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Pre-slaughter management in Northeast Brazil and the effects on thermophysiological indicators in pigs and pH_{45}[1]

Manejo pré-abate no Nordeste do Brasil e efeitos nos indicadores termofisiológicos dos suínos e pH_{45}

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HIGHLIGHTS:

Transport and unloading period presumably affected pig rectal temperature and respiratory rate.
The enthalpy index showed that the pigs were under heat stress.
The pH_{45} of the pork showed tendencies toward the DFD (dark, firm, and dry) type defect.

ABSTRACT: Inadequate pre-slaughter handling practices may compromise animal welfare and pork quality. This study aimed to evaluate the effects of transport period (TT), unloading period (UT), and pre-slaughtering period (WT) on animal thermophysiological indicators and pork pH using a multivariate approach. This study was conducted in a slaughterhouse located in Aquiraz, Ceará state, Brazil. A total of 60 mestizo pigs with 107 ± 5 kg body weight were distributed into three experimental groups: group A (TT = 180 min, UT ≤ 10 min, and WT = 24 hours), group B (TT = 60 min, UT ≤ 15 min, and WT = 18 hours), and group C (TT = 45 min, UT ≤ 5 min, and WT = 24 hours). Canonical discriminant analysis was implemented and the differences between treatments are represented graphically. The first two components accounted for 97% treatments. The discriminant analysis showed that group C presented multivariate differences in relation to others, particularly with high rectal temperature before slaughter (38.4 ± 0.23 °C) and respiratory rate (131 ± 7.0 breaths min^{-1}) and rectal temperature (39.7 ± 0.18 °C) upon arrival at the slaughterhouse. Transport period (TT), unloading period (UT), and thermal stress of the facilities influenced physiological variables, such as rectal temperature and respiratory rate. Furthermore, the physiological stress variables also favored cuts with basic pH, resulting in quality loss.

Key words: swine industry, pork quality, animal transport

RESUMO: Práticas inadequadas de manejo antes do abate podem comprometer o bem-estar animal e a qualidade da carne suína. Este estudo visou avaliar os efeitos do período de transporte (TT), período de descarga (TD), e período de pré-abate (TE) nos indicadores termofisiológicos animais e no pH da carne suína, utilizando uma abordagem multivariada. Este estudo foi realizado num matadouro localizado em Aquiraz, Ceará, Brasil. Um total de 60 suínos mestiços com 107 ± 5 kg de peso corporal foram distribuídos em três grupos experimentais: grupo A (TT = 180 min, TD ≤ 10 min, e TE = 24 horas), grupo B (TT = 60 min, TD ≤ 15 min, e TE = 18 horas), e grupo C (TT = 45 min, TD ≤ 5 min, e TE = 24 horas). A análise discriminante canônica foi implementada e as diferenças entre tratamentos são representadas graficamente. Os dois primeiros componentes foram responsáveis por 97% dos tratamentos. A análise discriminante mostrou que o grupo C apresentou diferenças multivariadas em relação aos outros, particularmente com temperatura retal elevada antes do abate (38,4 ± 0,23 °C) e frequência respiratória (131 ± 7,0 respirações min^{-1}) e temperatura retal (39,7 ± 0,18 °C) à chegada ao matadouro. O período de transporte (TT), período de descarga (TD) e o stress térmico das instalações influenciaram variáveis fisiológicas, tais como a temperatura retal e a frequência respiratória. Além disso, as variáveis de estresse fisiológico também favoreceram cortes com pH básico, resultando em perda de qualidade.

Palavras-chave: indústria suína, qualidade da carne suína, transporte animal

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Introduction

Pre-slaughter operations comprise complex steps from farms to moments before animal slaughter in abattoirs (Faucitano, 2018). Currently, the efficiency of these operations has become a concern for the swine industry due to eminent animal welfare issues and associated production losses (Voslarova et al., 2017; Romero et al., 2022).

Brazilian swine industry loses approximately US$160,000 annually per slaughterhouse (Reis et al., 2015). In this sense, most studies have focused on proposing solutions for this stage. However, other knowledge gaps (such as waiting period management) in pre-slaughter management practices may pose risks to animal welfare and industry profitability.

Pre-slaughter waiting mainly aims to recover the animals from stress and/or physical exhaustion caused by previous stages (Faucitano, 2018). Studies have associated increased economic losses caused by meat defects with the inadequate management of animal dwell time during the waiting period and the thermal comfort of the facilities during this period (Rioja-Lang et al., 2019; Driessen et al., 2020; Machado et al., 2022).

Furthermore, pre-slaughter pig operations comprise a set of actions with multifactorial and dynamic storylines. Thus, the analysis of animal physiological data in this area is complex and can be explained only by univariate analysis, which does not consider all the (co) variations between the previous operations and variables studied. This study aimed to evaluate the effects of transport period (TT), unloading period (UT), and pre-slaughtering period (WT) on animal thermophysiological indicators and pork pH using a multivariate approach.

Material and Methods

The study was conducted between March 10 and March 30, 2020, in a slaughterhouse located in Aquiraz municipality, Fortaleza metropolitan region, Ceará state, Northeast Brazil (3° 54' 9" S, 38° 23' 19" W and 14 m altitude). All procedures were performed without compromising the company's usual rhythm and following the guidelines prescribed by the Animal Care and Use Committee of the Universidade Federal do Ceará under protocol no. 5293280720.

Sixty 150-day-old crossbred pigs (Large White × Landrace × Duroc, average body weight: 107 ± 5.0 kg) collected from three farms of a single company were used in the study. The animals were separated into three groups, A, B, and C, adopting the following criteria: 45 min ≤ TT ≤ 180 min, 5 min ≤ UT ≤ 15 min, and 18 hours ≤ WT ≤ 24 hours.

The travel distances from the farms of origin to the slaughterhouse were 79, 50, and 30 km for groups A, B, and C, respectively. The investigation was conducted using a randomized block design. Each animal was evaluated in an experimental unit.

Animals were identified with non-toxic ink marks on the dorsal region in the lairage compartment 10 ± 3 min after unloading and assigned to the three experimental treatments described in Table 1.

Table 1. Characteristics of the groups in the pre-slaughter routine according to the transport, unloading, and waiting periods

Groups	Transport period (min)	Unloading period (min)	Waiting period (h)
Group A	180	≤10	24
Group B	60	≤15	18
Group C	45	≤5	24

Pig management in the pre-loading and transport periods was standardized in all trips according to the company commercial standards. Feeding was stopped before the loading started (120 ± 20 min). During loading, all animals were guided using flags. Trips were made in a cargo truck, Ford® 1519, with a double-deck Triel® - HT body model containing six compartments per level floor and 13 ton load capacity. More details are described in previous studies (Machado et al., 2021).

The vehicle was not equipped with an air-conditioning system or environmental controls, and the compartments did not contain any type of bedding or drinking water supply system. During pig loading, "load cooling" occurred, which involved wetting the pigs loaded on the truck with water by a farm employee using a hose. This is a standard practice in the region, which aims to reduce the impact of heat stress during transport (Pinheiro et al., 2020).

During the waiting period, the animals were housed in masonry facilities with aluminum tile roofs, 2.25 m high, 3.0 m ridge and 0.2 m eaves, with seven stalls 4.60 m wide and 5.0 m long (area of 28 m^2), without air-conditioning. The stalls were equipped with a drinking fountain to promote a hydric diet for the housed animals.

The thermophysiological indicators of thermal stress used were rectal temperature (°C), skin temperature (°C), and respiratory frequency (breaths min^{-1}). All indicators were measured approximately 15 ± 10 min after unloading the trailer and 30 ± 10 min before slaughter. Respiration rate and skin temperature were measured before immobilizing the pigs to avoid the effect of immobilization stress.

Respiration rate was measured by trained experts by counting the flank movements for 15 s and multiplying it by four to obtain the respiration rate as movements per minute (Sampaio et al., 2021). Both observers recorded respiration rate measurements simultaneously; therefore, an average was calculated for each animal based on the two measurements provided by the experts, which were then used in the statistical analysis (Machado et al., 2021).

A Fluke® model TiS10 infrared thermal imager was used to obtain the skin temperature. Thermographic images were captured at 1 m distance from the animal. The emissivity of the equipment was adjusted to 0.98, which is the value indicated for biological tissues (Soerensen et al., 2014). Subsequently, the thermographic images were analyzed using FlukeView® software.

Finally, the rectal temperature of the pigs was measured after restraint using a "pipe" (instrument routinely used in swine farms) with a digital skewer thermometer with ± 0.5 °C accuracy (−10 to 100 °C, AK05, AKSO), inserted directly into the rectum at a depth where the bulb was in contact with the rectal mucosa (Pereira et al., 2022).

The thermophysiological indicators were defined as follows: RRA, respiratory rate after waiting (30 min before slaughtering); RRB, respiratory rate before waiting (15 min after unloading); RTA, rectal temperature after waiting; RTB, rectal temperature before waiting; STA, skin temperature after waiting; and STB, skin temperature before waiting.

Inside the pre-slaughter waiting facilities, air temperature and relative air humidity were recorded every 10 min by data loggers (Onset, U23-001 HOBO Pro v2, Massachusetts, USA) positioned at the center of the lairage at the height of the pigs. The microclimate was characterized by the specific enthalpy (Rodrigues et al., 2011) using Eq. 1.

$$H = 1.006AT + \frac{RH}{Pb} \times 10^{7.5AT(237.3+AT)^{-1}} \times \left(71.28 + 0.052AT\right) \quad (1)$$

where:

H - enthalpy (kJ kg^{-1} dry air);
AT - air temperature (°C);
RH - relative humidity of the air (%); and,
Pb - local barometric pressure (mm Hg).

In this study, the barometric pressure used was 760 mm Hg, and the critical enthalpy limit range for the species was considered an excellent thermal comfort value for pigs: 18–21 °C air temperature and 50–70% relative air humidity, according to Silva et al. (1999). The pigs were placed in a stunning box, stunned by electronarcosis, and slaughtered by exsanguination. At the end of bleeding (approximately 40 s), pH was measured at 45 min using a pH meter (Hanna Instruments® model HI981036 and ±2% reading plus 0.02 NTU) to measure the longissimus dorsi and semispinalis capitis.

The hypothesis of normality and homoscedasticity of residual variances was verified using the Shapiro–Wilk and Levene tests ($p \leq 0.05$) in the statistical program Statistical Analysis System - SAS® (SAS, 2012). The univariate procedure was used to perform a basic descriptive statistical analysis of the groups. Tukey's adjustments were used to compare treatment means ($p \leq 0.05$). Canonical discriminant analysis was performed using the CANDISC procedure to understand the degree of multivariate similarity between groups using the statistical model (Eq. 2). A graph showing 95% confidence ellipses of the mean vectors for each treatment with the first two canonical variables (Can1 and Can2) to visualize the multivariate trends of all treatments together was created using R Core Team (2015).

$$Y_{iktr} = \mu_t + B_{kt} + T_{it} + E_{iktr} \quad (2)$$

where:

Y_{iktr} - multivariate vector of observations on variable t for treatment i, in replicate r, of block k;

μ_t - general means multivariate vector for variable t;

B_{kt} - multivariate vector of effects in block k on variable t (random effect of the collection week);

T_{it} - multivariate vector of effects in treatment i on variable t; and,

E_{iktr} - multivariate vector for random errors associated with the observation vector Y_{iktr}.

Results and Discussion

The data in Table 2 shows the averages of the variables studied to understand the changes in physiology of the animals evaluated. The STB does not provide significant interpretation, probably due to "load cooling," where the pigs are wetted with a hose before transport. However, the RRB, which is directly associated with unloading- and transport-mediated stress in all animals, was high, with a significant difference between groups B and C. This result can be attributed to transport-mediated stress and short UT. This may have resulted in less calm and possible more aggressive handling by the handlers (Machado et al., 2022).

It is worth noting that the RTB and RRB of the animals in group C were higher than those in the animals in group A. This result suggests that this is directly related to the short TT (less time available for animal homeostasis) and particularly fast disembarkation (≤5 min), as rough handling increases stress on animals and accident rates during these operations (Dalla Costa et al., 2019).

There is no ideal UT due to the various conditions and combinations in which this operation can be performed (such as animal loading/unloading facility type, driving method, employees involved, environmental conditions, operation duration, and country legislation). However, previous studies suggest the hypothesis that rapid and abrupt animal handling tends to increase stress, particularly if questionable handling methods (such as electric prods) are used for animal welfare (EFSA et al., 2020).

However, according to Dalla Costa et al. (2007), the normal recommended TT for pigs is between one and three hours. Short periods may be insufficient for the animals to recover from stress caused by multiple factors during loading and transport, including inadequate microclimatic conditions (Miranda-de la lama et al., 2021), mechanical vibrations (Donofre et al., 2014), and human–animal interactions (such as handling) (Mota-Rojas et al., 2020). Thus, the usual conditions of the slaughtering operations may have negatively influenced

Table 2. Average skin temperature before waiting (STB), respiratory rate before waiting (RRB), rectal temperature before waiting (RTB), skin temperature after waiting (STA), respiratory rate after waiting (RRA), rectal temperature after waiting (RTA), pH_{45} of the shoulder and loin, air temperature (AT), relative air humidity (RH), and specific enthalpy of the pre-slaughter facility (H)

Variables	Groups A	Groups B	Groups C	CV (%)
STB (°C)	38.2±0.32 a	37.6±0.42 a	36.3±0.69 a	6.00
RRB (breaths min^{-1})	126±6.00 ab	122±6.00 b	131±7.00 a	27.0
RTB (°C)	36.5±0.21 b	38.5±0.30 a	39.7±0.18 a	5.00
STA (°C)	35.5±0.34 a	35.9±0.49 a	35.1±0.60 a	6.00
RRA (breaths min^{-1})	88±4.00 a	89±4.00 a	73±3.00 b	25.0
RTA (°C)	35.8±0.14 b	37.5±0.46 a	38.4±0.23 a	4.00
Pork shoulder pH_{45}	6.33±0.08 a	6.30±0.09 a	6.21±0.06 a	6.00
Loin pH_{45}	6.34±0.07 a	6.25±0.08 a	6.21±0.08 a	5.00
AT (°C)	29.0±0.36 a	27.5±0.33 a	28.1±0.13 a	8.00
RH (%)	84±1.24 a	82±0.89 ab	77±0.60 b	13.0
H (kj kg^{-1} dry air)	81±0.75 a	74±1.37 b	73±0.84 b	10.0

Averages followed by same lowercase letters (horizontal) do not differ statistically from each other according to the Tukey test ($p \leq 0.05$); CV - Coefficient of variation the documented thermophysiological indicators upon arrival at the slaughterhouse, particularly in the animals that traveled ≥50 km; consequently, the TT will increase in these animals, which provides high stress due to the factors inherent to transport and a possible loss of pork quality when gauging pH 45 min. (Ochove et al., 2010; Pereira et al., 2017; Faucitano, 2018; Miranda-de La Lama et al., 2021).

Furthermore, high stress due to transport distance causes PSE (pale, soft, and exudative) and DFD (dark, firm, and dry) quality defects (Faucitano, 2018). However, the pH should be measured 24 hours after slaughter, which is not possible due to slaughterhouse logistics. Thus, further research on thermophysiological responses combined with behavioral analyses and load cooling aspects at the level of commercial swine transport is recommended for further elucidation of the results.

In this study, all pre-and post-transport practices (such as handling, infrastructure, employees, trucks) were standardized across all operations to minimize the risk of confounding factors, making the observed results genuine. This study provides information that can contribute to the development of efficient strategies for swine slaughterhouses in Northeast Brazil, as well as local guidelines and public policies for stress mitigation, including heat stress and its impact on production losses in the local swine industry.

Overall, the RRA was outside the ideal range for the species (resting pigs) in the three groups evaluated (88, 89, and 73 breaths min^{-1} in groups A, B, and C, respectively). According to Radostits et al. (2002), respiration rates should ideally range between 15 and 25 breaths min^{-1}. Ferreira et al. (2014) have also stated that respiratory rate > 40 breaths min^{-1} strongly indicates hyperthermia. In this study, the documented enthalpy of groups A, B, and C (81, 74, and 73 kj kg^{-1} dry air, respectively), are outside the optimal thermoregulation zone for pigs (60–68 kj kg^{-1} dry air; Silva, 1999).

Interestingly, air humidity of the environment strongly influences enthalpy, making the environment stressful for the swine species, considering its physiology of heat exchange as an environment (Rioja-Lang et al., 2019). Thus, the management of pre-transport and waiting load cooling, which is common in this region, compromises pig welfare by causing both cold and heat stress, which can be ratified by the RTA, RTB, STA, STB, and RRA.

Table 3 shows the standardized canonical coefficients for the total sample and the total variance explained by each canonical variable. In this study, the first canonical variable (Can 1) or Fisher discriminant function and the second canonical variable (Can 2) explained 73.18 and 23.82% variation, respectively; thus, the variables studied explained 97% variation, which can be considered reasonable, favoring accurate interpretation.

In the extraction process of Can 1, a high weighting was observed in the scores of the bioclimatological variables: temperature, relative air humidity, and enthalpy; the thermophysiological variables: RTB and RRA; and consequently, the pH of the shoulder. In Can 2, the highest coefficients observed were also those of the bioclimatological variables of the waiting facilities, in the thermophysiological

Table 3. Standardized canonical coefficients for the total sample and total variation explained by skin temperature before waiting (STB), respiratory rate before waiting (RRB), rectal temperature before waiting (RTB), skin temperature after waiting (STA), respiratory rate after waiting (RRA), rectal temperature after waiting (RTA), pH_{45} of the shoulder and loin, air temperature, relative air humidity, and enthalpy

Variable	Canonical variable (Can)	
	Can1	Can2
STB	0.023	0.034
RRB	−0.054	0.182
RTB	−0.615*	0.430*
STA	−0.190	0.028
RRA	0.294*	0.338*
RTA	−0.174	0.305*
Pork shoulder pH_{45}	0.266*	0.107
Loin pH_{45}	−0.165	−0.265*
Air temperature	−14.90*	−21.01*
Relative air humidity	−6.700*	−9.156*
Enthalpy	13.89*	19.32*
Variation (%)	73.18	23.82
Cumulative change (%)	73.18	97.00

* - Highly weighted scores of > 0.20

variables: RTB, RTA, RRA, and loin pH. These variables are related to the physical stress of transport, unloading management, and heat-mediated thermal stress.

Figure 1 shows a two-dimensional representation of the contribution of each variable to the dissimilarity between groups. According to Mingoti (2005), the reliability of graphic representation is evidenced when the percentage of accumulated variance is 80% (97% in this study), showing that group A, from a multivariate perspective, is similar to group B. Interestingly, the groups are intersected owing to their proximity and similarity. The variables that contributed the most to this assumption were RRA, RH, pH_{45} shoulder and loin, and enthalpy.

Therefore, the discriminant analysis highlighted the dissimilarity of groups A and B to group C. Groups A and B showed an assumed response in pork quality while evaluating the loin and shoulder pH_{45}, where mean vectors related to

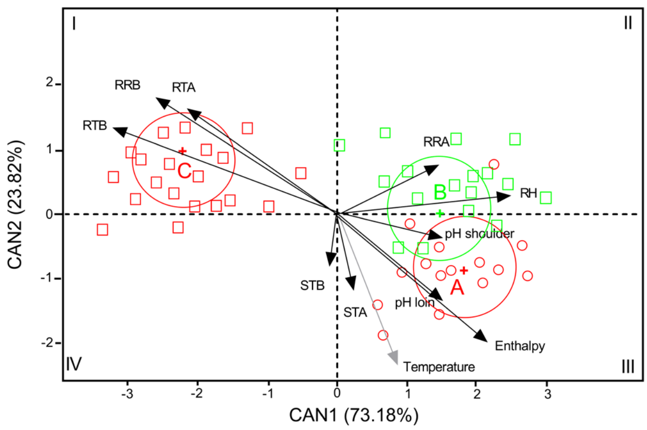

Figure 1. Dissimilarity between the groups evaluated by Can1 × Can2. STB, skin temperature before waiting; RRB, respiratory rate before waiting; RTB, rectal temperature before waiting; STA, skin temperature after waiting; RRA, respiratory rate after waiting; RTA, rectal temperature after waiting; air temperature, enthalpy, pH_{45} loin and shoulder

meat pH overlapped across the groups, particularly in group A. This result may be related to the transport and unloading periods associated with enthalpy values and high fasting period, suggesting a high probability of developing DFD-type defects in the shoulder (Faucitano, 2018; Driessen et al., 2020; EFSA et al., 2020).

Finally, the transport distance, whether short or long, negatively influences animal physiology and meat quality due to a short adaptation period to transport conditions, as well as the time when the animals arrive at the slaughterhouse (Ochove et al., 2010; Miranda-de la Lama et al., 2021). These situations can promote quality losses (Nannoni et al., 2014), implying reduced welfare conditions, particularly when associated with waiting in facilities that do not favor thermal comfort (Machado et al., 2022).

Conclusions

1. The first two components accounted for 97% treatments. The discriminant analysis showed that group C presented multivariate differences in relation to the others, particularly with high RTA (38.4 ± 0.23 °C), RRB (131 ± 7.0 breaths min^{-1}), and RTB (39.7 ± 0.18 °C).

2. TT and UT, as well as the thermal stress of the facilities influenced the physiological variables such as rectal temperature and respiratory rate.

3. The physiological stress variables also favored a tendency for cuts with basic pH, resulting in quality losses.

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