Influence of selection phase, fasting and transport on porcine stress and on the development of PSE pork

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Abstract. The objectives were to investigate the effect of selection phase, fasting time and transportation on porcine stress and on the occurrence of pale, soft and exudative (PSE) pork.

The first selected pigs had the lowest blood creatine kinase (CK) activity and were least susceptible to PSE development. Whereas, the second selected animals were most stressed during transport and showed the highest frequency of PSE. On the other hand, the last selected pigs had elevated CK values from the farm up to stunning, and also showed a slow pH fall in *M. longissimus dorsi* (LD) 45 min *postmortem* (p.m.). Furthermore, the pigs of the first and last selection had, respectively, the lowest and highest lean meat content.

Muscle traits such as glycogen and lactate content 0 min p.m. and pH 45 min p.m. suggested that the most acceptable pork quality for meat products manufacture could be obtained after a fast of 6—7 h if the pigs were fed twice a day. Whereas, overnight holding had a negative effect on muscle glycogen and lactate content.

During transport, pigs were under greatest stress if they were loaded just before feeding period, or if they were transported in warm weather (over 10°C). Moreover, high mortality and CK values occurred at the same external temperatures during transportation. In addition, increased transport time elevated the CK activity from the farm up to the abattoir.

Index words: selection phase, fasting, transport, creatine kinase, PSE meat

Introduction

Pigs fed a restricted ration twice a day showed much more variation in glucose flux rate, rate of formation and hydrolysis of glycogen than pigs fed *ad libitum* (RIIS and GRUMMER, 1969). Easily mobilized glycogen stores were exhausted in about 20 h. Pigs fed *ad libitum* most likely had an almost constant glycogen reserve, whereas the labile glycogen depots of pigs fed a restricted ration were presumably nearly exhausted in the morning before feeding (RIIS and GRUMMER, 1969). POSPIECH *et al.* (1981) observed that the highest number of pigs with muscle defects of PSE and DFD type occurred in groups of 90

and 120 kg live-weight. Lower meat quality was perhaps caused by the change in metabolism. In the group of 90 kg it was probably connected with the change of proteins to carbohydrates proportion in the fodder. The cause of the poorer meat quality of 120 kg pigs could be the slowing down of muscle tissue synthesis (POSPIECH *et al.,* 1981).

For practical purposes only those factors which affect energy reserves in the muscle at the point of slaughter were important for meat quality (BARTON-GADE, 1985). If PSE and DFD meat were considered equally important, then slaughter of unfed pigs immediately after unloading and slaughter of fed pigs after a holding period of about four hours gave the lowest frequency of both PSE and DFD (NIELSEN, 1981). Withholding food during the 12-hour period prior to transportation was a simple way to reduce death during transport (WILLIAMS, 1985).

Earlier work (HONKAVAARA, 1989) considered the influence of porcine stress on blood composition and early p.m. meat quality in pigs of different halothane genotypes. The purpose of this study was to investigate the effects of selection phase, fasting time and transportation of porcine stress and meat quality. In addition, it was studied the effect of transport temperature and time on mortality of pigs.

Material and methods

Test animals

The test animals consisted of 103 pigs of which one died during transport. They were from the following breeds: 39 Finnish Landrace (L) and 8 Finnish Yorkshire (Y) pigs, and 56 LxY crosses. 11—12 pigs were collected per month from September 1985 to May 1986. 31, 12, 8 and 4 pigs came respectively from 1, 2, 2 and 8 producers. Treatments were repeated 22 times, and the programme of one treatment is described earlier (HONKAVAARA, 1988). 28, 24 and 50 pigs were slaughtered at the abattoir in Nurmo, Forssa and Salo, respectively.

The pigs were selected in three phases. Thus the animals of the first, second and last selection gained the slaughter weight of 90—120 kg first, second and third, respectively. Farmers evaluated the weight of pigs by measuring or weighing. After blood sampling, marking and tattooing, the test animals were not separated from other pigs at farm. Moreover, all pigs were treated in the same way from the farm up to the point of stunning. Furthermore, data were collected from the last feeding time of day, loading time of day, transport temperature and duration.

In order to compare the influence of transport temperature and time on mortality with the CK values of the test pigs after transport, data were collected from a further 317 pigs which have died in trucks in 1986.

Evaluation of porcine stress and meat quality

Methods used for the evaluation of porcine stress and meat quality are presented earlier (HONKAVAARA, 1988).

Variables

The collected data included the following 20 continuous variables: loading time of day (LT), transport temperature (TT), transport duration (TD), duration of lairage (DL), temperature of lairage (TL), humidity of lairage (HL), stunning time of day (ST), fast duration (FD, time elapsed between feeding and stunning), carcass temperature 0 min p.m. (CT_0), blood creatine kinase (CK) activity on farm (CKF), CK activity during unloading (CKU), CK activity at exsanguination (CKE), serum glucose (SG), serum glycerol (SGL), serum lactate (SL), serum pH (SpH), muscle glycogen 0 min p.m. (MG_0), muscle lactate 0 min p.m. (ML_0), pH in the *M. longissimus dorsi* (LD) 45 min p.m. (pH_1) and carcass hot weight (CW). Also carcass grading class (CC) was included as an uncontinuous varia-

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ble in the analysis. The CKF, CKU and CKE values were $\log_{10}$ transformed.

Statistical analyses

Conventional statistical methods were used to calculate means, standard deviations and standard error of the means (SEM). In order to examine significant relations between the variables and differences among the means, data were analysed by simple regression and by analysis of variance, respectively. Furthermore, to evaluate the influence of the variables on porcine stress and meat quality, a stepwise linear regression analysis was performed (statistical program Pato for microcomputers, Mikrovuo Ky, Finland).

The regression model (1) included the dependent variable Y_i (i = 1—9), the 20 independent variables X_j (j = 1—20, j ≠ i) and the standardized regression coefficients B_j (j = 1—20, j ≠ i).

$$Y_i = B_1X_1 + B_2X_2 + B_3X_3 + \ldots + B_{20}X_{20} \qquad (1)$$

Regression analysis computed the combination of the most contributing independent variables to the regression model. The program added to the model that variable which fulfilled the F-ratio criterion (F > 3) and had the maximum coefficient of determination (R^2). The variables which made minor contributions (F < 2) were deleted from the model. Finally the program printed the chosen model which estimated the dependent variable best. This output included the coefficient of determination, F ratio, significance of contribution, chosen independent variables and their standardized regression coefficients, partial regression coefficients, F values, significance levels and contributions.

On the basis of stepwise regression, the prediction equations (2) were developed using the independent variables described above.

$$Y_i = B_0 + B_1X_1 + B_2X_2 + \ldots + B_nX_n \qquad (2)$$
$$(i = 1—9, \ n = 3—5)$$

Results

Prediction of porcine stress and meat quality

Table 1 shows the combined effects of the studied variables on porcine stress and meat quality. The former was evaluated by blood CK activity, serum values and carcass temperature, while the latter was estimated by muscle glycogen, lactate and pH_1 value. Thus the coefficient of determination (R^2 100) of the prediction equations was highest for carcass temperature (62.7 %), serum lactate (61.0 %) and glucose (60.7 %), and log CK at exsanguination (57.1 %). Whereas R^2 100 decreased for muscle lactate (35.3 %) and glycogen (28.5 %) serum glycerol (22.7 %) and pH (19.7 %), and log CK during unloading (16.4 %). The combined effects of the independent variables of the prediction equations are discussed below.

Selection phase

Table 2 shows the blood characteristics and the carcass traits studied in the three selection phases. The occurrence of reactors was 0, 7.6 and 4.2 % in the first, second and last selection, respectively. No significant differences were found between the three phases in log CK at exsanguination that was one of the most contributing components for carcass temperature (partial R^2 100 = 12.9 %, Table 1).

Pigs of the first selection were stressed least. Thus they had the lowest CK values and the lowest content of muscle lactate. In addition, they had a moderate pH fall in the LD muscle 45 min p.m. that made them most acceptable for meat products manufacture. This was based on the fact that carcasses which had a fast pH fall 45 min p.m. developed PSE meat, whereas those with a slow pH fall could develop dark, firm and dry (DFD) meat in higher frequencies than those with a moderate pH fall. Actually, our unpublished results indicated that about 20 % of the carcasses with pH_1 values over 6.4 developed DFD in the LD next day.

Table 1. The best stepwise regression models[a] for predicting the characteristics of porcine stress and meat quality.

Prediction equations[b]	$R^2 \cdot 100$[c] %	Df[d]
Log CK during unloading = $2.782 - 0.159LT - 0.129TT + 0.327TD + 0.141logCKF$	16.4	4/88
Log CK at exsanguination = $-9.933 - 0.308LT + 0.652CT_0 + 0.27SGL - 0.288MG_0 + 0.218logCKU$	57.1	5/47
Serum glucose = $1.027 - 0.164TD + 0.647SL - 0.201logCKE$	60.7	3/86
Serum glycerol = $-19.974 - 0.378TT + 0.246SL + 0.189SpH + 0.242logCKE$	22.7	4/85
Serum lactate = $-0.521 + 0.22FD + 0.665SG + 0.154SGL + 0.13logCKF$	61.0	4/85
Serum pH = $8.03 + 0.138TT + 0.167TD + 0.193SGL - 0.318SL$	19.7	4/85
Carcass temperature = $35.331 + 0.40LT + 0.501TT + 0.329logCKE$	62.7	3/53
Glycogen of LD = $6.435 - 0.241TT - 0.222FD - 0.189ML_0 + 0.202CW - 0.195logCKE$	28.5	5/89
Lactate of LD = $7.907 + 0.49TD - 0.216ST - 0.189MG_0 - 0.203pH_1 + 0.173CC$	35.3	5/89

[a] Regression models have significant F-values ($P < 0.01$).
[b] Abbreviations given in text.
[c] Coefficient of determination $\times 100$
[d] Degrees of freedom.

Table 2. Mean values of studied variables in three selection phases.

Trait	Selection phase			SEM
	First	Second	Last	
Log CK on farm, U/l	3.0	3.2	3.2	0.03
Log CK during unloading, U/l	3.0[a]	3.4[b]	3.2	0.04
Log CK at exsanguination, U/l	3.2	3.4	3.5	0.05
Serum glucose, mmol/l	5.6[a]	5.0[a]	7.8[b]	0.05
Serum glycerol, mmol/l	0.5	0.5	0.6	0.003
Serum lactate, mmol/l	13.3[a]	12.2[a]	16.7[b]	0.06
Serum pH	8.0	8.1[a]	7.8[b]	0.03
Carcass temperature, °C	39.3[a]	38.6	38.1[b]	0.13
Glycogen of LD, μmol/g	27.2[a]	30.2[a]	23.5[b]	0.20
Lactate of LD, μmol/g	38.9	45.6	45.6	0.11
Fasting time, h	22.3[a]	17.5[b]	21.2[a]	0.52
$pH_1 \leq 5.8$, %	8.3	22.7	8.3	—
$5.8 < pH_1 \leq 6.4$, %	66.7	45.5	58.4	—
$6.4 < pH_1$, %	25.0	31.8	33.3	—
Best grading class, %	8.3	38.5	45.8	—
Good grading class, %	50.0	24.6	41.7	—
Poor grading class, %	41.7	36.9	12.5	—
Carcass hot weight, kg	74.7	77.5	74.1	0.79
Number of pigs	12	66	24	—

[a,b] Means within a row with different superscripts are significantly different ($P < 0.05$).

The pigs of the second selection were under greatest stress during transportation (table 2). Consequently, they had the highest CK activities after transport and elevated muscle lactate content. However, these animals had the greatest muscle glycogen content which might be caused by their shortest fasting time and smallest increase in CK values during lairage. Actually, fast duration and carcass hot weight accounted for 8.4 and 2.6 % of the variation in muscle glycogen, respectively (Table 1). On the other hand, selection phase had no significant influence on carcass weight. Furthermore, the pigs of the second selection had the fastest pH fall in the LD muscle 45 min p.m. that explained their highest PSE frequency of 22.7 %. In addition, the percentage of the carcasses of the best grading class i.e. the highest lean meat content was greater in these pigs than in those of the first selection.

The pigs of the last selection were stressed most severely on the journey from producer up to the point of stunning (table 2). Therefore they had the highest CK values at farm and slaughter and the highest content of serum glucose, glycerol and lactate that led to the lowest serum pH value.

Among the variables listed in Table 1, serum lactate was the most predictive one for serum glucose (43.6 %) and pH value (9.4 %), while serum glucose provided the best single estimate of serum lactate (39.4 %). Furthermore, the pigs of the last selection had the lowest content of muscle glycogen (Table 2). This could be explained by fast duration (8.4 %) and by log CK at exsanguination (3.5 %). However, muscle lactate did not vary significantly between the selection phases. Moreover, the pigs of the last selection had the slowest pH fall in the LD muscle 45 min p.m. which accounted for their low PSE fre-

Table 3. Effect of fasting time on porcine stress and meat quality.

Trait		Fasting time, h							
		7	17	18	19	21	24	27	SEM
Log CK on farm,	U/l	3.2	3.4[a]	3.2	3.1	3.0[b]	3.3[a]	3.1	0.03
Log CK during unloading,	U/l	3.3	3.4[a]	3.5[a]	3.4	3.1[b]	3.3	3.2	0.04
Log CK at exsanguination,	U/l	3.5	3.5	3.7[a]	3.4[b]	3.2[b]	3.2[b]	3.6[a]	0.05
Change in log CK*, %		5.9	5.5	16.3[a]	8.2	6.4	− 2.2[b]	15.2[a]	1.68
Serum:									
glucose, mmol/l		5.6	5.0[a]	4.4[a]	5.6[a]	7.8[b]	7.2[b]	6.1	0.05
lactate, mmol/l		12.2	13.3[a]	12.2[a]	12.2[a]	15.6	16.7[b]	15.6	0.06
pH		7.9	7.9[a]	8.1	8.1	7.9[a]	7.9	8.2[b]	0.03
Carcass temperature, °C		38.8	38.0[a]	38.9	38.3[a]	37.6[a]	38.2[a]	39.4[b]	0.13
Glycogen of LD, μmol/g		34.0[a b]	37.0[a]	25.3[b c]	30.2[a b]	27.8[b]	24.1[b c]	18.5[c]	0.20
Lactate of LD, μmol/g		41.1	48.9	46.7	42.2[a]	35.6[a]	52.2[b]	51.1[b]	0.11
$pH_1 \leq 5.8$, %		9.1	25.0	5.0	21.7	8.3	41.7	16.7	—
$5.8 < pH_1 \leq 6.4$, %		54.5	33.3	50.0	30.4	83.4	58.3	66.7	—
$6.4 < pH_1$, %		36.4	41.7	45.0	47.9	8.3	0	16.6	—
Grading class:									
best, %		36.4	25.0	47.4	30.4	33.3	33.3	50.0	—
good, %		18.2	41.6	26.3	39.1	16.7	41.7	33.4	—
poor, %		45.4	33.4	26.3	30.5	50.0	25.0	16.6	—
Carcass hot weight, kg		76.6[a]	82.2[b]	79.1[a b]	76.3[a]	75.3[a]	63.9[c]	77.6[a b]	0.79
Number of pigs		12	12	20	23	12	12	12	—

[a, b, c] Means within a row with different superscripts are significantly different (P < 0.05).

*   Change in log $CK = 100 \times \dfrac{\log CK \text{ at exsanguination} - \log CK \text{ on farm}}{\log CK \text{ on farm}}$.

quency. Finally, the percentage of the carcasses of the best grading class was highest in this group.

Fasting time

The effects of fasting time on some blood characteristics and carcass traits are presented in table 3. The occurrence of reactors was 9.1, 0, 0, 17.4, 0, 0 and 8.3 % in the pigs fasted for 7, 17, 18, 19, 21, 24 and 27 h, respectively. Fast duration was one of the most contributing components for serum lactate (17.6 %) and muscle glycogen (8.4 %). While in the other models listed in Table 1, fasting made non-significant contributions.

The linear effect of fast duration on the studied traits (Table 3) was evaluated by simple regression analysis. Fasting time had a significant positive influence on serum lactate and glucose, and muscle lactate content. Consequently, it was estimated that a prolongation of fast duration (FD) from 6 to 18 h increased the level of serum lactate (SL) from 8.0 to 12.9 mmol/l, glucose (SG) from 4.0 to 6.1 mmol/l and muscle lactate (ML_0) from 39.2 to 44.6 µmol/g. The respective equations were $SL = 5.548 + 0.407FD$ (R^2 $100 = 17$ %, $P < 0.001$), $SG = 2.997 + 0.171FD$ (R^2 $100 = 11$%, $P = 0.001$) and $ML_0 = 36.507 + 0.449FD$ (R^2 $100 = 4$ %, $P < 0.05$).

In contrast, the prolonged fasting time reduced significantly muscle glycogen, pH_1 value and carcass hot weight in this study. So an increase in fast from 6 to 18 h reduced muscle glycogen from 37.9 to 28.8 µmol/g, pH_1 value from 6.45 to 6.23 and carcass hot weight from 80.4 to 76.1 kg. In addition, the respective equations were $MG_0 = 42.473 - 0.759FD$ (R^2 $100 = 11$ %, $P = 0.001$), $pH_1 = 6.559 - 0.018FD$ (R^2 $100 = 6$ %, $P < 0.05$) and $CW = 82.59 - 0.341FD$ (R^2 $100 = 5$ %, $P < 0.05$).

Pigs showed the highest blood CK values after an 18 or 27 h fasting period. On the other hand, fast times of 7—27 h were not significantly related to blood CK, serum glycerol or carcass temperature, however the highest CK activity and carcass temperature were found after a 27 h fast period. Furthermore, the fall in the LD muscle pH 45 min p.m. was fastest after a 24 h fast and slowest after an 18—19 h fast.

Transport

Table 1 shows that the loading time of day was one of the most contributing variables for log CK during unloading (3.9 %) and carcass temperature (14.6 %). In addition, transport temperature was one of the best predictors of carcass temperature (35.2 %), muscle glycogen (10.5 %), serum glycerol (11.2 %) and pH (3.2 %), and log CK during unloading (1.4 %). Furthermore, transport duration made major contributions of the variation in muscle lactate (15.7 %), serum glucose (13.7 %) and pH (5.5 %), and log CK during unloading (8.7 %).

The effects of loading time, transport temperature and duration on porcine stress were evaluated by the change in CK activity from the farm up to the unloading (figure 1). In general, pigs were fed a restricted ration twice a day at 8—9 o'clock a.m. and at 2—3 o'clock p.m. Results suggested that pigs were stressed most severely during transport if loaded just before the morning or afternoon feed. In contrast, transport was least stressful when pigs were loaded at 9—10 o'clock a.m. that was between the two feeding periods.

During transport, pigs were stressed significantly if a heavy frost ($-20°C$), still more significantly if a small frost ($-4°C$) and most significantly in warm weather ($15°C$). Whereas, transportation in cool weather ($8°C$) was least stressful for pigs (figure 1). These results were supported by the frequency of transport losses. Actually, the mortality was low during a heavy frost (-30 to $-20°C$), while it increased considerably in cold weather (-3 to $0°C$), and maximum mortality was found in warm weather (14 to $24°C$).

In general, increasing transport time resulted in elevated CK values (figure 1). Thus, pigs were stressed least during a trip of 17 min, while maximum stress was found during the
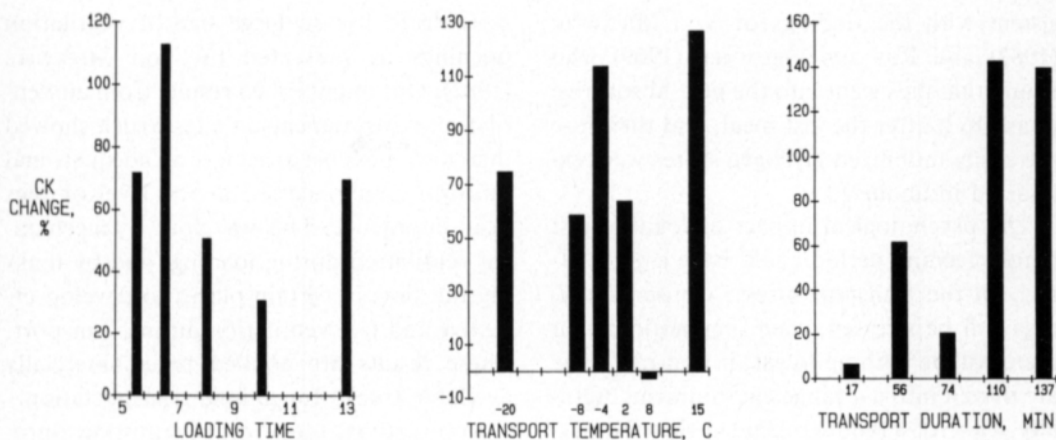
Fig. 1. Effect of loading time of day, transport temperature and duration on the increase of blood creatine kinase (CK) activity.

$$\text{CK change (\%)} = 100 \times \frac{\text{CK during unloading} - \text{CK on farm}}{\text{CK on farm}}$$

journeys of 110 and 137 min. An exception to these findings was the low CK change after a journey of 74 min that could be due to high CK activity of the pigs on farm. On the other hand, mortality was low during the 10—20 min transport, while maximum mortality occurred during the 45—80 min transportation. However, mortality decreased prominently during the journeys of 120 to 420 min.

Discussion

The pigs of the first selection were least stress-susceptible and had the best meat quality, however their carcasses had poorer grading class than those of the later selection phases. Whereas, the pigs of the second selection were heavier than the others, in addition, they were most susceptible to the development of PSE at slaughter. The pigs of the last selection were under stress throughout and therefore they had the slowest pH fall in the LD muscle 45 min p.m. Moreover, carcasses of this group had the highest percentage of the best grading class.

Results suggested that the best possible pork quality for the meat industry will be obtained when pigs are grown fast and evenly so that

there will be no significant differences in liveweights between the animals of the same group. Thus, the psychological impact of selecting pigs in different phases could have a significant effect on the development of porcine stress due to changes in the familiar group. These will result in hierarcial fighting and other negative behavioral changes in the rest of animals.

This study considered the fasting times that are used in commercial practice. Moreover, there will be a trend to shorten the lairage times nowadays, hence fast duration will also be reduced. This is made possible by the breeding activities which improved significantly the stress-resistance of the Finnish pigs since 6 years.

In this work, muscle pH_1 value, glycogen and lactate content suggested that the most acceptable pork quality for meat products was obtained after a fast of 6—7 h when pigs were fed twice a day. Because of increased risk of transport losses due to feeding, the last meal should be lighter by half than usual, in addition, pigs should have free access to drink water. Furthermore, it was not recommended to fast pigs for 17—27 h due to overnight holding which had a negative effect on muscle lactate and glycogen content. This result was con-

sistent with the findings of Von MICKWITZ (1982) and RIIS and GRUMMER (1969) who found that pigs went into the post-absorptive phase 16 h after the last meal, and therefore the easily mobilized glycogen stores were exhausted in about 20 h.

The psychological impact of loading just before feeding period could have a great effect on the transport stress. Consequently, pigs will be stressed when they noticed that there will be nothing to eat, in contrast, they are driven into a strange environment. Actually, GONYOU (1986) concluded that the psychological impact of the situation is more important in determining the response than the physical.

The present results suggested that the high transport losses and CK values in cold weather ($-6-0°C$) occurred due to fluctuating temperature and due to closed ventilation slots which were not opened until air temperature raised over $0°C$. Therefore, the improved cooling of pigs due to better ventilation could decrease porcine stress at temperatures of 5 to $10°C$. Above external temperatures of $10°C$ pigs were not able to maintain constant body temperature during trucking which accounted for the heat stress that led to increased CK activity and mortality during transport.

The weather is considerably colder in Finland than in Central Europe, therefore it is not possible to use so large size of ventilation openings as presented by Von MICKWITZ (1982). Our unpublished results from air ventilation measurements in a test truck showed that it was possible to achieve an adequate and uniform air circulation at the level of pigs head in practice. This was done by mechanical ventilation during loading, and by making the slots in certain places to develop effective and free ventilation during transport. These results are applied to commercially available trucks for animal transportation.

In this study, the effect of transport duration on mortality was consistent with the increase in CK activity during transport. Thus, it was found that mortality was low during the short transport times of 10—25 min, while maximum losses occurred during the 45—80 min transport. These results suggested that the most stress-sensitive pigs will already die during a relative short 45 min transport, whereas stress resistant animals will survive and showed an increased CK activity during unloading.

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SELOSTUS

Karsintavaiheen, paaston ja kuljetuksen vaikutus sian stressaantumiseen ja PSE-lihan muodostumiseen

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Työssä tutkittiin karsintavaiheen, paaston ja kuljetuksen vaikutusta sian stressaantumiseen ja PSE-lihan muodostumiseen.

Alkukarsinnassa sikojen veren kreatiinikinaasi- (CK-) aktiivisuus oli alhaisin ja tällöin niissä oli vähiten PSE:tä. Sen sijaan karsitut siat stressaantuivat kuljetuksessa eniten, niiden PSE-% oli korkein. Lopputyhjennyksessä sikojen CK-aktiivisuus oli korkea tilalta pistoon, edelleen niiden pitkän selkälihaksen pH laski muita hitaammin 45 min *postmortem*. Ruhon lihaprosentti oli korkein lopputyhjennyksen ja alhaisin alkukarsinnan sioissa.

Kahdesti päivässä ruokittujen sikojen paastotessa 6—7 h ennen teurastusta niiden lihassa oli niin paljon glykogeeniä ja niin vähän maitohappoa, että lihan laatu oli moitteeton. Sikojen lepo yön yli teurastamon navetassa alensi ruholihan glykogeenitasoa ja lisäsi sen maitohappopitoisuutta.

Kuljetus stressasi sikoja eniten silloin, kun ne lastattiin juuri ennen aamu- tai iltaruokintaa tai jos eläimet kuljetettiin lämpimällä ilmalla (yli 10°C). Sikojen autokuolleisuus ja veren CK-aktiivisuus kuljetuksessa olivat korkeimmillaan samalla ulkolämpötila-alueella. Mitä pitempi oli kuljetusaika sitä enemmän veren CK-aktiivisuus kohosi tilalta teurastamoon.