

Stunning slaughter pigs using the inert gas helium

Machtolf M.^{1*}, Moje M.¹, Troeger K.¹ and Bülte M.²

¹ Max Rubner-Institut, Department of Safety and Quality of Meat, Kulmbach, Germany

² Institute of Veterinary Food Science, Justus-Liebig-University, Gießen, Germany

*Muriel.Machtolf@mri.bund.de

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-values, muscle temperature at 45 min p.m. (pH₄₅/T₄₅) and electrical conductivity at 24 h p.m. (EC₂₄) were measured. After that, the presence of ecchymosis in common meat cuts was recorded. Three days p.m. 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 CO₂ hyperventilated, revealed retreat or escape attempts and vocalization. Stunning pigs with CO₂ 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 CO₂ stunning.

I. INTRODUCTION

In most countries, carbon dioxide (CO₂) 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 CO₂ 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 cross-breeds) 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/m³), 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.

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, Uta, 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 grilled at 160 °C for 3 min and 11 s. The grill loss 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 CO₂ showed retreat and escape attempts as well as hyperventilation, before the loss of balance took place. Also vocalizations occurred only during CO₂ 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 CO₂ 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 CO₂ -pigs showed grade 2. The convulsion duration during Helium stunning was longer than during CO₂ stunning. This effectively balance each other out and the differences are insignificant. (Fig. 2).

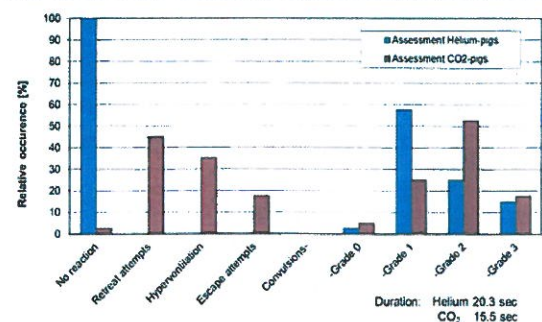


Figure 2: Percentage of pigs showing reactions and convulsions (grade 0 - 3) during stunning with helium (n = 40) and CO₂ (n = 40) published in Machtolf *et al.* [10]

The helium exposure time (180 s) was sufficient to ensure a state of unconsciousness and insensibility, until the pigs died of exsanguination. Immediately after stunning and during bleeding, reflex tests were negative for all animals.

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, $p < 0.001$).

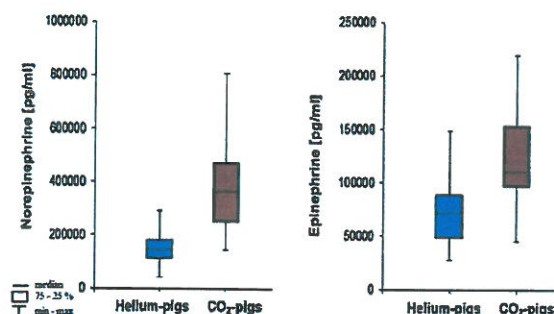


Figure 3: Norepinephrine and epinephrine concentrations [pg/ml] in sticking blood after helium (n = 40) and CO₂ (n = 40) stunning published in Machtoft *et al.* [10]

A similar result was detected for epinephrine, as the Helium-pigs had mean concentrations of $75\,580 \pm 34\,340$ pg/ml and CO₂-pigs $121\,630 \pm 42\,170$ pg/ml ($p < 0.001$). Because the treatment of the pigs before stunning was nearly the same, the results showed that CO₂ atmosphere effected 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 concentrations of these hormones do not permit an unequivocal answer the question of whether the stunning is the cause of stress to the pigs [11]. A endocrine reaction is found during stress and pain but also during low pH-values in the blood or hypoxia [12, 13,14, 15]. Even if the interpretation not result in a definitive statement, higher level of norepinephrine and epinephrine has an influence in meat quality. Because of the behavior observations, from the animal welfare point of view the use of helium, is a gentle stunning method.

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 pH₄₅ and muscle temperature T₄₅ values. After Helium stunning the pH₄₅ values were 6.6 ± 0.2 whereas after CO₂ stunning pH₄₅ of 6.4 ± 0.3 ($p < 0,01$) were found. The mean T₄₅ 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 pH₄₅-values are higher levels of catecholamines in the blood. The hormones norepinephrine and epinephrine stimulate the *premort* metabolism and led to a high *postmort* glycolysis rate [16,17]. This influenced the *postmort* pH extend and the carcass temperature.

Table 1: Meat quality parameters measured in the *Musculus semimembranosus* after helium- and CO₂ stunning, and sensory evaluation (1 = unacceptable to 6 = very good)

Parameter	Helium-pigs		CO ₂ -pigs		p
	mean	sd	mean	sd	
pH ₄₅	6.6	0.2	6.4	0.3	**
T ₄₅ (°C)	39.6	0.4	40.5	0.6	***
EC ₂₄ (mS/cm)	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 EC₂₄, 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 CO₂-stunning. In contrast to stunning with argon [5], carcass quality deficiencies in the form of ecchymosis did not occur in pigs stunned with helium. Based on the resulting meat and carcass quality, helium has a potential to be an alternative gas for stunning pigs.

IV. CONCLUSION

This study confirmed aversive behavior of pigs in the commercially used 90 % CO₂ atmosphere stunning. By applying helium (> 95 %) instead of CO₂, no aversive reactions can be observed. This makes helium stunning a more gentle method, resulting in an improved meat quality as measured by pH₄₅ and T₄₅. In contrast to stunning with argon, ecchymosis did not occur after helium stunning.

This research showed the feasibility to stun pigs in helium. For a commercial use of helium or inert gas-mixtures further investigation of economic and animal welfare aspects are recommended.

ACKNOWLEDGEMENTS

This study was supported by Tönnies Research, Organization for the Promotion of Research on the Future of Animal Welfare in Livestock. We would like to thank the staff of the Department of Safety and Quality of Meat for providing assistance in stunning pigs and for all the laboratory work.

REFERENCES

1. Raj, A.B.M., Gregory, N.G. (1990). Effect of rate of induction of carbon-dioxide anesthesia on the time of onset of unconsciousness and convulsions. *Research in Veterinary Science* 49: 360-363.
2. Hartung, J., Nowak, B., Waldmann, K. H., Ellerbrock, S. (2002). CO₂ stunning of slaughter pigs: Effects on EEG, catecholamines and clinical reflexes. *Deutsche Tierärztliche Wochenschrift* 109:135-139.
3. Dalmau, A., Rodriguez, P., Llonch, P., Velarde, A. (2010). Stunning pigs with different gas mixtures: aversion in pigs. *Animal Welfare* 19: 325-333.
4. Llonch, P., Dalmau, A., Rodriguez, P., Manteca, X., Velarde, A. (2012). Aversion to nitrogen and carbon dioxide mixtures for stunning pigs. *Animal Welfare* 21: 33-39.
5. Troeger, K., Machold, U., Moje, M., Behrschmidt, M. (2005). Gasbetäubung von Schweinen: Ein Vergleich von Kohlendioxid, Argon und einer Stickstoff-Argon-Mischung bezüglich der Schlachtkörper- und Fleischqualität. *Fleischwirtschaft* 85: 109-111.
6. Machold, U., Troeger, K., Moje, M. (2003). Gasbetäubung von Schweinen - Ein Vergleich von Kohlendioxid, Argon, einer Stickstoff-Argon-Mischung und Argon/ Kohlendioxid (2-stufig) unter Tierschutzaspekten. *Fleischwirtschaft* 83: 109-114
7. Raj, A.B.M. & Gregory, N.G. (1995). Welfare implications of the gas stunning of pigs. 1. Determination of aversion to the initial inhalation of carbon dioxide or argon. *Animal Welfare* 4: 273-280.
8. Döpjan, S., Schön, P.C., Puppe, B., Tuchscherer, A., Manteuffel, G. (2008). Differential vocal responses to physical and mental stressors in domestic pigs (*Sus scrofa*). *Applied Animal Behaviour Science* 114: 105-115.
9. Schäffer, D.v.B., (2005). Vokalisation von Schlachtschweinen. Ein Indikator für die Bewertung der Tiergerechtigkeit des Handlings an Betäubungsanlagen. *Fleischwirtschaft* 9: 37-44.
10. Machtof, M., Moje, M., Troeger, K., Bülte, M. (2013). Betäubung von Schlachtschweinen mit Helium und im Vergleich mit Kohlendioxid. *Fleischwirtschaft* 93: 118-124.
11. Nowak, B., Mueffling, T.V., Hartung, J. (2007). Effect of different carbon dioxide concentrations and exposure times in stunning of slaughter pigs: Impact on animal welfare and meat quality. *Meat Science* 75: 290-298.
12. Manteca, X. (1998). Neurophysiology and assessment of welfare. *Meat Science* 49, Supplement 1: S205-S218.
13. Forslid, A. und Augustinsson, O. (1988). Acidosis, hypoxia and stress hormone release in response to one-minute inhalation of 80% CO₂ in swine. *Acta Physiologica Scandinavica* 132: 223-231.
14. Shaw, F. D. und Tume, R. K. (1992). The assessment of pre-slaughter and slaughter treatments of livestock by measurement of plasma constituents—A review of recent work. *Meat Science* 32: 311-329.
15. Warriss, P. D., Brown, S. N., Adams, S. J. M., Corlett, I. K. (1994). Relationship between subjective and objective assessments of stress at slaughter and meat quality in pigs *Meat Science* 38: 329-340.
16. Hambrecht, E., Eissen, J.J., Nooijen, R.I.J., Ducro, B.J., Smits, C. H. M., den Hartog, L.A., Verstegen, M.W.A. (2004). Preslaughter stress and muscle energy largely determine pork quality at two commercial processing plants. *Journal of Animal Science* 82: 1401-1409.
17. D'Souza, D.N., Dunshea, F.R., Warner, R.D., Leury, B. J. (1998). The Effect of Handling Pre-Slaughter and Carcass Processing Rate Post-Slaughter on Pork Quality. *Meat Science* 50: 429-437.