Clinical and electrocardiographic evidence of death following use of a poll shot with a penetrating captive bolt as a single-step method for euthanasia of cattle

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Abstract

The objective of this study was to validate the effectiveness of a penetrating captive bolt when positioned behind the poll as a single-step method for humane euthanasia of cattle. Seventeen bovine animals of various ages (neonate to mature) that were eligible for euthanasia were enrolled in the study. Cattle were euthanized using a penetrating captive bolt gun with an extended bolt positioned flush against the parietal bone on midline just behind the poll and aimed toward the base of the tongue. Clinical and ECG parameters were monitored and recorded following application of the poll shot method. One animal required a second shot due to improper angle of the captive bolt device. All properly placed and aimed shots successfully induced immediate loss of consciousness without return to sensibility which was followed by clinical death. Once unconscious, none of the cattle required a secondary step to assure clinical death. Following death, the head was removed, transected sagittally on midline, and brain trauma was assessed and scored. A properly placed and aimed poll shot was determined to be an effective single-step method of euthanasia of cattle when using a penetrating captive bolt.

Key words: bovine, euthanasia, penetrating captive bolt

Introduction

Euthanasia is a term originating from the Greek words *thanatos,* which means death, and the word *eu,* which means good. Thus, euthanasia means "good death". As such, euthanasia is meant to describe termination of life in a manner that reduces or eliminates pain as well as distress the animal may experience prior to onset of unconsciousness. Selected euthanasia techniques should consistently produce rapid loss of consciousness proceeded by cessation of respiratory or cardiac function and brain death.[1] Human safety and mental health should also figure into the selection of a euthanasia method. The American Veterinary Medical Association (AVMA) Guidelines for Euthanasia and the American Association of Bovine Practitioners lists a frontal shot as the recommended anatomical location for both penetrating captive bolt (PCB) and gunshot euthanasia.[1,2] The frontal shot has been well-described in several publications (Gilliam et al. 2012; Dewell et al. 2016; Shearer, 2018; Shearer et al. 2018).[3,4,5,6]

Perhaps the simplest, most consistent, and reliable description of the frontal shot when using a PCB or firearm is to: "direct the trajectory of the PCB perpendicular or slightly downward to the frontal sinus plane on midline between the base of the ears at the level of the external acoustic meatus".[4] However, in some situations, access to the frontal site for PCB euthanasia is not possible (i.e. fractious animal, suboptimal restraint, etc.) or may cause excessive distress to the animal because frontal shot placement requires the operator to stand directly in front of the animal. Placement of the PCB shot behind the poll is an alternative to the frontal site and has been commonly used in bovine practice. Currently, there is a paucity of data about this technique in bovids in the peer-reviewed literature. Review of the poll shot technique in water buffalo indicates that proper placement of the shot is critical to achieve effective euthanasia.[7] However, the skull anatomy of the water buffalo is substantially different compared to domestic bovids, with significant deviations in the morphometry of the occipital bone, which could affect the placement, trajectory and outcome of poll shot euthanasia.[8] A recent study compared a poll shot to a frontal shot as a secondary step following initial use of a frontal shot PCB.[9] Although the poll shot was found to be comparable to the frontal shot as a secondary step, this study did not explore efficacy of poll shots alone as a primary means of euthanasia. Further, the study was limited to adult cattle. A similar poll shot approach has also been described in sheep and goats[10] and is a recommended method in the AVMA Guidelines for the Euthanasia of Animals.[1]

When selecting a euthanasia method for a bovine, it is important to consider whether the obex will be left intact. The obex is located in the caudal extent of the fourth ventricle as it narrows and transitions to the central canal.[11] Obex tissue is required for diagnosis of bovine spongiform encephalopathy (BSE). Thus, preservation of this structure is critical to meet ongoing USDA BSE surveillance program objectives including animals greater than 30 months of age that meet sampling criteria as well as cattle at least 12 months of age exhibiting clinical signs of a central nervous system (CNS) disorder.[12]

Compared to frontal placement of the PCB, the poll shot is advantageous as this technique may cause less distress to the animal. When using a frontal placement method, the operator must place the PCB flush against the frontal bone by positioning themselves in close proximity directly in front of the

animal where their flight zone is larger compared to other areas of their body.[13] This may be particularly true for cattle that are less acclimated to regular human encounters.

Use of the poll shot approach for a penetrating captive bolt euthanasia may also benefit human safety compared to frontal placement. In contrast to frontal placement, a poll shot does not require the operator to stand directly in front of the animal to properly place the trajectory. Standing directly in front of a bovine could be interpreted as a threat by the bovine and may also place the operator in danger if the animal is restrained in such a way that it is able to lunge forward. The authors have personal experience with the aforementioned occurrences when using the frontal approach. This potential may be exacerbated when cattle are not well acclimated to human contact. Of course, human safety is paramount and both approaches require that the animal be properly restrained by mechanical means and/or chemical sedation.

Surveys of dairy workers have identified that euthanasia has a significant emotional toll on livestock caregivers.[14,15] Compared to a frontal shot method, a poll shot can be less aversive to the operator since they are not looking directly at the animal's face and eyes when administering a PCB.

The objective of this study was to assess the use of a properly placed poll shot technique using a PCB device as a single-step method to render an animal unconscious and cause clinical death. Additionally, guidance for proper PCB placement for poll shot euthanasia was determined. Finally, this study assessed the euthanasia method's potential impact on obex tissue.

Materials and methods

This study was approved by Iowa State University (ISU) Institutional Animal Care and Use Committee (IACUC #18-330). Cattle eligible for euthanasia at ISU were enrolled in the study. Cattle were either being euthanized for other research projects or were identified for euthanasia by a clinical veterinarian because of illness or injury. Individual demographic data including identification, euthanasia date, sex, estimated age, breed type and reason for euthanasia was recorded. Cattle were restrained just prior to euthanasia using a bovine halter either in a headgate or pen as warranted for safety of cattle and investigators. Following restraint, Mortara ELI 230[a] electrocardiogram (ECG) leads were attached. Leads were connected using subcutaneously placed stainless steel wire loops and connected to alligator clips fixed onto the monitor cable. Isopropyl alcohol was applied to the skin in each lead area to improve conductivity. Leads were placed in the following locations:

- Yellow electrode in the left distal jugular furrow
- Black electrode in the right distal jugular furrow
- Green electrode on the left lateral thoracic wall at the level of the olecranon
- Red electrode was placed approximately 10 cm ventral from thoracic vertebra 10

An ECG tracing was run for approximately 2 minutes to establish accurate baseline readings prior to euthanasia.

A CASH® Special[b] captive bolt gun with a 11.4 mm diameter, 206.5 mm extended length bolt was then placed in the recess directly behind the animal's poll and directed toward the base of the tongue (performed by GD) (Figure 1). Appropriately sized bolt gun cartridges based upon the animal's stage of life were used according to manufacturer recommendations.

Figure 1: Correct placement of PCB initiated in the recess directly behind the animal's poll and directed toward the base of the tongue.

Time was noted at application of the bolt gun firing and clinical assessment was initiated.[9,16] Clinical assessment included presence or absence of: corneal reflex, palpebral reflex, righting reflex, vocalization, rhythmic respiration, involuntary spinal reflex, rhythmic heartbeat and auscultable heartbeat (performed by RD and KH). Clinical death was defined as the cessation of respiration and the absence of an auscultable heartbeat. Clinical scores were recorded every minute until clinical death was determined. The ECG was recorded from time of baseline until flatline was observed for at least 2 minutes or until the length of maximum paper recording (approximately 20 minutes). The ECG tracing was interpreted by a trained technician with a specialty certificate in cardiology (performed by LM). During examination of the ECG, heart rate and rhythm were evaluated. Notations were made regarding heart rate decreases and rhythm abnormalities as well as when a lack of recordable electrical activity ("flatline") occurred.

After determination of clinical death and cessation of ECG recording, the head was removed at the atlantooccipital joint and bisected sagittally with a bandsaw to permit evaluation

of bolt trajectory and tissue damage. Brain tissues were subsequently removed and subjected to gross evaluation of tissue damage by a boarded veterinary anatomic pathologist (performed by AM). Tissue destruction scores (0 = none, 1 <25%, 2 = 25-50%, 3 >50% damage) were assigned based on the degree of architectural disturbance within the medulla, pons, cerebellum, midbrain and cerebrum similar to that of Derscheid et al. (2016).[18] The obex was grossly evaluated for damage and acceptability for diagnostic testing for BSE.

Data collection and analysis

Cattle demographic information, clinical assessment data, ECG data and brain tissue trauma data were collated into a spreadsheet[c]. Since there was no null hypotheses to be tested, descriptive statistics were generated in Excel using the individual animal as the unit of analysis. Descriptive statistics included means, medians, ranges and standard deviation for the following variables: Rhythmic heartbeat, Auscultable heartbeat, ECG flatline and Brain tissue trauma scores. The individual brain trauma scores for each region (Cerebellum, Midbrain, Pons, Medulla and Occipital Cortex) were added together and categorized into a composite brain trauma score (adapted from Derscheid et al. 2016)[18] on a scale from 0 to 4 (0 = combined score of 0, 1 = combined score of 1 to 3, 2 = combined score of 4 to 6, 3 = combined score of 7 to 9, and 4 = combined score of 10 or more). One-way ANOVA was used to evaluate the association between age and time to clinical death and composite brain trauma score and time to clinical death.

Results

Seventeen animals were enrolled in the study (Table 1) and euthanized using the poll shot method. Sixteen of the 17 enrolled animals were successfully euthanized with a single poll shot. One animal required a secondary poll shot due to suboptimal positioning of the initial PCB placement (operator error) leading to removal of this animal's clinical and tissue trauma data from the study as it is confounded by 2 shots. All animals (16/17) that were correctly targeted with the PCB were rendered immediately unconscious (i.e. collapsed, no vocalization, and found to be absent of: a corneal reflex, a palpebral reflex, rhythmic respiration, or righting reflex). All but one animal achieved clinical death (lack of auscultable heartbeat) within 12 minutes of the poll shot application. Two animals clinical data were not included within the dataset for analysis due to lack of clinical assessment data.

For all animals with appropriate poll shot placement, the palpebral and corneal reflexes, righting reflex, vocalizations and rhythmic respirations were absent immediately following PCB application. A normal heart rhythm was auscultated for a

Table 1: Demographic data of euthanized cattle with time to loss of auscultable heartbeat (clinical death).

Animal ID	Number of shots	Reason for euthanasia	Breed type	Sex	Age	Complete data	Auscultable heartbeat (min)
1	1	Research	Holstein cross	Female	Weaned calf	Yes	8
2	1	Research	Holstein cross	Female	Weaned calf	Yes	8
3	1	Research	Holstein cross	Female	Weaned calf	Yes	9
4	1	Research	Holstein cross	Female	Weaned calf	Yes	9
5	1	Research	Holstein cross	Female	Weaned calf	Yes	8
6	1	Research	Holstein	Female	Adult	No	
7	1	Illness	Angus X	Male	Yearling	Yes	10
8	2	Illness	Angus X	Male	Yearling	N/A	N/A
9	1	Research	Holstein	Male	Neonate	Yes	11
10	1	Research	Holstein	Male	Neonate	Yes	12
11	1	Research	Holstein	Male	Neonate	Yes	17
12	1	Research	Holstein	Female	Adult	Yes	7
13	1	Research	Holstein	Female	Adult	Yes	8
14	1	Research	Holstein	Female	Adult	No	
15	1	Illness	Holstein	Female	Adult	Yes	10
16	1	Illness	Holstein	Female	Adult	Yes	12
17	1	Illness	Jersey	Female	Adult	Yes	9

N/A not applicable.

median time of 8 minutes (range 2 to 12 minutes) and clinical death (no auscultable heartbeat) median time was 9 minutes (range 7-17 minutes). There was a difference (*P* value = 0.03) in time to clinical death by age group (Figure 2). The mean time to ECG flatline was 13.6 minutes (median 12 minutes, range 10 to 19 minutes) (Table 2).

Of the 16 out of 17 enrolled animals that received a single shot, the cerebellum and rostral medulla had the most tissue damage with a mean score of 2.9 and 2.1; respectively (Table 3). All animals had greater than 25% damage to the cerebellum and 11 of the 16 animals had greater than 50% damage to the medulla which was transected in these animals (Figures 3, 4 and 5). There was no difference (*P* value = 0.44) in time to clinical death by composite brain trauma score (Figure 6). Only one animal of the 17 euthanized with poll shot exhibited mid- to caudal medullary damage. In this animal, the obex at the caudal-most extent of the 4th ventricle remained intact and was suitable for BSE testing. The remaining 16 animals had an intact obex that was also suitable for BSE testing.

Discussion

This study demonstrates that poll shot euthanasia is an effective one step method for cattle. In cases where the obex must be preserved for diagnostic testing, this method does not adversely affect sample integrity. Compared to a frontal approach with a PCB at close range, this method may be safer for the operator and elicit less stress on the animal as well as the human operator because it does not require standing directly in front of the animal at close range.

Of the seventeen total animals euthanized, only one animal did not achieve immediate loss of consciousness. This failure was due to suboptimal positioning of the PCB resulting from an operator error. More specifically, the error in positioning was due to angling the PCB too obtusely toward the muzzle instead of more acutely toward the base of the tongue. The PCB was immediately reloaded and a properly positioned poll shot was obtained within 60 seconds. As with all PCB and gunshot euthanasia, trained personnel are critical to obtain satisfactory results; however, occasional placement errors can occur even with experienced operators. The immediate loss of palpebral and corneal reflexes, righting reflex, rhythmic respiration and vocalization indicates a catastrophic disruption of central nervous function, and once absent, neither reflexes nor sensibility were ever regained. Thus, a properly placed poll shot meets criteria for a humane euthanasia method.

Few studies have reported cardiac activity after PCB; however, euthanasia guidelines recommend confirming death after euthanasia via auscultation of heartbeat. Cardiac output is, in part, a function of heart rate. When using a PCB, the decrease in cardiac output contributes to the damage caused by mechanical trauma because perfusion to the brain (and other organs) are decreased, leading to shock and death. Dewell et al. (2015) reported that clinical cardiac death, defined as cessation of rhythmic auscultable heartbeat, occurred in most calves at 10 minutes following application of PCB, while electrical cardiac death with ventricular standstill (ECG flatline) extended to 15 minutes.[16] In contrast to this study, animals in the 2015 study were euthanized using a frontal shot technique by a pneumatic captive bolt with a secondary air pithing

Figure 2: Scatter plot of time to clinical death in cattle euthanized by a poll shot PCB by age category (1 = neonate, 2 = weaned calf, 3 = yearling, 4 = adult), including median (clear box) and standard deviation (bars). Bold black bar and * indicate significant difference in time to clinical death by age category 1 and 2.

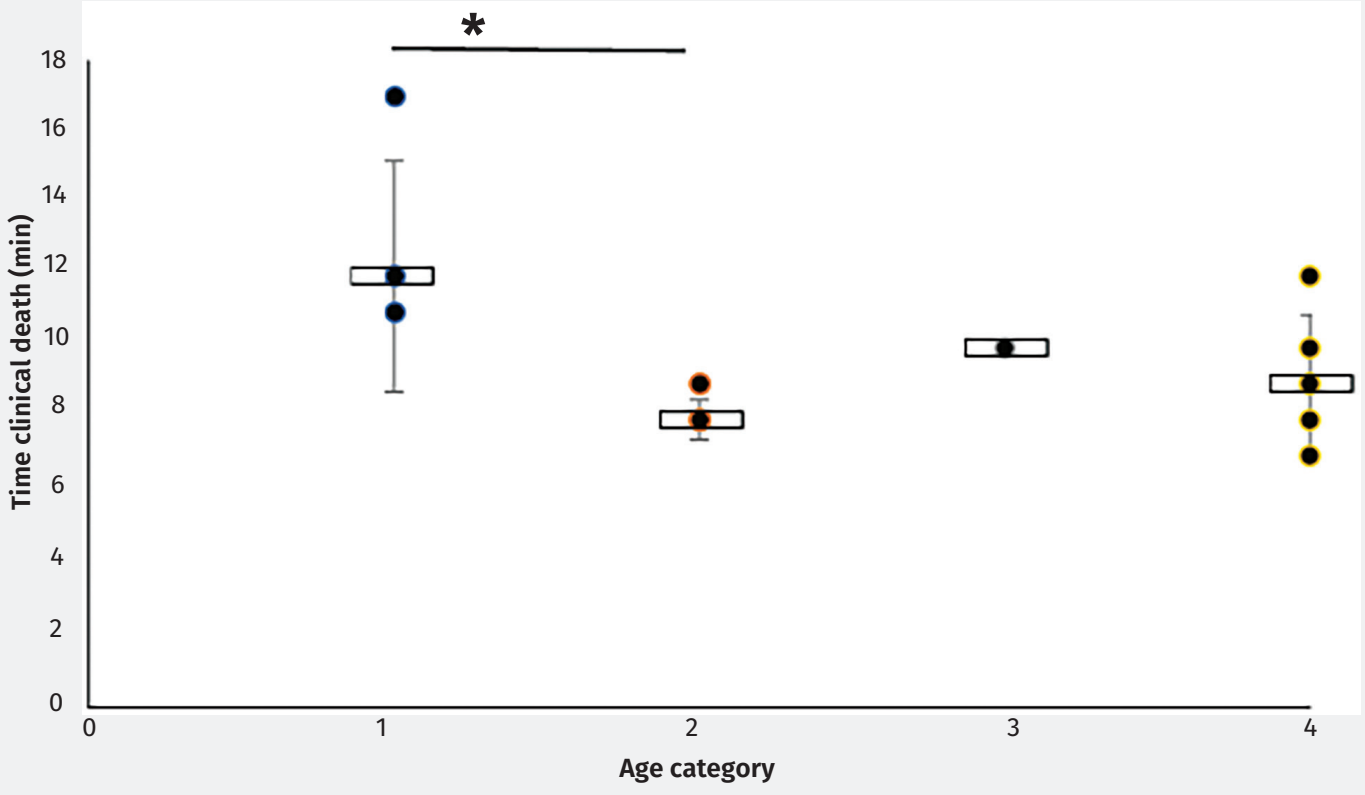


Table 2: Duration of cardiac parameters in cattle following poll shot PCB for euthanasia.

	Mean	Median	Minimum	Maximum
Rhythmic heartbeat (min)	N/A	8	2	12
Auscultable heartbeat (min)	N/A	9	7	17
ECG flatline (min)	13.6	12	10	19

N/A not applicable.

Table 3: Tissue destruction scores in brain tissue in cattle being euthanized by poll shot PCB.

	Mean	Median	Minimum	Maximum
Cerebellum	2.9	3	2	3
Midbrain	0.3	0	0	3
Pons	1.0	0	0	3
Medulla	2.1	3	0	3
Occipital cortex	0.1	0	0	1

Tissue destruction scores (0 = none, 1 < 25%, 2 = 25-50%, 3 > 50% damage).

Figure 3: Transection of adult cow skull showing PCB trajectory path from the recess directly behind the animal's poll and directed toward the base of the tongue as well as extensive damage to cerebellum and medulla.

Figure 4: Transection of an intact adult cow skull showing PCB trajectory path from the recess just behind the animal's poll and directed toward the base of the tongue.

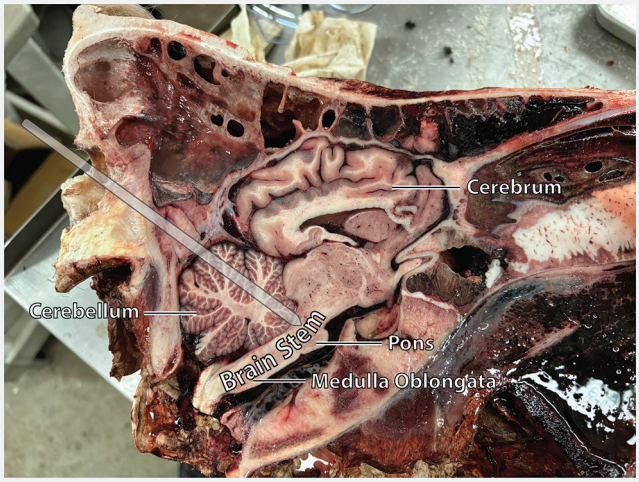


Figure 5: Calf brain shows damage to cerebellum and transection of brain stem at level of pons and medulla after euthanasia by poll shot PCB.

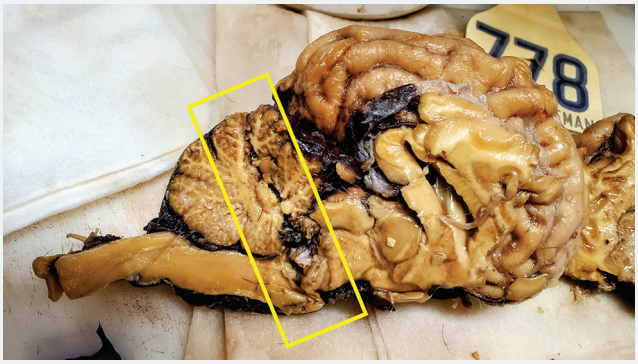
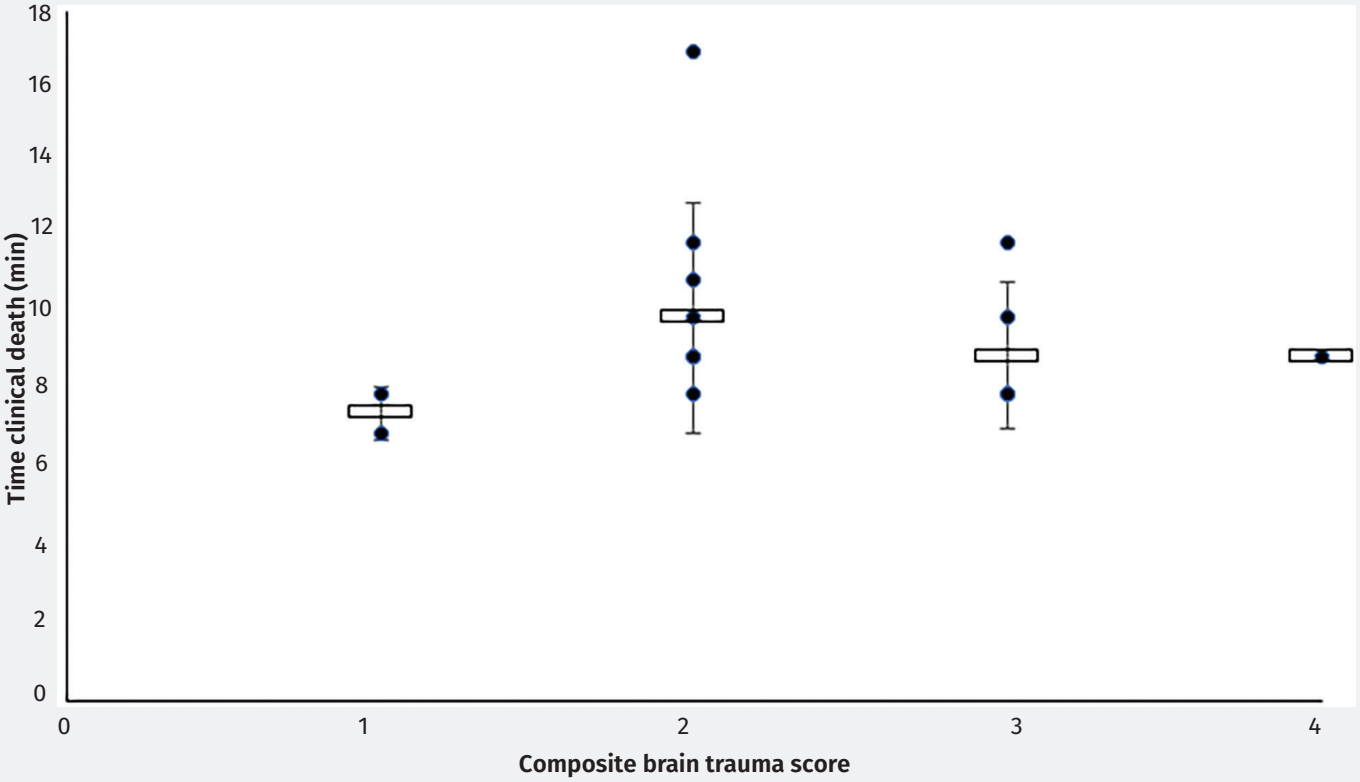


Figure 6: Time of clinical death and associated composite brain trauma score in cattle euthanized by poll shot PCB (1 = 1 to 3, 2 = 4 to 6, 3 = 7 to 9, 4 = 10 or more), including median (clear box) and standard deviation (bars).



mechanism, not a singular PCB application as performed in this study. In this present work, all but one animal reached clinical death by 12 minutes, which is comparable to the previous 2015 report which did not include younger animals. It is notable that neonatal calves had some of the longest clinical death and ECG flatline times. These three calves had clinical death at 11, 12 and 17 minutes and a corresponding ECG flatline of 18, 12 and 19 minutes, respectively. All of these calves had a transected medulla and greater that 50% damage to the cerebellum. The neonatal heart appeared to be more resilient to cessation of activity compared to older animals. Persistent cardiac function in neonatal animals after PCB euthanasia aligns with previously published data.[17]

Four of the study animals did not have ECG flatline observed by the trained cardiology veterinary technician. These were animals observed early in the study period and were found by the attending clinical veterinarians to have no auscultable heart rhythm and appearance of ECG flatline after poll shot application. Therefore, ECG recordings were terminated erroneously before true ECG flatline was obtained as determined by the trained cardiology veterinary technician. However, for these 4 study animals, the trained cardiology veterinary technician was able to observe the recorded ECG readings collected and determined the readings at the end of the recorded time period to be incompatible with life even though they were not a true flatline. After those initial study results, the study protocol was altered to allow ECG readings to run until the machine was out of paper – equating to approximately 20 minutes of recording time. This resulted in discernable ECG flatlines for the remaining study subjects.

The amount of tissue damage to the brainstem (especially the medulla) and the cerebellum was remarkable (Figures 3 and 4). Previous published studies evaluating a pneumatic PCB with a secondary air pithing mechanism revealed minimal tissue damage from the bolt with most animals having a less than 25% damage score to any structure.[18] Relative to frontal placement, the enhanced cerebellomedullary destruction observed with poll shot placement is consistent with the closer anatomical proximity and lower quantity of intervening calvarial bone. Our data is in contrast to Robbins et al (2021) that evaluated a poll shot as a secondary step following frontal shot euthanasia.[9] In the Robbins study, gross examination of bolt trajectory indicated that poll shot placement was angled more obtusely toward the muzzle, instead of more acutely ("downward") toward the base of tongue, thereby missing a trajectory directly impacting the brainstem. Although cattle had some damage to the brainstem, most pathology was associated with the frontal shot instead of the secondary poll shot due to shot angle placement toward the muzzle instead of the base of the tongue.

Proper placement and angle are critical to achieving effective euthanasia and operators should be competent in proper PCB application. The median time from application of the PCB to clinical death of 9 minutes and subsequent ECG flatline median time of 12 minutes indicates that a properly placed PCB poll shot can achieve effective euthanasia with a single step. Whenever euthanasia is performed an adjunctive method should be available in case it is required. Since neonatal bovids tended to have prolonged cardiac activity compared to more mature animals, it may be prudent to routinely incorporate a secondary step in their euthanasia protocol.

Study limitations and suggested future research

Limitations of this study include small sample size and omission of mature bulls in the study population. However, though the enrollment size is small, the study produced consistent results for important outcome variables. Though there were no bulls enrolled, it is unlikely that their inclusion would have produced different results. In contrast to the frontal shot approach with a PCB which must account for variations in thickness and density of the frontal bone based on bovine size and gender, the poll shot method does not require penetration through the frontal bone.

Future research efforts could include validation of this technique using a firearm. Though the use of a poll shot approach for euthanasia using a firearm is not uncommon in the field, it has not yet been validated and has not been published in peer-reviewed literature.

Conclusion

Our findings demonstrate that a correctly placed poll shot using a PCB is an acceptable single-step method to achieve humane euthanasia in cattle. The poll shot approach is a method already being used by practitioners. Similar to a frontal approach, it requires adequate restraint and/or sedation. Comparable to use of a frontal approach, proper placement, cartridge selection, and bolt length is critical to produce immediate unconsciousness. The use of a poll shot technique is not expected to interfere with diagnostic testing requiring the collection of obex tissue. In contrast to the use of the frontal shot method, the use of a poll shot may be safer for the human as well as less stressful for both the animal being euthanized and the human operator. For these reasons, the poll shot method may be preferable to the frontal shot approach for euthanasia using a penetrating captive bolt. Similar to other euthanasia methods caregivers should be prepared for the use of a secondary method, whether it be reapplication of the PCB or another method, if the initial PCB shot is not immediately effective.

End notes

[a] Mortara ELI 230 Electrocardiograph, Mortara Instrument Inc., Milwaukee, WI

[b] I CASH® Special captive bolt gun, Accles & Shelvoke, Minworth, United Kingdom

[c] Microsoft. Microsoft Excel. Microsoft, 2010. Redmond, WA

Conflicts of interest

Authors do not have any conflict of interest related to this project.

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