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Overview

Clinical Management of Pain in Rodents

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The use of effective regimens for mitigating pain remain underutilized in research rodents despite the general acceptance of both the ethical imperative and regulatory requirements intended to maximize animal welfare. Factors contributing to this gap between the need for and the actual use of analgesia include lack of sufficient evidence-based data on effective regimens, under-dosing due to labor required to dose analgesics at appropriate intervals, concerns that the use of analgesics may impact study outcomes, and beliefs that rodents recover quickly from invasive procedures and as such do not need analgesics. Fundamentally, any discussion of clinical management of pain in rodents must recognize that nociceptive pathways and pain signaling mechanisms are highly conserved across mammalian species, and that central processing of pain is largely equivalent in rodents and other larger research species such as dogs, cats, or primates. Other obstacles to effective pain management in rodents have been the lack of objective, science-driven data on pain assessment, and the availability of appropriate pharmacological tools for pain mitigation. To address this deficit, we have reviewed and summarized the available publications on pain management in rats, mice and guinea pigs. Different drug classes and specific pharmacokinetic profiles, recommended dosages, and routes of administration are discussed, and updated recommendations are provided. Nonpharmacologic tools for increasing the comfort and wellbeing of research animals are also discussed. The potential adverse effects of analgesics are also reviewed. While gaps still exist in our understanding of clinical pain management in rodents, effective pharmacologic and nonpharmacologic strategies are available that can and should be used to provide analgesia while minimizing adverse effects. The key to effective clinical management of pain is thoughtful planning that incorporates study needs and veterinary guidance, knowledge of the pharmacokinetics and mechanisms of action of drugs being considered, careful attention to individual differences, and establishing an institutional culture that commits to pain management for all species as a central component of animal welfare.

Abbreviations and Acronyms: BW, bodyweight; CFA, Complete Freund adjuvant; ED, Effective dose; FI, Food intake; PONV, postoperative nausea and vomiting;SX, surgical procedure; SR, sustained release.

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Clinical management of pain in research rodents remains an important ethical and moral issue for IACUC, researchers, and veterinarians today. This is not surprising—pain management in human patients is still poorly characterized and under managed and remains one of the most common reasons that patients seek medical attention.[7,22] Some aspects of inconsistency in rodent pain management may be attributable to unknown effective dosages of drugs for different strains of mice and rats, as well as challenges in assessing pain and pain mitigation in these animals. However, our review of the literature revealed that a large proportion of the inconsistent provision of adequate pain relief stems from either explicit or inferred socio-zoologic bias of the research community. For example, several studies have examined the methods of peer-reviewed papers that were published in highly ranked scientific journals and that involved surgery on research animals.[26,43,174,199] Repeatedly, these studies demonstrate a significant underuse of peri-operative analgesics in mice and rats[26,174,199] in contrast to much better reported use of analgesics in large animal (that is, primate, dog, and pig) surgical studies.[43,44] Follow-ups with authors of publications not reporting use of analgesics in mice and rats for painful surgical procedures has not significantly altered these findings, suggesting that it is unlikely to be due to under reporting of analgesic administration.[174,199]

To begin to address rodent pain consistently in research settings, there must be fundamental recognition that all mammals, at the very least, have near identical nociceptive pathways and pain signaling mechanisms.[158] Affective and cognitive processing of pain occurs as much in mice and rats as in primates and dogs, meaning that mice and rats are not somehow 'less sentient' species.[127,193] Recognizing and admitting this simple concept should give IACUC, researchers, and veterinarians pause before submitting or approving research protocols that do not specify adequate pain relief for mice and rats. An appropriate question to reflect upon in every instance should be, "Would this protocol be approved in a dog or a primate under these same conditions?". This question would go a long way toward improving consideration for pain management in mice and rats in research settings. An additional challenge for pain management in laboratory animal science has been the lack of objective pain indicators for some species.[84,137] Ongoing research has begun to address these gaps, resulting in the development of validated pain assessment tools for mice and rats.

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Considerations for types of pain based on underlying mechanisms

Recognition that mice and rats experience pain as much as other mammals is an important consideration when evaluating the types of pain (chronic or acute) to be managed. In both human and veterinary medicine, there is recognition of the importance of properly managing and treating acute pain, for ethical reasons and to prevent the condition from evolving into chronic pain— which is a more difficult condition to treat.[97,158] Working estimates are not available for the type, intensity, and duration of pain experienced with different research animal models; however, much of the pain that occurs in induced models is caused by acute peri-procedural pain. This would include most surgical models, models in which animals are instrumented with catheters, implants or other devices, initial injections of irritating substances, such as carrageenan, and many tissue biopsy or invasive sampling methods.

Strict guidelines do not distinguish acute and chronic pain, consistent with recognition that pain occurs along a continuum.[28] In human medicine, acute pain is considered to last up to 7 d after an initial event, but this limit can be modified by the severity, extent, and type of injury, and acute pain may last upward of 30 d or longer.[114] The pathophysiology of pain initiation and subsequent inflammation has been described previously, with no evidence of any biomarkers that distinguish acute from chronic pain, except that central sensitization is more common in chronic pain.[170]

Although acute pain may have evolved to provide a protective response to the host, a key distinguishing feature between acute and chronic pain is the lack of any physiologic benefits derived from chronic pain. Chronic pain, particularly when persistent and unrelieved, can severely and negatively impact quality of life[29] as a result of the onset of chronic maladaptive stress and hypothalamic-pituitary-adrenal gland axis activation, disruption of sleep, decreased functional and immune system performance, and impairment of social interactions.[29] Thus, unless chronic pain is the object of scientific study, this should be between this state and be avoided or minimized by managing pain in its acute stage.

General approaches to clinical pain management in rodents

Multiple pharmacologic agents are available to manage pain in research animals. These agents have different mechanisms and duration of action as well as varying potencies for providing pain relief (see below). This permits the veterinarian, research team, and IACUC to tailor treatments, based on the invasiveness of a given procedure and its potential to cause pain. While the use of standard operating procedures is helpful to ensure consistent pain management in research facilities, it can be counterproductive to take a 'one-size-fits-all' approach to pain management in rodents, for example, if only a specific nonsteroidal antiinflammatory drug (NSAID) is used for all painful studies in a facility.

The World Health Organization (WHO) provides a stepped approach to pain management for humans that can be useful to consider when treating research animals (Figure 1).[210] Certain steps in this 'pain ladder' can be skipped if the level of pain following a procedure is anticipated to be more severe, but it is useful to consider the full range of pharmacologic (and nonpharmacologic) options available for managing pain in research animals. Use of a systematic approach such as this allows treatment to be titrated to the amount of pain expected and observed. This type of approach helps to avoid both under and overuse of pain medications, both of which can be harmful to veterinary patients.

Setting realistic goals for pain management in laboratory rodents

Given the stated difficulties in managing pain adequately in humans, it may not be realistic to assume that all pain can be effectively treated in research animals all the time. This further emphasizes the importance of having a thoughtful plan that is tailored to the procedures being conducted. The plan should include anticipation of pain, early treatment to minimize sensitization, and evaluation of individual animals for a response to therapy. Companion animal pain management guidelines, such as the "PLATTER" approach, can provide a useful approach for systematic management of pain in research animals (see Figure 2).[53] Consistent use of this tool by the veterinary team for both clinical cases and research protocols would help to ensure better pain recognition and mitigation in laboratory rodents. Further, this approach could be built into the institutional animal user training program to ensure consistency in analgesia management. Where objective scoring tools do not exist, close observation of animal behavior is necessary and should be conducted noninvasively by an individual familiar with normal preprocedure behavior of the specific animals.

For new procedures or models with unknown outcomes, it can be useful to conduct detailed individual assessments on a few animals and then generalize these findings to develop a robust scoring system and appropriate pain treatment plan for the larger cohort.[58] Because pain and response to treatment can differ between sexes of animals, between animals of different ages, and even between genetically similar animals,[115,154] each rodent should be monitored directly after treatment for signs of comfort and wellbeing that indicate a pain-free state. This includes evaluating normal postures, social interactions, grooming, nest-building (in the case of mice), general activity, and food and water intake.

Evidence-based analgesia in rodents

Numerous formularies provide dosing regimens for pain management in rodents. These regimens are primarily based on studies evaluating analgesic efficacy, but also draw from commonly accepted historical practices. Analgesiometry assays used in these studies included variations of the tail flick assay, paw withdrawal, and the hot plate test. While these assays provide some information, they primarily test withdrawal reflexes indicative of nociception, and lack the ability to fully capture the more complex experience and central processing associated with surgical pain.[1] More recent studies have attempted to assess pain more comprehensively rather than just nociceptive responses.[14] These include assays such as behavioral assessments, grimace scales, vocalizations, and nesting behaviors (for a review, see reference 205).

Most analgesics used for mitigating pain in rodents fall into one of a few classes: opioids (or opioid-like), NSAID or local analgesics. Commonly used agents include buprenorphine, tramadol, meloxicam, carprofen, ketoprofen, ibuprofen, acetaminophen, lidocaine, and bupivacaine. Table 1). provides recommended dosing regimens for mice, rats and guinea pigs for each of these agents based on commonly referenced texts and guidelines.[60,83,119] An extensive literature review of pharmacokinetics of these drugs was also conducted. Table 2 presents data on establishment of therapeutic levels of

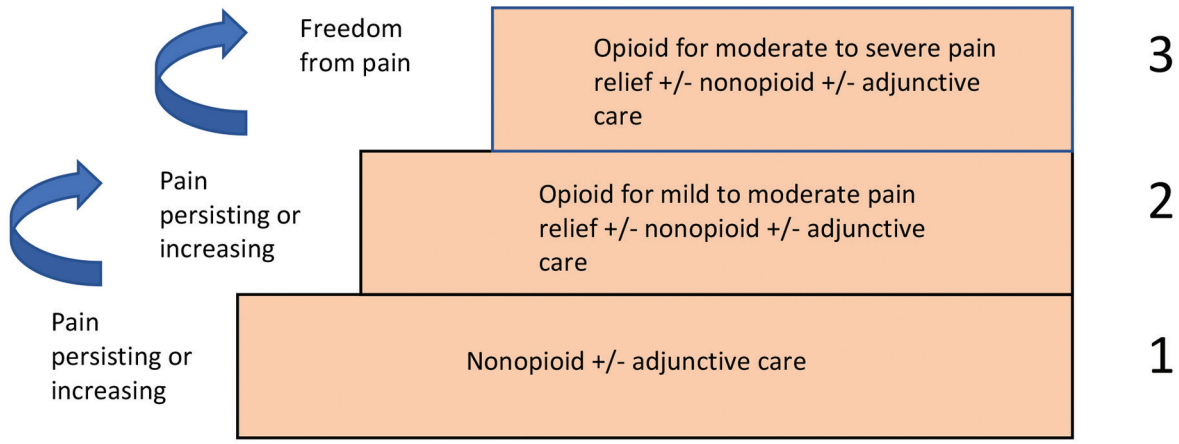

Figure 1. WHO's Pain Relief Ladder for Patient Management (modified from https://www.who.int/cancer/palliative/painladder/en/). This ladder models an approach for the veterinary clinician, IACUC, and research team to use for pain management in laboratory rodents, based on the anticipated level of invasiveness of procedures being conducted. For example, for a short recovery procedure, such as jugular vein cannulation being conducted by a skilled surgeon, the animal may require peri-operative NSAID treatment in addition to excellent postoperative nursing care. In all cases, regardless of the approved protocol or SOP, each patient should be assessed after the procedure to ensure that pain is being well managed. In the event that an animal appears uncomfortable, an escalation to the next higher level of care in the pain ladder should be considered.

*PL*an – every protocol or clinical case has a specific pain assessment and treatment plan

*A*nticipate – pain needs are anticipated, when possible, to minimize or prevent pain from occurring or, when pain is already present, to treat pain effectively

*T*rea*T* – appropriate treatments are given, considering the expected duration, severity, and type of pain

*E*valuate – treatment efficacy is evaluated using objective measures

*R*eturn – treatment is modified or discontinued based upon the clinical outcome

* Modified from 2015 AAHA/AAFP Pain Management Guidelines for Dogs and Cats (Epstein et al, 2015).

Figure 2. *PLATTER** Approach to Managing Pain in Research Animals.

commonly used analgesics. As shown, much of the literature has used "therapeutic levels" that are not based on well-proven studies in rodents, but rather extrapolated from other species, and the current dosing regimens do not appear to be based on achieving those therapeutic levels. The published ranges are also often very large, probably based on a range of analgesic efficacy from a small effect to a much more substantial dampening of pain responses. As such this information is currently of limited value and could benefit from more specific studies performed in rodents, but is included to provide currently available values.

An overview of the pharmacokinetic studies for mice, rats, and guinea pigs are summarized in Tables 3 to 5 respectively, and these are discussed in more detail below. The literature was further probed for efficacy studies, and results of these are summarized in a series of tables (Tables 6 to 12) , based on species (mouse, rat, or guinea pig) and analgesic drug or class (buprenorphine, nonopioid analgesics, and local anesthetics). The summary of the studies highlighted in these tables suggest, as discussed in more detail below, how the dosing regimens historically used for pain management in rodents may not be adequate,[66,105,112,113,130,149,163,211]

Buprenorphine

Buprenorphine, one of the most commonly used analgesics in rodents, is typically dosed subcutaneously twice a day, yet pharmacokinetic data demonstrate that mice and rats rarely achieve a plasma level greater than the purported therapeutic level beyond 4 to 6 h. Oral formulations provided continuously in feed or gels, and sustained-release formulations enhance the duration of action of buprenorphine. When buprenorphine is provided in MediGel or Nutella, the duration of effect can be up to 12 to 14 h [72,94]However, the mouse studies found considerable variation in the amount ingested.[94] Sustained-release formulations of buprenorphine regularly achieve a plasma level greater than therapeutic levels for more than 12 h, and often up to 24 h [37,103,112,202]However, manufacturer guidelines

Table 1. Common currently used analgesic dosing regimens for rodents

Species	Agent	Dose (mg/kg)	Route	Frequency
Mouse	Buprenorphine	0.05-0.1	SC	6-12 h
	Tramadol	5-40	SC, IP	ND
	Carprofen	2-5	SC	12-24 h
	Meloxicam	1-5	SC, PO	12 h
	Ketoprofen	2-5	SC	24 h
	Ibuprofen	30-40	PO	ND
	Acetaminophen	200	PO	ND
Rats	Buprenorphine	0.01-0.1	SC, IM	8-12 h
	Tramadol	5-20	SC, IP	ND
	Carprofen	2-5	SC	24 h
	Meloxicam	1-2	SC, PO	12-24 h
	Ketoprofen	2-5	SC	24 h
	Ibuprofen	15	PO	ND
	Acetaminophen	200	PO	ND
Guinea pig	Buprenorphine	0.05	SC	6-12 h
	Carprofen	2-5	SC, IM	12-24 h
	Meloxicam	0.1-0.3	SC, PO	24 h
	Ibuprofen	10	PO	4 h

ND = not determined.
Dosages drawn primarily from Flecknell 2016; Hawkins 2012; Kohn and colleagues 2007.

Table 2. Purported therapeutic plasma levels

Analgesic	Therapeutic plasma level (ng/mL)	Species studied	Reference
Buprenorphine	1.0	Human, rat	79
Carprofen	20,000-24,000	Human, cat, dog, in vitro	132
Ketoprofen	2,000-10,000	Human, rat	195
Meloxicam	390-911	Cat, dog, in vitro	69,102,131
Tramadol	100	mouse	56

(Zoopharm, Windsor, CO) suggest that dosing once every 72 h is sufficient. These findings suggest that the commonly used twice-daily dosing schedule of buprenorphine does not achieve an adequate duration of analgesia. Efficacy studies in rodents support these findings as they infrequently achieve clinical analgesia beyond 8 h, unless sustained-release formulations are used.

Nonsteroidal antiinflammatory drugs (NSAID)

The pharmacokinetics of nonopioid analgesics demonstrate similar pharmacokinetic and efficacy trends as buprenorphine. The commonly used dosages of NSAID in rodents fail to routinely provide plasma levels greater than therapeutic levels. Carprofen given at 5 mg/kg SC to mice has a duration of effect for 12 h;[112] however, the common dosing interval can be up to once daily. Efficacy studies demonstrate a minimal effect beyond the first 6 h postoperatively.[176] While carprofen administered in the drinking water achieved sustainable therapeutic levels up to 35 h, the study did not evaluate the efficacy of this route of administration in a postoperative model.[96] Meloxicam at 1 mg/kg SC in mice has a duration of effect of 4 h,[112] and when given orally at 10 mg/kg has a duration of action of 4 to 6 h.[21] However, when given at a higher oral dose of 20 mg/kg or in a sustained release formulation meloxicam has a duration of effect lasting up to 24 h.[96,112] Mice provided meloxicam in the drinking water refused to consume it.[96] Efficacy studies of meloxicam support pharmacokinetic studies in that 5 mg/kg appears to have no effect on postoperative analgesia in mice and clinically higher doses up to 20 mg/kg may be required for analgesia in mice.[130,212]Other NSAID have demonstrated similar findings of shorter duration of action in mice, which may be overcome with higher doses, such as ketoprofen at 10 to 20 mg/kg.[140] Doses of 1 or 4 mg/kg SC appear to be similarly ineffective in guinea pigs.[49,163] However, in rats, a 2 mg/kg SC dose reduced behavioral signs of pain in a laparotomy model.[160]

Local anesthetics

Local anesthetics have a short duration of action; 30 min with lidocaine and up to 60 min with bupivacaine. There are formulations that prolong the analgesic efficacy of local anesthetics, and these formulations can increase the duration of action to 24 to 48 h.

Recommendations

Dosing regimens for these analgesics should be carefully reconsidered in light of recent pharmacokinetic and efficacy studies . The frequency of dosing should be based on these pharmacokinetic studies as well as cage-side clinical assessments of pain, although clinical assessments should consider the ability of rodents to mask signs of pain. Table 13 provides our updated recommendations that address the inadequate dosing intervals that are widely used (and currently considered acceptable practice by many IACUC).[61] Given the inconsistent findings associated with the efficacy studies on NSAID, the dosing regimens recommended in Table 13 are based on current studies using more recent techniques to identify pain, such as

Table 3. Pharmacokinetics of analgesics used in mice

Analgesic	Dose (mg/kg)	Route	T_{max}	C_{max} (ng/mL)	Duration of action	Reference
Buprenorphine	5 µg/mL	PO-M	1 h	7.8	< 6 h	94
	15 µg/mL	PO-M	1 h	3.0	12 h	94
	0.03	SC	1 h	0.5	N/A	37
	0.05	SC	1 h	0.5	N/A	37
	0.1	SC	8 h	8.6	12 h	19
	0.1	SC	3 h	1.3	N/A	37
	0.1	SC	1 h	1.5	< 6 h	94
	0.1	SC	2 h	1.5	4 h	103
	0.6	SC	2 h	19.1	4 h	112
	2.0	SC	1 h	20.2	12 h	37
Buprenorphine SR	0.1[zp]	SC	4 h	14.5	24 h	112
	0.3[zp]	SC	6 h	0.8	N/A	37
	1.2[zp]	SC	0.5 h	5.0	12 h	37
	2.2[ih]	SC	2 h	11	24 h	103
	3.25[ag]	SC	6 h	16.3	72 h	202
	4.0[ih]	SC	24 h	ND	72-96 h	80
Carprofen	10	PO-G	2 h	20,300	N/A	96
	10	PO-W	12 h	17,000	N/A	96
	30	PO-W	24 h	32,000	N/A	164
	5	SC	2 h	525,000	12 h	112
Meloxicam	10	IV	5 min	365,000	4-6h	21
	10	PO	0.7 h	18,000	4-6h	21
	20	PO-G	4 h	16,700	24 h	96
	1	SC	2 h	4700	4 h	112
Meloxicam SR	6[zp]	SC	2 h	7300	12-24 h	112
Tramadol	25	IP	0.08 h	3010	4h	56
	25	IV	0.25 h	3710	2h	56
	25	PO-G	1 h	347	2 h	56
	25	PO	1 h	347	constant in water	56
	25	SC	0.25 h	1870	6 h	56
EMLA	18 mg/25g	Top	0.5 h	165	100 min Toxic at 21.2 mg/kg	6
	18 mg/25g	Top (open wound)	0.5 h	909	100 min	6
Bupivacaine	150 µL 0.5%	SC	1	1,000,000	approximately 4 h Toxic at >0.5%	73

Duration of action = time at which plasma level falls below therapeutic level (see Table 2)
N/A = plasma level did not exceed therapeutic level; ND = not determined
T_{max} = time to reach maximum concentration; C_{max} = maximum concentration
PO-W = oral in water; PO-M = oral in MediGel; PO-G = oral by gavage; Top = topical; SR= sustained release;
z*P* = manufactured by Zoopharm, Windsor, CO; ih= inhouse formulation; ag= manufactured by Animalgesics Laboratories, Millersville, MD

facial grimace score, and pharmacokinetic studies. Although several studies have evaluated voluntary ingestion of medical gels or feedstuff, routine use requires caution as rodents will reduce feed and water intake during the postoperative period and voluntary ingestion can be variable, resulting in inadequate dosing.

Multimodal analgesia

Another aspect of analgesic therapy that may overcome the current dosing challenges is multimodal analgesia. Multimodal analgesia combines multiple analgesics with different mechanisms of action into the treatment regimen, which often results in an increased efficacy while using lower dosages of the individual agents. Multimodal analgesia is commonly used in human and veterinary medicine for pain management.[12,13,17,42,50,126] Evidence that multimodal analgesia is effective in rodents is summarized in Table 14. In a tail-flick assay, the effects of ibuprofen were enhanced with opioids.[217] The effective dose of gabapentin and tramadol were both reduced when given in combination in a diabetic neuropathy model evaluating analgesia using the tail-flick assay, hot plate, and formalin test.[153] Similarly, the analgesic effect of tramadol was improved when ketoprofen was given concurrently using the writhing test, tail-flick assay, and formalin test.[150,152] **O**pioids also enhance the effects of tramadol.[59,175] In a murine laparotomy model, mice were treated with either buprenorphine alone or in combination with carprofen, administered in the drinking water.[164] The combination of buprenorphine and carprofen provided the best analgesia, compared with buprenorphine alone, and carprofen alone failed to provide any analgesia. A similar study was performed in a guinea pig ovariohysterectomy

Table 4. Pharmacokinetics of analgesics used in rats

Analgesic	Dose (mg/kg)	Route	T_{max}	C_{max} (ng/mL)	Duration of action	Reference
Buprenorphine	0.05	SC	0.5 h	1.5	2 h	72
	0.1	SC	4 h	2.7	8-24 h	64
	0.4	PO-N	2 h	1.25	14 h	72
	0.9 SR^{zp}	SC	4 h	2.8	24-48 h	64
	1.2 SR^{zp}	SC	4 h	2.8	24 h	64
	1.2 SR^{zp}	SC	24 h	1.01	24 h	160
Ketoprofen	2.5	IV	<5 min	10,000	48 h	181
	10	IV	<5 min	100,000	24 h	181
	3.2	PO	0.5 h	2730	24 h	143
	10	PO	0.5 h	11,700	90-360 min	4
	0.5	SC	ND	0.73	N/A	195
	1.0	SC	ND	1.79	N/A	195
	5.0	SC	ND	8.43	Measured at 2 h	195
Meloxicam	1	IV	< 0.25 h	5000	24 h	21
	0.3	PO	4.5-6.5 h	2300-3200	ND	21
Tramadol	20	IP	10 min	3187	300 min	186
	20	IV	< 10 min	23,314	300 min	186
Bupivacaine	2% 300 µL	SC	2 h	7000	Waned by 10 h	74

Duration of action = time at which plasma level falls below therapeutic level (see Table 2).
SR = sustained-release
N/A = indicates plasma level did not exceed therapeutic level
ND = not determined
T_{max} = time to reach maximum concentration
C_{max} = maximum concentration
PO-*n* = oral in Nutella
z*P* = manufactured by Zoopharm, Windsor, CO

Table 5. Pharmacokinetics of buprenorphine in guinea pigs

Dose (mg/kg)	Route	T_{max}	C_{max} (ng/mL)	Duration of action	Reference
0.2	IV	1.5 m	46.7	6 h	179
0.2	PO	1.2 h	2.4	3-6 h	179
0.05	SC	1 h	2.3	< 6 h	189
0.15 SR^{zp}	SC	1 h	2-2.3	6 h	216
0.3 SR^{zp}	SC	26 h	1.34	24-48 h	189
0.3 SR^{zp}	SC	1 h	6.9-11.5	48 h	216
0.48 SR^{ag}	SC	48 h	1.2	72-96 h	163
0.6 SR^{zp}	SC	1 h	64-71	72 h	216

Duration of action = time at which plasma level falls below therapeutic level (see Table 2).
SR = sustained-release formulation
T_{max} = time to reach maximum concentration
C_{max} = maximum concentration
z*P* = manufactured by Zoopharm, Windsor, CO; ag= manufactured by Animalgesics Laboratories, Millersville, MD

model.[163] Guinea pigs were treated at induction with an extended-release formulation of buprenorphine, carprofen or multimodal treatment. The frequency of behaviors indicative of pain was reduced in the multimodal treatment group compared with buprenorphine or carprofen alone.

Experiments assessing analgesic efficacy are challenging and complicated by species, strain, model, and environment. Nonetheless, studies evaluating alternative dosing regimens and multimodal therapies would further expand our knowledge base and provide better options for pain control. These studies must include proper control groups, including a "no treatment" group when not ethically precluded. However, sufficient data are available at this time to warrant the use of shorter dosing intervals for some of these drugs, and/or use of multimodal regimens. Many of the studies evaluating rodent pain have found that the most significant signs of pain occur within the first 12 to 24 h postoperatively. Multimodal therapies could be extremely beneficial during this critical postoperative time, including the administration of local anesthetic at the site of the incision, which could greatly reduce postoperative pain.[10,18]

Routes of administration

Administration of analgesic drugs to rodents must consider their small body size, stress associated with handling, the half-life of drugs, bioavailability, and factors that impact compliance with administration, such as difficulty in method of administration, time needed to administer the drug, and frequency of dosing required to achieve effective levels.

Table 6. Mouse efficacy studies of buprenorphine

Dose (mg/kg)	Route	Test	Duration of action	Comments	Reference
0.5-2	IP	HP, TF	105-135 min		3
0.5-6.8	IP	TF	ED70 at 0.5-2 mg/kg	Effective dose decreased with doses > 4.5 mg/kg	118
2.4	IP	SX	No effect	Dosed on day 1 and 7 postoperative	87
1	PO-N	SX		Reduced blood corticosterone	200
0.75	PO-F	HP, Lap	Up to 4 h	Suggest one SC dose followed by medicated feed for up to 20 h	155
4.2	PO-F	HP, Lap	Up to 4 h	Suggest one SC dose followed by medicated feed for up to 20 h	155
0.5-5.0	SC	HP, TF, WT		ED50 1.5 mg/kg	206
0.001-0.1	SC	Lap	Up to 90 min at 0.05-0.1 mg/kg		140
0.01, 0.05	SC	Lap		Partially effective at high dose	211
0.05	SC	Lap	5 h		148
0.05	SC	SX	Minimal effect	Dosed twice d for 2 d. Decrease BW, increase arterial pressure, decrease HR	173
0.1	SC	CLP	No effect		90
0.1	SC	HP, Lap	4 h	Dosed q8h for 24 h	103
0.1	SC	Lap	No effect	Dosed q12h for 3d	113
0.1	SC	Lap	No effect		124
0.1	SC	Lap, VF	2-8 h	Dosed q12h for 48 h. Suggest multimodal with carprofen	164
0.1	SC	SX	Partial efficacy to 12h	Dosed q12h for 3 d	203
0.25-5	SC	TF		ED50- 0.25 mg/kg ED30 1-5 mg/kg ED50- 10 mg/kg ED80- 50 mg/kg	171
0.3	SC	HP	No effect		25
0.5	SC	SX	No effect	Dosed q8h for 48 h	99
0.6	SC	SX		Low level pain up to 24 h	57
1.0	SC	HP	12 h		25
1.5	SC	HP, TF	4 h		88
2	SC	Lap	6 h	Dosed once, or q6h for 18 h. Increase in blood pressure at 6 h	70
2	SC	HP, TF	3-5 h		66
0.6 SR	SC	Lap	up to 24 h		113
1.0 SR	SC	CLP	24 h	Improved clinical score	90
1.5 SR	SC	HP, TF	up to 48 h		88
2.2 SR	SC	HP, Lap	24-48 h		103

Effects are based on a single dose of analgesic unless otherwise described in the comments.
PO-*n* = oral in Nutella; PO-F = oral in feed
CLP = cecal ligation and puncture; SX = surgical model; Lap = Laparotomy
HP = hot plate assay
TF = tail flick assay
VF = von Frey test
WT = writhing test
z*P* = manufactured by Zoopharm, Windsor, CO; ih= inhouse formulation

Parenteral administration

Parenteral routes remain the most common route of administration for analgesics. Based on retrospective reviews of analgesic administration reported in the literature, buprenorphine and carprofen are the most commonly used analgesics in rats and mice, and are most frequently administered subcutaneously.[91,199] Intraperitoneal and intramuscular injections have been reported but less commonly. Parenteral routes also offer more reliable and consistent rates of absorption and bioavailability, compared with oral administration.[204] While intraperitoneal injections might provide slightly faster absorption, subcutaneous injections are relatively easy for personnel to administer, can be performed with minimal and short-lasting restraint, and have less potential for adverse effects such as injection into an organ, and/or peritonitis. An often unrecognized characteristic of intraperitoneally administered substances is that absorption occurs largely through mesenteric vessels and are at least partially subject to first-pass hepatic metabolism.[136]

Buprenorphine, carprofen, and meloxicam, 3 commonly administered analgesics in rats and mice, are all available in injectable formulations but require dilution to be administered at appropriate dosages in mice. Carprofen and meloxicam were

Table 7. Mouse efficacy studies of nonopioid analgesics

Agent	Dose (mg/kg)	Route	Test	Duration of action	Comments	Reference
Acetaminophen	50	IP	Lap	1 h		149
	320	PO	SX		No effect on activity	87
	160,320	PO	CFA	Up to 90 min		156
	100-450	SC	Lap	No effect		140
Carprofen	30	PO-W	Lap, VF	In effective	Medicated water provided for 72 h	164
	5-25	SC	Lap	90 min at 20-25 mg/kg	Suggest 29 mg/kg	140
	5	SC	Lap		Burrowing latency similar to anesthesia alone	105
	5	SC	Lap		Activity and burrowing no different than anesthesia alone	104
	5	SC	Lap		Nest complexity improved slightly at high dose	106
	50					
Flunixin	2.5	SC	Lap	No effect		70
Gabapentin	1	IP	VF	3 h	Returned to baseline by 24 h	159
	3					
	3-100	IP	DN, FT, HP, TF		FT ED50 9.3 mg/kg	153
					HP ED50 16.5 mg/kg	
					TF ED50 17.6 mg/kg	
	50	IP	CCI, VF		ED50 7 mg/kg	45
Ibuprofen	200		TF	No effect		217
	40, 80	PO	CFA	150 min		156
	40	PO-W	SX		No effect on activity	87
	2.5-20	SC	CFA, VF		ED50 10 mg/kg	38
	200	SC	TF		In effective at 45 min	121
Ketoprofen			FT, WT		FT- ED50 100 mg/kg	68
					WT- ED80 10 mg/kg	
	30	IP	WT		ED50 30 mg/kg	151
	1-20	SC	Lap	90 min at 20 mg/kg	Suggest 65 mg/kg	140
Meloxicam	1	IP	HP, FT, WT		FT ED50 3 mg/kg, 10 mg/kg HP ED50 3 mg/kg,10 mg/kg	180
	3				WT- ED80 10 mg/kg	
	10					
	2	SC	Lap		Dosed once daily for 3 d. Reduced activity for 24 h postoperative	203
	2	SC	SX	Partially effective	Dosed with 2 mg/kg preoperative, then 1 mg/kg daily for 2 d. Improved BW, increase arterial pressures and HR	173
	5	SC	Lap	No effect		149
	5	SC	SX	No effect	Dosed once daily for 2 d	99
	5	SC	Lap	1 h		148
	5	SC	Lap		Corticosterone normalized at 20 mg/kg; All effective based on ethogram	212
	10					
	20					
	20	SC	SX	1 h	Reduced MGS and behaviors	130
Tramadol			FT, TF, WT		FT ED50 2.8 mg/kg	150
					TF ED25- 2.4 mg/kg	
					WT- ED50 1.86 mg/kg	
	20		Lap	No effect		123
	20	SC	SX	Minimal effect	Dosed daily for 2 d. Decrease BW, increase arterial pressure, decrease HR	173
	3-100	IP	DN, FT, HP, TF		FT ED50 3.5 mg/kg	153
					HP ED50 12.5 mg/kg	
					TF ED50 9.7 mg/kg	
	10-100	IP	CFA		ED50 25 mg/kg	152

Table 7. Continued

Agent	Dose (mg/kg)	Route	Test	Duration of action	Comments	Reference
	10-80	IP	HP	30-60 min	ED50 70 mg/kg	145
	10	IP	TF		Increased latency at 20 and 40 mg/kg	55
	20					
	40					
	50	IP	HP, TF	30-60 min	ED50 50 mg/kg; Trace minerals increased effectiveness	5
	40,80	PO	CFA	45-90 min		156
		SC	HP		ED50 14.8 mg/kg	175
					ED80 71.9 mg/kg	
	3.2	SC	WT		ED50 3.2 mg/kg	59

Effects are based on a single dose of analgesic unless otherwise described in the comments.
PO-W- oral in water; ED = effective dose; SX- surgical model; Lap- laparotomy
CLP = cecal ligation and puncture; CCI = chronic constriction injury
CFA = Complete Freund's adjuvant; DN = diabetic neuropathy; FT = Formalin test; HP = hot plate assay;
NP = neuropathic pain; PW = paw withdrawal test; TF = tail-flick assay; VF = von Frey test; WT = writhing test

Table 8. Mouse efficacy studies of local anesthetics

Agent	Dose	Route	Test	Duration of action	Comments	Reference
Bupivacaine	0.5%	Immer	TBX	No effect	Immersion for 30 s	48
	0.25% to 0.5% 50 µL	SC	HP, TF	5-15 min at 0.25 mg/kg 30-45 min at 0.5 mg/kg	Epinephrine at 1:200000 increased duration to 60 min	190
	0.5% 150 µL	SC	Electric	1-2 h		73
	10% in polymer	SC	HP, SNB	Up to 30 h		187
	333 mg/kg in polymer	SC	HP, SNB	Up to 48 h		192
	0.015% to 0.5% 150 µL	SC	Electric	15 min low dose; 60 min high dose		77
	0.12% 100 µL	SC	TF	30-45 min		191
	0.75% 20 µL	SC	TBX, TF	< 5 min	In effective for TBX	108
	1.1% 40 µL	SC	TF	45 min	Epinephrine increased duration to 80 min	75
	5 mg/kg	SC	Lap	Up to 60 min	Reduced mouse grimace scale	130
EMLA		Top	Tail vein injection	No effect		47
		Top	TBX	No effect		48
Lidocaine	2-4mM	Immer	TF	5 min		120
	0.5% 40 µL 1% 2%	SC	TF	5-30 min	Epinephrine at 1:200000 increased duration up to 100 min	76
	2% 20 µL	SC	TBX, TF	< 5 min	In effective for TBX	108

Effects are based on a single dose of analgesic unless otherwise described in the comments.
Immer = immersion; TBX = tail biopsy; SNB = sciatic nerve block; Electric = electrical stimulus

shown to be stable under a variety of environmental conditions (light compared with dark, and room temperature compared with 4 °C) for up to 7 d when diluted in reverse osmosis water.[96] Although this study evaluated oral administration, it provides evidence of the stability of these drugs, even after dilution.

Sustained-release formulations are increasingly available, and based on personal and listserv communications appear to be gaining widespread acceptance in the US. As early as 1994 investigators were exploring use of liposomal preparations to extend the duration of action of local anesthetics such as bupivacaine,[75] and systemic opioids such as morphine.[78] The first commercially available formulation of a systemically absorbed analgesic for use in rodents was Buprenorphine-SR-LAB (Zoopharm, Windsor, CO) and its use for analgesia in rats was first published in 2011.[64] Since that time, 14 other publications in rodents have included mice, rats, guinea pigs, and prairie dogs. Sustained-release meloxicam is also commercially available; however literature showing its efficacy and sustained plasma levels beyond 24 h in rodents are still lacking.[112,184] These sustained-release formulations, based on use of biodegradable polymers, offer many advantages including decreased handling (and thus stress) to the animal, decreased personnel time, and more consistent and sustained plasma and tissue drug levels, which decrease the potential for breakthrough pain that can occur if standard formulations are dosed too infrequently.[63] However, their use needs to be carefully considered and drawbacks weighed against their benefits. For example, current formulations require use of very small volumes for mice. This makes accurate dosing very challenging and over-dosing is a possibility. Also, absorption

Table 9. Rat efficacy studies of buprenorphine

Dose (mg/kg)	Route	Test	Duration of action	Comments	Reference
0.01	IM	Lap, TF	No effect	Dosed q12h for 72 h	41
0.1	IM	Lap, TF		Dosed q12h for 72 h. BW and food intake similar to saline treatment; TF increased latency	41
0.02-0.2	IP	TF	24 h at 0.2 mg/kg	Hyperalgesia at 0.01 mg/kg	207
8 µg/kg	IV	TF	4 h		161
0.4	PO	Lap	270-390 min	Observations limited to 390 min postoperative	176
0.5-10	PO-G	TF	2 h at 5-10 mg/kg		138
0.5	PO-G	HP	3-5 h		129
0.1-0.4	PO-J	Lap		Increased BW all treatment groups	62
0.5	PO-J	HP	1 h		129
0.5	PO-J	Lap		Dosed q12h for 36 h. Not effective based on BW	98
0.5- 2.0	PO-N	HP	60-120 at 1 mg/kg		92
0.4	PO-N	SX		No change in corticosterone; no change in activity 5h post op; BW loss less than control	72
0.3-3.0	SC	HP, TF		ED50 0.4 mg/kg	206
0.03	SC	Lap		Dosed q12h for 72 h. Decrease BW	20
0.03	SC	PW	24 h	Reduce RGS	133
0.05	SC	SX		Dosed q8-12h for 120 h. Improved gait	27
0.05	SC	HP, SX, VF	No effect	Dosed q12h for 72 h	34
0.05	SC	HP	1 h		107
0.05	SC	SX	No effect	Dosed preoperative and 18 h postoperative supplemented with 0.25 mg/kg POJ	117
0.05	SC	HP	3-5 h		129
0.05	SC		2 h		138
0.05	SC	HP		Dosed q12h for 60 h. PW latency increased; Minimal effect	141
0.05	SC	Lap	270-390 min postoperative	Observations limited to 390 min postoperative	176
0.05	SC	SX, VF, HP	Up to 96 h	Dosed q12h for 72 h. Reduced mechanical and thermal sensitivity.	184
0.05	SC	Lap		Lower ethogram score	160
0.1	SC	FT	6 h		1
0.1	SC	HP	30-240 min		92
0.1	SC	Lap		Dosed q12h for 72 h. Lower ethogram score	160
0.25-0.1	SC	VF		Increase threshold	196
0.2	SC	PW, SX		PW no effect at 24h; no effect on vertical rises	64
0.5	SC	SX		Increase corticosterone levels	72
0.5	SC	SX	No effect	Supplemented with 0.25 mg/kg POJ	117
0.5	SC	HP, TF	6-8 h		66
0.3 SR^{zp}	SC	HP, SX, VF	No effect		34
0.65 SR^{zp}	SC	HP	4-48 h		107
1.2 SR^{zp}	SC	HP, SX, VF		HP increase latency at 24h; VF no significant difference to baseline	34
1.2 SR^{zp}	SC	SX, PW	Up to 48 h	Increase vertical rises compared with buprenorphine	64
1.2 SR^{zp}	SC	HP	24-72h		107
1.2 SR^{zp}	SC	SX, VF, HP	Up to 96 h	Reduced mechanical and thermal sensitivity.	184
1.2 SR^{zp}	SC	Lap		Dosed q12h for 72 h. Lower ethogram score	160
4.5 SR^{zp}	SC	HP, SX, VF		HP increased latency at 24 h at 4.5 mg/kg; VF no effect; Sedative effect with 4.5 mg/kg	34

Effects are based on a single dose of analgesic unless otherwise described in the comments.
PO-*n* = oral in Nutella; PO-J = oral in gelatin; PO-G = oral by gavage
SX = surgical model; Lap = Laparotomy
HP = hot plate assay; PW = paw withdrawal test; TF = tail-flick assay; VF = von Frey test
z*P* = manufactured by Zoopharm, Windsor, CO

Table 10. Rat efficacy studies of nonopioid analgesics

Agent	Dose (mg/kg)	Route	Test	Duration of action	Comments	Ref
Acetaminophen	100, 300	PO	VF	No effect	Dosed daily for 2 d	196
	20-1000	PO	HP, TNT, VF	30-120 min at 100 and 1000 mg/kg	VF ED50 32.8 mg/kg	188
	4.48 mg/mL	PO-W	SX	No effect		27
Carprofen	2	PO-G	PW, VF	6-9 h		201
	5	PO-G	SX, VF, HP	Up to 48 h	Medicated feed provided 2 d preoperative and 2 d postoperative. Reduced mechanical pain, but not thermal.	184
	5	SC	Lap		Dosed preoperative and 4 and 24 h postoperative. Increased activity	23
	5	SC	Lap	270-390 min	Observation limited to 390 min postoperative	176
	5, 10	SC	CFA	No effect		166
Gabapentin	25-200	IP	FT		Effective at 100 and 200 mg/kg	157
	30-300	IP	CCI TF, VF		TF increase at 300 mg/kg; VF ED50 34 mg/kg; cold allodynia ED50 103 mg/kg	95
	5-20	IP	HP, VF		Increase thresholds 10-20 mg/kg	81
	300	PO	CFA	No effect		139
	30-300	PO-G	RS	1-4 h at 300 mg/kg 02-4 h at 100 mg/kg 3 h at 30 mg/kg		85
	10-100	SC	VF		Nominal effect at 100 mg/kg	167
	90	SC	TF	30-90 min		146
Ibuprofen	0.3-30	PO	CFA		No return to baseline gait	139
	20	PO	SX		Dosed q8-12h for 120 h. Improved gait	27
	31, 100	SC	CFA	WT bearing within 30-90 min;	Rearing increase at 100 mg/kg; Burrowing increased	178
Ketoprofen			HP, PW	6 h at 30-100 mg/kg		68
	3	IM	Lap, TF		Dosed q12h for 72 h. No effect	41
	3,5	IM	Lap		Dosed preoperative and 9-12 h postoperative. Reduced BW and FI; single and double dose have similar effect	40
	1,3.2,10	PO	HP	30-60 min	ED90 3.2 and 10 mg/kg	4
	0.5-10	SC	PW, VF		Guarding reduced 2-24 h at 5 and 10 mg/kg; no effect on PW or VF	195
	40	SC	Lap		Reduced RGS similar to morphine	111
Meloxicam	1	SC	Lap		Dosed daily for 3 d. Lower ethogram score; no difference from 2 mg/kg dose	160
	2	SC	Lap		Dosed daily for 3 d. Lower ethogram score	160
	2, then 1	SC	Lap		Dosed daily for 3 d. Improved BW, FI	20
	4.0 SR^{zp}	SC	SX, VF, HP	Up to 48 h	Reduced mechanical pain, but not thermal.	184
Naproxen	50-100	IP	CFA		Weight bearing increased at 30 min; increase burrowing	178
	50-150	IP	CFA		Effective at 50 mg/kg; higher dose no benefit	177
Tramadol	0.625-40	IP	HP, VF		ED50 10 mg/kg; ED80 40 mg/kg	144
	10	IP	HP		Dosed q12h for 60 h. In effective	141
	10-30	IP	HP		ED40 30 mg/kg	65
	10-40	IP	VF	15-30 min at 20 mg/kg; 15-120 min at 40 mg/kg		116
	11	IP	TF	75 min		218
	12.5	IP	Lap		Dosed preoperative and 4 and 24 h postoperative. No effect on activity, wheel running, BW	23

Table 10. Continued

Agent	Dose (mg/kg)	Route	Test	Duration of action	Comments	Ref
	1-25	IP	TF		Increase latency at 15 and 25 mg/kg; motor function impaired > 15 mg/kg	135
	4-50	IP	HP, TF		Increase latency at 12.5-50 mg/kg; heavy sedation > 25 mg/kg	24
	5-20	IP	CFA	60-90 min	Increase latency at 10 and 20 mg/kg	214
	5-40	IP	TF	30-120 min	ED50 20 mg/kg; ED80 40 mg/kg	100
	3-30	PO	HP, TNT, VF	30-120 min at 10 and 30 mg/kg	VF ED50 4.8 mg/kg	188
	4-50	PO-J	HP, TF	No effect		24
	0.45	SC	TF	30-90 min		162
	20	SC	FT		Reduced pain scores	67
	4-50	SC	HP, TF		Increased latency at 25-50 mg/kg; heavy sedation	24

PO-W = oral in water; PO-G = oral by gavage; PO-J = oral in gelatin;
SX = surgical model; Lap = Laparotomy; TNT = tibial nerve translocation; CCI = chronic constriction injury
CLP = cecal ligation and puncture; CFA = Complete Freund's adjuvant; DN = diabetic neuropathy; FT = Formalin test; HP = hot plate assay; RS = Randall-Selitto test; TF = tail-flick assay; VF = von Frey test; z*P* = manufactured by Zoopharm, Windsor, CO

Table 11. Rat efficacy studies of local analgesics

Agent	Dose	Route	Test	Duration of action	Comments	Ref
Bupivacaine		PN	SNB	7 h	Liposomal formulation increased duration to 21 h	54
	1-6 mg/kg liposomal formula	SC	VF	2 h		110
	2 mg/kg	SC	VF	—		110
	2% 300 µL	SC	VF	25 min		74
	2% liposomal Equation 300 µL	SC	VF	200 min		74
	5-15 mg/mL	SC	HP	120-200 min	Latency increased in dose dependent manner	93
Levobupivacaine	0.3% 50 µL	SC	SX	3-24 h		122
Lidocaine	2% 400 µL		HP, CCI		Reduced scratching behavior	15
	1.5-13.8 mmol/kg	SC	VF	15-30 min 13.8mmol	ED50 5.4 mmol/kg; ED75 8.0 mmol/kg	32
	2% 600 µL	SC	VF		ED50 0.13%	33
	4.4-62.2 mmol/kg	SC	VF	15-30 min at 62.2 mmol/kg	ED50 13.3 mmol/kg; ED80 36.7 mmol/kg	31
	2% gel	Top	TF	20 min		9
Pramoxine	12-120 mmol/kg	SC	VF	15-30 min at 120 mmol	ED50 42.1 mmol/kg; ED75 63.9 mmol/kg	32
Procaine	2% 600 µL	SC	VF		ED50 0.44%	33
Ropivacaine	2 mg/mL 300 µL	ID	Lap, VF	Up to 24 h	Less disturbed circadian rhythm, HR, BP	30

PN = perineural; SNB = sciatic nerve block; CCI = chronic constriction injury
ID = intradermal; Top = topical; HR = heart rate; BP = blood pressure; HP = hot plate assay; TF = tail-flick assay; VF = von Frey test

is variable and initial plasma concentrations can be quite high. Animals should be watched carefully during the first 4 to 8 h for signs of adverse opioid-induced effects, such as sedation, respiratory depression, and/or pica; however, other than pica in rats, other opioid-induced effects have not been appreciably seen in the authors' collective experiences. Lastly, the delay until an analgesic response is achieved must be factored into the pain management plan.

Regional anesthesia

Delivery of local anesthetics as a means of providing incisional or regional anesthesia and analgesia is a well-established and effective procedure. The relatively short duration of action and inability to redose in rodents has limited its utility to primarily 3 applications: (1) as part of a multimodal pain management plan, (2) as the sole pain management in minimally invasive procedures, such as small skin incisions for a subcutaneous implant, and (3) to provide some minimal analgesia when no systemic analgesia can be administered for scientific reasons.

Table 12. Guinea pig efficacy studies of buprenorphine, NSAIDs and local analgesics

Analgesic	Dose (mg/kg)	Route	Test	Duration of action	Comments	Reference
Buprenorphine						
	0.05	SC	RS	12-24 h	Dosed q12h for 72.	189
	1-5	SC	Pin prick		ED50 3.0 mg/kg; ED75 4-5 mg/kg at 30 min post administration	35
	0.3 SR[zp]	SC	RS	6 h		189
	0.48 SR[ag]	SC	Lap, VF	Up to 96 h	No change in behavior compared with analgesia only group	163
	0.6 mmol	IM	PW	4 h		213
Carprofen	1	SC	Lap, VF	Ineffective	Pain indices 2-8 h postoperative that resolved by 24 h	49
	4	SC	Lap, VF	Partially effective	Dosed daily for 3 d. Pain indices 8 h postoperative that resolved by 24 h	163
Meloxicam	0.2	SC	Lap		Dosed daily for 2 d. Received local bupivacaine and/or lidocaine. No effect.	52

Lap = laparotomy; PW = paw withdrawal assay; RS = Randall-Selitto test; VF = von Frey test;

Table 13. Updated analgesic dosing recommendations

Species	Agent	Dose (mg/kg)	Route	Frequency
Mouse	Buprenorphine	0.1-0.5	SC	4-6 h
	Buprenorphine SR[zp]	0.6	SC	48 h
	Tramadol	80	SC	24 h
	Carprofen	5	SC	12 h
		20	SC	24 h
	Meloxicam	5-10	SC	8-12 h
	Ketoprofen	20	SC	24 h
Rats	Buprenorphine	0.05-0.0.1	SC	6-8 h
		0.5-0.6	PO	24 h
	Buprenorphine SR[zp]	1.2	SC	48 h
	Tramadol	20-40	PO	24 h
		5	SC	24 h
	Carprofen	5	SC	24 h
	Meloxicam	1	SC	12-24 h
	Ketoprofen	5	SC	24 h
Guinea pig	Buprenorphine	0.05	SC	6 h
	Buprenorphine SR[zp,ag]	0.3-0.48	SC	48 h
	Carprofen	4	SC	12-24 h
	Meloxicam	0.2	SC	12-24 h

Modified from Flecknell 2018.[61]
SR = sustained release; # - provided in food treat, should be observed ingesting
z*P* = manufactured by Zoopharm, Windsor, CO; ag= manufactured by Animalgesics Laboratories, Millersville, MD.
Note: caution should be taken with higher doses of NSAIDs. Multimodal analgesia recommended to allow effective use of lower doses.

See Tables 8 and 11 for a summary of published efficacy studies in mice and rats respectively.

Oral administration

Bioavailability must be considered for any drug administered orally. Voluntary consumption will be variable between animals and both food and water consumption are often decreased after a surgical procedure.[8,87,197] If the drug is administered in a "treat" to encourage consumption, animals may need to be singly housed to ensure equal access and consumption. This could add another level of stress and an additional research variable. Absorption in the intestinal tract can be highly variable and affected by the amount of digesta in the tract, gastrointestinal motility, and other factors. The analgesics themselves may even impact GI motility.[125,165] Oral opioids are commonly used in humans but their primary use is for chronic pain, and there is a paucity of information on oral opioids in rodents. First pass metabolism is an impeding factor as opioids are degraded and lose a significant percentage of their bioavailability.

Oral gavage ensures exact dosing and delivery to all animals in the cohort. However, this method can be time consuming and the handling, restraint, and procedure itself may be stressful to the animals. Administration of analgesics in the drinking water is an attractive option and has been tested in a variety of paradigms in both mice and rats, but this method has numerous drawbacks to widespread use. Palatability and neophobia must be evaluated in each instance, as decreased

Table 14. Published multimodal analgesic efficacy studies

Species	Multimodal analgesics	Dose (mg/kg)	Route	Model	Comments	Reference
Mouse	Buprenorphine Carprofen	0.1 30	SC PO-W	Lap	Buprenorphine dosed q12h, carprofen medicated water provided for 72 h. Improved analgesia for 2-8 h postoperative	164
	Gabapentin	3-100	IP	TF, HP, FT	Reduced ED50 for each analgesic	153
	Tramadol	3-100	IP			
	Tramadol	10-100	IP	TF, HP, FT	ED50 reduced with Keto	152
	Ketoprofen	30-250	IP			
	Buprenorphine	0.05	SC	Lap	Buprenorphine dosed once preoperative. Melox was given 24 h postoperative	148
	Meloxicam	5	SC			
	Meloxicam	5	SC	Lap	No effect	149
	Acetaminophen	50	IP			
	Ibuprofen	200	IP	TF	Opioids enhanced latency	217
	Tramadol		SC	WT, HP	Opioids reduced ED50	59,175
Rat	Buprenorphine	0.03	SC	PW	Similar effect to buprenorphine alone	133
	Meloxicam	2	SC			
	Buprenorphine	0.05	SC	SX	Buprenorphine dosed q8-12h, meloxicam daily. No effect; 8 h dosing resulted in pica	183
	Meloxicam	2	SC			
	Acetaminophen	20-1000	PO	HP, VF	ED50 reduced of each	188
	Tramadol	3-30	PO			
	Carprofen	5	SC	Lap	Dosed preoperative and 4 and 24 h postoperative. Increased activity with tramadol	23
	Tramadol	12.5	IP			
	Gabapentin	5-20	IP	HP, VF	Potentiates opioids	81,146,162,167
	Tramadol	10	SC	HP	Tramadol dosed q12h for 60 h, gabapentin dosed daily. Minimal effect	141
	Gabapentin	80	SC			
	Tramadol	10	SC	SX	Tramadol dosed q8-12h and gabapentin dosed daily for 120 h, No effect	27
	Gabapentin	80	SC			
	Levobupivacaine	0.3% 50 µL	SC	SX	Enhanced with ibuprofen and epinephrine	122
	Ibuprofen	2 mg/mL 50 µL	SC			
	Lidocaine	22.6 mmol/kg	SC	VF	Increased threshold	31
	Naloxone	43.2 mmol/kg				
Guinea pig	Meloxicam	0.2	SC	Lap	No effect	52
	Bupivacaine	1	SC			
	Lidocaine	1	SC			
	Buprenorphine SR[ag]	0.48	SC	Lap	Improved analgesia compared with carprofen alone	163
	Carprofen	4	SC			

PO-W = Oral by water
ag= manufactured by Animalgesics Laboratories, Millersville, MD

water consumption will significantly impact the analgesic dosing.[16,194] Further, decreased consumption may compound an already negative hydration state due to the surgery and associated blood/fluid loss. The solubility of oral solutions is another consideration. Ibuprofen and acetaminophen in pediatric suspensions tend to settle out of solution and both are relatively insoluble in water.[63] A study evaluating rats given acetaminophen in drinking water found no difference in paw pressure latency compared with control rats and treated rats consumed less.[39] This same study also compared buprenorphine in drinking water to intramuscular injection. An increased latency response was measured in high dose buprenorphine (2.9 mg/kg/day equivalent to 0.02mg/ mL water) in drinking water comparable to that seen with IM buprenorphine, and neophobia was not seen. However, one group measured a decreased response to hot plate sensitivity in rats provided acetaminophen elixir at a concentration of 4.48 mg/mL in drinking water.[147] While consumption of acetaminophen treated water was greater than 50% less than tap water on Day 1, the neophobic response decreased

substantially by Day 2. In addition, rats drank significantly more acetaminophen the day after surgery compared with no-surgery controls. In a study by Ingrao and colleagues male C57BL/6 mice consumed carprofen willingly when diluted in their drinking water but not meloxicam.[96] Buprenorphine added to drinking water at 0.009 mg/mL (calculated to deliver approximately 10 times published subcutaneous doses) did not negatively affect volume of water consumed in female C57BL/6 mice, and resulted in therapeutic blood levels at many of the time points evaluated.[182] This differs from the results obtained from a study in rats in which a measurable neophobic response was seen.[101] Despite those encouraging results, interindividual differences in water consumption were seen as well as sporadic consumption during the daytime (light phase), resulting in variability in serum concentrations.

Delivery of analgesia by consumption in diet or a food treat has met with some success and offers the advantage of less stress on the animals since they do not need to be handled and restrained for dosing. Buprenorphine has been administered to rats in gelatin,[62,134] in hazelnut chocolate spread to rats and mice,[2,71,109] and in commercially available gels such as Medi-Gel (Clear H2O, Portland, ME) in mice.[94] Indeed, in some studies, oral consumption provided longer lasting blood levels of drug than subcutaneous injection,[109] for which the duration of action in mice is not long enough to provide continuous analgesia when dosed only every 8 to 12 h. However, consistent themes in all of these studies were variability in consumption, both in quantity and time of day that led the authors to conclude that these methods may be unreliable for provision of consistent and continuous analgesia. Further, in almost all of these studies animals were singly housed. NSAID have also been provided in "treat" forms, including carprofen-containing tablets (Rodent MDs, Bio-Serv, Frenchtown, NJ), and carprofen containing sucralose gel (MediGel CPF, Clear H2O, Portland, ME).

Another consideration for self-administration in water or food is the time of consumption. Mice and rats consume most of their feed and water during the dark cycle.[198] If surgery occurs in the morning of any given day, and the animals do not consume significant quantities of the medication until that night, they will lack pain management during the most crucial initial 12-h postoperative period. Therefore, beginning drug administration prior to the painful procedure (for example surgery) is recommended to overcome both the neophobic response and circadian rhythm impact on consumption to ensure that sufficient blood levels are attained preemptively.

Transdermal administration: Transdermal patches are effective for delivery of analgesia in humans and larger animals, but their practical application to rats and mice is so far limited. Two studies have evaluated the Buprederm patch (Samyang Pharmaceutical Center, Daejeong, Korea) in mice.[168,215] Analgesia, as measured by tail-flick latency, was most pronounced 3 to 6 h after application and an effect was measurable for 24 h.[215]

Timing of administration

The concept of preemptive analgesia is now well established in the pain management of human patients. A PubMed search conducted in December 2018 with keywords "pre-emptive" and "analgesia" produced 412 results. Many of these related to dental, spinal, and other orthopedic procedures. The clinical justification for preemptive analgesia is based on preventing central sensitization of nerve fibers by noxious stimuli occurring peripherally. This excitation results in a lowered pain threshold and hyperalgesia.[11,209] Indeed, a number of studies in humans have demonstrated that preemptive use of local anesthetics decreased the amount of analgesia required postoperatively and decreased hyperalgesia associated with some injuries.[46,51,142] Preemptive analgesia should provide similar benefits in animals by enhancing ability to ameliorate pain resulting in faster recovery periods. Preoperative administration of buprenorphine 30 min prior to surgery in rats resulted in less reduction in food intake than those given buprenorphine after surgery.[86] Preoperative administration of pethidine to rats undergoing ovariohysterectomy surgery prevented postoperative hyperalgesia.[128]

Analgesia should be administered preoperatively whenever short surgical periods are anticipated and an inhalant such as isoflurane or sevoflurane is the sole anesthetic used. The time to onset of action of the analgesic must be considered in planning time of administration. Even drugs given subcutaneously or intraperitoneally are expected to take 15 to 30 min to achieve therapeutic levels. Orally administered drugs will take even longer due to time needed for intestinal absorption and first-pass metabolism in the liver. The increased use of sustained-release formulations offers many advantages, as previously discussed; however, these agents generally take longer to reach effective plasma levels than their standard formulations. An animal that is anesthetized with isoflurane for a 30-min surgical procedure and does not receive a dose of SR-buprenorphine until after the surgery is completed will likely experience unrelieved pain for 30 to 60 min during the postoperative recovery period.

If the surgical period is sufficiently long that an analgesic can be administered under anesthesia and reach effective tissue levels before anesthetic recovery, then this provides another reasonable option. The advantage is that the animal will not be subject to an additional handling (and thus stress) event prior to anesthesia. Another advantage of preoperative or perioperative analgesia is the anesthetic-sparing effect that many of these drugs provide.[89,169] Thus, incorporating administration of analgesics into the anesthetic management plan is another method of providing balanced anesthesia that reduces some of the adverse effects of individual anesthetic agents.

Adjunct (nonpharmacologic) considerations

The goal of pain management is to keep patients as comfortable as possible. Nonpharmacologic interventions that may reduce pain should be considered during postoperative recovery in mice and rats . Animals subjected to procedures resulting in more chronic discomfort or pain are also good candidates for adjunct care. Training researchers in gentle handling techniques and methods to evaluate animals in a nonintrusive manner will minimize incidental stress. Taking time to habituate animals (particularly rats) to handling in advance of the invasive period can further reduce handling stress at both the time of surgery and during postoperative recovery. Skilled surgeons will minimize the degree to which the surgical procedure itself contributes unnecessarily to pain caused by excess tissue trauma, secondary infections, tissue swelling, or other inflammatory responses.[119] Selection of appropriate materials such as synthetic suture material and implanted materials that evoke less tissue response and effective sterilization of surgical materials are also important considerations.

General principles of supportive nursing care apply to all species, including laboratory rodents. Providing additional external heat will help with anesthetic recovery and prevent discomfort associated with hypothermia; soft dry bedding in a solid bottom cage similarly will provide a more comfortable recovery period. Bedding material that does not stick to the

animal's eyes, nose or mouth, such as a paper chip or shredded paper nesting material, should be used.[60] Housing animals in a quiet area that is not heavily trafficked will minimize another potential source of stress. Food and water should be easily accessible to the animals without having to stand up on their hind limbs and stretch to reach it, particularly for orthopedic, invasive abdominal or spinal cord surgeries. Food pellets can be placed on the cage floor and soaked with water to encourage consumption. For animals needing even more supportive care, a variety of high-calorie supplements are available as well as gels as a ready source of hydration. Animals that have had a procedure impacting their mouth, jaw, and/or surrounding tissues may benefit from a soft diet until healed. Administration of fluids either subcutaneously or intraperitoneally may also be beneficial, both in anesthetic recovery and also in preventing dehydration during a period of inappetence. Recommended volumes are 1 to 2 mL for mice and 5 to 10 mL for rats depending on body weight.[60] Larger volumes should be divided into two doses and administered at 2 separate sites.

Side effects of analgesia use in rodents

Analgesic drugs should be administered with care because of inherent side effects that result from their structure, chemical characteristics, and mechanism of action, and because of the potential for overdose effects when administered at high or extra-label doses. Even correct doses of analgesics can have unintended side effects if animals are not managed appropriately after a procedure. Figure 3 shows an example of renal tubular injury induced by flunixin meglumine. In addition to renal effects, other unintended side effects of some classes of NSAIDs include gastric and duodenal ulcers, and even intestinal perforations.[208] The higher dosages of NSAID currently being recommended narrow the therapeutic window and caution should be taken when dosing beyond 3 consecutive days. In addition, hydration status should be assured to minimize risk of acute kidney injury. Opioid use is similarly confounded by side effects, and intoxication is generally associated with cardiorespiratory depression, sedation, constipation, and cognitive impairment.[82] Different classes of opioids will have different mechanisms of action, and different side effects. For example, buprenorphine has been associated with pica and obstruction in rodents when used at high doses.[36] A common side effect of postoperative opioid administration in humans and other animal species that is rarely considered in mice and rats is postoperative nausea and vomiting (PONV).[172,185] Although most rodents cannot vomit, they may experience nausea after opioid administration. If rodents experience some version of PONV after opioid administration, then this could be associated with acute postprocedural weight loss. Dogs develop PONV more frequently after morphine administration than buprenorphine.[172]

Pain on injection may occur with some analgesic drugs and in particular NSAID. The intramuscular route is best avoided for injection in small rodents because swelling, necrosis and subsequent sloughing has been associated with administering acidic agents into their small muscle mass.[204] Some sustained-release formulations of buprenorphine have been associated with skin irritation and necrosis.[64]

Adverse effects of analgesics can be reduced with a number of strategies. Combining analgesic drugs with different mechanisms of action to reduce the overall dose required for any single agent, by using topical and local anesthetics, and incorporating other adjunctive forms of care for animals (see adjunct considerations above) all reduce the deficits associated with analgesia administration. Research teams should work together with their clinical veterinarian to select the safest and most appropriate analgesia plan for their studies.

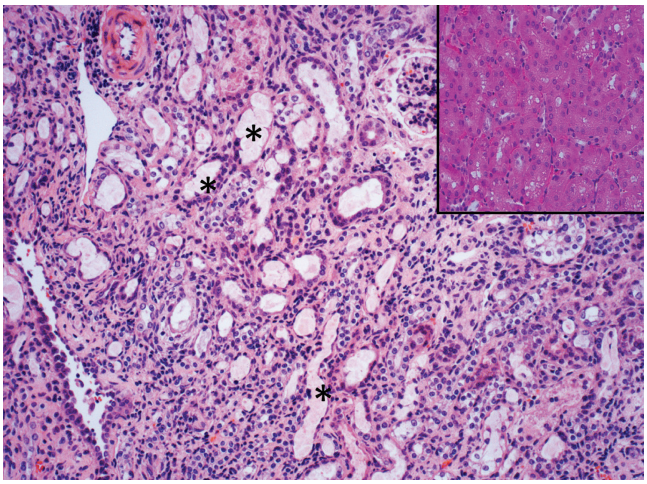

Figure 3. Photomicrograph of a mouse kidney demonstrating acute renal tubular dilatation and necrosis. This mouse was one of several animals that was treated with 2.5 mg/kg flunixin meglumine (Banamine) SC, a potent NSAID, following a 20-min surgical procedure conducted under ketamine/xylazine anesthesia. Fluids were not given after surgery, anesthesia was not reversed, and the cage of recumbent recovering mice was placed under a heat lamp. Animals were euthanized less than 24h after surgery following poor recovery. Renal injury was attributed to acute renal ischemia secondary to NSAID use that was compounded by mild to moderate clinical dehydration. Inset: Normal appearance of renal cortical tubules from an untreated mouse. (x100 H and E)

Conclusions

Like other mammals, laboratory rodents are sentient species and require the same considerations for peri-procedural treatment care that will minimize pain and suffering. The key to effective clinical management of pain is advance planning and anticipation of outcomes. A large number of pharmacologic and nonpharmacologic agents can be used alone or in combination to provide effective care while minimizing potential adverse effects. While the list of pharmacological agents considered most effective in rodents has not changed much in the last decade, we have presented an evidence-based approach to formulate our current recommendations, including consideration of dosing intervals, use of sustained-release formulations, and multimodal approaches to pain management. Based on the available evidence and dosing practices, rodents are often provided inadequate analgesia. However, a significant constraint is that more frequent dosing may require more frequent handling, restraint, and thus increased stress. Analgesics are potent agents with known side effects, and treatment plans should always be developed in conjunction with clinical veterinary input. More research is needed on the duration of effect of analgesia for many drugs and on better dose titrations for achieving and sustaining optimal analgesic blood drug levels. While valid scientific reasons may require withholding analgesic drugs to mice and rats after painful procedures, the vast majority of cases have no prohibitions to analgesic use. If potential experimental effects or interactions with specific analgesic agents are unknown or suspected, investigators and veterinarians should be encouraged to work collaboratively to design and conduct pilot studies before concluding that analgesia will not be provided. A commitment to appropriately managing pain in all research animals represents

a commitment to compassionate care and a goal that all those working with animals in research should be striving toward.

References

1. **Abbott FV, Bonder M.** 1997. Options for management of acute pain in the rat. Vet Rec **140:**553–557. https://doi.org/10.1136/vr.140.21.553.
2. **Abelson KSP, Jacobsen KR, Sundbom R, Kalliokoski O, Hau J.** 2012. Voluntary ingestion of nut paste for administration of buprenorphine in rats and mice. Lab Anim **46:**349–351. https://doi.org/10.1258/la.2012.012028.
3. **Ageel AM.** 1986. Effects of desipramine and chlorimipramine on buprenorphine analgesia in mice. Jpn J Pharmacol **41:**139–145. https://doi.org/10.1254/jjp.41.139.
4. **Aguilar-Carrasco JC, Rodríguez-Silverio J, Jiménez-Andrade JM, Carrasco-Portugal M del C, Flores-Murrieta FJ.** 2014. Relationship between blood levels and the antihyperalgesic effect of ketoprofen in the rat: ketoprofen pk/pd modeling in rats. Drug Dev Res **75:**189–194.
5. **Alexa T, Marza A, Voloseniuc T, Tamba B.** 2015. Enhanced analgesic effects of tramadol and common trace element coadministration in mice. J Neurosci Res **93:**1534–1541. https://doi.org/10.1002/jnr.23609.
6. **Al-Musawi A, Matar K, Kombian SB, Andersson L.** 2012. A pharmacokinetic study of a topical anesthetic (EMLA) in mouse soft tissue laceration. Dent Traumatol **28:**483–487. https://doi.org/10.1111/j.1600-9657.2012.01172.x.
7. **Apfelbaum JL, Chen C, Mehta SS, Gan TJ.** 2003. Postoperative pain experience: results from a national survey suggest postoperative pain continues to be undermanaged. Anesth Analg **97:**534–540. https://doi.org/10.1213/01.ANE.0000068822.10113.9E.
8. **Arras M, Rettich A, Cinelli P, Kasermann HP, Burki K.** 2007. Assessment of post-laparotomy pain in laboratory mice by telemetric recording of heart rate and heart rate variability. BMC Vet Res **3:**1–10. https://doi.org/10.1186/1746-6148-3-16.
9. **Atabaki R, Hassanpour-Ezatti M.** 2014. Improvement of lidocaine local anesthetic action using lallemantia royleana seed mucilage as an excipient. Iran J Pharm Res **13:**1431–1436.
10. **Bailey M, Corcoran T, Schug S, Toner A.** 2018. Perioperative lidocaine infusions for the prevention of chronic postsurgical pain: a systematic review and meta-analysis of efficacy and safety. Pain **159:**1696–1704. https://doi: 10.1097/j.pain.0000000000001273
11. **Bailey PM, Child CS.** 1987. Endocrine response to surgery. p 100–116. In: Kaufman L, editor. Anaesthesia review 4. London (United Kingdom): Churchill Livingstone.
12. **Barazanchi AWH, MacFater WS, Rahiri JL, Tutone S, Hill AG, Joshi GP, PROSPECT collaboration.** 2018. Evidence-based management of pain after laparoscopic cholecystectomy: a PROSPECT review update. Br J Anaesth **121:**787–803. https://doi.org/10.1016/j.bja.2018.06.023.
13. **Barker JC, Dibartola K, Wee C, Andonian N, Abdel-rasoul M, Lowery D, Janis JE.** 2018. Preoperative multimodal analgesia decreases postanesthesia care unit narcotic use and pain scores in outpatient breast surgery. Plast Reconstr Surg **142:**443e–450e. https://doi.org/10.1097/PRS.0000000000004804.
14. **Barrot M.** 2012. Tests and models of nociception and pain in rodents. Neuroscience **211:**39–50. https://doi.org/10.1016/j.neuroscience.2011.12.041.
15. **Batista LM, Batista IM, Almeida JP, Carvalho CH, de Castro-Costa SB, de Castro-Costa CM.** 2009. Preemptive analgesic effect of lidocaine in a chronic neuropathic pain model. Arq Neuropsiquiatr **67:**1088–1092. https://doi.org/10.1590/S0004-282X2009000600024.
16. **Bauer DJ, Christenson TJ, Clark KR, Powell SK, Swain RA.** 2003. Acetaminophen as a postsurgical analgesic in rats: a practical solution to neophobia. Contemp Top Lab Anim Sci **42:**20–25.
17. **Berry SH.** 2015. Analgesia in the perioperative period. Vet Clin North Am Small Anim Pract **45:**1013–1027. https://doi.org/10.1016/j.cvsm.2015.04.007.
18. **Bicket MC, Cohen SP.** 2018. Lidocaine infusions and preventative analgesia: can the answer to our prayers be hiding right under our noses? Pain **159:**1677–1678.
19. **Blankenship-Paris TL, Dutton JW, Goulding DR, McGee CA, Kissling GE, Myers PH.** 2016. Evaluation of buprenorphine hydrochloride Pluronic gel formulation in male C57BL/6NCrl mice. Lab Anim (NY) **45:**370–379. https://doi.org/10.1038/laban.1106.
20. **Bourque SL, Adams MA, Nakatsu K, Winterborn A.** 2010. Comparison of buprenorphine and meloxicam for postsurgical analgesia in rats: effects on body weight, locomotor activity, and hemodynamic parameters. J Am Assoc Lab Anim Sci **49:**617–622.
21. **Busch U, Schmid J, Heinzel G, Schmaus H, Baierl J, Huber C, Roth W.** 1998. Pharmacokinetics of meloxicam in animals and the relevance to humans. Drug Metab Dispos **26:**576–584.
22. **Buvanendran A, Fiala J, Patel KA, Golden AD, Moric M, Kroin JS.** 2015. The incidence and severity of postoperative pain following inpatient surgery. Pain Med **16:**2277–2283. https://doi.org/10.1111/pme.12751.
23. **Cannon CZ, Kissling GE, Goulding DR, King-Herbert AP, Blankenship-Paris T.** 2011. Analgesic effects of tramadol, carprofen or multimodal analgesia in rats undergoing ventral laparotomy. Lab Anim (NY) **40:**85–93. https://doi.org/10.1038/laban0311-85.
24. **Cannon CZ, Kissling GE, Hoenerhoff MJ, King-Herbert AP, Blankenship-Paris T.** 2010. Evaluation of dosages and routes of administration of tramadol analgesia in rats using hot-plate and tail-flick tests. Lab Anim (NY) **39:**342–351. https://doi.org/10.1038/laban1110-342.
25. **Carbone ET, Lindstrom KE, Diep S, Carbone L.** 2012. Duration of action of sustained-release buprenorphine in 2 strains of mice. J Am Assoc Lab Anim Sci **51:**815–819.
26. **Carbone L, Austin J.** 2016. Pain and laboratory animals: publication practices for better data reproducibility and better animal welfare. PLoS One **11:**1–24. https://doi.org/10.1371/journal.pone.0155001.
27. **Caro AC, Tucker JJ, Yannascoli SM, Dunkman AA, Thomas SJ, Soslowsky LJ.** 2014. Efficacy of various analgesics on shoulder function and rotator cuff tendon-to-bone healing in a rat (*Rattus norvegicus*) model. J Am Assoc Lab Anim Sci **53:**185–192.
28. **Carr DB, Goudas LC.** 1999. Acute pain. Lancet **353:**2051–2058. https://doi.org/10.1016/S0140-6736(99)03313-9.
29. **Chapman CR, Gavrin J.** 1999. Suffering: the contributions of persistent pain. Lancet **353:**2233–2237. https://doi.org/10.1016/S0140-6736(99)01308-2.
30. **Charlet A, Rodeau JL, Poisbeau P.** 2011. Radiotelemetric and symptomatic evaluation of pain in the rat after laparotomy: long-term benefits of perioperative ropivacaine care. J Pain **12:**246–256. https://doi.org/10.1016/j.jpain.2010.07.005.
31. **Chen YW, Shieh JP, Liu KS, Wang JJ, Hung CH.** 2017. Naloxone prolongs cutaneous nociceptive block by lidocaine in rats. Fundam Clin Pharmacol **31:**636–642.
32. **Chou AK, Chiu CC, Chen YW, Wang JJ, Hung CH.** 2018. Skin nociceptive block with pramoxine delivery by subcutaneous injection in rats. Pharmacol Rep **70:**1180–1184. https://doi.org/10.1016/j.pharep.2018.09.001.
33. **Chu CC, Wu SZ, Su WL, Shieh JP, Kao CH, Ho ST, Wang JJ.** 2008. Subcutaneous injection of inhaled anesthetics produces cutaneous analgesia. Can J Anaesth **55:**290–294. https://doi.org/10.1007/BF03017206.
34. **Chum HH, Jampachairsri K, McKeon GP, Yeomans DC, Pacharinsak C, Felt SA.** 2014. Antinociceptive effects of sustained-release buprenorphine in a model of incisional pain in rats (*Rattus norvegicus*). J Am Assoc Lab Anim Sci **53:**193–197.
35. **Cichewicz DL, Welch SP, Smith FL.** 2005. Enhancement of transdermal fentanyl and buprenorphine antinociception by transdermal Δ9-tetrahydrocannabinol. Eur J Pharmacol **525:**74–82. https://doi.org/10.1016/j.ejphar.2005.09.039.
36. **Clark JA Jr, Myers PH, Goelz MF, Thigpen JE, Forsythe DB.** 1997. Pica behavior associated with buprenorphine administration in the rat. Lab Anim Sci **47:**300–303.
37. **Clark TS, Clark DD, Hoyt RF Jr.** 2014. Pharmacokinetic comparison of sustained- release and standard buprenorphine in mice. J Am Assoc Lab Anim Sci **53:**387–391.
38. **Cobos EJ, Ghasemlou N, Araldi D, Segal D, Duong K, Woolf CJ.** 2012. Inflammation-induced decrease in voluntary wheel running in mice: A nonreflexive test for evaluating inflammatory

pain and analgesia. Pain **153:**876–884. https://doi.org/10.1016/j.pain.2012.01.016.
39. **Cooper DM, Delong D, Gilleti CS.** 1997. Analgesic efficacy of acetaminophen and buprenorphine administered in the drinking water of rats. Contemp Top Lab Anim Sci **36:**58–62.
40. **Cooper DM, Hoffman W, Tomlinson K, Lee HY.** 2008. Refinement of the dosage and dosing schedule of ketoprofen for postoperative analgesia in Sprague–Dawley rats. Lab Anim (NY) **37:**271–275. https://doi.org/10.1038/laban0608-271.
41. **Cooper DM, Hoffman W, Wheat N, Lee HY.** 2005. Duration of effects on clinical parameters and referred hyperalgesia in rats after abdominal surgery and multiple doses of analgesic. Comp Med **55:**344–353.
42. **Corletto F.** 2007. Multimodal and balanced analgesia. Vet Res Commun **31 S1:**59–63. https://doi.org/10.1007/s11259-007-0085-5.
43. **Coulter CA, Flecknell PA, Richardson CA.** 2009. Reported analgesic administration to rabbits, pigs, sheep, dogs and non-human primates undergoing experimental surgical procedures. Lab Anim **43:**232–238. https://doi.org/10.1258/la.2008.008021.
44. **Coulter CA, Flecknell PA, Leach MC, Richardson CA.** 2011. Reported analgesic administration to rabbits undergoing experimental surgical procedures. BMC Vet Res **7:**12. https://doi.org/10.1186/1746-6148-7-12.
45. **Crowe MS, Wilson CD, Leishman E, Prather PL, Bradshaw HB, Banks ML, Kinsey SG.** 2017. The monoacylglycerol lipase inhibitor KML29 with gabapentin synergistically produces analgesia in mice: KML29 and gabapentin synergistically reduce pain. Br J Pharmacol **174:**4523–4539. https://doi.org/10.1111/bph.14055.
46. **Dahl JB, Brennum J, Arendt-Nielsen L, Jensen TS, Kehlet H.** 1993. The effect of pre- versus postinjury infiltration with lidocaine on thermal and mechanical hyperalgesia after heat injury to the skin. Pain **53:**43–51. https://doi.org/10.1016/0304-3959(93)90054-S.
47. **David JM, Duarte Vogel S, Longo K, Sanchez D, Lawson G.** 2014. The use of eutectic mixture of lidocaine and prilocaine in mice (*Mus musculus*) for tail vein injections. Vet Anaesth Analg **41:**654–659. https://doi.org/10.1111/vaa.12177.
48. **Dudley ES, Johnson RA, French DC, Boivin GP.** 2016. Effects of topical anesthetics on behavior, plasma corticosterone, and blood glucose levels after tail biopsy of C57BL/6NHSD mice (*Mus musculus*). J Am Assoc Lab Anim Sci **55:**443–450.
49. **Dunbar ML, David EM, Aline MR, Lofgren JL.** 2016. Validation of a behavioral ethogram for assessing postoperative pain in guinea pigs (*Cavia porcellus*). J Am Assoc Lab Anim Sci **55:**29–34.
50. **Dunkman WJ, Manning MW.** 2018. Enhanced recovery after surgery and multimodal strategies for analgesia. Surg Clin North Am **98:**1171–1184. https://doi.org/10.1016/j.suc.2018.07.005.
51. **Ejlersen E, Andersen HB, Eliasen K, Mogensen T.** 1992. A comparison between preincisional and postincisional lidocaine infiltration and postoperative pain. Anesth Analg **74:**495–498.
52. **Ellen Y, Flecknell P, Leach M.** 2016. Evaluation of using behavioural changes to assess postoperative pain in the guinea pig (*Cavia Porcellus*). PLoS One **11:**1–20. https://doi.org/10.1371/journal.pone.0161941.
53. **Epstein M, Rodan I, Griffenhagen G, Kadrlik J, Petty M, Robertson S, Simpson W.** 2015. 2015 AAHA/AAFP pain management guidelines for dogs and cats. J Am Anim Hosp Assoc **51:**67–84. https://doi.org/10.5326/JAAHA-MS-7331.
54. **Epstein-Barash H, Shichor I, Kwon AH, Hall S, Lawlor MW, Langer R, Kohane DS.** 2009. Prolonged duration local anesthesia with minimal toxicity. Proc Natl Acad Sci USA **106:**7125–7130. https://doi.org/10.1073/pnas.0900598106. Erratum: Proc Natl Acad Sci USA 2011. **108:**4264
55. **Erhan E, Onal A, Kocabas S, Parlar A, Yegul I, Kosay S.** 2005. Ondansetron does not block tramadol-induced analgesia in mice. Methods Find Exp Clin Pharmacol **27:**629–632. https://doi.org/10.1358/mf.2005.27.9.939337.
56. **Evangelista Vaz R, Draganov DI, Rapp C, Avenel F, Steiner G, Arras M, Bergadano A.** 2018. Preliminary pharmacokinetics of tramadol hydrochloride after administration via different routes in male and female B6 mice. Vet Anaesth Analg **45:**111–122. https://doi.org/10.1016/j.vaa.2016.09.007.
57. **Faller KME, McAndrew DJ, Schneider JE, Lygate CA.** 2015. Refinement of analgesia following thoracotomy and experimental myocardial infarction using the Mouse Grimace Scale. Exp Physiol **100:**164–172. https://doi.org/10.1113/expphysiol.2014.083139.
58. **Fenwick N, Duffus SEG, Griffin G.** 2014. Pain management for animals used in science: views of scientists and veterinarians in Canada. Animals (Basel) **4:**494–514. https://doi.org/10.3390/ani4030494.
59. **Fernández-Dueñas V, Poveda R, Sánchez S, Ciruela F.** 2012. Synergistic interaction between fentanyl and a tramadol:paracetamol combination on the inhibition of nociception in mice. J Pharmacol Sci **118:**299–302. https://doi.org/10.1254/jphs.11161SC.
60. **Flecknell PA.** 2016. Laboratory animal anaesthesia. London (United Kingdom): Academic Press.
61. **Flecknell P.** 2018. Rodent analgesia: assessment and therapeutics. Vet J **232:**70–77. https://doi.org/10.1016/j.tvjl.2017.12.017.
62. **Flecknell PA, Roughan JV.** 1999. Use of oral buprenorphine ('Buprenorphine jello') for postoperative analgesia in rats—a clinical trial. Lab Anim **33:**169–174. https://doi.org/10.1258/002367799780578381.
63. **Foley PL.** 2014. Current options for providing sustained analgesia to laboratory animals. Lab Anim (NY) **43:**364–371. https://doi.org/10.1038/laban.590.
64. **Foley PL, Liang H, Crichlow AR.** 2011. Evaluation of a sustained-release formulation of buprenorphine for analgesia in rats. J Am Assoc Lab Anim Sci **50:**198–204.
65. **Fowler M, Clifford JL, Garza TH, Slater TM, Arizpe HM, Novak J, Petz LN, Loyd DR.** 2014. A rat model of full thickness thermal injury characterized by thermal hyperalgesia, mechanical allodynia, pronociceptive peptide release and tramadol analgesia. Burns **40:**759–771. https://doi.org/10.1016/j.burns.2013.10.011.
66. **Gades NM, Danneman PJ, Wixson SK, Tolley EA.** 2000. The magnitude and duration of the analgesic effect of morphine, butorphanol, and buprenorphine in rats and mice. Contemp Top Lab Anim Sci **39:**8–13.
67. **Gerçek A, Eti Z, Göğüş FY, Sav A.** 2004. The analgesic and anti-inflammatory effects of subcutaneous bupivacaine, morphine and tramadol in rats. Agri **16:**53–58.
68. **Girard P, Verniers D, Coppé M-C, Pansart Y, Gillardin J-M.** 2008. Nefopam and ketoprofen synergy in rodent models of antinociception. Eur J Pharmacol **584:**263–271. https://doi.org/10.1016/j.ejphar.2008.02.012.
69. **Giraudel JM, Diquelou A, Laroute V, Lees P, Toutain PL.** 2005. Pharmacokinetic/pharmacodynamic modelling of NSAIDs in a model of reversible inflammation in the cat. Br J Pharmacol **146:**642–653. https://doi.org/10.1038/sj.bjp.0706372.
70. **Goecke JC, Awad H, Lawson JC, Boivin GP.** 2005. Evaluating postoperative analgesics in mice using telemetry. Comp Med **55:**37–44.
71. **Goldkuhl R, Hau J, Abelson KSP.** 2010. Effects of voluntarily-ingested buprenorphine on plasma corticosterone levels, body weight, water intake, and behaviour in permanently catheterised rats. In Vivo **24:**131–135.
72. **Goldkuhl R, Jacobsen KR, Kalliokoski O, Hau J, Abelson KSP.** 2010. Plasma concentrations of corticosterone and buprenorphine in rats subjected to jugular vein catheterization. Lab Anim **44:**337–343. https://doi.org/10.1258/la.2010.009115.
73. **Grant GJ, Barenholz Y, Piskoun B, Bansinath M, Turndorf H, Bolotin EM.** 2001. DRV liposomal bupivacaine: preparation, characterization, and in vivo evaluation in mice. Pharm Res **18:**336–343. https://doi.org/10.1023/A:1011059131348.
74. **Grant GJ, Lax J, Susser L, Zakowski M, Weissman E.** 1997. Wound infiltration with liposomal bupivacaine prolongs analgesia in rats. Acta Anaesthesiol Scand **41:**204–207. https://doi.org/10.1111/j.1399-6576.1997.tb04666.x.
75. **Grant GJ, Vermeulen K, Langerman L, Zakowski M, Turndorf H.** 1994. Prolonged analgesia with liposomal bupivacaine in a mouse model. Reg Anesth **19:**264–269.
76. **Grant GJ, Zakowski MI, Vermeulen K, Langerman L, Ramanathan S, Turndorf H.** 1993. Assessing local anesthetic effect using the mouse tail flick test. J Pharmacol Toxicol Methods **29:**223–226. https://doi.org/10.1016/1056-8719(93)90029-E.

77. **Grant GJ, Piskoun B, Lin A, Bansinath M.** 2000. An in vivo method for the quantitative evaluation of local anesthetics. J Pharmacol Toxicol Methods **43:**69–72. https://doi.org/10.1016/S1056-8719(00)00079-4.
78. **Grant GJ, Vermeulen K, Zakowski M, Stenner M, Turndorf H, Langerman L.** 1994. Prolonged analgesia and decreased toxicity with liposomal morphine in a mouse model. Anesth Analg **79:**706–709. https://doi.org/10.1213/00000539-199410000-00015.
79. **Guarnieri M, Brayton C, DeTolla L, Forbes-McBean N, Sarabia-Estrada R, Zadnik P.** 2012. Safety and efficacy of buprenorphine for analgesia in laboratory mice and rats. Lab Anim (NY) **41:**337–343. https://doi.org/10.1038/laban.152.
80. **Guarnieri M, Tyler BM, DeTolla L, Zhao M, Kobrin B.** 2014. Subcutaneous implants for long-acting drug therapy in laboratory animals may generate unintended drug reservoirs. J Pharm Bioallied Sci **6:**38–42. https://doi.org/10.4103/0975-7406.124315.
81. **Hamidi GA, Jafari-Sabet M, Abed A, Mesdaghinia A, Mahlooji M, Banafshe HR.** 2014. Gabapentin enhances antinociceptive effects of morphine on heat, cold, and mechanical hyperalgesia in a rat model of neuropathic pain. Iran J Basic Med Sci **17:**753–759.
82. **Haroutounian S.** 2018. [Postoperative opioids, endocrine changes,and immunosuppression.] Schmerz **32:**374–380. https://doi.org/10.1007/s00482-018-0319-1. [Article in German].
83. **Hawkins M, Pascoe P.** 2012. Anesthesia, analgesia, and sedation of small mammals. p 429–451. Chapter 31. In: Quesenberry KE, Carpenter JW, editors. Ferrets, rabbits and rodents clinical medicine and surgery. St Louis (MO): Saunders. https://doi.org/10.1016/B978-1-4160-6621-7.00031-2
84. **Hawkins P.** 2002. Recognizing and assessing pain, suffering and distress in laboratory animals: a survey of current practice in the UK with recommendations. Lab Anim **36:**378–395. https://doi.org/10.1258/002367702320389044.
85. **Hayashida K, DeGoes S, Curry R, Eisenach JC.** 2007. Gabapentin activates spinal noradrenergic activity in rats and humans and reduces hypersensitivity after surgery. Anesthesiology **106:**557–562. https://doi.org/10.1097/00000542-200703000-00021.
86. **Hayes JH, Flecknell PA.** 1999. A comparison of pre- and post-surgical administration of bupivacaine or buprenorphine following laparotomy in the rat. Lab Anim **33:**16–23. https://doi.org/10.1258/002367799780578534.
87. **Hayes KE, Raucci JA, Gades NM, Toth LA.** 2000. An evaluation of analgesic regimens for abdominal surgery in mice. Contemp Top Lab Anim Sci **39:**18–23.
88. **Healy JR, Tonkin JL, Kamarec SR, Saludes MA, Ibrahim SY, Matsumoto RR, Wimsatt JH.** 2014. Evaluation of an improved sustained-release buprenorphine formulation for use in mice. Am J Vet Res **75:**619–625. https://doi.org/10.2460/ajvr.75.7.619.
89. **Hedenqvist P, Roughan JV, Flecknell PA.** 2000. Effects of repeated anaesthesia with ketamine/medetomidine and of pre-anaesthetic administration of buprenorphine in rats. Lab Anim **34:**207–211. https://doi.org/10.1258/002367700780457536.
90. **Herndon NL, Bandyopadhyay S, Hod EA, Prestia KA.** 2016. Sustained-release buprenorphine improves postsurgical clinical condition but does not alter survival or cytokine levels in a murine model of polymicrobial sepsis. Comp Med **66:**455–462.
91. **Herrmann K, Flecknell P.** 2019. Retrospective review of anesthetic and analgesic regimens used in animal research proposals. ALTEX **36:**65–80. https://doi.org/10.14573/altex.1804011.
92. **Hestehave S, Munro G, Pedersen TB, Abelson KSP.** 2017. Antinociceptive effects of voluntarily ingested buprenorphine in the hot-plate test in laboratory rats. Lab Anim **51:**264–272. https://doi.org/10.1177/0023677216668553.
93. **Hoare T, Young S, Lawlor MW, Kohane DS.** 2012. Thermoresponsive nanogels for prolonged duration local anesthesia. Acta Biomater **8:**3596–3605. https://doi.org/10.1016/j.actbio.2012.06.013.
94. **Hovard A, Teilmann A, Hau J, Abelson K.** 2015. The applicability of a gel delivery system for self-administration of buprenorphine to laboratory mice. Lab Anim **49:**40–45. https://doi.org/10.1177/0023677214551108.
95. **Hunter JC, Gogas KR, Hedley LR, Jacobson LO, Kassotakis L, Thompson J, Fontana DJ.** 1997. The effect of novel anti-epileptic drugs in rat experimental models of acute and chronic pain. Eur J Pharmacol **324:**153–160. https://doi.org/10.1016/S0014-2999(97)00070-8.
96. **Ingrao JC, Johnson R, Tor E, Gu Y, Litman M, Turner PV.** 2013. Aqueous stability and oral pharmacokinetics of meloxicam and carprofen in male C57BL/6 mice. J Am Assoc Lab Anim Sci **52:**553–559.
97. **Interagency Pain Research Coordinating Committee.**[Internet]. 2015. National pain strategy. [Cited 01 April 2019]. Available at https://iprcc.nih.gov/sites/default/files/HHSNational_Pain_Strategy_508C.pdf.
98. **Jablonski P, Howden B.** 2002. Oral buprenorphine and aspirin analgesia in rats undergoing liver transplantation. Lab Anim **36:**134–143. https://doi.org/10.1258/0023677021912415.
99. **Jacobsen KR, Fauerby N, Raida Z, Kalliokoski O, Hau J, Johansen FF, Abelson KS.** 2013. Effects of buprenorphine and meloxicam analgesia on induced cerebral ischemia in C57BL/6 male mice. Comp Med **63:**105–113.
100. **Jang HS, Jang IS, Lee MG.** 2010. The effects of tramadol on electroencephalographic spectral parameters and analgesia in rats. Korean J Physiol Pharmacol **14:**191–198. https://doi.org/10.4196/kjpp.2010.14.3.191.
101. **Jessen L, Christensen S, Bjerrum OJ.** 2007. The antinociceptive efficacy of buprenorphine administered through the drinking water of rats. Lab Anim **41:**185–196. https://doi.org/10.1258/002367707780378131.
102. **Jeunesse EC, Bargues IA, Toutain CE, Lacroix MZ, Letellier IM, Giraudel JM, Toutain PL.** 2011. Paw inflammation model in dogs for preclinical pharmacokinetic/pharmacodynamic investigations of nonsteroidal antiinflammatory drugs. J Pharmacol Exp Ther **338:**548–558. https://doi.org/10.1124/jpet.110.178350.
103. **Jirkof P, Tourvieille A, Cinelli P, Arras M.** 2015. Buprenorphine for pain relief in mice: repeated injections vs sustained-release depot formulation. Lab Anim **49:**177–187. https://doi.org/10.1177/0023677214562849.
104. **Jirkof P, Cesarovic N, Rettich A, Fleischmann T, Arras M.** 2012. Individual housing of female mice: influence on postsurgical behaviour and recovery. Lab Anim **46:**325–334. https://doi.org/10.1258/la.2012.012027.
105. **Jirkof P, Cesarovic N, Rettich A, Nicholls F, Seifert B, Arras M.** 2010. Burrowing behavior as an indicator of postlaparotomy pain in mice. Front Behav Neurosci **4:**165.
106. **Jirkof P, Fleischmann T, Cesarovic N, Rettich A, Vogel J, Arras M.** 2013. Assessment of postsurgical distress and pain in laboratory mice by nest complexity scoring. Lab Anim **47:**153–161. https://doi.org/10.1177/0023677213475603.
107. **Johnson RA.** 2016. Voluntary running-wheel activity, arterial blood gases, and thermal antinociception in rats after 3 buprenorphine formulations. J Am Assoc Lab Anim Sci **55:**306–311.
108. **Jones CP, Carver S, Kendall LV.** 2012. Evaluation of common anesthetic and analgesic techniques for tail biopsy in mice. J Am Assoc Lab Anim Sci **51:**808–814.
109. **Kalliokoski O, Jacobsen KR, Hau J, Abelson KSP.** 2011. Serum concentrations of buprenorphine after oral and parenteral administration in male mice. Vet J **187:**251–254. https://doi.org/10.1016/j.tvjl.2009.11.013.
110. **Kang SC, Jampachaisri K, Seymour TL, Felt SA, Pacharinsak C.** 2017. Use of liposomal bupivacaine for postoperative analgesia in an incisional pain model in rats (*Rattus norvegicus*). J Am Assoc Lab Anim Sci **56:**63–68.
111. **Kawano T, Takahashi T, Iwata H, Morikawa A, Imori S, Waki S, Tamura T, Yamazaki F, Eguchi S, Kumagai N, Yokoyama M.** 2014. Effects of ketoprofen for prevention of postoperative cognitive dysfunction in aged rats. J Anesth **28:**932–936. https://doi.org/10.1007/s00540-014-1821-y.
112. **Kendall LV, Hansen RJ, Dorsey K, Kang S, Lunghofer PJ, Gustafson DL.** 2014. Pharmacokinetics of sustained-release analgesics in mice. J Am Assoc Lab Anim Sci **53:**478–484.
113. **Kendall LV, Wegenast DJ, Smith BJ, Dorsey KM, Kang S, Lee NY, Hess AM.** 2016. Efficacy of sustained-release buprenorphine

in an experimental laparotomy model in female mice. J Am Assoc Lab Anim Sci **55:**66–73.

114. **Kent ML, Tighe PJ, Belfer I, Brennan TJ, Bruehl S, Brummett CM, Buckenmaier CC, Buvanendran A, Cohen RI, Desjardins P, Edwards D, Fillingim R, Gewandter J, Gordon DB, Hurley RW, Kehlet H, Loeser JD, Mackey S, McLean SA, Polomano R, Rahman S, Raja S, Rowbotham M, Suresh S, Schachtel B, Schreiber K, Schumacher M, Stacey B, Stanos S, Todd K, Turk DC, Weisman SJ, Wu C, Carr DB, Dworkin RH, Terman G.** 2017. The ACTTION-APS-AAPM pain taxonomy (AAAPT) multidimensional approach to classifying acute pain conditions. J Pain **18:**479–489. https://doi.org/10.1016/j.jpain.2017.02.421.
115. **Kest B, Wilson SG, Mogil JS.** 1999. Sex differences in supraspinal morphine analgesia are dependent on genotype. J Pharmacol Exp Ther **289:**1370–1375.
116. **Kimura M, Obata H, Saito S.** 2012. Antihypersensitivity effects of tramadol hydrochloride in a rat model of postoperative pain. Anesth Analg **115:**443–449. https://doi.org/10.1213/ANE.0b013e31825683c3.
117. **Kirsch JH, Klaus JA, Blizzard KK, Hurn PD, Murphy SJ.** 2002. Pain evaluation and response to buprenorphine in rats subjected to sham middle cerebral artery occlusion. Contemp Top Lab Anim Sci **41:**9–14.
118. **Kögel B, Christoph T, Straßburger W, Friderichs E.** 2005. Interaction of μ-opioid receptor agonists and antagonists with the analgesic effect of buprenorphine in mice. Eur J Pain **9:**599–611. https://doi.org/10.1016/j.ejpain.2005.02.002.
119. **Kohn DF, Martin TE, Foley PL, Morris TH, Swindle MM, Vogler GA, Wixson SK.** 2007. Guidelines for the assessment and management of pain in rodents and rabbits. J Am Assoc Lab Anim Sci **46:**97–108.
120. **Kolesnikov YA, Chereshnev I, Pasternak GW.** 2000. Analgesic synergy between topical lidocaine and topical opioids. J Pharmacol Exp Ther **295:**546–551.
121. **Kolesnikov YA, Wilson RS, Pasternak GW.** 2003. The synergistic analgesic interactions between hydrocodone and ibuprofen. Anesth Analg **97:**1721–1723. https://doi.org/10.1213/01.ANE.0000087801.20395.97.
122. **Korat PS, Kapupara PP.** 2017. Local infiltration of the surgical wound with levobupivacaine, ibuprofen, and epinephrine in postoperative pain: an experimental study. Biomed Pharmacother **96:**104–111. https://doi.org/10.1016/j.biopha.2017.09.131.
123. **Koutroli E, Alexakos P, Kakazanis Z, Symeon I, Balafas E, Voyiatzaki C, Kostomitsopoulos N.** 2014. Effects of using the analgesic tramadol in mice undergoing embryo transfer surgery. Lab Anim (NY) **43:**167–172. https://doi.org/10.1038/laban.518.
124. **Krueger KL, Fujiwara Y.** 2008. The use of buprenorphine as an analgesic after rodent embryo transfer. Lab Anim (NY) **37:**87–90. https://doi.org/10.1038/laban0208-87.
125. **Kurz A, Sessler DI.** 2003. Opioid-induced bowel dysfunction: pathophysiology and potential new therapies. Drugs **63:**649–671. https://doi.org/10.2165/00003495-200363070-00003.
126. **Lamont LA.** 2008. Multimodal pain management in veterinary medicine: the physiologic basis of pharmacologic therapies. Vet Clin North Am Small Anim Pract **38:**1173–1186. https://doi.org/10.1016/j.cvsm.2008.06.005.
127. **Langford DJ, Bailey AL, Chanda ML, Clarke SE, Drummond TE, Echols S, Glick S, Ingrao J, Klassen-Ross T, LaCroix-Fralish ML, Matsumiya L, Sorge RE, Sotocinal SG, Tabaka JM, Wong D, van den Maagdenberg AMJM, Ferrari MD, Craig KD, Mogil JS.** 2010. Coding of facial expressions of pain in the laboratory mouse. Nat Methods **7:**447–449. https://doi.org/10.1038/nmeth.1455.
128. **Lascelles BD, Waterman AE, Cripps PJ, Livingston A, Henderson G.** 1995. Central sensitization as a result of surgical pain: investigation of the pre-emptive value of pethidine for ovariohysterectomy in the rat. Pain **62:**201–212. https://doi.org/10.1016/0304-3959(94)00266-H.
129. **Leach MC, Forrester AR, Flecknell PA.** 2010. Influence of preferred foodstuffs on the antinociceptive effects of orally administered buprenorphine in laboratory rats. Lab Anim **44:**54–58. https://doi.org/10.1258/la.2009.009029.
130. **Leach MC, Klaus K, Miller AL, Scotto di Perrotolo M, Sotocinal SG, Flecknell PA.** 2012. The assessment of post-vasectomy pain in mice using behaviour and the mouse grimace scale. PLoS One **7:**1–9. https://doi.org/10.1371/journal.pone.0035656.
131. **Lees P, Giraudel J, Landoni MF, Toutain PL.** 2004. PK-PD integration and PK-PD modelling of nonsteroidal antiinflammatory drugs: principles and applications in veterinary pharmacology. J Vet Pharmacol Ther **27:**491–502. https://doi.org/10.1111/j.1365-2885.2004.00618.x.
132. **Lees P, Landoni MF, Giraudel J, Toutain PL.** 2004. Pharmacodynamics and pharmacokinetics of nonsteroidal anti-inflammatory drugs in species of veterinary interest. J Vet Pharmacol Ther **27:**479–490. https://doi.org/10.1111/j.1365-2885.2004.00617.x.
133. **Leung V, Zhang E, Pang DS.** 2016. Real-time application of the rat grimace scale as a welfare refinement in laboratory rats. Sci Rep **6:**1–12. https://doi.org/10.1038/srep31667.
134. **Liles JH, Flecknell P, Roughan J, Cruz-Madorran I.** 1998. Influence of oral buprenorphine, oral naltrexone or morphine on the effects of laparotomy in the rat. Lab Anim **32:**149–161. https://doi.org/10.1258/002367798780600025.
135. **Loram LC, Mitchell D, Skosana M, Fick LG.** 2007. Tramadol is more effective than morphine and amitriptyline against ischaemic pain but not thermal pain in rats. Pharmacol Res **56:**80–85. https://doi.org/10.1016/j.phrs.2007.04.003.
136. **Lukas G, Brindle SD, Greengard P.** 1971. The route of absorption of intraperitoneally administered compounds. J Pharmacol Exp Ther **178:**562–564.
137. **Magalhães-Sant'Ana M, Sandøe P, Olsson I.** 2009. Painful dilemmas: the ethics of animal-based pain research. Anim Welf **18:**49–63.
138. **Martin LBE, Thompson AC, Martin T, Kristal MB.** 2001. Analgesic efficacy of orally administered buprenorphine in rats. Comp Med **51:**43–48.
139. **Matson DJ, Broom DC, Carson SR, Baldassari J, Kehne J, Cortright DN.** 2007. Inflammation-induced reduction of spontaneous activity by adjuvant: a novel model to study the effect of analgesics in rats. J Pharmacol Exp Ther **320:**194–201. https://doi.org/10.1124/jpet.106.109736.
140. **Matsumiya LC, Sorge RE, Sotocinal SG, Tabaka JM, Wieskopf JS, Zaloum A, King OD, Mogil JS.** 2012. Using the mouse grimace scale to reevaluate the efficacy of postoperative analgesics in laboratory mice. J Am Assoc Lab Anim Sci **51:**42–49.
141. **McKeon GP, Pacharinsak C, Long CT, Howard AM, Jampachaisri K, Yeomans DC, Felt SA.** 2011. Analgesic effects of tramadol, tramadol–gabapentin, and buprenorphine in an incisional model of pain in rats (*Rattus norvegicus*). J Am Assoc Lab Anim Sci **50:**192–197.
142. **McQuay HJ, Carroll D, Moore RA.** 1988. Postoperative orthopaedic pain — the effect of opiate premedication and local anaesthetic blocks. Pain **33:**291–295. https://doi.org/10.1016/0304-3959(88)90287-4.
143. **Medina-López R, Vara-Gama N, Soria-Arteche O, Moreno-Rocha L, López-Muñoz F.** 2018. Pharmacokinetics and pharmacodynamics of (s)-ketoprofen co-administered with caffeine: a preclinical study in arthritic rats. Pharmaceutics **10:**20. https://doi.org/10.3390/pharmaceutics10010020.
144. **Merlos M, Portillo-Salido E, Brenchat A, Aubel B, Buxens J, Fisas A, Codony X, Romero L, Zamanillo D, Vela JM.** 2018. Administration of a co-crystal of tramadol and celecoxib in a 1:1 molecular ratio produces synergistic antinociceptive effects in a postoperative pain model in rats. Eur J Pharmacol **833:**370–378. https://doi.org/10.1016/j.ejphar.2018.06.022.
145. **Meymandi M-S, Keyhanfar F.** 2012. Pregabalin antinociception and its interaction with tramadol in actue model of pain. Pharmacol Rep **64:**576–585. https://doi.org/10.1016/S1734-1140(12)70853-8.
146. **Meymandi M-S, Sepehri G, Mobasher M.** 2006. Gabapentin enhances the analgesic response to morphine in acute model of pain in male rats. Pharmacol Biochem Behav **85:**185–189. https://doi.org/10.1016/j.pbb.2006.07.037.
147. **Mickley GA, Hoxha Z, Biada JM, Kenmuir CL, Bacik SE.** 2006. Acetaminophen self-administered in the drinking water increases the pain threshold of rats (*Rattus norvegicus*). J Am Assoc Lab Anim Sci **45:**48–54.

148. **Miller AL, Kitson GL, Skalkoyannis B, Flecknell PA, Leach MC.** 2016. Using the mouse grimace scale and behaviour to assess pain in CBA mice following vasectomy. Appl Anim Behav Sci **181:**160–165. https://doi.org/10.1016/j.applanim.2016.05.020.
149. **Miller AL, Wright-Williams SL, Flecknell PA, Roughan JV.** 2012. A comparison of abdominal and scrotal approach methods of vasectomy and the influence of analgesic treatment in laboratory mice. Lab Anim **46:**304–310. https://doi.org/10.1258/la.2012.012078.
150. **Miranda HF, Puig MM, Romero MA, Prieto JC.** 2009. Effects of tramadol and dexketoprofen on analgesia and gastrointestinal transit in mice. Fundam Clin Pharmacol **23:**81–88. https://doi.org/10.1111/j.1472-8206.2008.00636.x.
151. **Miranda HF, Prieto JC, Puig MM, Pinardi G.** 2008. Isobolographic analysis of multimodal analgesia in an animal model of visceral acute pain. Pharmacol Biochem Behav **88:**481–486. https://doi.org/10.1016/j.pbb.2007.10.005.
152. **Miranda HF, Romero MA, Puig MM.** 2012. Antinociceptive and anti-exudative synergism between dexketoprofen and tramadol in a model of inflammatory pain in mice: Synergy in analgesia. Fundam Clin Pharmacol **26:**373–382. https://doi.org/10.1111/j.1472-8206.2010.00922.x.
153. **Miranda HF, Sierralta F, Aranda N, Poblete P, Noriega V, Prieto JC.** 2018. Synergism between gabapentin-tramadol in experimental diabetic neuropathic pain. Fundam Clin Pharmacol **32:**581–588. https://doi.org/10.1111/fcp.12400.
154. **Mogil JS, Wilson SG, Bon K, Lee SE, Chung K, Raber P, Pieper JO, Hain HS, Belknap JK, Hubert L, Elmer GI, Chung JM, Devor M.** 1999. Heritability of nociception I: responses of 11 inbred mouse strains on 12 measures of nociception. Pain **80:**67–82. https://doi.org/10.1016/S0304-3959(98)00197-3.
155. **Molina-Cimadevila MJ, Segura S, Merino C, Ruiz-Reig N, Andrés B, de Madaria E.** 2014. Oral self-administration of buprenorphine in the diet for analgesia in mice. Lab Anim **48:**216–224. https://doi.org/10.1177/0023677214532454.
156. **Montilla-García Á, Tejada MÁ, Perazzoli G, Entrena JM, Portillo-Salido E, Fernández-Segura E, Cañizares FJ, Cobos EJ.** 2017. Grip strength in mice with joint inflammation: a rheumatology function test sensitive to pain and analgesia. Neuropharmacology **125:**231–242. https://doi.org/10.1016/j.neuropharm.2017.07.029.
157. **Munro G, Dyhr H, Grunnet M.** 2010. Selective potentiation of gabapentin-mediated antinociception in the rat formalin test by the nicotinic acetylcholine receptor agonist ABT-594. Neuropharmacology **59:**208–217. https://doi.org/10.1016/j.neuropharm.2010.05.010.
158. **National Research Council.** 2009. Recognition and alleviation of pain in laboratory animals, Washington (DC): National Academies Press
159. **Nishiyori M, Ueda H.** 2008. Prolonged gabapentin analgesia in an experimental mouse model of fibromyalgia. Mol Pain **4:**1–6. https://doi.org/10.1186/1744-8069-4-52.
160. **Nunamaker EA, Goldman JL, Adams CR, Fortman JD.** 2018. Evaluation of analgesic efficacy of meloxicam and 2 formulations of buprenorphine after laparotomy in female Sprague–Dawley rats. J Am Assoc Lab Anim Sci **57:**498–507. https://doi.org/10.30802/AALAS-JAALAS-17-000129.
161. **Ohtani M, Kotaki H, Sawada Y, Iga T.** 1995. Comparative analysis of buprenorphine- and norbuprenorphine-induced analgesic effects based on pharmacokinetic-pharmacodynamic modeling. J Pharmacol Exp Ther **272:**505–510.
162. **Okulicz-Kozaryn I, Leppert W, Mikolajczak P, Kaminska E.** 2013. Analgesic effects of tramadol in combination with adjuvant drugs: an experimental study in rats. Pharmacol **91:**7–11. https://doi.org/10.1159/000343632.
163. **Oliver VL, Athavale S, Simon KE, Kendall LV, Nemzek JA, Lofgren JL.** 2017. Evaluation of pain assessment techniques and analgesia efficacy in a female guinea pig (*Cavia porcellus*) model of surgical pain. J Am Assoc Lab Anim Sci **56:**425–435.
164. **Oliver VL, Thurston SE, Lofgren JL.** 2018. Using cageside measures to evaluate analgesic efficacy in mice (*Mus musculus*) after surgery. J Am Assoc Lab Anim Sci **57:**186–201.
165. **Pairet M, Ruckebusch Y.** 1989. On the relevance of nonsteroidal antiinflammatory drugs in the prevention of paralytic ileus in rodents. J Pharm Pharmacol **41:**757–761. https://doi.org/10.1111/j.2042-7158.1989.tb06360.x.
166. **Pais-Vieira M, Lima D, Galhardo V.** 2009. Sustained attention deficits in rats with chronic inflammatory pain. Neurosci Lett **463:**98–102. https://doi.org/10.1016/j.neulet.2009.07.050.
167. **Papathanasiou T, Juul RV, Gabel-Jensen C, Kreilgaard M, Heegaard AM, Lund TM.** 2017. Quantification of the pharmacodynamic interaction of morphine and gabapentin using a response surface approach. AAPS J **19:**1804–1813. https://doi.org/10.1208/s12248-017-0140-2.
168. **Park I, Kim D, Song J, In CH, Jeong SW, Lee SH, Min B, Lee D, Kim SO.** 2008. Buprederm™, a new transdermal delivery system of buprenorphine: pharmacokinetic, efficacy and skin irritancy studies. Pharm Res **25:**1052–1062. https://doi.org/10.1007/s11095-007-9470-6.
169. **Penderis J, Franklin RJ.** 2005. Effects of pre- versus post-anaesthetic buprenorphine on propofol-anaesthetized rats. Vet Anaesth Analg **32:**256–260. https://doi.org/10.1111/j.1467-2995.2005.00183.x.
170. **Peterson NC, Nunamaker EA, Turner PV.** 2017. To treat or not to treat: the effects of pain on experimental parameters. Comp Med **67:**469–482.
171. **Pick CG, Peter Y, Schreiber S, Weizman R.** 1997. Pharmacological characterization of buprenorphine, a mixed agonist–antagonist with κ3 analgesia. Brain Res **744:**41–46. https://doi.org/10.1016/S0006-8993(96)01069-4.
172. **Ramsey D, Fleck T, Berg T, Nederveld S, DeLong D, Tena J-K, Aleo M, McCall R.** 2014. Cerenia prevents perioperative nausea and vomiting and improves recovery in dogs undergoing routine surgery. Int J Appl Res Vet Med **12:**228–237.
173. **Rätsep MT, Barrette VF, Winterborn A, Adams MA, Croy BA.** 2013. Hemodynamic and behavioral differences after administration of meloxicam, buprenorphine, or tramadol as analgesics for telemeter implantation in mice. J Am Assoc Lab Anim Sci **52:**560–566.
174. **Richardson CA, Flecknell PA.** 2005. Anaesthesia and postoperative analgesia following experimental surgery in laboratory rodents: are we making progress? Altern Lab Anim **33:**119–127.
175. **Romero A, Miranda HF, Puig MM.** 2010. Antinociceptive effects of morphine, fentanyl, tramadol and their combination, in morphine-tolerant mice. Pharmacol Biochem Behav **97:**363–369. https://doi.org/10.1016/j.pbb.2010.09.005.
176. **Roughan JV, Flecknell PA.** 2004. Behaviour-based assessment of the duration of laparotomy-induced abdominal pain and the analgesic effects of carprofen and buprenorphine in rats. Behav Pharmacol **15:**461–472. https://doi.org/10.1097/00008877-200411000-00002.
177. **Rutten K, Robens A, Read SJ, Christoph T.** 2014. Pharmacological validation of a refined burrowing paradigm for prediction of analgesic efficacy in a rat model of sub-chronic knee joint inflammation. Eur J Pain **18:**213–222. https://doi.org/10.1002/j.1532-2149.2013.00359.x.
178. **Rutten K, Schiene K, Robens A, Leipelt A, Pasqualon T, Read SJ, Christoph T.** 2014. Burrowing as a nonreflex behavioural readout for analgesic action in a rat model of sub-chronic knee joint inflammation: Burrowing as a nonreflex behavioural readout. Eur J Pain **18:**204–212. https://doi.org/10.1002/j.1532-2149.2013.00358.x.
179. **Sadar MJ, Knych HK, Drazenovich TL, Paul-Murphy JR.** 2018. Pharmacokinetics of buprenorphine after intravenous and oral transmucosal administration in guinea pigs (*Cavia porcellus*). Am J Vet Res **79:**260–266. https://doi.org/10.2460/ajvr.79.3.260.
180. **Santos ARS, Vedana EMA, De Freitas GAG.** 1998. Antinociceptive effect of meloxicam, in neurogenic and inflammatory nociceptive models in mice. Inflamm Res **47:**302–307. https://doi.org/10.1007/s000110050333.
181. **Satterwhite JH, Boudinot FD.** 1992. Pharmacokinetics of ketoprofen in rats: effect of age and dose. Biopharm Drug Dispos **13:**197–212. https://doi.org/10.1002/bdd.2510130306.
182. **Sauer M, Fleischmann T, Lipiski M, Arras M, Jirkof P.** 2016. Buprenorphine via drinking water and combined oral-injection

protocols for pain relief in mice. Appl Anim Behav Sci **185:**103–112. https://doi.org/10.1016/j.applanim.2016.09.009.

183. **Schaap MWH, Uilenreef JJ, Mitsogiannis MD, van 't Klooster JG, Arndt SS, Hellebrekers LJ.** 2012. Optimizing the dosing interval of buprenorphine in a multimodal postoperative analgesic strategy in the rat: minimizing side-effects without affecting weight gain and food intake. Lab Anim **46:**287–292. https://doi.org/10.1258/la.2012.012058.
184. **Seymour TL, Adams SC, Felt SA, Jampachaisri K, Yeomans DC, Pacharinsak C.** 2016. Postoperative analgesia due to sustained-release buprenorphine, sustained-release meloxicam, and carprofen gel in a model of incisional pain in rats (*Rattus norvegicus*). J Am Assoc Lab Anim Sci **55:**300–305.
185. **Shaikh SI, Nagarekha D, Hegade G, Marutheesh M.** 2016. Postoperative nausea and vomiting: a simple yet complex problem. Anesth Essays Res **10:**388–396. https://doi.org/10.4103/0259-1162.179310.
186. **Sheikholeslami B, Gholami M, Lavasani H, Rouini M.** 2016. Evaluation of the route dependency of the pharmacokinetics and neuro-pharmacokinetics of tramadol and its main metabolites in rats. Eur J Pharm Sci **92:**55–63. https://doi.org/10.1016/j.ejps.2016.06.021.
187. **Shikanov A, Domb AJ, Weiniger CF.** 2007. Long acting local anesthetic–polymer formulation to prolong the effect of analgesia. J Control Release **117:**97–103. https://doi.org/10.1016/j.jconrel.2006.10.014.
188. **Shinozaki T, Yamada T, Nonaka T, Yamamoto T.** 2015. Acetaminophen and non-steroidal anti-inflammatory drugs interact with morphine and tramadol analgesia for the treatment of neuropathic pain in rats. J Anesth **29:**386–395. https://doi.org/10.1007/s00540-014-1953-0.
189. **Smith BJ, Wegenast DJ, Hansen RJ, Hess AM, Kendall LV.** 2016. Pharmacokinetics and paw withdrawal pressure in female guinea pigs (*Cavia porcellus*) treated with sustained-release buprenorphine and buprenorphine hydrochloride. J Am Assoc Lab Anim Sci **55:**789–793.
190. **Smith FL.** 1997. Regional cutaneous differences in the duration of bupivacaine local anesthesia in mice. Life Sci **60:**1613–1621. https://doi.org/10.1016/S0024-3205(97)00128-8.
191. **Smith FL, Davis RW, Carter R.** 2001. Influence of voltage-sensitive Ca^{++}channel drugs on bupivacaine infiltration anesthesia in mice. Anesthesiology **95:**1189–1197.
192. **Sokolsky-Papkov M, Golovanevski L, Domb AJ, Weiniger CF.** 2009. Prolonged local anesthetic action through slow release from poly (lactic acid co castor oil). Pharm Res **26:**32–39. https://doi.org/10.1007/s11095-008-9699-8.
193. **Sotocinal SG, Sorge RE, Zaloum A, Tuttle AH, Martin LJ, Wieskopf JS, Mapplebeck JC, Wei P, Zhan S, Zhang S, McDougall JJ, King OD, Mogil JS.** 2011. The Rat Grimace Scale: a partially automated method for quantifying pain in the laboratory rat via facial expressions. Mol Pain **7:**1–10.
194. **Speth RC, Smith MS, Brogan RS.** 2001. Regarding the inadvisability of administering postoperative analgesics in the drinking water of rats (*Rattus norvegicus*). Contemp Top Lab Anim Sci **40:**15–17.
195. **Spofford CM, Ashmawi H, Subieta A, Buevich F, Moses A, Baker M, Brennan TJ.** 2009. Ketoprofen produces modality-specific inhibition of pain behaviors in rats after plantar incision. Anesth Analg **109:**1992–1999. https://doi.org/10.1213/ANE.0b013e3181bbd9a3.
196. **St A Stewart L, Martin WJ.** 2003. Evaluation of postoperative analgesia in a rat model of incisional pain. Contemp Top Lab Anim Sci **42:**28–34.
197. **Stasiak KL, Maul D, French E, Hellyer PW, Vandewoude S.** 2003. Species-specific assessment of pain in laboratory animals. Contemp Top Lab Anim Sci **42:**13–20.
198. **Stephan FK, Zucker I.** 1972. Circadian rhythms in drinking behavior and locomotor activity of rats are eliminated by hypothalamic lesions. Proc Natl Acad Sci USA **69:**1583–1586. https://doi.org/10.1073/pnas.69.6.1583.
199. **Stokes EL, Flecknell PA, Richardson CA.** 2009. Reported analgesic and anaesthetic administration to rodents undergoing experimental surgical procedures. Lab Anim **43:**149–154. https://doi.org/10.1258/la.2008.008020.
200. **Sundbom R, Jacobsen KR, Kalliokoski O, Hau J, Abelson KSP.** 2011. Postoperative corticosterone levels in plasma and feces of mice subjected to permanent catheterization and automated blood sampling. In Vivo **25:**335–342.
201. **Teixeira FM, Castro LL, Ferreira RT, Pires PA, Vanderlinde FA, Medeiros MA.** 2012. High-frequency electroacupuncture versus carprofen in an incisional pain model in rats. Braz J Med Biol Res **45:**1209–1214. https://doi.org/10.1590/S0100-879X2012007500133.
202. **Traul KA, Romero JB, Brayton C, DeTolla L, Forbes-McBean N, Halquist MS, Karnes HT, Sarabia-Estrada R, Tomlinson MJ, Tyler BM, Ye X, Zadnik P, Guarnieri M.** 2015. Safety studies of post-surgical buprenorphine therapy for mice. Lab Anim **49:**100–110. https://doi.org/10.1177/0023677214554216.
203. **Tubbs JT, Kissling GE, Travlos GS, Goulding DR, Clark JA, King-Herbert AP, Blankenship-Paris TL.** 2011. Effects of buprenorphine, meloxicam, and flunixin meglumine as postoperative analgesia in mice. J Am Assoc Lab Anim Sci **50:**185–191.
204. **Turner PV, Brabb T, Pekow C, Vasbinder MA.** 2011. Administration of substances to laboratory animals: routes of administration and factors to consider. J Am Assoc Lab Anim Sci **50:**600–613.
205. **Turner P, Pang D, Lofgren J.** 2019. A review of pain assessment methods in laboratory rodents. Comp Med **69:**EPub ahead of print.
206. **Tyers MB.** 1980. A classification of opiate receptors that mediate antinociception in animals. Br J Pharmacol **69:**503–512. https://doi.org/10.1111/j.1476-5381.1980.tb07041.x.
207. **Wala EP, Holtman JR Jr.** 2011. Buprenorphine-induced hyperalgesia in the rat. Eur J Pharmacol **651:**89–95. https://doi.org/10.1016/j.ejphar.2010.10.083.
208. **Wallace JL, McKnight W, Reuter BK, Vergnolle N.** 2000. Nsaid-induced gastric damage in rats: requirement for inhibition of both cyclooxygenase 1 and 2. Gastroenterology **119:**706–714. https://doi.org/10.1053/gast.2000.16510.
209. **Woolf CJ, Wall PD.** 1986. Morphine-sensitive and morphine-insensitive actions of C-fibre input on the rat spinal cord. Neurosci Lett **64:**221–225. https://doi.org/10.1016/0304-3940(86)90104-7.
210. **World Health Organization.**[Internet]. 2019. WHO's pain relief ladder for patient management. [Cited 01 April 2019]. Available at https://www.who.int/cancer/palliative/painladder/en/.
211. **Wright-Williams S, Flecknell PA, Roughan JV.** 2013. Comparative effects of vasectomy surgery and buprenorphine treatment on faecal corticosterone concentrations and behaviour assessed by manual and automated analysis methods in C57 and C3H mice. PLoS One **8:**1–13. https://doi.org/10.1371/journal.pone.0075948.
212. **Wright-Williams SL, Courade J, Richardson CA, Roughan JV, Flecknell PA.** 2007. Effects of vasectomy surgery and meloxicam treatment on faecal corticosterone levels and behaviour in 2 strains of laboratory mouse. Pain **130:**108–118. https://doi.org/10.1016/j.pain.2006.11.003.
213. **Wu SZ, Liu KS, Chu KS, Cheng KI, Kuei CH, Wang JJ, Tzeng JI.** 2006. The depot of buprenorphine decanoate produced a dose-related long-lasting antinociceptive effect in guinea pigs. Acta Anaesthesiol Taiwan **44:**161–168.
214. **Xie H, Dong ZQ, Ma F, Bauer WR, Wang X, Wu GC.** 2008. Involvement of serotonin 2A receptors in the analgesic effect of tramadol in mono-arthritic rats. Brain Res **1210:**76–83. https://doi.org/10.1016/j.brainres.2008.02.049.
215. **Yun M, Jeong S, Pai C, Arndt S.** 2010. Pharmacokinetic-pharmacodynamic modeling of the analgesic effect of buprederm(TM) in mice. Health (N Y) **2:**824–831.
216. **Zanetti AS, Putta SK, Casebolt DB, Louie SG.** 2017. Pharmacokinetics and adverse effects of 3 sustained-release buprenorphine dosages in healthy guinea pigs (*Cavia porcellus*). J Am Assoc Lab Anim Sci **56:**768–778.
217. **Zelcer S, Kolesnikov Y, Kovalyshyn I, Pasternak DA, Pasternak GW.** 2005. Selective potentiation of opioid analgesia by nonsteroidal antiinflammatory drugs. Brain Res **1040:**151–156. https://doi.org/10.1016/j.brainres.2005.01.070.
218. **Zhang Y, Du L, Pan H, Li L, Su X.** 2011. Enhanced analgesic effects of propacetamol and tramadol combination in rats and mice. Biol Pharm Bull **34:**349–353. https://doi.org/10.1248/bpb.34.349.