



1. PURPOSE

This Standard Operating Procedure (SOP) intends to describe methods of assessing pain in rabbits and mitigating pain by administering analgesic medications.

2. RESPONSIBILITY

Principal investigator (PI) and their research staff.

3. GENERAL CONSIDERATIONS

- 3.1. A procedure expected to be painful in humans is considered painful in animals.
 - 3.2. When there is a question of whether a procedure is painful, the animal should benefit from 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 BGU ethical committee.
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4. PAIN RECOGNITION AND ASSESSMENT

- 4.1. Adapt the observation frequency to the procedure's invasiveness (minimum once a day).
- 4.2. Start by observing the animal from a distance, so the observer's presence does not alter the animal's behavior. Then proceed to observe the animal more closely.
- 4.3. Look for any changes in the behavior. Report animals that appear to be in pain to the vets.
- 4.4. Common clinical signs of pain or distress include (but are not limited to): avoidance, vocalization, aggressiveness, teeth grinding, isolation from the group, low spontaneous activity, hunched posture, lack of grooming, reduced appetite, and weight loss, and no feces in pen.

Note: *The most reliable signs of pain and distress are changes in behavior and no manipulation of enrichment materials.*

- 4.5. The Rabbit Grimace Scale (Keating et al. 2012): A poster demonstrating the scale is posted in Appendix 1.
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5. ANALGESIA PLAN

- 5.1. Specify the analgesia plan in your animal protocol.
- 5.2. Provide analgesia just before the painful stimulus whenever possible, as it is more effective in preventing

pain (e.g., give analgesic before surgery).

5.3. Use a combination of analgesics, often more effective than a single agent.

5.4. Extend analgesia from pre-op to 72 hours post-op for surgical procedures unless specified otherwise in the Animal Use protocol and approved by the BGU Ethical committee.

6. LOCAL ANESTHESIA

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

Analgesic	Dose	Route	Duration	Note
Lidocaine	< 2 mg/kg	SC, Infiltration of surgical wounds	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 must be prepared immediately before use and should not be stored. A diluted solution is as effective, but induction of analgesia is slightly prolonged. *Dilution with sodium bicarbonate is unnecessary if lidocaine is administered to an anesthetized animal.
EMLA cream	Thick spread	Topical	30–60 min.	Apply only to intact skin. Shave or pluck the fur. Ideally, 10 minutes before the painful procedure
Localine eye drops	1-2 drops	Ocular	30-60min	
Bupivacaine	< 2 mg/kg	SC, Infiltration of surgical wounds	3–4 hrs.	Use bupivacaine HCl 0.50% (5mg/ml) injectable solution. Same comment as for lidocaine.

7. SYSTEMIC ANALGESIA

7.1. Administration of non-steroidal anti-inflammatory drugs (NSAIDs):

7.1.1. NSAIDs include carprofen, ketoprofen, meloxicam, and dipyrone.

7.1.2. Ensure good water intake and monitor hydration status during the treatment period.

7.1.3. Suspend water restriction before administration of NSAIDs.

7.1.4. Do not administer NSAIDs to neonatal rodents.

Rabbit

Analgesic	Dose	Route	Frequency	Note
Buprenorphine	0.1 mg/kg	SC	4–8 hrs.	Mild to moderate pain. Controlled drug.
Buprenorphine long-acting	0.65 mg/kg	SC	72hrs	Mild to moderate pain. Controlled drug.
Carprofen	2 mg/kg	SC, PO	12–24 hrs.	Mild to moderate pain. Use carprofen 50mg/ml injectable solution. To prepare a 4mg/ml dilution: add 0.8 ml of carprofen 50mg/ml to 9.2 ml of sterile water for injection. Administer 5µL/g of body weight. Store at room temperature. Discard dilution after one month.
Meloxicam	1.5 mg/kg	SC, PO	24 hrs.	Mild to moderate pain.
Ketoprofen	15 mg/kg	SC	12-24 hrs.	Mild to moderate pain.
Dipyrone	50 mg/kg	PO	In water bottle	Mild to moderate pain.

8. Appendix

Appendix 1: The Rabbit Grimace Scale



National Centre
for the Replacement,
Refinement & Reduction
of Animals in Research



The Rabbit Grimace Scale

Research has demonstrated that changes in facial expressions provide a means of assessing pain in rabbits.

This specific facial action units (shown below) comprise the Rabbit Grimace Scale. These action units increase in their frequency in response to pain-provoking tests and can form part of a broader assessment alongside other behavioural aspects of pain.

The action units shown only for albino rabbit animals. Each animal should be observed for a short period of time to avoid causing undue changes in facial expressions that are unrelated to the specific test(s).

	Action units		
	Not present "0"	Moderately present "1"	Obviously present "2"
Orbital tightening <ul style="list-style-type: none"> • Closing of the eyelid • Flattening of orbital area • A wrinkling may be visible around the eye 			
Cheek flattening <ul style="list-style-type: none"> • Flattening of the cheeks • Flattening of the cheeks when the rabbit is present • Cheeks may be flattened • The face becomes more angular and less rounded 			
Nostril shape <ul style="list-style-type: none"> • Nostril shape has become vertically elongated 'Y' rather than 'U' shape • Nostril air is moved down towards the chin 			
Whisker shape and position <ul style="list-style-type: none"> • Whiskers are curled away from the face to 'stand on end' • Whiskers curl and bend from relaxed downward curve • Whiskers are curled up in the same direction. When obviously present whiskers show discomfort 			
Ear shape and position <ul style="list-style-type: none"> • Ears become more tightly flexed / curled (more 'cupped' or 'stuck') • Ears curled back being towards the position of ears in facing towards the researcher • Flattening the head down to the back of the neck 			

Newcastle University is a member of the European Association of Animal Experimentation (EAAC) and the European Association of University Veterinarians (EAUV).
 For a full list of the Rabbit Grimace Scale, visit the website: www.nc3rs.org.uk/rabbit-grimace-scale
 The Rabbit Grimace Scale is a tool for assessing pain in rabbits. It is based on the observation of facial expressions that are associated with pain. The scale is used to assess the level of pain in rabbits and to guide the treatment of pain.

SOP 105 RABBIT ANALGESIA
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