Stunning slaughter pigs using the inert gas helium

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Abstract – The objective of this study was to determine the stunning properties of the inert gas helium. Slaughter pigs were stunned in a helium containing dome (experimental facility) and in a commercial carbon dioxide (CO₂) dip-lift system, respectively. During exposure to the different gases, behavioral parameters were recorded. After stunning and sticking, blood samples were taken to analyze the hormones epinephrine and norepinephrine. Meat quality parameters such as pH and, muscle temperature were recorded at 45 min p.m. (pH_{45}/T_{45}) and electrical conductivity at 24 h p.m. (EC₂₄). After that, the presence of ecchymosis in common meat cuts Tree days p.m. was recorded. samples of semimembranosus muscles have been used for the determination of grill loss and sensory traits (tenderness, juiciness, flavour).

Pigs stunned with helium showed no aversive behavior inside the gas atmosphere, whereas pigs in CO_2 hyperventilated, revealed retreat or escape attempts and vocalization. Stunning pigs with CO_2 led to comparable higher epinephrine and norepinephrine levels resulting also in lower pH₄₅ and higher T₄₅ values. No significant differences were found for EC₂₄, grill loss and sensory quality. After helium stunning, none of the carcasses showed ecchymosis. In conclusion, stunning with helium can not only be considered an animal-friendly method but leads also to superior carcass and meat quality compared to CO_2 stunning.

I. INTRODUCTION

In most countries, carbon dioxide (CO_2) is used in gas stunning systems for commercial pig slaughter. Nevertheless, CO₂ has been criticized in terms of animal welfare. At gas contact, pigs show symptoms of respiratory distress [1,2] and some animals even display aversion in the form of escape attempts and vocalization [3,4]. According to Council regulation (EC) No. 1099/2009, the use of inert gases beside CO_2 is allowed for stunning pigs. So far, only high concentrations of the inert gas argon have been evaluated for stunning pigs. Pigs exposed to argon did not show any sign of aversion [4] and had a superior meat quality. However, according to Troeger et al. [5] the use of argon leads to a high incidences of blood splash and ecchymosis in carcasses, making it not acceptable for commercial use. A new alternative would be the application of helium in stunning systems.

II. MATERIALS AND METHODS

A total of 80 fattening pigs (commercial crossbreeds) were stunned and slaughtered in two groups. One group of forty pigs (**Helium-pigs**) were single stunned in a helium containing dome (Fig. 1, experimental facility). As helium has a lower density than air (helium: 0.18 kg/m³, air: 1.20 kg/m³) stunning was performed using a dome built from plexiglass (1.75 m x 0.90 m x 1.55 m, 15 mm wall thickness).

After filling the dome with helium up to a mean concentration of 98.5 %, it was lowered over a cage with a pig inside. The pig was exposed to the gas for 180 s.



Figure 1: Helium stunning design, longitudinal side; experimental animal already driven into the cage, shortly before the stunning cycle began

The **CO₂-pigs**, another group of forty pigs, were single stunned in a commercial CO₂ dip-lift system (Butina Company, Denmark). As CO₂ has a higher density than air (CO₂: 1.98 kg/m^3), pigs went down in a gondola to the bottom of a pit with high CO₂ concentrations > 90 %. The stunning cycle of the dip-lift system was 120 s.

Animal welfare

In order to evaluate the different stunning procedures from an animal welfare point of view, each animal was filmed with a video camera and sound levels of vocalization (in decibels) were recorded during the process. The behavior of the pigs was analyzed according to time required to overbalance, and uncontrolled muscular excitation as well as convulsion duration were measured. As behavoioural parameters retreat attempts, hyperventilation and escape attempts were recorded.

The convulsions were classified into four grades: (0) negligible, (1) up to five running motions, single head movements, (2) continuous running motions, head movements, (3) massive running motions, recurring movements of the whole body.

Samples of sticking blood were taken. The quantitative detection of norepinephrine and epinephrine was carried out by an enzyme-linked immunosorbent assay (ELISA) developed for human plasma (BTBA E-4500, Biotrend Company, Köln, Germany). Due to the high levels of catecholamines in the plasma of slaughter pigs, the samples had to be diluted 1:200 with distilled water before performing the test.

All parameters were analyzed statistically by NCSS program (Version 1.7.20, NCSS, LLC. Kaysville, Utha, USA). The data were checked for normal distribution using the Shapiro-Wilks-, Kolmogorov-Smirnov- and D'Agostino-test. The data of the Helium- and CO₂- pigs were subjected to a descriptive data analysis (t-test or Mann-Whitney-U-test).

Meat and carcass quality

For the determination of the meat quality, parameters in the *Musculus semimembranosus* (SM) were measured. The pH value (pH₄₅) and the muscle temperature (T₄₅) were collected 45 minutes *post mortem*. After 24 hours the electrical conductivity (EC₂₄) was measured in each left carcass sight.

After all carcasses were deboned and divided into commercial cuts. The presence of ecchymosis, defined as small blood areas located in the skeletal muscles, of all cuts was recorded.

After 3 days p.m. slices (1.5 cm thick) of SM were roasted at 160 °C for 3 min and 11 s. The weight loss after heating was measured and the sensory parameters tenderness, juiciness and flavor were evaluated by six panelists according to a six-point scoring system (1= unacceptable to 6 = very good).

III. RESULTS AND DISCUSSION

Animal welfare

Pigs stunned with helium showed no aversive behavior inside the gas atmosphere (Fig. 2). The pigs overbalanced after 20 s after preceding swaying movements. This is in accordance with results, which have been obtained by applying the inert gas argon [6, 7].

In contrast, pigs stunned with CO_2 showed retreat and escape attempts as well as hyperventilation, before the loos of balance took place. Also vocalizations occurred only during CO_2 stunning. The sound level had a maximum of 105.8 decibels. Düpjan *et al.* [8] suggested that very loud screams of pigs were a sign of intense psychological distress. Schaeffer *et al.* [9] argued that screams express anxiety as well as pain. In the CO_2 environment, loss of balance occurred after approximately 16 s.

Thereafter all pigs showed uncontrolled muscular excitation, lying in side position. In comparison Helium-pigs showed more often grade 1, the most CO_2 –pigs showed grade 2. The convulsion duration during Helium stunning was longer than during CO_2 stunning. This effectively balances each other out and the differences are insignificant. (Fig. 2).



Figure 2: Percentage of pigs showing reactions and convulsions (grade 0 - 3) during stunning with helium (n = 40) and CO_2 (n = 40) published in Machtolf *et al.* [10]

The helium exposure time (180 s) was sufficient to ensure a state of unconsciousness and insensibility, until the pigs died exsanguinated. Immediately after stunning and during bleeding, none of the pigs had a positive response to corneal reflex-test or to a painful stimulus (nose prick-test).

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Figure 3 shows the norepinephrine and epinephrine concentrations in the sticking blood of the slaughtered pigs. The concentration of norepinephrine in the blood of Helium-pigs was significantly lower (159 570 \pm 11 055 pg/ml) than that of CO₂-pigs (393 200 \pm 195 520 pg/ml, p < 0.001).



Figure 3: Norepinephrine and epinephrine concentrations [pg/ml] in sticking blood after helium (n = 40) and CO₂ (n = 40) stunning published in Machtolf *et al.* [10]

A similar result was detected for epinephrine, as the Helium-pigs had mean concentrations of 75 580 ± 34 340 pg/ml and CO₂-pigs 121 630 \pm 42 170 pg/ml (p < 0.001). Although the treatment of the pigs before stunning was not exactly the same, these preliminary results indicate that CO₂ atmosphere could produce a stronger endocrine reaction than helium stunning. High levels of catecholamines after CO₂ stunning were also reported by Machold *et al.* and Nowak *et al.* [6, 11].

The interpretation of the concentration of such hormones has to be performed with great care as endocrine reactions are not only found in response to stress and pain but also to low blood pH-values and hypoxia [11, 12, 13,14, 15]. Nevertheless, even if the hormone levels cannot directly be related to perceived stress of the animals, higher levels of norepinephrine and epinephrine have an influence on the quality of meat. Based on the behavior observations, the use of helium is a more gentle stunning method from the animal welfare point of view.

Meat and carcass quality

The meat quality parameters and the sensory evaluation are shown in Table 1. An influence of the stunning procedure was observed in the significant difference of pH_{45} and muscle temperature T_{45} values. After Helium stunning the pH_{45} values were 6.6 \pm 0.2 whereas after CO₂ stunning pH_{45} of 6.4 \pm 0.3 (p< 0.01) were found.

The mean T_{45} of the CO₂-pigs (40.5 °C) was significantly higher than that after helium stunning (39.6 °C, p < 0.001).

The cause of higher muscle temperatures and lower pH_{45} -values are higher levels of catecholamines in the blood. The hormones norepinephrine and epinephrine stimulate the *premortal* metabolism and led to a high *postmortal* glycolysis rate [16,17]. This influenced the *postmortal* pH extend and the carcass temperature.

Table 1: Meat quality parameters measured in the *Musculus semimembranosus* after helium- and CO_2 stunning, and sensory evaluation (1 = unacceptable to 6 = very good)

	Helium-pigs		CO ₂ -pigs		
Parameter	mean	sd	mean	sd	р
pH ₄₅	6.6	0.2	6.4	0.3	**
T ₄₅ (°C)	39.6	0.4	40.5	0.6	***
EC ₂₄ (<i>mS/cm</i>)	3.5	1.2	4.0	2.0	ns
Grill loss (%)	25.5	4.2	25.9	4.0	ns
Sensory evaluation					
Flavour	4.1	0.8	4.2	0.6	ns
Juiciness	4.7	0.6	4.6	0.7	ns
Tenderness	4.4	0.8	4.5	0.8	ns

 $_{45}$ = 45 minutes *post mortem*; $_{24}$ = 24 hours *post mortem*; T= muscle temperature; EC₂₄ = electrical conductivity; sd = standard deviation; p = significant differences (ns = not significant, *p < 0.05, **p < 0.01; ***p < 0.001)

The further measurements of EC_{24} , grill loss revealed no significant differences. The sensory assessment of both groups was almost identical.

After cutting the carcasses, ecchymosis were only found on one surface of *Musculus longissimus dorsi* after CO_2 -stunning. In contrast to stunning with argon [5], carcass quality deficiencies in the form of ecchymosis did not occur in pigs stunned with helium.

IV. CONCLUSION

This study confirmed aversive behavior of pigs in the commercially used 90 % CO_2 atmosphere stunning. By applying helium (> 95 %) instead of CO_2 , no aversive reactions can be observed. This makes helium stunning a more gentle method.

On the other hand, the pH_{45} and T_{45} presented higher values with Helium. In contrast to stunning with argon, ecchymosis did not occur after helium

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stunning. For a commercial use of helium or inert gas-mixtures further investigation of economic and animal welfare aspects are recommended.

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