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LDH₅ AND MEAT QUALITY IN PIGS

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Introduction

In 1964 Charpentier and Goutefongea focussed the attention on the possible etiological importance of LDH in exudative meat. LDH₅ is the muscle isozyme of lactic dehydrogenase. This oxidoreductase is ubiquitous in vertebrate tissues. It catalyzes the interconversion of pyruvate and lactate. LDH exists in several distinct varieties. With help of electroforesis the existence of five isozymes of LDH could be revealed (Markert, 1963; Zondag, 1964).

LDH₅ is predominant in muscle tissue. Heart tissue is a source of LDH₁. There is evidence that the aerobic state of the tissue involved, is related to the predominance of the isozyme type. Under the anaerobic conditions of striated muscle, LDH₅ is found.

The levels of these tissue enzymes in blood are increased not only in association with tissue damage, but also after many non specific physiological stresses without obvious organic lesions i.e. muscular exercise, acute exposure to cold, severe altitude, hypoxia, vibration, noise and confinement and exposure to acute heat stress (Bedrak, 1965).

Manz (1964) discussed the relation between the exudative meat condition and LDH increase in serum after transport.

It was our aim to evaluate the increase of LDH₅ in blood plasma and its significance for the prae-mortem diagnosis of meat quality aberrations (deviations).

Material and methods

The investigations were conducted with several pigs of the three different Dutch breeds, Pietrain, Large White and Dutch landrace. We used the Boehringer sets for estimation of the total LDH concentration. The plasma was assayed for their content of isozymes by agar-gel electrophoresis. The estimation of LDH₅

in percentage were determined by way of scanning the zymogram. The increase is given in percentage of the total amount LDH. Quantitative application of the plasma sample made it possible to compare between and within animals without estimating at the same time the total LDH.

Investigation

This paper is grouped in two sections.

The first investigation deals with the determination in blood of total LDH and isozymes in two different breeds which have a different sensitivity towards meat quality i.e. exudative meat (I).

The second investigation deals with the effect of stress of transport and muscular exercise on LDH₅ in blood; and the role of adrenalin and anti-stress drugs on the LDH₅ level (II).

I LDH levels in Large White and Pietrain

In a group of 31 Pietrains and 32 Large White pigs which were reared in the same barn blood samples were taken from the vena cava cranialis. The results are compiled in table 1.

Table 1. Mean LDH concentrations in Pietrain and Large White pigs (intern. units)

	number of animals	LDH _{tot.}	LDH ₁	LDH ₂	LDH ₃	LDH ₄	LDH ₅
Large White	32	1061	633	314	97	10	7
Pietrain	31	1740	982	457	124	18	158

With the Wilcoxon test there appeared a significant difference between the breeds as to the total LDH concentration and the different LDH isozymes values $P < 0.01$). The LDH₅ was detected in the blood of 24 Pietrain animals (75%) against 16 (50%) of the Large White Pigs.

In 9 Pietrain pigs a level of more than 10% LDH₅ was found. No Large White pig showed a higher LDH₅ than 3%.

These differences may be due to the higher stress-sensitivity of the Pietrain pigs. In this case the blood sampling can be held responsible for the stress. In general the Pietrain pigs show a higher incidence of meat degeneration (exudative meat).

Therefore it was worth to evaluate further the effect of more specific stresses on the level of LDH₅ and its significance as an indication for meat quality.

II.1. Transport and vasodilation

With pigs of the Pietrain breed the following experiments were conducted. Before and after the transport to the slaughterhouse (\pm 6 km.) blood samples were taken. Five animals were treated with 230 mg isoxsuprine lactate (Duphaspasmin) in order to promote the blood circulation i.e. the aerobic state in the muscles, five animals served as control. The meat quality of the m. long. dorsi and m. psoas major was determined with a visual classification method and the transmission value method (Hart, 1961). With the Fahellpho apparatus the colour of the m. long. dorsi was measured (Schröder et al., 1965).

Table 2 LDH₅ percentage in blood plasma of Pietrain pigs before and after application of isoxsuprine and transport

transport number	LDH ₅ % sample		meat quality				
	1 *	2 **	M. long. dorsi class.	trans.	colour	m. psoas major class.	transmission
76/30	0.2	13.1	3	80	71	2	29
77/31	0.6	6.4	3	84	70	2	49
78/32	1.1	38.9	4	100	80	4	75
79/33	2.8	15.6	4	90	78	2	48
82/36	1.3	3.5	3	89	60	4	65
total	1.2	15.5	3.4	89	72	2.8	53
tr.&treat- ment							
80/34	0.0	9.7	3	73	69	2	38
81/35	2.4	22.0	4	90	79	3	57
83/37	3.4	10.3	1	26	37	1	17
84/38	2.2	9.5	2	63	49	1	35
85/39	1.8	32.7	2	44	64	1	29
total	1.9	16.8	2.4	59	60	1.6	35

* before and ** after transport and treatment

From table 2 one finds that the rise in LDH₅ level is about the same in the two groups.

Though the LDH₅ level is low before slaughter, several pigs show a rather high percentage after transport. The treatment seems to have had no effect on the LDH₅ level. The meat quality however has improved in regard of classification, transmission value and colour.

Class 3 and 4 indicate exudative quality. A lot of the samples (in both groups) can be considered as abnormal. Especially in the control group a bad meat quality is associated with a high LDH₅ percentage.

II.2. Muscular exercise

A number of animals of different breeds were used in orientation experiments. Exercise was forced during five minutes. It was obvious that quantitatively they did not undergo the same stress. Before and after the exercise blood samples were taken. Together with the LDH₅, glucose and lactic acid was determined as an indication of resp. stress-effect and metabolic activity of the muscle. (table 4.).

Table 4 The effect of exercise on blood levels of LDH₅, lactic acid and glucose.

Nr.	Breed	Lactic acid mg%		Glucose mg %		LDH ₅ %		quality scoring m.long.dorsi
		sample 1	sample 2	sample 1	sample 2	sample 1	sample 2	
91	NL	94.5	206.1	108	308	4.0	18.3	3
82	NL	39.8	128.5	89	249	7.5	11.2	4
84	NL	26.6	88.1	99	120	10.4	16.0	4
103	NL	27.5	124.2	98	181	6.6	14.6	2
59	Pie	49.1	166.7	113	191	9.4	28.6	3
15	LW	76.7	183.7	111	235	0.2	1.0	1
35	LW	30.8	98.7	106	195	5.1	17.1	1
39	LW	3.2	193.8	113	221	12.3	23.4	4
mean		43.5	148.6	105	212	7.0	16.3	2.7

During the forced exercise, control animals were confined to a pen not familiar to them, outside the laboratory. In some of them unexpected high rises were found (table 5).

Table 5. Bloodlevels of LDH₅, Lactic acid and glucose without exercise

Nr.	Breed	Lactic acid mg%		Glucose mg%		LDH ₅		quality scoring m.long.dorsi
		sample 1	sample 2	sample 1	sample 2	sample 1	sample 2	
83	NL	18.4	93.8	103	134	3.2	3.9	4
22	NL	14.3	32.7	89	110	1.0	37.8	1
112	NL	25.1	94.6	85	139	1.3	1.0	3
45	Pie	72.3	54.4	98	99	29.2	52.4	2
58	Pie	13.6	72.1	82	94	1.1	3.9	2
16	LW	28.9	55.2	111	171	1.1	1.1	2
20	LW	54.3	45.1	108	120	18.4	24.7	1
40	LW	16.0	65.0	117	155	4.4	7.7	2
mean		30.4	64.1	99	128	7.4	16.7	2.1

The rise in lactic acid and glucose is more outspoken in the exercise group compared to the control group than the rise in LDH₅ percentage.

However a look at the individual figures reveals that 7 out of 8 have a higher LDH₅ level than 10% in the last blood sample. In the control group there are only three. In the exercise group 5 (60%) meat samples were considered as abnormal originating from 7 pigs with more than 10% LDH₅.

In the control group from three pigs with a high LDH₅ level at the last sampling two showed already high LDH₅ percentages in the first sample. These unexpected high LDH₅ levels could be due to stress of the transport to the laboratory.

In the control group a high LDH₅ was not associated with a bad meat quality. Two meat samples (25%) originating from pigs without a high LDH₅ level showed symptoms of meat degeneration.

From the differences between the groups the conclusion might be made that the physiological exercise stress was associated with a rise in LDH₅ and a decrease in meat quality.

II.3. Adrenalin and LDH₅

There is some evidence that catecholamine increases the level of LDH and LDH₅ in the blood (Bedrak, 1967).

The rise in blood glucose during exercise could be due to adrenalin action. So in three experiments the effects of adrenaline was tested.

In the first trial three animals of the Dutch Landrace were injected with 8 mg adrenalin bitartrate.

One and a half hour after application, the second blood sample was taken. The results are given in table 6.

Table 6 Adrenalin bitartrate and LDH₅

	nr.	Lactate mg %		Glucose mg %		LDH ₅ sample	
		sample		sample		sample	
		1	2	1	2	1	2
Control	3	19.9	48.8	85	99	1.5	2.5
	33	12.9	8.2	96	98	0.0	0.0
	24	36.3	23.5	104	103	0.0	0.0
Adrenalin bitartrate	34	50.6	85.5	103	178	1.5	6.5
	4	63.5	96.4	84	195	2.0	10.3
	23	36.5	107.6	96	278	2.0	14.0

As there was an indication that adrenalin increased the LDH₅ in the blood, a second experiment was undertaken with 16 mg adrenalin bitartrate to provoke the LDH₅ increase. Thirty minutes post injectionem some of the animals got in a preshock state.

From one pig 6 blood samples were collected with 5 minutes intervals during 30 minutes. The results were compiled in table 7 and 8.

Table 7 and 8 Effect of adrenalin on LDH₅

Nr.	Breed	Lactate mg %		Glucose mg %		LDH ₅ %	
		sample 1	sample 2	sample 1	sample 2	sample 1	sample 2
50	Pie	151	169	207	331	9.3	23.8
32	LW	91	106	118	284	1.2	3.5
104	NL	26	125	98	331	4.5	6.4

Nr.	Breed	sample	Lactate mg %	Gluc. mg%	LDH ₅
21	NL	1-0 min.	80	132	12.5
		2-5 min.	65	127	13.0
		3-10 min.	55	125	5.5
		4-15 min.	36	122	16.0
		5-20 min.	44	134	9.5
		6-25 min.	134	200	14.5

Nr. 104 was a remarkable case. There were symptoms of severe meat degeneration in the m. long. dorsi and the m. semimembranosus, but the LDH₅% was rather low.

Nr. 21 showed a rather high lactate and glucose and LDH₅ concentrations at the beginning of the blood sampling, indicating that a stress condition already existed.

The effect of adrenalin on the blood glucose level is obvious but no clear effect on the LDH₅ pattern could be found.

One of the reasons might be that the period of sampling (30 minutes) was too short for a marked increase of the LDH₅ level (Bedrak, 1965).

Discussion

The aim of our investigation was to evaluate the significance of the LDH₅ blood level for the early diagnosis of meat quality aberrations. One assumes that an increase of the blood level of tissue enzymes is caused by a higher cellular permeability (Bedrak, 1965; Hess, 1966). Physiological stress as there are transport and muscular exercise, were in our experiments associated with a higher LDH₅ level (table 2 and table 4).

In pigs confined to a strange pen some of them showed high LDH₅ percentages (table 5) without marked rises of lactic acid and blood glucose, which were used resp. as indicators for muscle metabolism and stress. A shift of LDH₅ from the muscle to the blood caused by a higher cellular permeability, could provide evidence about the state of anaerobioses in those muscle (Hess, 1966) (which could lead to exudative meat). A Vasodilatator used as blood circulation stimulator showed no effect on the LDH₅ level (table 2). Adrenalin which plays an important role in increasing enzyme levels showed no result under the used circumstances. The most sensitive breed for exudative meat showed the highest LDH levels (table 1). In the experiments with transport and muscular exercise there was evidence that physiological stress was associated, both with a higher incidence of exudative meat and a shift to a higher LDH₅ percentage. In the vasodilation group (table 2) and the control group of muscular exercise (table 5) some discrepancy existed between meat quality and LDH₅ shift. Meat degeneration without an increase of LDH₅ is quite well possible. To our opinion in normal slaughter pigs a LDH₅ increase is the result of the physiological stress on muscle.

Whether or not this LDH₅ increase is followed by meat degeneration. symptoms depends also on the susceptibility of the pig.

Summary

Several determinations of total LDH and LDH₅ were (in experiments with pigs of three different breeds) performed in order to evaluate the significance of the muscle tissue isozyme LDH₅ for a prae mortem diagnosis of the meat quality. In the first experiment 31 Pietrain pigs were compared with 32 Large White animals. There proved to be a significant difference of the level of LDH and LDH isozymes between the breeds, the Pietrain being higher. No Large White pig had a higher LDH₅ level than 3 %. In experiments with transport and muscular exercise as physiological stress factors, LDH₅ increased in blood. In some control animals without obvious stress, the LDH₅ unexpectedly increased. There was evidence that the physiological stress which causes LDH₅ increases affects also the meat quality.

Zusammenfassung

Es wurde das Isoenzym LDH₅ bestimmt in Blut von Schweinen verschiedener Rassen. Der Anstieg der LDH Aktivitäten ist nach aufsprechenden Untersuchungen u.m. abhängig von der physiologischen, muskulären Anaerobiose (Hess, 1966).

Es wurde untersucht ob der Anstieg der Muskelspezifischen LDH₅ auch voraussagenden Wert hatte über die Fleischqualität. Für Fleischdegeneration anfällige Schweine wie Pietraintiere, zeigten ein höheres Gehalt an LDH und LDH₅ im Blutplasma als Holländische Edelschweine. Keiner der Edelschweine zeigte ein höheres LDH₅-gehalt als 3%. Transport und Muskelarbeit erhöhten die LDH₅ Konzentration.

Man hatte den Eindruck dass Umstände die förderend wirkten auf die Blut-LDH₅, die Fleischqualität ungünstig beeinflussten. Adrenalin und Kreislauf stimulierende Mittel hatten in orientierende Versuche nicht ein deutlich erkennbare Einfluss auf das LDH₅-gehalt.

Résumé

Plusieurs experiments ont été entrepris avec porcs de trois races Néerlandaises pour déterminer la relation entre le taux sanguin de LDH₅ et la qualité de la viande.

On trouva une différence de niveau de LDH et LDH₅ sanguin entre la race Pietrain et la race Large White.

Aucun des Large White avait une concentration de LDH₅ de plus de 3 %. L'état anaérobie du tissu entraîne une augmentation sanguine de LDH (Hess, 1966) de la viande exudative est alors trouvée fréquemment.

L'exercice musculaire ainsi que le transport augmentait le LDH sanguin chez tous les animaux expérimentaux, mais le taux montait également chez les animaux de contrôle.

Il y a des indications que le stress physiologique, causant une augmentation du taux sanguin de LDH₅, influence également la qualité de la viande. En ce qui concerne les expériences d'orientation, il s'est avéré qu'adrenaline et une substance vasodilatatoire (isoxsuprine) n'avait pas d'effet sur LDH₅.

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