RELATIONSHIP BETWEEN GLYCOLYTIC POTENTIAL AND MEAT QUALITY OF DUROC PIGS WITH CONSIDERATION OF CARCASS CHILLING SYSTEM*

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Abstract

The objective of this study was to determine phenotypic relations between glycolytic potential (GP) measured 45 min postmortem and meat quality traits of stress-resistant fatteners, with consideration of carcass chilling system. The investigations involved 35 Duroc fattening pigs whose left halfcarcasses were chilled conventionally (4°C for 24 h) and right half-carcasses were rapidly chilled in a three-phase chilling tunnel (-10°C for 15 min, -15°C for 25 min and -5°C for 40 min with air velocity of 3 m/s). In this study we showed that rapid chilling significantly slows the rate of pH fall from 2 to 96 h after slaughter. The negative relationship between glycolytic potential and pH (especially 24 h postmortem) was stronger for conventionally chilled carcasses but the regression coefficient (b) does not suggest increased rate of pH fall in meat of conventionally chilled carcasses (especially compared to rapid chilling) at the later stages of conversion of muscle to meat (from 24 to 144 h after slaughter). In this investigation GP was positively correlated to drip loss at 48 h postmortem, and a stronger correlation was noted for rapidly chilled carcasses. Moreover, the regression coefficient indicates that rapid chilling to 48 h postmortem can cause a slightly higher drip loss from meat than when the carcasses are chilled conventionally (0.55 vs. 0.46 percentage points per 10 umol/g GP). At the later stages of conversion of muscle to meat (96 and 144 h postmortem) the correlation and regression coefficients were the same regardless of the chilling system.

Key words: Duroc, glycolytic potential, chilling system, meat quality

The conversion of muscle to meat is a complex process involving many biochemical and physical changes. The speed and extent of postmortem metabolism has an effect on muscle properties such as desirable colour, water holding capacity (WHC) or drip loss and palatability, and its subsequent use for food (Pösö and Poulanne, 2005). Glycogen is the predominant carbohydrate in muscles and the main metabolic fuel for anaerobic glycolysis that takes place postmortem when muscles are no

^{*}Work finansed from statutory activity.

longer supplied with oxygen. Measurement of glycogen combined with a measure of lactate, called glycolytic potential (GP), is also a way of predicting quality attributes of meat (Monin and Sellier, 1985). Chilling rate also influences the rate of glycolytic changes in meat tissue after slaughter and has an effect on pork meat quality (Josell et al., 2003; Zybert et al., 2007).

The objective of this experiment was to investigate the relation between glycolytic potential and meat quality traits of stress-resistant Duroc fatteners, including the chilling system.

Material and methods

The investigation covered 35 stress-resistant Duroc pigs. The animals were kept under the same environmental conditions and fed a complete diet. Pigs were slaughtered within 2–4 h of transport (300 km) using an electric stunner (MIDAS, Stork RMS, the Netherlands and INARCO constant voltage system) and bled lying down, in accordance with the technology applied at the meat plant. Thirty-five left halfcarcasses were chilled conventionally (4°C for 24 h) and 35 right half-carcasses were chilled in a three-phase chilling tunnel (-10° C for 15 min, -15° C for 25 min and -5° C for 40 min with air velocity of 3 m/s). The quality of meat was evaluated (from 35 min to 24 h postmortem) directly in carcasses in the *m. longissimus lumborum* (LL) behind the last rib, and at 48, 96 and 144 h postmortem in meat samples taken after 24-h chilling at the last rib and 1st lumbar vertebra. The samples were separated from the bone, external fat and epimysium and stored in plastic bags at 0–4°C. In this investigation the following parameters were measured:

– acidity of the muscle tissue (pH) measured at 35 min, 2 h, 3 h, 24 h, 48 h, 96 h and 144 h postmortem (pH₃₅, pH₂, pH₃, pH₂₄, pH₄₈, pH₉₆, pH₁₄₄, respectively), using a pistol pH-meter MASTER (Draminski, Olsztyn, Poland) calibrated with temperature compensation;

– electrical conductivity (EC) measured with an LF-Star conductometer (Ingenieurbüro Matthäus, Noblitz, Germany) 35 min, 2 h, 3 h, 24 h, 48 h, 96 h and 144 h postmortem (EC₃₅, EC₂, EC₃, EC₂₄, EC₄₈, EC₉₆, EC₁₄₄);

– colour lightness (L*) of the muscle tissue was measured using Minolta Portable Chroma Meter (model CR 310, Minolta, Osaka, Japan) and 50 mm orifice. The apparatus was calibrated using white calibration plate with D65 illuminant and 2° as standard observer. The colour parameters of the individual meat samples of 15 mm thickness (and, as mentioned above, taken after 24-h chilling at the 1st lumbar vertebra without bone, external fat and epimysium and stored in plastic bags at 0–4°C) were determined at 24 h, 48 h, 96 h and 144 h postmortem in CIELAB colour system;

– WHC, determined by the filter paper method according to the method of Grau and Hamm (1952) as modified by Pohja and Ninivaara (1957), 24 h postmortem;

- drip loss, determined according to Prange, Jugert and Scharner (1977), 48 h, 96 h, and 144 h postmortem.

The samples cut from LL muscle 45 min postmortem (immediately immersed into tubes with 1M HClO₄ and homogenized to inhibit glycolytic changes) were analysed for glycolytic potential (GP) and content of glycogen and lactate. Glycogen content was determined by the enzymatic method according to Dalrymple and Hamm (1973) and lactate content according to Bergmeyer (1974). The glycolytic potential was calculated as the sum of 2[glycogen] + [lactate] according to Monin and Sellier (1985) and expressed as umol of lactic acid equivalent per g of fresh muscle. The genomic DNA was isolated from white blood cells according to Kawasaki (1990) while the RYR1 C1843T polymorphic site was analysed with DNA test using the PCR/RFLP method, according to Fujii et al. (1991). The data were analysed by one-way analysis of variance using an orthogonal design in Statistica 6.0 (StatSoft, Tulsa, OK, USA). The significance of differences between means was calculated using Duncan's test. For the entire analysed population and for groups differentiated by the chilling system, phenotypic correlations were determined between glycolytic potential measured 45 min postmortem and meat quality traits.

Results

The average lean meat content in the analysed population of Duroc pigs was 57.64±1.87% with hot carcass weight of 93.55±4.10 kg, while average glycolytic potential (measured 45 min postmortem) was 129.25±22.75 umol/g. Table 1 shows the characteristics of the experimental population of animals, including the carcass chilling system. Meat acidification (especially up to 24 h postmortem), electrical conductivity and meat lightness indicate normal values of meat tissue. There were several significant effects of the chilling system on meat quality parameters. Results indicate that more rapidly chilled carcasses had significantly slower pH fall to 48 h after slaughter than those chilled conventionally. Moreover, rapid chilling not only slowed the acidification of *longissimus* muscle tissue but also significantly prolonged the time in which the tissue achieved the ulimate pH to 96 h postmortem. The results concerning the changes in pH fall depending on the chilling system were reflected in meat lightness and electrical conductivity (Table 1). Meat from conventionally chilled carcasses was paler at 24 h postmortem while 96 h after slaughter it was darker than meat from the rapid chilling system. The lighter colour of rapidly chilled meat is the result of the prolonged pH fall up to 96 h postmortem. There was no effect of the chilling system on WHC or drip loss.

Table 2 gives Pearson correlations between glycolytic potential (GP) and meat quality parameters analysed in this experiment. Results indicate that as glycolytic potential increased, pH of meat decreased (at 24, 48, 96 and 144 h postmortem). There was also a weak positive relationship between glycolytic potential and meat lightness at 24, 96 and 144 h postmortem, which suggests that an increase of glycogen stores in meat makes *longissimus* muscle paler. Increases in GP were also related to an increase in drip loss (P \leq 0.01).

Traits	Chilling system		T. (1	F-emp.
	conventional	fast	n = 70	Significance
	n = 35	n = 35		of differences
pH ₃₅	6.62±0.10	6.60±0.11	6.61±0.10	0.32 NS
pH ₂	6.36 A±0.14	6.51 B±0.10	6.44±0.14	25.72 **
pH ₃	6.17 A±0.12	6.40 B±0.12	6.29±0.16	59.79 **
pH ₂₄	5.63 A±0.05	5.67 B±0.07	5.65±0.06	7.75 **
$pH_{_{48}}$	5.40 A±0.07	5.46 B±0.09	5.43±0.08	11.54 **
pH ₉₆	5.51 B±0.09	5.45 A±0.09	5.48±0.09	7.69 **
pH ₁₄₄	5.55±0.10	5.52±0.10	5.53±0.10	2.32 NS
EC445 (mS/cm)	3.61±0.33	3.59±0.33	3.60±0.33	0.17 NS
EC ₂ (mS/cm)	2.54±0.41	2.59±0.48	2.57±0.44	0.23 NS
EC ₃ (mS/cm)	2.49±0.40	2.58±0.53	2.54±0.47	0.63 NS
EC ₂₄ (mS/cm)	2.58 a 0.59	2.94 b 0.68	2.76 0.66	5.59
EC ₄₈ (mS/cm)	10.0 11.17	10.64 1.51	10.33 1.38	3.75 NS
EC ₉₆ (mS/cm)	11.50 b	10.86 a	11.18	4.83
EC ₁₄₄ (mS/cm)	12.17 B 0.91	11.02 A 0.88	11.60 1.06	28.72 **
L*_24	56.91 B 2.54	54.16 A 2.34	55.53 2.79	22.23 **
L*_48	56.03 1.65	56.77 2.09	56.40 1.90	2.69 NS
L* ₉₆	56.57 a 1 99	57.86 b 2.40	57.22 2.28	5.98
L* ₁₄₄	57.08	57.20 2.99	57.14	0.05 NS
WHC (cm ²)	5.91	6.27 1.20	6.09	1.68 NS
Drin loss 48 h	5.09	4 59	4 84	0.93
(%)	2.17	2.12	2.14	NS
Drip loss 96 h	8.51	7.57	8.04	2.31
(%)	2.66	2.50	2.61	NS
Drip loss 144	10.76	10.13	10.44	1.03
<u>h (%)</u>	2.66	2.51	2.59	NS

Table 1. Characteristics of the analysed population of pigs with regard to the chilling system

Values in the table are given as means \pm standard deviations; means designated with capital letters A, B differ significantly at P \leq 0.01; means designated with small letters a, b differ significantly at P \leq 0.05; NS – not statistically confirmed.

Troita	Statistical	Glycolytic potential (µmol/g) (x)		
(y)	measure	Conventional	Fast chilling	Total
		chilling	<u> </u>	
pH ₃₅	r	0.15	-0.09	0.03
	b _{xy}	0.001	-0.0004	0.0001
pH ₂	r _{xy}	0.10	0.14	0.10
	b _{xy}	0.001	0.001	0.001
pH ₃	r _{xy}	-0.05	0.09	0.02
	b _{xy}	-0.002	0.001	0.0001
pH ₂₄	r _{xy}	-0.73**	-0.22	-0.42**
	b _{xy}	-0.002	-0.001	-0.001
pH ₄₈	r _{xv}	-0.71**	-0.62**	-0.61**
	b _{xv}	-0.003	-0.003	-0.002
pH ₉₆	r	-0.78**	-0.75**	-0.72**
20	b _{xy}	-0.003	-0.003	-0.003
pH ₁₄₄	r	-0.77**	-0.78**	-0.76**
* 144	b	-0.003	-0.003	-0.003
EC ₁ (mS/cm)	r	-0.24	-0.04	-0.14
45 \	b	-0.003	-0.001	-0.002
EC. (mS/cm)	r	-0.29	-0.20	-0.24
	b	-0.001	-0.004	-0.005
EC (mS/cm)	r xy	-0.34*	-0.34*	-0.34**
	b	-0.006	-0.008	-0.007
FC (mS/cm)	r r	-0.04	0.21	0.09
LC_{24} (IIIS/CIII)	h h	-0.04	0.006	0.003
$EC_{mS/am}$	v _{xy}	0.001	0.000	0.12
EC_{48} (IIIS/CIII)	h	0.20	0.04	0.13
$EC_{m}(mS/am)$	U _{xy}	0.015	0.11	0.000
EC_{96} (IIIS/CIII)	l _{xy}	-0.07	-0.11	-0.09
$\mathbf{E}\mathbf{C} = (\mathbf{u}, \mathbf{C}, \mathbf{L}, \mathbf{u})$	U _{xy}	-0.004	-0.000	-0.003
EC_{144} (mS/cm)	r _{xy}	-0.36*	0.20	-0.07
T.d.	D _{xy}	-0.014	0.008	-0.003
L* ₂₄	r _{xy}	0.19	0.39*	0.25*
	b _{xy}	0.021	0.039	0.030
L* ₄₈	r _{xy}	0.11	0.26	0.19
	b _{xy}	0.008	0.024	0.016
L* ₉₆	r _{xy}	0.24	0.31	0.27*
	b _{xy}	0.020	0.032	0.027
L* ₁₄₄	r _{xy}	0.15	0.46**	0.33**
	b _{xy}	0.012	0.059	0.036
WHC (cm ²)	r _{xv}	0.30	0.20	0.24*
	b _{xv}	0.046	0.010	0.012
Drip loss 48 h (%)	r _{xv}	0.49**	0.60**	0.54**
	b _{xv}	0.046	0.055	0.051
Drip loss 96 h (%)	r	0.67**	0.72**	0.68**
	b	0.078	0.078	0.078
Drip loss 144 h (%)	r,	0.66**	0.69**	0.67**
/	b	0.076	0.076	0.076

Table 2. Coefficients of phenotypic simple correlations between glycolytic potential measured 45 minutes after slaughter and analysed meat quality traits of pigs with regard to the chilling system

* – significant at P≤0.05; ** – significant at P≤0.01; NS – not statistically significant.

Another area of interest is the relationship between GP and meat quality traits including the chilling system. The expected negative relationship between glycolytic potential and pH24 was present but a higher and statistically confirmed correlation was noted for conventionally chilled carcasses ($r = -0.73^{**}$ vs -0.22^{NS}). On the other hand the regression coefficient (b) does not suggest increased rate of postmortem metabolism in the meat of conventionally chilled carcasses (especially compared to rapid chilling) during conversion of muscle to meat from 24 to 144 h postmortem. A positive correlation of GP with meat lightness of rapidly chilled carcasses was moderate and statistically confirmed at 24 and 144 h postmortem. Moreover, the regression coefficient shows that the increase of GP at about 10 µmol/g results in growth of meat paleness only for 0.39 and 0.59 units, respectively. We also showed a stronger correlation between GP and drip loss at 48 h postmortem for rapidly chilled carcasses (Table 2). The regression coefficient indicates that up to 48 h postmortem rapid chilling can create a slightly greater drip loss from meat than when the carcasses are chilled conventionally (0.55 vs. 0.46 percentage point per 10 µmol/g GP). At the later stages of conversion of muscle to meat (96 and 144 h postmortem) the correlation and regression coefficients were the same regardless of the chilling system (Table 2).

Discussion

Glycolysis is a fundamental biochemical process in the postmortem conversion of muscle to meat and simultaneously a key factor in pork meat quality (Pösö and Poulanne, 2005). After slaughter, when the circulation is stopped and buffering capacity is greatly reduced, anaerobic glycolysis results in pH fall. Glycolytic potential is an estimate of the amount of glycogen that is present in the muscle at the slaughter (or after slaughter) and its potential for lactic acid production, which causes a decline in postmortem pH (Monin and Sellier, 1985). Muscle glycogen level at death largely determines pH_u and glycolytic potential is often used as the indicator of the ultimate pH in meat (Greaser, 1986). A strong negative correlation between glycolytic potential (or glycogen content) and ultimate pH has been reported by Przybylski et al. (1994), van Laack (2001), Henckel et al. (2002) and Zybert et al. (2008) (-0.64, -0.61, -0.60 and -0.45 respectively). Although glycolytic potential is related to pHu it does not fully explain ultimate pH. van Laack (2001) demonstrated that glycolytic potential accounts for 37% of the difference in pH_u of pork loins, while our study (Koćwin-Podsiadła et al., 2009) showed that glycolytic potential accounts for 62 to 64% of the variation in pH during meat aging and storage.

The glycogen stores in the muscle (or GP) are also positively correlated with meat lightness. The results shown in the present experiment are in agreement with the studies of Meadus and MacInnis (2000), Oksbjerg et al. (2004) and Huff-Lonergan et al. (2002) who suggest that increases in glycolytic potential promote paleness of meat.

Muscle glycogen level at death, rate of pH drop and ultimate pH also have a strong relationship to WHC or drip loss (Purslow et al., 2001; Schäfer et al., 2002).

Water holding capacity is the ability of meat to hold all or part of its water during treatment while drip loss is typically reported as a percentage of the weight of the meat that is lost due to fluid that is released from the tissue during storage using the methods without external forces (Honikel, 2004). WHC is determined by structural and biochemical mechanisms (Offer, 1991). Normal ultimate pH is very close to the isoelectric point of the major proteins in muscle (5.3-5.5; Huff-Lonergan and Lonergan, 2005). This is the point at which electric charges on the amino and carboxyl groups on the proteins cancel each other. These positive and negative groups within the protein are attracted to each other and result in a reduction in the amount of water that can be held by the protein. Additionally, reduction of space within the sarcomere produces water loss from muscle postmortem (Offer, 1991). As shown by Rosenvold et al. (2001), muscle glycogen stores at the time of slaughter and postmortem glycolysis are important factors responsible for variation in drip. Schäfer et al. (2002) shows that glycogen content in muscle explains 60% of the differences in drip loss, while in our study (Koćwin-Podsiadła et al., 2009) glycogen and lactate content was responsible for 38% of the differences in drip loss of (Landrace × Yorkshire) × Duroc pigs. The positive correlations between glycolytic potential and drip loss (from 0.36 to 0.61) were noted by some authors (van Laack and Kauffman, 1999; Huff-Lonergan et al., 2002; Hamilton et al., 2003; Zybert et al., 2008). Moreover, results from regression analysis shown in Table 2 demonstrated that 10 µmol/g wet tissue increase in glycolytic potential results in 0.5-0.7 percentage point increase in drip loss and is similar to the results of Hamilton et al. (2003) obtained for progeny of sires and Camborough 22 dams (PIC, USA, Franklyn, KY).

Chilling is a factor that is known to affect pork meat quality (Savell et al., 2005). The principle of the chilling process is to remove heat from the carcass as quickly as possible after slaughter. Transfer of heat from carcasses to the atmosphere (or another material) reduces evaporative weight loss of carcasses and the rate of anaerobic metabolic process in meat after slaughter by decreasing the rate of pH decline (Savell et al., 2005). In comparison to conventional chilling, a slower rate of pH fall in rapidly chilled carcasses was noted by Josell et al. (2003) and Zybert et al. (2007). By slowing the rate of pH decline, severity of denaturation of myoglobin and other proteins is reduced and the colour and WHC of meat may be improved (Huff-Lonergan and Lonergan, 2005). Accelerated air chilling has been shown by Kerth et al. (2001) and Bertram et al. (2001) to improve WHC in pork.

In conclusion, we showed the influence of the chilling system on the rate of pH changes from 2 to 96 h after slaughter. The negative relationship between glycolytic potential and pH (especially 24 h postmortem) was stronger for conventionally chilled carcasses, but the regression coefficient (b) does not suggest changes in the rate of postmortem metabolism expressed by pH fall at the later stages of conversion of muscle to meat, depending on the chilling system, with the increase of GP in muscles. We did not statistically confirm the impact of chilling system on drip loss, but we obtained a statistically (P \leq 0.01) higher correlation for rapidly chilled carcasses at 48 h postmortem and the regression coefficient shows the possibility of deterioration of rapidly chilled meat by the increase fluid loss from meat and greater susceptibility of rapidly chilled meat to paleness with the increase in glycolytic potential.

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Accepted for printing 18 I 2013

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Związek potencjału glikolitycznego z cechami jakości mięsa tuczników rasy Duroc, z uwzględnieniem systemu chłodzenia tusz

STRESZCZENIE

Celem pracy było określenie związku potencjału glikolitycznego mierzonego w próbkach mięśnia *Longissimus lumborum* (LL) pobieranych w 45 minut po uboju z wybranymi cechami jakości mięsa z uwzględnieniem systemu chłodzenia tusz. Badaniami objęto 35 tuczników rasy Duroc, których półtusze prawe poddawano konwencjonalnemu wychładzaniu w temperaturze 4°C przez 24 godz. zaś półtusze lewe poddawano szybkiemu chłodzenia tusi, trójfazowym tunelu ($-10^{\circ}C - 15 \min, -15^{\circ}C - 25 \min i -5^{\circ}C - 40 \min$) przy prędkości powietrza 3m/s), a następnie do 24 godz. po uboju chłodzono w temperaturze 4°C. W niniejszych badaniach udowodniono, że szybkie chłodzenie istotnie spowalnia tempo spadku pH w mięśniu *Longissimus* w okresie od 2 do 96 godz. po uboju. Stwierdzono również istotne współdziałanie pomiędzy wartością potencjału glikolitycznego a stopniem zakwaszenia tkanki mięśnia *Longissimus*, które silniejsze (szczególnie 24 godz. *post mortem*) było w grupie tusz poddanych szybkiemu wychładzaniu. Uzyskane w niniejszych badaniach wartości współczynników regresji nie wskazują jednak na możliwość istotnego zwiększenia tempa spadku pH w przypadku tusz poddanych konwen-

cjonalnemu chłodzeniu (w porównaniu do tusz chłodzonych szybko) w okresie konwersji mięśnia w mięso od 24 do 144 godz. po uboju. W niniejszych badaniach stwierdzono również dodatnią zależność pomiędzy wartością potencjału glikolitycznego a wyciekiem naturalnym, przy czym silniejszy związek pomiędzy opisywanymi cechami występował w 48. godzinie *post mortem* w grupie tusz poddanych szybkiemu wychładzaniu. Uzyskane wartości współczynników regresji wskazują również, że szybkie wychładzanie tusz świń rasy Duroc może prowadzić do nieznacznego zwiększenia wycieku naturalnego z tkanki mięśnia *Longissimus*, 48. godz. *post mortem*. W późniejszym okresie konwersji mięśnia w mięso, tj. w 96. i 144. godz. po uboju wartości współczynników korelacji i regresji wyliczone pomiędzy wartością potencjału glikolitycznego a wyciekiem naturalnym były zbliżone w obu analizowanych grupach zróżnicowanych systemem chłodzenia.