Effect of amantadine on oxymorphone-induced thermal antinociception in cats

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This study examined the effect of amantadine, an N-methyl-D-aspartate receptor antagonist, on the thermal antinociceptive effect of oxymorphone in cats. Six adult healthy cats were used. After baseline thermal threshold determinations, oxymorphone was administered intravenously to maintain plasma oxymorphone concentrations of 10, 20, 50, 100, 200, and 400 ng/mL. In addition, amantadine, or an equivalent volume of saline, was administered intravenously to maintain a plasma amantadine concentration of 1100 ng/mL. Thermal threshold and plasma oxymorphone and amantadine concentrations were determined at each target plasma oxymorphone concentration. Effect maximum models were fitted to the oxymorphone concentration–thermal threshold data, after transformation in % maximum response. Oxymorphone increased skin temperature, thermal threshold, and thermal excursion (i.e., the difference between thermal threshold and skin temperature) in a concentration-dependent manner. No significant difference was found between the amantadine and saline treatments. Mean ± SE oxymorphone EC_{50} were 14.2 ± 1.2 and 24.2 ± 7.4 ng/mL in the amantadine and saline groups, respectively. These values were not significantly different. Large differences in oxymorphone EC_{50} in the saline and amantadine treatment groups were observed in two cats. These results suggest that amantadine may decrease the antinociceptive dose of oxymorphone in some, but not all, cats.

INTRODUCTION

Pain in cats has been reported to be largely undertreated (Dohoo & Dohoo, 1996; Capner *et al.*, 1999; Hugonnard *et al.*, 2004; Williams *et al.*, 2005). While opioids are commonly considered the first line of treatment for surgical pain in many species (Pascoe, 2000), they are not as widely used in cats (Dohoo & Dohoo, 1996; Capner *et al.*, 1999; Hugonnard *et al.*, 2004; Williams *et al.*, 2005). This may be because of, in part, the fact that, in cats, clinical doses of opioids may produce dysphoria (Pascoe, 2000; Robertson & Taylor, 2004). In addition, opioids also cause hyperthermia and sympathetic nervous system stimulation, particularly when high doses are used (Pascoe *et al.*, 1997; Niedfeldt & Robertson, 2006; Posner *et al.*, 2007,

2010). Nevertheless, because opioids have high efficacy and low toxicity (Pascoe, 2000), their use in cats should be encouraged.

N-methyl-D-aspartate receptor (NMDAR) antagonists have been used as an adjunct therapy to opioids and have been shown to reduce the dose of the opioid required to produce analgesia (Fischer *et al.*, 2005). Amantadine is an anti-viral agent used in the treatment of influenza A infections (Van Voris *et al.*, 1981; Dolin *et al.*, 1982). It has also been shown to antagonize NMDAR and is used to treat chronic pain (Hewitt, 2000). It has been reported to reduce the therapeutic dose of opioids in mice and rats (Fischer *et al.*, 2005; Snijdelaar *et al.*, 2005) and has anecdotally been combined with opioids in dogs and cats to improve analgesia (Lamont, 2008; Robertson, 2008). Amantadine has also been combined with nonsteroidal anti-inflamma-

tory drugs to alleviate refractory osteoarthritic pain in dogs (Lascelles *et al.*, 2008). Amantadine may therefore have the potential of reducing the analgesic dose of opioids in cats, and thereby of reducing the undesirable effects of these drugs.

The aim of this study was to evaluate the effect of amantadine on oxymorphone-induced antinociception in cats and to examine the relationship between plasma oxymorphone concentration, with or without amantadine, and antinociceptive effect. We hypothesized that administration of amantadine would decrease the antinociceptive dose of oxymorphone.

MATERIALS AND METHODS

Animals

Six healthy adult female spayed domestic shorthair cats weighing 4.2 ± 0.7 kg (mean ± SD) were used in the study. The pharmacokinetics of oxymorphone and amantadine had been determined in each cat in previous studies (Siao *et al.*, inpress a,b). Each cat was studied twice with a minimum interval of 2 weeks between successive experiments. This study was approved by the institutional animal care and use committee at the University of California, Davis. Cats were observed for behavioral changes and other visible drug effects during the study period.

Instrumentation and drug administration

The day before an experiment, cats were anesthetized with isoflurane in oxygen delivered in an acrylic chamber. After anesthesia was induced, the trachea was intubated and anesthesia was maintained with isoflurane in oxygen delivered via a coaxial Mapleson F circuit. A 22-gauge, 10-cm catheter was aseptically placed in a jugular vein. The catheter was capped with an infusion plug and sutured to the skin. A light bandage was then placed over the catheter insertion site. The lateral aspect of the thorax (between the caudal aspect of the scapula and the last rib, and from the sternum to the spine) was clipped, and cats were allowed to recover from anesthesia.

On the day of an experiment, a 22-gauge, 2.5-cm catheter was placed in a medial saphenous vein, and a light bandage was placed over the catheter insertion site. Oxymorphone (Opana; Endo Pharmaceuticals, Chadds Ford, PA, USA) was administered intravenously via the medial saphenous catheter using a target-controlled infusion system (RUGLOOP I; Demed, Temse, Belgium) and a syringe pump (Harvard PHD 22/2000; Harvard Apparatus, Holliston, MA, USA). With this system, the central compartment was rapidly loaded to the desired concentration. The infusion rate was then updated every 10 sec, as needed to maintain pseudo–steady-state plasma concentration, according to the following equation:

$$R = C_T \times V_1(k_{10} + k_{12}e^{-k_{21}t} + k_{13}e^{-k_{31}t})$$

where R is the infusion rate; C_T is the target plasma concentration; V_1 is the volume of the central compartment; t is the time; and k_{10}, k_{12}, k_{21}, k_{13}, and k_{31} are the microrate constants.

Individual pharmacokinetic data were used. Oxymorphone target plasma concentrations were 0, 10, 20, 50, 100, 200, and 400 ng/mL. Oxymorphone target concentrations were administered in an ascending order to decrease experimental time. In addition, amantadine (Spectrum Chemicals, Gardena, CA, USA), or an equivalent volume of saline, was administered intravenously in the medial saphenous catheter, using the same target-controlled infusion system to maintain a plasma concentration of 1100 ng/mL. Amantadine was dissolved in isotonic saline to a concentration of 5 mg/mL, immediately before administration. The solution was filtered through a 0.2-μm filter. Individual pharmacokinetic data were used. The order of treatments (amantadine vs. saline) was randomly selected.

Thermal threshold determination

Each cat was placed in an individual cage ($80 \times 80 \times 65$ cm) that had mirrors on each sidewall and a transparent acrylic door. Cats were acclimated to the cage and placement of the thermal threshold probe prior to the beginning of the study. These acclimation sessions included placing an elastic band and pressure cuff similar to those used during the study on the lateral aspect of the thorax. The cat remained in the cage used for the study for several hours with intermittent interaction with an investigator.

The thermal threshold system used in this study was identical to the system previously developed and validated for use in cats and used in various studies of the effects of analgesic drugs in cats (Dixon *et al.*, 2002; Robertson *et al.*, 2003; Lascelles & Robertson, 2004; Pypendop *et al.*, 2006, 2008; Steagall *et al.*, 2008). A probe containing a heating element and adjacent temperature sensor, both embedded in epoxy, was attached to a pressure cuff and held in position over the lateral aspect of the thorax by an elastic band. The pressure cuff was inflated to 100 mmHg to ensure proper contact between the probe and skin. The cuff bladder was inspected before each measurement and re-inflated if necessary. Before each measurement, the probe was connected to a control unit by a flexible cable. Each cat was allowed to move freely in the cage and had free access to food and water during the experiments.

Thermal probes were calibrated weekly during the study. For calibration, the probe was securely attached on top of a $9.0 \times 9.0 \times 0.5$ cm aluminum plate. A thermocouple was placed in a previously drilled horizontal hole so that the tip was directly below the probe and was connected to a digital thermometer. The accuracy of this digital thermometer had been checked against a certified thermometer. The aluminum block was placed on a standard laboratory hotplate that was heated to approximately 85 °C and then allowed to cool to 30 °C. As the hotplate temperature changed from 65 to 30 °C, measurements from the probe and thermocouple were recorded at decrements of 5.0 °C. The probe response was linear within that range [linear coefficient of determination (R^2) > 0.998]. A calibration curve was constructed by use of linear regression, and temperatures recorded during the experiments were mathematically corrected by use of the most recent curve for that probe.

For thermal threshold determination, skin temperature was measured, and the heater was activated (rate of temperature

increase, 0.6 °C/sec). The cat was observed until a reaction (e.g., jumping, turning the head toward the probe, or licking and biting the probe area or cable) was detected or a maximal temperature (55 °C) was reached. When a reaction was observed before the cutoff temperature was reached, the temperature was recorded and considered the thermal threshold, and the heater was turned off. Thermal thresholds were always determined by the same investigator (KTS), who was unaware of treatment (amantadine or saline) assignment. At least 30 min were allowed after probe placement for equilibration between probe and skin temperatures. Baseline skin temperature and baseline thermal threshold values were then determined in duplicate at 20-min intervals. Ten minutes after determination of baseline values, amantadine or saline administration was started. Oxymorphone was administered to reach and maintain the first target plasma concentration. Drugs were infused for 25 min before skin temperature and thermal threshold were determined in duplicate, allowing twenty minutes between each determination, with the drug infusions ongoing. After duplicate determinations, the target plasma oxymorphone concentration was increased to the next value, 25 min were allowed, measurements were taken as described above, and the sequence was repeated until the last target plasma oxymorphone concentration had been administered.

Blood sample collection

A blood sample (2 mL) was collected from the jugular catheter prior to oxymorphone and amantadine or saline administration and at each target plasma oxymorphone concentration, 10 min after the first of the duplicate thermal threshold determinations. At least 5 mL of blood was scavenged prior to collection of the sample. Scavenged blood was injected back through the catheter after sample collection, and the catheter was flushed with 2 mL of physiologic saline solution containing heparin (1 U of heparin/mL). Blood samples were transferred to tubes containing ethylenediaminetetraacetic acid, immediately placed on ice, and centrifuged at 3901 g for 10 min at 4 °C within 10 min of collection; the plasma was separated and stored at −20 °C until analyzed for oxymorphone and amantadine concentrations.

Drug analysis

Oxymorphone and amantadine were quantitated in feline plasma by liquid chromatography-mass spectrometry (LC-MS) analysis of protein-precipitated samples. The calibration standards were prepared as follows: stock solutions were prepared by dissolving 10.0 mg of oxymorphone (Toronto Research Chemicals, North York, ON, Canada) and amantadine (Sigma-Aldrich Co, St Louis, MO, USA) standards in 10.0 mL of methanol. Working solutions were prepared by dilution of the oxymorphone and amantadine stock solution with methanol to concentrations of 1000, 100, and 1.0 ng/mL. Plasma calibrators were prepared by dilution of the working oxymorphone and amantadine solutions with feline drug-free plasma to concen-

trations of 2.5, 5.0, 10, 50, 100, 150, 250, 500, 1000, 2000, 3000 and 4000 ng/mL. Calibration curves and negative control samples were prepared freshly for each quantitative assay. In addition, quality control samples (plasma fortified with analytes at concentrations midpoint of the standard curve) were routinely included as an additional check of accuracy. The concentration of oxymorphone and amantadine in each sample was determined by the internal standard (oxymorphone-D3) (Toronto Research Chemicals) method using the peak area ratio and linear regression analysis.

Quantitative analyses were performed on a mass spectrometer (TSQ Quantum Ultra triple quadrupole mass spectrometer; Thermo Scientific, San Jose, CA, USA) equipped with a heated electrospray ionization probe that was kept at 355 °C. All analyses were performed in the positive ionization mode with a spray voltage set at 5000 V. The sheath and auxiliary gas used was nitrogen at 45 and 10 arbitrary units, respectively. The system was operated in the selected reaction monitoring mode with argon as the collision gas at a pressure of 1.5-mTorr. The ion transfer tube was kept at 300 °C while the scan time and width were 0.25 sec and 0.1 m/z, respectively. Data were processed using LCQuan software version 2.6 (Thermo Scientific, San Jose, CA, USA). The triple quadrupole mass spectrometer was coupled with liquid chromatography (1100 Agilent LC system; Agilent, Santa Clara, CA, USA). Chromatographic separation employed a column (ACE C$_{18}$, 100 × 2.1 mm, 3 μm, column; Mac Mod, Chadds Ford, PA, USA) and a linear gradient of acetonitrile (HPLC grade; Burdick and Jackson, Muskegon, MI, USA) in water with a constant 0.2% formic acid (spectrophotometric grade; Aldrich, St Louis, MO, USA) at a flow rate of 0.35 mL/min. The acetonitrile concentration was held at 1% for 0.5 min, ramped up to 90% over 8.5 min. Prior to analysis, the plasma proteins, controls, and calibrators were fortified with 100 ng/mL of oxymorphone-D3. The injection volumes were 10.0 μL.

Detection and quantification employed full-scan LC-MS/MS transitions of initial product ions for oxymorphone [mass to charge ratio (m/z) 302.1] and for amantadine [mass to charge ratio (m/z) 152.1]. The response for the major product ion, for oxymorphone (m/z, 284.1, 227.0, 198.0, 242.1, and 181) and for amantadine (m/z, 135.1, 77.1, 93.1, 79.1, and 107.1) was plotted, and peaks at the proper retention time integrated using LCQuan. The software was used to generate calibration curves and quantitate the analytes in all samples.

The concentration of oxymorphone and amantadine in each sample (e.g., calibrators, quality control, and unknowns) was determined by an internal standard method using the peak area ratio and linear regression analysis. The response for oxymorphone and amantadine was linear and gave correlation coefficients (R^2) of 0.99 or better. The technique was optimized to provide a limit of quantitation at 2.5 ng/mL for both analytes. For oxymorphone, the accuracy (percentage of nominal concentration) was 102.7% and 102.3% at 50 and 1000 ng/mL, respectively. The precision (percentage relative standard deviation) was 8.3% and 7.5% at 50 and 1000 ng/mL, respectively. For amantadine, the accuracy was 93.4% and 105.5% at 50 and 1000 ng/mL, respectively. The

precision was 10.3% and 10.0% at 50 and 1000 ng/mL, respectively.

Pharmacodynamic analysis

Nonlinear least squares regression (WinNonlin 6.1; Pharsight, Cary, NC, USA) was performed on oxymorphone concentration–thermal threshold data. For this analysis, thermal threshold data were transformed to % maximum possible effect (% MPE). according to the following equation:

$$\% \, MPE = \frac{TT - Baseline}{Cutoff - Baseline} \times 100$$

where TT is the observed thermal threshold, baseline is the thermal threshold at baseline (i.e., before drug administration), and cutoff is the temperature at which the stimulus is discontinued if the cat does not respond (55 °C). Simple and sigmoid effect maximum models were fitted to the data. Observation of the residuals plot and Akaike's information criterion were used to select which model fitted the data best (Yamaoka *et al.*, 1978). Parameters estimated by the model were E_{max} (MPE of oxymorphone), EC_{50} (plasma oxymorphone concentration producing 50% of E_{max}), and γ (sigmoidicity factor).

Statistical analysis

Power analysis based on the results of other thermal threshold studies previously conducted in our laboratory (Pypendop *et al.*, 2006, 2008) suggested that six cats were needed to detect a thermal threshold increase of 5 °C, with an α level of 0.05 and a power of 0.8. Normal distribution of all data was verified using the Shapiro-Wilk test. Skin temperature, thermal threshold, and thermal excursion (the difference between thermal threshold and skin temperature) were analyzed for dose (target plasma oxymorphone concentration) and treatment (amantadine vs. saline) effects using a repeated-measures ANOVA. A Dunnett or Tukey test was used where appropriate for pairwise comparisons. Oxymorphone EC_{50} values with and without amantadine were compared using a paired *t*-test. Statistical significance was set at $P < 0.05$. Skin temperature, thermal threshold, thermal excursion, and plasma drug concentration data are presented as mean ± SD. Pharmacodynamic parameters are presented as mean ± SE.

RESULTS

No cat experienced an exaggerated response to the stimulus or developed any sign of skin burns during the study. Marked mydriasis and excessive salivation were observed following oxymorphone administration in both the amantadine and saline groups. Dysphoria commonly occurred at the higher target plasma oxymorphone concentrations (200–400 ng/mL).

Actual plasma oxymorphone concentrations were 0 ± 0, 13.3 ± 3.3, 19.5 ± 2.8, 40.3 ± 16.3, 86.6 ± 34.0, 161.0 ± 47.0, and 311.3 ± 116.3 ng/mL for the 0, 10, 20, 50, 100, 200, and 400 ng/mL target plasma concentrations, respectively, in the amantadine group, and 0 ± 0, 11.7 ± 3.2, 18.1 ± 2.1, 35.9 ± 6.0, 74.9 ± 16.0, 109.7 ± 7.5, and 293.1 ± 44.5 ng/mL for the 0, 10, 20, 50, 100, 200, and 400 ng/mL target plasma concentrations, respectively, in the saline group. Actual amantadine plasma concentrations were 919 ± 353 and 0 ± 0 ng/mL in the amantadine and saline groups, respectively.

Skin temperature, thermal threshold, and thermal excursion increased with increasing plasma oxymorphone concentration in both the amantadine and the saline groups (Table 1). There was no significant difference in skin temperature, thermal threshold, or thermal excursion between the amantadine and saline groups.

Thermal threshold–plasma concentration data for both oxymorphone with and without amantadine best fitted sigmoid effect maximum model with baseline effect. According to this model, the effect can be predicted by the following equation:

$$E_c = \frac{(E_{max} \times C^{\gamma})}{EC_{50}^{\gamma} + C^{\gamma}}$$

where E_c is the effect (%MPE) at plasma oxymorphone concentration C, E_{max} is the MPE of oxymorphone, EC_{50} is the plasma oxymorphone concentration producing 50% of E_{max}, and γ is the sigmoidicity factor. The model adequately fitted the concentration–effect data in five cats; neither the simple nor the sigmoid effect maximum model fitted the data from the sixth cat

Table 1. Mean ± SD skin temperature, thermal threshold, and thermal excursion in six cats receiving oxymorphone and amantadine or saline

| Oxy TPC | Amantadine | | | Saline | | |
	ST	TT	TE	ST	TT	TE
0	37.1 ± 0.7	44.4 ± 2.5	7.3 ± 2.4	37.3 ± 0.6	47.0 ± 4.7	9.7 ± 5.1
10	37.5 ± 1.0	50.0 ± 3.2*	12.5 ± 3.3*	37.7 ± 0.7	47.8 ± 5.0	10.1 ± 5.2
20	38.0 ± 1.1	51.6 ± 3.0*	13.6 ± 2.4*	37.8 ± 0.9	50.2 ± 4.5	12.3 ± 4.5
50	38.5 ± 0.9	54.0 ± 1.9*	15.5 ± 2.0*	38.1 ± 1.1	53.0 ± 2.9*	14.9 ± 3.5*
100	39.0 ± 1.0*	54.4 ± 2.0*	15.4 ± 2.0*	38.5 ± 1.2	53.4 ± 2.0*	14.9 ± 2.6*
200	39.4 ± 1.0*	55 ± 0*	15.6 ± 1.0*	39.0 ± 1.2*	54.8 ± 0.7*	15.8 ± 1.3*
400	39.9 ± 0.8*	55 ± 0*	15.1 ± 0.8*	39.5 ± 1.1*	54.8 ± 0.6*	15.3 ± 1.2*

Oxy TPC, oxymorphone target plasma concentration (ng/mL); ST, skin temperature (°C); TT, thermal threshold (°C); TE, thermal excursion (i.e., TT-ST; °C).
*Value is significantly different ($P < 0.05$) from its respective baseline (Oxy TPC 0).

adequately. E_{max}, EC_{50}, and γ were $104 \pm 2\%$ and $98 \pm 2\%$, 14.2 ± 1.2 and 24.2 ± 7.4 ng/mL, and 5.6 ± 2.0 and 6.4 ± 1.6 in the amantadine and saline groups, respectively. The mean equations therefore were

$$E_c = \frac{(104 \times C^{5.6})}{(2836709 + C^{5.6})}$$

for the amantadine group, and

$$E_c = \frac{(98 \times C^{6.4})}{(718486053 + C^{6.4})}$$

for the saline group. EC_{50} was not significantly different between groups. However, it was approximately half in the amantadine group compared with the saline group in one cat (10.9 vs. 22 ng/mL), and 4–5 times less in the amantadine group compared with the saline group in a second cat (11.9 vs. 53.4 ng/mL).

DISCUSSION

In this study, overall, amantadine did not potentiate the antinociceptive effect of oxymorphone in cats. These results should be interpreted in view of a few limitations. First, a type II statistical error cannot be excluded. While the study was adequately powered based on previous studies using similar measurements conducted in our laboratory, adequate power does not guarantee that a significant difference will be found if one exists. Indeed, a power of 0.8 indicates that one in five trials is expected not to find a difference when one exists. In addition, some of the data used for power analysis may not have been valid in the present study. For example, the baseline thermal threshold was higher and more variable in this study than in previous studies, particularly in the saline group; the skin temperature increased with increasing plasma oxymorphone concentrations, which limits the magnitude of the increase in thermal excursion for a given increase in thermal threshold. The magnitude of the effect of amantadine may also be smaller than that considered in the power analysis; this would possibly result in lower than assumed power. Finally, while power analysis was based on thermal threshold results rather than on difference in EC_{50}, the fact that data from one cat could not be modeled satisfactorily and that the pharmacodynamic analysis was therefore based on data from five rather than six cats limits the power of that analysis. Second, the thermal threshold model may not be adequate to test the effect of amantadine. This model tests for changes in thermal nociception, and it is possible that amantadine would affect other types of nociception differently. More importantly, while NMDAR antagonists have been shown to reduce acute pain and opioid consumption after surgery in several studies, their effect is likely related to the prevention of central nervous system sensitization to noxious stimulation (McCartney *et al.*, 2004; Annetta *et al.*, 2005), and they may have minimum effect on nociception. Third, while the plasma oxymorphone concentrations were similar in the amantadine and saline groups at most target oxymorphone concentrations, there was a larger difference at the 200 ng/mL target concen-

tration. The effect on the results is unknown; it could be argued that because of the difference in actual plasma oxymorphone at the 200 ng/mL target, the groups should not be directly compared. However, because each cat received the same dose of oxymorphone in both the amantadine and saline studies, the group comparison based on target rather than actual concentration represents an analysis of the dose–effect relationship. The pharmacodynamic modeling represents an analysis of the concentration–effect relationship. The actual concentrations were lower than their respective targets at target concentrations of 50 ng/mL and higher. These lower than targeted oxymorphone concentrations had no influence on the pharmacodynamic modeling, as individual actual oxymorphone concentrations were used for this analysis, and the range of concentrations was wide enough for good characterization of the effect on thermal threshold, as illustrated by the identical thresholds in both groups at the two highest plasma oxymorphone concentrations. The range of target plasma oxymorphone concentrations in this study was selected to reflect the range of peak concentrations likely produced with clinical doses. According to our previous pharmacokinetic study, 10 and 400 ng/mL are the peak concentrations produced after intravenous oxymorphone dose of approximately 0.005 and 0.2 mg/kg. Finally, the plasma amantadine concentration was lower than targeted and was variable both within and between cats. The target amantadine concentration of 1100 ng/mL was selected based on a previous pharmacokinetic study to maintain the peak concentration observed after oral dose of 5 mg/kg of amantadine. This dose has anecdotally been used in conjunction with opioids for the treatment of pain in cats. It is possible that an effect of amantadine would have been observed if higher concentrations had been reached.

While the effect of oxymorphone in combination with amantadine on thermal threshold was not statistically significantly different than that of oxymorphone alone, the results of this study provide indirect evidence that amantadine may potentiate the effect of oxymorphone in some, but not in all, cats. The variability of the effect of oxymorphone on thermal threshold appeared slightly less in the amantadine group, as illustrated by the fact that thermal threshold and thermal excursion values were significantly higher than baseline values for all oxymorphone concentrations in that group, and the cutoff temperature was reached without observing a response in all six cats at the two highest oxymorphone target concentrations, whereas it was reached in four of six cats in the saline group. Similarly, the mean EC_{50} of oxymorphone, while not statistically significantly different between groups, was 41% lower in the amantadine group, compared with the saline group. In the five cats in which oxymorphone EC_{50} could be determined, it was very similar in the amantadine and saline group in three cats (15.2 and 14.0, 17.3 and 13.8, and 15.8 and 18.0 ng/mL in the amantadine and saline groups, respectively), while it was largely lower in the amantadine than in the saline group in two cats (11.9 and 53.4, and 10.9 and 22 ng/mL, in the amantadine and saline groups, respectively). The plasma amantadine concentration in the two cats in which a reduction in oxymorphone EC_{50} was observed was not the highest. Taken

together, these observations may suggest that the lack of statistical significance in this study was related to inadequate statistical power and/or to a large variability in response between individual cats, with amantadine reducing the effective antinociceptive dose of oxymorphone in some, but not all, cats.

Skin temperature increased in cats in a manner dependent on the plasma oxymorphone concentration. While body temperature was not measured in the present study, opioid-induced hyperthermia has been reported in cats (Niedfeldt & Robertson, 2006; Posner *et al.*, 2007, 2010). The mechanism for this effect has not, to the authors' knowledge, been reported. It is likely that skin temperature increased as a result of increased body temperature.

In conclusion, in this study, overall, amantadine did not reduce the antinociceptive dose of oxymorphone. However, an effect was likely produced in some cats. These results suggest that amantadine may be useful to reduce the analgesic dose of opioids in cats in clinical practice, but that analgesic treatment will need to be tailored to the individual's response.

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