

Evaluation of a fracture pain model in domestic pigeons (*Columba livia*)

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Objective—To validate a model of postfracture pain in perching birds.

Animals—21 adult domestic pigeons (*Columba livia*).

Procedures—In each bird, a standardized osteotomy of 1 femur was performed and the fracture was immobilized with an intramedullary pin. Degree of postoperative pain was evaluated 6 times/d for 4 days by use of 3 methods: an electronic perch for assessment of weight-bearing load differential of the pelvic limbs, 4 numeric rating pain scales for assessment of pain (all of which involved the observer in the same room as the bird), and analysis of video-recorded (observer absent) partial ethograms for bird activity and posture. Measurements obtained were compared with data collected before the surgery to evaluate the ability of these methods to detect pain.

Results—The weight-bearing load differential was a sensitive, specific, reliable, and indirect measure of fracture-associated pain in the model used. Two of 4 tested pain scales (fractured limb position and subjective evaluation of degree of pain) were sensitive and specific for detecting pain and were reliable in a research setting. Interobserver reliability of the 4 pain scales was excellent. Partial ethograms were sensitive for identifying pain-associated behavior in pigeons, particularly during the first 2 days after surgery.

Conclusions and Clinical Relevance—The fracture pain model was reliable and reproducible and may be useful for experimental studies involving postsurgical pain in pigeons. Weight-bearing load differential was the most sensitive and specific means of determining degree of pain in pigeons during the first 4 days after hind limb fracture induction. (*Am J Vet Res* 2012;73:353–360)

Relieving pain in animals is an integral part of veterinary medicine, yet major gaps have been identified in our knowledge of animal pain and analgesia.¹ The lack of species-specific information in the avian literature is of great concern. Indeed, pain assessment in birds is a challenge for avian veterinarians. Painful conditions such as traumatic wing or limb fractures are common in wild and companion birds. Appropriate recognition and evaluation of the degree of pain are therefore important to provide appropriate analgesia.

Several models and pain assessment methods have been used in avian research. In poultry, for example,

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ABBREVIATION

DBW Distribution of body weight

the ability to cross an obstacle course, degree of activity, scores of lameness, and self-selection of an analgesic drug have all been used to assess lameness-associated pain.^{2–5} In dogs, use of a force plate is the reference standard for orthopedic research.⁶ The force plate technique has been used in chickens⁷ but is limited to ground-dwelling birds and has not yet been shown to be useful for analgesia studies.

Various nociceptive mechanical stimuli such as crest or digit pinching and feather removal have been used in conscious or anesthetized birds.^{8–10} Thermal and mechanical stimuli have also been used to evaluate the effect of administration of various opioids on pain threshold; however, responses to these types of stimuli can differ widely among birds.^{11–13} Quantifiable electric stimuli, which induce more predictable reactions than do the other types of stimuli, have been used in some studies^{11–14}; however, electrical stimulation induces nerve depolarization that is not specific to nociceptors. The limitation of experimental models that involve use of thermal, mechanical, or electric noxious stimuli is that these stimuli do not mimic clinical syndromes of pain; therefore, extrapolation of results from such stud-

ies to predict the degree of surgical and inflammatory pain in birds should be done carefully.

The analgesic efficacy of opioids has been evaluated by determining their influence on the minimum dose of isoflurane required to provide anesthesia in birds.^{15,16} However, dose reduction following drug administration does not necessarily indicate analgesia has been achieved because the drug used could also have nonanalgesic properties such as sedation that might also reduce the minimum anesthetic dose.¹⁵

Models of experimentally induced, short-term gout-like arthritis have been used for assessment of acute pain in chickens, pigeons, and parrots.^{17–21} Partial amputation of the beak has been used as a chronic pain model.^{22,23} Yet, despite the fact that several nociceptive experimental models have been reported for birds, few of these models have been thoroughly validated. Most such experimental models are designed for evaluating the analgesic effects of opioids and are often not useful in evaluating the effects of NSAIDs.

Mechanisms that drive fracture-associated pain are complex in mammals. The initial pain that follows fracture of the femur in humans is most commonly described as intense and is mainly caused by mechanical activation of mechanosensitive nociceptors (C and A- δ nerve fibers) that innervate the periosteum, mineralized bone, and marrow.^{24,25} On the other hand, avian femurs are pneumatic, and the vascularization and innervation differ from those in mammalian femurs. Nevertheless, it could be assumed that, as for mammals, stabilization of a fracture site by internal or external fixation results in a considerable attenuation of fracture-associated pain in birds.²⁶

Minutes to hours after an initial trauma, marked influx of inflammatory cells into the fracture site results in the activation of nociceptors that express receptors for acidity, cytokines, chemokines, and inflammatory factors (eg, bradykinin, nerve growth factor, or prostaglandins).²⁷ Consequently, drugs such as NSAIDs that affect prostaglandin production are useful to control fracture-associated pain.²⁵ Sensitization of the peripheral and central nervous systems may also occur after a fracture occurs, and this sensitization can be responsible for chronic pain that may be debilitating, particularly for wild birds.²⁶

Because the experimental models reported in the avian literature cannot reproduce these complex pain mechanisms, a specific fracture pain model would be useful to adequately assess the analgesic effects of drugs on postoperative orthopedic pain in birds. Fracture pain models have been developed in mammals,^{26,28} helping to increase our understanding of the mechanisms that generate and maintain fracture-associated pain and to translate these findings into the development of new analgesic treatments.²⁸ Given that birds differ from mammals in many anatomic and physiologic characteristics, creation of a fracture pain model in birds is needed to assess this common type of pain.

In mammalian pain models recently described, both femurs and tibias were used.^{26,28} The femur alone is a suitable bone for fracture pain modeling in birds because surgical reduction and immobilization of experimentally induced fractures with an intramedullary

pin can be easily standardized. Such a model would enable the use of differential weight bearing as an indirect measure of the degree of pain in affected birds. The degree of weight bearing on a fractured pelvic limb is recognized as a clinically relevant tool for evaluating fracture-associated pain because the ability of a subject to voluntarily bear weight on an affected extremity is commonly used in human and veterinary medicine as a measure of successful bone healing during and after rehabilitation.^{26,29}

The objectives of the study reported here were to validate a model of pelvic limb fracture in domestic pigeons (*Columba livia*) and to estimate construct (convergent) and predictive (pain present or absent) validities as well as the reliability (repeatability and interobserver reproducibility) of various quantitative methods of postoperative pain assessment in pigeons.

Materials and Methods

Animals—Twenty-one male domestic pigeons were used in the study. Male birds were chosen to decrease intraindividual variation in femoral density associated with physiologic osteomyelosclerosis in female birds.³⁰ Birds were allowed to acclimatize to the environment for 1 week before the study began. They were housed individually in cages with a solid floor covered with 5 cm of soft bedding during the preoperative period and the first 10 days after surgery. Afterward, the birds were assembled into large aviaries for the remainder of the study. A cycle of 12 hours of light and 12 hours of dark was used throughout the study. Birds were fed a pelleted diet and provided ad libitum access to water. All were considered healthy on the basis of unremarkable results of a complete physical examination, CBC, serum biochemical profile, and fecal examination.³¹ All procedures were approved by the Faculté de Médecine Vétérinaire Animal Care and Use Committee, which operates under the auspices of the Canadian Council on Animal Care.

Fracture induction and repair—All pigeons received an IM injection of 1 mg/kg of butorphanol.^a Afterward, anesthesia was gradually induced via face mask by use of isoflurane^b delivered in oxygen through a nonrebreathing Bain system. Once muscular relaxation was apparent (assessed as a loss of muscle tone in the pelvic limbs and jaw), endotracheal intubation was performed with a Magill uncuffed endotracheal tube (diameter, 3 to 4.5 mm). The concentration of isoflurane in oxygen was adjusted as needed in accordance with the bird's response to surgical stimuli.

Birds were manually ventilated to keep end-tidal CO₂ values between 30 and 45 mm Hg. Heart rate was monitored with an ultrasonic Doppler probe^c applied on a palatine artery. A circulating hot water blanket was used to provide supplemental heat for the duration of the anesthetic period, and body temperature was monitored with a cloacal thermometer. The use of transparent vinyl surgical drapes^d facilitated maintenance of an adequate cloacal temperature.

After aseptically prepared the skin overlying the left femur, an incision of approximately 10 mm was made in the skin along the lateral aspect of the thigh.

The femur was surgically approached by smooth dissection between the iliotalialis lateralis and iliofibularis muscles. A mid-diaphyseal, 45° oblique fracture of the femur was produced by use of an oscillating bone saw^e (5.5 × 11.5 × 0.38 mm). Normograde intramedullary pin insertion was used to reduce the fracture. Muscles were then reapposed with a simple interrupted suture pattern, and the skin incision was closed by use of a simple continuous suture pattern.^f All pigeons received butorphanol (1 mg/kg, IM) 4 and 8 hours after anesthetic induction. All surgeries were performed by the same investigator (MD) to control for differences in surgeon technique or skill.

Experimental design—To accommodate the recommendations of the Canadian Council on Animal Care for reducing the number of animals used in experiments, the pain model was performed in parallel with a study³² on the efficacy of an NSAID. Therefore, immediately after fracture induction and repair, pigeons were randomly allocated to 1 of 3 groups of 7 birds each. Two groups received an NSAID^g that differed only in dosage, and 1 group received an equal volume of saline (0.9% NaCl) solution IM. Details regarding the NSAID treatment and resulting data are reported elsewhere.³²

For validation of the pain model, the status of all pigeons before fracture induction and repair (ie, baseline) was used to represent pain-free birds. Pigeons from the group that did not receive an NSAID were used as negative control birds in that they were expected to have pain. Because this study involved a species for which the effectiveness of other common analgesics has not yet been determined, the recommended³³ use of a positive control group that received a drug with established efficacy in place of a negative control (saline solution) group was not feasible.

Study endpoints such as anorexia, weight loss, severe lethargy, and reluctance to move were evaluated in consultation with the institutional animal care and use committee. A protocol was used in which birds that had these signs would be euthanized or provided with medical support including rescue analgesia.

Pain assessment—Measurements were obtained the week before surgery and during the first 4 days after surgery. Differences in the distribution of weight bearing between the right (termed intact and contralateral) pelvic limb and the left (termed fractured and ipsilateral) pelvic limb were assessed by placing each bird on a perch mounted on an incapacitance meter^h modified for birds (Figure 1). Distribution of body weight was calculated as a percentage of total weight bearing by use of the following equation:

$$\text{DBW} = (\text{weight placed on contralateral limb} - \text{weight placed on ipsilateral limb}) / (\text{weight placed on contralateral limb} + \text{weight placed on ipsilateral limb})$$

A DBW of 0% corresponded to a bird that bore weight equally on both pelvic limbs, whereas a DBW of 100% indicated that the bird bore no weight on its surgically altered limb (or that it bore 100% of its weight on the contralateral limb).

Baseline measurements were obtained by measuring DBW at 6 time points every day (8:05 AM, 9:05 AM, 11:05 AM, 2:05 PM, 5:05 PM, and 8:05 PM) for 2 days prior to surgery. Postoperative DBW was measured 6 times/d for the first 4 days after surgery by use of the same schedule used before surgery. At each time point, 12 measurements were obtained during the preoperative period and 6 measurements during the postoperative period, with each measurement lasting 10 seconds. The mean of all 6 (postoperative) or 12 (preoperative) measurements was used as the value for each time point. When a pigeon left the perch during the 10-second measurement period, a test was repeated to obtain the same number of measurements for each bird. Two independent observers, who were unaware of each other's results, were used to determine interobserver reliability in a subsample (12%) of randomly chosen measurements during the preoperative period.

Four numeric rating pain scales were used to assess postoperative pain (Appendix). For each pain scale, a score of 0 represented the lack of behavior suggesting pain and a score of 5 corresponded to the presence of behavior suggesting intense pain. On the 2 days before surgery, baseline scores were assigned by 1 of 2 observers 6 times/d (8:00 AM, 9:00 AM, 11:00 AM, 2:00 PM, 5:00 PM, and 8:00 PM) with birds still in their cages. Postoperative scores were obtained 6 times/d (same schedule as baseline) for the first 4 days after surgery. Assessments were dually performed by 2 independent observers, who were unaware of each other's results, on a subsample (14%) of randomly chosen measurements from the preoperative and postoperative periods.

Twenty-minute video recordings were also obtained in the absence of any observer 6 times/d (at 7:40 AM,



Figure 1—Photograph of an incapacitance meter modified for birds. For each measurement, domestic pigeons (*Columba livia*) were placed on a perch composed of 2 separated sections, each linked to a scale to measure weight bearing on each limb.

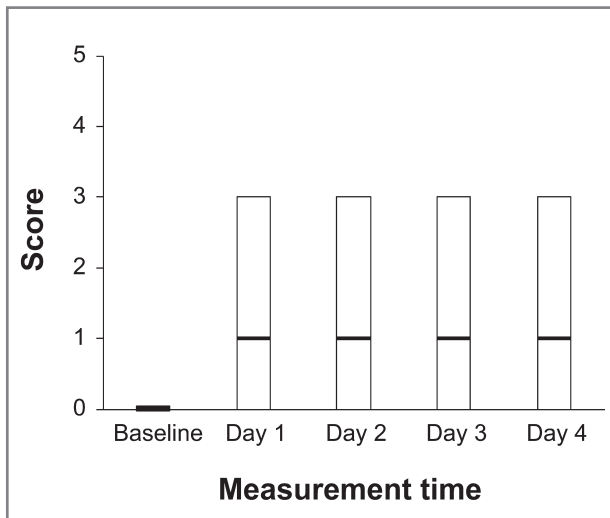


Figure 2—Attitude scores for 7 pigeons that underwent experimental fracture induction and repair in 1 limb and received no subsequent analgesic. Scores were assigned in the presence of the observer before fracture induction (baseline) and on days 1 to 4 after fracture induction. A score of 0 represented the lack of behavior suggesting pain, and a score of 5 corresponded to the presence of behavior suggesting intense pain. Lowest and highest values in the boxes represent the minimum and the maximum pain scores, respectively. Horizontal lines in the boxes represent median values.

8:40 AM, 10:40 AM, 1:40 PM, 4:40 PM, and 7:40 PM) on the 2 days preceding (baseline) and the 4 days following surgery. The last 10 minutes of each recording was analyzed. Twenty sequences of 30 seconds each were evaluated at each time point. The partial ethograms used for video analysis consisted of describing the activity of the bird and its posture. One of the 4 following behaviors was attributed to each sequence as being the main activity of a bird during these 30 seconds: exploring the environment, resting, eating or drinking, or preening. In parallel, 1 of the 4 following postures was similarly recorded for each 30-second period as representing bird posture throughout most of the period: perching, lying on the perch, lying on the floor, or standing on the floor. Results from 2 independent observers were compared to assess interobserver reliability in a subsample (12%) of randomly chosen observations made during the baseline and postoperative periods. Observers were again unaware of each other's results, and video assessments were never performed at the same time by both observers.

Euthanasia—Pigeons were euthanized 21 days after surgery by IV administration of euthanasia solutionⁱ (0.3 mL/kg) while birds were anesthetized with isoflurane. Lateral and ventrodorsal radiographic views of birds were obtained. A complete necropsy, including histologic evaluation of major organs and tissues, was performed on each pigeon.

Statistical analysis—The 2 independent observers were not blinded to period (preoperative vs postoperative) but were blinded to the treatment administered during the postoperative period. Interobserver agreement with respect to DBW, each numeric pain score (for attitude, observed fractured limb position, overall

assessment, and motor activity), and the 2 partial ethogram variables (activity and posture) was assessed from the previously described subsamples for each variable via Bland-Altman analysis and *t* test comparisons. Intraobserver agreement was assessed only for DBW by comparing 2 sets of DBW measurements obtained by the same observer at 30-second intervals and was also analyzed with the Bland-Altman technique.

To establish the number of DBW trials needed to obtain reliable values, mean values obtained from 6 randomly selected measurements were compared with the mean values of 12 measurements by use of the Bland-Altman technique. This comparison was possible only in the preoperative period. To assess how sensitive the various methods were for evaluating analgesic efficacy (ie, assessing the degree of pain), postoperative values (pain present) were compared with preoperative values (baseline; pain absent) for pigeons in the saline solution group. The ability of the methods to identify a lack of pain (ie, how specific the methods were) was tested only on the expression of values for each variable during the preoperative period.

Values of weight bearing and partial ethogram variables were analyzed by use of repeated-measures linear models, with group as a between-subject factor and time as a within-subject factor. The Bonferroni sequential procedure was used to adjust α values when multiple pairwise comparisons were made. Differences between bird groups, among observers, and within observers with respect to the 4 sets of pain-scale scores were analyzed by use of the Cochran-Mantel-Haenszel test followed by post hoc comparisons. Statistical analyses were performed by use of statistical software.^j Values of $P < 0.05$ were considered significant for all analyses. Results are reported as mean \pm SD.

Results

Animals—Data from 3 of the 21 pigeons used in this study were removed from the data set because of an error in sex determination (2 birds) or minor postoperative migration of the intramedullary pin (1 bird). The 2 birds with incorrect sex determination were removed in the preoperative period. The bird with pin migration was still included in the baseline data for pain scale validation because it still fit the criteria for the initial analysis. Therefore, 19 pigeons were included in the baseline data, 7 were included in the control group (no NSAID during the first 4 days after fracture induction and repair), 6 were included in the low-dose NSAID group, and 5 were included in the high-dose NSAID group.

Fracture pain model—The surgical approach used was easy to perform, and only minor trauma to the soft tissues was evident. The mean \pm SD duration of surgery was 14.4 ± 4.2 minutes. The mean interval from mask anesthetic induction to anesthetic recovery (indicated by the bird standing) was 34.8 ± 10.0 minutes. All birds from the first to the fourth postoperative day had a hematoma measuring approximately 1 cm at the fracture site. Surgical wound healing was without complication, and sutures were removed 1 week after the surgery.

Bone healing was complete by 3 weeks after fracture induction and repair in all birds.

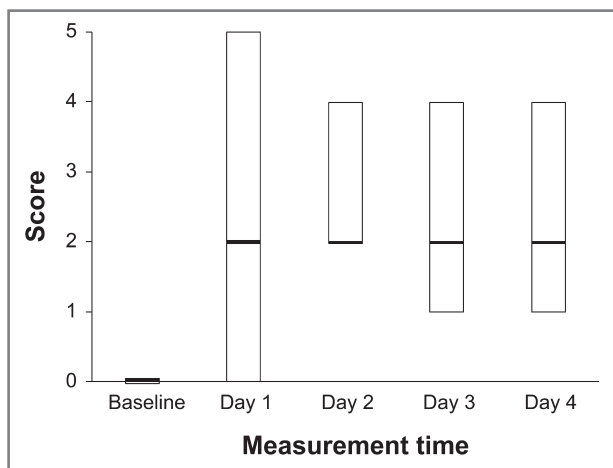


Figure 3—Weight-bearing scores for the pigeons in Figure 2. A bird's apparent ability to bear weight on the fractured limb was evaluated in the presence of the observer. See Figure 2 for remainder of key.

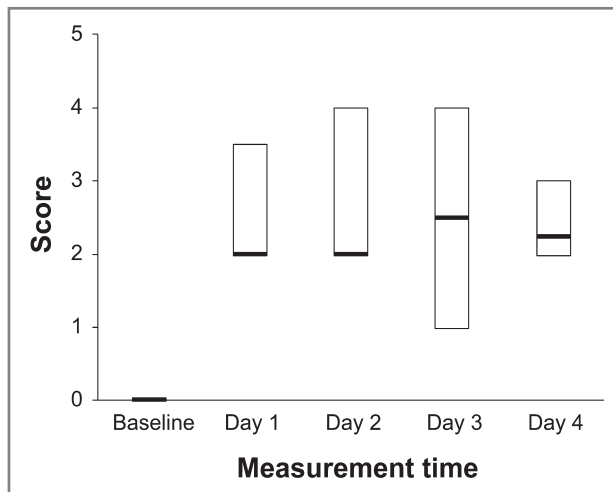


Figure 4—Subjective pain (overall assessment) scores for the pigeons in Figure 2, as evaluated in the presence of the observer. See Figure 2 for remainder of key.

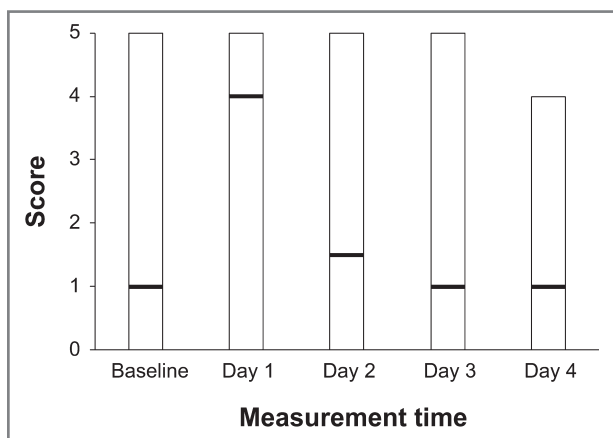


Figure 5—Motor activity scores for the pigeons in Figure 2. Degree of motor activity was assessed by review of 10-minute video recordings made at various assessment points. See Figure 2 for remainder of key.

Weight bearing—Mean DBW was higher during the postoperative period ($71.3 \pm 14.7\%$) than during the preoperative period (baseline; $-0.4 \pm 5.3\%$) for the 7 pigeons in the control group. Mean DBW was not significantly ($P = 0.85$) different from 0 during the preoperative period, although it was significantly ($P < 0.001$) different from 0 for each postoperative day, suggesting the responsiveness of the weight-bearing method to detect fracture-associated limb impairment.

Bland-Altman analysis revealed no systematic bias between mean DBW as calculated on the basis of 6 versus 12 measurements obtained during the preoperative period. Results of the test-retest Bland-Altman analysis showed no significant intraobserver bias for either of the 2 observers (mean difference for observer 1, $0.4 \pm 6.7\%$; mean difference for observer 2, $1.1 \pm 5.7\%$). Therefore, intraobserver agreement and interobserver reproducibility were judged as excellent, with a mean difference between the 2 observers of 0.4% and a 95% confidence interval of the differences of -5.2% to 6%.

Numeric rating pain scales—All pain scales used were specific, allowing identification of the absence of

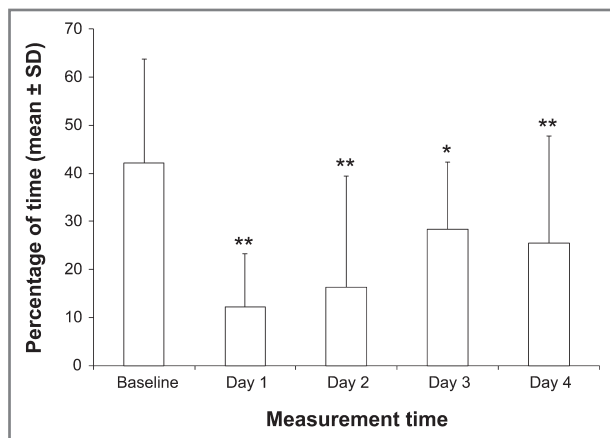


Figure 6—Percentage of 30-second sequences during which exploring the environment was the dominant observed behavior in the birds in Figure 2. *Value differs nonsignificantly (Bonferroni adjusted $P > 0.05$) from baseline value. **Value differs significantly (Bonferroni adjusted $P < 0.05$) from baseline value.

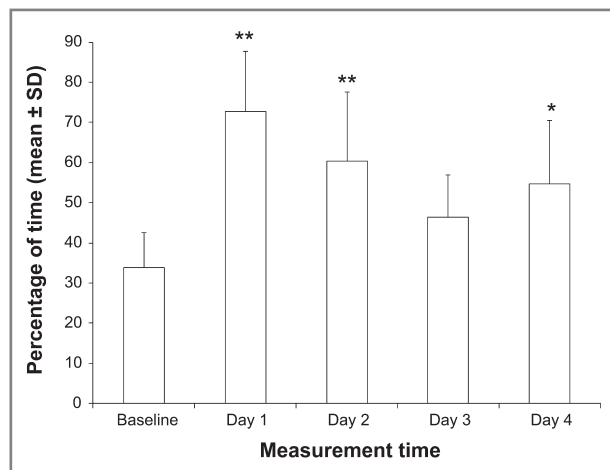


Figure 7—Percentage of 30-second sequences during which resting was the dominant observed behavior in the birds in Figure 2. See Figure 6 for key.

limb impairment (and probable pain). Scores for all baseline measurements were 0 except those for pigeon motor activity during 10 minutes of video recording (Figures 2–5). The scoring method that was most sensitive for evaluating pain was the observed position of the fractured limb as assessed in the presence of the observer, followed by subjective evaluation of the degree of pain (overall assessment). For both of these methods, all postoperative scores were ≥ 1 .

Perfect interobserver agreement was obtained for pain scores assigned during the preoperative period. For each of the 4 scales, *t* tests revealed no significant ($P > 0.05$) differences in scores between the 2 observers.

Partial ethograms—The degree of exploratory behavior was significantly lower on postoperative days 1 ($P < 0.001$), 2 ($P < 0.001$), 3 ($P = 0.006$; not significant after Bonferroni correction), and 4 ($P = 0.001$; Figure 6) than at baseline. The percentage of time spent resting was also significantly higher on postoperative days 1 ($P < 0.001$), 2 ($P < 0.001$), and 4 ($P = 0.003$; not significant after Bonferroni correction; Figure 7) than at baseline. The 7 pigeons in the control group spent

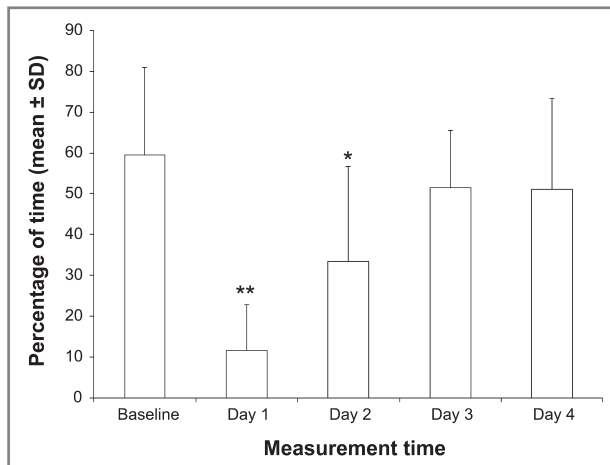


Figure 8—Percentage of 30-second sequences during which perching was the dominant observed behavior in the birds in Figure 2. See Figure 6 for key.

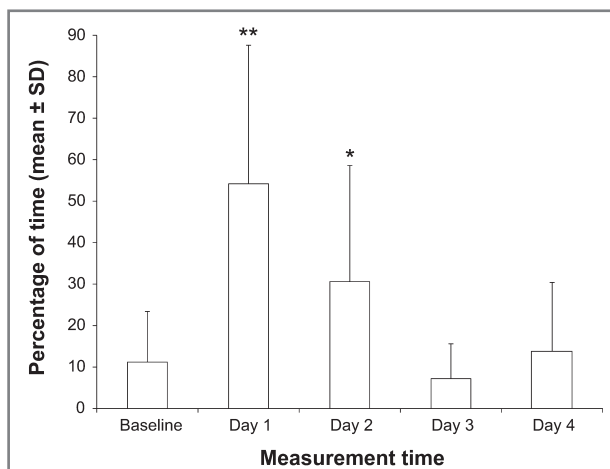


Figure 9—Percentage of 30-second sequences during which lying at the bottom of the cage was the dominant observed behavior in the birds in Figure 2. See Figure 6 for key.

significantly less time perching on postoperative day 1 ($P < 0.001$) and marginally less time on day 2 ($P = 0.01$; not significant after Bonferroni correction; Figure 8), compared with the amount of time spent perching at baseline. They also spent significantly more time lying on the floor on day 1 ($P < 0.001$) and marginally more time on day 2 ($P = 0.04$; not significant after Bonferroni correction; Figure 9), compared with the amount of time spent lying on the floor at baseline. No other significant differences were detected.

Mean duration for all 4 activities and 4 postures was calculated for each bird at each time and for each observer at baseline and during all postoperative periods. No systematic bias was evident for the activities of exploring, resting, and preening at baseline nor for preening during the postoperative period. The Bland-Altman analysis revealed a significant difference between observers for the activity eating or drinking at baseline ($3.6 \pm 15.7\%$) and during the postoperative period ($0.3 \pm 1.4\%$). A significant difference between observers was detected for the activity exploring ($7.5\% \pm 13.0\%$) only during the postoperative period. No systematic bias for interobserver agreement was evident for all 4 postures at any evaluation point.

Discussion

In the study reported here, the model of postsurgical pain associated with fracture and repair in pigeons was deemed to have good reproducibility in pigeons; variability in durations of anesthesia, surgery, and anesthetic recovery among surgeries was reasonable. Despite the lack of a control group that did not undergo the surgery, it could be reasonably presumed that the decrease in DBW on the fractured limb observed after the surgery was attributable to the surgical procedure. Even if the decrease could be explained in part by other surgical effects such as limb instability, it is highly likely the observed changes in bird posture were a consequence of the surgically induced pain. The increase in weight-bearing load following the administration of the NSAID meloxicam in another study³² supports this hypothesis. Therefore, the distribution of weight bearing between the pelvic limbs as measured by use of an incapitance meter adapted for perching birds was a sensitive and specific means of detecting limb impairment for pain evaluation during the first 4 days after surgery.

The good agreement between mean DBW values obtained with 6 or 12 measurements supports our decision to monitor DBW during the postoperative period with 6 trials at each evaluation point. This approach allowed us to decrease the time spent manipulating birds after surgery and the potential effect of manipulation on the pigeons' well-being. Intraobserver and interobserver agreement were high. The modified incapitance meter used in the study provided an objective, practical, and reproducible method to measure the distribution of weight-bearing load on each pelvic limb as an indicator of limb impairment. Similarly modified devices have been used to assess pain in various disease models in birds, including models of arthritic pain in parrots.^{19–21}

When considering the value of the modified incapitance meter for evaluation of pain in birds, several

limitations should be considered. First, the reliability and ability of the device to identify a lack of limb impairment or pain were tested only during the preoperative period, and both observers were aware that the birds had not yet undergone surgery. However, the impact of this lack of blinding to the objective DBW measurements was likely minimal. Moreover, both observers were blinded to the postoperative treatment received (saline solution vs meloxicam).

The subjective observer evaluation of pain (overall assessment) and the fractured limb position, both of which were conducted with an observer near the birds, were reliable and sensitive in postoperative pain assessment, presuming the fracture pain model had actually induced pain. Limitations of the DBW assessment apply to these evaluations, particularly because reliability of the method in the preoperative period and how specific the method is judged to be could be greatly biased with regard to a potential period effect. It remains that the postoperative interobserver agreement and the ability to identify postoperative pain for overall assessment (and fractured limb position) were excellent.

The finding that the pain scale for the pigeons' attitude in the presence of the observer was not specific was most likely attributable to the stress and visual stimulation associated with that presence. Pain-related behaviors in birds can be substantially altered by any environmental changes or stressors as well as by their emotional and motivation states.³⁴ The pain scale for pigeons' motor activity during 10 minutes of video recording was considered neither sensitive nor specific. The evaluation method was based on 10-minute periods of observation 6 times/d via video recordings, and results might have been attributable to the fact that the measurements were not obtained throughout the entire day. Additionally, some of the criteria chosen, such as sleeping on a perch (score, 3/5) or resting on the floor (scores, 4/5 and 5/5), might not have reflected the pain response because these behaviors can be observed in pain-free, undisturbed birds.

Comparing differences between the baseline and postoperative partial ethograms allowed detection of some variables that appeared sensitive for identifying postoperative pain, such as the time that a bird spent perching or lying on the floor and, mostly, the time spent exploring the environment or resting. Variables that were not identified as useful (ie, time spent eating or drinking, preening, and lying on the perch or standing on the floor) represented small proportions of the partial ethograms (ie, they occurred infrequently). Interobserver agreement was suboptimal for the variables exploring the environment and eating or drinking. Despite a clear definition of each variable, interobserver differences existed in the classification of measured periods during which the durations of eating or drinking or exploring were similar and therefore affected interobserver agreement. Actual timing of each behavior during each session could have limited the degree of this discrepancy.

Evaluation of the positioning of pigeons showed that after 2 days of recovering from surgery, pigeons were spending as much time on their perch as they did before the surgery. Activity patterns differed for a longer

period, as exploring time was still decreased and resting time was still increased 4 days after surgery, compared with patterns before surgery. Overall, the present study showed that the DBW method was reliable, sensitive, and specific for detecting limb impairment, presumably associated with orthopedic pain in birds. We therefore concluded that this method holds promise for use in future studies.

The fracture pain model used in the present study may need to be adapted before application in other avian species that differ from pigeons anatomically, physiologically, or behaviorally. Although developed to assess the efficacy of NSAIDs in pigeons, the model could also be used to assess the efficacy of other analgesics such as opioids. However, the assessment methods used would need to be able to discriminate analgesia from the sedation often associated with opioids and from lethargy.

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- a. Torbugesic, Wyeth Animal Health, St-Laurent, QC, Canada.
 - b. AErrane, Baxter, Toronto, ON, Canada.
 - c. Ultrasonic Doppler flow detector, model 811-B, Parks Medical Electronics Inc, Aloha, Ore.
 - d. VSP, Mission, Kan.
 - e. Stryker Canada, Hamilton, ON, Canada.
 - f. PDS 4-0, Ethicon, Johnson and Johnson, Sommerville, NJ.
 - g. Metacam, Boehringer Ingelheim, Burlington, ON, Canada.
 - h. Incapacitance meter for rats, IITC Inc Life Science, Woodland Hills, Calif.
 - i. T-61 Euthanasia Solution, Intervet-Schering-Plough Animal Health, Kirkland, QC, Canada.
 - j. SAS, version 9.1, SAS Institute Inc, Cary, NC.
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References

1. Paul-Murphy J, Ludders JW, Robertson SA, et al. The need for a cross-species approach to the study of pain in animals. *J Am Vet Med Assoc* 2004;224:692-697.
2. McGeown D, Danbury TC, Waterman-Pearson AE, et al. Effect of carprofen on lameness in broiler chickens. *Vet Rec* 1999;144:668-671.
3. Buchwalder T, Huber-Eicher B. Effect of the analgesic butorphanol on activity behaviour in turkeys (*Meleagris gallopavo*). *Res Vet Sci* 2005;79:239-244.
4. Kestin SC, Knowles TG, Tinch AE, et al. Prevalence of leg weakness in broiler chickens and its relationship with genotype. *Vet Rec* 1992;131:190-194.
5. Danbury TC, Weeks CA, Chambers JP. Self-selection of the analgesic drug carprofen by lame broiler chickens. *Vet Rec* 2000;146:307-311.
6. Quinn MM, Keuler NS, Lu Y, et al. Evaluation of agreement between numerical rating scales, visual analogue scoring scales, and force plate gait analysis in dogs. *Vet Surg* 2007;36:360-367.
7. Corr SA, McCorquodale C, McDonald J, et al. A force plate study of avian gait. *J Biomech* 2007;40:2037-2043.
8. Woolley SC, Gentle MJ. Physiological and behavioural responses in the hen (*Gallus domesticus*). *Comp Biochem Physiol* 1987;88A:27-31.
9. Machin KL, Livingston A. Assessment of the analgesic effects of ketoprofen in ducks anesthetized with isoflurane. *Am J Vet Res* 2002;63:821-826.
10. Gentle MJ, Hunter LN. Physiological and behavioural responses associated with feather removal in *Gallus gallus* var *domesticus*. *Res Vet Sci* 1990;50:95-101.
11. Paul-Murphy J, Brunson DB, Miletic V. A technique for evaluating analgesia in conscious perching birds. *Am J Vet Res* 1999;60:1213-1217.
12. Hoppes S, Flammer K, Hoersch K, et al. Disposition and analgesic effects of fentanyl in white cockatoos (*Cacatua alba*). *J Avian Med Surg* 2003;17:124-130.
13. Sladky KK, Krugner-Highby L, Meek-Walker E, et al. Serum

- concentrations and analgesic effects of liposome-encapsulated and standard butorphanol tartrate in parrots. *Am J Vet Res* 2006;67:775–781.
14. Rager DR, Gallup GG. Apparent analgesic effects of morphine in chickens may be confounded by motor deficits. *Physiol Behav* 1986;37:269–272.
 15. Curro TG, Brunson DB, Paul-Murphy J. Determination of the ED50 of isoflurane and evaluation of the isoflurane-sparing effect of butorphanol in Cockatoos (*Cacatua* spp.). *Vet Surg* 1994;23:429–433.
 16. Concannon KT, Dodam JR, Hellyer PW. Influence of a mu- and kappa-opioid agonist on isoflurane minimal anesthetic concentration in chickens. *Am J Vet Res* 1995;56:806–811.
 17. Hocking PM, Robertson GW, Gentle MJ. Effects of non-steroidal anti-inflammatory drugs on pain-related behaviour in a model of articular pain in the domestic fowl. *Res Vet Sci* 2005;78:69–75.
 18. Brune K, Bucher K, Walz D. The avian microcrystal arthritis II. Central versus peripheral effects of sodium salicylate, acetaminophen and colchicine. *Agents Actions* 1974;4:27–33.
 19. Paul-Murphy JR, Sladky KK, Krugner-Higby LA, et al. Analgesic effects of carprofen and liposome-encapsulated butorphanol tartrate in Hispaniolan parrots (*Amazona ventralis*) with experimentally induced arthritis. *Am J Vet Res* 2009;70:1201–1210.
 20. Paul-Murphy JR, Krugner-Higby LA, Tourdot RL, et al. Evaluation of liposome-encapsulated butorphanol tartrate for alleviation of experimentally induced arthritic pain in green-cheeked conures (*Pyrrhura molinae*). *Am J Vet Res* 2009;70:1211–1219.
 21. Cole GA, Paul-Murphy J, Krugner-Higby L, et al. Analgesic effects of intramuscular administration of meloxicam in Hispaniolan parrots (*Amazona ventralis*) with experimentally induced arthritis. *Am J Vet Res* 2009;70:1471–1476.
 22. Breward J, Gentle MJ. Neuroma formation and abnormal afferent nerve discharges after partial beak amputation (beak trimming) in poultry. *Experientia* 1985;41:1132–1134.
 23. Gentle MJ, Hunter LN, Waddington D. The onset of pain related behaviours following partial beak amputation in the chicken. *Neurosci Lett* 1991;128:113–116.
 24. Crandall M, Miaskowski C, Kools S, et al. The pain experience of adolescents after acute blunt traumatic injury. *Pain Manag Nurs* 2002;3:104–114.
 25. Greenfield GQ. Orthopaedic pain. In: Boswell MV, Cole BE, eds. *Weiner's pain management: a practical guide for clinicians*. 7th ed. Boca Raton, Fla: CRC, 2006;465–476.
 26. Freeman KT, Koewler NJ, Jimenez-Andrade JM, et al. A fracture pain model in the rat. *Anesthesiology* 2008;108:473–483.
 27. Haegerstam GA. Pathophysiology of bone pain: a review. *Acta Orthop Scand* 2001;72:308–317.
 28. Minville V, Laffosse JM, Fourcade O, et al. Mouse model of fracture pain. *Anesthesiology* 2008;108:467–472.
 29. Oni OA, Hui A, Gregg PJ. The healing of closed tibial shaft fractures. The natural history of union with closed treatment. *J Bone Joint Surg Br* 1988;70:787–790.
 30. Pollock CG, Orosz SE. Avian reproductive anatomy, physiology and endocrinology. *Vet Clin North Am Exot Anim Pract* 2002;5:441–474.
 31. International Species Inventory System. *Physiological data reference values* [CD ROM]. Eagan, Minn: International Species Inventory System, 2002.
 32. Desmarchelier M, Troncy E, Fitzgerald G, et al. Analgesic effects of meloxicam administration on postoperative orthopedic pain in pigeons (*Columba livia*). *Am J Vet Res* 2012;73:361–367.
 33. AVMA policy. Use of placebo controls in assessment of new therapies for alleviation of acute pain in client-owned animals. Available at: www.avma.org/issues/policy/placebo_controls.asp. Accessed Aug 12, 2010.
 34. Gentle MJ, Corr SA. Endogenous analgesia in the chicken. *Neurosci Lett* 1995;201:211–214.

Appendix

Numeric rating pain scales evaluated to assess severity of pain after surgical induction of lameness in pigeons.

Score	Behavioral observation
Pigeon's attitude in the presence of the observer	
0	Alert and attentive; tries to escape and fly with insistence.
1	A little curious; still tries to escape but with only mild effort.
2	Stays quiet on its perch with little reaction to the presence of the observer.
3	Stands on the floor and displays little reaction to the presence of the observer.
4	Stands on the floor and displays no reaction to the presence of the observer.
5	Lies on the floor and displays no reaction to the presence of the observer.
Fractured limb position in the presence of the observer	
0	Appears to bear equal weight on both limbs.
1	Bears weight on both limbs but appears to bear a little less weight on the fractured limb.
2	Bears weight on both limbs but obviously bears much less weight on the fractured limb.
3	Able to bear weight on both limbs but appears reluctant to do so on the fractured limb.
4	Does not bear weight on its fractured limb but stands on its nonfractured limb.
5	Lies on the floor.
Subjective observer evaluation of pigeon's degree of pain (overall assessment)	
0	No signs of pain; pigeon appears as it did before surgery.
1	Appears uncomfortable on 1 limb but discomfort not always obvious.
2	Evidence of discomfort on 1 limb but no other obvious sign of pain.
3	Overall, appears moderately disturbed by pain in its fractured limb.
4	Overall, appears highly disturbed by pain in its fractured limb.
5	Lies on the floor; does not appear able to stand.
Pigeon's motor activity during 10 minutes of video recording	
0	Highly active; perches, moves around, explores, preens, or eats.
1	Moderately active; moves a little, but mainly stays quiet on its perch; preens.
2	Awake but quiet; stays quiet on its perch, does not preen, and looks around.
3	Very quiet; sleeps on its perch or stands on the floor; does not preen.
4	Obvious decreased reaction; stands or lies on the floor and appears lethargic or asleep.
5	No reaction; lies on the floor; and does not react to any stimuli.