

Dairy Cow Sensitivity and Aversion to Short Duration Transient Currents

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Summary:

This paper summarizes research relating to the sensitivity of dairy cows to a variety of transient current waveforms. The phase duration of stimuli ranged from 10 micro seconds to 8.3 milliseconds (effective frequency from 50,000 to 60 Hz). Characterization of the effects of waveform (monophasic sinusoidal, monophasic exponential decay, biphasic and multiple cycle) transients are presented. Exposure pathways included: muzzle to four hooves, both with a direct application to the nose and via water bowl exposure, teat to hooves via milking machines and via direct skin contact. The effects of hoof condition and hoof trimming were also studied. The sensitivities of over 120 cows were determined for a single cycle 60 Hz transient current. The relationship between sensitivity to transient current and aversion to water were examined for a 6000 Hz transient current applied to a water bowl.

Keywords: Stray Voltage, Stray Current, Transient Voltage, Dairy Cows, Electrical Stimulation

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1. INTRODUCTION

Research conducted before 1989 on the effects of stray voltage on dairy cows and investigative methods are summarized in a U.S. Department of Agriculture stray voltage handbook (USDA, 1991). At the time that this handbook was published, research into the perception and avoidance by dairy cows of electrical phenomena had been confined largely to continuously applied, 60 cycle AC voltages. Recommendations for further research cited in this handbook include studies of:

- o How animals perceive and interpret current flow.
- o The variability in sensitivity and response to current for a large number of dairy animals.
- o Evaluation of short and long term exposure to transients on animals.
- o The effects of direct current on animals.

The response of dairy cattle electrical phenomena other than continuously applied 60 Hz stimuli has not been thoroughly investigated. Brennan and Gustafson, 1986 examined the sensitivity of dairy cows to short AC currents. Currence et al., 1987 examined the sensitivity of dairy cows and humans to 60 Hz AC currents with stimuli of 1, 10 and 100 cycles. Cows responded at a lower current for the 10 and 100 cycle stimuli than for single cycle stimuli. Studies performed by Gustafson et al, (1984, 1988), examined perception thresholds of monophasic rectangular current (DC) stimulation ranging in duration from 0.52 to 265 ms. These studies indicate that as the duration of exposure is decreased the voltage required for animal perception is increased. Another conclusion was that further study over expanded time and current ranges and with larger cow numbers was needed.

More recent work (Reinemann et al, 1994) has extended the range of measurements to include shorter duration transient currents. Dairy cow sensitivity to transient currents was found to follow the form of previously developed neurological models and experimental data from human subjects (Reilly, 1992, 1994). Sensitivity was found to be highly influenced by the phase duration of the stimulus (time from zero crossing to zero crossing). The range of wave forms used in previous studies was expanded further in the studies.

Behavioral indicators have previously been used to evaluate sensitivity of dairy cattle to electrical phenomena. Behaviors altered by exposure include shock induced suppression of a learned response (e.g., pressing a plate with the muzzle to receive grain, Norell et al, 1983), hoof raising or mouth opening to escape electrical shock (Gustafson et al, 1988) and activity during milking (Aneshansley, 1992, Lefcourt et al, 1982, 1985). Several authors report occurrence of other behavioral signs of annoyance such as hoof movement, muscle contraction, startled jerks, and shoulder shakes (Norell et al, 1983; Gustafson, et al, 1985). However, these appeared to be idiosyncratic to certain animals as well as to the route of current delivery. Work performed by Henke et al, (1982), indicate that behavioral response is as sensitive or more sensitive an indicator of perception than analysis of stress related hormones in blood. The behavioral observation techniques used by Reinemann et al, (1994) were shown to be repeatable and sensitive indicators of animal response and correlated well with electronic recording of changes

in animal activity in response to electrical stimuli .

Studies relating the sensitivity measures to aversive behavior (delay to drink and total water and feed consumption) have been performed for a single cycle 60 Hz transient current (Reinemann et al, 1995b). Further studies were performed to determine if the relationship between behavioral reaction and aversion reported previously for 60 Hz stimuli also existed for shorter phase duration (higher frequency) transient currents.

The research reported in this paper was initiated at the request of and funded by the Wisconsin Department of Agriculture, Trade and Consumer Protection (WDATCP) to provide data to address concerns of the agricultural community. The broad objectives of this study are to:

- 1) Characterize dairy cow sensitivity to a wide range of transient voltage and current
- 2) Determine the relationship between short term sensitivity response and water and feed intake, milk production and animal health responses to long term exposure to cow contact voltages.
- 3) Investigate potential behavioral response to magnetic fields.
- 4) Contribute to the stray voltage knowledge base by continuing research recommended in the USDA Handbook 696
- 5) Relate responses measured in these studies to steady state 60 Hz exposure previously reported.

Results from objectives 1, 2, 3, and 4 are presented in previous publications (Reinemann et al 1993, 1994, 1995a, 1995b). This paper presents results which address objectives 1, 2 and 4. Studies are currently underway to address objectives 4 and 5.

2. Animal Sensitivity

2.1 Animal Sensitivity Materials and Methods

2.1.1. Muzzle to hoof Exposure

The test apparatus and methods to measure response to transient voltages applied through the muzzle to hoof pathway has been described in Reinemann et al, 1994. The strength-duration relationship for dairy cow response to transient current pulses was found to follow the form of previously developed neurological models (Reilly 1992, 1994). However the time scale of the experimental curve was shifted to a longer strength duration time constant as compared with the theoretical model. Such a temporal shift is also observed in human electro-cutaneous sensitivity data. Previous results suggest that the horizontal time scale in the experimental plots may need to be scaled by a factor of about 2 for comparison with the theoretical model. Threshold levels for short duration pulses are mainly influenced by the charge of the pulse rather than the peak current. In the previous experiments the waveforms for short duration (10

microsecond phase duration) were not sinusoidal as the resolution of the waveform generator was 100 kHz. The 10 microsecond waveform developed in previous trials closely approximated a monophasic (unipolar) square wave.

The experimental apparatus was modified to provide better control of the waveform and symmetry of short duration pulses. A frequency generator was used to develop a constant frequency signal with sinusoidal, or square waveform. This signal was then fed through a zero-crossing switching device which would block a pre-set number of pulses and then allow another pre-set number of cycles to pass. This signal was fed into the power amplifier used previously. These transient currents were applied to the animal once per second over the 30 second observation period. The test apparatus was designed with a high source resistance to minimize the effects of varying hoof/floor contact resistance and control the current delivered to the cow. A direct measurement of the current flowing through the cow was made during all tests.

The tests reported previously (Reinemann et al, 1994) were all done on a concrete surface which had been cleared of all bedding and thoroughly wetted with a salt solution. The surface was chosen to represent the lowest possible contact resistance encountered in the field. Tests were also performed on a variety of surfaces including: clean and dry concrete, an expanded metal grate, and normal stall conditions to investigate the effects of varying hoof/floor contact resistance on the voltage and current relationships for different floor surfaces.

2.1.2. Measuring Animal Response

Methods developed in previous research were used to measure animal response (Reinemann et al 1994). Cows were fitted with a non-piercing ball end nose clip and allowed to become accustomed to the presence of this clip. Transient Currents were applied through the muzzle to hooves pathway with the cows standing in the specially constructed test stalls. Tests were repeated on each cow with consecutively increasing stimulus level until a clear positive response was established. This completed the testing on that cow on that day. Each cow was tested in this manner for two consecutive days and a third day if the results of the first two test differed by more than 10%. Observers were trained by pairing a new observer with an experienced observer. The observers independently observed and recorded responses. Paired observations were repeated until correlation of observed responses was in excess of 90% for several consecutive tests. The primary indicators used to measure response for these tests were facial twitches and hoof lifting. Previous results indicated that hoof lifting responses measured by trained human observers agreed well with measures made with electronic recording equipment and that hoof lifting and facial twitches were the most sensitive and repeatable measures of response.

2.1.3. Milking Machine Exposure

Several types of milk flow sensors use low-level, high-frequency electrical pulses to sense the presence of milk flow. Tests were performed on such a commercial milk flow sensor (BouMatic¹) to investigate concerns over the possible effects of these electrical pulses on dairy cows, and measure milk hose impedance with high frequency waveforms. The waveform of the electrical impulse used in this sensor is an exponential decay. The sensor was tested when installed according to the manufacturers recommended practice and under additional scenarios representative of improper installation. The sensor was also tested using an amplified sensing signal, far greater than that which could possibly occur in practice. This test was done to accurately measure milk hose impedance and to document the conditions under which cows would react to the signal. Gustafson et al, (1983) previously documented the impedance of a milk hose and milking unit under operating conditions as 20,000 to 30,000 ohms for 60 Hz alternating current. Tests were done with a monophasic sine wave to relate sensitivity of exponential waveforms to a monophasic pulse.

A schematic of the test apparatus and circuit is shown in Figure 1. Voltage measurements were made using a Yokogawa, oscillographic recorder with a sampling rate of 100 kHz. The accuracy of this instrument was checked against calibrated voltmeters before testing.

Voltages were measured in five locations:

- 1) from the signal ring of the sensor to ground,
- 2) from the grounded sensor ring to ground,
- 3) from the milking claw to ground,
- 4) from the teat skin to ground, and
- 5) across a 10-ohm resistor in series with the cow/sensor circuit

During tests the cows were standing on a metal grate placed on a specially constructed test stall suspended off the floor (Figure 1). This test apparatus made it possible to isolate the cow and sensor from all other electrical influences, such as voltages imposed on grounded objects from sources other than the milk flow sensor. This apparatus also allowed for accurate measurement of the current flow through the cow.

Tests were done under operating conditions while milking three cows. Measurements were made during the peak milk flow period. Measurements were made of the total milking time and pounds of milk harvested. Average milk flows over the course of milking ranged from 3.4 to 6.0 pounds per minute. A 3-foot length of 5/8" milk hose connected the flow sensor to a stainless steel Abarrel@ type claw. A 3-foot length of milk hose connected the sensor to a bucket milking system with pneumatic pulsation. The bucket milker was placed on an electrically isolated scale. The following scenarios were tested:

Scenario 1: The sensor was tested when operating with the grounded ring toward the cow with grounding both connected and lifted. When the sensor is installed as recommended

¹ Mention of Trade Names is for identification purposes only and does not imply endorsement by the University of Wisconsin, WDATCP or the US Department of Agriculture.

(with one ring connected to the DC common grounded), the current generated by the sensor has two paths to ground. One path is through the cow and the other is through the connection to ground. The impedance of the connection to ground is much lower than the path through the cow and will therefore carry most of the current generated. When this ground is lifted, the only path for current flow is through the cow. This situation creates the worst case scenario expected in the field, as the only path for the current is through the cow to ground.

Scenario 2: The sensor was tested with the signal ring toward the cow and the other ring both connected and disconnected to the DC common ground. When operating in this configuration, there may, theoretically, be a very brief period of time in which there is an electrical connection between the signal ring and the cow through the milk hose but the connection across the sensor rings to ground is not completed. This would presumably remove the connection to ground and result in only one flow path for current through the cow. It is likely, however, that a thin film of milk is always present between the two sensing rings and that this connection is never truly Aopen.@ This circuit configuration was examined to test the effect of the orientation of the sensor rings on current flowing through the cow.

Scenario 3: The sensor was tested with an amplified signal voltage, the signal ring toward cow and the other ring not grounded. In normal operation, the maximum peak voltage produced at the sensor ring is less than 5 volts when there is no milk flow. Based on previous studies, (Gustafson et al, 1983), it was not expected that this voltage would be sufficient to produce a measurable current through the milk hose / milking unit / cow pathway nor to illicit a response from a cow. The sensing signal was, therefore, passed through an isolated amplifier to increase its voltage. Note that this situation is far worse than could ever occur in the field.

Scenario 4: The effective pulse width of the waveform used by this milk flow sensor was estimated from the results of scenario 3 to be between 20 and 30 micro seconds. The transient current generating device described in section 2.1.1. was used to develop a 25 micro second sine wave which was applied from the teat to hooves pathway. These tests were performed to characterize the sensitivity relationship between sinusoidal and exponential wave forms. Measurements were also made of the potential developed across the cows skin in the presence of a know current flow. Areas on the cows rump, ankle and teat of approximately 2 inches square were shaved and EKG patches applied. The stimulus was applied through the EKG patch on the teat. The other terminus for current flow was an expanded metal grate on the floor. A direct measurement of current flow was made with one channel of an oscilloscope reading voltage across a 10 ohm resistor in series with the test circuit. Voltages from teat to floor and from rump to ankle were also recorded.

2.1.4. Hoof Trimming and Hoof Condition

The objective of this study was to determine if hoof trimming increased sensitivity to transient current. Secondary goals were to determine if cow resistance changed after trimming and the effect of abscesses on reaction level. Because current driven by stray voltage often travels through the hooves, the resistance and condition of the contact points

could be an important factor in a cow's sensitivity to currents.

Dry cows, in need of hoof trimming were selected by the UW herdsman for this study. The cows were tested by the muzzle to hoof pathway described in section 2.1.1 while standing on an expanded metal grate. Cows were exposed to an ascending series of a 1 cycle, 60 Hz transient applied at 30 second intervals as described above. The condition of the hooves were recorded before trimming. The cows were tested for reaction level (Pre) and then their hooves were trimmed. The cows were then placed in a stall and allowed to relax for approximately one hour before being tested again for reaction level after trimming (Post1). A third reaction level test was conducted seven days later (Post2). Two cows were not available seven days after trimming.

A sample size of 12 cows was estimated based on the main objective. Paired T-tests were used for the before and after comparisons for reaction level and cow resistance. Independent two tailed T-test were used for the comparisons for the cows with abscess and cows with no abscess. The power calculations were based on noncentral t distributions.

2.2. Animal Sensitivity Results and Discussion

2.2.1. Effects of phase duration and wave form

The efficacy of an electrical stimulus depends critically on its waveform (Reilly, 1992, 1994). Examples of waveform sensitivity are shown in figure 2 for three types of current waveforms: a monophasic (unidirectional current) rectangular pulse, a biphasic (current reversal) rectangular pulse, and a single cycle of a sine wave. This example was derived from a myelinated nerve model (Reilly 1992), in which a 20 μ m nerve fiber is excited by current from a small electrode within the biological medium. The horizontal axis is the phase duration of the current stimulus, i.e., the duration of the monophasic pulse, or the duration of one phase of the biphasic pulses. The right-hand vertical axis is the peak current magnitude (threshold current) needed to excite the nerve; the left-hand axis is the phase charge, i.e., the amount of charge in the monophasic current, or in one phase of the biphasic currents. The monophasic current curve in Figure 2 is often called a strength-duration curve, which may be fitted with the mathematical expression (1).

$$I_t = I_o \left(1 + \frac{\tau_e}{t} \right)$$

Where I_t is the threshold current, I_o is the minimum threshold current (called rheobase or rheobasic current), t is the duration of the stimulus, and τ_e is the strength-duration time constant, which describes the transition point along the t axis between falling and minimum plateau phases of the strength duration curve. For a given phase duration the threshold is affected by wave form and charge balance (area under positive and negative portion of current waveform). An interpretation of Figure 2 is that, for long duration monophasic stimuli, the threshold of excitation converges to a minimum current value referred to as the rheobase (I_o), and for short duration the threshold of excitation converges to a minimum

charge value. At short phase duration the neurological response is based on the amount of electrical charge released into the nerve. Note that for a rectangular monophasic pulse, $Q = I_t$, where Q is delivered charge.

Strength-duration threshold response data for **single-pulse** and **single cycle** stimuli are reported in Table I and Figure 3. Waveforms tested included **biphasic sinusoidal**, **monophasic sinusoidal** and **monophasic square** waves with a phase duration of 10 microseconds. The data points and curve fit in Figure 3 are for the mean response of the sample. Maximum, minimum and standard deviation of the samples are also reported in Table I. As predicted by the myelinated nerve model, the threshold current for biphasic pulse is higher than for a monophasic pulse. Likewise the threshold current for a monophasic, sinusoidal pulse was greater than for a monophasic square wave.

The same data is presented in Figure 4 in the form of threshold charge. Again the results agree with the form predicted by the myelinated nerve model. For short duration monophasic stimuli sensitivity is determined by threshold charge rather than peak current. For biphasic stimuli, however, the curve relating threshold phase charge and phase duration is not monotonic, ie., the minimum phase charge is attained at a particular value of phase duration. The ratio between the biphasic sine and monophasic square wave threshold charge at 10 microsecond phase duration is 5.6:1. The ratio between the threshold current for the biphasic sine and monophasic square wave is 7.9:1. Thus the threshold phase charge is less affected by waveform than peak current for short duration phase charge. With very short transient wave forms however, it is necessary to refer to a strength duration curve in order to accurately predict sensitivity.

2.2.2. Effect of Floor Surface

No difference in sensitivity to current was detected for the various floor surfaces tested. Estimates of the current flow though a cow made with a 4x4 inch copper plate on a wetted concrete surface, using a 500 Ohm shunt resistor, agreed well with the worst case (lowest impedance) floor conditions tested.

2.2.3. Effect of number of cycles

Figure 5 illustrates another type of waveform sensitivity through a strength-frequency curve, which represents the peak sinusoidal current threshold for neural excitation versus the frequency of a sinusoidal stimulus (Reilly 1992, 1994). The two solid curves were derived from the myelinated nerve model. Note that as the frequency is increased above a critical value, the threshold rises with frequency. For a brief single cycle stimulus, the frequency effect is greatest. For a continuous stimulus the threshold rises approximately in direct proportion to the frequency for frequencies above a critical value (f_c). The theoretical strength frequency curves may be fitted with the following expression over the range 10 Hz to 10 kHz (2).

$$I_t = I_o [1 - \exp(-f_e / f)]^a [1 - \exp(-f / f_o)]^b$$

Data comparing single-cycle sine and multiple cycle sine wave are presented in Table I and Figures 6. The threshold current was lower for the multiple cycle events than for single cycle events. The form of the response agrees well with the SENN model which predicts that the effect of the number of cycles will increase as the frequency of the stimuli increases.

2.2.4. Effects of Hoof condition and hoof trimming

The results of the T-tests for the comparison of Pre and Post hoof trimming Reaction Levels are shown in Table II. No significant difference was found in the reaction levels. There was 85 percent power to find a 3 mA difference in the Reaction Levels of cows at Pre trimming and Post1 trimming and 95 percent power to find a 3 mA difference in the reaction level of cows at per trimming and Post2 trimming.

Table II. Reaction Level Comparisons for before and after hoof trimming.

Treatment group	Number of cows	Average Dif. In Reaction	Std. Dev of dif..	Paired T-test P-value	Sig at 95% level
Pre-Post1	12	-1.3	3.4	0.213	No
Pre-Post2	10	-0.2	2.6	0.815	No

The results of the T-tests for cow resistance are shown in Table III. Cow resistance was found to be higher immediately after hoof trimming but returned to the pre-trimming level one week after trimming.

Table III Comparison of Cow Resistance before and after trimming.

Treatment group	Number of cows	Average Dif.. In Resistance	Std. Dev of Dif..	Paired T-test P-value	Sig at 95% level
Pre-Post1	12	-59.8	33.4	0.001	Yes
Pre-Post2	10	-4.3	56.1	0.82	No

Five of the 12 cows tested had at least one abscess on one hoof. The cows were divided into two groups based on the presents or absence of abscess. The results of an independent T-test comparison are shown in Table IV. No significant difference was found between these groups for Reaction Level or Cow resistance.

Based on these results it is unlikely that of hoof trimming will have a significant effect on cow sensitivity. The increased resistance immediately after trimming is probably due to the hoof being cleaned of debris

Table IV. Comparison of reaction level with and without hoof abscess.

Comparison	Mean No Abscess	Std. Dev No Abscess	Mean Abscess	Std. Dev Abscess	P-value Ind. Test	Sig at 95% level
Cow Reaction	11.1	2.7	13.7	5.1	0.28	No
Cow Resistance	309.1	50.9	317.6	64.6	0.80	No

The power to find a 50 percent change in cow reaction levels from 11.1 mA to 16.7 mA was 70 percent. The power to find a 25 percent change in cow resistance from 309.1 Ohms to 386 Ohms was 73 percent.

2.2.5. Milking Machine exposure Results

When the milk flow sensor was installed as recommended, (with one ring properly grounded), and supplied with the design sensing signal, there was no measurable current flow through the milk hose for either orientation of the sensor. With all grounding completely removed, and the sensor supplied with the designed sensing signal, the current flow through any cow was less than 0.1 mA for either orientation of the sensor. The sensing signal will not be perceptible to cows even when installed without grounding. The orientation of the sensor will also have no effect on dairy cow perception of the sensing signal.

When the recommended grounding was completely removed up to 1 volt could be measured between the claw and ground. This open circuit, claw to earth voltage during milking was not a reliable measurement of the current flow through the cow or milk hose, or of milk hose impedance.

When the sensing voltage was amplified over 1000 times (1056 volts on the sensing ring) one of three cows reacted to the stimulus. This voltage produced 50 to 66 mA of current through the milking unit/cow circuit. When the sensing voltage was amplified, the total impedance of the 3-foot milk hose, milking unit and cow circuit was measured at 16,000 to 21,000 ohms. Or 5300 to 7000 Ohms per foot of milk hose. This is somewhat lower than that predicted by Gustafson (1983) for peak milk flow in a low line system (26,000 Ohms). This lower impedance corresponds with reduced impedance at high frequencies in humans, cows and other biological materials.

When the 25 micro second sine wave was applied to the teat to hooves pathway no animal response was detected with a peak current flow of 30 to 40 milliamps. The contact voltage from teat to floor for a 30 milliamp pulse was 9.2 volts and teat to hooves of 300 Ohms. The rump-to-ankle voltage was 2.4 volts. When the teat to hooves voltage was increased to 55 Volts with a current flow of 200 milliamps an animal reaction was recorded. When a 42 milliamp current was applied from the rump-to-ankle pathway the rump-to-ankle voltage was 11.2 volts.

2.2.6 60 Hz Transient Current Sensitivity Distribution

The sensitivity of 120 cows to a single cycle 60 Hz transient current have been measured in these study. A cumulative frequency distribution is presented in Figure 7. Because of the wide range in sensitivities, it is recommended that animal response for a given level of current exposure be stated as the percentage of animals affected.

3. Animal Aversion and Health Response to Short Duration Transient Current

3.1. Aversion Materials and Methods

3.1.1. 6000 Hz Aversion Study

Tests were performed to evaluate the aversion response to short phase duration transients. A similar methodology was used as a previous study on 60 Hz transients (Reinemann et al, 1995b) to determine the relationship between short term behavioral response and aversion to longer term exposure. The stimulus current administered to the water bowls for these tests was a single cycle, sinusoidal transient with a phase duration of 83 microseconds (Effective frequency 6000 Hz). Before each test the sensitivity of all subject cows to the 83 microsecond transient current was determined as described in section 2.1.2.

Each test was performed with 8 cows of which 2 were controls, receiving no stimulus, 3 cows exposed at their reaction level (R) and 3 cows exposed at 1.5 times their reaction level (R*). Three groups of 8 cows were tested (24 cows total). Previous studies with 60 Hz transients indicated that changes in drinking behavior and water volume were observed on the first day of exposure, particularly in the time interval immediately after milking. The transient current was applied to the water bowl so that the first exposure occurred when the cows returned to their stalls after the morning milking. The stimuli were applied to the water bowl once per second for a period of 28 hours.

The Fishers Exact test was used to test for association between delay to drink and stimulus level with the null hypothesis H_0 : no association and the alternative hypothesis H_1 : association. The delay to drink categories are define as 0 to 3 hours for the controls. The mean delay to drink times for the control cows in a previous study with a single cycle 60 Hz transient study was 1.3 hours. It was expected that all the control cows would drink within three hours and was used for the first time interval of interest. The second time interval was chosen as 12 hours for the time of exposure which extended to two hours after the evening milking. The second time interval was chosen as 24 hours from the time of exposure. Previous studies indicated that if aversion continued for 24 hour, there was also a decline in milk production.

Independent non-paired one tailed T-test with unequal variances were used for the comparison of mean delay to drink for the control cows and treatment cows. A one tailed test was used because it was not expected that the treatment cows would have a shorter delay to drink, or more water consumed than the control cows. In the three trials run for this study, all the control cows drank in the first hour after the morning milking.

Paired two sided T-tests were used for the comparisons of and total daily water consumed for the control and treatment cows. Two measurements before and two measurements after the shock day were averaged and compared with the water volume on the treatment day. Two day averages before and after the shock were used to avoid confounding.

3.1.2. Blood Composition

In the previously reported experiment with three week exposure to single cycle 60 Hz transient current (Reinemann et al, 1995b), blood was collected from all cattle three times per week during the two week pre-treatment, three week treatment, and two week post-treatment periods. Serum was separated by centrifugation and frozen at -10 C until selected serum biochemical and electrolyte assays were performed (Johnson & Johnson Vitros 250 Analyzer, formerly known as Kodak Ektachem 250 Analyzer, Johnson & Johnson Clinical Diagnostics, Rochester, NY.). Serum electrolyte and biochemical analyses included sodium, potassium, chloride, enzymatic CO₂, calcium, phosphorus, magnesium, glucose, creatinine, albumin, and creatine kinase concentrations.

3.2 Aversion Results and Discussion

3.2.1 6000 Hz Aversion Results

A cross table with the associated frequencies used for the Fisher=s Exact Test is shown in Table V. The hypotheses tested were H₀, no association and H₁, association between stimulus level and delay to drink. The Fisher=s Exact Test shows a high probability of association between Delay To Drink times and Treatment level.

Table V. Contingency Table for the Fisher=s Exact Test.

	Delay To Drink			Totals
	0-3 hours	3.01-12 hours	12.01-24 hours	
Control	6	0	0	6
R	2	6	1	9
R*	1	4	4	9
Totals	9	10	5	16

Fisher=s Exact Test (2-Tail) P-Value=0.003
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The T tests also indicated a significant difference in delay to drink times between the control and the R and the control and R* cows (Table VI). The power to find a 100 percent deference between the R and the R* cows was approximately 63 percent--assuming equal variance. The actual difference between the R and R* delay to drink times was 85 percent when using

the R cows as a reference.

Table VI. Results of T-test comparison of delay to drink.

Comparison groups	T-test ^a P-value	Sig at 95% level
Controls to R	0.011	Yes
Control to R*	0.001	Yes
R to R*	0.065	No

^a One tailed independent T-test.

Table VII. Results of T-test comparison of total daily water consumed.

Treatment group	Number of cows	Average change in water consumption	Paired T-test P-value	Sig at 95% level
Controls	6	0.3 gallons more	0.88	NO
R	9	4.1 gallons less	0.27	NO
R*	9	9.6 gallons less	0.03	YES

Cows exposed at their reaction level (R) to 1 cycle of 6000 Hz (0.083 millisecond phase Duration) showed significant delay to drink, but did not show a significant reduction in water intake (Table VII). Cows exposed at R* showed a significant delay to drink and a significant reduction in water consumption.

The results of the 6000 Hz transient compared well with those using a 60 Hz transient, i.e. Some changes in drinking behavior but not for total water volume were observed for cows exposed at a level which first produced a behavioral response. Delay to drink and total water consumed were reduced when animal were exposed at levels 1.5 times higher than the level which produced a behavioral reaction.

3.2.2. Blood Composition Results

The measured blood parameters from individual cows remained within reference intervals defined for healthy cattle (Purdue University School of Veterinary Medicine Reference Intervals for healthy cattle, 1985) throughout the experimental period. No significant differences between treatment and either pre or post-treatment periods were found for any of the parameters.

The panel of electrolyte and serum biochemical parameters used in this trial were selected based on their established sensitivity to feed deprivation (McGuirk, 1988), changes in water intake (hydration status) or acid-base balance, muscle injury, stress, and kidney function of cattle. The absence of significant changes in these laboratory data in treatment cattle over

time (each cow serving as their own control) as well as the lack of difference between treatment and control cows indicate that there was no alteration in circulating volume or acid-base balance, nor was there significant stress (as measured by glucose concentration), or muscle injury inflicted by the treatment. The changes observed in individual animals remained within reference intervals and therefore has no biological significance. Food deprivation in non-lactating cattle for 48 hours caused changes in glucose, calcium, and enzymatic CO₂ concentrations in an earlier study (McGuirk, 1988). The absence of change in lactating cattle in this study suggests that feed intake changes were not sufficient to cause physiological response. The power to find clinically significant differences in the variables measured was in excess of 88% for all variables except Enzymatic CO₂ (25%) and Creatinine Kinase (40%). The power calculations were based on normal distributions.

Serum **sodium** concentrations reflect a ratio of the total body sodium content to total body water content. Lactating cows lose a little sodium in milk (concentration of sodium in milk is 1/10th of concentration in serum) which would lower sodium content. If a cow does not drink, it is expected that total body water decreases and serum sodium concentration increases. This effect was not observed in this study.

Albumin is another measure of hydration status. Albumin concentration increases when cows are deprived of water. This effect was observed in an another study (McGuirk, 1988) in which cows were deprived of food for 48 hours with the affect attributed to lower total water intake.

Serum **potassium** concentration depends on the total body potassium content and the acid-base status of the animal. Intake is primarily dietary and dietary restriction can lower total body potassium without affecting serum K concentration. Also, small changes in acid-base balance can occur without detecting changes in K. Total body potassium could go up in the face of some dehydration because the kidney would excrete less. Our study shows no significant dehydration so kidney elimination should not have been a problem. Intake could have been reduced but it wasn't significant enough to have an impact on our values.

Enzymatic CO₂ is an indication of acid-base balance. This test represents the total quantity of CO₂ that can be liberated from serum, approximately 95% of which is bicarbonate. This test is an estimate of bicarbonate: lower bicarbonate concentrations result from acidosis and increased bicarbonates results alkalosis. We saw no change in CO₂ in this study. The absence of change in this study indicates that acid-base balance did not change and therefore potassium did not change due to acid-base balance changes.

Serum **chloride** changes tend to follow temporal patterns in healthy animals. Hypochloremia is associated with hyponatremia and increased bicarbonate concentrations. Hyperchloremia is associated with hypernatremia or decreased serum bicarbonate concentrations. In the fasting cow study (McGuirk, 1988), Cl concentration did go up, consistent with a decrease in CO₂ concentration (as the bicarbonate indicator). There was no evidence of chloride, sodium or bicarbonate changes in this study.

Low **calcium**, and albumin can be due to inadequate diet and lactational drain. **Phosphorus** changes usually are in the opposite direction of calcium. Changes can be due again to diet, kidney function changes or failure to eliminate it in the gi tract. We saw no change in calcium, phosphorous or albumin in this study.

Glucose concentration can increase when cows are stressed (painful, sick, etc) or decrease, particularly in post partum cows, when feed intake is decreased. In the fasted cow study (McGuirk, 1988) glucose decreased even though the cows were not lactating. No change in glucose was observed in this study.

Creatinine is a nonprotein nitrogen compound in serum that is formed from degradation of muscle. Its concentration, like most other compounds in serum, depends on the rate of synthesis and excretion. Therefore, increased concentration could come from increased muscle damage (synthesis) and/or decreased excretion. The latter is due to dehydration or kidney damage. This was a good test of hydration effects on kidney. No change in creatinine were observed in this study.

Creatine Kinase (CK) was included in this study because it has a high specificity for damaged muscle. CK is released from cells by cell damage. In our study, we might have anticipated increased CK from prolonged recumbency, trauma, Intramuscular shots, or increased activity associated with the anticipation or avoidance of electric shock. The reference values vary widely because of varying activity and trauma levels unrelated to this study. There was a slight increase in the CK level during the treatment period but the increase was not significant.

4. Summary and Conclusions

The reaction threshold of dairy cows was determined for transient voltages ranging in phase duration from 8333 milliseconds to 10 microseconds (frequency from 60 to 50,000 Hz) and for single and multiple cycle stimuli. The strength-duration relationship for dairy cow response to transient voltage pulses followed the form of previously developed by neurological models. Sensitivity was affected primarily by phase duration with secondary effects of waveform. Cows responded at lower threshold current for multiple as compared to single cycle stimuli of the same phase duration.

Milkhose impedance was slightly lower for a short duration current pulse (16,000 to 20,000 Ohms) than that previously measured for 60 Hz, AC current (26,000 to 30,000 Ohms) for a 1 meter milk hose. The current pulse used in the milk flow sensor tested did pose a threat to cows because of its short phase duration, high milkhose impedance and low peak current level. Even in the worst case scenario, when all grounding is removed and for any orientation of the sensing rings, the sensing signal is more than 500 times lower than the minimum level perceptible by a cow.

Cow impedance increased slightly immediately after hoof trimming but returned to pre-trimming values after one week. The increased resistance immediately after trimming is probably due to the hoof being cleaned of debris. No difference in animal sensitivity to current was detected

after hoof trimming.

A significant difference was found in delay to drink times between the control cows and cows exposed at their reaction level (R) and 1.5 times their reaction level (R*) to single cycle, 6000 Hz stimuli applied to the water bowl. The p-values for the control group to R group was 0.011. The p-value for the control group to R* group was 0.001. A Fisher's exact test using a 3 by 3 table thus showed association between delay to drink and treatment level. A statistically significant drop in water consumption over the 24 hour treatment period was found for the R* cows. The p-value was 0.03. The relationship between behavioral reaction to brief exposure and aversion to long term exposure agreed well to previous studies using single cycle 60 Hz stimuli.

The measured blood parameters from individual cows remained within reference intervals defined for healthy cattle throughout a three exposure period to single-cycle, 60 Hz current applied to the water bowl. No significant differences between treatment and either pre or post-treatment periods were found for any of the parameters. All reactions were explainable by aversion to unpleasant stimuli rather than direct physiological response.

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Table I. Transient Current Test Summary.

Test	A	B	C	D	E	F	G	H	I	J	K	L
Phase duration, Tp (:s)	8333	8333	8333	1000	83	83	83	10	10	10	10	10
Frequency (Hz)	60	60	60	500	6k	6k	6k	50k	50k	50k	50k	50k
Number of cycles	1	5	9	1	1	9	100	1	1	1	5	833
Bi/Mono phasic	B	B	B	B	B	B	B	M	M	B	B	B
Waveform	Sine	Sine	Sine	Sine	Sine	Sine	Sine	Square	Sine	Sine	Sine	Sine
Number of Animals	120	24	24	24	24	23	12	12	8	8	8	12
Mean Threshold (Peak mA)	9.6	7.1	7.6	14	71	58	36	182	543	1308	695	186
Standard Deviation	3.0	1.5	2.6	4.9	15	19	5.8	37	42	164	85	38
Min Threshold (Peak mA)	2.8	4.4	2	6.3	41	30	28	129	480	1000	600	124
Max Threshold (Peak mA)	19.1	10.5	13	27	103	103	48	257	600	1500	820	246

Figure 1. Schematic of milking equipment and electrical exposure test apparatus.

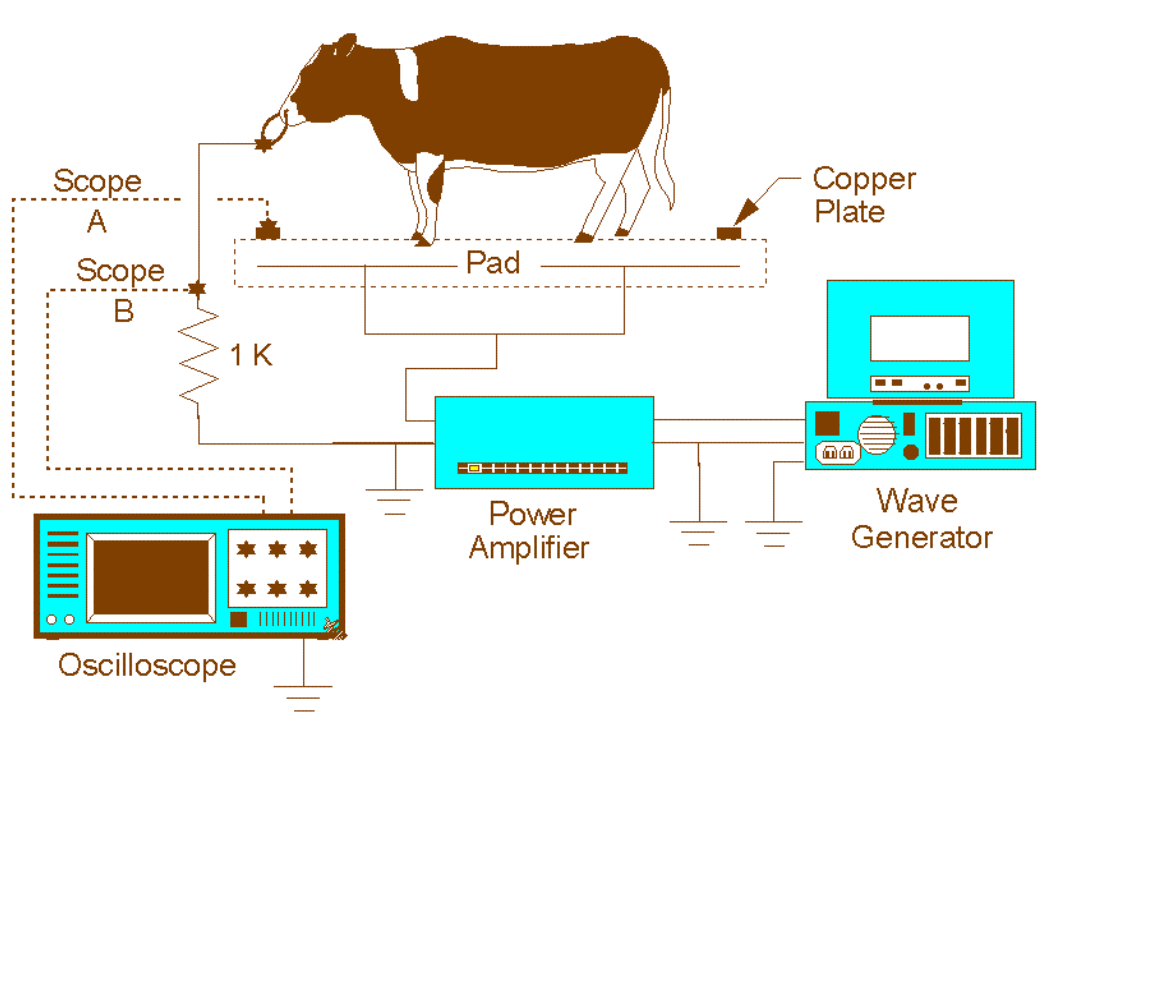


Figure 2. Strength-duration relationships derived from the myelinated nerve model: current thresholds and charge thresholds for single-pulse monophasic and for single-cycle biphasic stimuli with initial cathodal phase, point electrode 2 mm distant from 20 μ m fiber. Threshold current (T_p) refers to the peak of the stimulus waveform. Charge refers to a single phase for biphasic stimuli. (From Reilly, 1992).

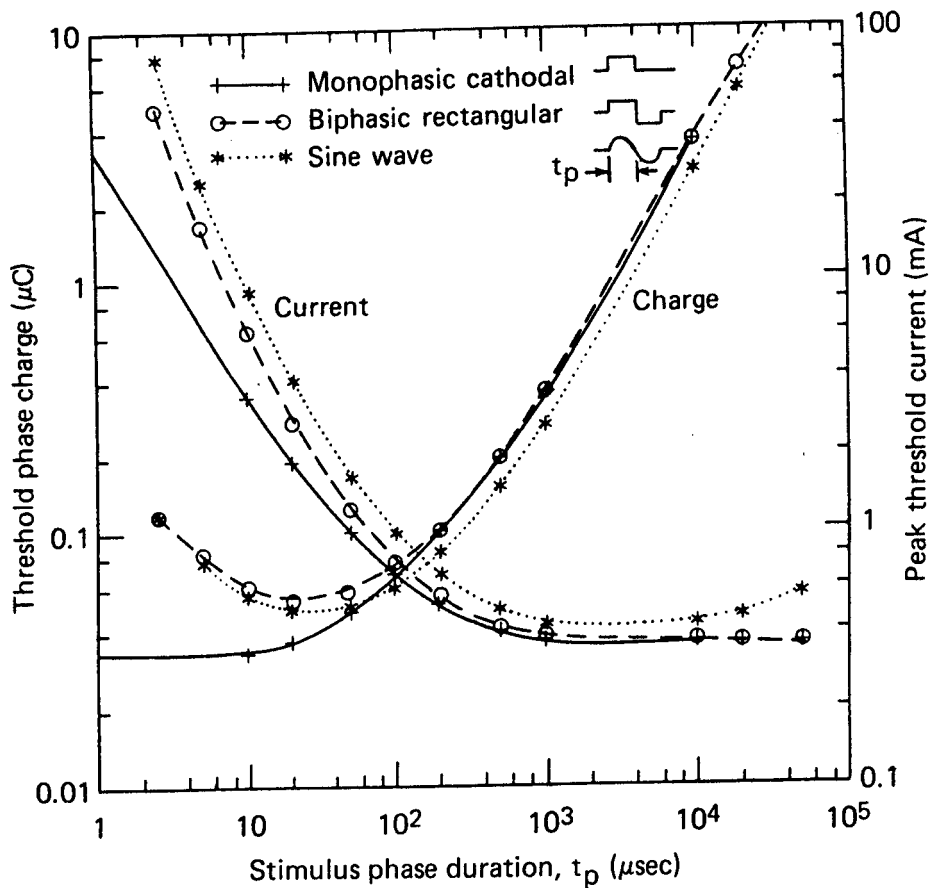


Figure 3. Strength-duration threshold current data for **single-pulse and single cycle** stimuli.

Threshold current reverts to the peak of the stimulus waveform. Data are for the mean response threshold for the sample group.

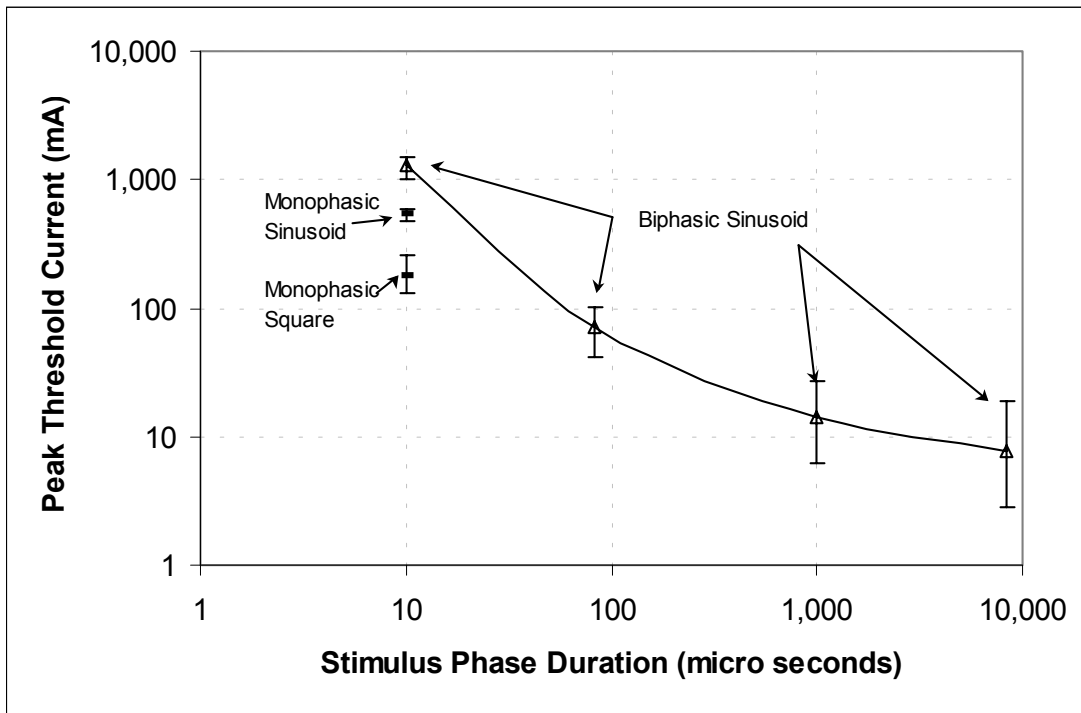
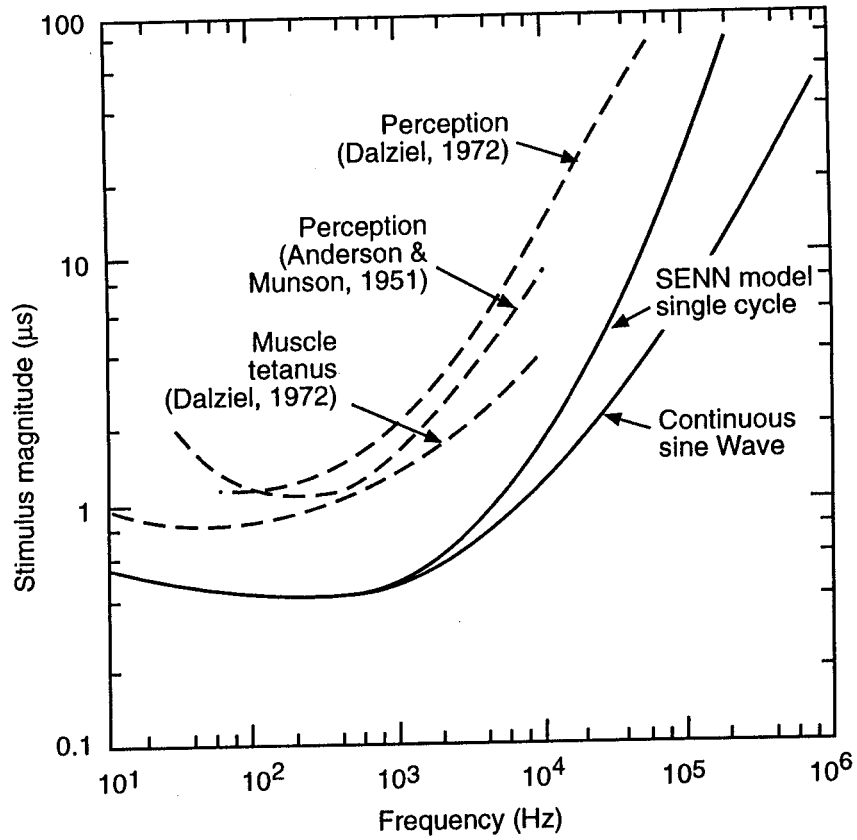


Figure 4. Strength-duration threshold charge data for **single-pulse and single cycle** stimuli. Charge refers to a single phase for biphasic stimuli. Data are for the mean response threshold for the sample group.

Figure 5. Strength-frequency curves for sinusoidal current stimuli. Dashed curves are from human experimental data. Experimental curves have been shifted vertically to facilitate comparisons. (From Reilly 1992).



Figures 6. Strength-frequency threshold current data for single and multiple cycle sinusoidal stimuli. Threshold current refers to the peak of the stimulus waveform where an animal response was recorded. Data are for the mean response threshold for the sample group.

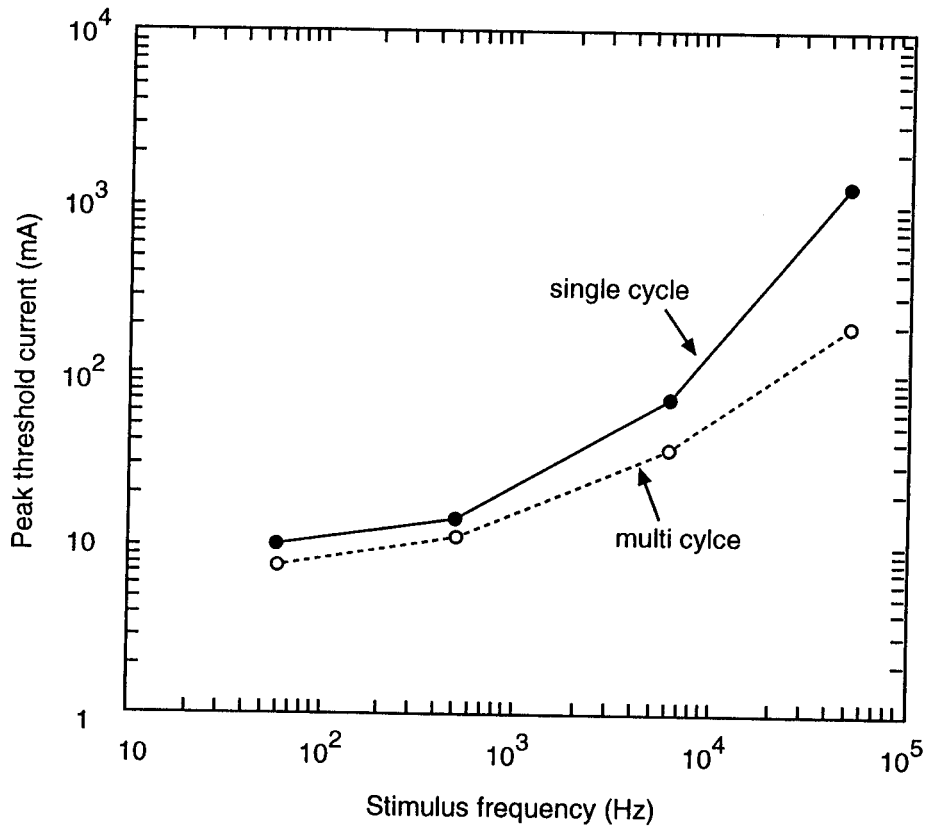


Figure 7. Cumulative frequency distribution for reaction threshold of 120 cows diary cows for

single-cycle, 60 Hz transient current. Threshold current refers to the peak of the stimulus waveform where an animal responded.

