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Glutathione peroxidase activity, tissue and soluble selenium content in beef and pork in relation to meat ageing and pig RN phenotype

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Abstract

Since glutathione peroxidase (GSHPx) may be important for meat quality, its activity was examined in relation to animal species (beef and pork), muscle type (oxidative and glycolytic), selenium content, meat ageing and RN phenotype in Hampshire crossbred pigs. The GSHPx activity in bovine *M. Longissimus dorsi* [LD; 1.9 (0.4) U/g, mean (S.D.)] was significantly higher ($P < 0.001$) than in *M. Psoas major* [PM; 1.5 (0.5) U/g] and the activities in the two muscles were correlated ($r = 0.66$; $P < 0.001$). Pork LD had a several fold lower GSHPx activity [0.4 (0.2) U/g] than bovine muscles but, in contrast, a somewhat higher content of selenium [113 (15) ng/g] than bovine LD [106 (12) ng/g] and PM [95 (17) ng/g]. The proportion of soluble selenium in bovine LD (72%) tended to be higher than that in bovine PM (64%), and it was significantly lower in pork LD (52%). Significant correlations between GSHPx activity and soluble selenium were obtained in both beef and pork. Thus, GSHPx activity varied among single animals, with species and muscle type, whereas meat ageing and pig RN phenotype had no effect.

Keywords: Glutathione peroxidase; Selenium; Meat ageing; RN phenotype; Lipid oxidation; Meat quality

1. Introduction

Meat is sensitive to lipid oxidation, which leads to quality deterioration and loss of both nutritional value and safety. Meat contains iron, either bound to proteins, such as myoglobin, hemoglobin, ferritin and transferrin, or non-protein-bound and different forms of iron can accelerate the rate of lipid oxidation (Kanner, Hazan & Doll, 1988; Monahan, Crackel, Gray, Buckley & Morrissey, 1993). On the other hand, endogenous antioxidants in the muscle can control the oxidation in several ways (Chan & Decker, 1994). These include the fat-soluble α-tocopherol and ubiquinone, along with the water-soluble ascorbic acid and histidine-containing dipeptides. Superoxide dismutase, catalase and glutathione peroxidase are antioxidative enzymes also contributing to the oxidative defence (Frankel, 1998).

Glutathione peroxidase (GSHPx; E.C. 1.11.1.9) is a selenium-containing enzyme, catalysing the reduction of lipid and hydrogen peroxides to less harmful hydroxides (Burk, 1997; Cohen & Hochstein, 1963). The mammalian glutathione peroxidase family contains four selenoproteins, cellular, extracellular, phospholipid hydroperoxide, and gastrointestinal GSHPx (Huang, 1996; Ursini et al., 1995). At least 10 other selenoproteins have been found in mammals, but their importance for meat quality is not known.

Another factor possibly influencing the susceptibility of muscle to lipid oxidation is the predominance of either oxidative or glycolytic fibres, but previous comparisons of GSHPx activity in these two types of muscles have not given consistent results (DeVore, Colnago, Jensen & Greene, 1983; Lee, Mei & Decker, 1996; Yamauchi, Yada, Ohashi & Pearson, 1984).

With respect to effects of post-mortem handling DeVore and Greene (1982) found that GSHPx retained its activity throughout the rigor process in beef semitendinosus muscle, but only limited data have been found in the literature concerning the stability of GSHPx during meat ageing. Another important parameter affecting the pig muscle quality in Hampshire crossbred pigs is the RN^- gene (Rendement Napole), but no previous results on GSHPx activity in relation to the RN phenotype have been reported.

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In this paper factors affecting the GSHPx activity are examined, including the effect of meat ageing in bovine *M. Longissimus dorsi* and *M. Psoas major* and pork *M. Longissimus dorsi*, the effect of phenotype (RN^- and rn^+rn^+) of Hampshire crossbred pigs, and the relation to tissue and soluble selenium.

2. Material and methods

2.1. Chemicals

Hydrochloric acid, nitric acid and perchloric acid were all of pure analytical grade. t-Butylhydroperoxide and glutathione reductase were purchased from Merck (Germany) and Boehringer Mannheim (Germany), respectively. Glutathione, NADPH and mercaptosuccinate were purchased from Sigma Chemical Co (USA). Other chemicals were of reagent grade.

2.2. Meat sources and ageing

2.2.1. Beef

M. Longissimus dorsi (LD) and *M. Psoas major* (PM; $n=9$) were obtained from steers weighing approximately 300 kg, from a slaughterhouse in Kävlinge, Sweden. The samples were provided 1 day after slaughter, and each muscle was divided into three parts — rear, middle and front part — and vacuum-packed in separate plastic bags (Multiseven 78). One part, from each muscle was immediately frozen at −20°C (1 day of cold storage). The others were placed in a cold room (4°C) and then frozen at −20°C after storage times of 7 and 14 days, respectively. The three parts (rear, middle and front parts) from each muscle were randomised to the three treatments. In this study, PM was considered to be an oxidative muscle and LD a glycolytic one (Hunt & Hedrick, 1977).

2.2.2. Pork

M. Longissimus dorsi ($n=14$) were obtained from a slaughterhouse in Kristianstad, Sweden. One slice (1 cm thick) from each muscle was taken for glycogen determination. LD was divided lengthways into two parts and separately vacuum-packed in plastic bags (Multiseven 78). One part was immediately frozen at −20°C (1 day of cold storage). The other part was kept at 4°C for 4 days and then frozen at −20°C.

2.3. Sample preparation

After thawing, the samples were kept on ice. All visible fat was removed prior to grinding through a plate with holes 6 mm in diameter. Samples were then diluted (1:4) with cold potassium phosphate buffer (80 mmol/l) containing 5 mmol/l EDTA, 2 mmol/l glutathione (pH 7.6) and homogenised in an Omnimixer (Sorvall Inc, Connecticut, USA) for 20 s at maximum speed. After centrifugation at 5000 ***g*** for 20 min (4°C) (Sorvall RC-5), the supernatant was filtered and frozen at −70°C.

2.4. Glutathione peroxidase activity

The activity of glutathione peroxidase was measured by a coupled assay procedure (Chen, Lindmark Månsson & Åkesson, 2000), recording the oxidation of NADPH by the decrease in absorbance at 340 nm. The reaction medium contained 50 mmol/l potassium phosphate buffer (pH 7.6), 5 mmol/l EDTA, 0.63 mmol/l glutathione, 0.25 mmol/l NADPH, 5 μg/ml glutathione reductase and 0.1 mmol/l t-butylhydroperoxide in a total volume of 0.8 ml. For every type of sample, the assay was run using a blank consisting of the complete incubation mixture plus mercaptosuccinate (4 mmol/l), an inhibitor of GSHPx (Chaudiere, Wilhelmsen & Tappel, 1984), to exclude possible interfering activity from other NADPH oxidizing enzymes. The NADPH oxidation, in such blanks, was very similar to that in blanks without added enzyme source. The GSHPx activity in meat was expressed as U/g (wet wt.), where one unit (U) was defined as 1 μmol oxidized NADPH/min. A serum control was included in every assay. The within-run c.v. was 2.9% and the between-run c.v. 7.8%.

2.5. Selenium analysis

The samples (meat 0.5 g; supernatant 3 g) were wet-digested using 1 ml perchloric acid and 4 ml nitric acid. After standing overnight at room temperature, the mixture was heated in an aluminium block (Tecator, Sweden) to 210°C using a temperature program (Autostep; 105°C, ramp 10 min, hold 30 min; 150°C, ramp 10 min, hold 30 min; 200°C, ramp 10 min, hold 30 min; 210°C, ramp 5 min, hold 60 min). Concentrated hydrochloric acid (2 ml) was added and selenate was reduced to selenite by treatment at 70°C for 15 min. After dilution to 25 ml in 1.2 M hydrochloric acid, the selenium content was measured using graphite furnace hydride generation atomic absorption spectrometry (GF-HG-AAS; Perkin Elmer Aanalyst 800) combined with flow-injection analysis (FIAS-400), using sodiumborohydride [0.2% (w/v) in 0.05% (w/v) NaOH] as a reducing agent. A selenate standard solution (Merck, Germany) was diluted and reduced to selenite. The final standard selenium concentrations were 0.4–1.6 μg/l. Bovine liver (NIST 1577b) was included as a reference material in every assay.

2.6. Phenotype assignment

Determination of RN phenotype was performed by measuring the amount of glycogen, expressed as μmol/g meat (Keppler & Decker, 1970).

2.7. Statistical analysis

The significance of differences were assessed by using analysis of variance (ANOVA) or Student's *t*-test. Correlations were obtained from the Pearson correlation matrix.

3. Results

3.1. Glutathione peroxidase activity and selenium content

The GSHPx activity was generally higher in beef LD than in PM ($P \leqslant 0.001$) but not in day 1 samples, and it was significantly higher in both beef muscles than in pork ($P < 0.001$; Table 1). Also, the selenium content was significantly higher in beef LD than in PM muscle, but pork LD contained more selenium than beef PM. The variation in GSHPx activity between animals was larger than that in selenium content, both in beef and pork. In beef LD, the activity ranged from 1.1 to 2.7 U/g, in beef PM from 0.6 to 2.5 U/g and, in pork LD, from 0.1 to 0.9 U/g. Interestingly, there was a significant correlation of GSHPx activity between LD and PM from the same beef animals ($r = 0.66$; $P < 0.001$).

Since GSHPx activity analysis was performed in the supernatant from meat homogenates, it was interesting to measure the amount of soluble selenium. This approach would also give an indication of the amount of insoluble seleno compounds. The proportion of soluble selenium tended to be higher in beef LD than in PM (Table 1). In pork LD, the proportion of soluble selenium was significantly lower than that in beef, accounting for only half of the tissue selenium.

The correlation of soluble selenium to GSHPx activity was significant in the bovine muscles (Table 2). Exclusion of one data point also resulted in a significant correlation between GSHPx activity and soluble selenium in pork LD. In contrast, the correlation between tissue selenium content and GSHPx activity did not reach statistical significance for either of the bovine muscles or pork. When these data from beef LD and PM were combined, however, a significant correlation appeared. Tissue selenium and soluble selenium were significantly correlated in beef PM and in pork, but not in beef LD.

3.1.1. Influence of meat ageing and pig RN phenotype

An important part of the present study was to investigate the effect of ageing on GSHPx activity. No significant change of its activity due to cold storage for 4 days (pork) and 14 days (beef) was found (Table 3). It was also ascertained that the GSHPx activity did not vary significantly among the different parts of a single muscle.

Muscles from carriers and non-carriers of the RN^- gene in Hampshire crossbred pigs differ in composition, but the influence of this gene on the activity of GSHPx and selenium content has not been previously investigated. In this study no significant difference was found, either in GSHPx activity or selenium content, between carriers and non-carriers of the RN^- gene (Table 4).

4. Discussion

Several factors can influence the susceptibility to lipid oxidation in muscle tissue. Previously, a higher susceptibility to lipid oxidation in oxidative compared to glycolytic muscle was observed, probably due to the higher content of heme proteins, iron and mitochondria in oxidative muscle (Kanner et al., 1988; Lawrie, 1979). Among the muscles used in the present study, PM was considered to be the oxidative muscle and LD the glycolytic one (Hunt & Hedrick, 1977), but a higher GSHPx activity was found in beef LD than in beef PM. Previously, a similar tendency with a higher GSHPx activity in chicken and pig breast muscle (glycolytic) than in thigh muscle (oxidative) was reported (Yamauchi et al., 1984), but other authors have made opposite findings in turkey (Lee et al., 1996) and chicken muscles (DeVore et al., 1983). It has also been proposed that a

Table 1
Tissue and soluble selenium (ng/g wet weight) in beef *M. Longissimus dorsi* (LD) and *M. Psoas major* (PM) and pork LD[a]

	Beef				Pork			
	n	LD	PM	*P*[b]	*N*	LD	*P*[c]	*P*[d]
GSHPx[e] activity (U/g)	9	1.8 (0.4)	1.5 (0.5)	0.115	14	0.4 (0.2)	< 0.001	< 0.001
Tissue selenium (ng/g wet wt.)	9	106 (12)	95 (17)	< 0.05	14	113 (15)	0.24	< 0.01
Soluble selenium (ng/g wet wt.)	9	76 (10)	60 (11)	< 0.01	14	59 (9)	0.001	0.84
Soluble selenium/tissue selenium (%)	9	72 (10)	64 (9)	0.14	14	52 (6)	< 0.01	< 0.01

[a] Data are expressed as mean (S.D.).
[b] Significance of difference between beef LD and PM.
[c] Significance of difference vs. beef LD.
[d] Significance of difference vs. beef PM.
[e] Glutathione peroxidase.

Table 2
Correlations of glutathione peroxidase (GSHPx) activity, tissue and soluble selenium content of beef *M. Longissimus dorsi* (LD) and *M. Psoas major* (PM) and pork LD[a]

	n	GSHPx vs. tissue selenium	GSHPx vs. soluble selenium	Tissue selenium vs. soluble selenium
Beef LD	9	0.51 ($P=0.16$)	0.78 ($P<0.05$)	0.30 ($P=0.43$)
Beef PM	9	0.58 ($P=0.10$)	0.79 ($P<0.05$)	0.80 ($P=0.01$)
Beef LD + PM	18	0.63 ($P<0.01$)	0.84 ($P<0.001$)	0.68 ($P<0.01$)
Pork LD	14	0.21 ($P=0.48$)	0.45 ($P=0.11$)[b]	0.60 ($P<0.05$)

[a] Linear correlation coefficients are shown.
[b] When eliminating one data point the value was 0.76 ($P<0.01$).

Table 3
Glutathione peroxidase activity (U/g) in beef *Longissimus dorsi* (PM) and *M. Psoas major* (LD) and pork LD after ageing for different times[a]

	Ageing time (days)				
	n	1	4	7	14
Beef LD	9	1.8 (0.4)		1.9 (0.5)[b]	1.8 (0.4)[c]
Beef PM	9	1.5 (0.5)		1.4 (0.5)	1.4 (0.6)
Pork LD	14	0.4 (0.2)[d]	0.4 (0.2)		

[a] Data are expressed as mean (S.D.).
[b] The statistical significance of the difference from beef PM (7 days) was $P\leqslant 0.001$.
[c] The statistical significance of the difference from beef PM (14 days) was $P<0.01$.
[d] The statistical significance of the difference from beef LD and PM (1 day) was $P<0.001$.

Table 4
Glutathione peroxidase activity (U/g) and selenium content (ng/g wet wt.) from carriers and non-carriers of the RN^- gene in Hampshire crossbred pigs. Mean (S.D.).

	n	Carriers	*n*	Non-carriers
GSHPx activity (U/g)	5	0.5 (0.2)	7	0.4 (0.3)
Tissue selenium (ng/g wet wt.)	5	115 (5)	7	116 (19)
Soluble selenium (ng/g wet wt.)	5	58 (5)	7	60 (11)
Soluble selenium/tissue selenium (%)	5	50 (4)	7	52 (7)

high GSHPx activity corresponds to low tocopherol content in chicken and porcine muscles and that this functions as a compensational relationship in the defence against lipid oxidation (Yamauchi et al., 1984). A basis for this proposal was the observed higher content of α-tocopherol in the oxidative muscle, which also holds when animals are fed with different amounts of α-tocopherol (Cheah, Cheah & Krausgill, 1995; Chan & Decker, 1994).

Another important parameter affecting the pig muscle quality is the RN^- gene (Rendement Napole) in Hampshire crossbred pigs. Approximately 70% of all slaughter pigs in Sweden are three-way crosses with Hampshire as a terminal sire (Lundström, Enfäldt, Tornberg & Agerhem, 1998), indicating a high probability that pigs may carry the dominant RN gene. Meat from carriers has a higher glycogen content but lower water holding capacity, Napole yield, protein content, and ultimate pH than meat from non-carriers (Estrade, Vignon & Monin, 1993; Fjelkner-Modig & Tornberg, 1986; Monin & Sellier, 1985). Due to these large differences in muscle composition, it was of interest also to study any possible relations to the activity of GSHPx and selenium content. The results in this paper imply that the RN^- gene had no significant effect, either on the GSHPx activity or the selenium content in pork.

GSHPx activities were higher in beef than in pork, which is in accordance with previous results (Mei, Crum & Decker, 1994). Also, GSHPx activity in turkey thigh muscle was found to be lower than that in beef but higher than that in pork (Lee et al., 1996). Others reported a somewhat lower GSHPx activity in beef and pork than found in this paper (Lee, Mei & Decker, 1997; Mei et al., 1994), but no data on selenium content were given in those studies. When comparing GSHPx activity results between studies, it is also important to note that different assays and units have been employed.

Divergent results are also observed when comparing selenium content in muscle from different animal species and countries (Table 5). The selenium content of beef reported in this paper was lower than recent data from Finland (Eurola, Ekholm, Ylinen, Koivistoinen & Varo, 1991) but higher than data obtained in Finland before 1985 and in Sweden (Agerhem, Kolar, Laser, Reuterswärd & Nickels, 1983; Jorhem, Sundström, Åstrand & Haegglund, 1989; Koivistoinen, 1980). Our data on the selenium content in pork was higher than the results obtained in Finland before 1985, Spain and Sweden (Díaz-Alarcón, Navarro-Alarcón, López-García de la Serrana & López-Martínez, 1996; Koivistoinen, 1980; Jorhem et al., 1989), but lower than other data from Sweden (Agerhem et al.) and more recent data from Finland (Eurola et al.). The higher selenium content in pork compared to beef found in this paper is in accordance with results from several other countries (Agerhem et al., 1983; Jorhem et al., 1989; Tinggi, 1999).

As shown by studies before and after initiation of selenium fertilization in Finland, feed composition has a marked effect on meat selenium content (Ekholm, Ylinen, Koivistoinen & Varo, 1990), but no data on the

Table 5
Selenium contents (μg/g wet wt.) in different species and from different studies[a]

	Bovine	Pig	Reference
Muscle	0.03[b]	0.133[b]	Agerhem et al. (1983)
	0.05–0.10	0.030–0.09	Koivistonen (1980)
		0.061–0.116	Díaz-Alarcón et al. (1996)
	0.106 (0.012)[b]	0.113 (0.015)[b]	Present study
	0.095 (0.017)[c]		Present study
Minced meat	0.077–0.142		Tinggi (1999)
	0.030 (0.020)	0.094 (0.017)	Jorhem et al. (1989)
	0.141–0.153[d]		Eurola et al. (1991)

[a] Data are presented either as mean (SD) or as range.
[b] *M. Longissimus dorsi*.
[c] *M. Psoas major*.
[d] Converted from dry weight to wet weight using a factor of 0.3 (70% water content).

feed composition were available in our study. To avoid muscle degenerative diseases depending on selenium deficiency, it has been legally permitted in Sweden since 1980 to supplement feed to farm animals with selenium as inorganic selenite. In controlled experiments, increasing doses of selenite in the cow feed increased selenium content and GSHPx activity in several organs, including skeletal muscle and myocardium (Pehrson, 1985). Further experiments showed higher selenium content in skeletal muscle from dairy cows supplemented with selenium yeast than in those supplemented with the same amount of selenium as selenite, indicating a higher bioavailability of organic selenium compared to inorganic selenium (Ortman & Pehrson, 1997).

Considering the correlations between selenium and GSHPx activity, most of the work done so far has concerned blood or serum, but not much work has been done on muscle selenium and GSHPx activity in different animals. A significant correlation between GSHPx activity and tissue selenium was found in bovine semitendinosus muscle (DeVore et al., 1983) and, in the present study for the combined bovine LD and PM samples (Table 2). Much higher correlations with GSHPx activity were found for soluble selenium than for tissue selenium. This indicates that GSHPx is an important part of the soluble selenium fraction. Moreover, the significant correlation between GSHPx activities in beef LD and PM indicates that GSHPx status was consistent in muscles within each animal.

The relationships between GSHPx and selenium vary with animal species as shown by the absence of a correlation between GSHPx and tissue selenium and the higher ratio of selenium to GHSPx in pig LD compared to bovine muscles observed in this paper. The latter finding, along with the lower percentage of soluble selenium, supports the conclusion that pork contains a higher proportion of non-GSHPx selenoproteins or other selenium compounds than beef. This has not been previously reported and thus, further studies on the nature of other selenoproteins are necessary. Similarly, Thompson, McMurray and Blanchflower (1976) found a significant correlation between cellular GSHPx activity and selenium in cow and sheep blood but not in pig blood, and they also found a higher ratio of selenium to GSHPx in pig blood than in bovine blood. These findings indicate that pig blood also contains a higher proportion of insoluble selenoproteins or other selenium compounds compared to bovine blood.

Holding of unprocessed meat above the freezing point is known as ageing and it is associated with an increase in tenderness. About 50% of the tenderisation occurs within 24 h after slaughter and the remaining 50% of tenderisation takes place during ageing (Dransfield, 1994). It is generally accepted that proteolysis by endogenous enzymes is an important part of tenderisation. The rate of ageing increases with higher temperatures, and faster rigor development, and differs significantly between species (Lawrie, 1979).

During ageing, enzymes may lose activity or be redistributed between cellular compartments (Spanier, McMillin & Miller, 1990). Considering alterations within the muscle starting at slaughter, denaturation, and thereby, inactivation of GSHPx with time is a possibility. Since GSHPx is almost exclusively a soluble enzyme (Pehrson, 1985), no increase in soluble enzyme activity would be expected during ageing due to redistribution. A main finding in the present paper was that GSHPx remained stable for 4 and 14 days of ageing in pork and beef, respectively. Only few previous studies have been made on this point. Renerre, Dumont & Gatellier (1996) found that GSHPx activity tended to decrease 10–30% in four bovine muscles during ageing for 8 days at 2°C. Two other antioxidative enzymes, superoxide dismutase and catalase, were investigated in the latter study and superoxide dismutase decreased significantly, but catalase retained its activity after 8 days of storage. Maybe the difference in GSHPx stability between the two studies is due to use of wrapping in an oxygen permeable film (Renerre et al., 1996) and vacuum packaging in the present study, respectively. Other studies on GSHPx activity at cold storage have been performed with sheep, cattle and pig plasma, which contain the extracellular form. GSHPx in pig plasma lost 40% of its activity during cold storage for 7–14 days (Zhang, Ku, Miller & Ullrey, 1986), but its stability in sheep and bovine plasma was markedly lower (Davidson, Kennedy, Hughes & Blanchflower, 1990). Since the enzymes usually have a high degree of homology, the surrounding matrix may be important for enzyme stability. These findings also necessitate further studies of GSHPx activity during meat processing.

5. Concluding remarks

GSHPx activity, tissue and soluble selenium content were lower in a bovine oxidative (PM) than in a glycolytic (LD) muscle. Differences were also observed between pork and beef, GSHPx activity and the proportion of soluble selenium compounds being significantly lower in pork, although the tissue selenium content was quite similar. The higher ratio of selenium to GSHPx, along with the lower percentage of soluble selenium in pig LD compared to bovine muscles, supports the conclusion that pork contains a higher proportion of non-GSHPx selenoproteins or other selenium compounds than beef. GSHPx activity was also correlated with soluble selenium in both species rather than with the tissue selenium content, which was expected since it is a soluble enzyme. Furthermore, the pig RN phenotype had no significant effect on the GSHPx activity, indicating that it is not necessary to consider this variable in future studies of this enzyme. GSHPx also remained stable after meat ageing in both beef and pork, which makes its participation possible in the oxidative defence of the muscle.

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References

Agerhem, H., Kolar, K., Laser Reuterswärd, A., & Nickels, C. (1983). Selenium content in animal food before and after heat treatment. *Köttforskningsinstitutets rapport, 1*(49), 2571.

Burk, R. F. (1997). Selenium-dependent glutathione peroxidases. In F. P. Guengerich, *Comprehensive toxicology* (pp. 229–242). London: Pergamon.

Chan, K. M., & Decker, E. (1994). Endogenous skeletal muscle antioxidants. *Critical Reviews in Food Science and Nutrition, 34*, 403–426.

Chaudiere, J., Wilhelmsen, E. C., & Tappel, A. L. (1984). Mechanism of selenium-glutathione peroxidase and its inhibition by mercaptocarboxylic acids and other mercaptans. *Journal of Biological Chemistry, 259*, 1043–1050.

Cheah, K. S., Cheah, A. M., & Krausgill, D. I. (1995). Effect of dietary supplementation of vitamin E on pig meat quality. *Meat Science, 39*, 255–264.

Chen, J., Lindmark-Månsson, H. & Åkesson, B. (2000). Optimisation of a coupled enzymatic assay of glutathione peroxidase activity in bovine milk and whey. *International Dairy Journal*, *10*, 347–351.

Cohen, G., & Hochstein, P. (1963). Glutathione peroxidase: the primary agent for elimination of hydrogen peroxides in erythrocytes. *Biochemistry, 2*, 1420–1428.

Davidson, W. B., Kennedy, D. G., Hughes, P. J., & Blanchflower, W. J. (1990). The stability of glutathione peroxidase activity in plasma from cattle, pigs and sheep on storage in the presence and absence of glutathione. *Veterinary Research Communications, 14*, 441–446.

DeVore, V. R., & Greene, B. E. (1982). Glutathione peroxidase in post-rigor bovine semitendinosus muscle. *Journal of Food Science, 47*, 1406–1409.

DeVore, V. R., Colnago, G. L., Jensen, L. S., & Greene, B. E. (1983). Thiobarbituric acid values and glutathione peroxidase activity in meat from chickens fed a selenium-supplemented diet. *Journal of Food Science, 48*, 300–301.

Díaz-Alarcón, J., Navarro-Alarcón, M., López-García de la Serrana, H., & López-Martínez, M. C. (1996). Determination of selenium in meat products by hydride generation atomic absorption spectrometry — selenium levels in meat, organ meats, and sausages in Spain. *Journal of Agricultural and Food Chemistry, 44*, 1494–1497.

Dransfield, E. (1994). Optimisation of tenderisation, ageing and tenderness. *Meat Science, 36*, 105–121.

Ekholm, P., Ylinen, M., Koivistoinen, P., & Varo, P. (1990). Effects of general soil fertilization with sodium selenate in Finland on the selenium content of meat and fish. *Journal of Agricultural and Food Chemistry, 38*, 695–698.

Estrade, M., Vignon, X., & Monin, G. (1993). Effect of the RN^- gene on ultrastructure and protein fractions in pig muscle. *Meat Science, 35*, 313–319.

Eurola, M. H., Ekholm, P. I., Ylinen, M. E., Koivistoinen, P. E., & Varo, P. T. (1991). Selenium in Finnish foods after beginning the use of selenate-supplemented fertilisers. *Journal of the Science of Food and Agriculture, 56*, 57–70.

Fjelkner-Modig, S., & Tornberg, E. (1986). Water distribution in porcine *M. Longissimus* dorsi in relation to sensory properties. *Meat Science, 17*, 213–223.

Frankel, E. N. (1998). *Lipid oxidation*. Dundee, Scotland: The Oily Press Ltd.

Huang, W. (1996). Extracellular glutathione peroxidase. *Purification, immunoassay, nutritional regulation and clinical aspects*. Doctoral thesis, University of Lund.

Hunt, M. C., & Hedrick, H. B. (1977). Profile of fibre types and related properties of five bovine muscles. *Journal of Food Science, 42*, 513–517.

Jorhem, L., Sundström, B., Åstrand, C., & Haegglund, G. (1989). The levels of zinc, copper, manganese, selenium, chromium, nickel, cobalt, and aluminium in the meat, liver and kidney of Swedish pigs and cattle. *Zeitschrift für Lebensmittel Untersuchung und Forschung, 188*, 39–44.

Kanner, J., Hazan, B., & Doll, L. (1988). Catalytic "free" iron ions in muscle foods. *Journal of Agricultural and Food Chemistry,* 36, 412–415.

Keppler, D., Decker, K. (1970). Glycogen. Determination with amyloglucosidase. In: H.U. Bergmeyer. *Methoden der enzymatischen analyse* Vol II, pp. 1127–1131. Weinheim, Germany: Verlag Chemie.

Koivistonen, P. (1980). Mineral element composition of Finnish foods: N, K, Ca, Mg, P, S, Fe, Cu, Mn, Zn, Mo, Co, Ni, Cr, F, Se, Si, Rb, Al, B, Br, Hg, As, Cd, Pb and ash. *Acta Agriculturæ Scandinavica, Supplementum 22*, 1–171.

Lawrie, R. A. (1979). *Meat science* (3rd ed.). Oxford: Pergamon Press.

Lee, S. K., Mei, L., & Decker, E. A. (1996). Lipid oxidation in cooked turkey as affected by added antioxidant enzymes. *Journal of Food Science, 61*, 726–728.

Lee, S. K., Mei, L., & Decker, E. A. (1997). Influence of sodium chloride on antioxidant enzyme activity and lipid oxidation in frozen ground pork. *Meat Science, 46*, 349–355.

Lundström, K., Enfältdt, A-C., Tornberg, E., & Agerhem, H. (1998). Sensory and technological meat quality in carriers and non-carriers of the RN^- allele in Hampshire crosses and in pure-bred Yorkshire pigs. *Meat Science, 48*, 115–124.

Mei, L., Crum, A. D., & Decker, E. A. (1994). Development of lipid oxidation and inactivation of antioxidant enzymes in cooked pork and beef. *Journal of Food Lipids, 1*, 273–283.

Monahan, F. J., Crackel, R. L., Gray, J. I., Buckley, D. J., & Morrissey, P. A. (1993). Catalysis of lipid oxidation in muscle model systems by haem and inorganic iron. *Meat Science, 34*, 95–106.

Monin, G., & Sellier, P. (1985). Pork of low technological quality with

a normal rate of muscle pH fall in the immediate post-mortem period: the case of the Hampshire breed. *Meat Science, 19*, 49–63.

Ortman, K., & Pehrson, B. (1997). Selenite and selenium yeast as feed supplements for dairy cows. *Journal of Veterinary Medicine, 44*, 373–380.

Pehrson, B. (1985). Selenium-dependent and non-selenium dependent glutathione peroxidase activity in tissues from young bulls. *Zentralblatt für Veterinär Medizine, 32*, 488–491.

Renerre, M., Dumont, F., & Gatellier, P. (1996). Antioxidant enzyme activities in beef in relation to oxidation of lipid and myoglobin. *Meat Science, 43*, 111–121.

Spanier, A. M., McMillin, K. W., & Miller, J. A. (1990). Enzyme activity levels in beef: effect of post mortem aging and endpoint cooking temperature. *Journal of Food Science, 55*, 318–322.

Tinggi, U. (1999). Determination of selenium in meat products by hydride generation atomic absorption spectrophotometry. *Journal of AOAC International, 82*, 364–367.

Thompson, R. H., McMurray, C. H., & Blanchflower, W. J. (1976). The levels of selenium and glutathione peroxidase activity in blood of sheep, cows and pigs. *Research in Veterinary Science, 20*, 229–231.

Ursini, F., Maiorino, M., Brigelius-Flohé, R., Auman, K. D., Roveri, A., Schomburg, D., & Flohé, L. (1995). Diversity of glutathione peroxidases. *Methods in Enzymology, 252*, 38–53.

Yamauchi, K., Yada, K., Ohashi, T., & Pearson, A. M. (1984). The interrelationship between polyunsaturated fatty acids, α-tocopherol and glutathione peroxidase in chicken and porcine skeletal muscles. *Agricultural and Biological Chemistry, 48*, 2831–2832.

Zhang, W. R., Ku, P. K., Miller, E. R., & Ullrey, D. E. (1986). Stability of glutathione peroxidase in swine plasma samples under various storage conditions. *Canadian Journal of Veterinary Research, 50*, 390–392.