

## RELATIONSHIPS AMONG PRE-SLAUGHTER STRESS, RIGOR MORTIS, BLOOD LACTATE, AND MEAT AND CARCASS QUALITY IN PIGS

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The aim of this study was to examine the relationships among pre-slaughter stress, *rigor mortis*, blood lactate, and meat and carcass quality in 100 pigs (cross between Naima sows and hybrids P-76 PenArLan boars). Before slaughter lairage time, handling and pig behavior were assessed for each animal. At exsanguination blood concentrations of lactate and cortisol were determined, while *post-mortem* were assessed: initial and ultimate pH value, temperature, drip, sensory and instrumental colour and marbling. On the carcasses the degree of *rigor mortis* and skin damage score were estimated, as well as carcass quality parameters. More developed ( $p < 0.01$ ) *rigor mortis* was observed after long lairage compared to short lairage. Higher intensity of rigor was found in pigs with higher blood lactate level ( $p < 0.05$ ) and with a greater thickness of subcutaneous fat tissue ( $p < 0.05$ ) and lower lean meat content ( $p < 0.01$ ). Higher blood lactate level was observed after long lairage compared to short ( $p < 0.05$ ) and after rough handling compared to gentle handling ( $p < 0.01$ ). In the group with blood lactate from 10 to 15 mmol/l meat temperature and skin blemishes score increased, while in the group with the highest blood lactate concentration ( $> 15$  mmol/l) initial pH decreased and  $L^*$  value increased. These results suggest that in groups with higher blood lactate concentrations meat quality deteriorates.

**Key words:** pigs, pre-slaughter stress, *rigor mortis*, blood lactate, carcass and meat quality

## INTRODUCTION

After slaughter blood circulation stops and levels of ATP in the cells decrease which is finally followed by *post-mortem* contraction [1]. At this point muscles become shortened and stiff, joints immobile, and this phenomenon is known as *rigor mortis* [2].

*Rigor mortis* occurs when reserves of adenosine triphosphate [ATP] become depleted and can not be anymore resynthesized from creatine phosphate and glycogen, therefore factors that affect their content at slaughter indirectly impact the time of onset and

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intensity of *rigor mortis*. These factors are often related to preslaughter stress [3]. Warriss *et al.* [4] showed that a rapid development of *rigor mortis* was associated with increased blood concentrations of cortisol, lactate and activity of creatine kinase in pigs. These authors found more developed *rigor mortis* in pigs with higher meat temperature and lower ultimate pH value. This suggests that the degree of *rigor mortis* can be used to measure the level of stress at slaughter and meat quality [5].

In addition, an increase in blood lactate concentration is associated with preslaughter stress which has been shown to have detrimental effects on pork quality [6-8]. Hambrecht *et al.* [7] determined that pigs exposed to aggressive handling just prior to stunning had a higher blood lactate concentration at slaughter and exhibited pork with a higher drip loss and thus proposed that lactate was a potential indicator of both physical and psychological stress associated with handling of pigs immediately before slaughter. Therefore, the aim of this study was to examine how *rigor mortis* and blood lactate were associated with preslaughter stress and meat and carcass quality in pigs and whether they could be used for their prediction.

## MATERIAL AND METHODS

### ***Animals, housing and feeding***

The experiment was conducted on 100 commercial pigs (31 gilts, 51 barrows and 18 boars), six months old and with live weight between 115 and 130 kg. All animals were of the same origin (cross between Naima sows and hybrids P-76 PenArLan boars) and originated from the same farm. Pigs were housed in a finishing facility on partially slatted floors, in pens with 20 animals per pen (stocking density = 1 m<sup>2</sup>/pig). Pigs were provided with *ad libitum* feed and water.

### ***Pre-slaughter handling***

Before transport feed and water were not withdrawn. Pigs from the same pen were transported together, without mixing with other pigs, in batches of 20 animals in transportation trailer (stocking density = 0.45 m<sup>2</sup>/pig). The distance between the pens and the vehicle was about 20 m and the slope of the loading ramp was 15°. Transport from the farm to the abattoir lasted 15 minutes. The surfaces of the vehicle and unloading area were on the same level. After unloading, the pigs entered a corridor long 10 m that led to the lairage pens. During loading and unloading sticks and electric prods were used to incite the animals.

The pigs were held in a lairage less than 3 hours - short lairage (on average 1.36 hours) or more than 14 hours - long lairage (on average 17.01 hours). During lairage pigs were not mixed and stocking density was 0.70 m<sup>2</sup> per pig. In the lairage water was provided. For each pig were assessed the exact time of lairage, handling procedure from lairage pen to the stunning area and behavior. Handling procedure was measured through the use of sticks and electric prods, slipping, falling, turning back and vocalization of pigs. If any of these handling or behavior parameters was present, handling was characterized as rough. Opposite to that, absence of these parameters indicated gentle

handling. In the group with short lairage animal behaviour was observed. According to their behavior animals were divided into “aggressive“ and “non - aggressive“ group. A pig was characterized as “aggressive“ if it exhibited fighting and agonistic acts (bites and head knocks) during lairage. Otherwise, it was considered as “non - aggressive“.

After lairage pigs were head-only electrically stunned in batches of 6 animals without restraining. Between lairage pens and the stunning area there was a 5 m single file corridor. During pig handling sticks and electric prods were used randomly. Following bleeding, the carcasses were processed using conventional practice.

### **Blood sampling and determination of blood lactate and cortisol content**

At slaughter blood samples were taken into plastic tubes and part of the material was transferred to heparinized vacutainer tubes. Thereupon blood lactate content was determined using a portable lactate analyzer (Lactate Scout, EKF Diagnostic, Magdeburg, Germany). The lactate analyzer was tested with a standard solution to ensure accuracy. After blood collection the vacutainer tubes were placed on ice and within 4 - 6 hours blood was centrifuged at 3000 rpm for 3 minutes. Supernatants (plasma) were collected into microtubes and stored at  $-20^{\circ}\text{C}$  until determination of cortisol concentration by radioimmunoassay (RIA-CT Cortisol, INEP, Belgrade, Serbia).

### **Meat and carcass quality analyses**

Meat quality measurements were carried out 60 minutes, 24 and 72 hours after slaughter on muscle *Longissimus dorsi* (LD), *pars lumbalis*. pH values were measured 60 minutes and 24 hours *post-mortem* ( $\text{pH}_{60\text{min}}$ ,  $\text{pH}_{24\text{h}}$ ), using a pH-meter “Testo 205” (Germany) while temperature was measured 60 minutes *post-mortem* ( $t_{60\text{min}}$ ). Before and during pH measuring the pH-meter was calibrated with pH 4.00 and 7.00 phosphate buffer (SRPS ISO 2917, 2004). Skin blemishes (SB) were assessed by three observers on three regions (from head to back of shoulder, from back of shoulder to hind-quarters and the region of hind-quarters) immediately after dressing using scores 1 (no damages), 2 (scratches or small wounds, less than 2 cm), 3 (bleeding wounds between 2 and 5 cm or a healed wounds of more than 5 cm) and 4 (deep and open wounds of more than 5 cm). The final score for each carcass was obtained by summing the scores for three regions and can range from 3 to 12. The intensity of *rigor mortis* (RM) was estimated on the right carcass side 3 hours *post-mortem* by measuring the degree of angle between body axis and foreleg [9]. For that purpose photographic images of carcasses were taken, at a distance of approximately 2 m and a height of 160 cm, parallel to the plane in which were the carcasses. The angle was calculated in AutoCAD program (Image 1). The size of the angle and intensity of rigor were inversely proportional, e.g. smaller angle meant a higher degree of *rigor mortis*. In addition, the rate of development of *rigor mortis* was determined measuring the angle after 30, 90, 140 and 180 minutes of slaughter for 12 pig carcasses.

For the determination of drip loss, colour and marbling 2.5 cm thick loin chops were taken 24 hours after slaughter from LD, between the 3th and 4th lumbar vertebrae. Meat samples were weighed and stored for 48 hours at  $4^{\circ}\text{C}$  in a container [10]. After storage meat samples were reweighed and drip loss (DL) was calculated as the difference



**Image 1.** Determination of the angle between body axis and foreleg

between sample weight before and after storage divided by the sample weight before storage. Drip loss was analyzed in duplicate. Sensory (SC) and instrumental color (CIE  $L^*$  - lightness;  $a^*$  - redness;  $b^*$  - yellowness) as well as marbling (Mar) were determined at 24 hours *post-mortem*, after approximately 60 minutes of blooming time [10].  $L^*$ ,  $a^*$  and  $b^*$  [11] values were determined using a Minolta Chroma Meter CR-400 (Minolta Co., Ltd., Osaka, Japan) using D-65 lighting, a 2° standard observer angle. Instrumental color was measured three times on each chop and average  $L^*$ ,  $a^*$  and  $b^*$  values were taken. An analytical panel of three members assessed sensory color and marbling of meat samples by using the scaling method [12]. Meat quality class (PSE - pale, soft, exudative; normal or RFN - red, firm, non-exudative; DFD - dark, firm, dry; RSE - reddish-pink, soft, exudative and PFN - pale, firm, non-exudative) was determined according to Kauffman *et al.* [13] using  $\text{pH}_{24\text{h}}$ , drip loss and  $L^*$  value (Table 1).

**Table 1.** Assessment of meat quality class according to Kauffman *et al.* [13]

Meat quality	$\text{pH}_{24\text{h}}$	DL (%)	$L^*$ value
PSE	< 6.0	$\geq 5$	$\geq 50$
RSE	< 6.0	$\geq 5$	42 – 50
RFN	< 6.0	< 5	42 – 50
PFN	< 6.0	< 5	$\geq 50$
DFD	$\geq 6.0$	< 5	< 42

$\text{pH}_{24\text{h}}$  - pH value measured 24 hours *post-mortem*; DL- Drip loss;  $L^*$  - Lightness; PSE - pale, soft, exudative; RSE - reddish-pink, soft, exudative; RFN - red, firm, non-exudative; PFN - pale, firm, non-exudative; DFD - dark, firm, dry meat

Hot carcass weight (HCW) was measured on a balance scale with an accuracy of 0.1 kg, while carcass backfat thickness at two points (between the 13th and 15th dorsal vertebrae - FTB and over *M. gluteus medius* - FTS) with a metal ruler (accuracy to 1.0 mm). Lean meat content (LMC) was determined according to Serbian Regulation [14].

### **Statistical analysis**

Statistical analysis of the results was elaborated using software GraphPad Prism version 5.00 for Windows, GraphPad Software, San Diego California USA, www.graphpad.com. All parameters were presented by descriptive statistical parameters (mean, standard deviation, minimum and maximum value). The rate of *rigor mortis* development was represented by regression analysis. Student t-test was used to examine the effects of lairage time (short vs. long), handling procedure (gentle vs. rough) and pig behavior (non-aggressive vs. aggressive) on the degree of *rigor mortis* and lactate concentration in pigs. The relationship between the degree of *rigor mortis* and stress, meat and carcass quality parameters was determined by Pearson correlation. One-way ANOVA with Tukey post test was performed to test the differences between the average degree of *rigor mortis* after 30, 90, 140 and 180 minutes of slaughter and to test effects of blood lactate concentration on meat quality parameters. Individual pig was used as experimental unit for all analyses.

## **RESULTS**

The characterization of the experimental population is presented in Table 2. In this study blood lactate ranged from 1.30 to 24.60 mmol/l, while blood cortisol showed greater variability (3.00 - 248.00 nmol/l). Values of pH were after 60 minutes from 5.64 to 6.81, while after 24 hours measured from 5.26 to 5.93. *Rigor mortis* was in average 124.80°. In the experimental group were carcasses without skin blemishes (score 3), as well as with maximum score for skin blemishes (score 12). Average hot carcass weight was 93.46 kg, while carcass backfat thickness ranged from 5.00 to 36.00 mm.

The rate of *rigor mortis* development was measured for 12 pig carcasses (Figure 1). The largest angle was determined after 30 minutes (129.7°), while significantly lower ( $p < 0.01$ ) was after 140 (123.9°) and 180 minutes (123.9°) of slaughter. Between other angles were not observed significant differences.

The degree of *rigor mortis* in relation to the lairage time, handling procedure and pig behavior is shown in Figure 2. In pigs with short lairage the angle of *rigor mortis* (126.7°) was significantly larger ( $p < 0.01$ ) than in pigs with long lairage (124.1°). In addition, more developed *rigor mortis* was determined after rough procedure (124.4°) and in aggressive pigs (126.5°) compared to gentle procedure (125.0°) and non-aggressive pigs (127.0°), although the differences were not significant.

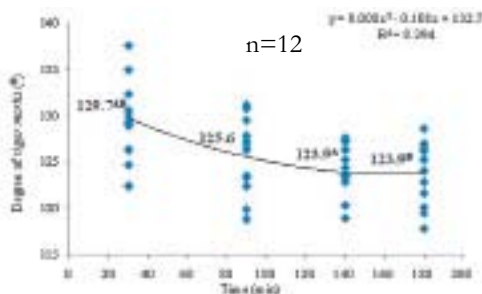
In pigs with long lairage blood lactate concentration (10.86 mmol/l) was significantly higher ( $p < 0.05$ ) compared to short lairage (8.04 mmol/l) (Figure 3). Also, significant difference ( $p < 0.01$ ) was observed between gentle (8.21 mmol/l) and rough procedure (13.61 mmol/l), while pig behaviour did not affect blood lactate concentration.

**Table 2.** Characterization of the experimental population: stress, meat and carcass quality parameters

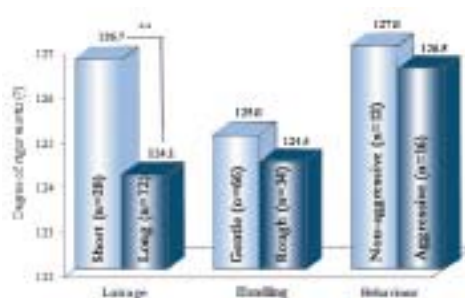
Parameter	$\bar{X} \pm Sd$	Min	Max
Blood lactate (mmol/l)	10.06±5.47	1.30	24.60
Plasma cortisol (nmol/l)	58.36±57.46	3.00	248.00
pH <sub>60 min</sub>	6.33±0.21	5.64	6.81
pH <sub>24h</sub>	5.55±0.13	5.26	5.93
t <sub>60 min</sub> (°C)	38.58±0.74	37.10	40.30
Rigor mortis (°)	124.80±4.62	115.10	136.60
Skin blemishes score	7.09±2.30	3.00	12.00
Drip loss (%)	6.30±1.40	3.01	9.50
L* value	50.20±3.02	35.11	60.22
a* value	7.71±1.26	4.15	11.83
b* value	4.21±0.96	2.23	7.41
Sensory colour score	2.44±0.52	1.00	4.00
Marbling	1.30±0.38	1.00	2.50
HCW (kg)	93.46±11.25	54.30	115.60
FTB (mm)	21.94±5.90	12.00	36.00
FTS (mm)	14.51±5.23	5.00	34.00
Lean meat content (%)	43.79±1.71	38.18	46.61

pH<sub>60min</sub> and pH<sub>24h</sub> - pH values measured 60 minutes and 24 hours *post-mortem*; t<sub>60min</sub> - Meat temperature measured 60 minutes *post-mortem*; L\* - Lightness; a\* - Redness; b\* - Yellowness; HCW - Hot carcass weight; FTB - Carcass backfat thickness between the 13th and 15th dorsal vertebrae; FTS - Carcass backfat thickness over *M. gluteus medius*

The angle between body axis and foreleg, that was used for measuring the degree of *rigor mortis*, negatively correlated with lactate concentration ( $r=-0.20$ ,  $p<0.05$ ) and had a tendency toward a positive correlation with drip loss ( $r=0.19$ ,  $p<0.06$ ) (Table 3). *Rigor mortis* was not significantly correlated with other meat quality parameters. Between the angle of *rigor mortis* and carcass backfat thickness at two points (FTB and FTS) were observed negative correlations ( $r=-0.24$ ,  $p<0.05$  and  $r=-0.25$ ,  $p<0.05$ , respectively), while the angle of *rigor mortis* positively correlated with lean meat content ( $r=0.27$ ,  $p<0.01$ ).



**Figure 1.** The rate of *rigor mortis* development (Legend: Means with a common superscript letter differ; A, B - ( $p<0.01$ ))



**Figure 2.** The degree of *rigor mortis* in relation to the lairage time, handling procedure and pig behavior (Legend: \*\* - ( $p<0.01$ ))

**Table 3.** Correlation between *rigor mortis* (the angle between body axis and foreleg) and stress, meat and carcass quality parameters

Parameters	Lac	Cor	pH <sub>60min</sub>	pH <sub>24h</sub>	t <sub>60min</sub>	SB	DL	L*
<i>Rigor mortis</i>	<b>-0.20*</b>	0.01	0.05	-0.10	-0.07	0.002	<b>0.19†</b>	0.12

Parameters	a*	b*	SC	Mar	HCW	FTB	FTS	LMC
<i>Rigor mortis</i>	-0.006	0.08	-0.11	-0.08	-0.08	<b>-0.24*</b>	<b>-0.25*</b>	<b>0.27**</b>

Lac - Lactate blood concentration; Cor - Cortisol blood concentration; pH<sub>60min</sub> and pH<sub>24h</sub> - pH values measured 60 minutes and 24 hours *post-mortem*; t<sub>60min</sub> - Meat temperature measured 60 minutes *post-mortem*; SB - Skin blemishes score; DL - Drip loss; L\* - Lightness; a\* - Redness; b\* - Yellowness; SC - Sensory colour; Mar - Marbling; HCW - Hot carcass weight; FTB - Carcass backfat thickness between the 13th and 15th dorsal vertebrae; FTS - Carcass backfat thickness over *M. gluteus medius*; LMC - Lean meat content; † - (p<0.06); \* - (p<0.05); \*\* - (p<0.01)

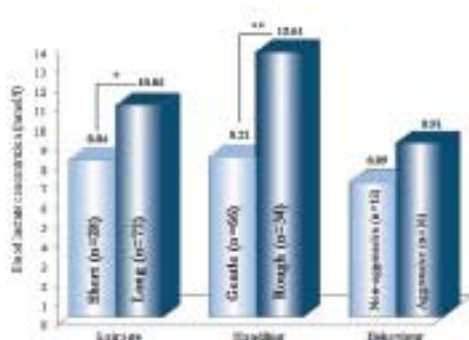
In Table 4 are presented meat quality parameters in relation to blood lactate concentration. In pigs with blood lactate concentration higher than 15 mmol/l a significantly lower (p<0.05, p<0.01) pH<sub>60min</sub> value was determined comparing to groups with lower concentrations of blood lactate. Meat temperature was significantly higher (p<0.01) in pigs with blood lactate over 10 mmol/l than in pigs with blood lactate concentration up to 5 mmol/l. Skin blemishes score was significantly higher (p<0.01) in the group of pigs with blood lactate from 10 to 15 mmol/l compared to the group with blood lactate concentration up to 5 mmol/l. L\* value of LD muscle was significantly higher (p<0.01) in the group with the highest blood lactate concentration compared to the group with blood lactate from 10 to 15 mmol/l. The concentration of lactate did not significantly influence carcass quality parameters (data not shown).

**Table 4.** Meat quality parameters in relation to blood lactate concentration

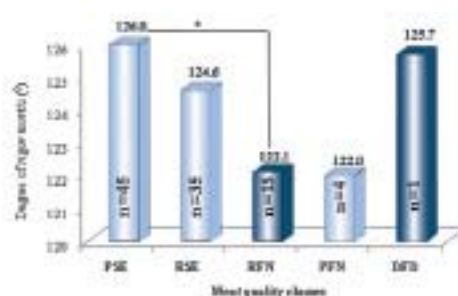
Parameters	Blood concentration of lactate (mmol/l)			
	< 5.00 (N=19)	5.01 - 10.00 (N=36)	10.01 - 15.00 (N=25)	> 15.01 (N=20)
pH <sub>60min</sub>	6.35 <sup>a</sup> ±0.18	6.37 <sup>^</sup> ±0.19	6.42 <sup>B</sup> ±0.16	6.18 <sup>aAB</sup> ±0.17
pH <sub>24h</sub>	5.54±0.09	5.55±0.15	5.54±0.15	5.54±0.13
t <sub>60min</sub> (°C)	38.07 <sup>AB</sup> ±0.60	38.46±0.68	38.88 <sup>^</sup> ±0.67	38.94 <sup>B</sup> ±0.74
SB	5.65 <sup>^</sup> ±2.05	7.04±1.95	8.07 <sup>^</sup> ±2.52	7.12±2.51
RM (°)	126.0±3.95	125.1±5.19	124.8±4.70	123.4±3.95
DL (%)	6.66±1.43	6.46±1.22	5.94±1.59	6.09±1.43
L*	50.40±2.56	50.31±1.94	48.45 <sup>^</sup> ±3.59	51.67 <sup>^</sup> ±3.54
a*	8.11±0.75	7.51±1.37	7.34±0.85	8.10±1.61
b*	4.57±0.67	4.09±0.80	3.70±0.78	4.60±1.32
SC	2.42±0.51	2.45±0.55	2.55±0.45	2.31±0.58

pH<sub>60min</sub> and pH<sub>24h</sub> - pH values measured 60 minutes and 24 hours *post-mortem*; t<sub>60min</sub> - Meat temperature measured 60 minutes *post-mortem*; SB - Skin blemishes score; RM - *Rigor mortis*; DL - Drip loss; L\* - Lightness; a\* - Redness; b\* - Yellowness; SC - Sensory colour score; Within a row, means with a common superscript letter differ: a - (p<0.05); A, B - (p<0.01)

The degree of *rigor mortis* in relation to meat quality class is shown in Figure 4. The highest value of *rigor mortis* was observed in PSE meat ( $126.0^\circ$ ) and the lowest in PFN meat ( $122.0^\circ$ ). Significantly more developed *rigor mortis* ( $p < 0.05$ ) was determined in RFN meat compared to PSE meat.



**Figure 3.** Blood lactate concentration in relation to the lairage time, handling procedure and pig behavior (Legend: \* - ( $p < 0.05$ ); \*\* - ( $p < 0.01$ ))



**Figure 4.** The degree of *rigor mortis* in relation to meat quality class (Legend: PSE - pale, soft, exudative; RSE - reddish-pink, soft, exudative; RFN - red, firm, non-exudative; PFN - pale, firm, non-exudative; DFD - dark, firm, dry meat; \* - ( $p < 0.05$ ))

## DISCUSSION

Intensity of *rigor mortis* is classified on the basis of angle between body axis and foreleg on: 1) presence of rigor ( $\leq 110^\circ$ ), 2) intermediate rigor ( $111-120^\circ$ ) and 3) absence of rigor ( $\geq 121^\circ$ ) [9]. According to this, in our study no carcass was in complete rigor, partial rigor was present in 18% of carcasses, and the absence of rigor was found in 82% of carcasses (Table 2). However, classification according to Davis *et al.* [9] can not be applied, since the time of onset, intensity and duration of *rigor mortis* vary within a wide range. Characteristics of *rigor mortis* depend on initial levels of ATP and CP and their levels during onset, pH value initially, at onset and ultimately, initial and residual stores of glycogen, as well as on activities of ATP-ases and the sarcoplasmic reticulum pump, known as intrinsic factors (species and type of muscle). The most important extrinsic factors that affect *rigor mortis* are the degree of exhaustion at time of death and rate of *post-mortem* carcass chilling [1]. Therefore, different values were determined for the time of onset and duration of *rigor mortis*. According to Savell *et al.* [15], *rigor mortis* in pigs appears from 15 minutes to one hour, and completes in 6 hours. Also, Taylor and Dant [16] have found that *rigor mortis* completes within three to six hours *post-mortem*, while Swatland [17] reported  $197 \pm 113$  minutes. Dransfield and Lockyer [18] have found greater variations in terms of onset and completion of *rigor mortis*. *Rigor mortis* began from three to seven and a half hours and completed from six and a half to 15 hours *post-mortem*. In our study *rigor mortis* was significantly more developed after 140 and 180 minutes *post-mortem* compared to the first measuring (after 30 minutes *post-mortem*) (Figure 1). As no significant difference was determined in rigor between 140 and 180 minutes of slaughter, it indicated that *rigor mortis* was completed after 140 minutes



*post-mortem.*

Since intensity of *rigor mortis* is positively correlated with blood lactate and cortisol as indicators of stress, *rigor mortis* could be used to assess preslaughter stress in pigs [4,5]. According to these authors [4,5], more developed *rigor mortis* after a long lairage compared to short observed in this study indicated that the procedure was more stressful because pigs were for a longer period exposed to stress (rough handling, fights, change of environment, food deprivation, etc.) [19-21] (Figure 2). In addition, although no significant difference was determined, a higher degree of *rigor mortis* was observed after rough handling compared to gentle, as well as in aggressive pigs compared to non-aggressive.

Similarly to Warriss *et al.* [4], in this study a negative correlation between the angle of *rigor mortis* and blood lactate concentration was determined (Table 3). This is one more proof that irrespective of the weak correlation between the intensity of *rigor mortis* and lactate concentration, *rigor mortis* is more intensive in pigs exposed to a greater stress before slaughter. Although Warriss *et al.* [4] found that more developed *rigor mortis* was associated with a higher meat temperature and lower ultimate pH value, in this study no significant relationship was determined between rigor and meat quality parameters, except a tendency toward a positive correlation with drip loss. Drip loss was higher in carcasses with less developed *rigor mortis*, which could be explained by the fact that during rigor "free" water became displaced from muscle fibres [1]. Thus, in carcasses with less developed rigor more water remained in muscles, which was released during storage. *Rigor mortis* was associated with carcass quality parameters, although Warriss *et al.* [4] found no relationship between these parameters. Higher intensity of rigor was found in carcasses with a greater thickness of subcutaneous fat tissue and lower lean meat content. Carcasses with more body fat slowly cool, because fat is a good thermal insulator, and higher meat temperature not only accelerates glycolysis and the development of *rigor mortis*, but also leads to a higher degree of shortening of muscle fibers. Minimum shortening of the muscles (10%) was observed at temperatures of 15 to 20 °C, while at temperatures of 20 to 40 °C shortening increased to 30% [22].

Although there was no significant difference, Warriss *et al.* [4] observed in the carcasses with more developed *rigor mortis* a higher incidence of PSE (20%) compared to DFD meat (9%) and meat of "normal" quality (7%). *Rigor mortis* develops sooner in DFD and PSE meat because levels of ATP and its sources (creatine phosphate and glycogen) rapidly fall. In DFD meat glycogen reserves are depleted prior to slaughter, while in PSE meat immediately after slaughter due to accelerated glycolysis [23]. Shiang Liang *et al.* [24] observed in a group with extreme *rigor mortis* the highest incidence of PSE meat compared to groups with moderate and slight *rigor mortis*. In contrast, we observed significantly less developed *rigor mortis* in carcasses with PSE meat compared to carcasses with RFN meat (Figure 4). PSE meat may vary widely with respect to the intensity of *rigor mortis*. In fact, shorter sarcomere or higher degree of muscle shortening is due to higher meat temperature, which is one of the characteristic of PSE meat. On the other hand, longer sarcomere in PSE meat, as was the case in this study, is the result of denaturation of sarcoplasmic and glycolytic proteins, as well as ATP-ases, so sarcomere shortens less due to a lack of available energy [25].

In this study blood lactate ranged from 1.3 to 24.6 mmol/l (Table 2), which is in accordance with results of other authors, from 1.1 to 20.6 mmol/l [26], from 4.0 to 19.7 mmol/l [27] and from 0.11 to 20.57 mmol/l [28]. Increase in blood lactate concentration is often associated with preslaughter stress, so many factors affect blood lactate, such as equipment and construction of the slaughterhouse, transport [29], genotype [30], lairage time [31] and pig handling [29, 32].

In pigs with long lairage significantly higher blood lactate was observed compared to short lairage (Figure 3), suggesting that long lairage was as a more stressful procedure. Perez *et al.* [33] did not find significant effect of lairage time on blood lactate, while Salajpal *et al.* [30] determined that one group of pigs after short lairage showed higher blood lactate. Further, significantly higher blood lactate was observed after rough handling compared to gentle handling. As in our study, Benjamin *et al.* [34] found a significant difference ( $p < 0.05$ ) in blood lactate between gentle (4.0 mmol/l) and rough pig handling (25.2 mmol/l). Blood lactate in fatigued pigs was higher (32.2 mmol/l) compared to non-fatigued pigs (11.1 mmol/l) [35]. Rough preslaughter treatment increases lactate concentration and adversely affects meat quality in pigs [7,8]. Although in this study a significant difference in lactate concentration between aggressive and non-aggressive pigs was not observed, lactate can increase during aggressive behavior in response to physical exercise and catecholamine release [36].

In pigs with high blood lactate (more than 15 mmol/l) the initial pH value significantly decreased (Table 4). According to Edwards *et al.* [27] elevated levels of lactate were associated with a lower initial pH and a higher drip loss, indicating a lower meat quality. Also, Hemsworth *et al.* [28] have found a negative correlation between blood lactate and initial pH. High preslaughter stress is accompanied by increased blood lactate [7, 8], decreased initial pH value [7,8,37,38] and increased meat temperature [8]. Therefore, stress before slaughter leads to rapid metabolism in muscles both before and after slaughter [39], which in this study resulted in higher meat temperature in pigs with lactate concentration of more than 10 mmol/l. In addition, skin damage score was higher in pigs with higher lactate level (from 10 to 15 mmol/l) than in pigs with blood lactate up to 5 mmol/l, as a stressful treatment increases lactate [32] and degree of injury [38]. The consequence of rapid metabolism in muscles due to preslaughter stress is protein denaturation and lighter color of meat [40,41]. Thus, lighter color was determined in the group with lactate above 15 mmol/l. Since lactate can be used to assess meat quality parameters in pigs [27], in groups with higher blood lactate concentrations meat quality deteriorated.

In the present study *rigor mortis* was completed after 140 minutes of slaughter. More developed *rigor mortis* was observed after long lairage compared to short lairage, while handling and pig behavior did not have any significant influence on the degree of *rigor mortis*. Higher intensity of rigor was found in pigs with higher blood lactate concentration, greater carcass backfat thickness and lower lean meat content, as well as in RFN compared to PSE meat. Higher blood lactate level was observed after long lairage compared to short and after rough handling compared to gentle. In groups with higher blood lactate concentration meat quality deteriorated.

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## **POVEZANOST IZMEĐU STRESA PRE KLANJA, MRTVAČKE UKOČENOSTI, KONCENTRACIJE LAKTATA U KRVI I KVALITETA MESA I TRUPOVA SVINJA**

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Cilj ovog rada je bio da se utvrdi povezanost između stresa pre klanja, mrtvačke ukočenosti, koncentracije laktata u krvi i kvaliteta mesa i trupova 100 svinja dobijenih ukrstanjem ženki Naima rase i P-76 PenArLan mužjaka. Za svaku životinju su pre klanja praćeni dužina boravka u stočnom depou, postupanje radnika i ponašanje svinja. Tokom iskrvarenja merene su koncentracije laktata i kortizola iz krvi, dok su postmortalno

određene početna i krajnja pH vrednost, temperatura, gubitak tečnosti, senzorna i instrumentalna boja, kao i mramoriranost mesa. Na trupovima svinja određeni su stepen *rigor mortis*-a, intenzitet ozleda kao i parametri mesnatosti. Više razvijen ( $p < 0,01$ ) *rigor mortis* utvrđen je kod svinja koje su duže boravile u stočnom depou u odnosu na kraći boravak. Jači intenzitet rigora nađen je kod svinja koje su imale veću koncentraciju laktata na iskrvarenju ( $p < 0,05$ ), deblje subkutano masno tkivo ( $p < 0,05$ ) i manju mesnatost ( $p < 0,01$ ). Veća koncentracija laktata u krvi utvrđena je kod svinja koje su duže boravile u stočnom depou u odnosu na one sa kraćim boravkom ( $p < 0,05$ ) i nakon grubog postupanja radnika sa svinjama u odnosu na blag postupak ( $p < 0,01$ ). U grupi svinja sa koncentracijom laktata u krvi od 10 do 15 mmol/l povećale su se temperatura mesa i stepen ozleda na trupu u odnosu na grupe sa manjom koncentracijom laktata. U grupi koja je imala najveće vrednosti za koncentraciju laktata (više od 15 mmol/l) početna pH vrednost se smanjila, a  $L^*$  vrednost boje povećala u odnosu na ostale grupe. Ovi rezultati ukazuju da se kod svinja sa većom koncentracijom laktata na iskrvarenju kvalitet mesa pogoršao.