EXTERNAL SCIENTIFIC REPORT

Review Of The Main Welfare Risks Related To Electrical Stunning Of Small Ruminants (Ovine And Caprine Species)[1]

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ABSTRACT

EFSA commissioned a comprehensive review of the welfare aspects of electrical stunning methods for small ruminants with an emphasize on low ampere stunning to establish the state of the art in the field and to assess whether scientific studies would address criteria outlined in an EFSA guidance on the assessment criteria for studies evaluating the effectiveness of stunning interventions regarding animal protection at the time of killing (EFSA Journal 2013;11(12):3486). The review was not formulated as a systematic review with a focused question instead the review followed the approach to assessing the literature described by the EFSA guidance. The key databases searched were: Science Citation Index (1900-2014), CAB Abstracts (1910-2014) and Medline (1990-2014). Key conferences proceedings and the bibliographies of review articles were manually searched. The search yielded 1599 records. 706 duplicate records were removed and 894 records assessed for relevance. Relevant studies reported electronic stunning of small ruminants and outcomes associated with onset and duration of unconsciousness. Eighteen papers reported electrical approaches to stunning in sheep. No goats were studied. None of the papers reported all of the parameters detailed in the EFSA guidance (EFSA, 2013) and a risk of bias assessment was not conducted. No studies reported the appearance of the electrodes. When the frequency (Hz) applied to the animal was reported, it was not specified whether this represented a minimum or maximum frequency. Only one study explicitly reported an effect size for amperes. The study suggested that the odds of a poor stun were higher for amperes of 0.6 (odds ratio (OD) of 6.27 with 95% confidence interval (CI) of 1.98-20.7) and 0.8 (OR of 24.4 with 95% CI of 6.98-85.2) when compared to a poor stun at 1.25 ampere.

KEY WORDS

animal welfare; electrical stunning; small ruminant slaughter

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BACKGROUND AS PROVIDED BY EFSA

The European Commission requested EFSA to prepare a scientific opinion on the use of a lower minimum current than 1 Ampere for electrical stunning of small ruminants.

In support of the evidence base of the scientific opinion concerning electrical stunning of lambs (M-2014-0041; EFSA-Q-2014-00109) EFSA wishes to carry out a systematic literature review.

The aim of this assignment is to undertake a systematic review and elucidate any studies related to head-only and head-to-body electrical stunning of small ruminants (ovine and caprine species). The systematic search shall present an overview of each of the areas listed below and shall follow the structure of a PICO (Population, Interventions, Comparators and Outcomes). The population under study are small ruminants, the interventions are electrical stunning described, comparators are different electrical stunning options as outlined in section 3.1.2.1 of the EFSA guidance (2013) and outcomes are immediate unconsciousness or absence of pain until onset of unconsciousness (according to sections 2.3.1.2., 3.2.2. and 3.2.3. of the EFSA guidance (2013)) (EFSA AHAW Panel (EFSA Panel on Animal Health and Welfare), 2013. Guidance on the assessment criteria for studies evaluating the effectiveness of stunning methods regarding animal protection at the time of killing. EFSA Journal 2013;11(12):3486, 41 pp. doi:10.2903/j.efsa.2013.3486).

OBJECTIVES AS PROVIDED BY EFSA

This assignment should cover the following area:

Overall objective:

To undertake a systematic review related to electrical stunning of small ruminants and to provide an excel sheet with the resulting reference titles and abstracts when available, of these studies in the form of an EndNoteTM Library (or format compatible with EndNoteTM) and an external consensus report detailing the search methodology and provide and interpretation of the results according to the areas listed under 1.1.

A systematic review on the effectiveness of electrical stunning of small ruminants (in particular on lowering the current for younger/smaller animals) in achieving unconsciousness, taking into account different electrical parameters including additional requirements possibly linked to the use of minimum currents lower than 1 Ampere, in particular in terms of maximum live weight and possibly of other conditions (minimum voltage, maximum frequency, time of exposure, stun-to-stick interval, etc.) and specifically considering different breeds and weight classes of lambs and kid goats.

Objective:

The objective of the specific contract resulting from the present reopening competition is as follows: A systematic literature review shall be carried out for the area listed above (section 1.1) appropriate databases including Web of Science and PubMed shall be used. The literature search protocol with appropriate review question(s) necessary to address the terms of reference of the mandate (M-2014-0041; EFSA-Q-2014-00109) and a justification for such questions based on a preliminary assessment of the available literature shall be provided with the start-up contract. The follow-up contract is

concerned with the execution of the proposed systematic review following the protocol provided in the start-up contract and provision of a final external scientific report as detailed in section 1.3.

This contract/grant was awarded by EFSA to: Annette O'Connor at the Iowa State University, Julie Glanville at the York Health Economics Consortium, University of York and Jan Sargeant at the University of Guelph.

Contract/grant title: Systematic review and provision of abstracts, when available, of studies related to electrical stunning of small ruminants (ovine and caprine species)

Contract/grant number: RC/EFSA/ALPHA/2014/03

1. Introduction

1.1. General background and rationale (from technical specifications provided by EFSA)

The European Commission requested the European Food Safety Authority (EFSA) to prepare a scientific opinion on the use of a lower minimum current than 1 Ampere for electrical stunning of small ruminants. In support of the evidence base of the scientific opinion concerning electrical stunning of lambs (M-2014-0041; EFSA-Q-2014-00109) EFSA asked for a literature review. The aim of this assignment was to undertake a review and identify any studies related to head-only and head-to-body electrical stunning of small ruminants (ovine and caprine species). The systematic search presented an overview of each of the areas listed below and followed the structure of a PICO (Population, Interventions, Comparators and Outcomes). The population under study were small ruminants, the interventions were electrical stunning described in section 3.1.2.1 of the EFSA guidance (EFSA, 2013), comparators were different electrical stunning options, and outcomes were immediate unconsciousness, absence of pain until onset of unconsciousness, and duration of unconsciousness (according to sections 3.2.1.2. 3.2.2. and 3.2.3. of the EFSA guidance (EFSA, 2013).

1.2. General objectives (from technical specifications provided by EFSA)

The overall objective of this project was to summarize the data available from studies on effectiveness of electrical stunning of small ruminants (in particular on lowering the current for younger/smaller animals) in achieving unconsciousness. Although EFSA requested that the review team to take into account different electrical parameters with particular focus on minimum currents lower than 1 Ampere, the review team was asked to include also studies that would enable comparison of outcomes for animal exposure to low amperes (< 1 A).

2. Materials and Methods

2.1. Protocol and registration

The overall aim of the review was to summarize the welfare outcomes associated with studies that reported electrical stunning of small ruminants. A protocol was developed prior to conducting the review through discussion between EFSA staff and the contractors. The protocol is not registered but is included in Appendix A. The approach to the review deviated from the steps and approach recommended EFSA guidance on systematic reviews (EFSA, 2010) for two reasons. First, the request from EFSA was for an overview of information about electrical stunning in goats rather than a standard systematic review that would adopt the specific PICO question format (population (P), the intervention (I) the comparison (C.) and the outcome (O)). That is, although EFSA was interested in the welfare outcomes associated with studies that assessed stun methods that used less than 1 A, EFSA did not want to limit the review to studies that compared <1 A to > 1 A. If this approach had been taken it would have been possible to define the review using the PICO format, and use this approach to define eligible studies, screening studies etc. Instead, EFSA requested a summary of all studies about electrical stunning in small ruminants, however because valid approaches to summaries of experimental studies require a comparison to adjust for study level effect then summation is difficult. The second reason, is that EFSA requested the review team follow the approach to reviewing the literature proposed in a prior EFSA document specific to stunning methods (EFSA, 2013) rather than the general method proposed in the systematic review guidance (EFSA, 2010). Note that hereafter, when referring to the EFSA Guidance we are referring to the 2013 document (EFSA, 2013). Although

a systematic review process was not followed, many steps associated with a systematic review were included and we used a reporting style consistent with PRISMA guidelines (Moher et al., 2009).

3. Eligibility criteria

Studies eligible for inclusion in the review used an eligible stunning method and also described metrics that measures unconsciousness in sheep and goats stunned with electrical stunning. Although Council Regulation (EC) No 1099/2099 and Annex 1 indicates that all ruminants should have 1 A for sheep and goat for either head only or head-to body electrical stunning, EFSA was interested in methods that used any amperes higher or lower than 1 A.

Further eligible studies, reported the measures of unconsciousness of interest as defined in the Section 3.2.1.2. of the EFSA guidance as listed below.

For laboratory studies

a. Induction of a generalised epileptiform activity in the brain, which can be recognised from the predominance of 8–13 Hz high-amplitude EEG activity, followed by a quiescent EEG.

   or

b. An immediate onset of a quiescent EEG

   or

c. No somatosensory, visual or auditory evoked responses or potentials in the brain immediately after the stunning

and for studies conducted in slaughter houses

d. Presence of tonic seizures after removal of the current and.

   and

e. Apnoea during tonic and clonic seizures

The review included studies that described metrics relevant to the duration of unconsciousness after stunning.

4. Information sources

A range of information sources indexing published research were searched for studies reporting on stunning methods (Table 1).

Information on on-going or recently completed trials, unpublished research, and research reported in the grey literature was identified by searching trial registers, databases indexing conference

proceedings, and specialised search engines as follows: TEKTRAN, CRIS, Conference Proceedings Citation Index – Science, Science.gov, ScienceResearch.com, Open Grey.

The following key conference web-pages from the last three years (where available) were also searched to identify additional conference abstracts: International Congress of Meat Science and Technology; International Workshop on Assessment of Animal Welfare at Farm and Group Level; OIE Global Conference on Animal Welfare; Humane Slaughter Association Centenary International Symposium.

Where possible, search results were downloaded from the information sources and imported into EndNote bibliographic management software. De-duplication was undertaken using a number of algorithms. In addition to the information sources described, the references of review papers were manually scanned by two reviewers to find any potentially relevant studies that were not captured by the electronic database searches.

5. Search

The search strategy used to identify studies indexed in CAB Abstracts (Web of Knowledge) is presented in Figure 1. The strategies used to search each information source along with the dates each database was last searched are presented in Appendix B. The strategy was comprised of two key elements:

- The population: goats and sheep (search lines 1 to 4);
- The exposure: electrical stunning methods (search lines 5 to 9). The search terms for the exposure key element did not include terminology specific to electrical water-baths, as this stunning method is not routinely used in ruminants.

After developing the search strategy for CAB Abstracts, the searches were adapted appropriately to perform efficiently in other information sources. This included consideration of database interface differences as well as adaption to different indexing languages and syntax. The results of the searches were uploaded into bibliographic management software (EndNote 7) and de-duplicated using several algorithms before uploading to DistillerSR® (Evidence Partners©, Canada, 2012), an internet-based systematic review software, for relevance screening, data extraction, and management of identified studies.

6. Study selection

There were two levels of screening. Two reviewers (ST and RD), both veterinarians with post-graduate training in epidemiology and with systematic review methodology experience, independently evaluated each citation. Any conflicts were resolved by reaching a consensus after discussion, and, when necessary, by consulting a third reviewer (AOC).

For Level 1 screening, the title and abstract (if available) of each citation was reviewed for relevance using the following criteria:

Q1. Does the title or abstract describe a study that evaluates electrical stunning in sheep or goats?

Yes—primary study (proceed to Level 2 screening)

Yes—review (exclude)

No (exclude)

Not discernible (proceed to Level 2 screening)

For Level 2 screening, the title and abstract (if available) of each citation was reviewed for relevance using the following criteria:

Q2. Does the title or abstract describe a study that assesses the efficacy of electrical stunning in inducing unconsciousness using the criteria in section 3.2.1.2 of the EFSA guidance and/or duration of unconsciousness using the criteria in 3.2.3 of the EFSA guidance in commercial sheep and / or goats? (studies assessing just not meat quality alone will be excluded)

Yes (proceed to eligibility assessment and outcome extraction)

No (exclude)

Not discernible (procure full text of citation)

Citations that were scored as 'not discernible' for question 2 were procured so that they could be assessed for relevance using the question 2 screening criteria based on the full text of the citation.

7. Data collection process

Data extraction was performed independently by two reviewers (ST and RD). Study and intervention data were extracted into forms created in DistillerSR® (Evidence Partners©, Canada, 2012). Disagreements in the data extraction were resolved by consensus and, when necessary, by consulting a third reviewer (AOC)

8. Data Items

As proposed in the protocol, studies that were considered relevant were assessed for eligibility based on report how comprehensively they reported the electrical stunning methods based on Table 3 of the EFSA guidance (EFSA, 2013). Note that differences in the use of terms used in systematic reviews (EFSA, 2010), can be confused in this aspect of the review. Commonly in systematic reviews, studies are considered eligible if they meet the relevant criteria (defined in PRISMA item 6), however, the EFSA guidance on stunning (EFSA, 2013), considers that studies are assessed for eligibility based on how comprehensively they report the stunning method (see Figure 1 of the EFSA guidance). As EFSA requested that the review team extract all the data reported, even when not comprehensive, we considered this step equivalent to data extraction, although the EFSA guidance on stunning refers to this as level 1 eligibility assessment. Based on Figure 1 of the EFSA guidance on stunning and the proposed protocol, the review team extracted details of the intervention and relevant outcomes. Subsequently, the review team only extracted details of comprehensive reporting and methodological quality for those studies that provided all the information requested in Table 3 of the EFSA guidance on stunning.

The study information data extraction form is shown in Appendix B. The intervention information data extraction form is presented in Appendix C. Outcome data were extracted directly into Excel (Microsoft®, Redmond, WA, USA, 2013). Outcome data extraction form is presented in Appendix D. For all studies that described these methods, the approach to stun was assessed for comprehensive reporting based on Table 3 in section 3.1.2.1 of the EFSA guidance

9. Risk of bias within individual studies

Based on the EFSA guidance, methodological quality was only to be extracted on studies that provided the complete set of information requested in Table 3 of the EFSA guidance (2013). The methodological information that would be extracted when such papers were identified is provided in the protocol (Appendix A.

10. Summary Measures

Consistent with the EFSA request to provide an overview of the characteristics of electrical stunning methods in small ruminants and all outcomes reported rather than to compare a specific outcome across a predefined intervention and comparison, no particular summary effect measure was of interest.

11. Synthesis of results

Consistent with the EFSA request to provide an overview of the characteristics of electrical stunning methods in small ruminants and all outcomes reported rather than to compare a specific outcome across a predefined intervention and comparison, it was not anticipated that quantitative meta-analysis would be performed. The approach to reporting therefore was to present the characteristics of the stunning methods and to indicate which aspects were not reported and to present the results of the studies.

12. Risk of bias across studies

Assessment of risk of bias across studies would require a comparative effective size; as such results were not of interest, assessment of risk of bias was not conducted.

13. Additional Analysis

No additional analyses are planned.

14. Results

15. Study selection

The searches yielded 1599 records. The source of these records is presented in Table 2. After de-duplication 706 records were removed and 894 records assessed for relevance using the Level 1 screening form. The number of records which were screened by title/abstract and by full-text, along with reasons for exclusion is presented in the PRISMA flowchart (Figure 2). Translations were not conducted for papers that were not available in English; therefore, data extraction was not performed on non-English-language papers. We included in the review, papers that could be obtained within one month after the start of the contract to enable the team to complete the remaining aspects of the review. The list of papers excluded based on full text assessment is provided in Appendix C.

16. Study characteristics

18 papers reported electrical approaches to stunning small ruminants. All 18 studies utilized sheep; no goats were studied. The type of sheep (i.e. meat, dairy, wool) was only reported by Berg et al. (2012) (meat). Bórnez et al. (2010) used only male sheep, whereas Blackmore & Newhook (1982), Lambooy (1982), Gregory & Wotton (1984), and Blackmore & Newhook, 1981) used both male and female sheep. The remaining studies did not report the sex of the sheep in their study population. The remaining characteristics of the study populations are reported in Table 3: Study characteristics of small ruminant stunning studies.

16.1. Intervention information

The information about the stunning interventions used is provided in Table 4 and in Table 5. None of the 18 extracted papers reported all of the parameters that the EFSA Guidance document (EFSA, 2013) recommends should be provided when applying head-only or head-to-body electrical stunning (section 3.1.2.1.). Namely, latency (how soon the minimum current was reached after the intervention was applied to the animal) and the appearance of the electrodes (i.e. their condition and cleanliness, including the method used to clean them between use on individual animals) were not reported for any of the 18 extracted papers. In papers where the frequency (Hz) applied to the animal was reported, none of the authors specified whether this represented a minimum or maximum frequency.

The EFSA guidance suggests that authors provide 'provide a description of the study population in relation to the wool/hair/feather cover, and cleanliness of the coat (e.g. clipped or not, breed, wet/dry head'. Dry skin and the presence of wool at the site where the stunning electrode is positioned can increase the electrical resistance and affect the efficacy of the stun (Velarde et al., 2002). The condition of the animals' skin at stunning was not reported for twelve of the papers (Anil and McKinstry, 1991); (Blackmore and Newhook, 1981); (Bornez et al., 2009); (Bornez et al., 2010); (Cook et al., 1995); (Croft and Hume, 1956); (Devine et al., 1986); (Gregory and Wotton, 1984); (Gregory and Wotton, 1985); (Gregory and Wotton, 1988); (Hoenderken et al., 1981) ; (Lambooy, 1982a). Velarde et al. (2000), who were testing the efficacy of stunning with different skin conditions, stunned lambs with a combination of clipped and unclipped wool, wet and dry skin. Berg et al. (2012) tested stunning efficacy on lambs with dry skin (wool length reported as short), whereas Velarde et al. (2000), Kuhne et al. (1979), Blackmore and Newhook (1981), and Gregory and Wotton (1984) (for head-to-back stunning, back was wet) wet the skin of the animals before applying the stunning electrodes. Frequency of calibration of the stunning equipment was reported in only two papers ((Berg et al., 2012) ; (Gregory and Wotton, 1984)), in which the equipment was calibrated before each stun.

17. Risk of bias within studies

For this project, the review team followed the EFSA guidance on assessment of stunning (EFSA, 2013) rather than the EFSA guidance of the conduct of systematic reviews (EFSA, 2010) and therefore risk of bias was only assessed on papers that reported all of the elements of the intervention requested by EFSA guidance. As no studies met that criterion, no risk of bias assessment was conducted.

18. Results of individual studies

We classified the reported outcomes into one of the following: (1) onset of unconsciousness and insensibility or (2) the duration of unconsciousness and insensibility, according to the EFSA Guidance

on the assessment criteria for studies evaluating the effectiveness of stunning methods regarding animal protection at the time of killing (EFSA, 2013).

18.1. Onset of unconsciousness

Onset of consciousness outcomes were reported in sixteen studies (Croft and Hume, 1956; Blackmore and Newhook, 1981; Hoenderken et al., 1981; Lambooy, 1982a; Gregory and Wotton, 1984, 1985, 1988; Anil and McKinstry, 1991; Cook et al., 1995; Velarde et al., 2000; Velarde et al., 2002; Bornez et al., 2009, 2010). The onset of unconsciousness outcome data are summarized in Table 6, with the exception of Lambooy (1982), which is reported here in the text rather than Table 6 because of the difference in this author's approach to reporting results compared with the other studies. Lambooy (1982) found that in an up-and-down experiment, the amperage necessary to stun 90 % of the sheep effectively was 0.33 A (95 % CI = 0.24 to 0.40 A; n = 67) with a corresponding average voltage of 98V ± 28 (SD), where an effective stun was defined by the author as generating an epileptiform insult as observed on elecctrocorticography (ECoG). This is consistent with EFSA criteria for determining unconsciousness (EFSA, 2013). In the 'up-and-down' method, a level of current is applied to a group of animals. If one animal in that group is not effectively stunned, the stunning current for the next group is increased by 0.1A compared to the previous group. If all of the animals are effectively stunned at a given current, the next group of animals receives a stunning current 0.1 A lower than the previous group (Lambooy, 1982). In the same study, for the 300 V, 3 seconds, 1.7 A treatment, the 600V 2 seconds, 4.3A treatment, and the 600 V, 3 seconds, 3.9 A treatments one sheep, three sheep and three sheep, respectively, were irreversibly stunned (died) (Lambooy, 1982).

Velarde et al. (2000) found that effectiveness of stunning was significantly higher when the stunning tongs were applied in a frontal position versus a caudal position ($p<0.05$), when the skin on the sheep's head was wet versus dry ($p<0.001$), and when wool was present on the sheep's head versus absent ($p<0.001$). Of the sixteen extracted studies that reported unconsciousness as an outcome, five did not define onset of unconsciousness using the criteria as outlined in EFSA Guidance section 3.2.1.2. Berg et al. (2012) described 'good stun quality', approximately three seconds after tongs were removed from heads of sheep and prior to neck cutting. Berg et al. (2012) defined a good stun quality as absence of all of the following: corneal reflex, eye movements (defined as both eyes co-ordinated, fixed at an object), rhythmic breathing (defined as at least two breaths), head-righting reflex and excessive kicking during the tonic phase (defined as any substantial kicking i.e. when more than a minor pull was seen during the general tonic phase. Bórnez et al. (2009) described an animal as 'correctly stunned' when 'unable to respond to normal stimuli, including pain, but have breathing not-rhythmic'. Bórnez et al. (2010) described an animal as 'correctly stunned' when an 'animal is unable to respond to painful stimuli, like a nose prick with a hypodermic needle, but are breathing, not arrhythmically'. Croft & Hume (1956) assumed unconsciousness had occurred if the animal showed signs of an electroleptic fit. Gregory & Wotton (1984a) did not describe criteria for assessing unconsciousness in the abattoirs surveyed but reported the number of number of sheep that had to be re-stunned.

One study (Berg et al., 2012) performed logistic regression analysis on their data. A summary of their model outputs is presented in Table 7. This study (Berg et al., 2012) found a dramatic increase in the odds of a poor stun as the ampere decreased. Using 1.25 A as the reference, the point estimates (95% CI) of the odds ratio increased to 1.75 (0.47-6.4), 6.27 (1.9-20.7) and 24.4 (6.98-85.2) for amperes of 1 A, 0.8 A and 0.6 A respectively. The advantage of this study is that it was perhaps the only study that

actually reported a comparison of the odds of success that enable calculation of an effect size that adjusts for study specific conditions. The comparative approach to assessing outcomes is more useful than that of studies without comparisons such as the (Hoenderken et al., 1981) which only assess one level of ampere. The absence of a comparator means is unclear how much experimental conditions influence the outcome in one level of ampere studies.

18.2. Duration of unconsciousness

Thirteen studies reported data on the duration of unconsciousness following electrical stunning (Kuhne et al., 1979; Blackmore and Newhook, 1981; Lambooy, 1982a; Gregory and Wotton, 1984, 1985; Devine et al., 1986; Gregory and Wotton, 1988; Anil and McKinstry, 1991; Cook et al., 1995; Velarde et al., 2000; Velarde et al., 2002). These data are summarized in Table 8. Additionally, Gregory & Wotton (1988) observed the time to return of a cortical response to stimulation of a tooth in the sheep's mouth. The authors apparently used this metric because tooth stimulation is one of the few painful stimuli that has almost no other sensory components. Other potentially painful stimuli may provoke a physical response for a variety of reasons besides being painful. Of the fourteen sheep examined in this way, a response to tooth stimulation (as assessed by ECoG), the time from stunning to seeing a cortical response to tooth stimulation was less than two minutes for one sheep, two to six minutes for five sheep and over 9.5 minutes for eight sheep. Note that in Table 8, we indicate if the EFSA criteria were met. Some authors used an EEG or ECoG but did not use EFSA's criteria exactly, however provided they used an EEG or EcoG to define unconsciousness these are indicated as yes in the table.

19. Synthesis of results

As only one paper provided a within study comparative effect size, (Berg et al., 2012) i.e., a comparison that adjusted within study baseline, it was not possible to conduct a comparative meta-analysis.

20. Risk of bias across studies

As only one study reported a comparative effect size it was not possible to evaluate the body of work for small study effects.

21. Additional analysis

CONCLUSIONS AND RECOMMENDATIONS

22. Summary of evidence

Numerous studies have been conducted to assess various aspects of electrical stunning efficacy in sheep, and none were identified in goats. It appears that only the Berg et al. (2012) study aimed to assess differences in stun efficacy based on different amperes. This study was missing some of the information requested by the EFSA guidance. The results of the Berg et al. (2012) study suggested that lower amperes were associated strongly with lower stunning success. For each ampere assessed below 1 A, the odds of a poor stun were statistically higher when compared to stun success at 1.25 ampere. Berg et al. (2012) used the higher ampere 1.25 as the referent and found no evidence to reject the null hypothesis that 1.25 A and 1 A had the same stun efficacy. Using 1.25 A as the reference, the point estimates (95 % CI) of the odds ratio increased to 1.75 (0.47-6.4), 6.27 (1.9-20.7) and 24.4

(6.98-85.2) for amperes of 1 A, 0.8 A and 0.6 A respectively. Unfortunately, the rationale for the sample size used was not reported and therefore it is unclear what magnitude of difference the study was designed to assess. It is not clear from the results that the stun efficacy is different between 1 and 0.8 A and 1 A and 0.6 A, because the referent was 1.25 A. However despite the wide confidence intervals, the review team would reach the pragmatic conclusion that the point estimates indicate an increase in poor stun with decreasing amperes. This conclusion is based on a consistent increase in the odds ratio, and documentation of a dose response based on point estimates with wide confidence intervals.

The results of the Berg et al. (2012) study suggested that lower amperes were associated strongly with lower stunning success. The odds of a poor stun at 0.6 amperes were estimated to be over 6 times when compared to the odds of a poor stun at 1.25 A (OR of 6.27 with 95% CI 1.98-20.7). At a current of 0.8 A the odds of a poor stun were estimated to be over 24 times higher when compared to a current of 1.25 A (OR of 24.4 with 95% CI 6.98 – 85.2).

23. Limitations

One of the major limitations of the review was the lack of comprehensive reporting of the methods of stunning. The EFSA guidance provides clear criteria for information to be reported however no studies met all these criteria. This is not surprising as the criteria are very extensive and were published after the majority of studies, so the authors would have been unaware of the standards or reporting required. In addition it has to be noted that the scientific works published were different in intention to the target studies received by EFSA for evaluation as outlined in the EFSA guidance.

Another limitation of the ability of the review to address any question about the impact of decreased amperes, was the lack of comparative studies i.e., studies with different amperes used with the same setting. Two types of studies existed with only one ampere. One type of study is a case report, where nothing is varied across a group of animals and all receive the same stun method. Such studies reported a percentage of successful stuns. Such studies are not capable of providing comparative estimates as they lack any control group, and therefore the value of the information about the impact of ampere on stunning success is low. The reason we consider such information from such studies to be low is that the observed stun efficacy is entirely confounded by all other factors associated with the stunning procedure. An example of such a study would be Blackmore and Newhook (1981).

Another type of study, that only uses one ampere level, is a study that varies a different factor holding the amperes constant. For example, the Velarde et al. (2000) study was not designed to assess the efficacy of lower amperes compared to higher amperes, but instead provides results that, conditional on a set lower ampere, what is the impact of tong position, skin condition and the presence of wool on stun efficacy. Although this study has a control group, it is not the control of interest to this review. However, the results from Velarde et al. (2000) do illustrate the huge variation in stun efficacy that can occur within studies of a single ampere, and validates the review teams opinion that case report studies with an assessment of a single method are of little value for informing decisions i.e., for extrapolation only within-study comparisons are useful.

Note that the limitations discussed should not be seen as a criticism of the authors work. The limitations relate to the application of the results to the question about the impact of lower amperes on stun efficacy. Often the authors were studying a different and valid aspect of stunning not related to

the review question. However, generally studies that are comparative and designed to assess a particular hypothesis are more useful, than case reports where all aspects of the stun approach are confounded by others.

24. Conclusions

The conclusion reached by the review team is that based on the paper by Berg et al. (2012), lower amperes are associated with lower levels of successful stunning. However, it is also clear that it is possible to have high levels of successful stunning under 1 A as shown by Velarde et al. (2000) . It is unclear why the success of stunning was so low in Berg et al. (2012), but we can only assume that it relates to other factors, perhaps some of those described by Velarde et al. (2000). Given the large number of confounders that can impact the efficacy of stunning, the review team can see the rationale of the list of factors requested by EFSA. However, we would propose that this valuable resource is perhaps not been adopted rapidly enough. It would be useful if EFSA made a greater effort to alert the community of researchers of the standards of reporting in the EFSA guidance (EFSA, 2013) and further if EFSA perhaps provided an explanation and elaboration document which outlines why the information being requested is needed. For example, the request for information about skin criteria is clearly an evidence based criteria, as the findings of Velarde et al. (2000) suggest this is important but this rationale is missing from the EFSA guidance (EFSA, 2013). It is strongly recommended that investigators intending to study the effects of electrical stunning in small ruminants consult the EFSA guidance document on the minimum reporting criteria before conducting their study. Also we would suggest that authors consult other guidelines for how to report comparative studies such as the REFLECT statement or the ARRIVE guidelines (Kilkenny et al., 2010; O'Connor et al., 2010). If EFSA does not already provide guidance on reporting of statistical results, we would suggest the SAMPL guidelines be promoted to authors (Lang and Altman, 2013).

When generating supporting data for modified or new stunning interventions, the use of live animals should be minimized as stated in Directive 2010/63/EU on the protection of animals used for scientific purposes. Potential pain, distress and suffering of animals subjected to experimental investigations must be avoided and the principles of replacement, reduction and refinement (the 3Rs) when using animals for scientific purposes should be applied. However, reduction is not a suitable rationale for conducting underpowered studies. Finally, reporting of effect sizes and sample sizes is critical for understanding the impact of interventions. P values simply convey the probability of the null hypothesis, whereas the magnitude of differences in outcomes of interest is far more informative for decision-making. Of course, knowing how precisely we understand these effect sizes is also important so clear reporting of measures of precision (SE for parameters and SD for descriptions of populations including confidence intervals in addition to probabilities) should be included in all reports.

25. Funding

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TABLES AND FIGURES

Table 1: Information sources searched to identify relevant studies

Database	Interface
Science Citation Index (SCI)	Web of Knowledge, Thompson Reuters
Conference Proceedings Citation Index – Science (CPCI-S)	Web of Knowledge, Thompson Reuters
CAB Abstracts	Web of Knowledge, Thompson Reuters
BIOSIS Citation Index	Web of Knowledge, Thompson Reuters
MEDLINE and MEDLINE In-Process	OvidSP
AGRIS	http://agris.fao.org/
AGRICOLA	http://agricola.nal.usda.gov/
TEKTRAN	www.ars.usda.gov/services/tektran.htm
CRIS	http://cris.nifa.usda.gov/
Science.gov	www.science.gov/
ScienceResearch.com	http://scienceresearch.com/
Open Grey	www.opengrey.eu/

Table 2: Number of records identified by information source

Information source	Number of records identified
Science Citation Index (SCI)	376
Conference Proceedings Citation Index – Science (CPCI-S)	25
CAB Abstracts	316
BIOSIS Citation Index	248
MEDLINE and MEDLINE In-Process	364
AGRIS	11
AGRICOLA	235
TEKTRAN	0
CRIS	0
Science.gov	0
ScienceResearch.com	20
Open Grey	4
Search of conference abstracts	0
Search of reference lists of relevant studies and reviews	2
Total	**1599**

Table 3: Study characteristics of small ruminant stunning studies

Study	Setting	Country	Sample Size	Breed	Age (weeks)	Weight (kg)
Anil & McKinstry (1991)	NR	NR	12	Mixed	NR	NR
Berg et al. (2012)	Commercial	Sweden	200 (Trial 1) 135 (Trial 2)	Texel, Crosses, Other Meat Types	8 to 12	30 (approx.)
Blackmore & Newhook (1981)	NR	NR	34	Romney, Cheviot cross	"two tooth to full mouth" and 1 week old	NR
Blackmore & Newhook (1982)	NR	NR	16	Romney, Romney cross	52 to 104 and 16 to 24	NR
Bórnez et al. (2009)	Commercial	Spain	103	Spanish Machengo	10	25
Bórnez et al. (2010)	Commercial	Spain	100	Spanish Machenga	4.3	12.80 (0.20)[b]
Cook et al. (1995)	Laboratory	NR	17	Romney Cross	52 to 104	32 to 49
Croft & Hume (1956)	Commercial	United Kingdom	31	NR	NR	NR
Devine et al. (1986)	NR	NR	35	Mixed	52	30 (approx.)
Gregory & Wotton (1984)	NR	NR	91	NR	NR	41 (15)[c]
Gregory & Wotton (1984)	Commercial	United Kingdom	10764	NR	NR	NR
Gregory & Wotton (1985)	NR	NR	22	NR	NR	40.5 (5.2)[d]
Gregory & Wotton (1988)	NR	NR	21	NR	NR	57 (15)[c]
Hoenderken et al. (1981)	NR	The Netherlands	NR	NR	NR	NR

Study	Setting	Country	Sample Size	Breed	Age (weeks)	Weight (kg)
Kuhne et al. (1979)	NR	NR	18	Merino, Karakul	2 (Merino), 0.1 to 0.28 (Karakul)	NR
Lambooy (1982)	NR	NR	67	Texel	NR	43 (8)[c]
Velarde et al. (2002)	NR	NR	24	Ripollesa	12 to 14	22.6 (0.45)[d]
Velarde et al. (2000)	NR	NR	89	Ripollesa	12 to 14	22 (18-28)[e]

(a):NR = Not reported or Not discernible
(b): Authors did not report whether the dispersion was a standard error or standard deviation
(c): Standard deviation
(d): Standard error
(e): Range

Table 4: Intervention information for small ruminant electrical stunning studies, Part 1

	Stunning Method	Current Type	Current Waveform	Minimum Current (A)	Exposed Minimum Voltage (V)	Delivered Minimum Voltage (V)	Frequency (Hz)	Minimum Time Exposure (s)
Anil et al. (1991)								
n=12 mature sheep	HTB[(a)]	NR[(b)]	NR	NR	300	NR	50	3
Berg et al. (2012)								
Trial 1, 0.6A	HO[(c)]	Sine AC	NR	0.6	Up to 230	NR	50	10.5
Trial 1, 0.8A	HO	Sine AC	NR	0.8	Up to 230	NR	50	10.5
Trial 1, 1.0A	HO	Sine AC	NR	1.0	Up to 230	NR	50	10.5
Trial 1, 1.25A	HO	Sine AC	NR	1.25	Up to 230	NR	50	10.5
Trial 2, 1.25A, 14s	HO	Sine AC	NR	1.25	Up to 230	NR	50	14
Trial 2, 1.25A, 3s	HO	Sine AC	NR	1.25	Up to 230	NR	50	3
Blackmore & Newhook (1981)								
Head-only stunned and slaughtered	HO	NR	NR	0.7 (approx.)	NR	150	50	NR
Head-only, not slaughtered	HO	NR	NR	0.7 (approx.)	NR	150	50	NR
Head-to-back, not slaughtered	HTB	NR	NR	0.7 (approx.)	NR	150	50	NR
Blackmore & Newhook (1982)								
Head-only	HO	NR	NR	0.9 (approx.)	300	NR	50	5[(d)]
Head-to-back stun	HTB	NR	NR	0.9 (approx.)	300	NR	50	5[(d)]
Bórnez et al. (2009)								
G5 electrical stun control group	HO	NR	NR	NR	110	NR	50	5

	Stunning Method	Current Type	Current Waveform	Minimum Current (A)	Exposed Minimum Voltage (V)	Delivered Minimum Voltage (V)	Frequency (Hz)	Minimum Time Exposure (s)
Bórnez et al. (2010)								
G5 electrical stun control group	HO	NR	NR	NR	110	NR	50	5
Cook et al. (1995)								
0.1s stun duration	HO	NR	NR	1.0	400	NR	50	0.1
0.2s stun duration	HO	NR	NR	1.0	400	NR	50	0.2
0.5s stun duration	HO	NR	NR	1.0	400	NR	50	0.5
2.0s stun duration	HO	NR	NR	1.0	400	NR	50	2.0
Cook et al. (1995) (continued)								
4.0s stun duration	HO	NR	NR	1.0	400	NR	50	4.0
8.0s stun duration	HO	NR	NR	1.0	400	NR	50	8.0
12.0s stun duration	HO	NR	NR	1.0	400	NR	50	12.0
20.0s stun duration	HO	NR	NR	1.0	400	NR	50	20.0
1.0s stun duration	HO	NR	NR	1.0	400	NR	50	1.0
Croft & Hume (1956)								
Head-only stunning	NR	NR	NR	0.1 to 0.34	90	78 to 82.5	NR	2, 3, 4, 5, 10, 11 and 14
Devine et al. (1986)								
Head-only stun with recovery	HO	NR	NR	1.0	400	NR	50	4
Head-only stun then throat cut	HO	NR	NR	NR	NR	NR	50	4
Head-to-back stun then throat cut	HTB	NR	NR	1.0	400	NR	50	4
Gregory & Wotton (1984b)								

	Stunning Method	Current Type	Current Waveform	Minimum Current (A)	Exposed Minimum Voltage (V)	Delivered Minimum Voltage (V)	Frequency (Hz)	Minimum Time Exposure (s)
Head-only, late stick	HO	NR	NR	1.00[(e)] (0.38)[(f)]	300	300[(e)] (0)[(f)]	50	3
Head-only, quick stick	HO	NR	NR	1.15[(e)] (0.53)[(f)]	300	300[(e)] (0)[(f)]	50	3
Head-to-back, late stick	HTB	NR	NR	1.01[(e)] (0.25)[(f)]	300 to 400	377[(e)] (39)[(f)]	50	3
Gregory & Wotton (1984a)								
Low voltage, low frequency	HO	NR	NR	NR	109[(e)](68)[(f)]	NR	50	4
Low voltage, high frequency	HO	NR	NR	NR	143[(e)](14)[(f)]	NR	1542[(e)](102)[(f)]	3
Gregory & Wotton (1985)								
Subjected to flashing light, Sheep with recognizable paroxysmal visual-evoked potentials	HO	NR	NR	0.59[(e)] (0.06)[(g)]	200	NR	50	3
Subjected to flashing light, Sheep without any recognizable paroxysmal visual-evoked potentials	HO	NR	NR	0.72[(e)] (0.15)[(g)]	200	NR	50	3
Not subjected to flashing light	HO	NR	NR	0.91[(e)] (0.41)[(g)]	200	NR	50	3
Gregory & Wotton (1988)								
Head-only stun	HO	NR	NR	0.46[(e)](0.12)[(f)]	150	150[(e)] (25)[(f)]	50	3.5
Hoenderken et al. (1981)								
Head-only stun	HO	NR	NR	0.32	NR	100	50	NR
Kuhn et al. (1979)								

	Stunning Method	Current Type	Current Waveform	Minimum Current (A)	Exposed Minimum Voltage (V)	Delivered Minimum Voltage (V)	Frequency (Hz)	Minimum Time Exposure (s)
Karakul and Merino lambs	HO	Square wave AC	Square wave	0.0975 to 0.3	110	90 to 130	50	5
Lambooy 1982								
Up-and-down method	HO	NR	NR	0.2 to 0.6	NR	98[(e)] (28)[(f)]	50	1
300V, 1s, 1.6A	HO	NR	NR	1.6[(e)] (0.6)[(f)]	300	NR	50	1
300V, 3s, 1.7A	HO	NR	NR	1.7[(e)] (0.4)[(f)]	300	NR	50	3
600V, 1s, 4.3A	HO	NR	NR	4.3[(e)] (0.6)[(f)]	600	NR	50	1
600V, 2s, 4.3A	HO	NR	NR	4.3[(e)] (0.7)[(f)]	600	NR	50	2
600V, 3s, 3.9A	HO	NR	NR	3.9[(e)] (1.6)[(f)]	600	NR	50	3
Velarde et al. (2000)								
Frontal tong position	HO	Sine AC	Sinusoidal	0.485[(e)] (0.035)[(g)]	250	NR	50	0.2
Caudal tong position	HO	Sine AC	Sinusoidal	0.343[(e)] (0.043)[(g)]	250	NR	50	0.2
Wet skin	HO	Sine AC	Sinusoidal	0.446[(e)] (0.031)[(g)]	250	NR	50	0.2
Dry skin	HO	Sine AC	Sinusoidal	0.349[(e)] (0.056)[(g)]	250	NR	50	0.2
Wool clipped	HO	Sine AC	Sinusoidal	0.433[(e)] (0.044)[(g)]	250	NR	50	0.2
Wool	HO	Sine AC	Sinusoidal	0.375[(e)] (0.044)[(g)]	250	NR	50	0.2
Velarde et al. (2002)								
Ripollesa lambs	HO	Sine AC	Sinusoidal	2.14[(e)] (0.47)[(h)]	NR	250	50	3.0

(a): HTB = Head-to-body
(b): NR = Not reported or not discernible
(c): HO = Head-only
(d): This was the maximum reported exposure time. Minimum time not reported.
(e): Mean
(f): Standard deviation
(g) Standard error
(h) Authors did not report whether this was a standard deviation or standard error

Table 5: Intervention information for small ruminant electrical stunning studies, Part 2

	Electrode Position	Electrode Type and Characteristics	Max. stun to stick interval (seconds)[a]	Prevention of Shock/Restraint Method
Anil et al. (1991)	Head-to-back	Hand-held head-to-back electrode set	NR[b]	NR
Berg et al. (2012)	Between the eye and the ear on each side of the head	Scissor-type stunning tongs (Electronic Stunning Equipment BTR 108, Freund, Germany)	11.9[c] (2.59)[d]	Lambs remained in a pen with a small group of other lambs when stunned. No additional method of restraint was used.
Blackmore & Newhook (1981)				
Head-only stunned and slaughtered	Two electrodes applied to the occipital region of the head.	NR	NR	NR
Head-only, not slaughtered		NR	NA[e]	NR
Head-to-back, not slaughtered	As above, with a third electrode placed on the back in the mid-thoracic region	NR	NA	NR
Blackmore & Newhook (1982)				

	Electrode Position	Electrode Type and Characteristics	Max. stun to stick interval (seconds)[(a)]	Prevention of Shock/Restraint Method
Head-only	two electrodes placed 6 cm apart, to the occiput	Two probe electrodes made up the head assembly	NR	The animals were placed in a slatted wooden restraining crate of similar cross-sectional dimensions to the crush conveyors used for restraining sheep and calves while being stunned in New Zealand meat works.
Head-to-back stun	As above, with a third electrode the back electrode was applied to the mid-thoracid region	A third electrode was a curved metal plate (10 X 2.5 cm) attached by a 39 cm extension to the head electrode assembly	NR	
Bórnez et al. (2009)	On both sides of the head, behind the ears	Plate electrodes	Immediate	NR
Bórnez et al. (2010)	On both sides of the head, behind the ears	Plate electrodes (Electronarcosis Panel, MAC-01, Bernard, S.L.)	Immediate	NR
Cook et al. (1995)	Across the head at an approximate	Hand-held stunner with flat button electrodes (1.5 cm diameter) 7 cm apart through	NR	Animals were restrained in sternal recumbency, in a polypropylene net restrainer using a velcro strip around the trunk with foam rubber padding

	Electrode Position	Electrode Type and Characteristics	Max. stun to stick interval (seconds)[(a)]	Prevention of Shock/Restraint Method
	midpoint between the eyes and the ears	which a saline pulse was delivered immediately prior to stunning		positioned at all body contact points to minimize animal discomfort.
Croft & Hume (1956)	Electrode 2 over the nose of the sheep and electrode 3 above the eye, between the eye and the base of the ear	Two electrodes joined by a rigid handle with an on-off switch. The applicator is designed for one hand. Electrode 2 is a curved perforated metal plate pressed against the nose of the sheep. Electrode 3 is a metal tube with serrated end. This tube contains a sponge. The distance between electrode 2 and the centre of electrode 3 is 7 inches and electrode 3 is about 1 inch in diameter. The whole apparatus is dipped in a bucket of water at intervals.	NR	NR
Devine et al. (1986)				
Head-only stun with recovery	NR	NR	NA	Sheep were restrained in a V-shaped box insulated from the floor to reduce the 50 Hz interference picked up via blood and water.
Head-only stun then throat cut	NR	NR	10 to 14	
Head-to-back stun then throat cut	Head-to-back	NR	300	
Gregory & Wotton (1984b)				

	Electrode Position	Electrode Type and Characteristics	Max. stun to stick interval (seconds)[(a)]	Prevention of Shock/Restraint Method
Head-only, late stick	Immediately rostral to the EEG electrodes	Dry electrodes	42	Each animal was placed in a hammock.
Head-only, quick stick		Dry electrodes	10	Each animal was placed in a hammock.
Head-to-back, late stick	As above, with the back electrode 38 cm distant over the vertebrae	A set of Thornton head-to-back stunning electrodes (dry)	43	Each animal was placed in a hammock.
Gregory & Wotton (1984a)				
Low voltage, low frequency	"applied to the head or neck of the animal"	In two abattoirs they used single-handed tongs and in the remaining 31 they used scissor type tongs	21[(c)] (10)[(f)]	In 36 abattoirs the sheep were stunned whilst standing in a pen. At three abattoirs the sheep were held in a restraining conveyor during stunning, and in one abattoir they were manually restrained on a cradle during stunning.
Low voltage, high frequency			21[(c)] (9)[(f)]	
Gregory & Wotton (1985)	NR	Scissor-type tongs bearing dry electrodes	NR	Each animal was placed in a hammock for the duration of the experiment.
Gregory & Wotton (1988)	NR	NR	600	NR
Hoenderken et al. (1981)	"eye to ear" position	Two tongs	NR	Restrainer was used but was not described except to say that the electrodes at the end of the restrainer were so positioned that no animal could escape the contact.
Kuhn et al. (1979)	Bilaterally	Metal tong electrodes mounted	Immediately	Each lamb was restrained either on an insulated

	Electrode Position	Electrode Type and Characteristics	Max. stun to stick interval (seconds)[(a)]	Prevention of Shock/Restraint Method
	on the temporal line midway between the lateral canthus of the eye and the base of the external ear; in some cases electrodes were placed in the external auditory meatus	in a pair of insulated spring-loaded callipers	or not at all	table or in a specially designed sling.
Lambooy 1982	On either side of the head between the eye and the ear	Scissors model stunning tongs with spiked electrodes	120	Each sheep was placed in a hammock.
Velarde et al. (2000)				
Frontal tong position	Between the eyes and ears on	Scissor-type dry stunning tongs with flat button electrodes with a diameter of 2.5 cm	NA	To minimize its discomfort, each lamb to be stunned was placed in sternal recumbency in a net restrainer. The animal's limbs were

	Electrode Position	Electrode Type and Characteristics	Max. stun to stick interval (seconds)[(a)]	Prevention of Shock/Restraint Method
	either side of the head			approximately 30 to 40 cm above the ground to ensure electrical isolation.
Caudal tong position	Behind the ears on the occipital condyle on either side of the head	Scissor-type dry stunning tongs with flat button electrodes with a diameter of 2.5 cm	NA	
Wet skin	Frontal or Caudal (see above)	Scissor-type dry stunning tongs with flat button electrodes with a diameter of 2.5 cm	NA	
Dry skin	Frontal or Caudal (see above)		NA	
Wool clipped	Frontal or Caudal (see above)		NA	
Wool	Frontal or Caudal (see above)		NA	
Velarde et al. (2002)	NA	Scissor-type dry stunning tongs (Stork, MPG, SA, Spain) with flat button electrodes 2.5 cm in diameter		Each lamb to be stunned was placed in a net restrainer in sternal recumbency to minimize discomfort. The animal's limbs were approxmately 30 to 40 cm above the ground to ensure electrical isolation.

(a): Maximum time interval (in seconds) after application of the stunning electrodes until the sheep is killed

(b): NR = Not reported or not discernible
(c): Mean
(d): The authors did not report whether this was a standard deviation or standard error
(e): NA = Not applicable (sheep was not killed)
(f): Standard deviation

Table 6: Onset of unconsciousness in studies of electrical stunning of sheep

	Sample size (n)	# Successfully stunned	% Successfully stunned	EFSA criteria met?[a]
Anil & McKinstry (1991)	**Authors defined a successful stun as an epileptiform ECoG**			
HTB[b], 300V, 3s	11[c]	11	100%	Yes
Berg et al. (2012)	**"Good stun quality" was assessed 3s after removal of tongs and defined as absence of ALL of the following: corneal reflex, eye movements (defined as both eyes co-ordinated, fixed at an object), rhythmic breathing (defined as at least two breaths), head-righting reflex and excessive kicking during the tonic phase (defined as any substantial kicking i.e. more than a minor pull) during the general tonic phase)**			
0.6A, 10.5s	50	17	34%	No[d]
0.8A, 10.5s	50	32	64%	
1.0A, 10.5s	50	43	86%	
1.25A, 10.5s	48	44	91.7%	
1.25A, 14s	58	52	89.7%	
1.25A, 3s	75	42	56%	
Blackmore & Newhook (1981)	**Unconsciousness was assessed using EEG**			
HO[e] stun then slaughtered	6	6	100%	Yes
HO not slaughtered	7	7	100%	
HTB not slaughtered	4	4	100%	
Blackmore & Newhook (1982)	**Satisfactory stun assessed using EEG**			
HO	7	6	85.7%	Yes
HTB	4	4	100%	
Bórnez et al. (2009)	**"animal is unable to respond to normal stimuli, including pain, but have breathing not-rhymic" [sic]**			
HO	20	20	100%	No
Bórnez et al. (2010)	**"animal is unable to respond to painful stimuli, like a nose prick with a hypodermic needle, but are breathing, not arrhythmically" [sic]**			
HO	20	20	100%	No
Cook et al. (1995)	**Correct stun defined as occurrence of seizure, which was considered to have occurred if post-stun EEG amplitude was at least five times greater than pre-stun amplitude**			
0.1s stun	1	0	0%	Yes
0.2s stun	NR	NR	100%	Yes
0.5s stun	NR	NR	100%	Yes
1.0s stun	NR	NR	100%	Yes
2.0s stun	NR	NR	100%	Yes

	Sample size (n)	# Successfully stunned	% Successfully stunned	EFSA criteria met?[(a)]
4.0s stun	NR	NR	100%	Yes
8.0s stun	NR	NR	100%	Yes
12.0s stun	1	1	100%	Yes
20s stun	1	1	100%	Yes
Croft & Hume (1956)	**Observation of tonic and clonic phase (presence of apnea not specified)**			
HO	31	27	87.1%	No
Gregory & Wotton (1984b)	**Correct stun defined by induction of epileptiform activity on EEG**			
HO late stick	30	30	100%	Yes
HO quick stick	30	30	100%	
HTB late stick	30	30	100%	
Gregory & Wotton (1984a)	**Criteria for assessing unconsciousness not reported. Authors only reported number of sheep that had to be re-stunned.**			
Low voltage, low frequency	6735	81[(f)] (sheep only had to be stunned once)	98.8%	No
Low voltage, high frequency	2654	28[(f)]	98.9%	
Gregory & Wotton (1985)	**Successfully stun defined as induced epileptiform activity in ECoG**			
Subjected to flashing light, easily recognizable paroxysmal visual-evoked potentials (PVP)	8	8	100%	Yes
Subjected to flashing light, no obvious PVP	4	4	100%	
Not subjected to flashing light	10	10	100%	
Gregory & Wotton (1988)	**Onset of unconsciousness defined as epileptiform phase in ECoG**			
HO	24	24	100%	Yes
Hoenderken et al. (1981)	**Effective stunning defined by induction of a generalised epileptic insult (determined by EEG) within one second through electrical current application**			
HO, 0.32A, 100V	NR	NR	90%	Yes
Lambooy (1982)	**An effective stun was defined by the author as generating an epileptiform insult as observed on ECoG. This is consistent with EFSA criteria for determining unconsciousness (EFSA, 2013).**			
Up-and-down expt.	Please refer to the text of the report for the results			Yes
Velarde et al. (2000)	**Animals were considered to be correctly stunned by EEG and by the presence of tonic clonic activity AND the absence of spontaneous breathing and absence of corneal reflex**			
Frontal tong position	42	30	71.4%	Yes

	Sample size (n)	# Successfully stunned	% Successfully stunned	EFSA criteria met?[(a)]
Caudal tong position	47	22	46.7%	Yes
Wet skin on head	44	38	86.7%	Yes
Dry skin on head	44	13	29.5%	Yes
No wool on head	45	33	73.3%	Yes
Wool present on head	42	18	43.2%	Yes
Frontal tongs, wet skin, no wool on head	10	10	100%	Yes
Frontal tongs, wet skin, wool on head	11	11	100%	Yes
Frontal tongs, dry skin, no wool on head	11	9	81%	Yes
Frontal tongs, dry skin, wool	10	0	0%	Yes
Caudal tongs, wet skin, no wool on head	12	10	83%	Yes
Caudal tongs, wet skin, wool on head	12	8	66.6%	Yes
Caudal tongs, dry skin, no wool on head	12	4	33.5%	Yes
Caudal tongs, dry skin, wool on head	11	0	0	Yes
Velarde et al. (2002)	**A successful stun was defined by the presence of tonic-clonic activity, absence of spontaneous breathing and corneal reflex and amplitude of EEG**			
HO	24	24	100%	Yes

(a): Did the authors assess unconsciousness in sheep using the criteria in section 3.2.1.2 of the EFSA Guidance (EFSA, 2013)?
(b): HTB = Head-to-back stunning
(c): There were 12 sheep in this study; however, one sheep produced a "noisy" ECoG signal that could not be interpreted, so it is not included in this table.
(d): Under slaughterhouse conditions (as was the case in this study), EFSA says there must be apnea during seizures for an electrically stunned animal to be considered unconscious. Berg et al.'s (2012) criteria included absence of rhythmic breathing, but not apnea per se.
(e): HO = head-only stun
(f): Some of these sheep fell out of their shackles between stunning and sticking and this was why they had to be re-stunned.

Table 7: Logistic regression models of the association between current and stunning time with stunning efficacy in lambs (Berg et al., 2012)

	Poor Stun[(a)]		Corneal Reflex[(b)]		Eye Movement[(c)]	
	OR (95% CI)[(d)]	**P-value**	**OR (95% CI)**	**P-value**	**OR (95% CI)**	**P-value**
Trial 1 (n=181)						
1.25A, 10.5s	1	NA	1	NA	1	NA
1.0A, 10.5s	1.75 (0.476 - 6.44)	0.40	2.52 (0.46–13.8)	0.29	1.45 (0.288 – 9.2)	0.70
0.8A, 10.5s	6.27 (1.90 - 20.7)	0.003	9.86 (2.04 – 47.6)	0.004	7.26 (1.46-36.2)	0.016
0.6A, 10.5s	24.4 (6.98 – 85.2)	< 0.001	33.6 (6.88-164)	< 0.001	6.31 (0.288 – 9.2)	0.028
Trial 2 (n=133)						
1.25A, 14s	1	NA	1	NA	1	NA
1.25A, 3s	8.06 (2.95 - 22.0)	< 0.001	(0.313 -34.1)	0.32	28.8 (3.72 – 22.3)	0.001

(a): Poor stun quality (assessed 3s after removal of stunning tongs and defined as the presence of ANY one of the following: corneal reflex, eye movements (defined as both eyes co-ordinated, fixed at an object), rhythmic breathing (defined as at least two breaths), head-righting reflex and excessive kicking during the tonic phase (defined as any substantial kicking i.e. more than a minor pull during the general tonic phase)
(b): Corneal reflex present after stunning
(c): Both eyes co-ordinated and fixed on an object
(d): 95% Confidence interval

Table 8: Duration of unconsciousness in sheep following electrical stunning

	Sample size (n)	Duration of Unconsciousness (seconds)	EFSA criteria?[a]
Anil & McKinstry (1991)			
Time from application of head-to-back stunner to end of epilepsy as assessed on ECoG	12[b]	13 ± 2.5[c]	Yes
Blackmore & Newhook (1981)			
Length of time when there were low frequency fast amplitude waves on EEG lower than 10µV and higher than 35µV (Head-only stun)	7	33 (16 to 40)[d]	No
Length of time when there were low frequency fast amplitude waves on EEG lower than 10µV and higher than 35µV (Head-to-back stun)	4	NA (all sheep died as a result of the stun)	
Blackmore & Newhook (1982)			
Length of time when there were low frequency fast amplitude waves on EEG lower than 10µV and higher than 35µV (Head-only stun)	6	35.8 (18 to 42)[d]	No
Length of time when there were low frequency fast amplitude waves on EEG lower than 10µV and higher than 35µV (Head-to-back stun)	4	NA (all sheep died as a result of the stun)	
Cook et al. (1995)			
Time from delivery of stun until the amplitude of the EEG signal was less than half that seen at the start of the seizure (0.1s stun)	1	0	No
Time from delivery of stun until the amplitude of the EEG signal was less than half that seen at the start of the seizure (0.2s stun)	NR	18 ± 0.25[e]	No
Time from delivery of stun until the amplitude of the EEG signal	NR	19.5 ± 0.5[e]	No

	Sample size (n)	Duration of Unconsciousness (seconds)	EFSA criteria?[(a)]
was less than half that seen at the start of the seizure (0.5s stun)			
Time from delivery of stun until the amplitude of the EEG signal was less than half that seen at the start of the seizure (1.0s stun)	NR	22 ± 1[(e)]	No
Time from delivery of stun until the amplitude of the EEG signal was less than half that seen at the start of the seizure (2.0s stun)	NR	25 ± 1[(e)]	No
Time from delivery of stun until the amplitude of the EEG signal was less than half that seen at the start of the seizure (4.0s stun)	NR	32 ± 2.5[(e)]	No
Time from delivery of stun until the amplitude of the EEG signal was less than half that seen at the start of the seizure (8.0s stun)	NR	27 ± 2[(e)]	No
Time from delivery of stun until the amplitude of the EEG signal was less than half that seen at the start of the seizure (12s stun)	1	32 ± 0[(e)]	No
Time from delivery of stun until the amplitude of the EEG signal was less than half that seen at the start of the seizure (20s stun)	1	22.5 ± 0[(e)]	No
Devine et al. (1986)			
Duration of Epileptic fit (s) from the time of stunning (The end of the epileptic fit was determined via EEG from the abrupt change that occurred in the peak to peak amplitude which was at least twice the prestun EEG and did not include the quiescent phase (Head-only stun with recovery)	6	46.8 ± 13.4[(c)]	No
Duration of Epileptic fit (s) from the time of stunning (The end of the epileptic fit was determined via EEG from the abrupt change that occurred in the peak to peak amplitude which was at least twice the prestun EEG and did not include the quiescent phase (Head-only stun with throat cut 10 to 14 seconds post-stun)	8	39 ± 14[(c)]	No
Duration of Epileptic fit (s) from the time of stunning (The end of the epileptic fit was determined via EEG from the abrupt	11	22.1 ± 3.8[(c)]	No

	Sample size (n)	Duration of Unconsciousness (seconds)	EFSA criteria?[a]
change that occurred in the peak to peak amplitude which was at least twice the prestun EEG and did not include the quiescent phase (Head-to-back stunning with throat cut 5 minutes after stun)			
Time when EEG amplitude drops below < 10 uV from stun (Head-only stun with recovery)	6	NR	No
Time when EEG amplitude drops below < 10 uV from stun (Head-only stun with throat cut 10 to 14 seconds post-stun)	8	53.2 ± 8.1[c]	No
Time when EEG amplitude drops below < 10 uV from stun (Head-to-back stun with throat cut 5 minutes post-stun)	11	52.0 ± 25.0[c]	No
Gregory and Wotton (1984)			
Duration of epileptiform EEG (end of this phase was defined as occurring 0.4 seconds before the amplitude first fell to a sustained level which was less than double the resting value) (Head-only, late stick)	30	50 ± 20[c]	No
Duration of epileptiform EEG (end of this phase was defined as occurring 0.4 seconds before the amplitude first fell to a sustained level which was less than double the resting value) (Head-only, quick stick)	30	21 ± 5[c]	No
Duration of epileptiform EEG (end of this phase was defined as occurring 0.4 seconds before the amplitude first fell to a sustained level which was less than double the resting value)	30	23 ± 8[c]	No
Gregory & Wotton (1985)			
Duration of epileptiform activity on ECoG (Sheep subjected to flashing light, easily recognizable paroxysmal visual-evoked potentials)	8	56 ± 6[e]	Yes
Duration of epileptiform activity on ECoG (Sheep subjected to	4	65 ± 3[e]	

	Sample size (n)	Duration of Unconsciousness (seconds)	EFSA criteria?[a]
flashing light without any obvious paroxysmal visual-evoked potentials)			
Duration of epileptiform activity on ECoG (Sheep not subjected to flashing light)	10	42 ± 7[e]	
Gregory & Wotton (1988)			
Duration of epileptiform phase in ECoG (Head-only stun)	24	39 ± 20[c]	Yes
Time from stun to return of spontaneous breathing (Head-only stun)	14	43 ± 14[c]	Yes
Time from stun to return of palpebral reflex (Head-only stun)	8	45 ± 16[c]	No
Time from stun to response to slapped snout (Head-only stun)	13	92 ± 29[c]	No
Time from stun to return of menace response (Head-only stun)	13	146 ± 47[c]	No
Time from stun to response to ear pinch (Head-only stun)	16	190 ± 38[c]	No
Hoenderken et al. (1981)			
Minimum duration of epileptic insult as assessed by EEG (Head-only stun)	NR	21	Unclear
Kuhn et al. (1979)			
Duration of unconsciousnes following stunning (defined by absence of a cardiac response on EKG to a nose-prick pain stimulus in a sheep that had a cardiac response to pain when awake	12	60 to 150[f]	No
Lambooy (1982)			
Duration of entire epileptiform insult including tonic, clonic and subsequent quiescent phase as seen on ECoG (up-and-down experiment)	67	43 ± 16[c]	Yes

	Sample size (n)	Duration of Unconsciousness (seconds)	EFSA criteria?[a]
Duration of entire epileptiform insult including tonic, clonic and subsequent quiescent phase as seen on ECoG (300V 1s 1.6A)	18	39 ± 16[c]	Yes
Duration of entire epileptiform insult including tonic, clonic and subsequent quiescent phase as seen on ECoG (300V 3s 1.7A)	10	36 ± 12[c]	Yes
Duration of entire epileptiform insult including tonic, clonic and subsequent quiescent phase as seen on ECoG (600V 1s 4.3A)	7	66 ± 17[c]	Yes
Duration of entire epileptiform insult including tonic, clonic and subsequent quiescent phase as seen on ECoG (600V 2s 4.3A)	11	39 ± 7[c]	Yes
Duration of entire epileptiform insult including tonic, clonic and subsequent quiescent phase as seen on ECoG (600V 3s 3.9A)	6	32 ± 6[c]	Yes
Velarde et al. (2000)			
Epileptic EEG duration (was considered to begin when the EEG amplitude was at least four times greater than that before the stun and considered to be over when the EEG signal amplitude was less than half that recorded at the start of the seizure) (Frontal tong position)	30	30.3 ± 3.2[e]	No
Return to spontaneous breathing (Frontal tong position)	30	20.1 ± 1.20[e]	Yes
Return of corneal reflex (Frontal tong position)	30	23.6 ± 1.0[e]	Yes
Return of sensibility to pain (Front tong position)	30	308 ± 48[e]	No
Epileptic EEG duration (was considered to be over when the EEG signal amplitude was less than half that recorded at the start of the seizure (Caudal tong position)	22	17.2 ± 3.2[e]	Yes
Return to spontaneous breathing (Caudal tong position)	22	24.8 ± 1.5[e]	Yes
Return of corneal reflex (Caudal tong position)	22	24.2 ±1.6 [e]	Yes

	Sample size (n)	Duration of Unconsciousness (seconds)	EFSA criteria?[(a)]
Return of sensibility to pain (Caudal tong position)	22	142 ± 13 [(e)]	No
Epileptic EEG duration (was considered to be over when the EEG signal amplitude was less than half that recorded at the start of the seizure (wet skin on head)	38	35.3 ± 2.6[(e)]	Yes
Return to spontaneous breathing (wet skin on head)	38	24.4 ± 1.06[(e)]	Yes
Return of corneal reflex (wet skin on head)	38	24.9 ± 1.0[(e)]	Yes
Return of sensibility to pain (wet skin on head)	38	258 ± 38[(e)]	No
Epileptic EEG duration (was considered to be over when the EEG signal amplitude was less than half that recorded at the start of the seizure (dry skin on head)	13	11.7 ± 3.0[(e)]	Yes
Return to spontaneous breathing (dry skin on head)	13	18.6 ± 1.97[(e)]	Yes
Return of corneal reflex (dry skin on head)	13	20.8 ± 1.5[(e)]	Yes
Return of sensibility to pain (dry skin on head)	13	177 ± 36[(e)]	No
Epileptic EEG duration (was considered to be over when the EEG signal amplitude was less than half that recorded at the start of the seizure (wool clipped on head)	33	29.9 ± 3.2[(e)]	Yes
Return to spontaneous breathing (wool clipped on head)	33	21.9 ± 1.2[(e)]	Yes
Return of corneal reflex (wool clipped on head)	33	22.9 ± 1.0[(e)]	Yes
Return of sensibility to pain (wool clipped on head)	33	261 ± 46[(e)]	No
Epileptic EEG duration (was considered to be over when the EEG signal amplitude was less than half that recorded at the start of the seizure (wool on head)	18	17.1 ± 3.1[(e)]	Yes
Return to spontaneous breathing (wool on head)	18	25.0 ± 1.5[(e)]	Yes

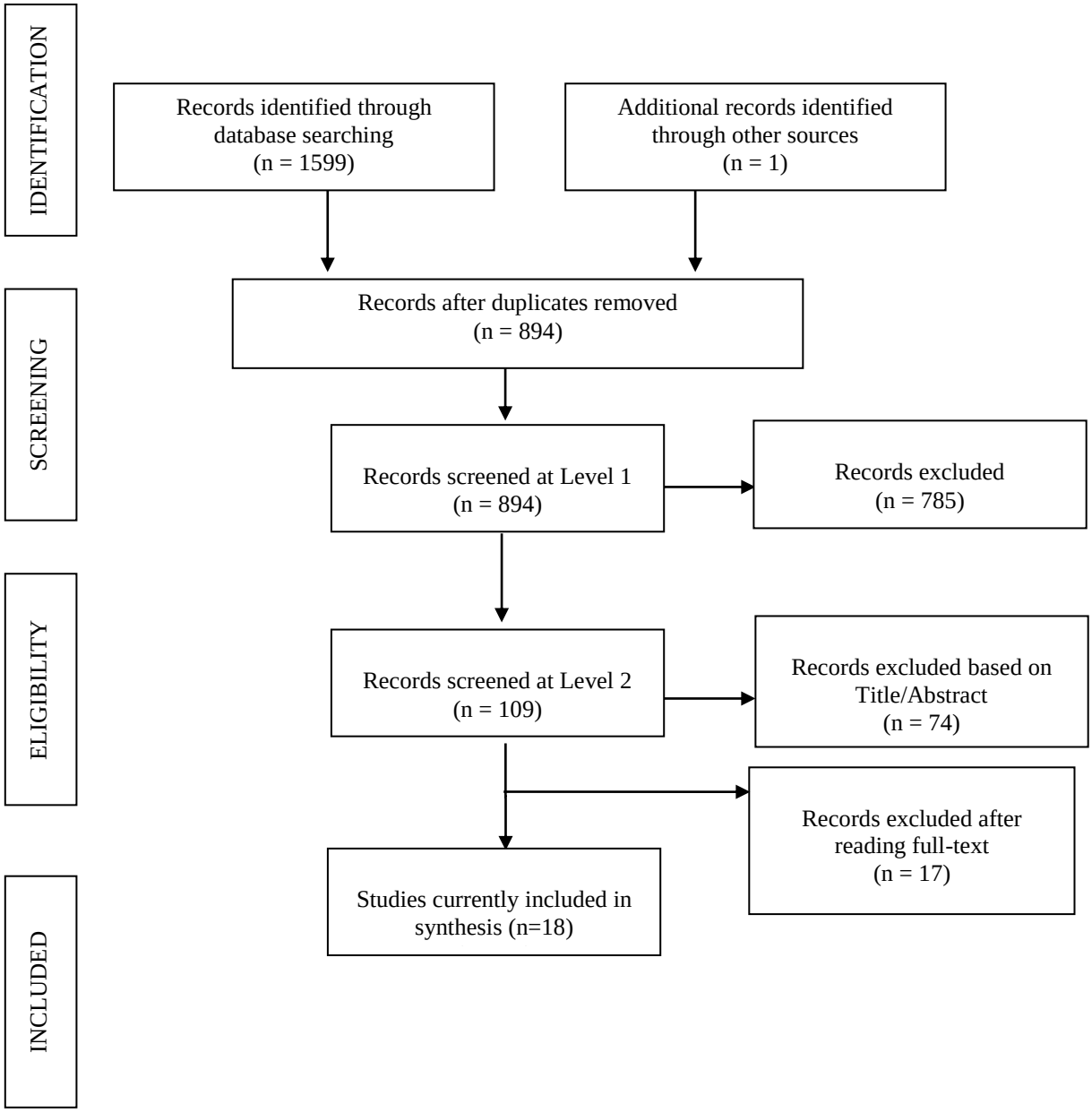
	Sample size (n)	Duration of Unconsciousness (seconds)	EFSA criteria?[(a)]
Return of corneal reflex (wool on head)	18	25.5 ± 1.7[(e)]	Yes
Return of sensibility to pain (wool on head)	18	198 ± 21[(e)]	No
Velarde et al. (2002)			
Return of spontaneous breathing	24	29.5 ± 1.55[(e)]	Yes
Return of corneal reflect	24	38.5 ± 1.75[(e)]	Yes
Return of responsiveness to pain	24	240 ± 1.34[(e)]	No

(a): Did the authors assess the duration of unconsciousness using one or more of the criteria outlined in Section 3.2.3. (EFSA, 2013)?
(b): One sheep produced a "noisy" ECoG signal, so it was not used to calculate the duration of unconsciousness
(c): Mean ± standard deviation
(d): Mean (range) calculated from raw data
(e): Mean ± SE
(f): Range

Figure 1: Search strategy to identify studies reporting on stunning of small ruminants in CAB Abstracts (Web of Knowledge, Thompson Reuters)

10 #9 AND #4
9 #8 OR #7 OR #6 OR #5
8 TS=((“stunning” OR “stun” OR “stunned” OR “stuns” OR stunner* OR restun* OR unstun* OR unconscious* OR euthan* OR "narcosis" OR "narcoses" OR insensib*) AND (electric* OR electrif* OR electro* OR voltage* OR "volts" OR "current" OR "currents" OR "wave form" OR "waveform" OR frequenc* OR “amps” OR “amperage”))
7 TS=(("head" OR "body" OR "back" OR "cardiac" OR "heart") AND ("stunning" OR "stun" OR "stunned" OR "stuns" OR stunner*))
6 TS=((electric* OR electrif* OR electro* OR stun*) AND ("wand" OR "wands" OR "tong" OR "tongs"))
5 TS=(“electronarcosis” OR “electro-narcosis” OR “electronarcoses” OR “electro-narcoses”)
4 #3 OR #2 OR #1
3 TS=("small animal*” OR “small ruminant*”)
2 TS=("sheep" OR "ovine" OR “ovis aries” OR “o aries” OR "lamb" OR "lambs" OR "ewe" OR "ewes" OR "ram" OR "rams" OR "mutton" OR "hogget*" OR wether*)
1 TS=(“goat” OR “goats” OR “capra aegagrus” OR “c aegagrus” OR “capra hircus” OR “c hircus” OR caprinae* OR caprine* OR "caprid" OR "caprids" OR “doe” OR “kid” OR “kids” OR “nanny” OR “nannies” OR “buck” OR “bucks” OR “billy” OR “billies”)

Figure 2: Flow diagram showing study identification process



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APPENDICES

Appendix A. Protocol

The overall aim of the review is to summarize the welfare outcomes associated with studies that report electrical stunning of small ruminants. For EFSAs purposes it is more consistent to use the reviewing methodology proposed by the EFSA guidance (EFSA, 2013) that to conduct the review based on the approach consistent with the EFSA "Application of Systematic Review Methodology to Food and Feed Safety Assessments to Support Decision Making" (EFSA, 2010) The rationale for this approach is that although the population and the intervention are clearly defined and enable clear identification of the relevant studies, however the comparison and outcomes are only broadly defined therefore the review question is not defined in a close-frame PICO format usually used in systematic reviews. However, consistent with the EFSA guidance of systematic reviews the aim is to ensure a comprehensive search is conducted and that the explicit data reported by studies is extracted. Such approaches are entirely consistent with transparent and comprehensive review of the literature.

Brief summary of the steps in the review

1) Conduct an extensive literature search (Extensive literature search section of the protocol)

2) Screen the literature for relevant publications (Relevant studies selection section of the protocol)

3) Assessing the relevant publications for eligibility (Eligibility assessment section of the protocol)

4) For studies that pass step 3 report all of the information extract the data sources of clinical heterogeneity and methodological quality. (Assessing heterogeneity section of the protocol)

5) Summarize (Summary section of the protocol)

Extensive Literature search

Information sources

Searches of the electronic citations databases listed in Table 1 will be conducted. In addition to searches of published literature in bibliographic databases, we will also include searches on the International Workshop on Assessment of Animal Welfare at Farm and Group Level. We will not extract data from conference papers with fewer than 500 words. The reference lists of relevant studies will also be hand-searched for additional evidence.

Table 9: Electronic citation databases to be searched for the review

Information resources	Interface
Science Citation Index (SCI)	Web of Knowledge, Thompson Reuters
Conference Proceedings Citation Index – Science (CPCI-S)	Web of Knowledge, Thompson Reuters
CAB Abstracts	Web of Knowledge, Thompson Reuters
BIOSIS Previews	Web of Knowledge, Thompson Reuters
MEDLINE and MEDLINE In-Process	OvidSP

AGRIS	http://agris.fao.org/
AGRICOLA	http://agricola.nal.usda.gov/
TEKTRAN	www.ars.usda.gov/services/tektran.htm
CRIS	http://cris.nifa.usda.gov/
Science.gov	www.science.gov/
ScienceResearch.com	http://scienceresearch.com/scienceresearch/
Open Grey	www.opengrey.eu/

Search strategy

The proposed search strategy is listed in Figure 1. This strategy will be translated to other citations bases as Figure 1. Search strategy designed for CAB Abstracts (via Web of Knowledge, Thomson Reuters). 1910 to latest update.

Proposed search strategy

10 #9 AND #4
9 #8 OR #7 OR #6 OR #5
8 TS=((“stunning” OR “stun” OR “stunned” OR “stuns” OR stunner* OR restun* OR unstun* OR unconscious* OR euthan* OR "narcosis" OR "narcoses" OR insensib*) AND (electric* OR electrif* OR electro* OR voltage* OR "volts" OR "current" OR "currents" OR "wave form" OR "waveform" OR frequenc* OR “amps” OR “amperage”))
7 TS=(("head" OR "body" OR "back" OR "cardiac" OR "heart") AND ("stunning" OR "stun" OR "stunned" OR "stuns" OR stunner*))
6 TS=((electric* OR electrif* OR electro* OR stun*) AND ("wand" OR "wands" OR "tong" OR "tongs"))
5 TS=(“electronarcosis” OR “electro-narcosis” OR “electronarcoses” OR “electro-narcoses”)
4 #3 OR #2 OR #1
3 TS=("small" NEAR/3 (ruminant* OR animal*))
2 TS=("sheep" OR "ovine" OR “ovis aries” OR “o aries” OR "lamb" OR "lambs" OR "ewe" OR "ewes" OR "ram" OR "rams" OR "mutton" OR "hogget*" OR wether*)
1 TS=(“goat” OR “goats” OR “capra aegagrus” OR “c aegagrus” OR “capra hircus” OR “c hircus” OR caprinae* OR caprine* OR "caprid" OR "caprids" OR “doe” OR “kid” OR “kids” OR “nanny” OR “nannies” OR “buck” OR “bucks” OR “billy” OR “billies”)

We have used this search in CAB Abstracts and identified 703 references, of which 32 were identified as potentially relevant primary studies by title/abstract screening. Some of these papers may be review papers, but there was insufficient information in the citation to determine if they were primary studies or reviews. Eleven of the 32 potentially relevant studies specifically evaluated unconsciousness or absence of pain as an outcome. Eight potentially relevant reviews were also identified. It is difficult to know how many additional papers are likely available but we might expect to have to screen another 1000 abstracts (based on prior experience) when the search is expanded to other databases, and identify another 2-10 relevant papers.

Search considerations

Translations will not be conducted for papers that are not available in English. We will include in the review, papers that could be obtained within 1 month after the start of the contract to enable the team

to complete the remaining aspects of the review. Relevant papers identified but not obtained within that time frame will be indicated in the final review report.

Search results and analysis

The results of the searches will be downloaded into bibliographic management software (EndNote 7) and de-duplicated using several algorithms before uploading to DistillerSR® (Evidence Partners©, Canada, 2012), an internet-based systematic review software, for relevance screening, data extraction, and management of identified studies.

Relevant Study Selection

Definition of relevant studies

Studies relevant for inclusion in the review will describe metrics that measure unconsciousness in sheep and goats stunned with electrical stunning.

When the animals used in the study are adults, the methods of head only or head-to-body stunning must be consistent with Regulation (Council Regulation (EC) No 1099/2099 and Annex 1). When the animals used in the study are younger animals (kids/ lambs) the methods of stunning should include studies that used the methods consistent with Regulation (Council Regulation (EC) No 1099/2099 and Annex 1)) studies that use a lower minimum current than 1 Ampere for electrical stunning of small ruminants.

The measures of unconsciousness of interest are those defined in the EFSA "Guidance on the assessment criteria for studies evaluating the effectiveness of stunning interventions regarding animal protection at the time of killing." (3.2.1.2. outcome electrical stunning and section 3.2.3. duration of unconsciousness) i.e.,

a. Induction of a generalised epileptiform activity in the brain, which can be recognised from the predominance of 8–13 Hz high-amplitude EEG activity, followed by a quiescent EEG. or

b. An immediate onset of a quiescent EEG or

c. No somatosensory, visual or auditory evoked responses or potentials in the brain immediately after the stunning or

d. Presence of tonic seizures after removal of the current and apnoea during tonic and clonic seizures.

e. Duration of unconsciousness

Approach to identifying relevant studies.

Forms for relevance screening will be created in DistillerSR® (Evidence Partners©, Canada, 2012). Based on the following proposed screening questions. There will be one level of screening based only on the abstract and title. 2 reviewers will assess each abstract, and conflicts will be resolved by discussion.

Q1: "Does the title/abstract describe head only or head-to body electrical stunning in sheep and goat raised for commercial use (NOT pet euthanasia or mass depopulations) using the stunning approaches defined in section 3.1.2.1 of the EFSA guidance (EFSA, 2013)

- Yes (Go to Q2)
- No- exclude

Q2. "Does the title/abstract describe the assessment of unconsciousness and insensibility or duration of unconscious as defined in section

- Yes (obtain full text)
- No-Exclude

Studies that pass the 1st level of screening will be obtained and the 2nd level of eligibility assessed.

Eligibility assessment

For each full text obtained, 2 reviewers will assess the fulfilment criteria (yes-no) based on Table 3 of (EFSA, 2013) One reviewer will extract the relevant information when reported. Outcome data will also be extracted using a template form listed in Appendix A of this document and based on the outcomes described for onset of unconsciousness and insensibility for electrical stunning methods (section .3.2.1.2 of the EFSA guidance (EFSA, 2013) and the duration of unconsciousness and insensibility in section 3.2.3 of the EFSA guidance (EFSA, 2013)

Assessing heterogeneity

Forms used:

For studies that provide all the information requested in section 3.4 of this document, we will then extract information about sources of clinical heterogeneity using the template form listed in Appendix B of this document and assess the methodological quality using the methodological quality form from EFSA "Guidance on the assessment criteria for studies evaluating the effectiveness of stunning interventions regarding animal protection at the time of killing." (EFSA, 2013).

Process of extracting data

Data extraction forms will be designed in DistillerSR®. Initial forms will be designed and piloted on several papers and modified as required for use. Two reviewers will extract data that are numerical, checkbox, radio, and list-based. Text extracted fields will be extracted by only one reviewer. Responses to numerical, checkbox, radio, and list-based will be compared between the reviewers and one review will consult the paper for evidence of minor issues. If any conflicts remain, these will be resolved through discussion. For the methodological quality, each reviewer will provide an rationale for the assessment independently and these will be discussed and a single rationale provided after discussion

Summarize

Study selection

We will use a flow chart to present the number of relevant papers screened based on the abstract and title, the number of papers assessed for eligibility and the numbers of papers with full data extraction

Relevant studies

We will provide an Excel Spread sheet with the fulfilment criteria for the interventions and the outcomes for any study considered relevant. For each study assessed we will provide tables of reporting the information in Table 3 of the EFS guidance and the outcome data extracted (Appendix a)

Eligible studies

For those studies that report all the of the eligibility criteria, we will also provide the data extracted that described potential sources of clinical heterogeneity (appendix B) and the methodological (Appendix C)

Summary effect sizes

Given that there is no specific PICO question we do not anticipate conducting a meta-analysis or summary effect size required because it is unlikely that sufficient studies will be available on a single outcome to enable such an approach. However, if several studies are available that report the same interventions and the same animal based metric with measures of variation, then we will consider if a meta-analysis should be conducted as an appropriate way of summarizing the data. We will conduct a pairwise meta-analysis – consistent with the approaches described in the EFSA guidance for systematic reviews (EFSA, 2010)

Other deliverables

For each form we will provide the data extracted in an Excel spread sheet and the SR distiller form legend corresponding to that form. For relevant publications (pass Screening level 1) will also provide a spread-sheet and EndNote library of the bibliographic information required for EFSA to uniquely identify the citation consistent with the rules governing our licence.

Outcomes extraction form

Question	Response type	Notes
Unconsciousness outcomes measured -	Checkbox	Induction of a generalised epileptiform activity in the brain, which can be recognised from the predominance of 8–13 Hz high-amplitude EEG activity, followed by a quiescent EEG. An immediate onset of a quiescent EEG. No somatosensory, visual or auditory evoked responses or potentials in the brain immediately after the stunning. Presence of tonic seizures after removal of the current and apnoea during tonic and clonic seizures. Duration of unconsciousness
Outcome definition (as described by authors)	Text box	Copied from paper
R (if proportion data is described)	Text box	Number only
N (if proportion data is described)	Text box	Number only
Mean for continuous data	Text box	Number only
Dispersion descriptor	Radio	SD SEM Not discernible or not reported
Value of dispersion	Text box	Number only
Summary effect	Text box	Number only
P value	Text box	Number only

Study level information form

Question	Style	Response
Q1. Is the full text available in English?	Radio	Yes No (end here? Are we translating non-English articles?)
Q2. Setting	Checkbox	Commercial Laboratory / Experimental Not discernible or not reported
Q3. Country	Radio	List countries
Q4.Species	Radio	Goat, Sheep
Q5. Animal type	Checkbox	Meat Dairy Not discernible or not reported
Q6: Sample size N		
Q8. Age (Weeks)	Text	
Q9. Descriptor of age	Radio	Mean Range Not discernible or not reported
Q10. Dispersion descriptor for age	Radio	SD SEM Not discernible or not reported Not applicable
Q11. Value of dispersion	Text	
Q12. Weight (kg)	Text	
Q13. Descriptor of age	Radio	Mean Range Not discernible or not reported
Q14. Dispersion descriptor for weight	Radio	SD SEM Not discernible or not reported
Q15. Value of dispersion	Text	

Q16. Sex	Checkbox	Male Females Mixed Not discernible or not reported

METHODOLOGICAL QUALITY (EFSA, 2013)

Question	Style	Responses
Q1. Information bias for the exposure — Was the extent of information bias on the exposure variable likely to be non-differential? (e.g., were different evaluations of the exposure applied to different groups?)	Radio	Yes No Unclear
Rationale	Text	
Q2: Selection bias — Was the approach to enrolment likely to be associated with differential selection probabilities for different outcome groups? (e.g., farm with indoor management systems with high prevalence of lameness were more likely to be enrolled than other groups	Radio	Yes No Unclear
Rationale	Text	
Q3. Confounding — Were known confounders identified *a priori* and controlled for, either by restriction, matching, or multivariable analysis?	Radio	Yes No Unclear
Rationale	Text	

Appendix B. Search strategies

A1. Database: Science Citation Index Expanded (Web of Science, Thomson Reuters). 1900 to present. Last updated 27/08/14. Searched 29/08/14.

10 #9 AND #4 376

9 #8 OR #7 OR #6 OR #5 7,608

8 TS=((“stunning” OR “stun” OR “stunned” OR “stuns” OR stunner* OR restun* OR prestun* OR unstun* OR unconscious* OR euthan* OR "narcosis" OR "narcoses" OR insensib*) AND (electric* OR electrif* OR electro* OR voltage* OR "volts" OR "current" OR "currents" OR "wave form" OR "waveform" OR frequenc* OR “amps” OR “amperage”)) 5,070

7 TS=(("head" OR "body" OR "back" OR "cardiac" OR "heart") AND ("stunning" OR "stun" OR "stunned" OR "stuns" OR stunner*))2,741

6 TS=((electric* OR electrif* OR electro* OR stun*) AND ("wand" OR "wands" OR "tong" OR "tongs")) 414

5 TS=(“electronarcosis” OR “electro-narcosis” OR “electronarcoses” OR “electro-narcoses”) 116

4 #3 OR #2 OR #1 246,742

3 TS=(("small" NEAR/3 animal*) or ruminant*) 39,479

2 TS=("sheep" OR "ovine" OR “ovis aries” OR “o aries” OR "lamb" OR "lambs" OR "ewe" OR "ewes" OR "ram" OR "rams" OR "mutton" OR "hogget*" OR wether*) 174,841

1 TS=(“goat” OR “goats” OR “capra aegagrus” OR “c aegagrus” OR “capra hircus” OR “c hircus” OR caprinae* OR caprine* OR "caprid" OR "caprids" OR “doe” OR “kid” OR “kids” OR “nanny” OR “nannies” OR “buck” OR “bucks” OR “billy” OR “billies”) 58,085

A2. Database: Conference Proceedings Citation Index – Science. (Web of Science, Thomson Reuters). 1990 to present. Last updated 27/08/14. Searched 29/08/14.

10 #9 AND #4 25

9 #8 OR #7 OR #6 OR #5 799

8 TS=((“stunning” OR “stun” OR “stunned” OR “stuns” OR stunner* OR restun* OR prestun* OR unstun* OR unconscious* OR euthan* OR "narcosis" OR "narcoses" OR insensib*) AND (electric* OR electrif* OR electro* OR voltage* OR "volts" OR "current" OR "currents" OR "wave form" OR "waveform" OR frequenc* OR “amps” OR “amperage”)) 514

7 TS=(("head" OR "body" OR "back" OR "cardiac" OR "heart") AND ("stunning" OR "stun" OR "stunned" OR "stuns" OR stunner*))270

6 TS=((electric* OR electrif* OR electro* OR stun*) AND ("wand" OR "wands" OR "tong" OR "tongs")) 72

5 TS=("electronarcosis" OR "electro-narcosis" OR "electronarcoses" OR "electro-narcoses") 3

4 #3 OR #2 OR #1 34,200

3 TS=(("small" NEAR/3 animal*) or ruminant*) 5,173

2 TS=("sheep" OR "ovine" OR "ovis aries" OR "o aries" OR "lamb" OR "lambs" OR "ewe" OR "ewes" OR "ram" OR "rams" OR "mutton" OR "hogget*" OR wether*) 18,855

1 TS=("goat" OR "goats" OR "capra aegagrus" OR "c aegagrus" OR "capra hircus" OR "c hircus" OR caprinae* OR caprine* OR "caprid" OR "caprids" OR "doe" OR "kid" OR "kids" OR "nanny" OR "nannies" OR "buck" OR "bucks" OR "billy" OR "billies") 12,040

A3. Database: Biosis Citation Index (Web of Knowledge, Thomson Reuters). 1969 to 2014. Last updated 22/08/14. Searched 29/08/14.

10 #9 AND #4 248

9 #8 OR #7 OR #6 OR #5 799

8 TS=(("stunning" OR "stun" OR "stunned" OR "stuns" OR stunner* OR restun* OR prestun* OR unstun* OR unconscious* OR euthan* OR "narcosis" OR "narcoses" OR insensib*) AND (electric* OR electrif* OR electro* OR voltage* OR "volts" OR "current" OR "currents" OR "wave form" OR "waveform" OR frequenc* OR "amps" OR "amperage")) 4,324

7 TS=(("head" OR "body" OR "back" OR "cardiac" OR "heart") AND ("stunning" OR "stun" OR "stunned" OR "stuns" OR stunner*))3,360

6 TS=((electric* OR electrif* OR electro* OR stun*) AND ("wand" OR "wands" OR "tong" OR "tongs")) 203

5 TS=("electronarcosis" OR "electro-narcosis" OR "electronarcoses" OR "electro-narcoses") 41

4 #3 OR #2 OR #1 252,551

3 TS=(("small" NEAR/3 animal*) or ruminant*) 36,049

2 TS=("sheep" OR "ovine" OR "ovis aries" OR "o aries" OR "lamb" OR "lambs" OR "ewe" OR "ewes" OR "ram" OR "rams" OR "mutton" OR "hogget*" OR wether*) 184,042

1 TS=(“goat” OR “goats” OR “capra aegagrus” OR “c aegagrus” OR “capra hircus” OR “c hircus” OR caprinae* OR caprine* OR "caprid" OR "caprids" OR “doe” OR “kid” OR “kids” OR “nanny” OR “nannies” OR “buck” OR “bucks” OR “billy” OR “billies”) 57,181

A4. Database: Ovid MEDLINE(R) In-Process & Other Non-Indexed Citations and Ovid MEDLINE(R) <1946 to Present> Searched 29/08/14

1 ruminants/ or exp goats/ or exp sheep/ (124629)

2 (goat or goats or capra aegagrus or c aegagrus or capra hircus or c hircus or caprinae* or caprine* or caprid or caprids or doe or kid or kids or nanny or nannies or buck or bucks or billy or billies).ti,ab. (33225)

3 (sheep or ovine or ovis aries or o aries or lamb or lambs or ewe or ewes or ram or rams or mutton or hogget* or wether*).ti,ab. (106985)

4 ((small adj3 animal*) or ruminant*).ti,ab. (25603)

5 or/1-4 (187451)

6 Electronarcosis/ or Electroshock/ or Electricity/ (23616)

7 (electronarcosis or electro-narcosis or electronarcoses or electro-narcoses).ti,ab. (213)

8 ((electric* or electrif* or electro* or stun*) and (wand or wands or tong or tongs)).ti,ab. (103)

9 ((head or body or back or cardiac or heart) and (stunning or stun or stunned or stuns or stunner*)).ti,ab. (1706)

10 ((stunning or stun or stunned or stuns or stunner* or restun* or prestun* or unstun* or unconscious* or euthan* or narcosis or narcoses or insensib*) and (electric* or electrif* or electro* or voltage* or volts or current or currents or wave form or waveform or frequenc* or amps or amperage)).ti,ab. (4886)

11 or/6-10 (29711)

12 5 and 11 (374)

13 humans/ not animals/ (12214979)

14 12 not 13 (368)

15 remove duplicates from 14 (364)

A5. Database: CAB Abstracts (Web of Knowledge, Thomson Reuters). 1910 to 2014. Last updated 28/08/14. Searched 01/09/14.

10 #9 AND #4 316

9 #8 OR #7 OR #6 OR #5 2,568

8 TS=((“stunning” OR “stun” OR “stunned” OR “stuns” OR stunner* OR restun* OR prestun* OR unstun* OR unconscious* OR euthan* OR "narcosis" OR "narcoses" OR insensib*) AND (electric* OR electrif* OR electro* OR voltage* OR "volts" OR "current" OR "currents" OR "wave form" OR "waveform" OR frequenc* OR “amps” OR “amperage”)) 2,371

7 TS=(("head" OR "body" OR "back" OR "cardiac" OR "heart") AND ("stunning" OR "stun" OR "stunned" OR "stuns" OR stunner*))486

6 TS=((electric* OR electrif* OR electro* OR stun*) AND ("wand" OR "wands" OR "tong" OR "tongs")) 77

5 TS=(“electronarcosis” OR “electro-narcosis” OR “electronarcoses” OR “electro-narcoses”) 666

4 #3 OR #2 OR #1 316,361

3 TS=("small animal*" OR "small ruminant*") 16,505

2 TS=("sheep" OR "ovine" OR “ovis aries” OR “o aries” OR "lamb" OR "lambs" OR "ewe" OR "ewes" OR "ram" OR "rams" OR "mutton" OR "hogget*" OR wether*) 238,847

1 TS=(“goat” OR “goats” OR “capra aegagrus” OR “c aegagrus” OR “capra hircus” OR “c hircus” OR caprinae* OR caprine* OR "caprid" OR "caprids" OR “doe” OR “kid” OR “kids” OR “nanny” OR “nannies” OR “buck” OR “bucks” OR “billy” OR “billies”) 100,309

A6. Database: National Agriculture Library Catalog [AGRICOLA] 1970-Current http://agricola.nal.usda.gov/ Searched 02/09/14

Limited number of search characters allowed – a series of smaller searches undertaken

Advanced: Article Citation Database

(goat? OR "capra aegagrus" OR "c aegagrus" OR "capra hircus" OR "c hircus" OR caprinae? or caprine? OR caprid? OR doe OR kid OR kids OR nanny OR nannies OR buck OR bucks OR billy OR billies OR sheep OR ovine OR "ovis aries" OR "o aries" OR lamb OR lambs OR ewe OR ewes OR ram OR rams OR mutton OR hogget? OR wether?) AND (electronarcos? or "electro narcos?") 4 results

(goat? OR "capra aegagrus" OR "c aegagrus" OR "capra hircus" OR "c hircus" OR caprinae? or caprine? OR caprid? OR doe OR kid OR kids OR nanny OR nannies OR buck OR bucks OR billy OR billies OR sheep OR ovine OR "ovis aries" OR "o aries" OR lamb OR lambs OR ewe OR ewes OR ram OR rams OR mutton OR hogget? OR wether?) AND (electric? OR electrif? OR electro? OR stun?) AND (wand OR wands OR tong OR tongs) 15 results

(goat? OR "capra aegagrus" OR "c aegagrus" OR "capra hircus" OR "c hircus" OR caprinae? or caprine? OR caprid? OR doe OR kid OR kids OR nanny OR nannies OR buck OR bucks OR billy OR billies OR sheep OR ovine OR aries OR lamb OR lambs OR ewe OR ewes OR ram OR rams OR mutton OR hogget? OR wether?) AND (head OR body OR back OR cardiac OR heart) AND stun? 15 results

(sheep OR ovine OR aries OR lamb OR lambs OR ewe OR ewes OR ram OR rams OR mutton OR hogget? OR wether?)AND(stunning OR stun OR stunned OR stuns OR stunner? OR restun? OR prestun? OR unstun? OR unconscious? OR euthan? OR narcosis OR narcoses OR insensib?)AND(electric? OR electrif? OR electro? OR voltage? OR volts OR current? OR wave? OR frequenc? OR amp OR amps OR amperage) 156 results

(goat? OR "capra aegagrus"OR"c aegagrus"OR"capra hircus"OR"c hircus"OR caprinae? or caprine? OR caprid? OR doe OR kid OR kids OR nanny OR nannies OR buck OR bucks OR billy OR billies)AND(stunn? OR stun OR stuns OR restun? OR prestun? OR unstun? OR unconscious? OR euthan? OR narcosis OR narcoses OR insensib?)AND(electr? OR volt? OR current? OR wave? OR frequenc? OR amp?) 42 results

Advanced: Books Catalog

(goat? OR "capra aegagrus" OR "c aegagrus" OR "capra hircus" OR "c hircus" OR caprinae? or caprine? OR caprid? OR doe OR kid OR kids OR nanny OR nannies OR buck OR bucks OR billy OR billies OR sheep OR ovine OR "ovis aries" OR "o aries" OR lamb OR lambs OR ewe OR ewes OR ram OR rams OR mutton OR hogget? OR wether?) AND (electronarcos? or "electro narcos?") 0 results

(goat? OR "capra aegagrus" OR "c aegagrus" OR "capra hircus" OR "c hircus" OR caprinae? or caprine? OR caprid? OR doe OR kid OR kids OR nanny OR nannies OR buck OR bucks OR billy OR billies OR sheep OR ovine OR "ovis aries" OR "o aries" OR lamb OR lambs OR ewe OR ewes OR ram OR rams OR mutton OR hogget? OR wether?) AND (electric? OR electrif? OR electro? OR stun?) AND (wand OR wands OR tong OR tongs) 0 results

(goat? OR "capra aegagrus" OR "c aegagrus" OR "capra hircus" OR "c hircus" OR caprinae? or caprine? OR caprid? OR doe OR kid OR kids OR nanny OR nannies OR buck OR bucks OR billy OR billies OR sheep OR ovine OR aries OR lamb OR lambs OR ewe OR ewes OR ram OR rams OR mutton OR hogget? OR wether?) AND (head OR body OR back OR cardiac OR heart) AND stun? 0 results

(sheep OR ovine OR aries OR lamb OR lambs OR ewe OR ewes OR ram OR rams OR mutton OR hogget? OR wether?)AND(stunning OR stun OR stunned OR stuns OR stunner? OR restun? OR prestun? OR unstun? OR unconscious? OR euthan? OR narcosis OR narcoses OR insensib?)AND(electric? OR electrif? OR electro? OR voltage? OR volts OR current? OR wave? OR frequenc? OR amp OR amps OR amperage) 3 results

(goat? OR "capra aegagrus"OR"c aegagrus"OR"capra hircus"OR"c hircus"OR caprinae? or caprine? OR caprid? OR doe OR kid OR kids OR nanny OR nannies OR buck OR bucks OR billy OR billies)AND(stunn? OR stun OR stuns OR restun? OR prestun? OR unstun? OR unconscious? OR

euthan? OR narcosis OR narcoses OR insensib?)AND(electr? OR volt? OR current? OR wave? OR frequenc? OR amp?) 0 results

A7. Database: International Information System for the Agricultural Sciences and Technology [AGRIS] 1975 to date http://agris.fao.org/ Searched 02/09/14

Export options not working – records added to EndNote manually. Duplicate records and obviously irrelevant records not added.

(goat* "capra aegagrus" "c aegagrus" "capra hircus" "c hircus" caprinae* caprine* caprid* doe kid kids nanny nannies buck bucks billy billies sheep ovine "ovis aries" "o aries" lamb lambs ewe ewes ram rams mutton hogget* wether*) AND (electronarcos* "electro narcos*") 15 results – 4 unique records added to EndNote

(goat* "capra aegagrus" "c aegagrus" "capra hircus" "c hircus" caprinae* caprine* caprid* doe kid kids nanny nannies buck bucks billy billies sheep ovine "ovis aries" "o aries" lamb lambs ewe ewes ram rams mutton hogget* wether*) AND (electric* electrif* electro* stun*) AND (wand wands tong tongs) 4 results – 2 duplicates, 2 obviously irrelevant. No records added to EndNote.

(goat* "capra aegagrus" "c aegagrus" "capra hircus" "c hircus" caprinae* caprine* caprid* doe kid kids nanny nannies buck bucks billy billies sheep ovine "ovis aries" "o aries" lamb lambs ewe ewes ram rams mutton hogget* wether*) AND (head body back cardiac heart) AND stun* 18 results - 9 duplicates, 8 obviously irrelevant. 1 record added to EndNote.

(sheep ovine aries lamb lambs ewe ewes ram rams mutton hogget* wether* goat* "capra aegagrus" "c aegagrus" "capra hircus" "c hircus" caprinae* caprine* caprid* doe kid kids nanny nannies buck bucks billy billies) AND (stunning stun stunned stuns stunner* restun* prestun* unstun* unconscious* euthan* narcosis narcoses insensib*) AND (electric* electrif* electro* voltage* volts current* wave* frequenc* amp amps amperage) 94 results. 82 duplicates, 6 obviously irrelevant. 6 records added to EndNote.

A8. Database: TEKTRAN: The ARS Manuscripts Database http://www.ars.usda.gov/services/tektran.htm Searched 02/09/14

Browse: Measure & Evaluate Animal Well-Being, Animal Behavior

Search: stun, prestun, restun, unstun, slaughter (appears to automatically truncate terms)

Records manually scanned; 0 potentially relevant records identified and added to EndNote

A9. Database: National Institute of Food and Agriculture Current Research Information System [CRIS] http://cris.nifa.usda.gov/ Searched 02/09/14

CRIS Assisted Search (automatic truncation)

Fulltext Terms: stun; prestun; restun; unstun; slaughter

AND

Fulltext Terms: goat; sheep

Not these: Fulltext Terms: Stunt

Records manually scanned; 0 potentially relevant records identified and added to EndNote

A10. Database: Open Grey http://www.opengrey.eu/ Searched 03/09/14

(stun OR stunning OR stunned OR stuns OR stunner* OR prestun* OR restun* OR unstun* OR electronarco* OR electro-narco*) AND (goat* OR "capra aegagrus" OR "c aegagrus" OR "capra hircus" OR "c hircus" OR caprinae* OR caprine* OR caprid* OR doe OR kid OR kids OR nanny OR nannies OR buck OR bucks OR billy OR billies OR sheep OR ovine OR "ovis aries" OR "o aries" OR lamb OR lambs OR ewe OR ewes OR ram OR rams OR mutton OR hogget* OR wether*) 1 result-

(stun OR stunning OR stunned OR stuns OR stunner* OR prestun* OR restun* OR unstun* OR tong* OR wand*) AND (electric* OR electrif* OR electro*) 123 results – manually scanned, only potentially relevant/non duplicate records added to EndNote. 4 new records added.

A11. Database: Science.gov http://www.science.gov/ Searched 03/09/14

(stun OR stunning OR stunned OR stuns OR stunner* OR prestun* OR restun* OR unstun* OR electronarco* OR electro-narco*) AND (goat* OR "capra aegagrus" OR "c aegagrus" OR "capra hircus" OR "c hircus" OR caprinae* OR caprine* OR caprid* OR doe OR kid OR kids OR nanny OR nannies OR buck OR bucks OR billy OR billies OR sheep OR ovine OR "ovis aries" OR "o aries" OR lamb OR lambs OR ewe OR ewes OR ram OR rams OR mutton OR hogget* OR wether*)

As not all collections seem to support Boolean/truncation/phrase searching – simple searches undertaken to try and capture any that may be otherwise missed.

sheep stun* electr*

lamb* stun* electr*

goat stun* electr*

Search full record: Science.gov websites, Biology and Nature, General Science. Agriculture and Food not searched as AGRICOLA and TEKTRAN searched separately.

Results scanned in databases – any potentially relevant records already identified by previous database searches. No records added to EndNote.

A12. Database: Scienceresearch.com http://scienceresearch.com/ Searched 03/09/14

Full Text: (stun OR stunning OR stunned OR stuns OR stunner* OR prestun* OR restun* OR unstun*) AND (sheep OR lamb* OR goat*) AND (electric* OR electrif* OR electro*)

In Biology and Nature and Agriculture

Results scanned in databases – any potentially relevant records already identified by previous database searches. 20 records added to EndNote.

Conference searches:

International Congress of Meat Science and Technology 2013, August 18-23 Izmir Turkey http://www.sciencedirect.com/science/journal/03091740/95 Searched 03/09/14

Proceedings available as a journal supplement; presentations manually scanned. 0 abstracts added to EndNote.

International Congress of Meat Science and Technology 2012, August 12-17 Montreal, Canada http://www.sciencedirect.com/science/journal/03091740/92/3 Searched 03/09/14

Proceedings available as a journal supplement; presentations manually scanned. 0 abstracts added to EndNote.

International Congress of Meat Science and Technology 2011, August 7-12 Ghent, Belguim http://www.sciencedirect.com/science/journal/03091740/89/3 Searched 03/09/14

Proceedings available as a journal supplement; presentations manually scanned. 0 abstracts added to EndNote.

International Congress of Meat Science and Technology 2010, August 15-10 Jeju, Korea http://www.sciencedirect.com/science/journal/03091740/86/1 Searched 03/09/14

Proceedings available as a journal supplement; presentations manually scanned. 0 abstracts added to EndNote.

International Conference on Assessment of Animal Welfare at Farm and Group Level, 2011, August 8-1 Guelph, Ontario http://www.uoguelph.ca/csaw/wafl/documents/WAFLproceedingsweb.pdf Searched 03/09/14

Proceedings available online; presentations manually scanned. 0 abstracts added to EndNote.

Conference was not held in 2010, 2012 or 2013 (takes place every 3 years) so proceedings from these years could not be searched. 2014 conference not due to take place until late September 2014.

Humane Slaughter Association Centenary International Symposium. Recent Advances in the Welfare of Livestock at Slaughter. 30 June-1 July 2011 Portsmouth, UK. http://www.hsa.org.uk/symposium%202011.html Searched 03/09/14

Proceedings available online; presentations manually scanned. 0 abstracts added to EndNote.

This was a one-off event, proceedings from 2010 and 2012 not available to search. Next Symposium 2015.

OIE Global Conference on Animal Welfare. 6-8 November 2012 Kuala Lumpur, Malaysia http://www.oie.int/eng/AW2012/presentations.htm Searched 03/09/14

Proceedings available online; presentations manually scanned. 0 abstracts added to EndNote.

Conference was not held in 2010, 2011, 2013 or 2014 so proceedings from these years could not be searched.

Appendix C. Studies excluded at full text assessment

Reference	Reason for exclusion
(Sanchez-Barrera et al., 2014)	Evaluated EEG in sheep after electrical and captive bolt stunning. Authors looked at the EEG profiles of lambs—they did not assess onset or duration of unconsciousness, only the characteristics of the EEG traces after stunning. Not relevant, since they aren't assessing efficacy of stunning.
(Grandin, 1998)	The authors report efficacy of stunning at US slaughter plants; however, data is reported by slaughterhouse. Sample of sizes of animals and outcomes per individual animal are not reported, nor are the specific voltages and currents used for stunning at each slaughterhouse reported. There is NO EXTRACTABLE DATA here.
(Gregory, 2001)	Not relevant. The authors looked only at current profiles (current vs time) and amount of back hemorrhage in lambs at stunning. They did not assess onset or duration of unconsciousness following stunning.
(Blackmore et al., 1979)	This is a letter to the editor-type article, talking about research that is "in preparation"
(Rao, 2014)	This is a letter to the editor, not an original research article.
(Gregory et al., 1983)	Unclear if the authors actually stunned the sheep electrically or induced cardiac fibrillation by application of direct shock to the heart (no intervention data are given)—nothing to extract and not sure if it's relevant. This paper is just one paragraph long.)
(Heal, 1999)	This is a description of a product (a monitor) to help make stunning more effective for cattle, sheep and goats. It is not a primary research study.
(Blackmore and Newhook, 1983)	This is a review paper.
(Leach, 1978)	No extractable data (no sample size of sheep given in the one instance where unconsciousness data were reported).
(Mickwitz et al., 1989)	This is a decree, not primary research. No extractable data.
(Vonmickwitz et al., 1989)	German
(Lambooy, 1982b)	Dutch
(Ween, 1972)	Norwegian
(Paleari et al., 1993)	Italian
(Lambooy et al., 1983)	German
(Mickwitz et al., 1989)	German

(Kallweit et al., 1989)	German