

USE OF NITROUS OXIDE FOR PIG STUNNING. PRELIMINARY RESULTS

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INTRODUCTION

Stunning of pigs for slaughter is performed most frequently using electrical current or carbon dioxide/air mixtures. From the humane point of view, the use of CO<sub>2</sub> is still controversial. The animals show frequently excitation and increased motor activity when exposed to CO<sub>2</sub>. These signs can be interpreted as conscious flight reactions which suggests that the pigs could experience some unease at least at the beginning of gas-exposure (Troeger and Wolsterdorf, 1991). On the other hand, Forslid (1991) reported from EEG recordings that the increased motor activity is unconscious. Gregory (1992) concluded that "clearly this issue still needs investigating as there is no consensus".

CO<sub>2</sub> stunning was reported to give less bone fractures and blood splashing than electrical stunning (Larsen, 1983). Concerning meat quality, its effects have been controversial. CO<sub>2</sub> stunning causes acidification of the organism, due to lactic acid production resulting from muscle activity and hypoxia, or/and to a direct effect of CO<sub>2</sub> (the latter is the anhydride of carbonic acid). This acidification could contribute to lower the post-mortem muscle pH and so favour the occurrence of PSE meat. An alternative gas for anaesthesia of slaughter pigs could be the nitrous oxide N<sub>2</sub>O (laughing gas), which is frequently used in surgical anaesthesia in humans and animals. This gas is authorized in the food industry. Its density is close to that of CO<sub>2</sub>, and so it could be used with similar equipments. Troeger and Wolsterdorf (1991) found that unconsciousness could not be achieved using N<sub>2</sub>O/CO<sub>2</sub>/air mixtures, even after gas-exposure for three minutes. However, Delpuech (1976) obtained full anaesthesia in chicken and piglets with N<sub>2</sub>O/air mixtures at 70 to 80% N<sub>2</sub>O.

The aim of the present study was to investigate the efficiency of N<sub>2</sub>O as a stunning agent and its effects on meat quality, particularly in comparison with CO<sub>2</sub>.

## MATERIALS AND METHODS

### Animals

The pigs were Large White or Large White x French Landrace, weighing 95 to 105kg. They were bought from a pig deal one week before experiments. All the pigs in one given experiment came from the same farm.

### Stunning equipment

Electrical stunning was performed using a manual Etim apparatus (Etim Company, France). The current of 300V and around 0.5A was delivered for one to two seconds.

Gas stunning was performed in a closed cage of approximately 400dm<sup>3</sup> volume. The animal was introduced into the cage, then pure CO<sub>2</sub> or N<sub>2</sub>O was blown till a residual oxygen concentration of 5% in case of CO<sub>2</sub> and 4% in case of N<sub>2</sub>O, corresponding approximately to concentrations of 75% CO<sub>2</sub> and 80% N<sub>2</sub>O respectively. These concentrations had been determined in preliminary trials where concentrations of 75 to 85% were tried. Oxygen was continuously measured with a Beckman 777 apparatus. The animal reactions were observed throughout exposure to stunning gases.

### Experiment 1

Six Large White pigs were stunned using N<sub>2</sub>O, then again a week later using CO<sub>2</sub>. As soon as unconsciousness was judged to be achieved, the animals were pulled out of the cage. Blood was obtained immediately by *vena cava* puncture and analyzed for pH, bicarbonate, pCO<sub>2</sub> and pO<sub>2</sub> with a Radiometer BMS2 blood gas analyzer. The animals were allowed to recover in air. They were considered to have recovered consciousness as soon as they manifested perception of shocks from an electric rod of a type usually used for pig handling. Times to obtain the final gas concentrations, to achieve apparent anaesthesia and to get recovery were registered.

### Experiment 2

Twenty LW pigs were used. Ten of them were stunned by electric current and the 10 others by N<sub>2</sub>O. The latter were kept in the cage of two minutes after the beginning of exposure to stunning gas. All the pigs were exsanguinated and the carcasses were dressed according to commercial procedures. At one hour after slaughter, a sample was taken from the *gluteus superficialis* and put in liquid nitrogen for later measurement of pH (pH<sub>1</sub>), lactic acid and ATP. The day after slaughter, the left ham was scored for meat quality (0=PSE to 4=good quality), then a sample of *biceps femoris* was taken for determination of ultimate pH (pH<sub>2</sub>), water holding capacity (press method of Goutefongea, 1966) and reflectance at 630nm. The hams from six pigs of each group were processed into cured cooked ham.

Frozen samples of *gluteus superficialis* were ground with liquid nitrogen in a Waring blender. Two grams of frozen powder were homogenized in 18ml of 0.005M iodoacetate and pH was measured on the homogenate. Five grams of powder were homogenized in 0.6M perchloric acid for determination of ATP and lactic acid by classical enzymatic techniques (Bergmeyer, 1974).

### Experiments 3 and 4

Twenty LW x Landrace pigs were used in each of these experiments. Ten of them were stunned by CO<sub>2</sub> and then 10 others by N<sub>2</sub>O. All the animals were kept in the cage for two minutes after the beginning of exposure to stunning gas. Then they were exsanguinated and measurements of meat quality were performed in the same way as in experiment 2. Times to obtain the final gas concentrations and to achieve apparent anaesthesia were registered in experiment 3.

## RESULTS and DISCUSSION

### Anaesthesia

Time to achieve apparent unconsciousness was found to be slightly shorter with N<sub>2</sub>O in experiment 1, but the contrary was observed in experiment 3 (Table 1). This time was much longer than that reported by Troeger and Wolsterdorf (1991) for CO<sub>2</sub> stunning, i.e., 30 to 40 seconds. This difference was undoubtedly due to the differences between both studies in stunning equipment. In the experiments of Troeger and Wolsterdorf, pigs were lowered in a box filled with the CO<sub>2</sub>/air mixture, so the gas concentrations were kept constant throughout gas-exposure. In our conditions, the final gas concentrations were reached only after 40 to 55 seconds. Times for recovery of apparent consciousness were similar with both gases. Muscular activity during bleeding was more marked in N<sub>2</sub>O stunned pigs. None of the CO<sub>2</sub> stunned pigs showed activity during bleeding, while five of the 10 N<sub>2</sub>O pigs struggled more or less.

There was a striking difference in blood acid-base parameters between CO<sub>2</sub> and N<sub>2</sub>O treatments. With N<sub>2</sub>O, the values of pH, pCO<sub>2</sub> and HCO<sub>3</sub><sup>-</sup> were kept almost normal after around 80 seconds of gas-exposure. By contrast, a marked respiratory acidosis developed in CO<sub>2</sub> stunned pigs. The values observed for blood acid-base parameters after CO<sub>2</sub> stunning agreed with the observations by Forslid and Augustinsson (1988). It appears clearly that acidosis observed in CO<sub>2</sub> stunning was due almost completely to a direct effect of CO<sub>2</sub> itself, and not to the hypoxia induced by lowering oxygen concentration. Also, these observations suggest that the physiological stress is less with N<sub>2</sub>O than with CO<sub>2</sub> stunning. This is supported by the trend to a lower lactic acid level in the musculature of N<sub>2</sub>O stunned pigs (Table 2).

### Meat quality

Post-mortem pH fall was faster in electrically stunned pigs than in N<sub>2</sub>O stunned pigs (Table 2). However, no significant difference was found for the other meat quality criteria between both stunning techniques.

In experiment 3, post-mortem glycolysis was slower and meat quality was better in N<sub>2</sub>O stunned pigs than in CO<sub>2</sub> stunned pigs. No significant difference was found between both treatments in experiment 4, however there was again a slight trend to a better meat quality in N<sub>2</sub>O stunned pigs.

## CONCLUSION

The results of the present study show that stunning of slaughter pigs can be achieved using the laughing gas N<sub>2</sub>O. The physiological stress seems to be lower with N<sub>2</sub>O stunning than with CO<sub>2</sub> stunning. This could explain the slower post-mortem glycolysis observed after N<sub>2</sub>O stunning in experiment 3 (similar trend in experiment 4). The indications that meat quality would be better with N<sub>2</sub>O are encouraging and deserve further investigation. However, the present experiments were insufficient to allow conclusions on the very important issue of welfare. Indeed, the conditions of gas-exposure were far from the practical abattoir conditions, since the time needed to reach the high gas concentrations required for stunning was unusually long. Comparison of the effects of CO<sub>2</sub> and N<sub>2</sub>O on animal welfare and meat quality in conditions close to those prevailing in abattoirs is needed.

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Table 1. Effects of stunning gas on anaesthesia parameters and acid-base parameters.

	Experiment 1		Experiment 3	
	CO <sub>2</sub> (n=6)	N <sub>2</sub> O (n=6)	CO <sub>2</sub> (n=6)	N <sub>2</sub> O (n=6)
Time for final gas concentration (s) (1)	52±6	52±6	44±7	48±10
Anaesthesia (s) (2)	102±20 <sup>a</sup>	81±12 <sup>b</sup>	76±20 <sup>a</sup>	95±12 <sup>b</sup>
Recovery (s) (3)	207±20	191±23		
Blood pH	6.72 ± 0.10 <sup>a</sup>	7.36 ± 0.07 <sup>b</sup>		
Blood pCO <sub>2</sub> (kPa)	21.6 ± 7.1 <sup>a</sup>	4.7 ± 0.9 <sup>b</sup>		
Blood HCO <sub>3</sub> <sup>-</sup> (meq/l)	19± 3	19± 2		
Blood pO <sub>2</sub> (kPa)	3.7± 0.7	3.2± 2.1		

1 Time to reach an oxygen concentration of 5% in the case of CO<sub>2</sub> and 4% in the case of N<sub>2</sub>O.

2 Time to reach apparent anaesthesia.

3 Time to reach apparent consciousness recovery.

Table 2. Effect of different stunning techniques on meat quality.

Traits of meat quality <sup>1,2</sup>	Experiment 2		Experiment 3		Experiment 4	
	Elec.	N <sub>2</sub> O	CO <sub>2</sub>	N <sub>2</sub> O	CO <sub>2</sub>	N <sub>2</sub> O
ATP, $\mu\text{mol/g}$	1.8 $\pm$ 0.7 <sup>a</sup>	3.4 $\pm$ 0.7 <sup>b</sup>	3.5 $\pm$ 0.7	4.0 $\pm$ 1.0	2.1 $\pm$ 1.2	2.5 $\pm$ 1.9
Lactic acid, $\mu\text{mol/g}$	52 $\pm$ 8 <sup>a</sup>	30 $\pm$ 9 <sup>b</sup>	40 $\pm$ 12 <sup>a</sup>	29 $\pm$ 9 <sup>b</sup>	49 $\pm$ 15	43 $\pm$ 26
pH <sub>1</sub>	6.1 $\pm$ 0.2 <sup>a</sup>	6.5 $\pm$ 0.3 <sup>b</sup>	6.5 $\pm$ 0.3	6.6 $\pm$ 0.7	6.2 $\pm$ 0.3	6.4 $\pm$ 0.5
pH <sub>2</sub>	5.8 $\pm$ 0.1	5.7 $\pm$ 0.2	5.5 $\pm$ 0.2	5.7 $\pm$ 0.2	5.6 $\pm$ 0.5	5.6 $\pm$ 0.5
Quality score	1.7 $\pm$ 0.8	2.2 $\pm$ 0.8	1.5 $\pm$ 0.7 <sup>a</sup>	2.3 $\pm$ 0.8 <sup>b</sup>	-	-
WHC, %	25 $\pm$ 6	23 $\pm$ 3	26 $\pm$ 4 <sup>a</sup>	22 $\pm$ 4 <sup>b</sup>	27 $\pm$ 4	26 $\pm$ 4
Reflectance, %	73 $\pm$ 10	68 $\pm$ 16	91 $\pm$ 6	80 $\pm$ 18	86 $\pm$ 5	84 $\pm$ 6
Cooking yield, %	69.5 $\pm$ 2.9	69.1 $\pm$ 3.5	68.5 $\pm$ 2.3 <sup>a</sup>	68.5 $\pm$ 2.6 <sup>b</sup>	66.4 $\pm$ 3.9	67.0 $\pm$ 1.6

<sup>1</sup> ATP, lactic acid and pH<sub>1</sub> in *gluteus superficialis*; the other traits in *biceps femoris*.

<sup>2</sup> n=10 in all experimental groups except for cooking yield of ham (n=6).