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A new method for determining levels of sedation in dogs: A pilot study with propofol and a novel neuroactive steroid anesthetic

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ABSTRACT

Background: Different levels of consciousness are required in order to perform different medical procedures. Sedation scales established to objectively define various levels of sedation in humans have not been thoroughly characterized in non-human species. Postural changes in rats or dogs are useful as gross measures of sedation but are inadequate for quantitative assessment since graded levels of sedation are difficult to delineate and obscured by movement abnormalities.

New method: A new canine sedation scoring (CSS) method was developed based on the modified observer's assessment of alertness and sedation score (MOAA/S) used in humans. The method employed a combination of physical, auditory and somatosensory stimuli of increasing intensity. Cardiovascular, respiratory, and a neurophysiological measure of sedation (bispectral index: BIS) data were recorded. Validation studies were performed following intravenous loading and constant rate infusion of propofol or a novel synthetic neuroactive steroid (SGE-746).

Results: Four levels of consciousness were identified: 1) Awake, 2) Moderate Sedation (MS), 3) Deep Sedation (DS) and 4) General Anesthesia (GA). Cardiorespiratory measurements obtained after bolus administration of propofol and SGE-746 and at the end of each CRI remained within normal limits. Canine sedation scores correlated with BIS for SGE-746. SGE-746 exhibited a more gradual exposure-response relationship than propofol. Larger increases in the plasma concentration from awake values were required to achieve different levels of sedation with SGE-746 compared to propofol.

Comparison with existing methods: No other canine sedation scoring methods are widely accepted.

Conclusion: A CSS method, based on the human MOAA/S scale defined four levels of consciousness in dogs and provided better resolution of sedation depth than BIS alone.

1. Introduction

Over 50 million medical procedures, including endoscopy (*e.g.,* colonoscopy, bronchoscopy), plastic surgery, and dental procedures, are performed in the US each year (Cullen et al., 2009). Therapeutic sedation is required for patients undergoing these procedures. The depth of sedation must be precisely targeted based on the type of procedure performed in order to minimize patient stress and discomfort, while avoiding the cardiorespiratory risks encountered when administering any sedative.

Most approved sedatives commonly used for short, out-patient procedures exhibit significant limitations (Sneyd and Rigby-Jones, 2010). Anesthetics, such as propofol, produce cardio/respiratory effects that often require an anesthesiologist for safe administration, which is not ideal for outpatient procedures requiring brief sedation time (Cooper et al., 2013). Alternative sedative/anxiolytics, such as midazolam, have improved safety profiles but require long recovery times and may not provide effective sedation (Antonik et al., 2012; Conway et al., 2016; Sneyd and Rigby-Jones, 2010). Given these limitations, there is continued interest in developing novel sedative compounds with predictable pharmacokinetic/pharmacodynamic properties that allow for graded levels of sedation/anesthesia required for a particular procedure and improve transition into and out of sedation.

Neuroactive steroids (NAS) are a class of compounds, some of which

Abbreviations: NAS, neuroactive steroid; CRI, constant rate infusion; MAP, mean arterial pressure; MS, moderate sedation; DS, deep sedation; GA, general anesthesia; BIS, bispectral index; MOAA/S, human modified observer's assessment of alertness and sedation; RR, respiratory rate

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possess sedative/anesthetic properties (Selye, 1941), with potent modulatory activity at GABA receptors. These compounds induce anesthesia by positively modulating central nervous system $GABA_A$ receptors (Majewska et al., 1986). A combination of two neuroactive steroids (alfaxalone and alphadolone), was first introduced as an anesthetic in both humans (Althesin®) and animals (Saffan®) in 1971. Neither product was marketed in the US and the human product was eventually removed from further development due to immunologic reactions most likely attributed to the excipient cremophor (Child et al., 1972, 1971; Hogskilde et al., 1987). The emergence of cyclodextrin excipients has renewed interest in developing novel products for use in humans and animals (Challa et al., 2005), due to their more favorable safety profiles. For example, alfaxalone formulated in aqueous 2-hydroxpropyl-β-cyclodextrin (HPBCD) was developed in 2001 and approved for veterinary use as a sedative-anesthetic in dogs and cats in Australia (Alfaxan-CD RTU) in 2012 and the United States in 2015 (Brewster et al., 1989; Estes et al., 1990). SGE-746 is a novel NAS sedative/anesthetic with similar in vitro and in vivo properties to a previously developed molecule, SGE-516 (Martinez Botella et al., 2015). SGE-746 is a potent positive allosteric modulator of both synaptic ($\alpha1\beta2\gamma2$) and extra-synaptic ($\alpha4\beta3\delta$) $GABA_A$ receptors, and possesses facile absorption and clearance, which would potentially support rapid emergence from sedation (Martinez Botella et al., 2015).

One of the challenges of developing novel sedatives is the lack of translatable animal models with sufficient dynamic range to establish a detailed dose-response effect. Current guidelines and objective measurements of physical signs (respiratory patterns, somatic muscle tone, ocular reflexes) or physiologic changes (respiratory rate, heart rate, arterial blood pressure) do not provide adequate criteria for differentiating discrete levels ("depths") of sedation in dogs (Bednarski et al., 2011). The loss of righting reflex in rodents and the induction of lateral recumbency in dogs are useful as gross measures of sedation, but do not discriminate graded levels of sedation. Moreover, dogs, a common species used for the preclinical development of novel sedatives and anesthetics, pose a specific challenge in that they are uniquely sensitive to transitions between awake and moderate degrees of sedation, often exhibiting movement abnormalities, paddling, and fictive running behaviors (Herron et al., 2008). This sensitivity, which has been observed with all currently available hypnotics (*e.g.,* propofol, etomidate, alfaxalone) including the ultra-short acting thiobarbiturate anesthetic thiopental, has a significant impact on determining the dose-response relationship for a given compound (Muir and Mason, 1989; Watney and Pablo, 1992). In the absence of an appropriate model for differentiating levels of sedation, it is difficult to rank a compound's sedative effects, and compounds are ultimately advanced into clinical trials based on pharmacokinetic parameters or surrogate endpoints, such as electroencephalographic (EEG) effects (Antonik et al., 2012; Upton et al., 2010).

We propose a new canine sedation scoring (CSS) method in order to differentiate different levels of consciousness in beagle dogs. This protocol paired changes in test article infusion rates with serial testing of responses to physical, auditory, and somatosensory stimuli of increasing intensity. Importantly, the method depended on rapidly achieving GA followed by stepwise reduction of the infusion rate for determining the doses and exposures required to achieve lesser degrees of sedation. This approach reduced the impact of species-specific and individual animal sensitivity to transitions from awake to moderate degrees of sedation. We hypothesized that our multiparameter CSS method would provide predictive utility for determining the dose-sedation response relationship for propofol and a newly developed neuroactive steroid (SGE-746) sedative-anesthetic. The primary goal of this study was to establish the multiparameter CSS method for evaluating sedation level in dogs.

2. Materials and methods

2.1. Compliance statement

All procedures were approved by the facilities Institutional Animal Care and Use Committee (IACUC). Specifically, these studies were conducted in accordance with the Animal Welfare Act and Regulations (as amended) and the National Research Council Guide for the Care and use of Laboratory Animals (Eighth Edition).

2.2. Dose-range finding pilot studies

Pilot studies were performed in three healthy male Beagle dogs (14.1 ± 0.5 kg, range: 12.2–15.6 kg) to determine the optimal dose range and associated plasma concentrations of both propofol and SGE-746 required to produce general anesthesia. These data were also used to establish initial bolus doses and continuous rate infusion (CRI) dose rates for propofol and SGE-746. At least a three-day washout period was maintained between dosing.

2.3. Study design

Canine infusion studies were performed in four healthy male beagle dogs (14.0 ± 0.6 kg, range: 11.0 to 16.0 kg). Beagle dogs were acclimated to the testing facility during a 7-day quarantine period. Each dog was surgically implanted with a Data Sciences International radio telemetry transmitter (TL11 M2-D70-PCT or TL11 M3-D70-PCTP, Data Sciences Int., New Brighton MN) at least 14 days prior to the study for the continuous collection of systemic arterial blood pressure, heart rate (derived from blood pressure waveform), ECG and body temperature. Indwelling venous catheters were transcutaneously placed in peripheral veins (e.g., cephalic; saphenous) for administration of the test articles and obtaining blood samples. Different venous catheters were used for drug administration and obtaining blood samples for determination of plasma drug concentrations. On the day of study, needle electrodes were placed subcutaneously at the base of the tail for electrical stimulation and on the dog's head for obtaining electroencephalographic signals (BIS, VISTA™ Monitoring System, Aspect Medical Systems, Natick, MA).

Baseline cardiovascular data were collected 10 min prior to test article administration and continuously throughout the study. General anesthesia was produced by administering propofol (6 mg/kg, n = 3) or SGE-746 (7.5 mg/kg, n = 5) over 60 s. Additional bolus doses of propofol and SGE-746 were administered to one dog in each group to meet the initial criteria for general anesthesia. Orotracheal intubation was performed with a cuffed tracheal tube and a CRI of propofol (400 μg/kg/min) or SGE-746 (576 μg/kg/min) was initiated. Behavioral assessments were performed at 2–3 min after the bolus dose and again at 45 min after the start of each CRI step. Each rate of infusion was continued for approximately 50 min followed by a step-wise 20% decrease in the CRI (Fig. 1).

2.4. Adverse events

Test article adverse events were recorded: 1) apnea for greater than 60 s or respiratory rate (RR) ≥ 30; 2) mean arterial pressure (MAP) ≤ 50 mm of mercury (mm Hg or ≥ 140 mm Hg; 3) heart rate (HR) ≤ 60 beats per minute (bpm) or ≥ 180 bpm; 4) end tidal carbon dioxide ($ETCO_2$, mm Hg) ≥ 60 mm Hg or blood oxygen saturation (SpO2%) < 90%; and, 6) cardiac arrhythmia.