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The effect of transportation and pre-slaughter detention on quality of pig meat

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ABSTRACT

This research aimed to determine the influence of stress of various etiologies in pigs caused by transportation and pre-slaughter conditions on meat quality. For this purpose, pigs were divided into 11 groups within two meat processing enterprises, depending on the duration of transportation (short, long without breaks and long with breaks) and the conditions of keeping animals before slaughter. Also, within the two groups were created two subgroups with pigs of different breeds. A total of 156 pigs were studied. Blood was collected from all pigs to determine cortisol and lactate levels, and a sample of meat from the longest back muscle. The pH of the meat was determined at different stages of its maturation, and the weight loss of the sample was determined. As a result of the research, it was found that the highest quality pork was obtained from pigs that experienced lower levels of stress before slaughter. Keeping pigs for 10 – 14 hours before slaughter without access to water and food resulted in higher stress levels, which were probably expressed in higher blood concentrations of cortisol and lactate. Pigs' access to food and water during pre-slaughter retention allows for high-quality meat by reducing the influence of stress. Pigs' access to water before pre-slaughter does not affect the stress level but positively affects the loss of meat weight during maturation. The higher the concentration of lactate in pigs' blood, the faster the pH of the meat decreases after slaughter, which negatively affects its quality and moisture retention. If there is a long-term transport of pigs, there is no rest stop that can significantly reduce stress levels in pigs. Duration of transportation of pigs does not correlate with stress levels, as the conditions before slaughter content.

Keywords: pork, meat quality, pig stress, cortisol, lactate

INTRODUCTION

According to the concept of "The Only Health", the quality of life of people depends on the safety and quality of the food they consume. For the safety and quality of meat to meet consumer expectations, one should relentlessly control it at all stages of production and circulation, as the final result depends on many factors. In particular, the conditions of keeping animals, feeding, veterinary treatments, pre-slaughter **[1]**, **[2]**, **[3]**, **[4]** and post-mortem factors **[5]**, **[6]**. Much remains unknown despite the significant amount of research on various factors directly or indirectly affect pork quality. Today it is known that the quality of pork depends on the amount of stress he had placed before the slaughter of pigs **[7]**, **[8]**, **[9]**, **[10]**, **[11]**. In particular, muscle carbohydrates (glucose and glycogen) under anaerobic oxidation are metabolised to lactate, which lowers the pH of the meat. Under stress, the lactate level in the blood increases significantly, and muscle glycogen content decreases. In this case, lowering the pH of the meat during its maturation will be too fast. As a result of fast hardening, muscle fibers are damaged in their structure. There is excessive loss of moisture and discolouration, thus decreasing the quality and expiration date of the meat **[10]**, **[12]**. Depending on the geographical location of the farm and technologies of preparation of pigs for slaughter, various methods of transportation and pre-slaughter keeping are practised. If the farm is located at a considerable distance from the slaughterhouse, it can be practised as transportation without stops and with breaks (long or short) for the rest of the animals. Also, in some cases, the

animals are transferred to a starvation diet; in others – the animals are fed and given access to water. There is also a different approach to the pre-slaughter keeping of animals.
In some cases, the slaughter of animals is practised immediately upon arrival; in others – animals are sent to quarantine of various durations – from several hours to one or several days. Animals are often accumulated during the day and slaughtered in the morning. In this period, at some slaughterhouses, animals are given access to water; on others to food and water, third animals are not watered or fed. In addition, they sometimes practise an approach in which they try not to mix animals from different farms or even different piggeries of one farm for transportation and pre-slaughter **[13]**. In others, cases are neglected, especially when forming a kept pig batch on small household farms.

Scientific Hypothesis

Theoretically, we assume that all of these factors can affect the stress experienced by animals. The level of stress will be different depending on the animal's conditions. In addition, we assume that the breed of pigs will directly influence stress. Hence the quality of pork will be different. This work includes the research results on the pig meat quality, depending on transport conditions, before slaughter maintenance and breed, which was our goal.

MATERIAL AND METHODOLOGY

The study was performed at the Department of Veterinary and Sanitary Inspection laboratory of S. Z. Gzhytskyi National University of Veterinary Medicine and Biotechnologies in Lviv.

Samples

The material for the research was pig blood and meat. In general, it was samples taken from 156. Blood samples were taken during the bleeding of the carcass in sterile tubes, and the meat was up to 30 minutes after the slaughter of pigs. One meat sample was taken from each carcass (weighing 450 – 500 g) from the longest back muscle in the area of 9 – 12 thoracic vertebrae. Meat samples were stored in a refrigerator at 4 ±2 °C. To obtain plasma, the blood was immediately centrifuged at three thousand rpm.

Chemicals

The cortisol content was determined in blood plasma by enzyme-linked immunosorbent assay using DRG (Germany) test kits. Hydrazine-glycine buffer pH 9.0, NAD+ (0.3 molar solution, pH 6.0) and lactate dehydrogenase solution (protein content 2 $mg.mL^{-1}$) were used to determine lactate (Khimreaktiv, Ukraine).

Animals and Biological Material

Samples were taken from two slaughterhouses located in the Lviv region of Ukraine. The experiment was performed from July – to September 2021. Before sampling, the attendants studied documents and conducted the initial inspection and weighing of pigs. Total experimental pigs were divided into 11 groups, as shown in Table 1. In 1 and 8 experimental groups, the number of studied animals was twice as large as in others because, in these groups, pigs of two breeds were included – Ukrainian Steppe White and Landrace. In the article, comparisons of the indicators received from pigs of the Ukrainian steppe were carried out white breed except for the part where the data of breed features are given.

Table 1 Division of pigs into groups, depending on the duration of transportation and detention conditions until slaughter.

Group No.	Number of samples, n	Transportation time		Breaks for transportation		Shutter speed		Diet		
		6 – 8 hours	0.5 – 1 hour	Without breaks	0.5 hours every 2 hours	up to 1 hour	10 – 14 hours	Without water and food	Only water	Food and water
1.	24	+		+		+		+		
2.	12	+		+			+	+		
3.	12	+		+			+		+	
4.	12	+		+			+			+
5.	12	+			+	+		+		
6.	12	+			+		+		+	
7.	12	+			+		+			+
8.	24		+	+		+		+		
9.	12		+	+			+	+		
10.	12		+	+			+		+	
11.	12		+	+			+			+

Before slaughter, all animals were analogues by sex (pigs), live weight kg and clinically healthy. All animals were kept unattended until the slaughter pig keeping system.

Transportation of pigs was carried out with the help of a specialised transport at the rate of 0.8 m^2 area per animal at a temperature environment not higher than 28 °C, for unloading animals used a bridge, which was installed so that there were no cracks and the angle of inclination to the surface did not exceed 20 °C. Upon arrival, the pigs crossed a corridor about 8 m long to the pre-slaughter room. To increase efficiency in unloading and moving pigs within the enterprise, employees' meat sticks are used by meat processing companies.

Before slaughter, all experimental animals were stunned with the help of an electric current.

Instruments

Stat-Fax analyzer (model 4300 ChroMate; USA).

Electronic laboratory scales (TBE-0.5, producer (Inter-Synthesis) Limited Liability Company; Ukraine).

pH meter TESTO 205 (Germany).

Centrifuge MICROmed (China).

Spectrophotometer ULAB 102 (China).

Laboratory Methods

The cortisol content was determined in blood plasma by enzyme-linked immunosorbent assay. Whole blood determined lactate content **[14]**. Meat samples were tested for pH at the first, third, and 12^{th} hours of storage using a pH meter. The quality of the meat was visually inspected and palpated, dividing it into three categories: 1) NOR (close to optimal quality indicators); 2) PSE (pale, soft, exudative); and 3) DFD (dark, solid, dry).

Sample preparation

Blood samples were taken during the bleeding of the carcass in sterile tubes, and the meat was up to 30 minutes after the slaughter of pigs. One meat sample was taken from each carcass (weighing 450 – 500 g) from the longest back muscle in the area of 9 – 12 thoracic vertebrae. Meat samples were stored in a refrigerator at 4 ±2 °C. To obtain plasma, the blood was immediately centrifuged at three thousand rpm.

Number of samples analysed: we analysed 156 blood samples and 156 samples of meat.

Number of repeated analyses: All measurements of instrument readings were performed two times.

Number of experiment replication: The number of repetitions of each experiment to determine one value was two times.

Design of the experiment: The experiment was performed from July to September 2021. Before sampling, the attendants studied documents and conducted the initial inspection and weighing of pigs. Research teams were formed based on the information received from suppliers of live pigs and available in the documents. The period of pigs' transportation (long or short) to the meat processing plant and the transportation conditions (with breaks for rest or without stops) were considered. In addition, we paid respect to the requirements of pre-slaughter keeping of pigs, access to feed and water, length of stay before slaughter. To ensure the purity of the experiment, the sex, breed and live weight of pigs were taken into account. In total, 11 experimental groups of pigs with different transport and pre-slaughter conditions were formed, as shown in Table 1. A total of 11 experimental groups of pigs were formed with different combinations of transport and pre-slaughter conditions, as shown in Table 1. Blood samples and the samples of the longest back muscle were taken after slaughter. After the preparation of the samples, studies were performed on the content of cortisol and lactate in the blood as markers of pre-mortem stress and the pH of meat and its natural weight loss. An organoleptic assessment of the quality of the meat was also performed.

Statistical Analysis

The ChroMate Manager software was used. The obtained digital data were processed in Excel (2010 professional +), determining the average arithmetic value (M), statistical error of the arithmetic mean values (m), the probability of the difference between the arithmetic means of the two variations series and correlation coefficient (r). The difference between the comparable values was significant for $p < 0.05$. The statistical reliability of the research results was provided by analyzing samples with the number from 12 to 24.

RESULTS AND DISCUSSION

As can be seen from the data in Table 2, the highest concentration of cortisol in the blood plasma of slaughter pigs was after their quarantine for 10 – 14 h at the slaughter point under starvation conditions. Thus, compared with blood samples taken from pigs slaughtered shortly after arrival, cortisol levels were higher by 36 – 41.4% ($p < 0.05$).

Table 2 The content of cortisol in pigs' blood, depending on the pre-slaughter conditions of storage and transportation.

Terms		Transportation					
		long without breaks		long with breaks of 0.5 hours every 2 hours		short	
Before slaughter		Slaughter on arrival	Slaughter in 10 – 14 hours	Slaughter on arrival	Slaughter in 10 – 14 hours	Slaughter on arrival	Slaughter in 10 – 14 hours
Without water and food	M ±m	245.2 ±21.31	333.4 ±22.84	217.2 ±22.94		208.6 ±13.14	294.9 ±14.97
	Min	122.1	190.4	78.6		107.9	155.2
	Max	369.0	408.8	310.0		250.4	355.7
Only water	M ±m		314.1 ±23.44		252.5 ±16.2		247.8 ±19.78
	Min		162.3		108.2		94.5
	Max		410.8		321.8		305.4
Food and water	M ±m		180.8 ±17.25		170.5 ±20.10		143.2 ±12.27
	Min		102.1		75.4		77.6
	Max		302.1		295.4		210.4

Note: lactose concentration (mmol.L^{-1}); n = 12.

In the group of pigs with free access to before slaughter water, the plasma cortisol concentration was slightly lower than in the blood of pigs without such access. However, the difference was in incredible values. The lowest hormone levels were found in the blood plasma of pigs that had access to water and food before slaughter. Yes, compared to animals, who were without water and food, the figure was lower by 45.8 – 51.4% (p <0.05), and compared to animals that had access only to water, by 42% (p <0.05).

We found that the time of transportation of pigs to the slaughter point slightly affected the level of cortisol in the blood plasma of pigs. In particular, comparing the average level of cortisol in the blood of transported pigs to the slaughterhouse for 6 – 8 hours and scored in a short period on arrival, we found a lower (15%) content compared to the figure obtained from pigs that were delivered to slaughter within 1 hour, but this difference was statistically unlikely.

Our research showed (Table 2) that the transportation of animals for an extended period with the use of pauses for rest allowed to reduce the level of cortisol in the blood of slaughter animals (by 5.7 – 19.6%), but probably a significant result (p <0.05) was recorded only when comparing groups in which they kept the exposure for 10 – 14 h at the slaughterhouse and free access of pigs to water. Unfortunately, it was impossible to form a group of pigs, transported for a long time with breaks for rest and kept based on pre-slaughter keeping without access to water and food.

In general, the cortisol level in the blood plasma of slaughter animals ranged quite widely (from 75.4 to 408.8 nmol.L^{-1}), depending on transportation conditions and before slaughter.

Studies of the lactate content in the blood of slaughter pigs showed (Table 3) that the highest concentration was found in pigs that were quarantined for 10 – 14 hours at the slaughterhouse. Thus, the content of lactate in the blood of such pigs was higher by 34.7 – 37.9% (p <0.05) compared with its content in the blood of pigs that were slaughtered immediately upon arrival. Notably, in the blood of pigs that had access to water and food during quarantine, the lactate concentration was 2 – 3 times lower (p <0.05). The access of pigs to water did not significantly reduce the lactate level in the blood.

Analyzing the lactate level in the blood of pigs, depending on the duration of their transportation to slaughter, we can say about the ambiguity of the results. Among the groups of pigs slaughtered immediately upon arrival at the slaughterhouse, the highest rate was obtained in the blood of animals after short-term transportation and the lowest among those given rest during long-term transport. However, the difference was incredible. There was also no significant difference in the lactate level in pigs' blood when comparing the two approaches to long-term transportation: with rest breaks and without them.

The obtained results showed that the level of lactate in pigs' blood ranged from 2.9 to 24.5 mmol.L^{-1}, which is above the upper limit of physiological fluctuations for this species. Statistical analysis showed a positive correlation between medium (r = 0.4) and high (r = 0.9) levels between the concentration of cortisol and lactate in the blood of pigs.

Table 3 The lactate content in pigs' blood, depending on the pre-slaughter conditions and transportation.

Terms		Transportation long without breaks		long with breaks of 0.5 hours every 2 hours		short	
Before slaughter		Slaughter on arrival	Slaughter in 10 – 14 hours	Slaughter on arrival	Slaughter in 10 – 14 hours	Slaughter on arrival	Slaughter in 10 – 14 hours
Without water and food	M ±m	12.4 ±1.70	16.7 ±1.09	11.4 ±1.50		13.2 ±1.20	18.2 ±1.04
	min	4.3	9.5	5.5		6.5	12.4
	max	24.5	23.8	20.1		19.1	21.6
Only water	M ±m		16.6 ±0.91		16.1 ±1.08		15.8 ±1.08
	Min		9.7		9.8		8.5
	max		20.5		21.0		20.1
Food and water	M ±m		8.3 ±0.97		7.5 ±1.23		5.8 ±0.95
	min		6.1		3.2		2.9
	max		18.5		19.5		13.4

Note: lactose concentration (mmol.L^{-1}); n = 12.

Figure 1 shows that the highest pH level of the longest back muscle was found in the group of pigs with lower levels of cortisol and lactate in the blood immediately after slaughter. The pH of pig meat with medium and high cortisol and lactate levels in the blood was more acidic (0.3 units; p <0.05). A similar trend persisted at other stages of meat maturation. From the first to the twelfth hour of meat maturation, the pH of the longest back muscle of pigs with medium and high levels of cortisol and lactate in the blood was more acidic (p <0.05), but the most pronounced difference was (0.7 – 0.8 units) one hour after slaughter. At 12 hours of meat maturation, the difference was 0.4 pH.

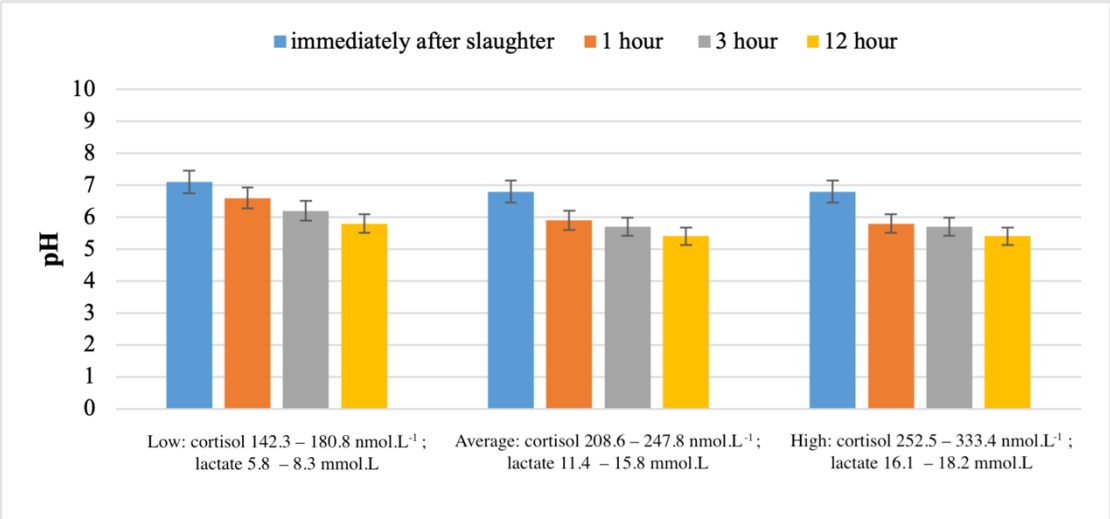

Figure 1 pH of the longest back muscle during maturation, depending on the level of cortisol and lactate in the blood. Note: n = 12. When dividing pigs into groups according to the level of cortisol and lactate into low, medium and high, our results were taken into account, not the physiological norm of these substances.

In meat selected from pigs with lower cortisol and lactate levels, a gradual decrease in pH to 12 hours after slaughter was observed (Figure 1). In contrast, in pig meat with medium and high levels of cortisol and lactate, there was a sharp decrease in pH during the first hour of maturation and a gradual up to 12 hours. No significant results were found between pigs with high and medium cortisol and lactate levels.

Table 4 shows data comparing the stress level in different breeds of pigs. The blood of Landrace pigs showed slightly lower cortisol and lactate concentrations than Ukrainian steppe white pigs, but the difference was unbelievable. The pH of the meat of both breeds of pigs at different stages of its maturation was also similar. Only the probably lower pH values (p <0.05) of the meat of pigs transported to the slaughterhouse within 0.5 – 1 h compared to those transported 6 – 8 h are noteworthy. Thus, one hour after slaughter, the meat's pH difference ranged from 0.6 to 0.7 units. However, the difference in performance is probably due to the peculiarities of the delivery of pigs, not their breed.

Table 4 The content of cortisol and lactate in the blood and pH of pig meat of different breeds.

Indicator		Transportation			
		6 – 8 hours without breaks		0.5 – 1 hour	
		Breed			
		Ukrainian white steppe	Landras	Ukrainian white steppe	Landras
Cortisol nmol.L^{-1}	M ±m	245.2 ±21.31	207.5 ±25.49	208.6 ±13.14	185.9 ±14.80
	min	122.1	81.4	107.9	103.6
	max	369.0	305.4	250.4	278.6
Lactate mmol.L^{-1}	M ±m	12.4 ±1.70	11.3 ±1.74	13.2 ±1.20	12.5 ±1.57
	min	4.3	3.2	6.5	4.1
	max	24.5	20.1	19.1	20.5
pH of meat after slaughter	M ±m	6.8 ±0.07	6.9 ±0.08	6.8 ±0.10	6.8 ±0.04
	min	6.4	6.1	5.7	6.5
	max	7.1	7.1	7.0	7.1
pH of meat 1 hour	M ±m	6.4 ±0.10	6.4 ±0.11	5.7 ±0.07	5.8 ±0.08
	min	5.9	5.9	5.3	5.1
	max	7.0	7.0	6.0	6.2
pH of meat 3 hours	M ±m	6.0 ±0.06	5.9 ±0.10	5.7 ±0.10	5.7 ±0.03
	min	5.84	5.4	5.3	5.3
	max	6.35	6.3	6.7	5.8
pH of meat 12 hours	M ±m	5.3 ±0.05	5.4 ±0.09	5.5 ±0.06	5.5 ±0.07
	min	5.1	5.0	5.2	5.1
	max	5.8	6.0	5.8	5.7

Note: n = 2.

Figure 2 shows the natural weight loss results of the longest back muscle sample during 12 hours of maturation. The most pronounced decrease in the mass of the longest back muscle samples was found in the groups of pigs that experienced the most significant pre-slaughter stress and had high cortisol and lactate levels in the blood. Thus, during the 12-hour maturation of pig meat with a lactate level in the blood above 11 mmol.L^{-1}, the samples lost an average of 6.12 g, which was 1.15%, and samples of pig meat with a lactate level in the blood above 16 mmol.L^{-1} – 6.44 g, which was 1.23%. Instead, a similar weight loss of the sample taken from pigs with low blood lactate levels (up to 8.3 mmol.L^{-1}) was 0.97%, respectively. In addition, a more uniform natural weight loss of samples taken from pigs with low cortisol and lactate levels is noteworthy.

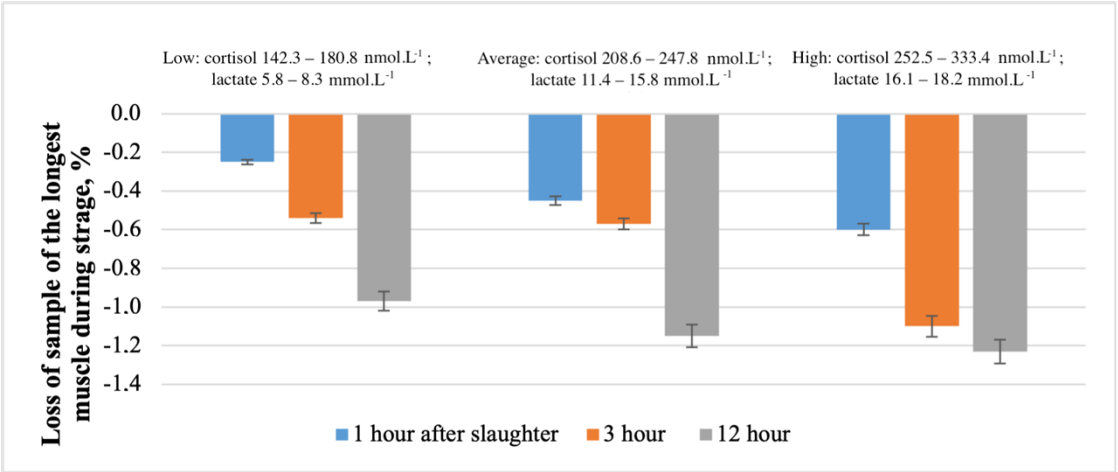

Figure 2 Loss of sample of the longest pig back muscle during 12 hours of storage, depending on the level of cortisol and lactate in the blood, %. Note: when dividing pigs into groups according to the level of cortisol and lactate into low, medium and high, our results were taken into account, and not the physiological norm of these substances.

As shown from the data shown in Figure 3, pigs' access to water during the quarantine period of slaughter allowed to reduce (p <0.05) the natural weight loss of samples compared to samples obtained from animals

without access to water and food. The difference was 0.36% after one hour of storage, 0.59 on the third and 0.23 on the twelfth. Compared to pigs that received water and food, the difference was unlikely.

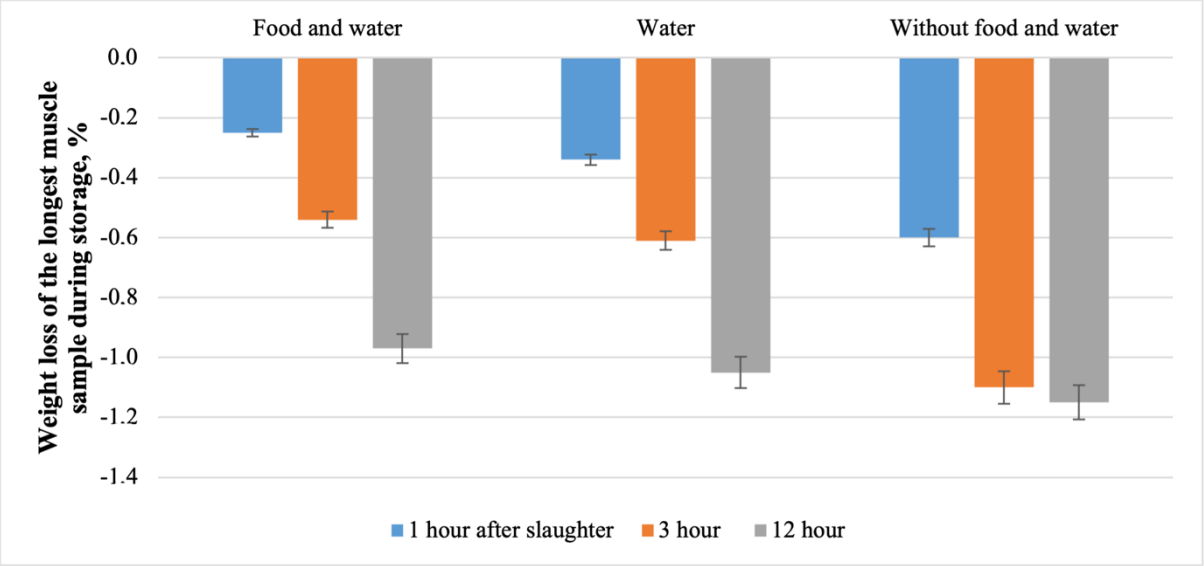

Figure 3 Weight loss (%) of the longest back muscle sample during 12 hours of storage, depending on the conditions before slaughtering keeping pigs.

After 12 hours of maturation, meat obtained from pigs with high lactate levels and more acidic pH was paler, softer, and exudative than meat obtained from lower lactate levels and less acidic pH (Table 5). Thus, most meat samples (75 – 83%) classified as low quality (PSE) were obtained from pigs slaughtered after 10 – 14 hours in a slaughterhouse without access to food. Meat obtained from pigs that experienced lower stress levels was pink, soft and non-exudative (NOR). As a result, the percentage of high-quality meat samples (NOR) in the groups of pigs that had access to water and food ranged from 67 to 75%. The quality to poor quality meat ratio in the animals with access to water ranged from 25:75 to 33:67%. It is noteworthy that in the groups in which the pigs were slaughtered before one o'clock on arrival, the ratio of quality to low-quality meat ranged from 58:42 to 50:50%.

Table 5 Quality of pig meat, depending on transport conditions and before slaughter; several samples.

Meat quality category	Conditions	Transportation: Long without breaks		Transportation: Long with breaks 0.5 hour. every 2 hours		Transportation: Short	
	Before slaughter	Slaughter on arrival	Slaughter in 10 –14 hours	Slaughter on arrival	Slaughter in 10 – 14 hours	Slaughter on arrival	Slaughter in 10 – 14 hours
DFD	Without water and food	1	0	0		1	0
NOR		6	3	6		7	2
PSE		5	9	6		4	10
DFD	Only water		0		0		1
NOR			4		3		4
PSE			8		9		7
DFD	Food and water		0		2		1
NOR			9		8		9
PSE			3		2		2

It should be noted that individual meat samples obtained from pigs of different groups were classified as DFD (dark, hard, dry).

Given that cortisol is a glucocorticoid hormone, which is the trigger for the development of a chain of biochemical stress reactions, it can be said that the most significant stress was experienced by pigs, which were kept for 10 – 14 hours without food and water after delivery to the meat processing plant. In our opinion, this stress factor has several components. First of all, it is a change of habitual location, transport in which the animal can not feel at ease because in a very limited space it does not find a safe place, the use of electric sticks to keep pigs indoors. In addition, the new model of the day adds its influence because the animal does not receive food at the usual time in the usual place. Mixing animals from different farms also increases stress. There is evidence in

the literature that this is also a significant stressor **[13]**. In particular, hungry pigs in a state of stress become aggressive and conflict with each other. Also important is the number of animals in the same group **[15]**.

In pigs slaughtered immediately upon arrival, cortisol levels were significantly lower, indicating that the magnitude of the stress factor that occurs during ante-mortem maintenance without food is higher than the level of "transport" stress. The lowest cortisol level was found in the blood plasma of pigs that had access to water and food during their stay at the slaughterhouse. This suggests that food is a calming factor for pigs. It can be assumed that releasing endorphins from meeting physiological needs can slow down the cascade of stressful biochemical reactions. It should be noted that cortisol was high in pigs with access to water while kept at the slaughterhouse—watering the animals before slaughter did not significantly reduce stress. Data in the literature indicate the optimal pre-slaughter time of pigs in reducing stress and improving the quality of meat. In particular, according to researchers **[16]** – it is three hours.

Significant fluctuations in the concentration of cortisol in the blood plasma of pigs in one experimental group indicate the individual characteristics of the response to the stress factor. In particular, there are data in the literature on the dependence of the amount of stress, and therefore the quality of meat, on the nature of the animal, its age and sex **[10]**, **[17]**.

Under stress, the activity of metabolic processes in the body increases significantly. Carbohydrates are used to meet the needs of energy metabolism. Muscle carbohydrates (primarily glucose and glycogen) are metabolized to lactate by anaerobic oxidation. Accordingly, the greater the amount of stress, the higher the lactate level in the body and the lower the level of muscle carbohydrates. Our studies have shown a moderate to strong positive correlation between cortisol and lactate levels in pig blood. Thus, the lactate concentration was the highest in the blood of pigs kept for 10 – 14 hours in the slaughterhouse without access to water and food.

In contrast, the lowest lactate levels were in pigs' blood with lower cortisol levels. These were pigs that had access to water and food. The lactate level in the blood of pigs that were slaughtered immediately upon arrival had an intermediate position. These patterns support the assumption of researchers that measuring the lactate content in the blood of slaughter pigs can predict the quality of the meat obtained **[18]**, **[19]**. We are also inclined to think about the high informativeness of this indicator. Other researchers point to the high diagnostic value of lactate dehydrogenase **[20]** and glucose **[21]** as prognostic markers of meat quality.

The pH of meat is one of the main indicators of its quality. In the case of high concentrations of lactate in pigs, the pH of the meat will shift to the acidic side. However, if the pH of the meat decreases too quickly during maturation, the quality of the pork will decrease. Rapid hardening of muscle fibers damages their structure causes excessive moisture loss and discolouration, and thus reduces the quality and shelf life of meat **[17]**, **[22]**. We found that when the lactate concentration in the blood of slaughter pigs is above 11 $mmol.L^{-1}$, there is a sharp decrease in the pH of the longest back muscle during the first hour after slaughter. In addition, the higher the blood lactate content, the more acidic the starting pH of the meat. The lower the content of cortisol and lactate in the blood, the more evenly the pH of the meat decreases as it matures. Similar results have been established by other researchers **[23]**.

As a result of a sharp decrease in the pH of the meat of pigs, which, judging by the high concentration of cortisol in the blood, experienced a high level of stress before slaughter, the products were of low quality. The review showed that the meat obtained can mainly be classified as PSE (pale, soft, exudative) with some exceptions when individual meat samples corresponded to the NOR category (close to the optimal quality indicators). In contrast, the predominant number of meat samples (9 – 11 samples) obtained from pigs with blood concentrations of cortisol and lactate in the range of 142.3 – 180.8 $nmol.L^{-1}$ and 5.8 – 8.3 $mmol.L^{-1}$, respectively, corresponded to the NOR quality category, and fewer samples (2 – 3 samples) of PSE category. In this case, single samples of meat in different groups of pigs were classified by us as DFD (dark, hard, dry). In our opinion, the main reason for this is the chronic stress experienced by pigs in the place of rearing. Accordingly, with low amounts of muscle carbohydrates, the pH of meat during maturation will be high, protein denaturation is inactive, water is tightly bound, and there is little or no exudate formation, which leads to dry, hard and dark meat (DFD) **[10]**, **[24]**, **[25]**.

After 12 hours of maturation of the meat, the highest percentage of weight loss was found in those samples in which there was a sharp decrease in pH and assigned to low-quality categories.

Interestingly, the pigs' access to water did not reduce the stress level but prevented significant weight loss of the sample during 12 hours and obtained products of slightly higher quality than on a completely starving diet—watering pigs during quarantine at the slaughterhouse. Similar results have been established by other researchers **[26]**.

The highest levels of stress and, consequently, the lowest quality of meat are characteristic of pigs kept in quarantine before slaughter without access to water and food. Feeding and watering pigs before slaughter positively affects the quality of the meat, but the positives of this scheme have their negatives. In particular, the animal's intestines filled with food can adversely affect the safety of the meat, as careless handling can

contaminate the contents of the intestines. The more filled the gastrointestinal tract during slaughter, the higher the risk of rupturing these tissues during ingestion and contamination of the carcass. It is known that the rate of salmonella secretion in animals increases both with the time of food withdrawal and with stress **[27]**, **[28]**, **[29]**. Equally important is that after ingestion, the food will be absorbed in the small intestine in four to eight hours, and most of the nutrients will be absorbed into the bloodstream in nine hours **[29]**. In addition to the above, it should be borne in mind that the filled intestine creates problems for the technological processing of the intestine itself. For example, for its picking and making a natural sausage casing. In addition to contaminating the product's contents, such intestines will be well filled with blood, not allowing us to obtain high-quality products. Lower levels of stress are experienced by pigs slaughtered immediately upon arrival. Still, such a scheme makes it impossible to observe animals before slaughter for clinical signs of infectious or non-infectious diseases.

Our research results indicate no dependence of stress levels on the breed of pigs. However, in our research, we selected material only from pigs of two breeds: Ukrainian steppe white and landrace. Perhaps other breeds of pigs have more pronounced stress resistance. In particular, some researchers point to this, associating it with the presence of a specific genotype that exhibits a high sensitivity to halothane **[7]**, **[30]**, **[31]**.

The duration of transporting pigs to the slaughterhouse probably did not affect the stress level. It also did not significantly reduce the stress level of alternating rest and transportation during long trips. During transport, the main stress factor for pigs is not the feeling of "road", but being in an unusual and limited space where the animal does not find a safe place. However, convincing evidence can be found in the literature that the amount of stress during pig transport depends on the season **[11]**, **[32]**.

In conclusion, feeding pigs before slaughter reduces the negative impact of stress on meat quality. To date, there are no clear time limits for a starvation diet for pigs before slaughter. But pork must be safe first and foremost, so the logistics of determining the duration of a starvation diet for pigs should consider many factors: the remoteness of the farm from the meat processing plant, feed digestion time, technical capacity of the enterprise; season and many others.

CONCLUSION

The highest quality pork was obtained from pigs that experienced lower stress levels before slaughter. Keeping pigs before slaughter without access to water and food caused higher stress levels, probably expressed in higher blood concentrations of cortisol and lactate. The higher the lactate concentration in pigs' blood, the faster the pH level of meat after slaughter, which adversely affects its quality and moisture retention. Access to pigs to food and water during pre-slaughter keeping allows them to obtain high-quality meat by reducing the impact of stress factors. Pigs' access to water during pre-slaughter ageing does not affect the stress level but positively affects the weight loss of meat during maturation. For the needs of long-term transportation of pigs, rest stops do not significantly reduce the level of stress in pigs. The duration of transportation of pigs does not correlate with the level of stress, as the conditions of pre-slaughter keeping have a decisive influence.

REFERENCES

1. Olsson, V., Pickova, J. (2005). The influence of production systems on meat quality, with emphasis on pork. Ambio (Vol. 34, Issue 4-5, pp. 338–343). https://doi.org/10.1639/0044-7447(2005)034[0338:tiopso]2.0.co;2
2. Choe, J., Moyo, K. M., Park, K., Jeong, J., Kim, H., Ryu, Y., Kim, J., Kim, J. M., Lee, S., Go, G. W. (2017). Meat Quality Traits of Pigs Finished on Food Waste. Korean Journal for Food Science of Animal Resources (Vol. 37, Issue 5, pp. 690–697). https://doi.org/10.5851/kosfa.2017.37.5.690.
3. Ba, H. V., Seo, H. W., Seong, P. N., Cho, S. H., Kang, S. M., Kim, Y. S., Moon. S. S., Choi, Y. M., Kim, J. H. (2019). Live weights at slaughter significantly affect the meat quality and flavor components of pork meat. Journal of Animal Science (Vol. 90, Issue 5, pp. 667–679). https://doi.org/10.1111/asj.13187
4. Kukhtyn, M., Salata, V., Pelenyo, R., Selskyi, V., Horiuk, Y., Boltyk, N., Ulko, L., Dobrovolsky, V. (2020a). Investigation of zeranol in beef of Ukrainian production and its reduction with various technological processing. Potravinarstvo Slovak Journal of Food Sciences (Vol. 14, pp. 95–100). https://doi.org/10.5219/1224
5. Kukhtyn, M., Salata, V., Berhilevych, O., Malimon, Z., Tsvihun, A., Gutyj, B., Horiuk, Y. (2020b). Evaluation of storage methods of beef by microbiological and chemical indicators. Potravinarstvo Slovak Journal of Food Sciences (Vol. 14, pp. 602–611). https://doi.org/10.5219/1381
6. Gallego, M., Arnal, M., Talens, P., Toldrá, F., Mora, L. (2020). Effect of Gelatin Coating Enriched with Antioxidant Tomato By-Products on the Quality of Pork Meat. Polymers (Basel) (Vol. 12, Issue 5, pp. 1–18). https://doi.org/10.3390/polym12051032.

7. Hambrecht, E., Eissen, J. J., Newman, D. J., Smits, C. H. M., den Hartog, L. A., Verstegen, M. W. A. (2005). Negative effects of stress immediately before slaughter on pork quality are aggravated by suboptimal transport and lairage conditions. Journal of Animal Science (Vol. 83, Issue 2, pp. 440–448). https://doi.org/10.2527/2005.832440x
8. Tang, R., Yu, B., Zhang, K., Chen, D. (2008). Effects of supplementing two levels of magnesium aspartate and transportation stress on pork quality and gene expression of micro-calpain and calpastatin of finishing pigs. Archives of Animal Nutrition (Vol. 62, Issue 5, pp. 415–425). https://doi.org/10.1080/17450390802214183.
9. Dokmanovic, M., Ivanovic, J., Janjic, J., Boskovic, M., Laudanovic, M., Pantic, S., Baltic, M. Z. (2017). Effect of lairage time, behaviour and gender on stress and meat quality parameters in pigs. Animal Science Journal (Vol. 88, Issue 3, pp. 500–506). https://doi.org/10.1111/asj.12649
10. Stajković, S., Teodorović, V., Baltic, M., Karabasil, N. (2017). Pre-slaughter stress and pork quality. Earth and Environmental Science (Vol. 85, pp. 1–5). https://doi.org/10.1088/1755-1315/85/1/012034
11. Dalla Costa, F. A., Dalla Costa, O. A., Coldebella, A., Mello Monteirode Lima, G. J., Ferraudo, A. S. (2019a). How do season, on-farm fasting interval and lairage period affect swine welfare, carcass and meat quality traits? International Journal of Biometeorology (Vol. 63, Issue 11, pp. 1497–1505). https://doi.org/10.1007/s00484-018-1527-1
12. Simonetti, A., Perna, A., Giudice, R., Cappuccio, A., Gambacorta, E. (2018). The effect of high pre-slaughter environmental temperature on meat quality traits of Italian autochthonous pig Suino Nero Lucano. Animal Science Journal (Vol. 89, Issue 7, pp. 1020–1026). https://doi.org/10.1111/asj.13007
13. Troeger, K. (2003). Slaughter. In: Encyclopedia of Food Sciences and Nutrition, 2nd ed. USA: Elsevier Science Ltd. https://doi.org/10.1016/B0-12-227055-X/00751-3
14. Vlizlo, V. V., Fedoruk, R. S., Ratych, I. B. (2012). Laboratory methods of research in biology, livestock and veterinary medicine. A reference book, ed. by V.V. Vlizlo. Lviv, Spolom.
15. Dalla Costa, F. A., Dalla Costa, O. A., Di Castro, I. C., Gregory, N. G., Di Campos, Me. S., de Medeiros Leal, G. B., de Castro Tavernari, F. (2019b). Ease of Handling and Physiological Parameters of Stress, Carcasses, and Pork Quality of Pigs Handled in Different Group Sizes. Animals (Basel) (Vol. 14, Issue 10, pp. 1–9). https://doi.org/10.3390/ani9100798
16. Zhen, S., Liu, Y., Li, X., Ge, K., Chen, H., Li, C., Ren. F. (2013). Effects of lairage time on welfare indicators, energy metabolism and meat quality of pigs in Beijing. Meat Science (Vol. 93, Issue 2, pp. 287–291). https://doi.org/10.1016/j.meatsci.2012.09.008
17. Jama, N., Maphosaa, V., Hoffmanb, L. C., Muchenje, V. (2016). Effect of sex and time to slaughter (transportation and lairage duration) on the levels of cortisol, creatine kinase and subsequent relationship with pork quality. Meat Science (Vol. 116, pp. 43–49). https://doi.org/10.1016/j.meatsci.2016.02.001
18. Edwards, L. N., Grandin, T., Engle, T. E., Ritter, M. J., Sosnicki, A. A., Carlson, B. A., Anderson, D. B. (2010). The effects of pre-slaughter pig management from the farm to the processing plant on pork quality. Meat Science (Vol. 86, Issue 4, pp. 938–944). https://doi.org/10.1016/j.meatsci.2010.07.020
19. Rocha, L. M., Dionne, A., Saucier, L., Nannoni, E., Faucitano, L. (2015). Hand-held lactate analyzer as a tool for the real-time measurement of physical fatigue before slaughter and pork quality prediction. Animal (Vol. 9, Issue 4, pp. 707–714). https://doi.org/10.1017/S1751731114002766
20. Čobanović, N., Stanković, S. Dj., Dimitrijević, M., Suvajdžić, B., Grković, N., Vasilev, D., Karabasil, N. (2020). Identifying Physiological Stress Biomarkers for Prediction of Pork Quality Variation. Animals (Basel) (Vol. 10, Issue 4, pp. 1–18). https://doi.org/10.3390/ani10040614.
21. Choe, J. H., Kim, B. C. (2014). Association of blood glucose, blood lactate, serum cortisol levels, muscle metabolites, muscle fiber type composition, and pork quality traits. Meat Science (Vol. 97, Issue 2, pp. 137–142). https://doi.org/10.1016/j.meatsci.2014.01.024
22. Simonetti, M., Cannas, D. M., Just-Baringo, X., Vitorica-Yrezabal, I. J., & Larrosa, I. (2018). Cyclometallated ruthenium catalyst enables late-stage directed arylation of pharmaceuticals. Nature chemistry (Vol. 10, Issue 7, pp. 724–731). https://doi.org/10.1038/s41557-018-0062-3
23. Dokmanovic, M., Baltic, M. Z., Duric, J., Ivanovic, J., Popovic, L., Todorovic, M., Markovic, R., Pantic, S. (2015). Correlations among Stress Parameters, Meat and Carcass Quality Parameters in Pigs. Asian-Australasian Journal of Animal Sciences (Vol. 28, Issue 3, pp. 435–441). https://doi.org/10.5713/ajas.14.0322.
24. Adzitey, F. Huda, H. (2011). Pale soft exudative (PSE) and dark firm dry (DFD) meats: Causes and measures to reduce these incidences – A mini review. International Food Research Journal (Vol. 18, Issue 1, pp. 11-20).

25. Zou, B., Zhao, D., He, G., Nian, Y., Da, D., Yan, J., Li, C. (2020). Acetylation and Phosphorylation of Proteins Affect Energy Metabolism and Pork Quality. Journal of Agricultural and Food Chemistry (Vol. 68, Issue 27, pp. 7259–7268). https://doi.org/10.1021/acs.jafc.0c01822

26. Petrolli, T. G., Junqueira, O. M., Pereira, A. S. C., Domingues, C. H. F., Artoni, S. M. B., Santos, E. T. (2017). Pork carcass injury in slaughterhouse waiting area and nutrient addition in pre-slaughter fasting. Revista MVZ Córdoba (Vol. 22, Issue 1, pp. 5619–5630).

27. Isaacson, R. E.; Firkins, L. D.; Weigel, R. M.; Zuckermann, F. A.; Di Pietro, J. A. (1999). Effect of transportation and feed withdrawal on shedding of Salmonella typhimurium among experimentally infected pigs. American Journal of Veterinary Research (Vol. 60, pp. 1155–1158).

28. Reid, C.A.; Avery, S.M.; Warriss, P.; Buncic, S. (2002). The effect of feed withdrawal on Escherichia coli shedding in beef cattle. Food Control (Vol.13, pp. 393–398).

29. Driessen, B., Freson, L., Buyse, J. (2020). Fasting Finisher Pigs before Slaughter Influences Pork Safety, Pork Quality and Animal Welfare. Animals (Vol. 10, Issue 12, pp. 1–10). https://doi.org/10.3390/ani10122206

30. Weschenfelder, A. V., Torrey, S., Devillers, N., Crowe, T., Bassols, A., Saco, Y., Piñeiro, M., Saucier, L., Faucitano, L. (2012). Effects of trailer design on animal welfare parameters and carcass and meat quality of three Pietrain crosses being transported over a long distance. Journal of Animal Science (Vol. 90, Issue 9, pp. 3220–3231). https://doi.org/10.2527/jas.2012-4676

31. Salas, R. C. D., Mingala, C. N. (2017). Genetic Factors Affecting Pork Quality: Halothane and Rendement Napole Genes. Animal Biotechnology (Vol. 28, Issue 2, pp. 148–155). https://doi.org/10.1080/10495398.2016.1243550

32. Sommavilla, R., Faucitano, L., Gonyou, H., Seddon, Y., Bergeron, R., Widowski, T., Crowe, T., Connor, L., Scheeren, M. B., Goumon, S., Brown. J. (2017). Season, Transport Duration and Trailer Compartment Effects on Blood Stress Indicators in Pigs: Relationship to Environmental, Behavioral and Other Physiological Factors, and Pork Quality Traits. Animals (Basel) (Vol. 7, Issue 2, pp. 1–8). https://doi.org/10.3390/ani7020008

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