Comparison of CO₂ versus mixed CO₂:Argon Gas at Different Flow Rates Using the Smart Box Euthanasia Device as an Effective Method of Piglet Euthanasia

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Summary and Implications

The objective of this study was to compare the effectiveness and response of weaned piglets to 100% CO₂ gas relative to a 50:50 CO₂:Argon gas mixture as an effective tool for euthanasia. A total of 180 piglets, BW 4.6 \pm 0.7 kg, were utilized. Piglets were 16 to 24 days of age. Two gas mixtures (100% CO2 and 50:50 CO2:Argon) and 4 flow rates (slow, medium, fast, and prefill; 20%, 35%, 50%, and prefill with 20%, chamber volume per minute, respectively) were examined. Two piglets were placed in a modified Smartbox[™] (Euthanex Corp, Palmer, PA) chamber, in which the lid and one side are composed of clear plastic to facilitate behavior observations. Piglets were scored using direct observation for latency to perform three behaviors associated with insensibility: loss of posture, last movement and gasping. Open mouth breathing occurred prior to insensibility and was used as an indicator of distress. The CO₂:Argon gas mixture and slow flow rates prolonged the duration of insensibility, as measured by last movement and did not confer advantages for measures of distress.

Introduction

The U.S. swine industry euthanizes piglets when their chances of survival are low and they are suffering due to injury or illness. This results in million of piglets being euthanized annually, and tools are needed to accomplish euthanasia quickly, economically and safely, as a repeatable humane process.

Carbon Dioxide (CO_2) gas to euthanize young pigs is acceptable by National Pork Board guidelines and is increasingly common. CO_2 is economical, relatively safe and readily available. CO_2 is a colorless, odorless gas, which stuns by lowering the pH of the central nervous system. It is capable of doing this because it is mildly acidic. However, this acidity may cause sensation and distress. Argon has been proposed as a more humane alternative, since it is unreactive throughout the body's systems, stuning through asphyxiation. Evidence from other species suggests that argon may be less aversive than the standard CO_2 methods. Furthermore, little empirical research exists to support best management practices for onfarm CO_2 euthanasia, in terms of gas flow rate, concentration or duration of exposure. Therefore the objective of this study was to compare the effectiveness and response of weaned piglets to 100% CO_2 gas relative to a 50:50 CO_2 :Argon gas mixture as an effective tool for euthanizing.

Materials and Methods

The protocol for this experiment was approved by the Iowa State University Institutional Animal Care and Use Committee (11-09-6825-S). The experiment was conducted from May to September, 2010.

Animals and housing: A total of 180 mixed sex piglets (90 barrows and 90 gilts) were used from commercial PIC genetic lines. Piglets were obtained and housed at the Iowa State University Swine Nutrition Farm. Piglets weighed 4.6 \pm 0.7 kg and were 16 to 24 days of age.

Experimental design: Piglets were compared as mixed sex pairs. The experimental design for this study was a 2×4 factorial arrangement of treatments. Two gas mixtures; 100% CO₂ (**CO**₂) and 50:50 CO₂:Argon (**CA**) with four different chamber exchange rates: slow, medium and fast, allowing 20%, 35%, 50% chamber volume turnover per minute respectively, and pre-fill of the chamber with 20% chamber volume turnover per minute.

Euthanasia protocol: Piglets were placed into a plastic chamber (inside dimensions 43 wide, x 60 long, x 30 height, cm), with 2 clear sides facilitating behavior observations. The floor was fitted with a black rubber mat to prevent slipping. Gas was supplied utilizing a Euthanex AgProTM (V-ast, Mason City, IA; Figure 1) and a constant gas flow was provided by a compressed gas regulator (Western Enterprises, Westlake, OH). Between each treatment the chamber was blown out with ambient air.

Figure 1. Smartbox[™]



Behavioral measures: Piglets were observed directly for latency to behavioral indicators of stunning and death (Table 1).

Parameter	Definition		
Open Mouth	Piglets mouth is open, taking in		
Breathing (OMB)	quick breaths, with distinct		
	thoracic movements; panting;		
	upper and lower jaw being held		
	open with the top lip pulled back,		
	exposing gums or teeth and		
	panting (pronounced inhalation		
	and exhalation observed at the		
	flanks ^{2,3}		
Gasping (GASP)	Rhythmic breaths characterized by		
	very prominent and deep thoracic		
	movements, with long latency		
	between, may involve stretching		
	of the neck; often occurs right		
	before or after loss of posture ^{1,2}		
Loss of posture (LP)	Piglet is slumped down, making		
	no attempt to right itself, follows a		
	period of attempts to maintain		
	posture; loss of attitude of position		
	of the body		
Last movement	No movement is observed by the		
(LM)	piglet of any type		

Statistical analysis: Analysis was performed in SAS. OMB, GASP, and LP were analyzed as Univariate productlimit estimation of the survival curves. LM data was log transformed and analyzed as a mixed model with fixed effects of sex and treatment, and blocked by day of treatment. Raw means were calculated using Proc Means.

Results and Discussion

When comparing gas types, differences were observed for LM with CA treatment taking longer relative to CO₂. Differences were not observed for OMB, LP or GASP between the two gas types. Within gas types, as expected, gas flow rate significantly (P < 0.001) affected LM, with the slow flow rate taking longer than the medium, fast, or prefill. No differences were observed for all other reported measures. In conclusion, CA and slow flow rate prolonged the duration of insensibility, as measured by LM and did not confer advantages for measures of distress (OMB).

Table 2. Latency to last movement by gas type and flow rates	,
P-value within gas type over flow rates <i>P</i> =0.003.	

	Gas Mixture					
Flowrate	CO2	SE	CA	SE		
Slow	529	181	774	216		
Medium	312	40	467	37		
Fast	274	27	397	32		
Prefill	269	73	451	209		

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