Evaluation of Medicated Gel as a Supplement to Providing Acetaminophen in the Drinking Water of Sprague Dawley Rats After Surgery

L E Riddle, S Raiciulescu, A B Mullins, C D Foster

Citation

L E Riddle, S Raiciulescu, A B Mullins, C D Foster. *Evaluation of Medicated Gel as a Supplement to Providing Acetaminophen in the Drinking Water of Sprague Dawley Rats After Surgery* . The Internet Journal of Veterinary Medicine. 2018 Volume 15 Number 1.

DOI: 10.5580/IJVM.53539

Abstract

Numerous studies have tested the suitability of specific analgesics to ameliorate post-procedural pain in laboratory rodents with varying results. The drug selected and the route of administration are two major factors in whether an analgesic may provide sufficient, reliable pain relief. Acetaminophen is an easily obtainable analgesic that can be self-administered to rodents in both water and gel products. This study compared the consumption of acetaminophen-treated water and/or gel offered to male Sprague Dawley rats following a sham surgical procedure to determine if there was a preference for acetaminophen delivery and to estimate whether the rats ingested a therapeutic dose of acetaminophen. Rats were assigned to one of three treatment groups post-surgery based on the drug delivery system provided: acetaminophen in water (AW), acetaminophen in gel (AG), and acetaminophen in water and gel (AWG). Body weight measurements were taken one day prior to surgery (day -1) and for three days post-operatively (days 1-3), while water and gel weights were measured over three days All three groups ingested acetaminophen doses that were higher than the 200 mg/kg targeted therapeutic dose, with the AW group ingesting the lowest average daily dose (350.9 mg/kg), and the AG group ingesting the highest average daily dose (619.6 mg/kg). There was no significant difference among the groups for the average daily acetaminophen dose ingested. The findings of this study suggest that water and/or gel could be used by rats for self-administration of oral acetaminophen as a post-procedural analgesic.

Abbreviations and Acronyms:

AG, acetaminophen in gel; AW, acetaminophen in water; AWG, acetaminophen in gel and water

INTRODUCTION

Regulations and performance-based standards pertaining to animal research stipulate that pain and distress must be alleviated or reduced to improve the welfare of animals being used in scientific studies. The Guide for the Care and Use of Laboratory Animals states that: “pain is a stressor and, if not relieved, can lead to unacceptable levels of stress and distress in animals.” (13) Further, the Public Health Service Policy on Humane Care and Use of Laboratory Animals states that: “Procedures that may cause more than momentary or slight pain or distress to the animals will be performed with appropriate sedation, analgesia, or anesthesia, unless the procedure is justified for scientific reasons in writing by the investigator.” (20)

Methods used to deliver analgesics in laboratory animals can be as varied as the number of animal species utilized in the research itself. Various drugs, such as acetaminophen, buprenorphine, fentanyl, and flunixin meglumine have been used in rodents with each having advantages and disadvantages. (1,3,5,14,16,18,22,23) For example, analgesics that are delivered parenterally may be more efficiently utilized when working with a smaller number of animal subjects. (5) While injectable analgesics provide the most reliable method of drug administration, their use requires more handling and disruption to the animals. (22) Conversely, if an analgesic can be self-administered orally by the animal, this can further reduce distress by eliminating handling and potentially stressful methods of administration; this method of delivery can also prevent disruption of an animal’s diurnal rhythm and that of other animals in the room. (5,8,10,22) One method of providing oral analgesics is

in the drinking water, particularly for laboratory rodents. (1,8,18) Buprenorphine has been used in the drinking water of rats (5) and it is widely used for pain control in many species due to its long-acting duration and minimal effects on the cardiovascular and respiratory systems. (23) For these reasons, buprenorphine has become one of the treatments of choice in laboratory rodents for postoperative pain. (8,12) However, since buprenorphine is a controlled substance, obtaining and storing it may be problematic for some facilities. Further, buprenorphine has also been associated with opioid-induced hyperalgesia which can occur once opioid treatment is quickly tapered or stopped. (23) For these reasons, it may be more beneficial to both the animals and the researchers to utilize an alternative treatment regimen for post-procedural pain.

Acetaminophen is a commonly used rodent analgesic, as it is inexpensive, easy to obtain, and it is not a controlled substance. It has been used effectively in the water of postsurgical rats (9) fairly extensively, though sometimes with mixed results, as neophobia (1,3,18) has been reported. The introduction of new tastes may result in an aversion to new objects/food items introduced into the home cage, leading to decreased food/water consumption. This can make the self-administration of oral medication less effective.

In addition to the administration of drugs in drinking water, many new gel products are available for rodents that can deliver various drugs and nutrients. Several studies have examined the ingestion of oral medication in the drinking water of rats (1,3,5,18,22,23) and mice (2,3), but no research has been done to evaluate the ingestion of oral medication in gel products by rats. Acetaminophen consumption may differ if provided to rodents in a gel versus the drinking water. This could provide another method of pain relief in these species, which would add to the body of knowledge on improving the welfare of laboratory animal species used in research.

The goal of this research was to evaluate the preference for consumption of self-administered acetaminophen in water and/or a gel delivery system (MediGel™) by Sprague Dawley rats following surgery. A similar study was previously performed in mice (2,3), but it has not been explored in laboratory rats. The focus of the study was not to determine whether acetaminophen provides sufficient postsurgical pain relief, but to establish whether the rats displayed a preference for medicated water, gel, or a combination following a surgical procedure. Further, the amount of acetaminophen ingested in each of the treatment groups was used to determine if the dosages ingested would be expected to provide postsurgical pain relief and to ascertain if a therapeutic dose could be reached. We hypothesized that each delivery system would be consumed equally as well and that providing both delivery methods (water and gel) would increase the likelihood that the animals received a targeted therapeutic dose.

MATERIALS AND METHODS

Subjects. Animals in this study (n=30) consisted of male Sprague Dawley rats (Rattus norvegicus). The age range was 3-6 months, and the weight range was 344-547 g, with the average weight being 482 g. The rats were obtained from Taconic Farms (Germantown, NY). The animals had not undergone any previous surgical procedures, and were singly housed during the course of the study in order to accurately measure the amount of medicated water and/or gel that was ingested individually. Animals were acclimated to single housing for 3 days prior to the start of the study.

The rats were individually housed in static polycarbonate shoebox-type cages with filter tops (Allentown, Allentown, NJ) on rodent hardwood bedding (catalog no. 7090M, Laboratory-Grade Teklad Maple SaniChips, Harlan Teklad, Madison, WI). Rats were provided pelleted rodent food (Envigo Teklad Global 18% protein 2018 Rodent Diet). In place of ad libitum tap water, rats were given acetaminophen-treated water and/or gel. The room was kept on a 12:12-h light:dark cycle (lights on, 0600: lights off, 1800) in a temperature-controlled room maintained between 68-79°F (20-26°C). Relative humidity was maintained at 33-64%. All procedures were approved by the Uniformed Services University of the Health Sciences IACUC, which is an AAALAC International accredited facility.

The health status was monitored using a sentinel program. The rat colony was negative for Mycoplasma pulmonis, Pneumocystis carinii, Toolan's H1 virus, Hantaan virus, Kilham's rat virus, lymphocytic choriomeningitis virus, mouse adenovirus, pneumonia virus of mice, rat coronavirus, sialodacryoadentis virus, reovirus, rat minute virus, rat parvovirus, rat theilovirus, Sendai virus, Aspiculuris tetraptera, Myocoptes, Radfordia/Myobia, and Syphacia muris.

Surgical Procedure. A laparotomy was performed using a modified version of an established surgical model of postoperative pain in rats. (15) Rats were induced and

maintained under anesthesia by intraperitoneal administration of ketamine (80 mg/kg) and xylazine (40 mg/kg) using a 25 gauge, ¾ inch needle. A withdrawal reflex was used to ensure an adequate depth of anesthesia prior to beginning surgery. The hair of the lower left quadrant of the abdomen was clipped and disinfected with three alternating scrubs of povidone-iodine solution followed by an alcohol wipe. A 2 cm incision was made through the abdomen 0.5 cm caudal to the last rib on the left flank of the animal. The musculature was manipulated and stretched by inserting approximately 2 cm of the index finger into the incision. Muscle layers were then closed with

4-0 Monocryl® suture in a simple interrupted pattern, and the skin edges were closed using wound clips. Immediately following the procedure, the rats were placed in a warm, dry cage and monitored until they were conscious and ambulatory.

Study Design. The rats were randomly assigned to 3 treatment groups post-surgery with n=10/group. The acetaminophen in water group (AW) received acetaminophen in filtered tap water, the acetaminophen in gel group (AG) received acetaminophen in gel, and the acetaminophen in water and gel group (AWG) received acetaminophen in both filtered tap water and gel. The day of surgery was designated as day 0 and various measurements were taken from the day prior to surgery (day -1) to three days post-surgery (days 1-3). Rats were weighed daily at approximately 0730 on day -1 and days 1-3. Water bottles were weighed the morning of day 0 and day 3. Gel cups were weighed the morning of day 0 and 24 hours after being placed into the cage. A new gel cup was weighed and placed into the cage daily for rats assigned to the AG and AWG groups; the old gel cup was also weighed and then discarded daily. The rats were provided acetaminophen treatments per assigned group immediately following surgery. The acetaminophen dose provided was calculated to fall within the recommended oral dosage range of 110 to 305 mg/kg for rats (7,8). The typical amount of water consumed daily by rats is 8-11 mL/100 g of body weight. (10) With an anticipated daily consumption of 38-52 mL of water per rat, the desired concentration of the water and gel was 2 mg/mL to achieve a targeted dose of 200 mg/kg, which falls within the therapeutic range. (2,3) The amount of water and/or gel ingested in mL was then multiplied by the acetaminophen concentration in the water or gel. To prepare the acetaminophen-treated water, 25 mL of cherry-flavored acetaminophen liquid (Children's Mapap™ Acetaminophen Liquid, Major, Livonia, MI) was added to 400 mL of filtered tap water in a standard rat water bottle. The gel (MediGel Sucralose, Clear H2O, Portland, ME) was prepared with acetaminophen according to the manufacturer's instructions. 3.7 mL of the acetaminophen liquid was drawn into a 5mL syringe and, using a 21-gauge needle, the drug was injected into the 2 oz. cup. The cup was then vigorously swirled for 10-15 seconds to ensure equal distribution of the medication throughout the gel.

Statistical analysis. The study design had 80% power to detect differences of 1.3 standard deviations. A two-way repeated measures ANOVA (version 20, SPSS, Chicago, IL) followed by a Tukey multiple-comparisons procedure was used to compare weight of the rats among the treatment groups, as well as gel consumption. A one-way ANOVA was used to compare the mean water consumption per rat, mean gel consumption per rat, and mean acetaminophen dose per rat among the 3 treatment groups. (4) Paired t test analysis was also performed to compare the difference in water and gel consumption within each group. (4) All calculations were based on a 5%, two-sided significance level.

RESULTS

Water consumption. The average daily consumption of water per rat post-surgery was 38.48 ± 5.11 mL (Table 1). Water intake was measured on day 0 and day 3, so the average intake per day was calculated for each rat. The average water consumption per rat did not differ significantly between the AW and AWG groups.

Gel consumption. The average daily consumption of gel per rat post-surgery was 51.8 ± 8.65 mL (Table 1). Gel consumption between the AG and AWG groups differed significantly (P = 0.049), with the AG group consuming more gel. The average gel consumption within the AG group differed significantly (P = 0.009) from the AWG group on day 1. The amount of gel consumed increased each day during the course of the study within both the AG and AWG groups (Table 2). Within the AWG group, the average daily consumption of gel differed significantly (P = 0.002) from day 1 to day 3.

Water and gel comparison. The overall daily consumption of gel per rat was greater than that of water (Table 1). Within the AWG group, the consumption of water and gel did not differ significantly.

Weight. The average weight of rats differed significantly within the groups when comparing day -1 weights with day 1 and also between day -1 weights compared with day 2 (Table 3). The AG and AWG groups also significantly differed within each group when comparing day 1 to day 2. The average weight of each group did not differ significantly when comparing day 2 to day 3 (Table 3). The average weights of the AG and AWG groups differed significantly when comparing day -1 weights to day 3 (Table 4). The average overall weight differed significantly (P = 0.009) between groups regardless of the time period compared.

Acetaminophen dose. Rats in the AW group ingested the lowest average daily dose of acetaminophen (350.9 mg/kg), while the AG group ingested the highest average daily dose (619.6 mg/kg). The AWG group ingested an average daily dose of 562.3 mg/kg (Figure 1). There was no significant difference among the groups for the average daily acetaminophen dose ingested. All 3 groups ingested an average daily dose that was greater than the therapeutic threshold dose of 200 mg/kg.

Table 1

Daily water and gel ingestion per rat (mL; mean ± SEM) by treatment group

	Water	Gel
Group	Daily Average	Daily Average
AW	42.25 ± 7.94	-
AG	-	71.1 ± 13.63
AWG	34.7 ± 6.65	32.5 ± 6.86
Overall	38.48 ± 5.11	51.8 ± 8.65[a]

[a]Significant ($P = 0.049$) difference of gel ingestion between AG and AWG groups.

Table 2

Daily intake of water and gel per rat (mean ± SEM) by treatment group

	Water	Gel		
Group		Day 1[a]	Day 2	Day 3
AW	42.25 ± 7.94		-	
AG	-	69.8 ± 17.22	72.4 ± 12.8	72.8 ± 9.72
AWG	34.7 ± 6.65	16.8 ± 5.83	48.2 ± 11.34	66.8 ± 9.56[b]
Overall	38.48 ± 5.11	43.3 ± 10.74	60.3 ± 8.77	69.8 ± 6.67

[a]The average gel consumption within the AG group differed significantly (P = 0.009) from the AWG group day 1.
[b]Within the AWG group, the average daily consumption of gel differed significantly ($P = 0.002$) from day 1 to day 3.

Table 3

Average weight of rat per treatment group

Table 3. Average weight (g) of rat per treatment group[a,b]

					P			
Group	Day -1	Day 1	Day 2	Day 3	Day -1 compared with Day 1	Day -1 compared with Day 2	Day 1 compared with Day 2	Day 2 compared with Day 3
AW	500.8 ± 8.66	484.3 ± 8.07	485.6 ± 9.45	488.6 ± 10.93	**<0.001**	**0.002**	0.57	0.31
AG	460.3 ± 17.1	434.5 ± 14.78	425.9 ± 11.86	425.9 ± 12.73	**<0.001**	**<0.001**	**0.03**	0.99
AWG	483.9 ± 10.99	467.5 ± 9.73	462.1 ± 9.71	463.6 ± 11.25	**0.002**	**<0.001**	**0.005**	0.54

[a]Significant *P* values ($P < 0.05$) are indicated in bold type.
[b]Significant ($P = 0.009$) difference of average overall weight between groups regardless of time period.

Table 4

Comparison of average weight (g) of rat within treatment groupa,b

			Mean difference	95% Confidence Interval for difference		P
Group	Time *i*	Time *j*	Mean difference	Lower Bound	Upper Bound	
AG	Day -1	Day 1	25.8	15.8	35.8	**<0.001**
		Day 2	34.4	20.67	48.13	**<0.001**
		Day 3	34.4	17.68	51.12	**<0.001**
	Day 1	Day -1	-25.8	-35.8	-15.8	
		Day 2	8.6	1.59	15.61	**0.01**
		Day 3	8.6	-5.01	22.21	0.41
	Day 2	Day -1	-34.4	-48.13	-20.67	**<0.001**
		Day 1	-8.6	-15.61	-1.59	**0.01**
		Day 3	0	-10.75	10.75	1.00
	Day 3	Day -1	-34.4	-51.12	-17.68	**<0.001**
		Day 1	-8.6	-22.21	5.01	0.41
		Day 2	0	-10.745	10.75	1.00
AW	Day -1	Day 1	16.5	6.5	26.5	**<0.001**
		Day 2	15.2	1.47	28.93	**0.02**
		Day 3	12.2	-4.52	28.92	0.256
	Day 1	Day -1	-16.5	-26.5	-6.5	**<0.001**
		Day 2	-1.3	-8.31	5.71	0.996
		Day 3	-4.3	-17.91	9.31	0.942
	Day 2	Day -1	-15.2	-28.93	-1.47	**0.02**
		Day 1	1.3	-5.71	8.31	0.996
		Day 3	-3	-13.75	7.75	0.967
	Day 3	Day -1	-12.2	-28.92	4.52	0.256
		Day 1	4.3	-9.31	17.91	0.942
		Day 2	3	-7.75	13.75	0.967
AWG	Day -1	Day 1	16.4	6.4	26.4	**<0.001**
		Day 2	21.8	8.07	35.53	**0.001**
		Day 3	20.3	3.58	37.02	**0.01**
	Day 1	Day -1	-16.4	-26.4	-6.4	**<0.001**
		Day 2	5.4	-1.61	12.41	0.206
		Day 3	3.9	-9.7	17.51	0.963
	Day 2	Day -1	-21.8	-35.53	-8.07	**0.001**
		Day 1	-5.4	-12.41	1.61	0.206
		Day 3	-1.5	-12.25	9.25	0.999
	Day 3	Day -1	-20.3	-37.02	-3.58	**0.01**
		Day 1	-3.9	-17.51	9.71	0.963
		Day 2	1.5	-9.25	12.25	0.999

[b]Significant ($P = 0.009$) difference of average overall weight between groups regardless of time period.
[a]Significant *P* values ($P < 0.05$) are indicated in bold type.

Figure 1

Daily average acetaminophen dose per rat in each treatment group by gel and/or water consumption (mg/kg; mean ± SEM). AW group is depicted by circles, AG group is depicted by squares, and AWG group is depicted by triangles. There was no significant difference in average daily acetaminophen dose between the treatment groups.

DISCUSSION

The purpose of this study was to provide acetaminophen in

two different delivery systems to rats after a surgical procedure to determine if the rats displayed a preference for one form of acetaminophen delivery over the other. A laparotomy was performed as a surgical model of postoperative pain to provide a representation of acetaminophen analgesic efficacy. (5)

Neophobia has been observed in other studies in which acetaminophen was provided in drinking water to alleviate pain. (1,3,18,22) Based on the average daily water consumption by rats of 8-11 mL/10g of body weight (10), it was expected that the rats would consume 38-52 mL of water and/or gel per day. Each of the three treatment groups fell within or exceeded this expected range. The AG group had the highest consumption with a daily average of 71.1 ± 13.63 mL. It was not unexpected that the AWG group consumed more than the AW group (67.2 ± 13.51 and 42.25 ± 7.94 mL, respectively), given that these animals had two sources of hydration available to them and that similar results were observed in comparable study involving mice (3). Based on these data, neophobia did not appear to be a factor. This may have been due to the novelty of the treated gel and water leading the rats to possibly ingest it as a form of enrichment. In a study by Bauer et al, 75% of male rats displayed neophobia for 24 hours, but female rats displayed no neophobia at all. (1) Since all of the subjects in this study were male, the initial consumption of water and gel could have been increased if the test subjects had been female.

Regardless of gender, the animals in this study consumed sufficient water and gel to prevent dehydration and to provide therapeutic doses of acetaminophen. It is important to note that there was no significant difference among three treatment groups, so a preference of delivery system was not observed. With ingested doses ranging from 350.9 mg/kg (AW group) to 619.6 mg/kg (AG group), all three treatment groups reached the targeted therapeutic dose of 200 mg/kg. Further, because acetaminophen has a recommended oral dosage range of 110-305 mg/kg for rats (6-8), the three treatment groups all surpassed the higher end of the therapeutic range. Depending on the length of time that rats are administered acetaminophen at this dose for a potential study, toxicity concerns could be raised, though a toxic level of acetaminophen in rats may be challenging to ascertain. The primary toxicity of acetaminophen metabolism occurs in hepatic tissues (11) and an overdose may cause damage in renal tissues by decreasing antioxidant enzymes. (4,21) In a previous study with a similar design to this one (3), mice ingested what may have appeared to be toxic levels of acetaminophen, but because the drug was ingested over a 24-hr period, toxicity was likely less of an issue. Further, it has been known for some time that rats are resistant to the liver-damaging effects of acetaminophen and are actually not a suitable model species for hepatotoxicity. (19) This was illustrated in a study by McGill et al. in which Sprague Dawley rats were administered acetaminophen at doses of 1.0, 1.5, and 2.0 g/kg of body weight resulting in little to no liver injury observed histologically. (17) The lowest of the doses given in the McGill study was 50% higher than the highest ingested dose observed in this study. Despite discernable clinical signs of hepatotoxicity, it is advisable to stay within published therapeutic dose ranges of acetaminophen for rats to be able to accurately compare results across published literature.

The weights of the rats in each of the treatment groups significantly decreased the day following surgery. This was not unexpected, as previous studies have demonstrated a decrease in food consumption by rats following acetaminophen administration. (1) By day 2, the weight of the rats in the AW group started to increase, and by day 3 there was not a significant difference in weight from before surgery. By the end of the study, there was no significant difference in any of the three treatment groups when comparing the weight of rats within each group at day 3 to day 2. The weights of the rats in the AW group increased more quickly than that of rats in the other groups. Conversely, the rats in the AG group had the slowest amount of weight gain by day 3 among the three groups despite having ingested the greatest average daily amount of an acetaminophen vehicle. Even though the rats in the AG group had the greatest hydration, the acetaminophen dose may have been high enough and lead to appetite suppression such that the weights of the rats did not improve as quickly as in the other two treatment groups. There was a significant difference in weights of the AG and AWG groups when comparing day 3 to day -1 weights, but the difference was less significant in the AWG group. In long term studies, it may be beneficial to provide both acetaminophen-treated water and gel, as a preference was not observed and this may allow for an increased rate of weight gain beyond what was observed when the rats were provided gel alone.

Another outcome that was observed for the AG and AWG groups involved the gel cups themselves. The gel cups were left in the cage for 24 hours and then were replaced each day

throughout the duration of the study, for a total of 3 gel cups being placed into each of the assigned cages. Over the course of the three days post-surgery, several were noted to have been chewed or shredded. This resulted in small fragments of plastic being found throughout the bedding. Pieces were removed as much as possible whenever a new gel cup was placed into a cage. For future studies, it is advisable to replace the cups more frequently, i.e. every 12 hours, to prevent the possible ingestion of foreign material, as this could have deleterious effects on the health and well-being of the research animals, though this was not observed in this study.

In summary, the findings of this study support the hypothesis that each delivery system would be consumed equally as well. Providing both delivery methods (water and gel) together did not necessarily increase the likelihood that the rats received a targeted therapeutic dose of acetaminophen at 200 mg/kg, as all 3 treatment groups exceeded this dose, with the AG group receiving the highest dose. Similar studies in mice have recommended the use of only one delivery system to avoid potential toxicity (3); however, in this study, using only the gel delivery system resulted in the highest acetaminophen dosage consumed. Therefore, any of these delivery methods could be suitable to provide oral acetaminophen at a concentration lower than 2 mg/mL to achieve a targeted therapeutic dose that still falls within the published dosage range of 110-305 mg/kg.

ACKNOWLEDGMENTS

We especially thank Efrain Salazar, Cara Reiter, Amory Koch, Nicole Rowley, Mike Junio, Jay Andrews, Grady Cline, Ally Oliver, Justin Brown, and Cara Olsen for their technical assistance in completing this project. The opinions, interpretations, conclusions, and recommendations are these of the authors and are not necessarily endorsed by the Uniformed Services University of the Health Sciences, the Department of Defense, or the United States Federal Government.

References

1. Bauer DJ, Christenson TJ, Clark KR, Powell SK, Swain RA. 2003. Acetaminophen as a postsurgical analgesic in rats: a practical solution to neophobia. Contemporary Topics 42(2):20-24.
2. Brunell MK, Olsen CH, Christy AC, Maxwell BM, Bentzel DE. 2017. Evaluation of consumption of self-administered acetaminophen in drinking water and two gel delivery systems in C57BL/6 mice. Internet Journal of Veterinary Medicine 14(1). doi: 10.5580/IJVM.52393.
3. Christy AC, Byrnes KR, Settle TL. 2014. Evaluation of medicated gel as a supplement to providing acetaminophen in the drinking water of C57BL/6 mice after surgery. Journal of the American Association for Laboratory Animal Science 53(2):180-184.
4. Cohen SD, Hoivik DJ, Khairallah EA. Acetaminophen-induced hepatotoxicity. In: Toxicology of the Liver. Plaa GL, Hewitt WR, editors. Philadelphia (PA): Taylor and Francis.
5. Cooper DM, DeLong D, Gillett CS. 1997. Analgesic efficacy of acetaminophen and buprenorphine administration in the drinking water of rats. Contemporary Topics 36(3): 58-62.
6. Fish RE, Brown MJ, Danneman PJ, Karas AZ, editors. 2008. Anesthesia and analgesia in laboratory animals, 2nd ed. New York (NY): Elsevier.
7. Flecknell PA. 1984. The relief of pain in laboratory animals. Laboratory Animals 18:147-160.
8. Flecknell PA. 1984. Laboratory animal anesthesia. London (UK): Academic Press.
9. Flecknell PA, Orr HE, Roughnan JV. 1999. Comparison of the effects of oral or subcutaneous carprofen or ketoprofen in rats undergoing laparotomy. Veterinary Record 144:65-67.
10. Fox JG, Anderson LC, Otto GM, Pritchett-Corning KR, Whary MT, editors. 2015. Laboratory animal medicine. 3rd ed. San Diego (CA): Academic Press.
11. Gu J, Cui H, Behr M, Zhang L, Zhang QY. 2005. In vivo mechanisms of tissue-selective drug toxicity: effects of liver-specific knockout of the NADPH-cytochrome P450 reductase gene on acetaminophen toxicity in kidney, lung and nasal mucosa. Molecular Pharmacology 67: 623-630.
12. Hubbell JA, Muir WW. 1996. Evaluation of a survey of diplomates of the American College of Laboratory Animal Medicine on use of analgesic agents in animals used in biomedical research. Journal of the American Veterinary Medical Association 209: 918-921.
13. Institute for Laboratory Animal Research. 2011. Guide for the care and use of laboratory animals. 8th ed. Washington (DC): National Academies Press.
14. Liles JH, Flecknell PA. 1993. The effects of surgical stimulus on the rat and the influence of analgesic treatment. British Veterinary Journal 149: 515-525.
15. Martin TJ, Buechler NL, Kahn W, Crews JC, Eisenach MD. 2004. Effects of laparotomy on spontaneous exploratory activity and conditioned operant responding in the rat: a model for postoperative pain. Anesthesiology 101: 191-203.
16. Matsuyima 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. Journal of the American Association for Laboratory Animal Science 51:42-49.
17. McGill MR, Williams CD, Xie Y, Ramachandran A, Jaeschke H. 2012. Acetaminophen-induced liver injury in rats and mice: comparison of protein adducts, mitochondrial dysfuntion, and oxidative stress in the mechanism of toxicity. Toxicology and Applied Pharmacology 264: 387-394.
18. 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). Journal of the American Association for Laboratory Animal Science 45(5): 48-54.
19. Mitchell JR, Jollow DJ, Potter WZ, Davis DC, Gillette JR, Brodie BB. 1973. Acetaminophen-induced hepatic necrosis. I. Role of drug metabolism. Journal of Pharmacology and Experimental Therapeutics 187:185–194.

20. National Institutes of Health. 2015. Public health service policy on humane care and use of laboratory animals. Office of Laboratory Animal Welfare.
21. Roy S, Pradhan S, Das K, Mandal A, Mandal S, Patra A, Samanta A, Sinha B, Nandi DK. 2015. Acetaminophen induced kidney failure in rats: a dose response study. Journal of Biological Sciences 15: 187-193.
22. Speth RC, Smith MS, Brogan RS. 2001. Regarding the inadvisability of administering postoperative analgesics in the drinking water of rats (Rattus norvegicus). Contemporary Topics 40(6): 15-17.
23. Stewart LA, Martin WJ. 2003. Evaluation of postoperative analgesia in a rat model of incisional pain. Contemporary Topics 42(1):28-34.

Author Information

Laura E. Riddle
Center for Laboratory Animal Medicine, Uniformed Services University of the Health Sciences
Bethesda, Maryland

Sorana Raiciulescu
Department of Preventive Medicine and Biostatistics, Uniformed Services University of the Health Sciences
Bethesda, Maryland

Anna B. Mullins
Center for Laboratory Animal Medicine, Uniformed Services University of the Health Sciences
Bethesda, Maryland

Chad D. Foster
Center for Laboratory Animal Medicine, Uniformed Services University of the Health Sciences
Bethesda, Maryland