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Use of Anaesthetized Animals to Test Humaneness of Killing Traps

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Abstract: Using anaesthetized animals to test potential humaneness of killing traps was initiated in the 1970s based on the knowledge that anesthesia minimizes animal stress. The study focus was to determine if time-to-loss-of-sensibility resulting from tests on anaesthetized animals were predictive of time-to-loss-of-sensibility of results from tests on unanaesthetized animals. Time-to-loss-of-sensibility of anaesthetized animal tests (median = 75.5 seconds, n = 198) were less than unanaesthetized animal tests (median = 102 seconds, n = 333). However, the relationship between anaesthetized animal tests and unanaesthetized animal tests is not predictive ($r^2 = 0.07$, $P = 0.244$) based on species and trap types tested by the Fur Institute of Canada's Trap Effectiveness Project between 1985 and 1997. Therefore, we do not recommend use of anaesthetized animals to test humaneness of killing traps.

Key words: anaesthetized animals, furbearers, humaneness, killing traps, survival analysis, time-to-loss-of-sensibility, trapping standards

Wildlife Society Bulletin 00(0): 000-000

New Canadian (Canadian General Standards Board 1996) and international (Agreement on International Humane Trapping Standards 1997) humane trapping standards require time-to-loss-of-sensibility (TTLS) information from killing trap tests on animals. For killing traps to pass these standards, a percentage of animals must be rendered irreversibly insensible (unconscious) within species-specific time limits. Use of anaesthetized animals (AA) to test the potential humaneness of killing traps was initiated in the 1970s by the Federal Provincial Committee for Humane Trapping (1981). Anaesthetized animals were used as a safeguard to minimize animal stress under the premise that AA were not conscious of pain, were relaxed, and less physically able to resist the impact and clamping forces of the trap. The assumption was made that a trap not able to kill AA within predetermined time limits should not be tested on unanaesthetized animals (UA). Similarly, AA tests were used in New Zealand to determine the mechanical power threshold for effective kill traps for possum (*Trichosurus vulpecula,* Warburton and Hall 1995).

The Trap Effectiveness Project (formerly the Humane Trapping Program), initiated in 1985 by the Fur Institute of Canada, conducted research to develop humane traps using AA and UA tests. The protocol used in testing the traps involved approach, pre-selection (AA), and kill tests (UA, Proulx et al. 1989*a*). If a predictive relationship could be established between the 2 types of tests, then UA tests would not be required and animal stress, number of animals used, and costs associated with trap testing would be reduced. Consequently, the objective of this study was to determine if the TTLS for AA tests are predictive of the TTLS for UA tests.

Methods

Trap testing

Data used in this study were collected by the Fur Institute of Canada's Trap Effectiveness Project at the Alberta Research Council (formerly the Alberta Environmental Centre) in Vegreville, Alberta, Canada, between 1985 and 1997. We conducted all animal care and use under approved research protocols according to the Canadian Council of Animal Care guidelines (Olfert et al. 1993). We used information on 198 AA tests and 333 UA tests on 10 furbearing species [arctic fox (*Alopex lagopus*), badger (*Taxidea taxus*), Canada lynx (*Lynx canadensis*), ermine (*Mustela erminea*), fisher (*Martes pennanti*), marten (*Martes americana*), mink (*Mustela vison*), muskrat (*Ondatra zibethicus*), raccoon (*Procyon lotor*), and red squirrel (*Tamiasciurus hudsonicus*)] with 35 different designs of traps. The 3 basic types of killing traps tested included rotating-jaw, mousetrap, and planar (Figure 1).

For AA tests, staff anaesthetized animals with an intramuscular injection of 10–20 mg/kg of ketamine hydrochloride (Kreeger 1996). After the initial injection, staff injected additional drug or allowed time to elapse until animals were supine with strong palpebral and corneal reflexes in both eyes. Palpebral reflexes are blinking reflexes in responses to touching the eyelids and corneal reflexes are blinking responses to touching the cornea (Kreeger 1996). Staff then positioned animals in the traps to receive the same strike locations dictated by the approach tests. After traps were fired, staff continuously monitored the palpebral and corneal reflexes of test animals. Animals were considered irreversibly insensible once all palpebral and corneal reflexes were lost.

If animals maintained eye reflexes at 300 seconds, staff euthanized them with an intra-cardiac injection of 200 mg/kg of sodium pentobarbital. Once the eye reflexes were irreversibly lost, staff monitored the heartbeat to cessation with a stethoscope. Because the exact TTLS was unknown for euthanized animals, TTLS of these animals (for which only a lower bound of the TTLS is known) were right censored (Kalbfleisch and Prentice 1980). Thus, AA tests resulted in either uncensored values (exact TTLS) or right censored values.

For the UA tests, staff set traps in test pens simulating natural conditions (Nolan and Barrett 1990). Staff then viewed the interactions between the animals and the traps by remote infrared video cameras. When the animal fired the trap, staff ran immediately to the test pen and began monitoring the eye reflexes. Time from trap firing to initiation of eye reflex monitoring varied between 25 and 75 seconds depending on the distance from the core building to the test pen used. Procedures continued as in AA tests. If animals had already lost sensibility (eye reflexes) upon arrival of staff, these TTLS (for which only an upper bound is known) were left censored. Thus, UA tests resulted in uncensored values, left censored values, or right censored values.

Statistical analysis

We compared the distributions of the TTLS for AA and UA tests with survival functions estimated using Turnbull’s nonparametric estimation method based on doubly censored data (Turnbull 1974). A SAS® (SAS Institute 1997) macro, written by David Camp and Michael Province from the Division of Biostatistics, Washington University Medical School, was used to calculate open-ended survival curves. We used the curve shape to determine similarity between survival functions for AA and UA tests and to determine appropriate modeling procedures.

Because each animal was used in only one of the 2 types of tests, the AA and UA test data were unpaired. To arrive at a pairing to model the relationship between these tests, we grouped animals based on species, trap type, and strike location(s). These characteristics maximized number of groups, while keeping animals within each group relatively homogeneous. We used only means of matched data between AA and UA groups in these analyses. Because the data set contained left and right censored values and followed a one-parameter exponential distribution, we used the methods of Sarhan and Greenberg (1962) to determine $\hat{m}$, the best linear unbiased estimate of the mean, as follows:

$$\hat{m} = \frac{1}{K}\left\{\left[\sum_{i=1}^{r_1+1}\frac{1}{n-i+1}\Big/\sum_{i=1}^{r_1+1}\frac{1}{(n-i+1)^2} - (n-r_1)\right]x_{(r_1+1)} + r_2 x_{(n-r_2)} + \sum_{i=r_1+1}^{n-r_2} x_{(i)}\right\}$$

where

$$K = \left[\left(\sum_{i=1}^{r_1+1}\frac{1}{n-i+1}\right)^2 \Big/ \sum_{i=1}^{r_1+1}\frac{1}{(n-i+1)^2} + (n-r_1-r_2-1)\right]$$

and

n is the total number of observations in the sample,

r_1 is the number of left censored observations,

r_2 is the number of right censored observations,

$x_{(i)}$ is the TTLS for the i^{th} ordered observation.

We then determined the relationship between TTLS for AA and UA tests using linear (PROC REG) and nonlinear (PROC NONLIN) model fitting procedures (SAS Institute 1989). Because data were approximately normally distributed, we used the Pearson correlation coefficient to measure association.

Results

Overall, 26 of 198 AA tests had right censored values and 63 and 144 of 333 UA tests had right and left censored values, respectively. Distributions of the TTLS for uncensored observations were skewed to the right (skewness = 0.86) for AA tests and moderately skewed to the right (skewness = 0.59) for UA tests (Figure 2). Median TTLS including censored observations for the AA tests (75.5 seconds) was shorter than that of the UA tests (102.0 seconds). Estimated survival functions (Figure 3) showed the probability of a particular animal surviving beyond a given time “t”. For time interval 0 to 300 seconds, UA tests’ survival probability is greater than that of AA tests indicating that an anaesthetized animal has a greater probability of having a shorter TTLS than an unanaesthetized animal. The premise that AA were relaxed and thus less physically able to resist the impact and clamping forces of the trap appears plausible. The negative log of the survival function versus time indicated that an exponential model would be appropriate for AA and UA tests (Kalbfleisch and Prentice 1980).

For 90% (19 of 21) of the cases, the mean TTLS for the AA tests was shorter than that for the UA tests (Table 1). The lack of correlation between the 2 types of tests ($r = 0.27$, $P = 0.24$) indicated no significant linear association. The relationship between the TTLS of AA and UA tests also lacked predictive strength (Figure 4). Nonlinear curves fit to the relationship proved no better fit than the linear curve. The linear regression equation relating TTLS of the UA and AA tests was:

$$TTLS_{UA} = 116.7595 + 0.5324 TTLS_{AA}$$

The overall model was not significant ($F = 1.446$, $P = 0.244$) and only 7% of the variation in the TTLS for UA tests is accounted for by the AA tests ($r^2 = 0.07$). The model assumption of independent normally distributed errors was satisfied; however, predicted values were not accurate. For example, estimated means of 3 of the 4 groupings for marten for AA tests were $\leq$120 seconds (Table 1), indicating a potential to pass the Agreement on International Humane Trapping Standards. However, none of the estimated means for the corresponding UA tests indicated a potential to meet this standard.

Discussion

Although sample sizes were small, estimated means of AA and UA tests seemed more similar for Canada lynx and Arctic fox than for other species (Table 1). Additionally, the mechanical power threshold for effective kill traps for possum developed from AA tests by Warburton and Hall (1995) were reasonably predictive of results obtained from UA tests. These contrasts in findings indicate the existence of different relationships between AA and UA tests across animal species. Some inconsistency may be because of anesthesia variability obtained on individuals within a species or varying physiology between species. We suggest that AA tests could be used to identify less humane traps. However, unless a sound predictive relationship between AA and UA tests for the species tested is established, we believe that AA testing cannot ascertain if a killing trap meets humane trapping standards.

With the advent of new trapping standards (Canadian General Standards Board 1996, Agreement on International Humane Trapping Standards 1997) and trap testing standards (International Organization for Standardization 1999), killing trap testing on animals will be required. Testing agencies and often regulatory agencies would prefer using anaesthetized animals for this testing because of animal welfare reasons. Our data indicates little benefit in this approach, therefore we do not recommend using anaesthetized animals to rate killing traps.

The predictive relationship between increased mechanical power of traps and lowered TTLS is well established (Federal Provincial Committee for Humane Trapping 1981, Proulx et al. 1989*b*, Hiltz and Roy 1998). Hiltz and Roy (1998) developed accurate computer models to predict whether traps met standards using as few as 37 unanaesthetized test animals and mechanical properties of 4 trap designs. These models can now be used to rate most new trap designs without the need for animal-based testing. This approach has led to cost savings and reduced the need to use animal test subjects. In conclusion, we advise limited use of testing on anaesthetized animals. Such testing is only justifiable with species where no trap testing data is available on the same or similar species. Moreover, we recommend judicious use of testing on unanaesthetized animals aimed towards model development that will lead towards the elimination of animal use for this purpose.

Acknowledgments. This research was supported by the Fur Institute of Canada, the National Research Council's Industrial Research Assistance Program, and the Alberta Research Council. We would also like to acknowledge the assistance of the Trap Effectiveness Research Team (Alfred Kolenosky, Sandra Melenka, Colin Twitchell, and Luke Nolan). Finally, we would like to thank Dr. L.Z. Florence, Dr. J. Schieck, Dr. S. Song, and Neal Jotham for reviewing earlier drafts of this manuscript and Marion Herbut for technical assistance.

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Associate editor: Ford

Table 1. Best linear unbiased mean time-to-loss-of-sensibility (seconds) for species and strike location groupings of traps tested using anaesthetized animals (AA) and unanaesthetized animals (UA) completed at the Alberta Research Council facility in Vegreville, Alberta, between 1985 and 1997. Kania are planar type traps, Victor 1 1/2 Long Spring is a mousetrap type, and all other trap designs tested are rotating-jaw.

Species	**Trap Design**	**Strike Location**	**AA**		**UA**	
			N	**Mean**	**N**	**Mean**
arctic fox	S 2001-8 [a]	atlanto-occipital [b]	2	37.00	4	37.69
arctic fox	S 2001-8	neck	3	55.67	3	85.37
Canada lynx	C330 [c]	neck	5	135.60	6	142.42
Canada lynx	Modified C330	neck	5	44.00	7	39.31
ermine	Victor 1 1/2 LS [d]	head	5	12.75	7	66.55
ermine	Victor 1 1/2 LS	neck	1	5.00	4	71.52
ermine	Victor 1 1/2 LS	neck and thorax	3	28.00	2	268.00
fisher	S 2001-8	atlanto-occipital	3	81.34	5	142.21
fisher	S 2001-8	head	3	147.33	4	269.00
marten	C120	atlanto-occipital	2	162.00	4	128.29
marten	C120	atlanto-occipital and thorax	2	47.50	3	197.00
marten	C120	neck and thorax	1	46.00	6	194.80
marten	Kania	atlanto-occipital	2	24.00	2	122.00
mink	C120 Magnum	atlanto-occipital and thorax	1	43.00	3	94.00
mink	C120 Magnum	neck	1	47.00	5	197.25
muskrat	C120	head and abdomen	3	36.34	2	320.00
muskrat	C120	neck	3	18.00	3	64.36
muskrat	C120	neck and abdomen	2	44.50	1	289.00
muskrat	C120	thorax	1	24.00	9	90.80
muskrat	C120	thorax and abdomen	3	44.67	14	182.70

red squirrel	Kania	head	2	11.50	3	32.73

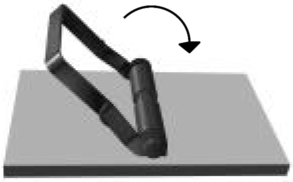
[a] S = Sauvageau

[b] Atlanto-occipital relates to the atlas and occipital bones

[c] C = Conibear

[d] LS = Long spring


Rotating-jaw


Mousetrap

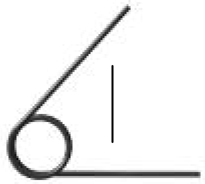
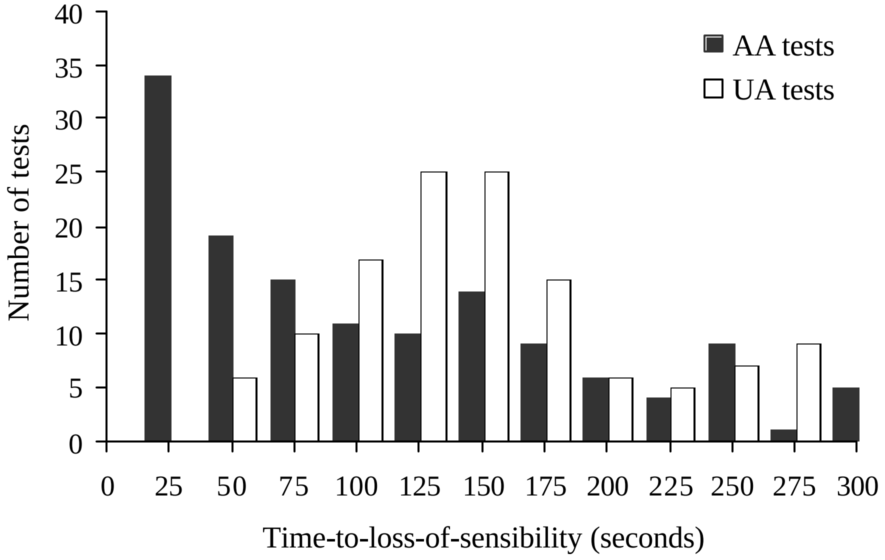

Planar

Figure 1. Rotating-jaw, mousetrap, and planar trap types were tested on anaesthetized and unanaesthetized animals at the Alberta Research Council facility in Vegreville, Alberta, between 1985 and 1997.



40
35
30
25
20
15
10
5
0
Number of tests
AA tests
UA tests
0
25
50
75
100
125
150
175
200
225
250
275
300
Time-to-loss-of-sensibility (seconds)

Figure 2. Distribution of uncensored time-to-loss-of-sensibility values obtained from trap tests on anaesthetized animal (AA) tests and unanaesthetized animal (UA) completed at the Alberta Research Council facility in Vegreville, Alberta, between 1985 and 1997.

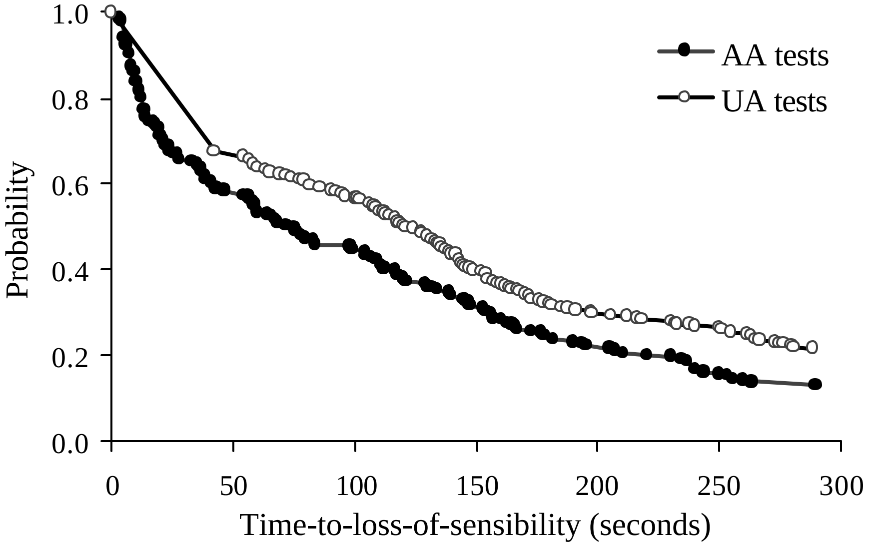

1.0
0.8
0.6
0.4
0.2
0.0
Probability
AA tests
UA tests
0
50
100
150
200
250
300
Time-to-loss-of-sensibility (seconds)

Figure 3. Estimated survival functions (probability of an animal surviving beyond time “t”) obtained from trap tests on anaesthetized animal (AA) and unanaesthetized animal (UA) completed at the Alberta Research Council facility in Vegreville, Alberta, between 1985 and 1997.

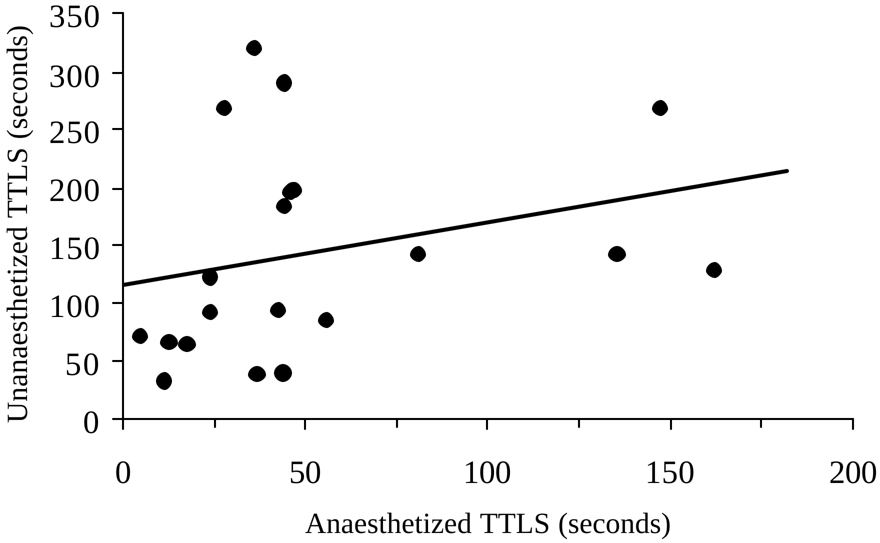

350
300
250
200
150
100
50
0
0
50
100
150
200
Unanaesthetized TTLS (seconds)
Anaesthetized TTLS (seconds)

Figure 4. Time-to-loss-of-sensibility (TTLS) relationship obtained from trap tests on unanaesthetized and anaesthetized animals at the Alberta Research Council facility in Vegreville, Alberta, between 1985 and 1997.