

**1. PURPOSE**

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The intent of this Standard Operating Procedure (SOP) is to describe methods of assessing pain in fish, amphibians and reptiles, and mitigating pain by administration of analgesic medications.

**2. RESPONSIBILITY**

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Principal investigator (PI) and their research staff, veterinary care staff.

**3. GENERAL CONSIDERATIONS**

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- 3.1. A procedure which would be expected to be painful if it were done on humans must be considered painful to the animal.
- 3.2. When there is a question of whether or not a procedure is painful, the animal should receive the benefit of analgesia.
- 3.3. Analgesia should be provided at an appropriate dose and frequency to control pain.
- 3.4. Any deviation from this procedure must be justified by the investigator and approved by the appropriate Facility Animal Care Committee (FACC).

**4. PAIN RECOGNITION AND ASSESSMENT**

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- 4.1. Adapt the frequency of observation to the protocol (minimum once a day).
- 4.2. Start by observing the animal from a distance so the animal's behavior is not altered by the presence of the observer. Then proceed to observe the animal more closely.
- 4.3. Look for any changes in the behavior. Report animals which appear to be in pain to the veterinary care staff.

**Note:** The most reliable signs of pain and distress are the changes in behavior. This implies a good knowledge of species and individual normal behavior by the observer.

- 4.4. Fish, amphibians and reptiles do not exhibit obvious clinical signs of pain. Because fish and amphibians can experience pain as mammals do, the assumption is made by extrapolation from human observation.

**5. ANALGESIA PLAN**

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- 5.1. If possible, provide analgesia before the painful stimulus, as it is more effective in preventing pain (e.g. give analgesic before surgery).
- 5.2. Try to use a combination of analgesics, which is often more effective than using a single agent.  
For example, combination of buprenorphine, carprofen, and local infiltration of a local analgesic.
- 5.3. For surgical procedures, extend analgesia from pre-op to 72 hours post-op, unless specified otherwise in the Animal Use Protocol (AUP) and approved by the FACC.

## 6. LOCAL ANALGESIA

- 6.1. Infiltrate or apply local analgesic to areas where a painful stimulus may be induced. Repeat application of local agent at specified intervals to maintain analgesia. In some cases a sedative is recommended when using local analgesia.

Analgesic	Dose	Duration	Note
Lidocaine	< 2 mg/kg	30–60 min.	Use lidocaine HCl 2% (20mg/ml) injectable solution. Because this drug is acidic, it is recommended to dilute it 3:1 with sodium bicarbonate injectable solution (at 5 or 8.4%). Dilution with sodium bicarbonate is not necessary if lidocaine is to be administered to an anesthetized animal. Dilution must be prepared immediately before use and should not be stored. Diluted solution is as effective but induction of analgesia is slightly prolonged.
Bupivacaine	< 2 mg/kg	3–4 hr.	Use bupivacaine HCl 0.50% (5mg/ml) injectable solution. Same comment as for lidocaine.
* Lidocaine-bupivacaine mixture	< 2 mg/kg	30min. to 4 hrs.	Same comment as for lidocaine. Combining both drugs allows for rapid induction and prolonged effect. Discard mixture after 3 months.
EMLA cream	Thick spread	30–60 min.	Apply a thick layer of cream ideally 10 minutes before the painful procedure.

\*most commonly used

## 7. GENERAL ANALGESIA

### Fish

Analgesic	Dose	Route	Frequency	Note
*MS222	25-300 mg/L	immersion	immersion	
Morphine	10 mg/ L	immersion	immersion	Controlled drug.

### Amphibians

Analgesic	Dose	Route	Frequency	Note
Buprenorphine	75 mg/kg	SC	> 4 hr.	Controlled drug.

### Reptiles

Analgesic	Dose	Route	Frequency	Note
*Buprenorphine	0.01 mg/kg	IM	immersion	Controlled drug.
*Carprofen	2-4 mg/kg	PO, SC, IM		Followed by 1-2 mg/kg q24-72 hr.
Ketoprofen	2 mg/kg	SC, IM	q24-48 hr.	
Meloxicam	0.1-0.2 mg/kg	PO, SC, IM	q24 hr.	

\*most commonly used

- 7.1. Administration of non-steroidal anti-inflammatory drugs (NSAIDs):
- 7.1.1. NSAIDs include carprofen, ketoprofen and meloxicam.
  - 7.1.2. Ensure good water intake and monitor hydration status during the treatment period.
  - 7.1.3. Suspend water restriction prior to administration of NSAIDs.
  - 7.1.4. To minimize chances for adverse drug interactions, a washout period of 5-7 days is recommended before switching between NSAIDs.

## SOP REVISION HISTORY

DATE	PREVIOUS VERSION	NEW VERSION
2015.04.22	6.1 (NO TEXT)	6.1 Use lidocaine HCl 2% (20mg/ml) injectable solution.
2015.04.22	6.1 (NO TEXT)	6.1 Use bupivacaine HCl 0.50% (5mg/ml) injectable solution.
2015.04.22	6.1 (NO TEXT)	6.1 Lidocaine-bupivacaine mixture: <b>Discard mixture after 3 months.</b>
2016.09.06	7. (NO TEXT)	7. Carprofen, ketoprofen and meloxicam: <b>Ensure good water intake and monitor hydration status.</b> <b>Suspend water restriction prior to administration.</b>
2016.09.06	5.2 For example, administer a combination of buprenorphine, ketoprofen, and local infiltration of lidocaine	5.2 For example, administer a combination of buprenorphine, <del>ketoprofen</del> <b>carprofen</b> , and local infiltration of <del>lidocaine</del> <b>a local analgesic.</b>
2017.01.27	7.1 (NO TEXT)	<b>7.1. Administration of non-steroidal anti-inflammatory drugs (NSAIDs):</b> <b>7.1.1. NSAIDs include carprofen, ketoprofen and meloxicam.</b> <b>7.1.2. Ensure good water intake and monitor hydration status during the treatment period.</b> <b>7.1.3. Suspend water restriction prior to administration of NSAIDs.</b> <b>7.1.4. To minimize chances for adverse drug interactions, a washout period of 5-7 days is recommended before switching between NSAIDs.</b>