

ORIGINAL ARTICLE

Pharmacokinetics of intravenous, oral transmucosal, and intranasal buprenorphine in healthy male dogs

Hiroko Enomoto¹ | Lydia Love² | Melanie Madsen² | Amber Wallace² |
Kristen M. Messenger^{1,2}

¹Comparative Medicine Institute, North Carolina State University, Raleigh, North Carolina, USA

²Department of Molecular Biomedical Sciences, North Carolina State University, Raleigh, North Carolina, USA

Correspondence

Kristen M. Messenger, Department of Molecular Biomedical Sciences, College of Veterinary Medicine, North Carolina State University, 1060 William Moore Drive, Raleigh, North Carolina 27607, USA.
Email: kmmessen@ncsu.edu

Funding information

Center for chemical Toxicology Research and Pharmacokinetics of North Carolina State University; Zoetis generously provided the Simbadol™ used in this study.

Abstract

Effective management of pain in animals is of critical importance but options are limited for treating acute pain in dogs on an outpatient basis. The objective of this study was to compare the plasma concentrations and pharmacokinetics of a concentrated solution of buprenorphine, 1.8 mg/ml (Simbadol™) administered intravenously, intranasally, and via the oral transmucosal (OTM) route in healthy male dogs. Five healthy castrated adult male Beagle-cross dogs were included in this randomized blocked crossover study. The dogs received 0.03 mg/kg body weight buprenorphine intravenously, intranasally, or via the OTM route, with a minimum 72-h washout period between treatments. Blood samples were collected at multiple intervals up to 24 h post administration and buprenorphine plasma concentrations were determined by liquid chromatography tandem mass spectrometry. Non-compartmental pharmacokinetic analysis revealed that the area under the curve of intravenous, intranasal, and OTM routes were 28.0 (15.1–41.3) h × ng/ml, 16.1 (3.4–28.7) h × ng/ml and 10.8 (8.8–11.8) h × ng/ml, respectively. The bioavailability of intranasal and OTM routes were 57.5 (22.7–93.7)% and 41.1 (25.5–69.4)%, respectively. Intranasal and OTM routes of administration of concentrated buprenorphine in dogs may allow for the provision of analgesic care at home.

KEYWORDS

buprenorphine, dog, intranasal, oral transmucosal, pharmacokinetics, Simbadol™

1 | INTRODUCTION

Effective outpatient pain management options for dogs are limited. Non-steroidal anti-inflammatory drugs (NSAIDs) may be contraindicated in some animals, including those with a risk of gastroduodenal ulceration and erosion, kidney disease, or hepatic dysfunction (Kore, 1990). Orally administered opioids are not well-absorbed in dogs and have not been shown to be efficacious in clinical studies (Benitez et al., 2015a,b). Acetaminophen has been used in dogs for the management of acute pain, but to date, there is still very little

evidence for analgesic efficacy of this drug in canines (Hernández-Avalos et al., 2020; Leung et al., 2021), or that effective concentrations can be maintained (Madsen et al., 2022).

Buprenorphine is a relatively long-acting and potent partial μ -agonist opioid analgesic used clinically for treatment of mild to moderate pain in dogs and cats (Brodbelt et al., 1997; Watanabe et al., 2018; Watanabe et al., 2020). Oral bioavailability of buprenorphine is low because of extensive first-pass hepatic metabolism; however, it has favorable physiochemical properties, such as high lipophilicity, which meets criteria for transmucosal penetration

This is an open access article under the terms of the [Creative Commons Attribution-NonCommercial-NoDerivs](https://creativecommons.org/licenses/by-nc-nd/4.0/) License, which permits use and distribution in any medium, provided the original work is properly cited, the use is non-commercial and no modifications or adaptations are made.

© 2022 The Authors. *Journal of Veterinary Pharmacology and Therapeutics* published by John Wiley & Sons Ltd.

(Johnson et al., 2005). It is commonly administered to cats via the oral transmucosal route (OTM) for outpatient analgesic care (Robertson et al., 2003). Oral transmucosal buprenorphine is not as bioavailable in dogs as compared to intravenous administration, hence a high dose may be required (Abbo et al., 2008). Intranasal (IN) buprenorphine administration for non-invasive drug delivery, for example, in the home environment or when an IV catheter is not present, is attractive because the highly vascularized large surface area of the canine nasal mucosa facilitates high systemic absorption and bypasses first-pass hepatic metabolism. In addition, drugs administered by the nasal route can directly reach the central nervous system (CNS) via the trigeminal and olfactory nerves (Erdo et al., 2018). The IN route of administration in dogs has been investigated for midazolam, dexmedetomidine, epinephrine, naloxone, and ketamine (Bleske et al., 1992; Charalambous et al., 2017; Charalambous et al., 2019; Dretchen et al., 2020; Santangelo et al., 2019; Tuttle et al., 2020; Vlerick et al., 2020; Wahler et al., 2019). These studies reported the intranasal route of administration to be clinically effective and/or results in adequate plasma concentrations of the investigated drug. Intranasal (IN) drug administration has not been investigated for buprenorphine in dogs.

A commercially available, highly concentrated (1.8 mg/ml) formulation of buprenorphine (Simbadol™, Zoetis, Florham Park, NJ, USA) is FDA-approved for use in feline patients subcutaneously every 24 h for up to three days for the management of postoperative pain. Transmucosal administration of other commercial solutions of buprenorphine (0.3 mg/ml) limit the use in dogs because high volumes are needed for administration, but the 1.8 mg/ml concentrated solution allows for small volumes to be administered to dogs. These properties of buprenorphine led us to consider this investigation in dogs.

The purpose of the present study was to compare the plasma concentrations and the pharmacokinetics of a concentrated buprenorphine solution when administered intravenously, intranasally, or via the OTM route to dogs. We hypothesized that IN and OTM administration of concentrated buprenorphine would result in a favorable pharmacokinetic profile, comparable to IV administration.

2 | MATERIALS AND METHODS

2.1 | Animals

Five male castrated Beagle-mix dogs (mean age 9.2 ± 3.5 years; mean weight; 12.9 ± 2.4 kg), assessed as healthy based on physical examination findings and routine laboratory analyses, including complete blood count, chemistry profile, and fecal exam, were enrolled in the study. The subjects were housed in the North Carolina State University Laboratory Animal Resources facility, where a maintenance diet was provided twice daily, and water was provided ad libitum. Animals were acclimated to the study environment for a minimum of seven days before the beginning of the data collection. The study was approved by the Institutional Animal Care

and Use Committee at North Carolina State University (protocol # 20-480-O).

2.2 | Study design/treatments

A prospective, randomized, within-subjects crossover experimental design was used for this study. Dogs were assigned by a random number generator (GraphPad Prism 6; GraphPad Software Inc., La Jolla, CA, USA) to receive any of the 3 treatments: IV 1.8 mg/ml buprenorphine (Simbadol™; Zoetis), OTM 1.8 mg/ml buprenorphine, and IN 1.8 mg/ml buprenorphine. All treatments used the same dose of 0.03 mg/kg. The mean volume of solution administered to each dog for all routes was 0.21 (0.16–0.26) ml. There was a minimum 96-h washout period between treatments. Intravenous administration was accomplished via a 24-g cephalic IV catheter (Surflo 1.25 inch; Terumo Medical Corporation, Elkton, MD, USA). This catheter was removed immediately after drug administration. Oral transmucosal administration was achieved by applying the full volume of drug to the buccal mucosa at one time. Intranasal administration was accomplished by applying the full volume of drug to the nostril on one side using a nasal mucosal atomization device (MAD) (Teleflex, Morrisville, NC, USA) by a single investigator (LL). Dogs were gently restrained, and the head was tipped dorsally prior to superficial insertion of the MAD into the nares. The pH of the IN cavity and buccal pouch were measured by using pHDrion insta-check 0–13 (Micro Essential Laboratory, NY, USA) before drug administration.

2.3 | Catheter placement

Because blood sample collection from the jugular vein can falsely increase the bioavailability from drugs administered to the oral or nasal mucosa (Sohlberg et al., 2013), we collected blood from another peripheral vein. Twenty-four hours prior to the first study day, following a 12-h fast, the dogs were sedated with dexmedetomidine (10 mcg/kg IV) and a peripherally inserted central catheter (Jorgensen Laboratory, Inc, USA) was placed in the medial or lateral saphenous vein. Atipamezole 100 mcg/kg was administered intramuscularly to reverse the dexmedetomidine. The catheters were rinsed with a heparin solution; however, we were not able to maintain patency for all of the catheters throughout the study time period. For dogs with catheters that did not remain patent, a 20- or 18-g, 1.25-inch cephalic catheter (Surflo 1.25 inch; Terumo Medical Corporation) was placed the day of drug administration for venous blood sampling (in the opposite thoracic limb if a cephalic IV catheter was placed for IV drug administration).

2.4 | Sample collection

Dogs were fasted for 12 h prior to drug administration on each study day. Blood samples were collected prior to drug administration to be

used as a negative control in the buprenorphine assay. After drug administration, blood samples (1–1.5 ml) were then collected at 0.03-, 0.08-, 0.17-, 0.25-, 0.5-, 0.75-, 1, 1.5-, 2-, 4-, 6-, 8-, 12-, and 24-h time points. After collection, samples were transferred into tubes containing lithium heparin (BD Vacutainer; Franklin Lakes, NJ, USA) and placed on ice. Samples were centrifuged at 3500g for 10 min at 4°C within 60 min of collection. The plasma from centrifuged samples was separated and stored at -80°C until analysis within 1 month of collection.

2.5 | Physiologic parameters

Temperature, pulse, and respiration were recorded at baseline and at 2 h after drug administration. Obvious side effects such as sedation, disinterest in surroundings, obtaining lateral recumbency, ptialism, vomiting, and defecation were noted at each blood sampling time point. The regular diet was offered 4 h post drug administration. If the subjects did not eat their regular diets of dry kibble, higher value canned food was offered, and appetite was recorded.

2.6 | Buprenorphine assay

All reagents except ammonium hydroxide were of LC/MS grade. Acetonitrile, methanol, formic acid, 29% ammonium hydroxide and acetic acid were supplied by Fisher Scientific (Hampton, NH). Buprenorphine reference standard was purchased from Sigma-Aldrich (Sigma-Aldrich RTC, Laramie, WY, USA). Ultrapure water was supplied by Water Corporation (Milford, MA, USA). Phosphoric acid was supplied by Aldrich Chemistry. Analysis of buprenorphine was carried out via ultra-performance liquid chromatography (UPLC) and tandem mass spectrometric (MS/MS) detection (Waters Corporation). The UPLC-MS/MS system consisted of a Waters Acquity UPLC I class Binary Solvent Manager, Acquity UPLC Sample Manager FTN and a Xevo TQD tandem mass spectrometer (Waters Corporation).

2.6.1 | Preparation of standard working solutions and calibration standard preparation

Standards of 1.75, 3.5, 8.75, 17.5, 35, 175, 350, 3500, 50,000, 100,000, and 500,000 ng/ml buprenorphine were prepared in the maximum recovery vial (Water Corporation) by dilution of buprenorphine stock solution (1 mg/ml) with 100% methanol.

Each standard solution of buprenorphine (1.75, 3.5, 8.75, 17.5, 35, 175, and 350 ng/ml) was diluted in glass tubes with blank canine plasma to give concentrations 0.05, 0.1, 0.25, 0.5, 1, 5, and 10 ng/ml for the calibration curve. Blank (zero) canine plasma was injected with every batch.

2.6.2 | Sample preparation

Plasma (350 µl) was pipetted to a clean glass tube (disposable culture tubes, borosilicate glass, VWR). Three hundred fifty microliter

of 5% phosphoric acid in water was added to each tube to pretreat the plasma. Solid phase extraction was then performed on an Oasis MCX 96 well µElution plate (Water Corporation). The plate was prepared by first conditioning it with 200 µl of methanol followed by 200 µl of ultrapure water. And then, 700 µl of pretreated plasma was loaded on the µElution plate and passed through the plate under vacuum (<5 mm). The pressure of the vacuum was increased as necessary to pull the samples through the plate. The plate was then washed with 200 µl of 0.2% acetic acid in water followed by 200 µl 100% methanol under vacuum. The plate was dried under vacuum for 1 min. The analyte was eluted into a clean 96-well collection plate (700 µl round 96 well samples plate, Waters Acquity UPLC, Water Corporation) by vacuum with addition of 50 µl of elution solution (acetonitrile:methanol:29% ammonium hydroxide = 57%:38%:5%) (Regina & Kharasch, 2013). Fifty microliter of water was added to the eluent and mixed thoroughly. Some samples from the IV group that were collected at 0.03-, 0.08-, 0.17-, 0.25-, and 0.5-h time points were diluted sixfold with additional blank canine plasma to achieve adequate volume for analysis and sample preparation was performed as described above. Some IV samples for 0.75–1.5-h time points were diluted 2–3.5 times with additional blank canine plasma before sample preparation.

2.6.3 | UPLC-MS/MS conditions

Chromatographic separation was performed by a gradient elution on the ACQUITY UPLC HSS T3 1.8 µm column (2.1×100mm) with VanGuard pre-column (Waters Corporation). The mobile phase solvents were 0.1% formic acid in water (A) and 0.1% formic acid in acetonitrile (B) at a flow rate 0.4 ml/min for 5 min. The gradient program mobile phase conditions were 85% of A and 15% of B for the first 2.5 min, then changed linearly to 10% of A and 90% of B from 2.5–4.0 min, then immediately back to 85% of A and 15% of B from 4.0–5.0 min to re-equilibrate at the initial conditions. The column temperature was 40°C, the autosampler temperature was maintained at 10°C and the injection volume was 8 µl. The positive electrospray ionization mode (ESI [+]) was used with the multiple reactions monitoring (MRM). The tune page source voltages were 0.7 kV and 84 V for the capillary and cone, respectively. The source desolvation temperature was 500°C. The source desolvation gas flow was 800 L/h and the cone gas was 50 L/h. The MS file cone voltage setting was 74 V with collision energy setting of 48 V. Argon was used as the collision gas and nitrogen as the desolvation and cone gases. Quantification was performed using the transition Parent (m/z): 468.34 and Daughter (m/z): 83.77 with a retention time of 2.43 min.

2.6.4 | The lower limit of quantification and the lower limit of detection

Lower limit of quantification (LLOQ) was defined as the lowest concentration that produced a peak area 5 times the blank peak area,

with an accuracy within 15% of the nominal value, and a precision of no more than 15% of coefficient variation. The LLOQ of buprenorphine was 0.1 ng/ml. The lower limit of detection (LLOD) was the lowest concentration that produced a peak area >3 times blank peak area. The LLOD of buprenorphine was 0.05 ng/ml.

The calibration curve was fitted with a weighted (1/concentration) linear equation. The calibration range of 0.05–10 ng/ml was linear with a coefficient of determination, R^2 , greater than or equal to 0.99. Each calibration standard concentration could be back calculated to within 15% of the true concentration.

A total of 6 replicates at low, medium, and high concentration (0.3, 3 and 7 ng/ml) were tested on 3 days and interday and intraday precision and accuracy were calculated. The precision ranged from 3–12% with accuracy between 87 and 108%.

2.7 | Pharmacokinetics

Non-compartmental pharmacokinetic analysis of buprenorphine in plasma were performed using commercially available software (Phoenix WinNonlin, version 8.3; Certara, St. Louis, MO, USA). Only the concentrations above LLOQ were included in the pharmacokinetic analysis. The pharmacokinetic parameters estimated for buprenorphine in plasma after IV, IN, and OTM administration included the elimination rate constant (λ_z), terminal half-life (HL_{λ_z}), the time to maximum concentration (T_{max}), the maximum concentration (C_{max}), the area under the curve from time zero to the last time point (AUC_{last}), the area under the curve from time zero to infinity (AUC_{inf}), which were calculated using the linear log trapezoidal method, the volume of distribution (V), and clearance (CL). The bioavailability was calculated by using the following equation:

$$\text{Bioavailability IN or OTM}(\%) = (AUC_{IN \text{ or } OTM} / AUC_{IV}) \times 100$$

2.8 | Statistical analysis

Pharmacokinetic parameters were compared between IV, IN, and OTM routes. Statistical analysis was performed using commercially available software (Prism version 7.04, GraphPad Software Inc.). A Friedman ANOVA with Dunn's multiple comparisons test were performed to compare the pharmacokinetic parameters. Differences were considered significant at $p < .05$.

3 | RESULTS

Mean \pm standard deviation of age and weight were 9.2 ± 3.5 year and 12.9 ± 2.4 kg, respectively. All dogs completed the study and no severe adverse reactions to buprenorphine via any route of administration was observed. Dogs developed similar side effects regardless of administration route, which included subjective sedation, hypersalivation, ataxia, and decreased rectal temperature (96.8 – 99.5°F); these effects were observed up to 6 h after drug administration.

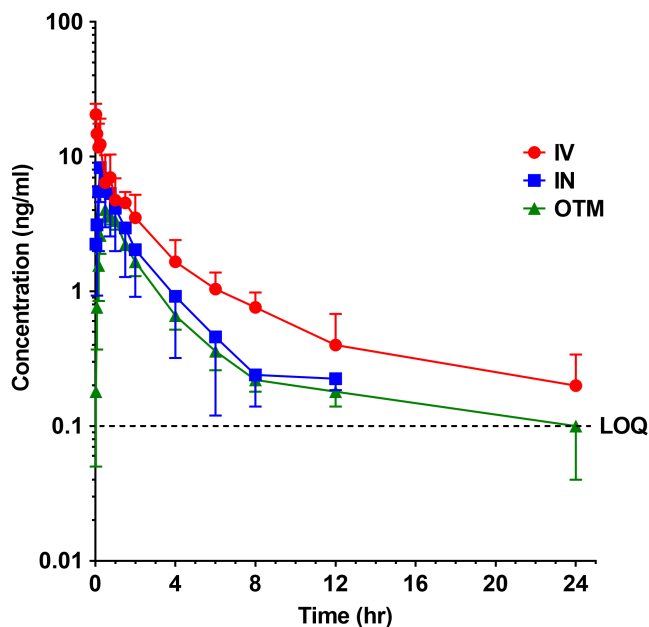


FIGURE 1 Buprenorphine plasma concentration (mean \pm standard deviation, ng/ml) versus time (hours) after intravenous (IV), intranasal (IN) and oral transmucosal (OTM) administration (Simbadol™; 0.03 mg/kg) in dogs ($n = 5$). The dotted line represents the limit of quantification (LOQ, 0.1 ng/ml) of the assay. The plasma concentration (0.6 ng/ml) that has been reported to be associated with analgesic effects in dogs for ovariectomy (Ko et al., 2011)

Loss of appetite was observed up to 12 h after all routes of administration; soft stool and diarrhea were observed 24 and 48 h after drug administration. Sneezing was observed after IN administration in 2/5 dogs, but was temporary. Serous nasal discharge was observed directly following IN administration in one animal. The pH of the intranasal cavity and buccal pouch ranged from 7–7.5 and 7.5–8.0, respectively.

3.1 | Buprenorphine concentrations in plasma

The plasma concentrations of buprenorphine versus time curves after IV, IN, and OTM administration are shown in Figure 1. The plasma concentration of buprenorphine versus time curve after just the IN and OTM administrations is shown in Figure 2.

3.2 | Pharmacokinetic parameters

Pharmacokinetic parameters for the IV, IN, and OTM routes are presented in Table 1. Significant differences were observed between groups for AUC and C_{max} . The AUC of the IV group was significantly greater than that of the OTM group ($p = .034$), but there was no significant difference between IV vs. IN or IN vs. OTM groups. The C_{max} of the IV group was significantly higher than the OTM groups ($p = .034$), but significant difference was not found between

IV vs. IN group. While the C_{max} of the IN group (median 8.7 ng/ml) was approximately double that of the OTM group (median 4.2 ng/ml), there was no significant difference between these values or any other parameters. The median bioavailability of IN was 57.5 (22.7–93.7)% and was 41.1 (25.5–69.4)% following OTM administration.

4 | DISCUSSION

This is the first report of the plasma concentrations and pharmacokinetics of intranasal concentrated buprenorphine in dogs. The plasma concentrations of buprenorphine measured in these dogs (0.9 to

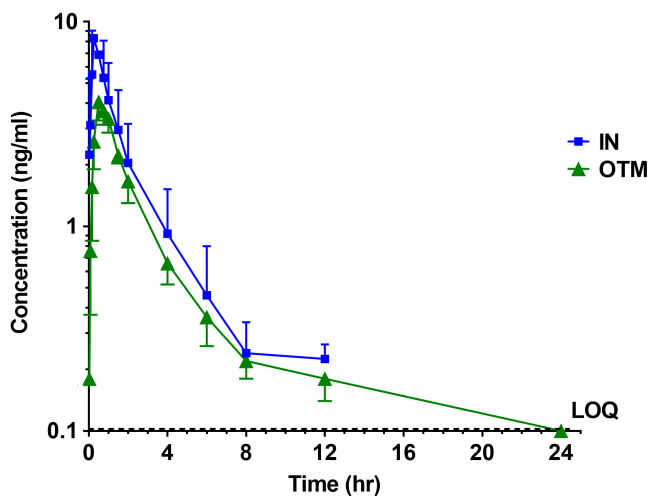


FIGURE 2 Buprenorphine plasma concentration (mean \pm standard deviation, ng/ml) versus time (hours) after only intranasal (IN) and oral transmucosal (OTM) administration (Simbadol™; 0.03 mg/kg) in dogs ($n = 5$). See legend in Figure 1 for the remainder of information

8.3 ng/ml) are within the range reported to be analgesic for up to 5 h (Ko et al., 2011), with short T_{max} and a moderate bioavailability of 57.5%. The plasma buprenorphine concentrations associated with analgesia have not been fully established in dogs; however, plasma concentrations greater than 0.6 ng/ml have been reported to be associated with analgesic effects in an ovariohysterectomy model in dogs (Ko et al., 2011). In the current study, the average plasma buprenorphine concentration of IN and OTM group was greater than 0.6 ng/ml for up to approximately 5 and 4 h, respectively. The IV route of administration of the same dose maintained plasma concentrations greater than 0.6 ng/ml for over 9 h.

Previously, the pharmacokinetics and the efficacy of concentrated buprenorphine administered intravenously, intramuscularly, and subcutaneously at 0.02 mg/kg, concurrently with carprofen, in dogs undergoing ovariohysterectomy (OVH) were reported (Steagall et al., 2020). The pharmacokinetics following an IV dose of 0.02 mg/kg, similar to our dose of 0.03 mg/kg, are remarkably similar compared with the dogs undergoing OVH. For example, clearance was nearly identical at 1.1 vs. 1.2 L/h/kg, volume of distribution at steady state was 4.4 vs. 4.5 L/kg, and terminal (elimination) half-life was 3.2 vs. 3.69 h in our study vs. the study by Steagall et al. (2020), respectively. A different study reported the pharmacokinetics of a high dose of concentrated buprenorphine in healthy dogs and is less comparable because of differences in dog populations (specifically ages and breeds) and differences in the pharmacokinetic analysis (Hansford et al., 2021). Despite these important methodological differences, a similar clearance (1.1 vs. 1.5 L/h/kg) is reported following IV administration supporting consistent and perhaps linear pharmacokinetics of buprenorphine in dogs across different doses.

No differences were found in all pharmacokinetic parameters, including bioavailability between IN and OTM groups. This finding is likely due to high inter-individual variability in drug concentrations

Parameter	Units	Administration route		
		IV	IN	OTM
λ_z	1/h	0.22 (0.13–0.29)	0.18 (0.14–0.24)	0.17 (0.12–0.31)
HL_{λ_z}	h	3.2 (2.4–5.4)	3.9 (2.9–4.8)	4.1 (2.2–6.0)
T_{max}	h		0.5 (0.3–0.8)	0.5 (0.5–0.8)
C_{max}	ng/ml		8.7 (1.8–12.6)	4.2 (3.5–4.7)
AUC_{last}	h \times ng/ml	25.6 (14.7–34.3)	14.7 (3.0–27.1)	9.6 (8.2–10.9)
AUC_{inf}	h \times ng/ml	28.0 (15.1–41.3)	16.1 (3.4–28.7)	10.8 (8.8–11.8)
AUC_{extrap}	%	6.0 (2.3–17.1)	8.7 (5.3–12.3)	11.0 (5.6–14.6)
V_{ss}	L/kg	4.4 (3.7–7.3)		
CL	L/h/kg	1.1 (0.7–2.0)		
F	%		57.5 (22.7–93.7)	41.1 (25.5–69.4)

Note: Data shown as median (range).

Abbreviations: λ_z , elimination rate constant; AUC_{extrap} , extrapolation of AUC; AUC_{inf} , area under the curve from time zero to infinity; AUC_{last} , area under the curve from time zero to the last time point; CL, clearance; C_{max} , maximum concentration; HL_{λ_z} , terminal half-life; F, bioavailability; T_{max} , time to the maximum concentration; V_{ss} , volume of distribution at steady state.

TABLE 1 Pharmacokinetic parameters following intravenous (IV), intranasal (IN) and via oral transmucosal (OTM) administration of buprenorphine (Simbadol™; 0.03 mg/kg) in 5 dogs

in plasma and small sample size, which has been reported by other investigators (Steagall et al., 2020), and observed in people (Johnson et al., 2005). In one dog, we suspect the total IN dose was not administered, as we observed nasal discharge immediately after drug administration, and this dog's plasma buprenorphine concentrations (and AUC) were less than half those of other dogs. This indicates that likely more than 50% of the administered drug was lost during administration and accounts for most of the variability in the IN data, given this small population of study animals. In accordance with our hypothesis, IN administration of concentrated buprenorphine resulted in a favorable pharmacokinetic profile compared with IV administration that may allow for effective outpatient analgesia in dogs.

There are many factors that could influence absorption of buprenorphine across the nasal mucosa, including breed variations in the anatomy; the presence, viscosity, or flow of mucus; drug residence time in nasal cavity; loss anteriorly from the nose or posteriorly into the esophagus; and administration technique (Erdo et al., 2018; Gizurarson, 1990; Kavoi et al., 2010). The highly lipophilic nature of the buprenorphine (LogP 4.98) (Kandimalla & Donovan, 2005) and the large surface area of the dog's nasal cavity are ideal characteristics for rapid drug absorption and delivery to the CNS (Bleske et al., 1992; Erdo et al., 2018). Intranasal or oral mucosal delivery of the commercial human formulation (0.3 mg/ml) is hampered by the large volume of solution required to deliver a therapeutic dose to dogs. However, the concentrated veterinary solution of 1.8 mg/ml (Simbadol™) allows for a small volume of administration (approximately 0.16–0.26 ml for a 10 kg dog), decreasing the likelihood of swallowing of buprenorphine or spillage from the administration site (Santangelo et al., 2019).

Intranasal administration of concentrated buprenorphine was performed by the same investigator using a MAD. The MAD creates a fine mist of particles in order to enhance drug absorption and, therefore, bioavailability (Charalambous et al., 2017; Charalambous et al., 2019). There was high variability in IN bioavailability among our dogs; however, our results were affected by the suspected loss of drug following administration in one dog that had observed nasal discharge after buprenorphine was administered, which resulted in an IN bioavailability of approximately 22% in this dog. Sneezing is reported during IN drug administration and could lead to drug loss and lack of drug effect, as well as owner exposure to the drug (Charalambous et al., 2017; Santangelo et al., 2019).

Following OTM administration, our results report a mean bioavailability of 41.1%, which was only slightly higher than a previous study (38%) that evaluated the OTM administration of a different formulation of buprenorphine to dogs at 0.02 mg/kg (Abbo et al., 2008). Many factors, including pH of saliva, drug volume, size of drug molecule, lipophilicity, mucosal permeability, drug formulation, metabolism, and physicochemical reactions pertaining to oral retention could affect buprenorphine absorption across the oral mucosa (Abbo et al., 2008; Ko et al., 2011). This study is different from previous studies in dogs or cats because we used a concentrated

solution of buprenorphine. We anticipated that a more concentrated solution would produce higher bioavailability compared with a more dilute solution, but there does not appear to be a substantial difference. Nevertheless, the drug volume (0.16–0.26 ml) of concentrated buprenorphine is small and would, therefore, be easier for pet owners and veterinary nurses to administer a therapeutic dose to clinical patients.

There are limitations associated with the present study. Notably, the effect of a small sample size could have resulted in Type 2 statistical error, preventing our detection of significance. However, small sample sizes of 5–8 animals are common in such studies in research animals. In addition, breed, dog size, and gender account for variability in pharmacokinetic estimates; our dogs were Beagle x Maltese crosses and ranged in age but were adult to geriatric (Fleischer et al., 2008; Santangelo et al., 2019; Wahler et al., 2019). Our subjects were all research dogs, deemed healthy based on our evaluation, but cannot speculate if the results reported here would be similar in clinical patients.

5 | CONCLUSIONS

Both the IN and OTM routes of delivery of a commercially available concentrated buprenorphine solution produced plasma concentrations that are within a therapeutic range in this group of healthy research dogs.

ACKNOWLEDGEMENTS

The Simbadol™ use in this study was generously provided by Zoetis. This project was funded by the Center for Chemical Toxicology Research and Pharmacokinetics of North Carolina State University. The authors thank Dr. Mark Papich for his assistance with study design and manuscript review.

ANIMAL WELFARE AND ETHICS STATEMENT

The study was approved by the Institutional Animal Care and Use Committee at North Carolina State University (protocol # 20-480-O). [Correction added on 03 September 2022, after first online publication: The Animal Welfare and Ethics Statement was included in this current version.]

CONFLICT OF INTEREST

KMM and MGP have been paid consultants for Zoetis and received gifts and honoraria from Zoetis. Other authors have no competing interests related to this study.

AUTHOR CONTRIBUTION

HE developed the buprenorphine analytical method, performed PK analysis, conducted the data collection and management, and wrote the manuscript. MM participated in data collection and sample analysis. AW contributed to data collection and manuscript writing and review. LL participated in study design, data collection, manuscript writing and review. KM conceived and designed the study,

collected data, assisted with PK analysis, contributed to, and approved the final manuscript. All authors reviewed the manuscript before submission.

DATA AVAILABILITY STATEMENT

The findings of this study are available from the corresponding author upon reasonable request.

ORCID

Hiroko Enomoto  <https://orcid.org/0000-0001-5095-3430>

Kristen M. Messenger  <https://orcid.org/0000-0001-8161-4482>

REFERENCES

- Abbo, L. A., Ko, J. C. H., Maxwell, L. K., Galinsky, R. E., Moody, D. E., Johnson, B. M., & Fang, W. B. (2008). Pharmacokinetics of buprenorphine following intravenous and oral transmucosal administration in dogs. *Veterinary therapeutics*, 9(2), 83–93.
- Benitez, M. E., Roush, J. K., McMurphy, R., Kukanich, B., & Legallet, C. (2015a). Clinical efficacy of hydrocodone-acetaminophen and tramadol for control of postoperative pain in dogs following tibial plateau leveling osteotomy. *American Journal of Veterinary Research*, 76(9), 755–762.
- Benitez, M. E., Roush, J. K., Kukanich, B., & McMurphy, R. (2015b). Pharmacokinetics of hydrocodone and tramadol administered for control of postoperative pain in dogs following tibial plateau leveling osteotomy. *American Journal of Veterinary Research*, 76(9), 763–770.
- Brodgelt, D. C., Taylor, P. M., & Stanway, G. W. (1997). A comparison of preoperative morphine and buprenorphine for postoperative analgesia for arthroscopy in dogs. *Journal of Veterinary Pharmacology and Therapeutics*, 20(4), 284–289. <https://doi.org/10.1046/j.1365-2885.1997.00060.x>
- Bleske, B. E., Warren, E. W., Rice, T. L., Shea, M. J., Amidon, G., & Knight, P. (1992). Comparison of intravenous and intranasal administration of epinephrine during CPR in a canine model. *Animals of Emergency Medicine*, 21(9), 1125–1130.
- Charalambous, M., Volk, H. A., Tipold, A., Erath, J., Huenerfauth, E., Gallucci, A., Gandini, G., Hasegawa, D., Pancotto, T., Rossmel, J. H., Platt, S., de Riso, L., Coates, J. R., Musteata, M., Tirrito, F., Cozzi, F., Porcarelli, L., Corlazzoli, D., Cappello, R., Vanhaesebrouck, B. J. G., Broeckx, L. V. H., Van Ham, L., Bhatti, S. F. M. (2019). Comparison of intranasal versus intravenous midazolam for management of status epilepticus in dogs: A multi-center randomized parallel group clinical study. *Journal of Veterinary Internal Medicine*, 33, 2709–2717.
- Charalambous, M., Bhatti, S. F. M., Van Ham, L., Platt, S., Jeffery, N. D., Tipold, A., Siedenburg, J., Volk, H. A., Hasegawa, D., Gallucci, A., Gandini, G., Musteata, M., Ives, E., & Vanhaesebrouck, A. E. (2017). Intranasal midazolam versus rectal diazepam for the management of canine status epilepticus: A multicenter randomized parallel group clinical trial. *Journal of Veterinary Internal Medicine*, 31(4), 1149–1158. <https://doi.org/10.1111/jvim.14734>
- Dretchen, K. L., Mesa, Z., Robben, M., Slade, D., Hill, S., Crutch, C., Kappeler, K., & Mesa, M. (2020). Intranasal epinephrine in dogs: Pharmacokinetics and heart rate effects. *Pharmacology Research and Perspectives*, 8, 1–8. <https://doi.org/10.1002/prp2.587>
- Erdo, F., Bors, L. A., Farkas, D., Bajza, A., & Gizutarson, S. (2018). Evaluation of intranasal delivery route of drug administration for brain targeting. *Brain Research Bulletin*, 143, 155–170.
- Fleischer, S., Sharkey, M., Mealey, K., Ostrander, E. A., & Martinez, M. (2008). Pharmacogenetic and metabolic differences between dog breeds: Their impact on canine medicine and the use of the dog as a preclinical animal model. *The Journal of American Association of Pharmaceutical Scientists*, 10(1), 110–119.
- Giaurarson, S. (1990). Animal models for intranasal drug delivery studies. *Acta Pharmaceutica Nordica*, 2(2), 105–122.
- Hansford, J., Henao-Guerrero, N., Machado, M., & Pypendop, B. (2021). Pharmacokinetics of a high-concentration formulation of buprenorphine (Simbadol) in male dogs. *Veterinary Anaesthesia and Analgesia*, 48(4), 509–516.
- Hernández-Avalos, I., Valverde, A., Ibancovich-Camarillo, J. A., Sánchez-Aparicio, P., Recillas-Morales, S., Osorio-Avalos, J., Rodríguez-Velázquez, D., & Miranda-Cortés, A. E. (2020). Clinical evaluation of postoperative analgesia, cardiorespiratory parameters and changes in liver and renal function tests of paracetamol compared to meloxicam and carprofen in dogs undergoing ovariohysterectomy. *PLoS One*, 15(2), e0223697.
- Johnson, R. E., Fudala, P. J., & Payne, R. (2005). Buprenorphine: Considerations for pain management. *Journal of Pain and Symptom Management*, 29(3), 297–326.
- Kore, A. M. (1990). Toxicology of nonsteroidal anti-inflammatory drugs. *The Veterinary Clinics of North America. Small Animal Practice*, 20(2), 419–430.
- Kandimalla, K. K., & Donovan, M. D. (2005). Localization and differential activity of P-glycoprotein in the bovine olfactory and nasal respiratory mucosae. *Pharmaceutical Research*, 22(7), 1121–1128. <https://doi.org/10.1007/s11095-005-5420-3>
- Ko, J. C., Freeman, L. J., Barletta, M., Weil, A. B., Payton, M. E., Johnson, B. M., & Inoue, T. (2011). Efficacy of oral transmucosal and intravenous administration of buprenorphine before surgery for postoperative analgesia in dogs undergoing ovariohysterectomy. *Journal of the American Veterinary Medicine*, 238(3), 318–328.
- Kavoi, B., Makanya, A., Hassanali, J., Carlsson, H. E., & Kiama, S. (2010). Comparative functional structure of the olfactory mucosa in the domestic dog and sheep. *Annals of Anatomy*, 192, 329–337.
- Leung, J., Beths, T., Carter, J. E., Munn, R., Whittam, T., & Bauguier, S. H. (2021). Intravenous acetaminophen does not provide adequate postoperative analgesia in dogs following ovariohysterectomy. *Animals*, 11, 3609. <https://doi.org/10.3390/ani11123609>
- Madsen, M., Enomoto, H., Messenger, K., & Papich, M. (2022). Effects of housing environment on oral absorption of acetaminophen in healthy beagles. *American Journal of Veterinary Research*, 83(1), 80–85. <https://doi.org/10.2460/ajvr.21.06.0075>
- Robertson, S. A., Taylor, P. M., & Sear, J. W. (2003). Systemic uptake of buprenorphine by cats after oral mucosal administration. *Veterinary Record*, 152(22), 675–678.
- Regina, K. J., & Kharasch, E. D. (2013). High sensitivity analysis of buprenorphine, norbuprenorphine, buprenorphine glucuronide, and norbuprenorphine glucuronide in plasma and urine by liquid chromatography mass spectrometry. *Journal of Chromatography B: Analytical Technologies in the Biomedical and Life Sciences*, 15(939), 23–31. <https://doi.org/10.1016/j.jchromb.2013.09.004>
- Steagall, P. V., Ruel, H. L. M., Yasuda, T., Monteiro, B. P., Watanabe, R., Evangelista, M. C., & Beaudry, F. (2020). Pharmacokinetics and analgesic effects of intravenous, intramuscular or subcutaneous buprenorphine in dogs undergoing ovariohysterectomy: A randomized, prospective, masked, clinical trial. *BMC Veterinary Research*, 16, 154–163.
- Santangelo, B., Harel, M., Fourel, I., Micieli, F., Cataldi, M., & Weisse, E. S. (2019). Intranasal dexmedetomidine in healthy beagles: An echocardiographic and pharmacokinetic/pharmacodynamic study. *The Veterinary Journal*, 251, 105346. <https://doi.org/10.1016/j.tvjl.2019.105346>
- Sohlberg, E., Halldin, M. M., Annas, A., Königsson, K., Jansson, B., Pehrson, R., & Borg, N. (2013). The impact of the site of blood sampling on pharmacokinetic parameters following sublingual dosing to dogs. *Journal of Pharmacological and Toxicological Methods*, 67(1), 1–4.

- Tuttle, R., Popescu, L., Hill, S., Slanczka, A., Jankowski, J., Barre, K., Krueger, E., Slade, D., Croutch, C., Robben, M., Mesa, Z., Mesa, M., & Dretchen, K. L. (2020). Intranasal epinephrine effects on epinephrine pharmacokinetics and heart rate in a nasal congestion canine model. *Respiratory Research*, 21, 78. <https://doi.org/10.1186/s12931-020-01343-x>
- Vlerick, L., Devreese, M., Peremans, K., Dockx, R., Croubels, S., Duchateau, L., Polis, I. (2020). Pharmacokinetics, absolute bioavailability and tolerability of ketamine after intranasal administration to dexmedetomidine sedated dogs. *PLoS One*, 15(1), e0227762. <https://doi.org/10.1371/journal.pone.0227762>
- Watanabe, R., Monteiro, B. P., Evangelista, M. C., Castonguay, A., Edge, D., & Steagall, P. V. (2018). The analgesic effects of buprenorphine (Vetergesic or Simbadol) in combination with carprofen in dogs undergoing ovariohysterectomy: A randomized, blinded, clinical trial. *BMC Veterinary Research*, 14(1), 304. <https://doi.org/10.1186/s12917-018-1628-4>
- Watanabe, R., Marcoux, J., Evangelista, M. C., Dumais, Y., & Steagall, P. V. (2020). The analgesic effects of buprenorphine (vetergesic or simbadol) in cats undergoing dental extractions: A randomized, blinded, clinical trial. *PLoS One*, 15(3), e0230079. <https://doi.org/10.1371/journal.pone.0230079>
- Wahler, B. M., Lerche, P., Pereira, C. H. R., Bednarski, R. M., KuKanich, B., Lakritz, J., Aarnes, T. K. (2019). Pharmacokinetics and pharmacodynamics of intranasal and intravenous naloxone hydrochloride administration in healthy dogs. *American Journal of Veterinary Research*, 80(7), 691–701. <https://doi.org/10.2460/ajvr.80.7.696>

How to cite this article: Enomoto, H., Love, L., Madsen, M., Wallace, A. & Messenger, K. M. (2022). Pharmacokinetics of intravenous, oral transmucosal, and intranasal buprenorphine in healthy male dogs. *Journal of Veterinary Pharmacology and Therapeutics*, 45, 358–365. <https://doi.org/10.1111/jvp.13056>