (0.814–1.65)	78	0.978	25
Meperidine	432 (400–467)	166	1.000	22
Med:mep 1:3	6.33 (3.60–11.2)	73	0.962	25
Medetomidine	3.06 (2.26–4.14)	65	0.989	31

Dose-response curves were constructed for fixed ratios of medetomidine and fentanyl, morphine, and meperidine in the rat tail flick test. The drug ratios are identified as medetomidine to opioid. The A₅₀ values, representing the dose producing one half the maximal possible effect in each test, were calculated from the logdose-response curve by linear regression using the opioid dose. The medetomidine dose at each combination may be derived from the ratio (as described in the text). Each dose was administered by iv injection and the animals were tested at the time of peak effect.

R = regression coefficient of the dose-response curve; n = total animals per dose-response curve, minimum of five per dose.

Fen = fentanyl; mor = morphine; mep = meperidine; med = medetomidine.

The isobolographic analysis of drug interactions was described by Loewe^{33,34} and later by Gessner.³⁵ Others³⁶ showed that spinal plus supraspinal morphine was synergistic because the isobols were clearly nonlinear and concave. Roerig and Fujimoto³⁷ used isobols to show that intracerebroventricular and intrathecal morphine were synergistic in the mouse, defining synergy as when the ED₅₀ points and 95% confidence limits fell below the line of additivity. Tallarida *et al.*²² recently provided a statis-

TABLE 4. Potency of it Medetomidine and Opioids Alone and in Combinations in the Hot Plate Test

	A ₅₀ (μg, 95% confidence limits)	Slope	R	n
Fentanyl	1.59 (0.98–2.57)	143	0.959	20
Med:fen 3:1	3.60 (3.5–3.7)	144	1.0	15
Med:fen 10:1	5.88 (4.84–7.15)	308	0.920	33
Morphine	0.72 (0.39–1.34)	67	0.951	25
Med:mor 10:1	3.86 (2.86–5.28)	155	0.982	21
Meperidine	46% MPE at max dose of 800 μg			22
Med:mep 1:3	26.8 (19.6–36.6)	116	0.992	20
Medetomidine	13.7 (7.34–25.6)	83	0.967	21

Dose-response curves were constructed for fixed ratios of medetomidine and fentanyl, morphine, and meperidine in the rat hot plate test. The drug ratios are identified as medetomidine to opioid. The A₅₀ values, representing the dose producing one half the maximal possible effect in each test, were calculated from the logdose-response curve by linear regression using the opioid dose. The medetomidine dose at each combination may be derived from the ratio (as described in the text). Each dose was administered by iv injection and the animals were tested at the time of peak effect. R = regression coefficient of the dose-response curve; n = total animals per dose-response curve, minimum of five per dose.

Fen = fentanyl; mor = morphine; mep = meperidine; med = medetomidine.

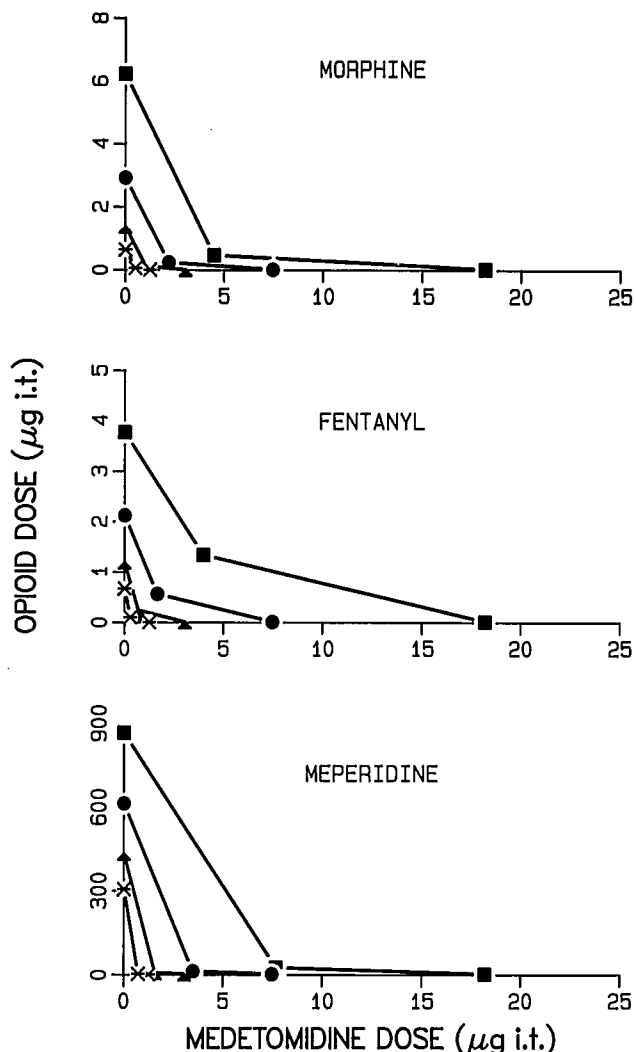


FIG. 4. Isobolograms for the 25 (*), 50 (▲), 75 (●), and 100% (■) effective doses for medetomidine plotted against morphine (*top*), fentanyl (*middle*), and meperidine (*bottom*) after it injection in the rat tail flick test. The isobol points are shown for ratios of medetomidine to morphine (10:1), fentanyl (3:1), and meperidine (1:3).

tical interpretation of isobolographic data in which the experimental ED₅₀ of a mixture could be compared with a theoretic additive ED₅₀. The advantages of this method include statistically derived values independent of graphic analysis and statistical validity without the need to establish parallelism of the dose-response curves.

In our study, an additive effect was seen after intravenous administration and in the supraspinally integrated HP response after intrathecal administration, whereas a synergistic interaction occurred in the spinal reflexive TF test after intrathecal administration. These results were consistent with the view that the site of interaction between opioids and alpha-2 adrenergic agonists was at the spinal level.^{14,28}

The absence of synergism after intravenous administration might be explained by the observation that spinal and supraspinal morphine were synergistic.³⁶ Systemically administered morphine acts spinally and supraspinally and activates descending inhibitory projections, many of which are noradrenergic and can act on spinal alpha-2 adrenoceptors.^{10,38} Consequently, the spinal alpha-2 sites were already activated and the further addition of medetomidine would fail to produce any additional effect. Pharmacokinetic factors should also be considered. After intravenous administration, the drug was distributed throughout the body, and, because of different metabolic pathways, redistribution to various compartments, and bioavailability to the central nervous system (CNS), the ultimate drug ratio actually attained in the CNS may not be the same as that injected systemically. In contrast, intrathecal administration placed the drugs in close proximity to the spinal site of action.

Medetomidine (either as the d-enantiomer dexmedetomidine or as the racemic mixture) was of interest because its analgesic and anesthetic, MAC-reducing, and thiopental-sparing properties were reported to be mediated by central alpha-2 adrenoceptors.^{39,40-42} Dexmedetomidine blocked spinal neuronal responses to painful stimuli.⁴³ An antinociceptive role for intrathecal or intravenous medetomidine was also supported by our data. The antinociceptive effect of medetomidine may also contribute to its MAC-reducing activity, because MAC measurement depends on the observation of a response to a nociceptive stimulus.

The synergistic interaction between opioids and medetomidine results may have therapeutic significance by decreasing the dose requirements of either drug to achieve an acceptable level of analgesia. Opioids and alpha-2 adrenergic agonists share analgesia as a common property, whereas the primary untoward effect of opioids is respiratory depression and that of alpha-2 agonists is decreased cardiovascular function.³ Because medetomidine did not cause respiratory depression,⁴⁴ and opioids did not significantly depress cardiovascular function at analgesic doses, then the combined effect of an opioid and alpha-2 adrenergic agonist should be analgesia with reduced hemodynamic and respiratory effects. The addition of clonidine to the anesthetic regimen improved hemodynamic stability.^{1,2} Combinations of fentanyl and clonidine³ produced similar analgesic intensity but less respiratory depression than did fentanyl alone. Dexmedetomidine in combination with alfentanil did not produce clinically significant depression of hemodynamic or respiratory function associated with alfentanil.⁴⁵

The data presented in this article clearly indicate that a positive interaction existed between the alpha-2 agonist medetomidine and opioid agonists. Moreover, this interaction was additive after systemic administration and syn-

ergistic when confined to the spinal level of antinociceptive action (*i.e.*, spinal administration and spinal reflex). This result was similar to observations made with clonidine and opioids (unpublished observations) and further supported the belief that the site of interaction between alpha-2 adrenergic and opioid receptors with regard to antinociception was at the spinal level. The therapeutic advantages of combinations of opioids and medetomidine are envisioned as increased analgesia with reduced respiratory and cardiovascular involvement.

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