GLYCOGEN, LACTIC ACID AND PH IN MEAT (META ANALYSIS)

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Background

Lactic acid has a pK_a-value of 3.86. This means that 90% is dissociated at pH 4.9, and 99% at pH 5.9, i.e. practically totally. Consequently, only ca. $3*10^{-5}$ M lactic acid solution would decrease the pH of distilled water to a value of 5.5. Analyses have shown that in meat the relationship between pH (a logarithmic measure) and the lactic acid content (a linear measure) is practically linear. On the assumption of a linear relationship, the buffering capacity of meat has been calculated from titration curves, to be 50–60 mmol H^+/(kg*pH) over the pH range of 7 to 5.5. Consequently, ca. 80 to 100 moles lactic acid per kilogram lean meat is needed to decrease the pH value from 7.2 of living muscle to 5.5.

In a simplified model glucose (glycogen) is broken down to lactic acid causing the pH to decrease. The protons formed are bound to buffering substances, such as proteins, organic and inorganic phosphates and dipeptides. The buffering capacity of all of these depend on pH. The buffering capacity of proteins is from about 20 mmol H^+/(kg*pH) (pH 7.0) to 35 mmol H^+/(kg*pH) (pH 5.5). The effect of the other substances ranges from ca. 30 mmol H^+/(kg*pH) (pH 6.8) to 15 mmol H^+/(kg*pH) (pH 5.5). At lower pH values, lactic acid becomes increasingly important, as its buffering capacity increases, while the capacity of other components decreases. Proteins, however, have another buffering capacity maximum at pH 4, due to the pK_a of the nonpeptidyl carboxyl group. Consequently, the buffering capacity is different at different pHs (Kivikari, review 1996).

In addition to the buffering capacity, there are difficulties obtaining unbiased determination of pH value of meat, as well as determining the glycogen, its derivates and lactic acid, especially in the small and inhomogenous samples generally used in meat research. The amount of water added during sample preparation and titration, added salt etc. have effects on the pH and also on the buffering capacity. This makes it understandable that the data given in the literature is somewhat variable.

Objectives

The purpose of this study was to analyse the relationships between glycogen and lactic acid contents and pH, based on the data presented in the literature (meta analysis).

Materials and methods

The data used in this meta analysis were collected from the literature relating to porcine *M. longissimus dorsi* (i) pH value, (ii) lactic acid content and (iii) glycogen content at (iv) different times post mortem. We used the value 7.2 (Kylä-Puhju, Ruusunen, Kivikari and Puolanne 2004) for the pH of zero lactate meat and buffering capacity of 52 mmol H^+/(kg*pH) (Kivikari 1996; Puolanne and Kivikari 2000). All data were converted into same units and calculated as follows (the underlinings refer to Table 1.):

The Measured pH value was the pH value that was given in the reference at the given time
The Calculated pH value was obtained as follows: pH calc. = 7.2 – lactic acid (mmol/kg)/52 mmol/(pH*kg)
Difference pH = pH measured – pH calculated
Measured Δ lactic acid LA (mmol/kg) = lactic acid time 2 – lactic acid time 1
Calculated Δ lactic acid LA (mmol/kg) = (glycogen time 1 – glycogen time 2)*2
Difference lactic acid LA (mmol/kg) = measured Δ lactic acid – calculated Δ lactic acid

Results and discussion

The differences between the measured and calculated lactic acid values did not show any consistent pattern, varying between -0.14 and +0.66 (Table 1.). The calculated pH values, however, tended to be lower than the measured.

The data in Table 1 are only a sample of the data available in the literature, and the following discussion is based on a more general analysis of the literature data. It was clearly seen in this study that the determination of pH values as well as lactic acid content and glycogen content are subject to large variation which may sometimes have lead to even biased results. This does not necessarily mean that the analyses have not been carefully done. The measurement of pH value is particularly difficult just after slaughter. Therefore it is recommended to utilise iodoacetate method to stop the glycolytic reactions which would also allow the measurement to be carried out at the room temperature. The sample sizes are sometimes small, from 10 mg (2 mg freeze dried) to several grams. Sometimes blood, fat and visible connective tissue had been carefully removed, but not always.

The intermediates of the glycolytic pathway which are not determined by the normal methods for detemining glycolytic potential may represent less than 10 mmol/kg as lactic acid (Kastenschmidt et al., 1968; Hamm and Fischer, 1980). These do not seem to be the major cause of the variability in the results. It was found that the value for buffering capacity is also very variable. Kivikari (1996) and Puolanne and Kivikari (2000) gave a buffering capacity of 52 mmol H^+/(pH*kg), but in her review Kivikari (1996) had found values from 42 to 64 mmol H^+/(pH*kg). In a previous study the value was 49 mmol H^+/(pH*kg) (Kylä-Puhju, Ruusunen and Puolanne, 2004), and in the current study the best fit was obtained with a value of 62 mmol H^+/(pH*kg). This was done by solving the value of the regression equation y = -0.1927x + 1.3843 at pH 5.5 (Figure 1). When the measured pH value x was 5.50, y was 0.32, meaning that the calculated pH value was 5.18 (5.50 – 0.32 = 5.18). From this it was possible to calculate that if the buffering capacity value was 62 mmol H^+/(pH*kg), the measured pH value and calculated pH value would be the same. The 62 mmol H^+/(pH*kg) gives ΔpH values between measured and calculated values that are on average zero independently of the pH value of the meat at the time of the determinations (Figure 2.). The buffering capacity values, however, should not be directly compared because they are from different sources. This still shows the uncertainty of the methodology involved.

Conclusions

It was concluded that much effort should be put on the analyses of pH values, glycogen contents and lactic acid contents. If they all are determined at the same times, it might be worthwhile to do the simple calculations in order to see, how well the figures fit together as glycolytic potential. It must, however, be pointed out, that during the post mortem reactions the calculations discussed above may be a over-simplification, and also other factors and components may have a marked role. In addition, it is recommended always to use the same control sample within the study to check the variation between the different sets of analyses. The buffering capacity of *M. longissimus dorsi* seems to depend on the method of determination, the values varying between 49-62 mmol H^+/(pH*kg).

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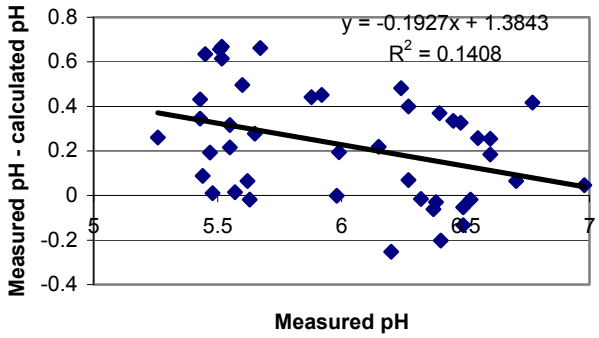

Figure 1. The difference of measured pH and calculated pH related to measured pH. Buffering capacity 52 mmol H^+/(pH*kg).

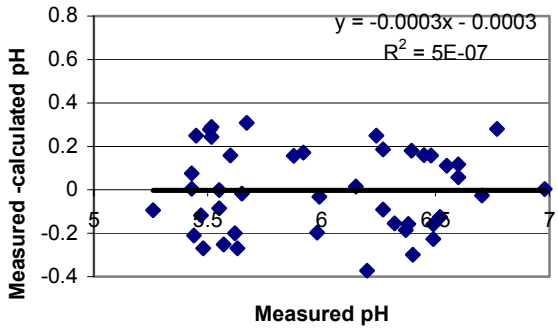

Figure 2. The difference of measured pH and calculated pH related to measured pH. Buffering capacity 62 mmol H^+/(pH*kg).

Table 1. Studies from the literature containing analytical data on pH, lactic acid content and glycogen content of porcine *M. longissimus dorsi* at different times post mortem. Calculations, see Materials and methods.

	pH				Lactic acid			
Reference	**Time**	**Meas.pH**	**Calc. pH**	**Diff.pH**	**Δ timeLA**	**Δ meas.LA**	**Δ calc.LA**	**Diff.LA**
Ahn et al. 1992	45 min	5.65	5.37	0.28				
	2 h	5.55	5.35	0.20	45–120 min	1	14	-13
	4 h	5.46	5.28	0.18	2–4 h	4	2	2
	24 h	5.45	4.80	0.65	4–24 h	25	6	19
							Total	8
D´Souza et al. 1999	40 min	6.48	6.15	0.33	5–40 min	11	24	-13
	24 h	5.55	5.23	0.32	40–-24 h	48	38	10
							Total	3
D`Souza et al. 1998a	45 min	6.4	6.60	-0.20	5–45 min	7	27	-20
	70 min	6.2	6.45	-0.25	45–70 min	8	27	-19
	24 h	5.62	5.55	0.07	70 min–24 h	47	39	8
							Total	-31
D´Souza et al. 1998b	40 min	6.6	6.35	0.25	5–40 min	2	11	-9
	24 h	5.48	5.47	0.01	40 min–24 h	46	50	-4
							Total	-13
Fernandez et al. 2002	40 min	6.52	6.54	-0.02	0–40 min	26	15	11
Hammelman et al. 2003	1 min	6.77	6.36	0.41				
	30 min	6.27	6.19	0.08	1–30 min	8	-5	13
	60 min	5.99	5.80	0.19	30–60 min	21	50	-29
	24 h	5.6	5.09	0.51	60 min–24 h	36	64	-28
							Total	-44
Henckel et al. 2002	24 h	5.60	5.18	0.42	1–15 min	10	6	4
					15–30 min	3	-6	9
					30–45 min	8	6	2
					45–60 min	4	6	-2
					1–3 h	29	30	-1
					3–6 h	12	10	1
					6–24 h	19	17	2
							Total	15
Klont et al. 1994	45 min	6.60	6.42	0.18	45 min–2 h	15	15	0
	18 h	5.57	5.56	0.01	2–4 h	9	17	-8
					4–18 h	20	24	-4
							Total	-12
Lambooij et al. 2004	0 h	6.49	6.54	-0.05				
	1 h	5.98	5.98	0.00	0–1 h	29	33	-4
	4 h	5.63	5.65	-0.02	1–4 h	17	13	4
	24 h	5.44	5.35	0.09	4–24 h	15	12	3
							Total	3
Schäfer et al. 2002	1 min	6.48	6.62	-0.14				
	15 min	6.38	6.43	-0.05	1–15 min	10	14	-4
	30 min	6.33	6.32	0.01	15–30 min	6	2	4
	1 h	6.16	5.95	0.21	30 min–1 h	19	16	3
	2 h	5.92	5.49	0.43	1 h–2 h	24	20	4
	24 h	5.51	4.85	0.66	2 h–24 h	33	30	3
							Total	10

Table 1 cont.

	pH				Lactic acid			
Reference	**Time**	**Meas.pH**	**Calc.pH**	**Diff.pH**	**Δ timeLA**	**Δ diff.LA**	**Δ calc.LA**	**Diff.LA**
Sayre et al. 1963								
Chester	0 h	6.45	6.12	0.33	0–24 h	54	96	-42
White	24 h	5.43	5.00	0.43				
Hampshire	0 h	6.38	6.41	-0.03	0–24 h	73	143	-70
	24 h	5.26	5.00	0.26				
Poland	0 h	6.27	5.87	0.40	0–24 h	45	68	-23
China	24 h	5.43	5.00	0.43				
Tarrant et al.	15 min	6.98	6.93	0.05				
1972	1 h	6.70	6.64	0.06	15 min–1 h	15	12	3
	2 h	6.55	6.29	0.26	1–2 h	18	21	-3
	3 h	6.39	6.02	0.37	2–3 h	14	12	2
	4h	6.24	5.76	0.48	3–4 h	14	28	-14
	5 h	5.88	5.44	0.44	4–5 h	17	26	-9
	6 h	5.67	5.01	0.66	5–6 h	22	14	8
	7 h	5.52	4.85	0.67	6–7 h	8	18	-10
	8 h	5.52	4.90	0.61	7–8 h	-3	6	-9
							Total	-32