This edition of the BRL Bulletin will provide an overview of large animal and nonhuman primate anesthetic guidelines at the University of Illinois at Chicago. The minimization of pain and distress in research animals is an ethical and legal obligation for investigators. Both the Animal Welfare Act and Public Health Service policy require that procedures causing more than momentary pain and distress be performed with appropriate sedatives, analgesics, and anesthetics unless an exemption is scientifically justified and approved by the IACUC. This Bulletin is intended as a resource for investigators, providing general guidelines for sedation, anesthesia, and analgesia. Because there may be significant variation in specific requirements a veterinarian should be consulted when designing large animal or nonhuman primate anesthetic protocols.

OVERVIEW OF ANESTHESIA

The major goals of balanced anesthesia are loss of consciousness, lack of movement, and prevention of pain. This can be accomplished in a multitude of ways. Planning an anesthetic event requires careful consideration of variables that include characteristics of the animal (species, age, health condition, etc.), the research objective, and the procedure. Drugs should be selected with the goal of providing balanced anesthesia. The side effects of drugs are often dose dependent. A balanced anesthetic protocol utilizes small amounts of multiple drugs in combination to optimize the desired effects while mitigating the negative effects associated with a larger dose of any individual drug.

When designing an anesthetic protocol for large animals or nonhuman primates, there are four general phases in which drugs are given in order to accomplish specific tasks: pre-anesthetic sedation, anesthetic induction, anesthetic maintenance, and postoperative analgesia. More invasive or painful procedures may necessitate use of drugs in all four phases, while minor or non-painful procedures often require a less complex anesthetic protocol. This Bulletin will examine each phase of anesthesia with a focus on commonly used drugs and the rationale behind their use.

PRE-ANESTHETIC SEDATION

Pre-anesthetic sedation reduces anxiety in the animal and facilitates restraint by handlers. In the case of nonhuman primates it is necessary for the safety of staff. Depending on the drugs used, pre-anesthetic sedation may result in muscle relaxation and immobility. For procedures that only require chemical restraint and do not involve significant pain, such as an IV infusion or imaging, this may be all that is needed. Pre-anesthetic sedation aids in a smoother induction and recovery, and decreases the amount of drugs needed for anesthetic induction. Sedation is accomplished using drugs that can be given subcutaneously or intramuscularly to a fully conscious animal. An IV catheter may be placed after animals are sedated so that subsequent drugs may be given intravenously.

Types of drugs given during this phase include dissociatives and alpha-2 agonists. Analgesics can be included as a component of pre-anesthetic sedation. This reduces the amount of anesthetic drugs needed to produce the desired level of anesthesia as well as reducing pain in the immediate postoperative period for short procedures.

Ketamine is classified as a dissociative anesthetic, referring to the seeming dissociation of an animal from its environment due to the interruption of CNS impulses. The animal may move its limbs spontaneously, the eyes remain open, and the laryngeal, pedal, and pinna reflexes remain intact. Benefits of ketamine include a wide margin of safety, with little effect on cardiac output and respiratory function. It is metabolized by the liver, so an animal with hepatic dysfunction will remain anesthetized for a relatively longer time period. Of note, ketamine may result in seizure-like activity and does not provide good visceral analgesia or muscle relaxation. It is the most common drug used for chemical restraint in nonhuman primates and may be used in other species including swine and rabbits.
Alpha-2 agonists are commonly administered with ketamine. While alpha-2 agonists do not cause a loss of consciousness independently, the two drugs in combination result in balanced anesthesia. The most commonly used drugs are xylazine and dexmedetomidine, with dexmedetomidine having more specific alpha-2 activity. These drugs work by inhibiting presynaptic calcium influx and neurotransmitter release, resulting in the desired effects of sedation, analgesia, and muscle relaxation. However, alpha-2 agonists cause a decrease in heart rate, blood pressure, and body temperature. Therefore, they are not the ideal agents to use when it is important to maintain cardiac output. Respiration often becomes shallow, which may result in poor oxygenation. One benefit of alpha-2 agonists is the availability of specific reversal agents. The reversal agents for xylazine are yohimbine and tolazoline, while the reversal agent for dexmedetomidine is atipamezole (Antisedan<sup>®</sup>). However, any of these reversal agents can be used to partially reverse the effects of either alpha-2 agonist.

Telazol<sup>®</sup> is an anesthetic/tranquilizer agent composed of tiletamine, a dissociative anesthetic related to ketamine, and zolazepam, a benzodiazepine, that can be given intramuscularly. It acts similarly to ketamine/diazepam and will induce anesthesia with muscle relaxation and mild to moderate analgesia. Telazol reduces heart rate and blood pressure and may reduce respiratory activity. It can also lead to severe hypothermia and animals may have prolonged recovery. Palpebral, laryngeal, and pharyngeal reflexes remain intact after administration, and eyes remain open necessitating the use of ophthalmic ointment. After reconstitution Telazol remains stable for four days at room temperature or 14 days when refrigerated. Telazol may also be combined with an alpha-2 agonist to increase the degree of muscle relaxation and visceral analgesia.

**ANESTHETIC INDUCTION**

Anesthetic induction utilizes drugs that cause unconsciousness and muscle relaxation, allowing an animal to be intubated and prepared for surgery. Depending on the anesthetic protocol additional drugs may not be needed following pre-anesthetic sedation if the animal has sufficient muscle relaxation to allow intubation. This is common in anesthetic protocols used in rabbits and pigs. In other circumstances induction drugs are given intravenously, which is common in dogs and nonhuman primates. If induction requires administration of additional drugs, propofol or a combination of ketamine and diazepam are used most commonly.

Propofol is a hypnotic drug that rapidly produces a short period of anesthesia with smooth, rapid recovery. It produces marked muscle relaxation, but does not provide analgesia sufficient for major surgery. The most problematic adverse effect associated with propofol is respiratory depression, and it may cause apnea when administered quickly. Therefore, propofol should not be given to an animal unless it is possible to support ventilation. Propofol rapidly redistributes to body tissues and must be given intravenously in order to reach anesthetic concentration in the brain.

While propofol produces rapid muscle relaxation and unconsciousness, it also has a very quick duration. If longer acting induction drugs are required or there are concerns about respiratory depression, a combination of ketamine and diazepam may be used. Diazepam is a benzodiazepine that can be used as an anxiolytic, muscle relaxant, hypnotic, appetite stimulant, and anti-convulsant. Benzodiazepines work by enhancing the effects of GABA and glycine, two of the main inhibitory neurotransmitters in the CNS. When combined with ketamine for anesthetic induction a rapid loss of consciousness and muscle relaxation will result. Laryngeal reflexes remain with ketamine/diazepam induction. Like propofol, these drugs must be given intravenously to produce a rapid loss of consciousness and muscle relaxation.

**ANESTHETIC MAINTENANCE**

The goal of anesthetic maintenance is to keep the animal at the desired plane of anesthesia for the duration of the procedure. When animals are anesthetized unconsciousness occurs at a lighter depth than a lack of pain perception. Therefore, it is important to supplement anesthetic drugs with analgesics for invasive or painful procedures.

Anesthetic maintenance can be accomplished with inhalant agents such as isoflurane, injectable agents either given as boluses or as a continuous infusion, or multiple agents in combination. During this phase the animal’s depth of anesthesia and physiologic stability are monitored and the anesthetic agents are titrated accordingly.
Anesthetic depth is monitored in large animals and nonhuman primates using palpebral reflex, muscle tone, and response to painful stimuli. The physiologic status of the patient is monitored during this time period using parameters that include heart rate, oxygen saturation, expired carbon dioxide levels, blood pressure, mucus membrane color, capillary refill time, and body temperature. The goal of anesthetic maintenance is to keep the animal at an appropriate plane of anesthesia without compromising physiologic stability.

Isoflurane is the primary agent used for anesthetic maintenance of large animal and nonhuman primate species at the BRL. Animals are intubated, and isoflurane is delivered using a precision vaporizer. Isoflurane can easily be titrated to alter anesthetic depth. While isoflurane is a convenient drug to use and appropriate for a wide range of species and procedures, it has the potential to cause serious adverse effects including a decrease in blood pressure due to dilation of peripheral blood vessels and depression of respiratory function. Isoflurane provides very poor analgesia. Therefore, an adjunct analgesic such as an opioid or nonsteroidal anti-inflammatory must be given prior to a painful stimulus.

**ANALGESIA**

Any procedure that would cause pain in a human should be assumed to cause pain in an animal and appropriate analgesics must be given accordingly. Analgesics can be given before, during, or after surgery. Early use of analgesics prevents wind-up pain, a condition where pain is perceived more intensely following repeated painful stimuli. With appropriate intraoperative use the amount of inhalant anesthetic required is decreased, thus minimizing the associated negative physiologic effects. Intraoperative analgesia also reduces the need for additional analgesic drugs later in recovery.

Analgesia can be provided using opioids, nonsteroidal anti-inflammatory drugs, or both in combination. Drug selection should be based on the expected degree of discomfort, duration of action, and compatibility with the research goal. A minimally invasive procedure may only require a nonsteroidal anti-inflammatory. As procedures become more invasive, multimodal analgesia, an approach that targets different pain pathways thus enhancing the overall analgesic effect, may be required for appropriate pain control. Multimodal analgesia commonly consists of an opioid combined with a nonsteroidal anti-inflammatory.

Surgical procedures that cause severe pain require strong opioid analgesics. Opioid drugs with short durations of action have limited use in the research setting outside of the operating room. However, several formulation are available with dosing intervals of once daily or less, making them attractive options for postoperative pain control. Commonly used opioids include hydromorphone, fentanyl, burpenorphine, and sustained release buprenorphine.

Hydromorphone is a semi-synthetic opioid with duration of 2–4 hours that can be used preoperatively, intraoperatively, or postoperatively for the relief of pain. It is a pure mu agonist which causes analgesia and antitussive activity, but may also lead to respiratory depression, sedation, emesis, and gastrointestinal effects including constipation or defecation. Hydromorphone, like other opioids, produces analgesia at lower doses and sedation at higher doses. Hydromorphone increases respiratory rate which manifests as panting in dogs. Its analgesic potency is about 5x that of morphine and is appropriate for animals undergoing procedures resulting in significant pain.

Fentanyl is a potent short-acting synthetic opioid with pure mu agonist activity. Fentanyl has a short duration of action. It should be administered as a continuous infusion for intraoperative analgesia or via a transdermal patch for postoperative analgesia. Fentanyl patches are not appropriate for providing intraoperative analgesia so an additional analgesic must be used. Absorption from a transdermal fentanyl patch can take up to 12 hours to reach therapeutic plasma concentration. As such the patch should be applied >12 hours prior to anesthetic recovery, or an additional opioid drug must be given until the patch has been adhered for 12 hours. Preparation of the skin and application of external heat can affect the absorption of fentanyl from a transdermal patch. Fentanyl is appropriate for animals undergoing procedures resulting in significant pain.

Buprenorphine (Buprenex®) is a synthetic opioid with duration of 8–12 hours. Buprenorphine is a partial mu agonist. It has a lower analgesic ceiling than pure mu agonists, but has a greater affinity for mu receptors resulting in a longer duration of
action. For this reason it is the opioid analgesic that is most commonly used in the research setting. Buprenorphine causes sedation at higher doses and may cause decreases in blood pressure and heart rate. Buprenorphine is appropriate for animals undergoing procedures resulting in moderate pain. Sustained release buprenorphine (Buprenex SR®) is formulated to provide at least three days of analgesia after a single subcutaneous injection and is commonly used in nonhuman primates. Sustained release buprenorphine may cause sterile granulomas, the potential for which varies based on species.

Nonsteroidal anti-inflammatory drugs (NSAIDs) are aspirin-like compounds having analgesic, antipyretic, and anti-inflammatory actions. They exert their effects through inhibition of the production of prostaglandins. NSAIDs are effective against mild to moderate pain, but do not relieve visceral pain. There is the potential for serious side effects, including gastrointestinal ulceration, delayed platelet clotting time, and renal dysfunction. The most commonly used nonsteroidal anti-inflammatory in the research setting is meloxicam (Metacam®). This drug is available in parenteral and oral formulations. One advantage associated with the use of meloxicam is that it has a long half-life and is dosed once daily. Meloxicam is indicated for mildly to moderately painful procedures. For procedures that are expected to cause severe pain, meloxicam is frequently administered with an opioid to achieve a greater degree of pain control than either drug alone.

Local anesthetic drugs can be used as an adjunct to analgesia in order to prevent pain sensation to a particular region. By infiltrating an incision line with a local anesthetic agent, the dose of systemic analgesic needed is decreased. With proper use, the addition of local anesthesia refines the analgesic regimen by keeping the animal comfortable during the immediate postoperative period. The most commonly used local anesthetic is bupivacaine. It works by blocking voltage-gated sodium channels in neural membranes, preventing nerve conduction. Bupivacaine is effective up to 6-8 hours but takes up to 20 minutes to block nerve transmissions.

ADDITIONAL INFORMATION
Pricing and availability of drugs can be discussed with BRL veterinary technical staff in conjunction with surgical scheduling and planning. For procedures requiring anesthesia that are approved to take place outside the BRL’s centralized facility, non-controlled drugs such as isoflurane, alpha-2 agonists, meloxicam, and local anesthetics may be purchased through the BRL surgery department. These items are regularly stocked and may be purchased after 1:00pm Monday through Friday. Please have the investigator name and BRL account number to purchase supplies.

Certain anesthetic and analgesic drugs are controlled substances due to their potential for abuse. These agents include ketamine, all opioids, and benzodiazepines. For more information on the BRLs controlled drug policy, please refer to: http://www.brl.uic.edu/?q=node/29#controlled%20drugs.

Guidelines for large animal and nonhuman primate anesthesia have been developed by the veterinary staff. These guidelines along with recommended anesthetic and analgesic regimens can be found on the BRL website at: https://www.brl.uic.edu/?q=anesthetic_guidelines. Please consult with a BRL veterinarian when developing specific anesthetic protocols for large animal and nonhuman primate protocols.

RESOURCES