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An independent animal welfare assessment of mass destruction methods for pigs on-farm

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Acknowledgement of Country

We acknowledge the Traditional Custodians of Australia and their continuing connection to land and sea, waters, environment and community. We pay our respects to the Traditional Custodians of the lands we live and work on, their culture, and their Elders past and present.

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Introduction

The purpose of this review was to provide a critical assessment of the methods available for the destruction of pigs, including their suitability in situations that call for mass depopulation. This review has been prepared for the Department of Agriculture, Water and the Environment. The review also aims to provide a scientific opinion on methods that are suitable for inclusion in Australian Standards and Guidelines.

1 Project objectives

The project was separated into two parts.

Part 1 of the project delivered the following objectives:

- Identification and critical review of the methods recommended for the destruction of pigs documented in national and international standards and guidelines
- Critical review of the methods specifically outlined in the statement of requirement
- Scientific opinion on suitable methods for inclusion in Australian standards and guidelines for the humane destruction of pigs.

Part 2 of the project developed the findings of Part 1 to:

- Specify the methods that are suitable for large-scale (mass) depopulation, taking into account aspects of commercial production systems including housing design, availability of personnel, stage of production and the number of animals involved
- Provide evidence-based information on the use of the recommended methods, to ensure that the correct parameters could be further developed to enable an optimum animal welfare outcome. The development of detailed methodologies for the application of the recommended methods for large-scale depopulation was not completed as part of this project, however, this can be undertaken as a separate step incorporating the recommendations from this review.

2 Methodology

Pigs at different stages of production may have to be killed on-farm for reasons other than slaughter for human consumption. This may include both individual animals (for example, an injured or non-viable pig) or large scale (mass) destruction (for example, for disease control). Mass destruction can involve pigs from one or more production sites and is usually in response to an emergency situation. References to 'humane destruction' within this document are usually in relation to methods of killing undertaken during the production phase (on the farm) rather than in a commercial processing environment.

2.1 Project statement of requirement

The search framework followed established and recognised steps for a systematic literature review. The project statement of requirement referenced several methods used for the destruction of pigs that were to be included in the review. They were:

- Carbon dioxide gas for all classes of pig
- Inert gases (nitrogen and argon) for all classes of pig
- Foam for pigs less than 30kg. Consider both water-based air-filled foam and dry foam filled with gas (carbon dioxide or nitrogen)
- Electrocution for all classes of pigs (both single-step head-to-body and two-step in which head-only stunning is followed by current delivered to the heart)
- Non-penetrative captive bolt for neonatal pigs
- Sodium nitrite administered orally to all classes of pig
- Penetrative captive bolt - whether a follow up procedure is required.

There was also a requirement for information related to methods used for the destruction of pigs to be extracted (and added to the review) from several reference standards and guidelines (Section 2.2).

2.2 Selected standards and guidelines

A selection of national and international Standards and Guidelines were accessed. Table 1 shows the documents selected and the reason for inclusion.

Table 1 Identified Standards and Guidelines documents and reasons for inclusion

Document	Reason for inclusion
AUSVETPLAN Operational Manual: Destruction of animals	Objective in the Customer Statement of Requirement
Primary Industries Standing Committee Model Code of Practice for the Welfare of Animals: Pigs, 3rd edition	Objective in the Customer Statement of Requirement
OIE Killing of Animals for Disease Control Purposes	Specific chapter within the Terrestrial Animal Health Code. Provides a global perspective of suitable methods for the humane destruction of pigs on-farm (both individual animals and large-scale depopulation). Includes conditions of use and acceptable parameters.

Document	Reason for inclusion
EFSA Welfare of pigs during killing for purposes other than slaughter, 2020	Includes information from other significant national and international Standards and Guidelines, such as AVMA Guidelines for the Depopulation of Animals: 2019 Edition, DEFRA, OIE.

The Australian Veterinary Emergency Plan (AUSVETPLAN) (administered by Animal Health Australia) outlines the nationally agreed approach to managing animals during emergency disease responses. It includes guidance on acceptable methods of destruction (Animal Health Australia 2015). The AUSVETPLAN Operational manual: Destruction of animals refers to the Primary Industries Standing Committee Model Code of Practice for the Welfare of Animals: Pigs, 3rd edition (MCOP) (Primary Industries Standing Committee 2008). The AUSVETPLAN Operational manual: Destruction of animals currently recognises the following methods (Animal Health Australia 2015):

- Carbon dioxide (CO_2) for pigs up to 5kg liveweight
- Barbiturate overdose delivered IV, IC or IP - <25kg, IV - >25kg
- Firearms (free projectile) for pigs >25kg
- Penetrative captive bolt for >5kg
- Manual blunt force trauma for neonates
- Electrical stunning (followed by a terminal method)
- Electrocution, although other methods preferred.

With the exception of CO_2 (and under some conditions, the use of firearms) the methods recognised for the mass depopulation of pigs in Australia all require animals to be handled and individually restrained. Table 2 provides a summary of the methods used for the destruction of pigs as detailed in the referenced source documents. The colour key represents the positions presented within the documents themselves.

Table 2 Recommended methods for the destruction of pigs in identified Standards and Guidelines documents

Method	AUSVETPLAN Operational manual: Destruction of animals	Primary Industries Standing Committee Model Code of Practice for the Welfare of Animals: Pigs, 3rd edition	OIE Killing of Animals for Disease Control Purposes	EFSA Welfare of pigs during killing for purposes other than slaughter, 2020
Carbon dioxide	Recognised method (under certain conditions), for one or more classes of pig	Recognised method (under certain conditions), for one or more classes of pig	Recognised method (under certain conditions), for one or more classes of pig	Recognised method (under certain conditions), for one or more classes of pig
Nitrogen or inert gas	Not referenced	Not referenced	Recognised method (under certain conditions), for one or more classes of pig	Recognised method (under certain conditions), for one or more classes of pig

Method	AUSVETPLAN Operational manual: Destruction of animals	Primary Industries Standing Committee Model Code of Practice for the Welfare of Animals: Pigs, 3rd edition	OIE Killing of Animals for Disease Control Purposes	EFSA Welfare of pigs during killing for purposes other than slaughter, 2020
Gas mixture (inert gas + CO_2)	Not referenced	Not referenced	Recognised method (under certain conditions), for one or more classes of pig	Recognised method (under certain conditions), for one or more classes of pig
Foam (gas- or air-filled)	Not referenced	Not referenced	Recognised method (under certain conditions), for one or more classes of pig	Use is questionable or needs further research, other methods preferred
Injectable agent	Use is questionable or needs further research, other methods preferred	Recognised method (under certain conditions), for one or more classes of pig	Recognised method (under certain conditions), for one or more classes of pig	Recognised method (under certain conditions), for one or more classes of pig
Firearm (free bullet)	Recognised method (under certain conditions), for one or more classes of pig	Recognised method (under certain conditions), for one or more classes of pig	Recognised method (under certain conditions), for one or more classes of pig	Recognised method (under certain conditions), for one or more classes of pig
Penetrative captive bolt	Recognised method (under certain conditions), for one or more classes of pig	Recognised method (under certain conditions), for one or more classes of pig	Recognised method (under certain conditions), for one or more classes of pig	Recognised method (under certain conditions), for one or more classes of pig
Non-penetrative captive bolt	Not referenced	Not recommended or should be avoided	Recognised method (under certain conditions), for one or more classes of pig	Recognised method (under certain conditions), for one or more classes of pig
Manual blunt trauma	Use is questionable or needs further research, other methods preferred	Recognised method (under certain conditions), for one or more classes of pig	Not referenced	Not recommended or should be avoided
Electrical two-step	Use is questionable or needs further research, other methods preferred	Not referenced	Recognised method (under certain conditions), for one or more classes of pig	Recognised method (under certain conditions), for one or more classes of pig
Electrical single stage	Use is questionable or needs further research, other methods preferred	Not referenced	Recognised method (under certain conditions), for one or more classes of pig	Not referenced

2.3 Literature search strategy

The literature search utilised the CSIRO library database subscriptions. The electronic literature databases included were:

- Web of Science®
- Scopus®
- Agricola®
- Derwent Innovations Index®

The search was completed between 6th - 9th April 2021. Articles identified during the search were uploaded to EndNote reference manager and duplicates automatically detected and removed. This was followed by manual removal of any additional duplicates (for example, publications published in more than one format or indexed in more than one database). Articles identified in the search were also exported to an Excel spreadsheet for sorting, alignment and synthesis. The initial keywords used for the search are summarised in Table 3. Note: '*' indicates variations in spelling or suffix.

Table 3 Key words used in initial search

Category or primary keywords	Secondary or qualifier keywords
Livestock	Pig, piglet, pigs, piglets, swine, porcine, sow, hog, boar
Euthan*	Kill*, stun*, depop*, destr*, terminate, slaughter, euthan*

This returned 48,202 hits in Web of Science®, which indexes the entire paper as well as abstract, title, and keywords, so the search was limited to TOPIC, returning 22,133 hits. From this initial search, irrelevant literature pertaining to 'guinea pig' or 'meat quality' was removed. Papers in languages other than English, French or German were also excluded. Subsequently, additional literature was specifically identified using Web of Science®, Scopus®, Derwent Innovations Index® and Agricola®, using the key words in Table 4:

Table 4 Key words used to refine literature search

Category keywords
Emergency killing, emergency culling, mass killing, mass culling, mass euthan*, mass slaughter, on-farm killing, carbon dioxide, foam, gas-filled foam, gas, electrocut*, gunshot, process, box

The list was further refined by excluding 'mosquito'; 'feed'; 'planting'; 'scalding' and 'swine virus' and by removing duplicates.

2.4 Article screening and selection

After the initial search and screen, which excluded philosophical and opinion type papers, 293 records were identified (comprising 203 articles and 90 patents). These were subject to additional screening through evaluation of each title and abstract to identify target documents for critical appraisal.

2.4.1 Scope

The scope of the review is the year 2000 to 2021, although the final review contains reference to older papers where these represent seminal works, or if there has been insignificant work on a particular topic in recent years. Additional documents were also added to the database if identified during the review process.

2.4.2 Geographical location

The literature search methodology protects against unintentional bias in selection of papers for inclusion in the review. The papers that have been included come primarily from Europe and North America. In reading the review it should be acknowledged that factors relating to Australian conditions, environment and established farming practices may not be fully represented in the literature, however, this is considered by the authors in Part 2.

2.4.3 Overview of the identified literature

The majority of literature reviewed was focused on the use of inhalational agents (such as CO_2) as a method for the destruction of pigs. There were fewer studies on the use of mechanical methods, electrical methods, oral agents and injectable agents. Many studies were based on a small sample size in a research or ‘laboratory-scale’ context and assumed a scalable process if mass depopulation was to be considered. There was also a range of research methodologies, methods used for assessment of unconsciousness and a degree of subjectivity around the determination of death; with fewer studies focusing on the induction period and the time to loss of consciousness. Studies that did focus on the induction period tended to be carried out in the proposed context of commercial slaughter processing for human consumption as opposed to on-farm killing or mass depopulation.

3 Part 1 – Animal welfare considerations

The objective of Part 1 was to review methods available for pig destruction to identify potentially suitable methods for inclusion in Australian standards and guidelines for the humane destruction of pigs. This section considers the suitability of a method for an individual pig, taking into account the class of pig. It focuses on the animal welfare outcome. The application of the method for mass depopulation is not considered in Part 1. For a method to be deemed suitable for the humane destruction of pigs, we firstly need to establish the meaning of the term ‘humane’. Fundamental to the concept of humane is the meaning of ‘consciousness’ and ‘unconsciousness’.

There are many definitions of consciousness, but in general it is associated with the awake state and the ability to perceive, interact and communicate with the environment and others (Zeman 2006). Unconsciousness (the opposite to consciousness) is defined as:

> a state of unawareness (loss of consciousness) in which there is temporary or permanent disruption to brain function. As a consequence of this disruption, the unconscious animal is unable to respond to normal stimuli, including pain (EFSA Scientific Panel on Animal Health and Welfare 2006).

If an animal is conscious or if it regains consciousness pain, fear, and distress can be experienced.

Humane destruction methods should ideally induce an immediate state of general unconsciousness that lasts until death occurs (EFSA Scientific Panel on Animal Health and Welfare et al. 2020a; EFSA Scientific Panel on Animal Health and Welfare 2004). Under practical conditions, EFSA (EFSA Scientific Panel on Animal Health and Welfare 2006; EFSA Scientific Panel on Animal Health and Welfare 2004) has defined immediate (or instantaneous) as “unconsciousness occurring within 1 second” of the intervention being applied. For methods that do not induce immediate unconsciousness, any alternative procedure should ensure: 1) the absence of pain, distress and suffering until the onset of unconsciousness, and 2) that the animal remains unconscious and insensible until death. Controlled atmosphere methods fall into this category as unconsciousness is induced gradually (Gerritzen & Raj 2009). The method must either kill animal whilst it is unconscious, or result in a duration of unconsciousness that is longer than the time needed for a secondary (or terminal) procedure to kill the animal (Gerritzen & Raj 2009).

Part 1 of the review focuses on the ability of the identified methods to produce a state of unconsciousness, without the animal feeling pain, fear or distress, which lasts until the animal is dead. To achieve this in a practical situation, to align with existing literature and guidelines (EFSA Scientific Panel on Animal Health and Welfare et al. 2020a; EFSA Scientific Panel on Animal Health and Welfare 2004; Gerritzen & Raj 2009; EFSA Scientific Panel on Animal Health and Welfare et al. 2020b; Berg 2012; Gavinelli, Kennedy & Simonin 2014) we need to consider the following:

- The type of handling and restraint required to perform the method

- The time to loss of consciousness and likelihood that the pig experiences pain and distress prior to loss of consciousness, and
- Maintaining a state of unconsciousness until the animal is dead.

The killing methods that have been identified (through the statement of requirement and in the four source documents) as relevant for pigs can be grouped into five categories:

1) Inhalational agents
2) Oral agents
3) Injectable agents
4) Mechanical
5) Electrical.

For the electrical methods, single stage head-only electrical stunning (followed by a killing method, such as exsanguination) and electrocution (single-step and two-step) are considered. Mechanical methods include penetrative captive bolt, non-penetrative captive bolt, manual blunt force trauma and the use of a firearm with free projectile. Inhalational agents include gases administrated in containers or gas-filled foams. Injectable agents include anaesthetic drugs, whist oral agents study the use of sodium nitrite. Some of the methods will directly result in death, whist others will need to be followed as by a secondary (terminal) procedure to ensure death.

If a method is deemed to be acceptable based on the criteria discussed, then its suitability as a method for mass depopulation will be considered in Part 2.

3.1 Inhalational agents

The use of inhalational agents, mainly carbon dioxide (CO_2) for the purpose of killing pigs has been studied mainly in the context of processing for human consumption. Commercially, CO_2 systems are used for stunning pigs before slaughter, whilst for the purpose of mass depopulation there may be opportunities to use other gas mixtures and alternative methods of delivery. In the context of this review, inhalational agents include:

- Carbon dioxide
- Nitrogen and inert gases
- Gas mixtures (inert gases + CO_2)
- Foams (gas or air-filled)

3.1.1 Carbon dioxide

Controlled atmosphere stunning (CAS) is used during pig processing for human consumption. Research undertaken over the last 30 years has highlighted some of the animal welfare concerns associated with the use of this method in abattoirs, however, it remains in common use throughout Australia. Under commercial processing conditions, the dominant method involves lowering individuals or small groups of pigs in a gondola into a pit that is pre-filled with a high concentration of more than 80% CO_2. It is usual for the systems to be used for slaughter weight pigs, between 50-100kg, although they are also sometimes used for heavier weight pigs. In commercial processing systems, pigs are moved quickly into a high concentration of carbon dioxide, with some industry standards requiring that conveyance to the maximum concentration of gas occurs within 30 seconds (RSPCA Australia 2018). An advantage with some of the commercial CAS systems is that pigs can be handled and stunned in small groups rather than separated and restrained individually (EFSA Scientific Panel on Animal Health and Welfare 2004; Nowak, Mueffling & Hartung 2007a; Nowak, Mueffling & Hartung 2007b). Handling pigs in a group rather than using systems which require animals to move in single file has been shown to result in lower stress and improved movement through the system.

The use of inhalational agents for depopulation in pig production has been comprehensively reviewed in recent publications (EFSA Scientific Panel on Animal Health and Welfare et al. 2020b; Arruda et al. 2020). Inhalational euthanasia systems for poultry were the first systems developed, and research into inhalational euthanasia for pig depopulation has been based on species-specific validation and up-scaling of such systems. The gases used are carbon dioxide (CO_2), nitrogen (N_2), oxygen (O_2), nitrous oxide (N_2O), argon (Ar) or mixtures of these. The concept involves using the desired gas mixture to displace atmospheric air in the container holding the animals, such that a lethal anoxic (low oxygen) or hypercapnic (high CO_2) situation is generated and maintained until the animals have died. The gas may also be administered in the form of a foam. One of the advantages of this type of system is that the pigs do not have to be individually restrained and handling stress is minimised (EFSA Scientific Panel on Animal Health and Welfare 2004; Nowak, Mueffling & Hartung 2007a; Nowak, Mueffling & Hartung 2007b).

Disadvantages, however, include the facts that the onset of unconsciousness is not immediate, and there is a period of time during which the animals perform behavioural responses indicative of aversion, stress and distress (Nowak, Mueffling & Hartung 2007a; Nowak, Mueffling & Hartung 2007b; Gerritzen et al. 2012; Verhoeven et al. 2016; Sutherland, Bryer & Backus 2017; Kells et al. 2018). Furthermore, if animals are not killed by the procedure, there is a risk of return to consciousness, one study reporting 10% of pigs returning to consciousness after 15 minutes of exposure to a CO_2 concentration of 63.5% in a transport truck 'sealed' using polythene sheets and duct tape (Meyer & Morgan Morrow 2005).

An expert elicitation process conducted by the European Food Safety Authority (EFSA) prioritised the following hazards to animal welfare when using inhalational agents for depopulation on-farm (EFSA 2019):

- Exposure time too short
- CO_2 concentration too low
- No clear operating instructions, leading to the method not being applied properly
- Inhalation of a high CO_2 concentration
- Temperature too low
- Low foam production rate
- Jet stream of gas at animal level, and
- Bubble size too small.

These hazards are discussed more fully in a later EFSA review, but briefly (EFSA Scientific Panel on Animal Health and Welfare et al. 2020b):

- If the exposure time is too short, there is a risk that pigs may not be killed and will return to consciousness
- If the CO_2 concentration is too low, the pigs may not be rendered unconscious, and the duration of distress prior to induction of unconsciousness can be prolonged (Raj & Gregory 1996)
- Operating instructions - for example the stocking rate can affect the CO_2 concentration in the chamber (Fiedler et al. 2014); leaking seals can reduce the CO_2 concentration (Meyer & Morgan Morrow 2005); and non-vapourised (liquid) CO_2 can lead to painful freeze-burns (Meyer & Morgan Morrow 2005). Staff training in the operation of the equipment was recommended in the EFSA review (EFSA Scientific Panel on Animal Health and Welfare et al. 2020b), including monitoring and adjusting gas flow rates, assessing pigs for death and application of back-up killing if required
- A high CO_2 concentration is aversive, and painful to inhale (Sutherland, Bryer & Backus 2017). Therefore, research continues into alternative processes, gases or gas mixtures that may be less aversive (see Section 3.1.2)
- CO_2 is supplied in the form of solid pellets (dry ice) or compressed gas (liquid form). These convert to gas as the pressure decreases and temperature rises. In liquid and solid state and during gasification, the temperature of the CO_2 is less than 0°C. This can result in contact freeze-burns to skin and is painful to inhale
- In gas-filled foams, the rate of release of gas from the foam is critical to ensure rapid unconsciousness and death. The gas is released when the foam bubbles burst, which can be precipitated by animal movement within the foam. Foam production rates must ensure that gas concentrations are maintained, because if the gas concentration is too low, the pigs may not be rendered unconscious, and the duration of distress prior to induction of unconsciousness can be prolonged (Raj & Gregory 1996)
- The positioning of the jet stream relates to both concentration and temperature aspects – the gas exiting the jet stream is highly concentrated and low temperature, so animal welfare may be compromised if animals are directly in the jet stream
- In gas filled foams, a high water-content, indicated by small bubble size, presents the risk of animals inhaling liquid, particularly as foam levels reach the head. This will result in respiratory distress and drowning, as opposed to inhalational euthanasia.

3.1.2 Alternative gas mixtures

A variety of gas parameters have been evaluated for pig stunning in slaughterhouses or euthanasia on-farm, including CO_2 in concentrations from 30% to 90% in air; 40% CO_2 with 30% O_2 in air; 30% CO_2 with 70% N_2O; 15 - 30% CO_2 in N_2 with <2% O_2; 50% N_2O in air; 100% CO_2; 90 to 100% Ar; 60% Ar with 40% CO_2; 50% CO_2:Ar. No mixture has been shown to eliminate behavioural or physiological signs of stress or distress in pigs, and an important factor maybe the finding that increasing concentrations of CO_2 result in earlier onset of behavioural signs, and earlier induction of unconsciousness – but the interval between the onset of behavioural signs and loss of consciousness is not affected (Nowak, Mueffling & Hartung 2007a; Nowak, Mueffling & Hartung 2007b; Atkinson et al. 2020). One of the challenges in drawing firm conclusions or ranking various gas mixtures lies in the large variety of research methodologies and assessment parameters used in published literature. There is no standardized protocol, so comparisons between published data are difficult.

Nitrous oxide (N_2O) is an anaesthetic gas used in human and veterinary surgery applications and is known for its sedative and analgesic properties (Clark & Brunick 2003). Recently, interest has been shown in adding N_2O to the gas mixture for pig stunning (Terlouw, Deiss & Astruc 2021), using N_2O alone (Cavusoglu et al. 2020) or applying a two-stage approach in which pigs are first exposed to N_2O before being exposed to the CO_2 mixture (Steiner et al. 2019). Adding N_2O to the gas mixture at 70% N_2O:30% CO_2 did not appear to provide any welfare benefits over 80% CO_2 in air (Terlouw, Deiss & Astruc 2021), but use of N_2O alone (gradual fill to 95% concentration) was considered to offer a potential alternative to CO_2 (gradual fill to 95% concentration) for weaner piglets (aged approximately 21 days), escape attempts being reduced although vocalisation increased (Cavusoglu et al. 2020). Using a two-stage process, in which pigs were exposed first to N_2O for 6 minutes and then to CO_2 (both gradual fill to 95% concentration), researchers observed that piglets (0-7 days of age) in the staged treatment group took longer to lose consciousness than pigs in a CO_2-only system, and displayed more vocalisation, escape attempts and righting responses (Smith et al. 2018). This led them to conclude that the two-stage system was not more humane than the single-stage CO_2 treatment.

3.1.3 Gas-filled foam

High expansion N_2- or CO_2-filled foams have been considered first for use in poultry shed depopulation and more recently for pigs. In poultry, use of CO_2 in the foam did not appear to provide a welfare benefit over N_2 and was more challenging to deliver through foam generators due to the extreme cold produced (Gerritzen et al. 2010; McKeegan et al. 2013). N_2-filled foams have been evaluated for pig euthanasia, but behavioural responses indicative of aversion have been recorded as the O_2 concentration in the container decreases, and particularly when the foam level reached the head of pigs (Lindahl et al. 2020).

3.1.4 Air-filled foam

Foam generated using ambient air has been used to depopulate poultry houses. The aim is to generate a blanket of foam over the birds, with bubble sizes suitable to block the airways and asphyxiate the birds (Thornber, Rubira & Styles 2014). Evaluations in poultry species, including ducks and turkeys, have indicated that water-based air-filled foam is no more aversive than CO_2-filled foams, and may provide faster euthanasia when dealing with larger numbers of birds (Benson et al. 2007, Benson et al. 2009). When compared with CO_2 gas systems, foam systems provided a shorter time to unconsciousness in two studies (Rankin et al. 2013; Benson et al. 2018), but a longer time to cessation of movement in a third study (Gurung et al. 2018). Reasons for the conflicting outcomes are not clear but may be attributable to differences in the research methodologies. Foam containing CO_2 provided shorter time to unconsciousness and brain death than air-filled foams in one study (Caputo et al. 2012), but this was not supported by another, which found no significant difference between air-filled or CO_2-filled foams (Benson et al. 2018).

Air-filled foams have not been tested on pigs, but challenges with achieving obstruction of the airways (which are larger in pigs than in poultry) without inhalation of fluid and associated respiratory distress or drowning may preclude its use in pigs.

In summary, inhalational agents have advantages in that animals can be euthanised in groups, but questions remain around the risk to welfare, particularly where rates of exposure to increasing concentrations cannot be adequately controlled. Although much is known about use of CO_2 gas systems in slaughter-age pigs, and there is a developing body of knowledge around euthanasia of neonates, there has been insufficient investigation or validation of alternative gas mixtures or foams in the broader range of pig ages and weights that exist in production premises to draw clear conclusions on welfare aspects.

3.2 Injectable agents

3.2.1 Mode of action

In the context of this review, injectable agents include anaesthetic compounds, such as barbiturates and barbituric acid derivatives. Other injectable agents are reviewed in depth by the American Veterinary Medical Association (AVMA) 2020 guidelines for the euthanasia of animals (AVMA 2020). An injectable anaesthetic is a compound which, when administered appropriately, acts rapidly and induces a smooth transition to unconsciousness and death. The type of active ingredient will affect the mode of action. Barbiturates depress the central nervous system, resulting in anaesthesia. With an overdose, deep anaesthesia progresses to apnoea due to depression of the respiratory centre, followed by cardiac arrest (AVMA 2020). Sodium pentobarbital is the injectable agent registered and most widely used in Australia for the euthanasia of pigs (APVMA, 2021). Within Australia, anaesthetic compounds are scheduled substances under the Poisons Standard 2021 (Standard for the Uniform Scheduling of Medicines and Poisons (SUSMP) No. 33) and regulatory requirements specify that these agents can be administered under the authority of a registered veterinarian. Administration of injectable agents requires handling and restraint of individual animals, and it may also be necessary to reduce animal activity by the use of sedation prior to injection (AVMA 2020; EFSA 2018). Intravenous (IV) administration in particular may be difficult in small, active piglets (Kells et al. 2018), particularly if injecting into an ear vein. Mixing of pentobarbital with a neuromuscular blocking agent is not considered to be an acceptable approach to euthanasia because of the potential for the neuromuscular blocking agent to induce paralysis prior to onset of unconsciousness (AVMA 2020).

3.2.2 Route of administration

When injectable agents are administered, the route of administration will affect the outcome. Intravenous (IV) delivery is usually preferred as it achieves more rapid distribution of the agent; whilst placement in other areas of the body may reduce speed and efficacy as well as the extent of associated irritation or pain (Kells et al. 2018). The pain experienced by animals when IV administration is used is thought to be minimal or confined to the transient pain associated with venipuncture (AVMA 2020). However, it should be noted that products containing concentrated pentobarbital solutions are viscous and therefore a large bore needle is required to administer the injection (Caputo et al. 2012).

The AVMA list the use of barbiturate or barbituric acid derivative (administered IV) as preferred methods of euthanasia for pigs. Intracardiac, intramuscular (IM), intrahepatic, and intrarenal injections are only recommended if the animal is unconscious or anaesthetised (AVMA 2020). In a field study of early weaned piglets, the researchers found the failure rate of intraperitoneal (IP) pentobarbital injection high when compared with mechanical euthanasia methods. In injected piglets, 6.7% were assessed as alive after 60 mins, with 2% showing return of consciousness (Whiting et al. 2011). The authors attributed the high failure rate to accidental injection into the bladder or faecal mass, or administration of a sub-lethal dose (as piglets were not weighed before administration). There is also concern that pentobarbital preparations may cause pain or irritation, prior to the loss of awareness, when delivered intraperitoneally (IP) (Kells et al. 2018). This is particularly apparent when euthanasia preparations are used, which have a higher concentration of active ingredient compared to solutions manufactured for anaesthesia (Kells et al. 2018). No other studies investigating the noxiousness of IP pentobarbital euthanasia in pigs were identified. There were no studies on the refinement of IV techniques to improve administration and further investigations in this area may be beneficial.

Despite the skill required to administer injectable agents, they were regarded as a possible alternative to existing euthanasia techniques in a Brazilian study of stockpersons' attitudes towards euthanasia (Dalla Costa et al. 2019); with most stockpersons (96%) taking part in the study declaring that they felt uncomfortable with the act of dispatching pigs at the farm, and 26% of stockpersons suggesting 'anaesthetics' as a suitable alternative to manual blunt force trauma.

3.2.3 Injectable agents as terminal procedures

Although unacceptable when used in conscious pigs, a solution of potassium chloride, magnesium chloride, or magnesium sulphate injected IV or intracardially in an animal that is unconscious or under general anaesthesia will induce cardiac arrest and death. It is therefore considered to be an acceptable terminal procedure in unconscious animals (AVMA 2020). These substances are not scheduled under the Poisons Standard and can be easily acquired. After administration, the carcasses are potentially less toxic for scavengers and predators than after the use of other injectable agents. The use of other injectable agents, such as opioids, would also be effective terminal procedures, however, are strictly regulated, require special licensing to obtain and use.

3.3 Oral agent – sodium nitrite

In Australia, sodium nitrite was first identified as a possible oral euthanasia agent for feral pigs in the 1980s; being reviewed again in 2008 as a potential method for feral pig control (Cowled, Elsworth & Lapidge 2008). In December 2019, Animal Control Technologies (Australia) Pty Ltd registered HOGGONE microencapsulated sodium nitrite (MeSN) with the Australian Pesticides and Veterinary Medicines Authority (APVMA) as a bait for the reduction of feral pig populations, though it is not yet registered for use in domestic pigs. The use of sodium nitrite is also recognised as a conditional method of euthanasia for pigs by the AVMA (AVMA 2020) under constrained circumstances. A model for assessing the relative humaneness of pest animal control (Edition 2) was published in 2011 (Sharp & Saunders 2011). Using this model, an assessment of the methods available for the destruction of feral pigs ranked the overall welfare outcome of the use of sodium nitrite as exceeding that of a chest shot, second only to a head shot with a firearm.

Sodium nitrite ($NaNO_2$) is commonly used at very low concentrations in the food industry, however at high doses it is a red blood cell toxin, causing central nervous system anoxia, lethargy and death (Shapiro et al. 2016). It has a saline, slightly bitter taste which is aversive to pigs unless it is microencapsulated (Humphrys 2017). In earlier studies, where microencapsulation was not used, pigs were reluctant to eat the bait, resulting in lower mortality rates and therefore efficacy (Shapiro, Hix & Eason 2009). Microencapsulation also provides stability of the compound, which in its unprotected form is hygroscopic, meaning that it absorbs water from the air and readily degrades to non-toxic compounds.

Pigs are very susceptible to sodium nitrite poisoning, with doses of >90mg/kg (gavage feeding) and >400mg/kg (free feeding) resulting in lethal outcomes (Shapiro et al. 2016). However, these trials used the granular (non-encapsulated) form of the ingredient, which is known to be taste aversive to pigs. In further trials on penned feral pigs, using microencapsulated sodium nitrite in paste, average doses of 760mg/kg resulted in 'rapid death with little distress evident' (attending veterinarian observations) (Shapiro et al. 2016). This study reported that 8/9 pigs in the pen trial consumed a lethal dose of bait. Two US industry reports, evaluated the use of microencapsulated sodium nitrite in domestic pigs delivered in feed, drinking water and using oral drenching (Pepin 2020), with lethal outcomes and time to death dependent on dose rate. An oral drench of 1200mg/kg in sows achieved 90% lethal outcomes in an average of 31 minutes. Vomiting was reported as an undesirable side effect, however, this could possibly be managed by refining the dose rate to provide an acceptable balance between time to death and side effects. There have been limited trials using sodium nitrite in drinking water. Pigs were averse to consuming water with granular sodium nitrite dissolved and there was minimal intake and no clinical signs (Lower et al. 2020). An Australian study (APL, 2021 - unpublished) on the use of microencapsulated sodium nitrite as an oral agent for domestic pigs has recently been completed, however, the results are not yet published. It demonstrated that the use of sodium nitrite in domestic pigs is very effective, with no aversion, no individual restraint required, acceptable welfare (attending veterinarian observations) and high mortality in a short period of time.

Within Australia, other oral agents are also registered for use in feral pig baits. Brodifacoum is a hydroxycoumarin which, like the anti-coagulant Warfarin, interferes with the production of vitamin K–dependent coagulation. Sodium fluoroacetate (1080 poison) and phosphorus are poisons that cause circulatory collapse. Time to death with these particular agents can be variable, sometimes taking days after ingestion to kill the animal; with affected individuals experiencing discomfort and sometimes central nervous system disturbance prior to coma and death. Studies on the use of sodium nitrite indicate that it may induce unconsciousness and death more quickly, offering a potentially more humane solution (Cowled, Elsworth & Lapidge 2008; Shapiro et al. 2016; Snow et al. 2017; Snow et al. 2021). The RSPCA Australia also recognise sodium nitrite as a 'more humane and effective toxic bait' than 1080 poison in the control of feral pigs (RSPCA Australia 2021).

3.4 Mechanical methods

The principal mode of action for mechanical methods is the induction of concussion resulting in unconsciousness, through the impact of a percussive blow to the head. In the context of this review, mechanical methods include:

- Firearm (free projectile)
- Penetrative captive bolt and non-penetrative captive bolt
- Manual blunt force trauma.

3.4.1 Firearm (free projectile)

The use of a firearm (with free projectile) involves the passage of one or more projectiles into the cranium causing immediate unconsciousness and extensive damage to the brain, ultimately resulting in the death of the animal (Humane Slaughter Association 2016). The physical principle behind killing with free projectiles is the transfer of high levels of kinetic energy in an extremely short time from the projectile to the animal's brain. The free projectile may be a bullet (used in a rifle or handgun) or a charge of lead (used in shot guns). There are few scientific studies on the use of different firearms for killing pigs in the field, although industry guidance documents from the Humane Slaughter Association are available (Humane Slaughter Association 2016), which recommend firearms suited to different pig types (summarised in Table 5). Muzzle energies of 400 N-m or more are recommended for killing adult sows, boars and growing/finishing pigs, because of the thickness of the pig's skull (AVMA 2020). Shooting the animal in the head is essential to ensure an acceptable animal welfare outcome when using firearms (Gerritzen & Raj 2009). Effective killing using a firearm is dependent on the shot position (and angle of penetration) and the kinetic energy of the projectile (Whiting & Will 2019). Most recommendations refer to the optimal shot position being on the frontal region of the head, aiming to penetrate the skull damage the important structures of the brain (EFSA Scientific Panel on Animal Health and Welfare et al. 2020b), however, alternative positions have been used successfully (Humane Slaughter Association 2016). When alternate site behind the ear is chosen, a 0.22 calibre firearm loaded with a solid-point bullet is effective (EFSA Scientific Panel on Animal Health and Welfare et al. 2020b). To improve energy transfer, some bullets are constructed to fragment within the skull, resulting in more widespread brain damage than seen with high velocity bullets (Humane Slaughter Association 2016; Whiting & Will 2019).

Table 5 Recommendations for the use of firearms arranged by pig type

Firearm	Piglet/weaner	Grower/finisher <100kg	Adult sow/boar >100kg
12-bore shotgun[1]	Recommended	Recommended	Recommended
16-bore shotgun[1]	Recommended	Recommended	Recommended
20-bore shotgun[1]	Recommended	Recommended	Recommended
28-bore shotgun[1]	Recommended	Recommended	Not recommended
.410 shotgun[1]	Recommended	Recommended	Not recommended
.22 rifle[1]	Recommended	Recommended	Not recommended
.243 rifle[2]	Not recommended	Recommended	Recommended
.270 rifle[2]	Not recommended	Recommended	Recommended
.308 rifle[2]	Not recommended	Recommended	Recommended
.32 humane killer[3]	Recommended	Recommended	Recommended

[1] Fired short distance (5-25cm) from the pig's head with safe backdrop
[2] Fired from a distance with safe backdrop
[3] Single shot pistol placed against the pig's head

For pigs reared in an outdoor system, it may be possible to shoot them from a distance, rather than having to catch and restrain animals. This can be done using a free bullet gun, such as a rifle with telescopic sight. The use of a silencer, or subsonic rounds, is advised to reduce the noise and risk of disturbance to other animals (Whiting et al. 2011; Gerritzen & Gibson 2016). A study under field conditions in early weaned piglets (around 21 days old and approximately 5kg) found the use of a free bullet gunshot (.22 rim fire charge) challenging, due to incidences where the bullet passed through the target pig and wounded a non-target pig (Whiting et al. 2011).

Adult pigs can be difficult to kill with free bullet, due to the anatomy of the skull and frontal sinus (Gerritzen & Gibson 2016). The use of shotguns has been suggested as an alternative and recommendations for their use is covered by industry guidance (Humane Slaughter Association 2016). If used properly, they are regarded as equally effective and safer than free bullet guns. It is recommended that they are used a short distance from the target (5-25 cm), therefore would require the pig to be restrained or confined in a small area. The use of a 12-gauge shotgun was studied in a preliminary trial on the cadaver heads of large sows and boars to establish whether the damage caused would be sufficient to cause immediate unconsciousness and death in a live animal. Following the preliminary investigation on cadaver heads, a single live large sow was shot behind the ear, collapsing immediately and showing pupillary dilation indicative of death (Blackmore et al. 1995).

3.4.2 Penetrative captive bolt

Penetrative captive bolt devices are designed to fire a retractable steel bolt through the cranium and into the brain of the animal. Penetrative captive bolts used on-farm are normally powered by a blank cartridge, however, pneumatically powered devices are also available. The desired outcome is for the impact of the bolt on the skull to result in concussion and the associated immediate loss of consciousness (EFSA Scientific Panel on Animal Health and Welfare et al. 2020a; EFSA Scientific Panel on Animal Health and Welfare 2004).

Pigs can be difficult to stun with a penetrative captive bolt pistol due to the position and size of the brain relative to the head and the shape of the head (EFSA Scientific Panel on Animal Health and Welfare 2004; Gavinelli, Kennedy & Simonin 2014; Humane Slaughter Association 2016; Humane Slaughter Association 2017; Humane Slaughter Association 2013). Efficacy is thought to be directly impacted by the ability of the bolt to penetrate the skull. As pigs age, the thickness of the skull increases, the frontal sinuses expand, and some breeds may develop a bony ridge on the midline of the head (Humane Slaughter Association 2013). For mature pigs, such as breeding sows and boars, operatives may be reluctant to use a penetrative captive bolt (Gerritzen & Gibson 2016) and a firearm (with free projectile) has been found to be more successful (Humane Slaughter Association 2016).

Bolt diameter, velocity and penetration depth are important determinants of stunning outcome (EFSA Scientific Panel on Animal Health and Welfare 2004). The structural damage to the brain may lead to rapid death of the animal (AVMA 2020), however some animals may not die immediately depending on the degree of damage to the brain (Lambooji & Algers 2016). Shot position, and its influence on the areas of the brain that are damaged, will also determine the stunning outcome (EFSA Scientific Panel on Animal Health and Welfare 2004; Humane Slaughter Association 2013). Research on the use of a penetrative captive bolt as a method for cattle indicates that the probability of damaging the brainstem and causing death can be increased if the shot position is adjusted (Gilliam et al. 2016). In pigs, researchers (Kramer et al. 2021) investigated the use of different positions for the placement of the devices, evaluating frontal, temporal, and behind-the-ear placement in anaesthetised adult pigs. The study demonstrated that devices with a higher power load and bolt penetration depth were more likely to result in brain trauma and death. Research studies on a commercially available euthanasia kit also demonstrated that penetrative captive bolt pistols could be an effective single step killing method for piglets, with the placement of the device and bolt length determining the nature of the damage to the brain and the overall outcome in the animal (Woods 2012). In these studies, the pigs consistently died after the application of a penetrative captive bolt; however visible traumatic brain injury was only present in one pig. This finding supports the common understanding that direct penetration of the brain with the bolt is not associated with efficacy (Humane Slaughter Association 2013) and visual evaluation of brain injury is not a reliable indicator of the effectiveness of a penetrative captive bolt. In a study on pigs weighing 2-30kg, penetrative captive bolt devices were trialled and were found to be effective for all animals (Jacobson, Schulz & White 2011), resulting in the immediate death of all animals 13kg and under.

With larger pigs, it is recommended that a secondary (or terminal) method is used to ensure death (AVMA 2020). An EFSA opinion paper on the killing of pigs for purposes other than slaughter (EFSA Scientific Panel on Animal Health and Welfare 2020b) recommends that a terminal procedure is performed for all pigs shot with a captive bolt pistol, citing pithing, lethal injection (with barbiturate or potassium chloride) and bleeding as possible suitable methods. Pithing is a relatively simple method that involves inserting a flexible wire or polypropylene rod through the hole in the head made by a penetrative captive bolt. The movement of the rod is used to physically destroy the brainstem and upper spinal cord, ensuring death, and also reducing convulsions. Disposable pithing rods that remain within the head and plug the bolt hole, reducing the leakage of body fluids, are useful during a disease outbreak (Gerritzen & Gibson 2016). The application of an electrical current across the chest (to induce cardiac arrest) can also be used as a terminal procedure after the use of a penetrative captive bolt (Gerritzen & Raj 2009). The Farm Animal Welfare Council (UK) also support the use of a secondary (terminal) procedure after captive bolt stunning (Farm Animal Welfare Committee 2017).

Poor maintenance is a major cause of captive bolt device failure (Humane Slaughter Association 2013). Repeated firing of a captive bolt device for extended periods may also reduce effectiveness (Gibson et al. 2015). The effect of the quality and consistency of blank cartridges on the stunning outcome has also been studied (Grist et al. 2020; Grist et al. 2019), with large differences in cartridge performance being found.

3.4.3 Non-penetrative captive bolt

Non-penetrative percussive stunning involves the controlled application of a blunt force trauma to the head of the pig. It was introduced as a potential killing method for neonatal pigs as a replacement for manual blunt force trauma (Grist et al. 2018) (Section 3.4.4). In a study comparing the two methods in early weaned piglets, blunt force trauma using a non-penetrative mechanical device was more effective (0% failure, n = 108) than manual blunt force trauma (24% failure, n = 50) (Whiting et al. 2011)

The effectiveness of non-penetrative percussive devices as a killing method has been studied in a range of pig sizes; though all focusing on neonatal (pre-weaning) or nursery (post-weaning) pigs during the early stages of production: < 72 hours (Casey et al. 2013); 4-49 days/3-9 kg (Casey et al. 2014); < 10kg (Woods 2012) < 11 kg (Grist et al. 2017); 5.8 days mean age (Grist et al. 2017); 2-30kg (Jacobson, Schulz & White 2011). There are very few studies that investigate the use of non-penetrative devices on larger animals, with most studies using piglets of around 10kg or less. In the trial performed on piglets between 2-30 kg (Jacobson, Schulz & White 2011), the non-percussive device was tested on one larger animal (30 kg) and did not result in death. The pig was stunned effectively, however a second step was required to kill the animal. This observation was consistent with another study which also described non-penetrating devices as less effective than penetrative captive bolts as a single step killing method (Kramer et al. 2021). Severity of traumatic brain injury has also been found to be much less with non-penetrative devices compared with penetrative captive bolts (Finnie et al. 2003). The results of that study also showed that acceleration/deceleration of the porcine head after temporal impact with a non-penetrative device causes only minimal brain damage compared with that seen in sheep after an equivalent stunning intervention.

The use of a non-penetrative device for killing piglets is supported in various advisory documents, standards and codes of practice (AVMA 2020; Farm Animal Welfare Committee 2017; WOAH 2019). In most of the experiments on neonatal pigs, the use of a non-penetrative captive bolt device rendered the animals immediately unconscious, with no return of sensibility before death occurred. In one study, sustained convulsions or irregular heartbeat prompted researchers to apply a secondary (terminal) procedure as a precautionary measure (Casey-Trott et al. 2013).

In a study of low-viability newborn piglets (n = 175), the use of a non-penetrative device appeared less effective when compared to the application of a manual blow to the head (blunt force trauma) (Widowski, Elgie & Lawlis 2008). In the blunt force trauma group (n = 76), all piglets were killed without showing any return to consciousness. In the non-penetrative captive bolt group (n = 99), 13.1 % showed signs of returning consciousness. It was concluded that the effectiveness of the non-penetrating captive bolt varied between operatives, whilst manual blunt force trauma was consistently effective for low-viability neonatal piglets. Unfamiliarity with the use of the device could partially explain the inconsistent outcome between operatives. Similar inconsistency between operatives was found in other studies, where time to brain death and cardiac arrest differed significantly between stockpersons (Casey-Trott et al. 2013; Casey-Trott et al. 2014). This indicates that, like manual blunt force trauma, the efficacy of the technique is highly dependent on the competency of the operative.

Despite their apparent effectiveness in piglets, non-penetrative captive bolts are not considered an effective method for slaughter weight or breeding pigs (AVMA 2020) and there is little scientific evidence to support their use in this context.

3.4.4 Manual blunt force trauma

This method is widely used in neonatal pigs (EFSA 2018). It is usually performed by holding the body or legs and delivering a percussive blow to the forehead with a hard object. Successful application of this method is dependent on the skill of the operative and their ability to impart sufficient force to the blow (Arruda et al. 2020; Grist et al. 2017; Dalla Costa et al. 2020). With sufficient force and accuracy, irrecoverable concussion is produced, however the percussive blow may not always be of a magnitude to produce death. Given this uncertainty around the outcome, it is stipulated in the EU that the method must only be used when no other methods are available, must be limited to < 70 animals (< 5kg liveweight) per day and must be followed by exsanguination (Gavinelli, Kennedy & Simonin 2014). Furthermore, the probability of consistently achieving an immediate kill is low and misapplication can lead to pain and distress to the animal (Grist et al. 2018; Dalla Costa et al. 2020). It is also a method of killing that is aesthetically unpleasant for both the operator and any bystanders (Dalla Costa et al. 2019; Grist et al. 2018; Husheer et al. 2020).

During the study of an actual emergency response, manual blunt force trauma was associated with a 24 % failure rate (n = 50), where 5 pigs failed to die within a 2-5 minute assessment period and one pig returned to consciousness (Whiting et al. 2011).

3.5 Electrical methods

In the context of this review, electrical methods include:

- Electrical stunning (followed by a killing method or terminal procedure such as exsanguination)
- Electrocution (single-step or two-step methods).

3.5.1 Electrical stunning

In a commercial abattoir setting, particularly in small throughput facilities, the use of electrical stunning is a method used for stunning pigs prior to slaughter. The aim of electrical stunning is to pass an electrical current across the brain of the pig, resulting in an epileptic fit during which the pig is unconscious. Effective head-only electrical stunning is characterised by immediate collapse and onset of tonic seizures during exposure to the current (EFSA Scientific Panel on Animal Health and Welfare 2004; McKinstry & Anil 2004). Electrical stunning requires the animal to be individually handled and restrained (on a restrainer or in a pen) to allow electrodes to be applied to the head. Individual animal handling and restraint of this nature can be stressful (Brandt & Aaslyng 2015).

Head-only electrical stunning does not kill the animal, but results in a recoverable state of unconsciousness (EFSA Scientific Panel on Animal Health and Welfare 2004). Therefore, this method must always be followed by a secondary (terminal) procedure, such as exsanguination. If exsanguination is used, it is recommended that the stun to stick (bleeding) interval is kept as short as possible to reduce the risk of the pig recovering from the stun (a maximum of 15 seconds is recommended (EFSA Scientific Panel on Animal Health and Welfare 2004). Head-only electrical stunning can be used as part of a two-step process of electrocution (Section 3.5.2).

Reviewed literature and guidance (EFSA Scientific Panel on Animal Health and Welfare 2004; Gavinelli, Kennedy & Simonin 2014; WOAH 2019; Galvin et al. 2005; Velarde et al. 2000) concur that effective electrical systems require:

- Selection of equipment that ensures electrodes are an appropriate fit for the size of the animal
- Accurate electrode placement to ensure good electrical contact (without pre-stun shocks)
- Sufficient restraint, such that contact can be maintained for the duration of the stun
- Regular maintenance and calibration to ensure correct electrical parameters are being used.

The required electrical parameters for an effective head-only stun are defined in a number of guidance documents (EFSA Scientific Panel on Animal Health and Welfare 2004; Humane Slaughter Association 2017; WOAH 2019; Galvin et al. 2005). The magnitude of the applied current should be at least 1.25-1.3 A at a low (50 Hz) frequency to meet the recommendations outlined in guidance for slaughter weight pigs. The implementation of constant current rather than constant voltage systems also improves animal welfare as they ensure that the minimum current required to stun the animal is consistently applied (Gerritzen & Raj 2009; Husheer et al. 2020; Wotton & O'Callaghan 2002).

3.5.2 Electrocution

Electrocution involves electrical stunning (as described in Section 3.5.1) with subsequent or concomitant electrical fibrillation of the heart. Two types of technique are used: a two-step method with head-only electrical stunning followed by ventricular fibrillation and the single-step method of head-to-body application. Two-step and single-step applications have been recognised as effective methods in pigs over one week old (Galvin et al. 2005). In a review of methods suitable for disease control, electrocution (single-step or two-step) was recommended over head-only stunning as it induces ventricular fibrillation leading to cardiac arrest and rapid onset of brain death (Gerritzen & Raj 2009).

When the two-step method is used, the application of current to the body should occur without delay (Gerritzen & Raj 2009; Vogel et al. 2011). Since the method is intended to kill the animals, the cardiac arrest current cycle should be of a frequency and magnitude that will effectively cause cardiac fibrillation (EFSA Scientific Panel on Animal Health and Welfare 2004; Gerritzen & Raj 2009). Successful fibrillation is thought to depend on a number of factors affecting current flow, such as the design of the electrodes, the applied pressure and the diameter of the pig's chest (Gerritzen & Raj 2009). In a study of the two-step method, researchers evaluated head-only stunning for 3 seconds immediately followed by a 3 second application of the same stunning electrodes to the cardiac region of the animal (Vogel et al. 2011). In both current cycles 2.3 A was applied using 60 Hz sine wave alternating current (AC). These current parameters eliminated rhythmic breathing, spontaneous blinking, eye tracking to moving objects and righting reflex in pigs. A cardiac fibrillation process may be a preferred terminal method after any non-terminal electrical stunning method, as exsanguination is not usually recommended during killing for disease control purposes. Alternatively, a lethal intravenous (IV) injection of barbiturate or potassium chloride could be used (EFSA Scientific Panel on Animal Health and Welfare et al. 2020b; AVMA 2020).

Electrical killing methods can also be applied in a single-step method as the electrodes span the head and the heart at the same time. Researchers studied a single-step electrocution method for nursery piglets (Beusekom et al. 2001). Two types of equipment were tested: a clamp applied to the ear and opposite flank; a copper wire snare and copper wire encircling the groin area. Both arrangements were designed to allow the passage of an electric current through the body of the piglet, resulting in cardiac fibrillation, however the researchers did not evaluate the immediacy of unconsciousness. In a similar experiment, testing a single-step electrocution method, researchers assessed piglets for signs of unconsciousness (McKinney et al. 2011). In some piglets, indicators of consciousness persisted after application of the method, however the researchers concluded that the system was an effective killing method.

If a two-step process is used (head-only followed by cardiac arrest), restraint may not be necessary for the application of the second current cycle during the tonic seizures induced by head-only current application (Gerritzen & Raj 2009). For the application of single-step hand-held head-to-body electrodes, movement of the animal may need to be restricted in such a way that accurate placement and consistent contact of the electrodes against the animal is ensured.

3.6 Recommendations

Part 1 provides an overview of methods that are acceptable or unacceptable on welfare grounds. Interestingly, many of the articles that study the suitability of methods for humane killing on-farm tend to focus on the capacity of the method to kill of the pig, rather than whether the method induces immediate unconsciousness. Consequently, some of the methods recommended in the literature and guidelines would not satisfy the criteria around 'humaneness' of the process, that being the production of unconsciousness without causing pain, fear or distress. The objective of Part 1 was to provide scientific opinion on the suitability of methods for inclusion in Australian standards and guidelines for the humane destruction of pigs. It was evident from the review that the ability of each method to deliver an acceptable animal welfare outcome is largely dependent on the animal type. With the exception of air-filled foam, there is sufficient evidence that the methods reviewed in Part 1 are suitable for pigs of a particular type (age, size etc.), but there is not one individual method that is suitable for all types of pig in all situations. Air-filled foam is regarded as unacceptable at present, due to the absence of underpinning science covering its use. Air-filled foams have not been tested on pigs, but challenges with achieving obstruction of the airways without inhalation of fluid and associated respiratory distress or drowning may preclude its use in pigs. All the other methods could be regarded as conditionally acceptable, provided that appropriate conditions for their use in a production environment or as part of an emergency response are established. The ultimate decision regarding the selection of methods for mass depopulation does not only require scrutiny from an animal welfare perspective, but additional process requirements, for example biosecurity, cost, aesthetics and safety also need to be considered. This is discussed in Part 2.

4 Part 2 – Suitability of methods for mass depopulation

In general, on-farm killing involves killing of animals that are injured, diseased (and unlikely to recover) or for disease control purposes, on their production site. Pigs might also be killed for economic reasons, deteriorating husbandry conditions or in the event of other unforeseen emergency situations, for example reduced slaughtering capacity due to the COVID-19 pandemic or other supply chain disruption (Grandin 2021; Marchant-Forde & Boyle 2020). Two main scenarios exist: large-scale killing (or mass depopulation) and killing of small numbers of individual animals. Large-scale (or mass) depopulation refers to the destruction of large numbers of pigs and may not only include animals affected by disease, but also healthy animals of varying ages in different production systems. This discussion section considers the identified potential methods for euthanasia of pigs, in the context of mass depopulation. The efficacy of the methods for large-scale depopulation may not always reflect that observed when the same methods are used in an abattoir or when they are applied to individual or small numbers of pigs. This can be due to the lack of specialist handling, restraining and killing infrastructure and equipment in an on-farm situation. The two phases involved in on-farm killing of pigs are 1) handling (including moving and restraint) and 2) killing. During on-farm killing, some welfare consequences are related to handling and moving of pigs and some others are experienced by pigs during the killing process (Gerritzen & Raj 2009; EFSA 2019; EFSA 2018; Brandt & Aaslyng 2015; Galvin et al. 2005).

The general principles for the welfare of livestock during humane destruction for disease control are detailed in the OIE Terrestrial Animal Health Code (WOAH 2019) and summarised in a review of killing animals for disease control purposes (Thornber, Rubira & Styles 2014). Animal welfare considerations during the selection of methods for mass destruction, based on existing literature and guidelines (Berg 2012; EFSA 2019; AVMA 2020; WOAH 2019; Galvin et al. 2005) include:

- Suitability for animal type - Age, size of animal and influence on effectiveness
- Required competencies - Knowledge and skill requirements
- Handling and restraint - Degree of handling and restraint required for application
- Induction and immediacy - Efficacy of induction of unconsciousness in a mass destruction scenario
- Confirmation of death - Ease of confirming death prior to disposal
- Disease control objectives - Impact of the method on biosecurity and spread of disease
- Animal welfare concerns if applied inappropriately - Specific risk of poor animal welfare outcome with ineffective application.

Process considerations during the selection of methods for mass destruction include (Berg 2012; AVMA 2020; WOAH 2019; Galvin et al. 2005):

- Human safety - Physically safe with low psychological impact for the human operator other bystanders

- Suitability for production system - ease of application and carcass removal after killing
- Availability - Access to equipment, personnel and any additional necessary resources
- Efficiency of the process - Ability to complete the whole process in a timely manner
- Environmental impact - Specific impact of disposal of carcasses and process waste on the environment
- Aesthetics - Acceptability for operators, public (media), community impact
- Cost - Capital and operating costs.

The methods deemed as suitable for the destruction of individual pigs (Part 1) were considered against these objectives and conditions of mass depopulation to identify a group of methods that are not only acceptable in relation to animal welfare but are also appropriate during a large-scale depopulation under different field conditions. The findings are discussed in Sections 4.1 – 4.14. With the exception of air-filled foam, which has not been demonstrated to result in an acceptable animal welfare outcome, there was sufficient evidence in Part 1 for the other methods to be considered further in Part 2. Conditions of use and assessment criteria for the application of the recommended methods under field conditions were also considered, however, the development of detailed methodologies for the application of the recommended methods for large-scale depopulation was not completed as part of this project. It is therefore recommended that this is undertaken in the future.

4.1 Suitability for animal type

A challenge with pig production sites is that there are often different ages of animals on a single site. There are few methods that are suitable for all ages of pig. It is likely that multiple methods may need to be used on multi-age sites.

When using inhalational methods, the mode of action makes the process suitable for all pig types (EFSA Scientific Panel on Animal Health and Welfare et al. 2020b) and this has been demonstrated in a commercial processing environment. However, in a production environment, it may only be practical for pigs that can easily be handled into and removed from containers (American Association of Swine Veterinarians 2016) and more research into the logistics of the operation is required. There is no commercial use of alternative gas mixtures for pigs, although different gases and mixtures have been researched under experimental conditions in a range of sizes of pig, up to 95 kg. Using foam as a delivery method for gas has only been researched in limited studies using pigs, between the weight ranges of 23-32kg and there is currently insufficient information to recommend it as a method for mass depopulation.

The efficacy of sodium nitrite relies on the timely consumption of a toxic dose. On this basis, the use of this method is best suited to animals that are readily consuming feed and have a good appetite. Sodium nitrite is therefore not considered to be a suitable method where adequate feed consumption cannot be assured, for example, sick or very young pigs. Sodium nitrite is also likely to be an effective method for sows and boars due to their appetite and feeding regime, however, there is limited information on this class of pig and additional scientific validation is required.

The use of a firearm (with a free projectile) is suitable for all pig types (EFSA Scientific Panel on Animal Health and Welfare et al. 2020b), though may not be practical for smaller piglets (<5kg). With larger pigs (finishers and adult breeders), a penetrative captive bolt is less likely to result in death and therefore may require the use of a follow-up or terminal procedure. For large boars in particular, production of an effective stun with a penetrative captive bolt device can be difficult and the use of another method should be considered (for example, firearm or intravenous injection). Non-penetrative devices provide a practical alternative for smaller piglets of <10kg (EFSA Scientific Panel on Animal Health and Welfare et al. 2020b; Grist et al. 2017; Grist et al. 2018). It is recommended that a non-penetrating device is used to replace manual blunt force trauma, which should only be used for piglets <5kg and when other more suitable methods are not available (EFSA Scientific Panel on Animal Health and Welfare et al. 2020b).

When appropriate parameters are established and employed, electrical stunning (head-only) has been demonstrated to be effective on all pig types (EFSA Scientific Panel on Animal Health and Welfare et al. 2020b), although design and size of existing electrical stunning tongs may not be appropriate for neonates (Arruda et al. 2020) and other methods (such as non-penetrating captive bolt) would provide a more practical solution. In large sows and boars, the impedance of the head requires the application of higher voltages (>240V) in order to induce an effective stun (EFSA Scientific Panel on Animal Health and Welfare et al. 2020b). If piglets are electrocuted using a head-to-body electrical method, it may be difficult to induce cardiac ventricular fibrillation as the electrical current passes through the body tissues (of lower resistance) rather than through the heart (EFSA Scientific Panel on Animal Health and Welfare 2004; AVMA 2020). For this reason, additional care should be taken to ensure effective electrocution in piglets, or an alternative method should be sought.

On some production sites, pregnant sows will require destruction. The potential welfare issues associated with killing pregnant animals has not been covered in detail in this review, though is covered in detail in an EFSA scientific opinion (EFSA Panel on Animal Health and Welfare et al. 2017). The conclusion was that it is likely that the livestock foetus is not capable of perceiving pain or other negative affect whilst in utero and therefore should be left undisturbed in utero for 30 min after the death of the dam by which time the foetus should also be dead.

4.2 Required competencies

During mass destruction of pigs within a production environment, the activities around handling, restraining, stunning and killing are broadly comparable with the equivalent operations used in by qualified stock-people on-farm, and in an abattoir environment. If necessary, some competent personnel might therefore be sourced from within the slaughter industry, pending their availability. The competencies around general pig handling are similar for all methods used, although the need to handle and restrain individual animals will vary between methods. Skill, attitude and knowledge is essential in performing effective humane killing (EFSA Scientific Panel on Animal Health and Welfare et al. 2020a; EFSA Scientific Panel on Animal Health and Welfare et al. 2020b; Thornber, Rubira & Styles 2014; Campler et al. 2020; Mullins et al. 2017), with some methods requiring more complex skills and understanding (Arruda et al. 2020). For all methods it is essential to ensure competent, responsible and accountable personnel are present to confirm signs of death.

Handling of some chemicals will require specific chemical safety competency or certification and others may require veterinary registration to enable procurement and administration, for example administration of injectable and oral agents (Gavinelli, Kennedy & Simonin 2014; Galvin et al. 2005). Injectable agents usually need to be administered by a vet or under direct veterinary supervision only and require the operator to have knowledge of pig anatomy to successfully administer at the appropriate injection site. The use of inhalational methods will require operators to be able to monitor gas mixtures (and adjust flow rates) and subsequently confirm death in individual animals. When using gas-filled foam, operators will require additional knowledge of foaming equipment and the ability to monitor and adjust foam production rates. Specific competencies required will depend on the complexities of the system and equipment used.

Mechanical methods, such as the use of firearms and captive bolt devices, require operators to possess skills as a marksman and/or the ability to select the correct shot position. There is also a requirement for operators to have a firearms license for all guns that fire a free projectile, with possible additional licensing requirements if using a silencer/suppressor. Specific requirements for firearm ownership and use may differ between jurisdictions. The use of manual blunt force trauma does not require specialised equipment; however, it requires skill and confidence to apply it successfully and repeatably. Individuals need to be able to apply the physical blow accurately and with sufficient force to kill the piglet. Operatives often find it difficult to assess whether the blow has been effective due to the nature of the post-application convulsions (Whiting et al. 2011).

When electrical methods are employed, operatives need to have an understanding of appropriate electrical parameters (for example, the applied electrical current, correct electrode position, pressure and duration of application and the necessary cleaning and maintenance of equipment). As with all the other methods, they must also be able to recognise signs of effective stunning and death.

4.3 Handling and restraint

Destruction activities may involve handling individual animals and sometimes moving them from production pens, sheds and farrowing crates to a central killing point. The duration and nature of handling will affect animal welfare outcomes (Gavinelli, Kennedy & Simonin 2014; Brandt et al. 2013), through the possibility of causing pain and fear. Pain is used to describe an unpleasant sensory and emotional experience associated with, or resembling that associated with, actual or potential tissue damage; whilst fear is an unpleasant emotional state induced by perception of danger or potential danger, that threatens the integrity of the animal (Von Holleben et al. 2010). Handling and restraining pigs in isolation or restricting their physical movement can cause fear (EFSA Scientific Panel on Animal Health and Welfare et al. 2020b) and pigs can be difficult to approach individually (Gavinelli, Kennedy & Simonin 2014). Potential methods of mass destruction can be divided simply into two categories: those that require individual animal handling and those that do not. To reduce the likelihood of pain and fear related to moving and handling, selecting a killing method that requires less handling (and restraint) could be favourable. Animal welfare hazards and animal-based measures associated with the handling and restraint process are discussed in the context of on-farm destruction of pigs in the EFSA review (EFSA Scientific Panel on Animal Health and Welfare et al. 2020b). The need to handle and restrain pigs individually poses significant logistical, time and human resourcing challenges and risks for response personnel. Further, there is potential for pain, fear and distress to be caused to pigs in association with individual restraint and intervention.

The two methods which can be applied to animals in groups are the inhalational methods and the use of oral agents. Inhalational methods require animals to be handled and placed into the unit, however, it does not require individual animal restraint as seen with other methods, such as the use of injectable agents, mechanical methods and manual electrical stunning. Oral agents have the potential to be used in the production environment, with pigs either being killed in the pens or being moved out into a corral as the agent starts to take effect.

When animals need to be handled and restrained individually for IV injection, prior sedation can sometimes make the application of the process easier for the operator, noting that the sedation process represents an additional individual intervention. When mechanical methods are used, it is important that animal movement is controlled to enable accurate targeting. This often requires animals to be restrained individually using a snare, in a race, crush or pen. Firearms (using a free projectile) have been used with minimal restraint, for example, in a field or corral, however, accurate shooting requires a great deal of skill. Unpublished industry information from the US, reported that the use of firearms for killing groups of housed animals resulted in high levels of distress and activity in the pigs, impacting on the efficacy of shots and resulting in panic and piling behaviours.

When manually applied two-step electrical methods are used, it may be possible to stun free-standing pigs confined in a small pen. Electrical stunning whilst pigs are in a group might reduce fear since animals are not individually handled and isolated. Pigs in a group generally do not respond to other pigs being killed (Gerritzen & Raj 2009; DEFRA 2003). For single step electrocution, individual handling and restraint will be required. For the application of hand-held head-to-body electrodes, the movement of animals needs to be restricted in such a way that placement and constant contact of the electrodes is guaranteed. Restraint needs to be sufficient for the electrical current to be applied continuously and long enough to ensure the death of the animal. Group systems that integrate single-step electrocution have been successfully trialled and used for pigs >1 week of age in the EU and US but are not currently commercially available in Australia (Thornber, Rubira & Styles 2014; Grandin 2021).

4.4 Induction and immediacy

By definition, a humane killing process involves a painless and stress-free death (described in detail for each method in Part 1). During mass depopulation it is important that we aim to achieve this ideal (Berg 2012; WOAH 2019; Galvin et al. 2005). Methods of mass destruction for pigs on-farm should cause loss of consciousness followed by death without pain, fear or distress (EFSA Scientific Panel on Animal Health and Welfare et al. 2020b). To induce unconsciousness, the methods should target one or more of the brain regions involved in consciousness; namely the cerebral cortex, thalamus and brainstem (Verhoeven et al. 2015). There are established animal-based indicators to assess the effectiveness of mechanical, gaseous and electrical stunning methods (EFSA Scientific Panel on Animal Health and Welfare 2004; EFSA Scientific Panel on Animal Health and Welfare et al. 2020b). Conditions of mass euthanasia may influence immediacy and induction. Practically, it is difficult to monitor animal-based measures during the use of inhalational agents. Therefore, important parameters such as gas concentration and exposure time should be monitored to ensure sufficient exposure. When using oral agents, pigs need to be actively eating, and in the case of killing diseased animals, the presence of disease may reduce intake of oral agent and therefore reduce effectiveness.

4.5 Confirmation of death

In addition to the requirements of Section 4.4, the effectiveness of any killing process should be able to be monitored at each step, including the confirmation of death before carcass disposal (Berg 2012). The operators should be able to conduct an assessment of the permanent absence of brain stem reflexes to confirm brain death. For some methods, it may be necessary to employ a terminal procedure to ensure death. For all methods, a back-up procedure or second intervention needs to be readily available in the event on unsuccessful application or animals showing signs of recovery.

When inhalational methods are used, pigs can remain in the system until death is produced. With oral agents, time to death is dependent on whether a lethal dose has been ingested. A terminal procedure should be available to kill animals that have not died within the required timeframe.

The use of a penetrative captive bolt will kill a large proportion of animals in a single step, though cannot be relied upon to kill every animal. A limited number of studies have evaluated the efficacy of penetrative captive bolt stunners on-farm and consequently, there is insufficient scientific evidence to support its use a one-step process for any class of pig other than small pigs (piglets and small growers) (EFSA Scientific Panel on Animal Health and Welfare 2004; Dalla Costa et al. 2021). Even then, animals should be checked for signs of incomplete concussion, with a second shot applied if necessary. For mature pigs (finishing pigs, sows and boars), a terminal procedure such as pithing or the intravenous (IV) or intracardiac (IC) injection of a saturated solution of potassium chloride (KCl) can be used to ensure death whenever captive bolt is used. Potassium chloride is cardiotoxic and when administered to unconscious animals by rapid intravenous injection it causes cardiac arrest (EFSA Scientific Panel on Animal Health and Welfare et al. 2020b; AVMA 2020). Further research on the use of penetrative captive bolt devices with higher power loads, alternative shot positions and bolt penetration depth (to result in brain trauma and death) is recommended, particularly for adult sows and boars, where there is limited scientific data to support their use.

When head-only electrical stunning is used to produce unconsciousness, a terminal step must be used to kill the pig. Under mass destruction conditions, when biosecurity is important, a preferred terminal method would be a second application of the electrodes across the chest of the animal to cause cardiac arrest. Successful cardiac arrest needs to be confirmed before disposal. This is indicated by complete and irreversible loss of muscle tone (EFSA 2013) and absence of heartbeat (Vogel et al. 2011). Immediate post-kill handling must be performed carefully, as forceful impacts have been known to resuscitate the heart (EFSA Scientific Panel on Animal Health and Welfare 2004; Galvin et al. 2005).

4.6 Disease control objectives

Movement of animals during an infectious disease outbreak is discouraged and, in many cases, prohibited for biosecurity reasons, therefore methods that allow animals to remain inside the production environment could be preferred in a mass destruction situation. For biosecurity reasons, exsanguination is not normally an option due to the risk of contamination of the site, equipment and infrastructure with potentially infectious material, therefore methods that require bleeding as a terminal procedure are less attractive options. The chosen methods should facilitate carcass (and disease) containment and reduce contact of people with animals and infectious material. Most of the methods have no direct impact on transmission of disease agents, where effective control of biosecurity relies on subsequent disposal methodology. Methods that result in the spillage of body fluids (for example, blood and vomit) could in theory increase the chance of contamination if this is not appropriately managed.

4.7 Animal welfare concerns if applied inappropriately

The primary objective of a selected method is to kill the animals without causing undue pain or distress. The consequences of inappropriate use may be more significant for some methods compared to others. The likelihood and severity of risk associated with inappropriate use has been considered by the EFSA reference group (EFSA Scientific Panel on Animal Health and Welfare et al. 2020b).

When inhalational methods are used, inappropriate flow rates or poorly sealed containers can lead to failure to euthanise animals (gas concentration too low or residual oxygen too high) (EFSA 2019) or animals asphyxiating rather than losing consciousness first (for example when CO_2 gas concentration increased too quickly). A short dwell time in the gas can also lead to animals being unconscious but not dead, and a risk of recovery (EFSA 2019). CO_2 gas is supplied as compressed liquid, and direct rapid application at pig level can result in cold burns to the skin. It is important that inhalational agents are supplied in purified form without contaminants, preferably from a commercially supplied source as this can increase aversion during induction or reduce efficacy. When gas is delivered in a foam, insufficient foam production rates or short dwell times can lead to failure to euthanise animals (insufficient intake of gas). Excessive foam production rates can lead to pigs inhaling the liquid portion and 'drowning'.

The risk of inappropriate application of mechanical methods increases with operator and equipment fatigue and the difficulty of the task (number of animals, environmental conditions and nature of restraint). This increases the chance of non-lethal wounding or animals regaining consciousness before death. Repeated firing of a captive bolt in quick succession will lead to overheating of its barrel and, as consequence, it becomes difficult to hold and apply correctly, or will cease to function effectively. Therefore, captive bolt devices should be allowed to cool periodically, rotated and serviced during use.

When injectable agents are used there is a risk of a non-lethal dose being administered (EFSA 2019) with possible pain associated with injection site. There is also risk of an inappropriate route of administration being used, which can be associated with pain (EFSA 2019). For any method that is applied to animals in a group (for example, inhalational or oral agents) there is a risk that affected animals may climb on and lay down on already recumbent animals, increasing the chance of crushing induced hypoxia. This is unlikely to occur with the use of sodium nitrite as during pen trials pigs continued to display typical behaviours post-consumption until they became recumbent.

When electrical methods are used, incorrect application may increase the chance of pain associated with pre-stun shock or an ineffective stun (associated with incorrect placement of electrodes or insufficient current) (EFSA 2019). The induction of cardiac arrest without unconsciousness is also a risk when electrocution methods are used (EFSA Scientific Panel on Animal Health and Welfare et al. 2020b).

4.8 Operator health and safety

Protecting operator health and safety is an imperative, especially many of the methods for the destruction of pigs are equally lethal to humans. Minimising human-animal interaction is advisable, particularly during zoonotic disease outbreaks, therefore methods which limit contact with animals are preferred. The impact of mass depopulation activities on mental health is also important.

The use of inhalational agents can be particularly hazardous as the gases are colourless and odourless. Specialist ventilation and monitoring are needed to ensure operator safety, both during application and post-application during disposal of carcasses. Visible and audible gas monitors in personnel areas are recommended. The acquisition of injectable agents is usually restricted by law and generally they must only be used by a veterinarian or under direct veterinary supervision. With any injectables, there is a risk of needlestick and self-injection. In field situations, where floors may be slippery, operators should limit the quantity of pentobarbital held in the syringe to avoid a lethal dose if self-injected.

Sodium nitrite has the potential to kill and cause sub-lethal effects in humans. HOGGONE microencapsulated sodium nitrite feral pig bait is registered as a Schedule 6 poison with the APVMA. Schedule 6 refers to substances with a moderate potential for causing harm, the extent of which can be reduced through the use of distinctive packaging with strong warnings and safety directions on the label. An antidote, methylene blue, is available (Shapiro et al. 2016). HOGGONE paste is deemed less toxic to humans and non-target animals than conventional vertebrate pesticides such as 1080 and brodifacoum. HOGGONE paste also contains peanuts and should not be used or handled by people with nut allergies.

Ownership and use of firearms require appropriate licensing. With free bullet there is a risk of ricochets from bone or solid objects, therefore animals should not be shot in enclosed spaces or areas with substantial solid objects (for example, metal gates) (Whiting et al. 2011; Gerritzen & Gibson 2016). One field study examining the use of free bullets to kill piglets, rarely found the bullet in the carcass during postmortem examination, but frequently found flattened bullets in the pen when carcasses were being removed, indicating that the projectile had passed through the body of the target pig, representing a serious safety risk. This is also risk of injury from convulsing animals, particularly if operators are attempting to apply a terminal procedure. Operators must also be carefully managed to avoid heightened risk of injury associated with fatigue. People and animals should therefore be cleared from the immediate area; with possible civil aviation safety authority (CASA) non-fly zone declaration, and police notification if high-calibre firearms are being used near populated areas (Animal Health Australia 2015). The use of mechanical methods, such as firearms and captive bolt pistols in a mass depopulation scenario can also have psychological effects on the operators.

The use of electrical stunning exposes the operator to the risk of electric shock; however, risks are considered to be low with commercially manufactured equipment and appropriate processes.

4.9 Suitability for production system

Differences in farm size, location and housing type will influence the choice of method. In some production systems, it may be necessary to move animals out of the housing to perform the killing process, as removal of large or many dead animals could be difficult. Methods that require individual handling of pigs, for example use of captive bolt, manual electrical stunning or injectable agents, can be used inside or outside the pig housing. The use firearms (free projectile) may have safety implications if used in a confined space, due to the risk of ricochet.

The use of inhalational methods is generally performed outside the pig housing in gas filled containers. Using gases or foams inside housing systems has been researched in poultry (Benson et al. 2007), but appears to be problematic, due to complications around effectively sealing the building.

The use of oral agents is suited to a variety of production systems. Pigs were successfully administered the treatment in their pens, through the normal feeding systems in an Australian study (APL, currently unpublished). Current regulatory requirements stipulate that existing feral pig bait must be delivered using HOGGONE hoppers to prevent consumption by non-target species. Consumption by non-target species would not be a concern for indoor housing systems, though would need to be considered if using the product in outdoor systems. As pigs are not immediately affected by sodium nitrite, there may also be an opportunity to administer the treatment and then move animals out of the production system before the toxin takes effect, to streamline carcass disposal.

4.10 Availability

The availability of equipment and resources will influence how quickly mass destruction can be completed and consequently influence the associated animal welfare outcome. Some methods may require specialist restraint or container systems which may not be readily available in the locality. Methods involving compressed gases or chemicals are limited by the supply of these agents. There may not be sufficiently large volumes of the required agents available in the locality. CO_2 gas is available from bottled-gas suppliers such as BOC. Supplies may be limited and variable. In addition, the use of gas-filled foam requires specialised foam-generating equipment and electricity supply.

Access to injectable agents is restricted and they usually require administration by veterinarian. Therefore, availability depends on access to veterinary services and on the availability at short notice of large quantities from pharmaceutical suppliers.

Microencapsulated sodium nitrite (HOGGONE) is produced exclusively by Animal Control Technologies Australia and is commercially available from rural merchants. Microencapsulation is considered a key factor in supporting palatability and stability, and hence effective lethal outcomes. It is therefore important to differentiate microencapsulated sodium nitrite from the volatile, unpalatable commercial grade granular sodium nitrite, which is not recommended.

The use of firearms requires access to trained marksmen with appropriate licensing. Equipment and ammunition are relatively easy to obtain by licensed personnel but access and approvals to obtain large quantities of both devices and ammunition may be constrained. Recognising the psychological impact of the use of mechanical methods (such as firearms, captive bolt pistols and blunt force trauma), it is necessary to ensure access to a sufficient number of personnel to avoid fatigue and reduce the emotional impact.

4.11 Efficiency of the process

Methods for mass depopulation should result in the death of a large population of animals in a quick and effective manner (Animal Health Australia 2015; Gerritzen & Raj 2009; Thornber, Rubira & Styles 2014; AVMA 2020). Delays in the time taken to complete the process can have numerous welfare implications. For example, in diseased animals, delays can lead to further suffering as the disease progresses. The overall efficiency of the process will be determined by:

- the time required to handle and restrain pigs for humane killing
- the ability to kill pigs in groups using the selected method
- the time needed to produce and confirm death after administration of the method, and
- the time needed to retrieve and dispose of carcasses.

The outcome of the methods can be stunning or killing. If the method only stuns the animal, then a second (or terminal) step is required. From an efficiency perspective, single step methods are generally more attractive, however, some single step methods can be relatively more time or labour intensive. The use of an injectable method would be a slow process for large numbers of animals, possibly requiring multiple operators. If the quantity of solution held in the syringe is restricted for operator safety, then syringes would also need to be regularly refilled.

Methods that can be applied to groups of animals, avoiding individual restraint and handling are also generally more efficient. However, if this involves killing pigs inside the housing system this may increase the time required to remove and dispose of carcasses.

When sodium nitrite is used, introduction of the baited feed may need to occur after pigs have become accustomed to the change in feed substrate. Pigs must also be sufficiently hungry to consume feed offered quickly. Depending on pig class this may necessitate a feed adjustment in the days preceding delivery of the oral agent. For example, grower pigs with unrestricted access to feed may require a period of time on maintenance rations to modify feeding behaviours, such that they eat sufficient volume rapidly upon delivery. In contrast sows that are already being fed once daily would not require this transition as they quickly eat feed when it is offered. Pigs can also be neophobic (avoiding novel objects and experiences), so it may take time for them to actively feed from a novel delivery system (bait box) in outdoor systems.

Killing large numbers of animals with mechanical devices (firearms and captive bolts) is likely to be a protracted process, requiring multiple operators to restrain, kill and confirm death, and to manage fatigue. Repeated firing of captive bolts in quick succession will lead to overheating and failure of the device (Gibson et al. 2015), therefore there must be a sufficient number of devices to allow rotation.

The efficiency of electrical methods will depend on the design and complexity of the system. Group killing units may result in a more efficient process, though will still require animals to be handled and moved into the system. A manual two-step method (involving electrode application to the head followed by the heart) can achieve rates of around 30-60 pigs per hour (EFSA Scientific Panel on Animal Health and Welfare 2004) with a single operator.

4.12 Environmental impact

The environmental impact of chemicals must be considered in terms of the final disposal method on a locality basis.

With inhalational methods, gases are vented to the atmosphere, and the long-term environmental consequences of releasing concentrated quantities of each of the suggested gases into the atmosphere is not known. Foams are generated using soap or detergent, which can lead to environmental contamination.

Injectable barbiturates can persist in animal carcasses. When carcasses have been insufficiently buried or left uncovered, these can cause secondary toxicosis (sedation and death) in animals that consume the remains. Unlike other poisons, the residues of sodium nitrite in carcasses pose no risk to scavengers as the compound degrades quickly leaving low levels in the edible tissues. Measures are taken to avoid consumption by non-target species when HOGGONE is used for feral pig control. This includes the use of customised delivery hoppers that non-target species cannot open.

4.13 Aesthetics

Killing of any animal can be confronting, particularly when on a large scale. Particular aspects that are aesthetically challenging are behavioural responses such as vocalisation, escape attempts and gasping, and visible blood or carcass damage. Methods performed outside the pig housing may, if possible, need to be screened from onlookers, including from the air (drones). Mechanical killing methods are often considered to be aesthetically displeasing as they generally involve extensive physical trauma to the head of the animal (AVMA 2020). The use of firearms can also be noisy, which may also attract public attention. Mechanical methods often result in the pig displaying physical convulsions (even after death), which can be challenging psychologically for operators and observers. The application of blunt force trauma, in particular, is aesthetically unpleasant for both operators and wider society, being generally unacceptable to the general public (Dalla Costa et al. 2021).

The use of CO_2 causes signs of respiratory distress and escape behaviour in pigs, which can be confronting to witness. However, the use of containers to hold the pigs means that this type of activity is not usually witnessed by onlookers.

When sodium nitrite is used, conclusions from pen trials described signs of toxicity and death to be unremarkable. The attending veterinarian stated that sodium nitrite caused a rapid death with little distress evident from the pigs (Shapiro et al. 2016).

4.14 Cost

A detailed costing of each method is beyond the scope of this project, as many considerations influence the final cost per pig (for example, availability of chemical agents and gases can fluctuate; location of the farm relative to suppliers of gases/chemicals/equipment; size of operation; class of pigs; labour requirements).

For inhalational methods, defining a price per pig is challenging, and requires a case-by case approach. Cost components include but are not limited to: Number of pigs processed per container; size of airspace; quality of seals, which impacts the flow rates needed to maintain the gas concentration for the required duration; source of the gas (whether supplied in compressed-gas cylinders or in a bulk tanker). Inert gases may also be more expensive than CO_2. When using foam application, the cost components are similar, with the added cost of foam generating equipment.

The use of injectable agents potentially has the highest cost per pig, associated with veterinary involvement and drug costs (EFSA 2018). For example, barbiturate for euthanasia (Lethabarb - 325 mg/mL solution of pentobarbitone) administered at 1ml/2kg body weight currently costs approximately $150 for 450ml. Sodium nitrite (HOGGONE feral pig bait) costs approximately $5 per pig (information from commercial product developers).

For mechanical methods, the cost of the firearm or captive bolt device, licensing, servicing, ammunition and suitably skilled operators need to be considered. Captive bolt pistols are available at a range of prices between $700 for a basic model and $5000 for a full euthanasia kit, with cartridge costs around 40-50 cents per cartridge depending on manufacturer, shipping costs etc.

4.15 Recommendations

When the decision has been made to humanely kill an animal, the method employed should result in the rapid loss of consciousness (or induce unconsciousness without pain, fear and distress) followed by cardiac or respiratory arrest (and ultimately the loss of brain function). In addition, animal handling and restraint should aim to minimise any pain, fear and distress experienced by the animal prior to unconsciousness.

When considering methods for mass depopulation, it is important to remember that we are dealing with an atypical situation, where our ideal choice of method may not be available or may be affected by the conditions in which it is being used. Achieving an acceptable animal welfare outcome is a multi-faceted challenge, particularly when trying to balance with safety of personnel, biosecurity and environment requirements and production conditions.

The ideal method for the mass destruction of pigs would allow for large numbers of animals to be killed in-situ in their production system, without individual handling; whilst still resulting in an acceptable animal welfare outcome. Unfortunately, there are few methods that can currently be successfully employed in a production environment to kill pigs in groups; though there is potential to further develop inhalational and oral agents for this purpose. The following sections 4.15.1 - 4.15.5 summarise the findings of Part 1 and 2, outlining the advantages and drawbacks of potential methods. This section also provides recommendations for changes to the AUSVETPLAN Operational manual. Section 4.15.6 provides an overall summary of the recommended methods.

4.15.1 Inhalational agents

Inhalational agents have a welfare advantage in that the animals can be treated in a group and individual restraint is not required, however there are welfare concerns around the delayed onset of unconsciousness, aversion to CO_2 and risk of return to consciousness. Loss of consciousness is more rapid if pigs are exposed to a high concentration of CO_2 from the outset. However, it is known that exposure to high concentrations (defined as higher than 80% by volume) can be aversive and distressing and is considered a serious welfare concern by the EFSA Panel. Despite this, if parameters and delivery methods could be effectively controlled on-farm, using a high concentration of CO_2 would deliver a welfare outcome equivalent to that achieved in a commercial controlled atmosphere stunning (CAS) system in a pig processing plant. Experimentally, the exposure to inert gases is less aversive as it causes less pain, fear and respiratory distress compared with gas mixtures containing carbon dioxide at high concentrations, however, there is insufficient information regarding practicalities during mass depopulation events.

In a production environment, the use of containers to hold CO_2 potentially limits its application to younger or smaller pigs due to constraints in processing volume, and the need to repeatedly load and empty the container which incurs both logistical and operator safety considerations. This is reflected in many of the recommendations for using CO_2 under mass depopulation conditions, which place limits on pig size. It is recommended that the AUSVETPLAN be amended to allow for the use of CO_2 in all sizes of animal, provided that equivalent conditions to those used during the commercial processing of pigs can be achieved. It is further recommended that work continues to identify suitable alternatives to CO_2. To-date, a variety of alternative gas mixtures, multi-stage processes (using rising concentrations of CO_2) and use of gas-filled foams have been investigated in a research context, but no clear optimal process for practical application has emerged. The conclusions drawn in a 2020 review (Arruda et al. 2020) therefore remain relevant. To quote:

> The use of CO_2 is the commonly reported method in literature, with higher concentrations causing faster unconsciousness at the expense of compromised welfare expressed by the provocation of air hunger, anxiety and fear. Despite research over three decades, a safe and reliable way to induce rapid unconsciousness and death in larger populations of swine appear to have not been found [sic].

4.15.2 Oral agent – sodium nitrite

The use of the oral agent sodium nitrite is a relatively undeveloped method of mass destruction for domestic pigs, despite showing promising results in the control of feral pigs and in limited published unpublished trials on domestic pigs. Using baited feed could potentially allow for large numbers of pigs to be euthanised in the production system, without the need for individual handling of live animals, which is an important welfare advantage over many of the other available methods. Presently, the use of oral agents is not recognised under AUSVETPLAN, which states: ‘there is no justification for using a poison on managed stock’. The ESFA report on methods for the destruction of pigs (EFSA Scientific Panel on Animal Health and Welfare et al. 2020b) also states that methods which involve the administration of toxic substances to feed or water, or injection of chemicals unauthorised for killing pigs must not be used. However, it is apparent that the mode of action of sodium nitrite is very different to that seen with other feral pig products and results in an acceptable animal welfare outcome (Section 3.3). There is currently insufficient published information on the practical use of this agent in domestic pigs, for example, the quantity of agent required to successfully kill the majority of pigs in a population, induction of unconsciousness and time to death. It is therefore important that the results of the trials on domestic pigs are published expediently. There should also be further research on the use of sodium nitrite in different classes of pig, including sows and boars. It should also be noted that microencapsulation appears to be key to the efficacy of this product as the unprotected granular product is unstable with poor palatability. Presently in Australia, there is only one commercial manufacturer of the microencapsulated product, which could affect access to an adequate supply during a mass depopulation situation. Extrapolating the results from feral pigs, although very useful, should be done with caution. It is therefore recommended that the approval of this method for mass depopulation be reconsidered after the publication and review of the recent scientific studies on domestic pigs.

4.15.3 Injectable agents

For the killing methods that require animals to be handled individually, the use of an injectable agent (administered intravenously) provides a humane option, resulting in a short time to death. However, the use of injectable agents for all animals in a mass depopulation event is not practical, therefore it should be reserved for use as a back-up or terminal procedure or for small groups of animals (for example, unweaned piglets). If trained personnel are available, the use of injectable agents can deliver consistently good animal welfare outcomes, though it is likely in many situations this will need to be combined with prior sedation to facilitate administration of the IV injection. The agents used are regulated substances that can only be administered under the authority of a registered veterinarian, which may influence availability and accessibility. It is recommended that the AUSVETPLAN is revised to allow for the use of barbiturate overdose delivered IV in conscious animals. An alternative route, such as Intracardiac (IC) or intraperitoneal (IP) may be permitted in unconscious animals (as a terminal procedure). Carcass disposal also needs to be carefully controlled to prevent potential poisoning of non-target animals. Disposal of carcases from animals killed with injectable preparations is becoming increasingly problematic, particularly as many rendering facilities will no longer accept animals contaminated with certain chemical residues. The use of injectable agents is the likely to be the most expensive method due to the veterinary involvement and the cost of the agents.

4.15.4 Mechanical methods

The use of firearms (free projectile) represents a humane method of destruction for all pig types, though practical application may not be suited to small piglets. The equipment and ammunition required is relatively easy to access and is not regarded as specialised equipment, though will require personnel to possess a firearms license. The health and safety aspects around the use of firearms is a significant concern.

Penetrative captive bolts are potential methods for the humane destruction of some classes of pig, however there are recognised difficulties associated with the anatomy of the pig head. To improve this method for pigs, further work needs to undertaken to evaluate the use of higher power loads and extended bolts (99), particularly if penetrative captive bolts are to be recommended as a single-step method or when used for adult sows and boars. Currently, to ensure a humane death, a terminal procedure is recommended when penetrative captive bolt pistols are used. Future research is required to further understand the influence of pig type (such as sex, weight and skull shape) on both the effectiveness of the device and the response in the animal following application. Penetrative captive bolt may be effective as a single step method in smaller pigs, but the upper weight limit for this application is unclear from published data.

It is recommended that the AUSVETPLAN be amended to allow for non-penetrating devices to be permitted to be used for piglets <10kg, particularly as an alternative to manual blunt force trauma.

4.15.5 Electrical methods

Electrical methods have the advantage of being suited to most pig types and can be used effectively in a range of production systems. They are non-invasive methods with fewer associated biosecurity concerns compared to other methods of humane destruction. Currently, electrical methods require animals to be handled and individually restrained, however, a mobile electrocution unit to facilitate killing as an automated process has been used in Europe and the US (Thornber, Rubira & Styles 2014;

Grandin 2021). There was no available literature on the specific parameters and resulting efficacy of this particular equipment, however the development and scientific verification of this type of portable electrical system warrants further research. Adult sows and boars may require specialised equipment (if conventional equipment is too small to apply across the chest).

4.15.6 Summary of preferred methods for mass depopulation

Under the conditions of mass depopulation, the following methods could deliver acceptable animal welfare outcomes using appropriate operational parameters (Table 6).

Table 6 Methods considered to currently be the most appropriate under conditions of mass depopulation

Method		Pig Type	Mass destruction conditions
Inhalational agent	Carbon dioxide	All	Requires system parameters that can produce an outcome equivalent to that achieved commercially
Oral agent	Sodium nitrite	Grower/finisher	Pigs must be feeding well and not affected by disease
Injectable agent	Barbiturate administered intravenously	All	Requires individual handling. Useful as a terminal procedure or as a back-up process to other methods. May be difficult to administer intravenously in small piglets. In some cases, pigs may require prior sedation for administration.
Mechanical methods	Firearms (free projectile)	All	Likely to require individual handling. Consider psychological impact on operators, particularly when killing large numbers. Using firearms for groups of unrestrained animals must be carefully considered as it can present both animal welfare and safety risks. May be practically difficult with small piglets, therefore consider for animals post-weaning. Useful as a terminal or back-up method.
	Penetrating captive bolt	Piglets, grower/finisher	Requires individual handling. An additional terminal procedure recommended, particularly when killing large numbers and opportunities to confirm death may be compromised. Useful as a back-up to oral and inhalational agents.
	Non-penetrating captive bolt	Piglets <10kg	Requires individual handling. Single-step method when appropriate equipment used. May only produce unconsciousness in larger piglets, requiring a second-step terminal procedure.
Electrical methods	Electrocution – one step	Grower/finisher	Manually applied, with some semi-automated systems being developed.
	Electrocution – two step	Grower/finisher, adult sows/boars	Requires individual handling. May require specialised equipment for sows and boars due to their size.

Based on a welfare assessment, in the context of mass depopulation, the following methods are considered suitable, however, they have characteristics which may preclude their use (or there is currently insufficient data to support their use) under conditions of mass depopulation. They remain potential options for use on small numbers of pigs or when preferred methods are not available:

- Manual blunt force trauma for piglets <5kg
- Penetrative captive bolt followed by a terminal procedure for adult sows and boars
- Carbon dioxide and inert gas for all classes of pig
- Nitrogen or inert gases for all classes of pig
- Head-only electrical stunning followed by bleeding for all classes of pig
- Electrocution for piglets and adult sows and boars
- Gas-filled (N_2) foam for piglets
- Sodium nitrite for adult sows and boars (likely to be suitable for breeding stock due to requirement for a good appetite, however validation data not currently available to confirm).

The following methods cannot currently be recommended for killing pigs under conditions of mass destruction, due to the absence of supporting science:

- Non-penetrative captive bolt for larger classes of pig (grower/finisher/adult sows and boars)
- Air-filled foam for all classes of pig
- Gas-filled foam larger classes of pig.

Preferred terminal procedures include:

- Injectable agents, for example, barbiturate or KCl administered intravenously (IV) or intracardiac (IC) in animals confirmed to be unconscious
- Pithing (only suited to methods that result in penetration of the skull such as firearms or penetrative captive bolt).

5 Conclusion

Current readily available equipment allows for mass humane destruction, however, it is clear that all methods investigated have advantages and disadvantages. Selection of the most appropriate method(s) for use in any particular situation will require a case-by-case evaluation of aspects such as animal type, availability of the required equipment, operational capabilities, environmental and disposal considerations and personnel safety.

The majority of studies describe the results of controlled experiments under laboratory conditions, using small sample sizes and assuming scalable process. There was also a range of research methodologies, methods used for assessment of unconsciousness and a degree of subjectivity around the determination of death; with fewer studies focusing on the induction period and the time to loss of consciousness. Studies that did focus on the induction period tended to be carried out in the proposed context of commercial slaughter processing for human consumption as opposed to on-farm killing or mass depopulation. There is value in validating the use of the methods in semi-commercial field studies. Many of the studies suggest that competent and conscientious personnel are the single most important factor in assuring a humane death. Risk of failure is likely to increase with operator fatigue and time pressures, therefore methods that remove the need for individual handling of animals need further research. The assessment of death can be difficult in a practical on-farm situation and protocols need to be developed for selected methods.

6 References

American Association of Swine Veterinarians, 2016, On-farm euthanasia of swine – recommendations for the producer, *National Pork Board*, Des Moines, Iowa, United States of America

American Veterinary Medical Association (AVMA) 2020, AVMA Guidelines for the Euthanasia of Animals: 2020 edition, *AVMA*, Illinois

Animal Health Australia (AHA) 2015, Operational manual: Destruction of animals (Version 3.2), Australian Veterinary Emergency Plan (AUSVETPLAN), Canberra

Arruda, AG, Beyene, TJ, Kieffer, J, Lorbach, JN, Moeller, S & Bowman, AS 2020, A systematic literature review on depopulation methods for swine, *Animals (Basel),* vol. 10, no. 11, pp. 2161 – 2180, DOI: 10.3390/ani10112161

Atkinson, S, Algers, B, Pallisera, J, Velarde, A & Llonch, P 2020, Animal welfare and meat quality assessment in gas stunning during commercial slaughter of pigs using hypercapnic-hypoxia (20% CO_2 2% O_2) compared to acute hypercapnia (90% CO_2 in air), *Animals (Basel)*, vol. 10, no. 12, pp. 2440-2456, DOI: 10.3390/ani10122440

Benson, E, Malone, GW, Alphin, RL, Dawson, MD, Pope, CR & Van Wicklen, GL 2007, Foam-based mass emergency depopulation of floor-reared meat-type poultry operations, *Poultry Science*, vol. 86, no. 2, pp. 219-224, DOI: 10.1093/ps/86.2.219

Benson, ER, Alphin, RL, Dawson, MD & Maline, GW 2009, Use of water-based foam to depopulate ducks and other species, *Poultry Science*, vol. 88, no. 5, pp. 904-901, DOI: 10.3382/ps.2008-00268

Benson, ER, Weiher, JA, Alphin, RL, Farnell, M & Hougentogler, DP 2018, Evaluation of two compressed air foam systems for culling caged layer hens, *Animals*, vol. 8, no. 5, pp. 61-73, DOI: 10.3390/ani8050061

Berg, C 2012, The need for monitoring farm animal welfare during mass killing for disease eradication purposes, *Animal Welfare*, vol. 21, no. 3, pp. 357–361. DOI:10.7120/09627286.21.3.357

Beusekom, EV, Weimer, DJ, Wellen, C, Daniels, CS 2001, Euthanasia by electrocution in 5kg to 15kg nursery pigs. Evidence-based practice: how do we get there? *American Association of Swine Veterinarians - 42nd Annual Meeting Proceedings*, pp. 341-342, Phoenix, Arizona, United States of America

Blackmore, DK, Bowling, MC, Madie, P, Nutman, A, Barnes, GR, Davies, AS, Donoghue, M & Kirk, EJ 1995, The use of a shotgun for the emergency slaughter or euthanasia of large mature pigs, *New Zealand Veterinary Journal*, vol. 43, no. 4, pp. 134-137, DOI: 10.1080/00480169.1995.35872

Brandt, P, Rousing, T, Herskin, M & Aaslyng, MD 2013, Identification of post-mortem indicators of welfare of finishing pigs on the day of slaughter, Livestock Science, vol. 157, no. 2-3, pp. 535-544, DOI: 10.1016/j.livsci.2013.08.020

Brandt, P & Aaslyng, MD 2015, Welfare measurements of finishing pigs on the day of slaughter: a review, *Meat Science*, vol. 103, pp. 13-23, DOI: 10.1016/j.meatsci.2014.12.004

Campler, MR, Pairis-Garcia, M, Rault, JL, Coleman, G & Arruda, A 2020, Interactive euthanasia training program for swine caretakers; a study on program implementation and perceived caretaker knowledge, *Journal of Swine Health and Production*, vol. 28, no. 5, pp.258-264, DOI: 10.54846/jshap/1173

Casey-Trott, TM, Millman, ST, Turner, PV, Nykamp, SG & Widowski, TM 2013, Effectiveness of a non-penetrating captive bolt for euthanasia of piglets less than 3d of age, *Journal of Animal Science*, vol. 91, no. 11, pp. 5477-5484, DOI: 10.2527/jas.2013-6320

Casey-Trott, TM, Millman, ST, Turner, PV, Nykamp, SG, Lawlis, PC & Widowski, TM 2014, Effectiveness of a non-penetrating captive bolt for euthanasia of 3kg to 9kg pigs, *Journal of Animal Science*, vol. 92, no. 11, pp. 5166-5174, DOI: 10.2527/jas.2014-7980

Caputo, MP, Benson, ER, Pritchett, EM, Hougentogler, DP, Jain, P, Patil, C, Johnson,AL & Alphin, RL 2012, Comparison of water-based foam and carbon dioxide gas mass emergency depopulation of White Pekin ducks, *Poultry Science,* vol. 91, no. 12, pp. 3057-3064, DOI: 10.3382/ps.2012-02514

Cavusoglu, E, Rault, JL, Gates, R & Lay, DC Jr 2020, Behavioural response of weaned pigs during gas euthanasia with CO_2, CO_2 with butorphanol or nitrous oxide, *Animals (Basel)*, vol. 10, no. 5, DOI: 10.3390/ani10050787

Clark, M & Brunick, A 2003, 'N_2O and its interaction with the body' in *Handbook of Nitrous Oxide and Oxygen Sedation - 4th Edition,* Elsevier Mosby, St. Louis, Missouri

Cowled, BD, Elsworth, P & Lapidge, S 2008, Additional toxins for feral pig (Sus scrofa) control: identifying and testing Achilles' heels, *Wildlife Research*, vol. 35, no. 7, pp. 651-662, DOI: 10.1071/WR07072

Dalla Costa, FA, Gibson, TJ, Oliveira, SEO, Gregory, NG, Coldebella, A, Faucitano, L & Dalla Costa OA 2019, On-farm pig dispatch methods and stockpeople attitudes on their use, Livestock Science, vol. 221, pp. 1-5, DOI: 10.1016/j.livsci.2019.01.007

Dalla Costa, FA, Gibson, TJ, Oliveira, SEO, Gregory, NG, Coldebella, A, Faucitano, L, Ludtke, CB, Buss, LP & Dalla Costa, OA 2020, Evaluation of physical euthanasia for neonatal piglets on-farm, *Journal of Animal Science*, vol. 98, no. 7, DOI: 10.1093/jas/skaa204

Dalla Costa, FA, Gibson, TJ, Oliveira, SEO, Gregory, NG, Faucitano, L & Dalla Costa OA 2021, On-farm culling methods used for pigs, *Animal Welfare*, vol. 30, no. 4, pp. 507-522, DOI: 10.7120/09627286.30.3.008

Department for Environment, Food & Rural Affairs (DEFRA) 2003, Getting it right. first time, every time. Practical guide on the management and operation of large scale humane killing of livestock during emergencies, *Countrywise Communications*

European Food Safety Authority (EFSA) Scientific Panel on Animal Health and Welfare 2004, Opinion of the Scientific Panel on Animal Health and Welfare on a request from the Commission related to welfare aspects of the main systems of stunning and killing the main commercial species of animals, *EFSA Journal,* vol. 2, no. 7, pp. 1-29, DOI: 10.2903/j.efsa.2004.45

European Food Safety Authority (EFSA) Scientific Panel on Animal Health and Welfare 2006, Opinion of the Scientific Panel on Animal Health and Welfare (AHAW) on a request from the Commission related with the welfare aspects of the main systems of stunning and killing applied to commercially farmed deer, goats, rabbits, ostriches, ducks, geese, *EFSA Journal*, vol. 4, no. 3, pp. 326 – 415, DOI: 10.2903/j.efsa.2006.326

European Food Safety Authority (EFSA) Panel on Animal Health and Welfare 2013, Scientific opinion on monitoring procedures at slaughterhouses for pigs, EFSA Journal, vol. 11, no. 12, DOI: 10.2903/j.efsa.2013.3523

European Food Safety Authority (EFSA) Panel on Animal Health and Animal Welfare, More, S, Bicout, D, Botner, A, Butterworth, A, Calistri, P, Depner, K, Edwards, S, Garin-Bastuji, B, Good, M, Gortazar Schmidt, C, Michel, V, Miranda, MA, Saxmose Nielsen, S, Velarde, A, Thulke, H-H, Sihvonen, L, Spoolder, H, Stegeman, JA, Raj, M, Willeberg, P, Candiani, D & Winckler, C 2017, Scientific opinion on the animal welfare aspects in respect of the slaughter or killing of pregnant livestock animals (cattle, pigs, sheep, goats, horses), *EFSA Journal*, vol. 15, no. 5, DOI: 10.2903/j.efsa.2017.4782

European Food Safety Authority (EFSA) 2018, Identification of elements for welfare risk assessment related to on-farm killing of animals, *EFSA Supporting Publications*, vol. 15, no. 8, pp. 1-32, DOI: 10.2903/sp.efsa.2018.EN-1453

European Food Safety Authority (EFSA) 2019, Hazard identification for pigs at slaughter and during on-farm killing, EFSA Supporting Publication 2019:EN-1684, vol. 16, no. 7, pp. 1-10, DOI: 10.2903/sp.efsa.2019.EN-1684

European Food Safety Authority (EFSA) Scientific Panel on Animal Health and Welfare, Nielsen, SS, Alvarez, J, Bicout, DJ, Calistri, P, Depner, K, Drewe, JA, Garin-Bastuji, B, Gonzales Rojas, JL, Gortázar Schmidt, C, Michel, V, Miranda Chueca, MÁ, Roberts, HC, Sihvonen, LH, Spoolder, H, Stahl, K, Viltrop, A, Winckler, C, Candiani, D, Fabris, C, Van der Stede, Y & Velarde, A 2020a, Scientific opinion on the welfare of pigs at slaughter, *EFSA Panel,* vol. 18, no. 6, pp. 1 -113, DOI: 10.2903/j.efsa.2020.6148

European Food Safety Authority (EFSA) Scientific Panel on Animal Health and Welfare, Nielsen, SS, Alvarez, J, Bicout, DJ, Calistri, P, Depner, K, Drewe, JA, Garin-Bastuji, B, Gonzales Rojas, JL, Gortázar Schmidt, C, Michel, V, Miranda Chueca, MÁ, Roberts, HC, Sihvonen, LH, Spoolder, H, Stahl, K, Viltrop, A, Winckler, C, Candiani, D, Fabris, C, Van der Stede, Y & Velarde, A, 2020b, Scientific Opinion on the welfare of pigs during killing for purposes other than slaughter, *EFSA Journal*, vol. 18, no. 7, pp. 1-72, DOI: 10.2903/j.efsa.2020.6195

Farm Animal Welfare Committee 2017, Opinion on the welfare of animals killed on-farm, *Farm Animal Welfare Committee*, London, England

Fiedler, KJ, Parsons, RL, Sadler, LJ & Millman, ST 2014, Effects of stocking rate on measures of efficacy and welfare during carbon dioxide gas euthanasia of young pigs, *Animal Welfare*, vol. 23, no. 3, pp. 309-321, DOI: 10.7120/09627286.23.3.309

Finnie, JW, Manavis, J, Summersides, GE & Blumbergs PC 2003, Brain damage in pigs produced by impact with a non-penetrating captive bolt pistol, *Australian Veterinary Journal*, vol. 81, no. 3, pp. 153-155, DOI: 10.1111/j.1751-0813.2003.tb11078

Galvin, JW, Blokhuis, H, Chimbombi, MC, Jong, D & Wotton, S 2005, Killing of animals for disease control purposes, *Revue scientifique et technique (International Office of Epizootics)*, vol. 24, no. 2, pp. 711-722, DOI: 10.20506/rst.24.2.1603

Gavinelli, A, Kennedy, TP & Simonin, D 2014, The application of humane slaughterhouse practices to large-scale culling, *Scientific and Technical Review,* vol. 33, no. 1, pp. 291-301, DOI: 10.20506/rst.33.1.2280

Gerritzen, MA & Raj, MAB 2009, 'Animal welfare and killing for disease control', in Welfare of production animals: assessment and management of risks, Wageningen Academic Publishers, Wageningen, Netherlands

Gerritzen, MA, Reimert, HGM, Hindle, VA, McKeegan, DEF & Sparrey, J 2010, Welfare assessment of gas-filled foam as an agent for killing poultry, Wageningen UR Livestock Research, Wageningen

Gerritzen, M, Lourens, S, Reimert, H, Gunnink, H, von Holleben, K, von Wenzlawowicz, M, Verhoeven, M & Eser, E 2012, Emergency killing of pigs in a carbon dioxide-nitrogen mixture, Wageningen UR Livestock Research, Lelystad, DOI: 10.13140/RG.2.1.1643.7925

Gerritzen, M & Gibson, TJ 2016, 'Animal welfare at depopulation strategies during disease control actions', in Animal welfare at slaughter, *5M Publishing Ltd.*, Essex, England

Gibson, TJ, Mason, CW, Spence, JY, Barker, H & Gregory, NG 2015, Factors affecting penetrating captive bolt gun performance, *Journal of Applied Animal Welfare Science*, vol. 18, no. 3, pp. 222-238, DOI: 10.1080/10888705.2014.980579

Giliam, JN, Shearer, JK, Bahr, RJ, Crochik, S, Woods, J, Hill, J, Reynolds, J & Taylor, JD 2016, Evaluation of brainstem disruption following penetrating captive bolt shot in isolated cattle heads: comparison of traditional and alternative shot placement landmarks, *Animal Welfare*, vol. 25, no. 3, pp. 347-353, DOI: 10.7120/09627286.25.3.347

Grandin, T 2021, Methods to prevent future severe animal welfare problems caused by COVID-19 in the pork industry, *Animals (Basel)*, vol. 11, no. 3, DOI: 10.3390/ani11030830

Grist, A, Murrell, JC, McKinstry, JL, Knowles, TG & Wotton, SB 2017, Humane euthanasia of neonates I: validation of the effectiveness of the Zephyr EXL non-penetrating captive-bolt euthanasia system on neonate piglets up to 10.9kg live-weight, *Animal Welfare*, vol. 26, no. 1, pp. 111-120, DOI: 10.7120/09627286.26.1.111

Grist, A, Knowles, TG & Wotton, SB 2018, Humane euthanasia of neonates II: field study of the effectiveness of the Zephyr EXL non-penetrating captive-bolt system for euthanasia of newborn piglets, *Animal Welfare*, vol. 27, no. 4, pp. 319–326, DOI:10.7120/09627286.27.4.319

Grist, A, Lines, JA, Knowles, TG, Mason, CW & Wotton, SB 2018, The use of a non-penetrating captive bolt for the euthanasia of neonate piglets, *Animals (Basel)*, vol. 8, no. 4, DOI: 10.3390/ani8040048

Grist, A, Bock, R, Knowles, TG, Lines, JA & Wotton, SB 2019, An examination of the performance of blank cartridges used in captive bolt devices for the pre-slaughter stunning and euthanasia of animals, *Animals (Basel)*, vol. 9, no. 8, DOI: 10.3390/ani9080552

Grist, A, Bock, R, Knowles, TG & Wotton, SB 2020, Further examination of the performance of blank cartridges used in captive bolt devices for the pre-slaughter stunning of animals, *Animals (Basel)*, vol. 10, no. 11, DOI: 10.3390/ani10112146

Gurung, S, Hoffman, J, Stringfellow, K, Abi-Ghanem, D, Zhao, D, Caldwell, D, Lee, J, Styles, D, Berghman, L, Byrd, J, Farnell, Y, Archer, G & Farnell, M 2018, Depopulation of caged layer hens with a compressed air foam system, *Animals (Basel)*, vol. 8, no. 1, DOI: 10.3390/ani8010011

Humane Slaughter Association 2013, Captive bolt stunning of livestock, *Humane Slaughter Association*, Wheathampstead, United Kingdom

Humane Slaughter Association 2016, Humane killing of livestock using firearms, *Humane Slaughter Association*, Wheathampstead, United Kingdom

Humane Slaughter Association 2017, On-farm killing for disease control purposes, *Humane Slaughter Association*, Wheathampstead, United Kingdom

Humphrys, S 2017, Development and delivery of a new feral pig toxin/HOGGONE®, *Meat and Livestock Australia Limited*, North Sydney

Husheer, J, Luepke, M, Dziallas, P, Waldmann, KH & von Altrock A 2020, Electrocution as an alternative euthanasia method to blunt force trauma to the head followed by exsanguination for non-viable piglets, *Acta veterinaria Scandinavica*, vol. 62, no. 1, DOI: 10.1186/s13028-020-00565-9

Jacobson, J, Schulz, L & White, R 2011, Penetrating vs. non-penetrating captive bolt euthanasia in nursery pigs. Evidence-based practice: how do we get there? *American Association of Swine Veterinarians - 42nd Annual Meeting Proceedings*, Phoenix, Arizona, United States of America, pp. 313-144

Kells, N, Beausoleil, N, Johnson, C & Sutherland, M 2018, Evaluation of different gases and gas combinations for on-farm euthanasia of pre-weaned pigs, *Animals,* vol. 8, no. 3, pp. 40 - 55, DOI: 10.3390/ani8030040

Kells, NJ, Beausoleil, NJ, Sutherland, MA & Johnson, CB 2018, Electroencephalographic responses of anaesthetised pigs to intraperitoneal injection of sodium pentobarbital, *Animal Welfare*, vol. 27, no. 3, pp. 205-214, DOI: 10.7120/09627286.27.3.205

Kramer, SA, Wagner, BK, Robles, I, Moeller, SJ, Bowman, AS, Kieffer, JD, Arruda, AG, Cressman, MD & Pairis-Garcia, MD 2021, Validating the effectiveness of alternative euthanasia techniques using penetrating captive bolt guns in mature swine (Sus scrofa domesticus), *Journal of Animal Science*, vol. 99, no. 3, DOI: 10.1093/jas/skab052

Lambooij, B & Algers, B 2016, Mechanical stunning and killing methods, in 'Animal welfare at slaughter, *5M Publishing Ltd.*, Essex, England

Lindahl, C, Sindhoj, E, Hellgren, RB, Berg, C & Wallenbeck, A 2020, Responses of pigs to stunning with nitrogen filled high-expansion foam, *Animals (Basel)*, vol. 10, no. 12, pp. 2210-2226, DOI: 10.3390/ani10122210

Lower, A, Lee, Y, Peterson, B, Silva, G & Connor, J 2020, Evaluation of sodium nitrite for mass euthanasia of commercial pigs, *National Pork Board*, Des Moines, Iowa

Marchant-Forde, JN & Boyle, LA 2020, COVID-19 effects on livestock production: a one welfare issue, *Frontiers in Veterinary Science*, vol. 7, DOI: 10.3389/fvets.2020.585787

McKeegan, DEF, Reimert, HGM, Hindle, VA, Boulcott, P, Sparrey, JM, Wathes, CM, Demmers, TGM & Gerritzen, MA 2013, Physiological and behavioural responses of poultry exposed to gas-filled high expansion foam, *Poultry Science,* vol. 92, no. 5, pp. 1145-1154, DOI: 10.3382/ps.2012-02587

McKinney, J, Miller, SP, Lower, A & Clark, S. 2011, Evaluation of a novel electrocution device in swine from birth to 15 pounds. Evidence-based practice: how do we get there? *American Association of Swine Veterinarians - 42nd Annual Meeting Proceedings*, Phoenix, Arizona, United States of America

McKinstry, JL & Anil, MH 2004, The effect of repeat application of electrical stunning on the welfare of pigs, *Meat Science*, vol. 67, no. 1, pp. 121-128, DOI: 10.1016/j.meatsci.2003.10.002

Meyer, R & Morgan Morrow, WE 2005, Carbon dioxide for emergency on-farm euthanasia of swine, *Journal of Swine Health and Production,* vol. 13, no. 4, pp. 210-217

Mullins, CR, Pairis-Garcia, M, George, K, Anthony, R, Johnson, AK, Coleman, G, Rault, J-L & Millman, ST 2017, Determination of swine euthanasia criteria and analysis of barriers to euthanasia in the United States using expert opinion, *Animal Welfare*, vo. 26, no. 4, pp. 449-459, DOI: 10.7120/09627286.26.4.449

Nowak, B, Mueffling TV & Hartung, J 2007a, Can CO2 stunning meet welfare of slaughter pigs?, Aland, A (editor), pp. 1038-1043

Nowak, B, Mueffling, TV & Hartung, J 2007b, Effect of different carbon dioxide concentrations and exposure times in stunning of slaughter pigs: impact on animal welfare and meat quality, *Meat Science,* vol. 75, no. 2, pp. 290-298, DOI: 10.1016/j.meatsci.2006.07.014

Pepin, B 2020, Determine effective oral dosing of sodium nitrite for efficient euthanasia of adult swine using oral drench technique, *National Pork Board*, Des Moines, Iowa

Primary Industries Standing Committee 2008, Model Code of Practice for the Welfare of Animals: Pigs, Third Edition, PISC Report No. 92, CSIRO Publishing, Collingwood

Raj, ABM & Gregory, NG 1996, Welfare implications of the gas stunning of pigs 2. Stress of induction of anaesthesia, *Animal Welfare*, vol. 5, no. 1, pp. 71-78, DOI: 10.1017/S0962728600018352

Rankin, MK, Alphin, RL, Benson, ER, Johnson, AL, Hougentogler, DP & Mohankumar, P 2013, Comparison of water-based foam and carbon dioxide gas emergency depopulation methods of turkeys, *Poultry Science*, vol. 92, no. 12, pp. 144-148, DOI: 10.3382/ps.2013-03341

Royal Society for the Prevention of Cruelty to Animals (RSPCA) Australia 2018, RSPCA Approved Farming Scheme Standards – Pigs, *RSPCA Approved Farming*, November

Royal Society for the Prevention of Cruelty to Animals (RSPCA) Australia 2021, Is sodium nitrite a more humane toxin than 1080 for feral pig control?, *RSPCA Knowledge Base*, Canberra

Sharp, T & Saunders, G 2011, A model for assessing the relative humaneness of pest animal control methods: second edition, *Australian Government Department of Agriculture, Fisheries and Forestry*, Canberra

Shapiro, L, Hix, S & Eason, CT 2009, Palatability and efficacy of sodium nitrite in a paste bait to possums and pigs in cage trials, *Connovation Ltd*. Unpublished.

Shapiro, L, Eason, CT, Bunt, C, Hix, S, Aylett, P & Macmorran, D 2016, Efficacy of encapsulated sodium nitrite as a new tool for feral pig management, *Journal of Pest Science*, vol. 89, no. 2, pp. 489-495, DOI: 10.1007/s10340-015-0706-7

Smith, RK, Rault, JL, Gates, RS & Lay, DC Jr 2018, A two-step process of nitrous oxide before carbon dioxide for humanely euthanising piglets: on-farm trials, *Animals (Basel)*, vol. 8, no. 4, pp. 52-67, DOI: 10.3390/ani8040052

Snow, NP, Foster, JA, Kinsey, JC, Humphrys, S, Staples, LD, Hewitt, DG & Vercauteren, K 2017, Development of toxic bait to control invasive wild pigs and reduce damage, *Wildlife Society Bulletin*, vol. 41, no. 2, pp. 256-263, DOI: 10.1002/wsb.775

Snow, NP, Wishart, JD, Foster, JA, Staples, LD & VerCauteren, KC 2021, Efficacy and risks from a modified sodium nitrite toxic bait for wild pigs, *Pest Management Science*, vol. 77, no. 4, pp. 161-1625, DOI: 10.1002/ps.6180

Steiner, AR, Flammer, SA, Beausoleil, NJ, Berg, C, Bettschart-Wolfensberger, R, Pinillos, RG, Golledge, HDW, Marahrens, M, Meyer, R, Schnitzer, T, Toscano, MJ, Turner, PV, Weary, DM & Gent, TC 2019, Humanely ending the life of animal: research priorities to identify alternatives to carbon dioxide, *Animals (Basel)*, vol. 9, no. 11, pp. 911-936, DOI: 10.3390/ani9110911

Sutherland, MA, Bryer, PJ & Backus, BL 2017, The effect of age and method of gas delivery on carbon dioxide euthanasia of pigs, *Animal Welfare*, vol. 26, no. 3, pp. 293-299, DOI: 10.7120/09627286.26.3.293

Terlouw, EMC, Deiss, V & Astruc, T 2021, Stunning of pigs with different gas mixtures: behavioural and physiological reactions, *Meat Science*, vol. 175, DOI: 10.1016/j.meatsci.2021.108452

Thornber, PM, Rubira, RJ & Styles, RK 2014, Humane killing of animals for disease control purposes, *Revue scientifique et technique (International Office of Epizootics)*, vol. 33, no. 1, pp. 303-310, DOI: 10.20506/rst.33.1.2279

Velarde, A, Gispert, M, Faucitano, L, Manteca, X & Diestre, A 2000, Survey of the effectiveness of stunning procedures used in Spanish pig abattoirs, *The Veterinary Record*, vol. 146, no. 3, pp. 65-68, DOI: 10.1136/vr.146.3.65

Verhoeven, MTW, Gerritzen, MA, Hellebrekers, LJ & Kemp, B 2015, Indicators used in livestock to assess unconsciousness after stunning: a review, Animal, vol. 9, no. 2, pp. 320-330, DOI: 10.1017/S1751731114002596

Verhoeven, M, Gerritzen, M, Velarde, A, Hellebrekers, L & Kemp, B 2016, Time to loss of consciousness and its relation to behaviour in slaughter pigs during stunning with 80 or 95% carbon dioxide, *Frontiers in Veterinary Science,* vol. 3, DOI: 10.3389/fvets.2016.00038

Vogel, KD, Badtram, G, Claus, JR, Grandin, T, Turpin, S, Weyker, RE & Voogd, E 2011, Head-only followed by cardiac arrest electrical stunning is an effective alternative to head-only electrical stunning in pigs, *Journal of Animal Science*, vol. 89, no. 5, pp. 1412-1418, DOI: 10.2527/jas.2010-2920

Von Holleben, K, von Wenzlawowicz, M, Gregory, N, Anil, H, Velarde, A, Rodriguez, P, Cenci Goga, B, Catanese, B & Lambooij, B 2010, Report on good and adverse practices – animal welfare concerns in relation to slaughter practices from the viewpoint of veterinary sciences, *DIALREL,* Cardiff, United Kingdom

Widowski, T, Elgie, R & Lawlis, P 2008, Assessing the effectiveness of a non-penetrating captive bolt for euthanasia of newborn piglets, in proceedings of the Allen D. Leman Swine Conference, St Pail, Minnesota, United States of America

Whiting, TL, Steele, GG, Wamnes, S & Green, C 2011, Evaluation of methods of rapid mass killing of segregated early weaned piglets, *Canadian Veterinary Journal,* vol. 52, no. 7, pp. 753-758

Whiting, TL & Will, D 2019, Achieving humane outcomes in killing livestock by free bullet I: Penetrating brain injury, *Canadian Veterinary Journal*, vol. 60, no. 5, pp. 524-531

World Organisation for Animal Health (WOAH) 2019, Terrestrial Animal Health Code, World Organisation for Animal Health, Paris, France

Wotton, SB & O'Callaghan, M 2002, Electrical stunning of pigs: the effect of applied voltage on impedance to current flow and the operation of a fail-safe device, *Meat Science*, vol. 60, no. 2, pp. 203-208, DOI: 10.1016/s0309-1740(01)00122-x

Woods, J 2012, Analysis of the use of the "CASH" Dispatch Kit captive bolt gun as a single stage euthanasia process for pigs, MS Thesis, DOI: 10.31274/ETD-180810-1851

Zeman, A 2006, What do we mean by "conscious" and "aware"?, *Neuropsychological Rehabilitation*, vol. 16, no. 4, pp. 356 – 376, DOI: 10.1080/09602010500484581