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CO2 STUNNING OF PIGS FOR SLAUGHTER - PRACTICAL GUIDELINES FOR GOOD ANIMAL WELFARE

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Introduction

Preslaughter stunning is used to ensure that animals do not suffer needlessly and are unconscious and insensible to the slaughter procedure. The stunning method itself should be painless and as close as possible to instantaneous in its effect. Furthermore, it should provide duration of unconsciousness and insensibility, which ensures that death from subsequent slaughter, intervenes before recovery of sensibility (Cook *et al.*, 1992). Concern for animal welfare is a major consideration in meat production and is based upon the belief that animals can suffer (Manteca, 1998). Scientific interest in farm animal welfare has rapidly grown in recent years. This has largely been due to the fact that meat consumers increasingly demand that animals are produced, transported and slaughtered in a humane way (Appleby and Huges, 1997).

In the European Union as well as in other countries, there is a legal requirement that all animals destined for meat consumption must be rendered insensible instantaneously and remain insensitive to pain until there is a complete loss of brain responsiveness due to exsanguination (Council Directive 93/119/EC). Under commercial conditions stunning of animals for slaughter may be achieved by a mechanical instrument (Penetrating captive bolt pistol, non-penetrating percussion stunner or free bullet) or by means of an electrical current (Head-only or head-to-back stunning). In addition, the induction of anaesthesia using carbon dioxide may be used to render pigs insensible to pain. Furthermore, alternative methods for stunning of pigs have been tested such as argon-induced unconsciousness (Raj and Gregory, 1995), microwaves (Lambooij *et al.*, 1990) and water jet (Lambooij, 1991).

Internationally, the two most commonly used methods for commercial pre-slaughter stunning of pigs are electrical stunning and CO_2 anaesthetization. The physiological effect of the two methods differs. CO_2 anaesthetization results in a lowering of the blood pH which leads to loss of consciousness (Eisele *et al.*, 1967) and the electric current used in electrical stunning produces an epileptiform activity in the brain leading to unconsciousness without a simultaneous lowering of the blood pH (Hoenderken, 1978).

None of the two methods are ideal and both have some advantages and disadvantages. But both of the methods are internationally recognised as humane and acceptable. Electrical stunning of pigs is, if performed correctly, in principle instantaneous and produces prompt insensibility. However the duration of an electrical stun is rather short and the first indication of a return towards consciousness is the onset of rhythmic breathing, which occurs on average 43-45 sec after end of the stun (Anil, 1991). Thus it is recommended to perform the exsanguination within 15 seconds after the stun to prevent regaining sensibility during debleeding. CO_2 stunning suffers from the disadvantage that loss of insensibility and consciousness is not immediate. In high capacity electrical stunning pigs must be lined up in a race to enter the stunning unit and have to be restrained during stunning to facilitate correct placement of the tongs. In the new Danish group-wise CO_2 stunning system restraint in driving pigs up to the stunning unit and during the exposure to CO_2 is completely eliminated and thus stress is reduced.

Alternatives to high concentrations of CO_2 have been suggested (Raj and Gregory, 1995, 1996). 90% argon in air or a mixture of 60% argon and 30% CO_2 in air eliminate oxygen and there is no reaction from the pig during the phase before loss of consciousness occurs. However, killing rather than reversible stunning is recommended with these gases, as return to consciousness can be rapid, and the time of exposure on more than 7 minutes needed to ensure killing means that existing CO_2 stunning systems cannot be used (Raj, 1999).

It is well known there are differences in opinion concerning the use of CO_2 versus argon or argon/ CO_2 mixtures for stunning or killing of pigs due to welfare considerations during the induction phase of the stunning procedure. CO_2 is known to be pungent to man, but I am however convinced that CO_2 is a humane and acceptable method for stunning of pigs under practical abattoir conditions. Research carried out under both laboratory and practical conditions in Germany, Sweden and Denmark has shown that pigs react very little to exposure to high CO_2 concentrations under practical conditions provided that exposure is rapid (Ring et al., 1988; Barfod, Ring and Erhardt, Barton Gade cited in Lambooij, 1990; Troeger and Woltersdorf, 1991; Barton Gade, 1996). As discussed later on in this presentation stunning with CO_2 furthermore has the great advantage that time to return of consciousness can be controlled and thereby stun-stick interval can be controlled without compromising animal welfare.

Reduced stress in pre-slaughter handling and CO_2 exposure techniques may lower both aversion and induction phases. Lowered stress from good pre-slaughter handling needs to be balanced against any potentially higher stress of the actual stun methods. Stunning related welfare runs through from the farm to the death by exsanguination, and the overall benefits of different stunning systems need this broader welfare consideration.

Evaluation of stunning efficiency and insensibility during shackling, exsanguination and debleeding is an important issue at abattoirs from a point of view of animal welfare. Practical guidelines and code of recommendations have been established for judgement of an

effective stun both by captive bolt stunner and electrical stunning (Anonymous, 1993; Anonymous, 1994; Anil *et al.*, 1997). However, practical guidelines for assessment of anaesthetic depth during CO₂ stunning of pigs have never been established.

The Danish Meat Research Institute has extensively researched the optimal conditions for CO_2 stunning of pigs. It is not my intention in this paper to discuss induction of CO_2 stunning but instead to discuss maintenance of CO_2 stunning of pigs from a point of view of animal welfare. Recommendations and practical guidelines for assessment of stunning efficiency and recognition of signs of recovery are proposed and discussed for use under practical abattoir conditions.

Maintenance of CO2 stunning

Stunning of pigs for slaughter in high concentration of CO_2 is generally a reversible anaesthesia. After the end of exposure to CO_2 , pigs begin to regain consciousness after contact with ambient air and can recover completely (Blomquist, 1957; Dodmann, 1977; Forslid, 1987; Ring *et al.*, 1988).

One minute of exposure to 80% CO₂ induced a cerebral depression definitely below the level used in conventional surgical anaesthesia (Forslid, 1987). The profound depression lasted for about one minute after end of exposure and during the second post-stun minute the EEG gradually attained a pattern characteristic of surgical anaesthesia. Forslid (1987) concluded that from an ethical point of view it could be acceptable to perform exsanguination during the first minute after exposure to CO₂ and the anaesthesia apparently lasts for yet another minute. Martoft *et al.* (2001) have found that the period of marked depression of CNS after emerging from one minute of exposure to 90% CO₂ was approximately 60 to 90 seconds.

Stunning is performed to provide a state of insensibility lasting until the pig is dead through a rapid loss of blood by severing major vessels in the chest near the heart (thoracic stick). The rapid flow of blood from the sticking wound should result in a rapid and permanent loss of blood pressure and lead to insensibility and death due to cerebral anoxia.

To ensure good animal welfare during post-stun handling of pigs, CO₂ stunning should always induce unconsciousness and insensibility of a sufficient duration. Insensibility should last not only through the stun-stick interval but also the time taken for the animal to obtain a sufficient degree of cerebral hypoxia to cause insensibility by exsanguination (Figure 1).

Temporary unconsc	iousness and insensibility		
	Permanent unconsciousness and insensibility		
End of exposure	Exsanguination	> Time	

Figure 1. In correctly performed CO_2 stunning and exsanguination of pigs, the period of unconsciousness and insensibility caused by anaesthesia overlaps the progressive loss of brain responsiveness caused by exsanguination. Thus, no pig regains consciousness during post-stun procedures.

Evaluation of insensibility from an effective CO_2 stunning of pigs can be evaluated by the methods normally employed to judge the effect of a chemical anaesthetic used for surgical procedures (Blackmore and Newhook, 1983). Gregory *et al.* (1987) and Raj (1999) have used brain stem reflexes such as absence of rhythmical breathing and absence of corneal reflex to assess the effectiveness of CO_2 stunning of pigs. These brain stem reflexes may indicate if positive, that the animal is recovering from the stun, but there is no indication of the speed at which the animal is recovering.

Under practical conditions with high slaughter capacity, it becomes more and more common to stun 2-3 pigs in each gondola, and the new Danish group-wise stunning system operates with 4-5 pigs in each gondola. This means that the time required for shackling and exsanguination of the last pig in groups of 3-5 pigs increases and thus the requirements for longer stun-stick intervals increases. From the point of view of animal welfare the most important condition is that no animal ever again regain consciousness before death intervenes from exsanguination. It is totally unacceptable if a stunning procedure gives rise to consciousness and sensibility during or after exsanguination. However, from a practical point of view there is no need to stun pigs deeper than absolutely necessary.

Under practical conditions the stunning must be carried out in such a way, which ensures not only good animal welfare but also a high capacity and a low consumption of CO_2 . This conflict between animal welfare and desire for high stunning capacity and low costs may sometimes lead to inadequate stunning of pigs. Especially, in older CO_2 stunning equipment (with lower CO_2

concentrations) where the initial capacity has gradually increased over the years, this may result in shortening of the exposure time in CO_2 and thus to a less efficient anaesthesia. The risk of pigs regaining consciousness and sensibility thereby increases.

Although it is impossible to judge the state of insensibility of an animal after stunning according to objective scientific criteria, certain stereotypic brain stem reactions are associated with a successful stun and should be assessed on a periodic basis. At the moment, assessment of anaesthetic depth during CO_2 stunning relies more on empirical methods than on evidence from experiments. There are no exact recommendations for the evaluation of anaesthetic depth after CO_2 stunning by testing brain stem reflexes such as rythmic breathing and corneal reflex under practical conditions at the slaughterhouse. It would be useful for employees at the slaughterhouses to have a code of practice to be used for example in a daily quality assurance scheme regarding animal welfare.

Own research

The Danish Meat Research Institute has researched the awakening phase after CO_2 stunning of pigs extensively. Two experiments will be described in the following. The aim of the first experiment was to evaluate the whole awakening phase and establish practical guidelines for assessment of anaesthetic depth during sticking and debleeding in a set of Good Stunning Practice to ensure that no pigs regain consciousness until they die from exsanguination (Holst, 2001a). The aim of the second study was to investigate the relationship between time of exposure to CO_2 and the stun-stick interval in order to establish optimal conditions for CO_2 stunning of pigs (Holst, 2001b).

Both experiments took place in a full-scale research set-up at a commercial abattoir (Barton-Gade *et al.*, 1995) and pigs (live weight approx. 100 kg) were randomly chosen from the normal delivery to the abattoir. Pigs were stunned in a large box, which was essentially a lift (dimensions $1.4 \times 2.4 \text{ m}$), which was lowered into the gas and then raised. The box was operated in a paternoster simulation close to practical slaughterhouse conditions, where the box stopped at appropriate intervals to simulate the loading and unloading of gondolas. Selecting the stop time at the bottom position controlled the total stunning time.

In the first experiment a total of 210 pigs were stunned one by one in a mixture of 90% CO₂ in air where the CO₂ concentration at the first stop was >70%. The first position was reached after 7 seconds and the bottom position after 40 seconds. The total stunning time was 132 seconds of which 120 seconds were in >70% CO₂.

After stunning the pigs were tipped out on to the receiving table and remained undisturbed while regaining consciousness. Each pig was continuously examined for behavioural and different physiological reflexes to evaluate the rate of regaining consciousness. The

following reflexes were noted: corneal reflex, cilia (eyelash) reflex, regular respiration, excitation, nystagmus (horizontal vibrating movements of eyeball), spontaneous blinking of the eye, conscious movements and attempts to stand up. Regular respiration was defined as being deep and having regular intervals, i.e. different from superficial and occasional gasps.

Time from end of CO_2 exposure to appearance of reflexes was registered for each pig. Pigs not showing corneal reflex after 150 seconds were assumed to be dead, shackled and exsanguinated. For practical purposes the total resting period after end of exposure to CO_2 was limited to 15 minutes before pigs were returned to the lairage area.

In a second experiment a total of 1,638 pigs were stunned with varying exposure times in a mixture of 90-93% CO_2 in air where the CO_2 concentration at the first stop was 79-83%. The pigs were stunned in groups of two to five in the box. The whole experiment was performed in nine separate sub-sections with different, but increasing total exposure time to CO_2 ranging from 112 to 192 seconds in intervals of 10 seconds.

Approximately 200 pigs were stunned in each sub-section, except for the shortest and longest holding times where only 158 and 100 pigs, respectively, were stunned. Pigs in each group were shackled and hoisted, but not immediately exsanguinated. Each pig was continuously examined for corneal reflex and when showing corneal reflex it was immediately exsanguinated by a skilled slaughterman. Time from end of CO_2 exposure until reappearance of corneal reflex was registered. Pigs not showing corneal reflex were exsanguinated after 150 seconds.

Results

In experiment 1 no pigs appeared to have reflexes just after end of CO_2 exposure. Corneal reflex was the first reflex to occur on average after 42 seconds, and was used to evaluate recovery from the deepest level of anaesthesia just prior to collapse and death. The ciliar reflex followed shortly after, at 51 seconds on average. Regular respiration occurred after 68 seconds and was used as the first sign of return to consciousness. Excitation was noted after 76 seconds and nystagmus after 86 seconds. Excitation and nystagmus did not occur in all pigs. 77% of the pigs showed excitation to a greater or lesser extent and only 42% showed nystagmus. Spontaneous blinking of the eye occurred after 93 seconds and was together with nystagmus used to indicate imminent return to consciousness. Conscious movements of head or legs were observed after 171 seconds and were evaluated as complete return of consciousness. Attempts to stand up were noted 387 seconds after the end of CO_2 exposure (Table 1).

	Appearance of different physiological reflexes (In seconds)				
Reflex	% pigs	Mean ± SD	Min. time	Max. time	
Corneal	100	42 ± 32	16	146	
Ciliar	100	51 ± 35	19	192	
Regular respiration	100	68 ± 34	24	179	
Excitation	77	76 ± 35	25	207	
Nystagmus	42	86 ± 33	38	190	
Spontaneous blinking	100	93 ± 52	37	261	
Conscious movements	100	171 ± 101	64	685	
Attempt to stand up	88 ¹⁾	$(387 \pm 276)^{11}$	67	900 ²⁾	

Table I. Time to appearance of different physiological reflexes in pigs regaining consciousness after CO2 stunning.

There was a considerable range in time to appearance of each reflex. The fastest appearance of corneal reflex was 16 seconds after end of CO_2 exposure and the slowest was 146 seconds. 69 pigs (33%) showed corneal reflex within 30 seconds of the end of exposure. 88 % of the pigs showed attempt to stand up before end of the resting period, i.e. within 15 minutes (900 seconds). 16 pigs (8%) died during stunning.

In the experiment 2 it was found, that at the shortest time of exposure of 112 seconds corneal reflex reappeared in 91% of the pigs within 150 seconds after end of exposure (Figure 2). With increasing time of exposure to CO_2 , the number of pigs with corneal reflex decreased and after the longest time of exposure at 192 seconds only 3% of the pigs showed corneal reflex.

The minimum time to first reappearance of corneal reflex increased from 20 seconds to 102 seconds with exposure time increasing from 112 to 192 seconds respectively (Figure 2) as an indication of deeper and longer lasting anaesthesia with increasing time of exposure to CO_2 .





Discussion

Under practical conditions at an abattoir it is impossible to evaluate more than a few of the mentioned reflexes. The most appropriate reflexes to be used are corneal reflex, regular respiration and spontaneous blinking of the eyes, as they are seen in all pigs and are clearly observable.

Using the results of the first experiment it was found that regular respiration occurred just under half a minute later than corneal reflex, and after yet another approximately half minute spontaneous blinking of the eye occurred. Forslid (1987) found that pigs exposed to 80% CO₂ for 60 seconds showed regular respiration 30-60 seconds after end of exposure and head movements after 2-3 minutes as the first sign of returning motoric control. Based on the recordings of EEG, Forslid (1987) furthermore found, that the pigs were anaesthetised below the level of surgical anaesthesia for about one minute after end of CO₂ exposure or until the reappearance of regular respiration and must have remained anaesthetised and insensible for at least one minute longer, or until just prior to spontaneous movements. In the present experiments pigs thus must have been insensible until after reappearance of regular respiration and even during the excitation phase until spontaneous blinking.

The death of pigs actually occurs due to exsanguination and a rapid loss of blood by severing the major vessels in the chest near the heart (Thoracic stick). After chest stick time to loss of brain responsiveness (Visual Evoked Potentials) has been found to be on average 18 seconds (range 14-23 seconds) in pigs (Wotton and Gregory, 1986). The same authors found a development of an isoelectric electrocorticogram (ECoG) 22-30 seconds after the chest stick. Hoenderken (1979) has reported that chest stick by a skilled slaughterman induces an isoelectric ECoG in 12-20 seconds. These investigations indicate that pigs must be anaesthetized by the stunning for at least 30 seconds after sticking to have a safety margin where they remain insensible until a permanent insensibility intervenes due to cerebral anoxia from loss of blood (See Figure 1).

This means, that with a safety margin of 30 seconds from sticking to expected insensibility, the sticking must be performed 30-45 seconds before the occurrence of excitation and spontaneous movements to ensure that the pigs remain unconscious during bleeding until death occurs. In practice the pigs then need to be stuck in the very deep stage of anaesthesia, at around the time when the corneal reflex reappears. As anaesthetizing below the level of surgical anaesthesia according to Forslid (1987) lasts until after reappearance of regular respiration, exsanguination may be tolerated after reappearance of corneal reflex.

To ensure worker safety it is recommended to exsanguinate the pigs before excitation appears. Due to the great variation in time to appearance of excitation pigs in this experiment should have been exsanguinated around 25 seconds after end of exposure to CO_2 . At that time corneal reflex was registered in around 5% of the pigs. Thus, exsanguination should be performed at the point where maximum 5% of the pigs show corneal reflex. At that time no pigs showed regular respiration or spontaneous blinking of the eye. These findings could be used in a set of guidelines for Good Stunning Practice.

The results of this study show that as exposure time to CO_2 increases, the time to regain consciousness is delayed as indicated by the longer time to first reappearance the corneal reflex. Within a specific time exposure to CO_2 there was a variation in the time from the end of stunning until reappearance of corneal reflex. There is always a biological variation, both within a breed and possibly between breeds (Grandin, 1988), and for a given CO_2 concentration and exposure time, there will be a variation in the depth of the anaesthesia that may be assumed to be due to different susceptibility to CO_2 . This is known to occur using chemical anaesthetics for surgery (Thurmon et al., 1996).

Because of this biological variation, corneal reflex in different pigs may for certain stunning conditions disappear at different times during exposure to CO_2 , and reappear at different times after end of exposure. Some animals may even die in the deep anaesthetic stage due to depression of the brain stem resulting in vascular collapse and cardiac arrest. This means that some pigs will never recover from certain stunning conditions while others will be reversibly stunned and show corneal reflex within a given time after the end of exposure.

The difference in susceptibility may also explain why minimum time to first reappearance of corneal reflex was earlier after one exposure time than after the subsequent exposure time as for example seen after exposure for 142 and 152 seconds respectively. Taking into account the different susceptibility to CO_2 , the minimum time to reappearance of corneal reflex after exposure for 132 and 142 seconds may be assumed to be lower than found in this experiment. Thus, a stun-stick interval of ≥ 60 seconds requires an exposure time of at least 142 seconds.

Practical experience has shown that with commercial slaughter speeds there is no need for stun-stick intervals longer than 90 seconds, and even in the new Danish group-wise stunning system a stun-stick interval of 60-70 seconds is sufficient. A maximum stun-stick interval of >80 seconds was in this experiment demonstrated to be fulfilled with an exposure time in CO_2 of 162 seconds without compromising animal welfare.

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In commercial CO₂ stunning equipment, the flooring as well as the sides of the gondolas is penetrable for gas to increase the intrusion of the gas. Furthermore, the gondolas are small and only intended for 1 or 2 pigs (very seldom 3 or 4 pigs). The boxes in the research set-up are much larger than the commercial gondolas, and are designed for 5 ± 1 pigs (maximum 7). In addition, the flooring in the boxes is solid. This means that intrusion of CO₂ into the centre of these large boxes may be a little delayed compared to the intrusion into commercial gondolas. Under practical abattoir conditions the stunning times indicated may therefore be a little shorter than

found in this research. On the other hand we have found that intrusion of CO_2 into the box used in the research set-up was very fast and the CO_2 concentration at the centre of the box was >70% only 1-2 seconds after the box reached the first stop position or equal to the concentration measured in the pit.

The results from these experiments indicate, that efficiency of CO₂ stunning of pigs can be evaluated by assessing physical reflexes during exsanguination and debleeding and that the stun-stick interval can be controlled by adjustment of the time of exposure to CO₂. Thus it is not necessary to set up limitations for maximum stun-stick intervals or legislate on stun-stick intervals with specified technical requirements, as seen in some countries (LD, 1995; BELF, 1997). Instead the stunning effect should be monitored by mean of functional requirements as Good Stunning Practice i.e. no pigs regain consciousness during post-stun handling.

If the equipment is such that the above-mentioned relation between time of exposure and stun-stick interval cannot be fulfilled, the stunning conditions must be controlled. Maybe Good Stunning Practice can be obtained either by increasing the CO_2 concentration or by increasing exposure times, but in other cases technical alterations will be necessary to adjust the discrepancy between the actually stunning and the required stun-stick interval to obtain good animal welfare.

Conclusions

To ensure Good Stunning Practice during CO_2 stunning, i.e. that no pigs regain consciousness during post-stun handling and exsanguination, the safe depth of anaesthesia at the time of sticking can be evaluated by the following criteria:

- No pig shows deep or regular respiration except for irregular abdominal gasping
- No pig shows excitation or kicking except for slow movements of legs
- No pig shows spontaneous blinking of the eye
- Maximum 5% of the pigs have a corneal reflex

Using these criteria just before sticking not only fulfils animal welfare concerns but also slaughterhouse workers safety during poststun handling of pigs, as the pigs are hanging completely still during exsanguination without any convulsions.

The results of this study have shown that with increasing exposure time to CO_2 the stun-stick interval increases. This means that by changing exposure time to CO_2 the length of the stun-stick interval can be controlled. Furthermore, it has been demonstrated that a stun-stick interval up to 90 seconds under practical conditions can be established without compromising animal welfare. It is not necessary to legislate with specified technical requirements on maximum stun-stick intervals after CO_2 stunning of pigs as seen in some countries. Instead the stunning effect should be monitored by mean of functional requirements as Good Stunning Practice i.e. no pigs regain consciousness during post-stun handling until they die from debleeding.

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