AGEING OF BEEF STUDIED BY USING DIFFERENT INSTRUMENTAL TECHNIQUES AND SENSORY TENDERNESS

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Introduction

Beef tenderness is affected by a number of variables such as the age and sex of the animal, pre-slaughter stress, electrical stimulation, rigor temperature and ageing etc. Probably the most crucial of these variables, if chilling during rigor is controlled, is ageing. The aim of this study has therefore been to follow the ageing process at two relevant temperatures, +4 and -1.5°C, respectively. The increase in the sensorially determined tenderness was followed and compared with the results from different instrumental methods that reflect the textural and structural changes going on. The methods used in this study were Warner-Bratzler shear force measurements, myofibrill length measurements, nuclear magnetic resonance spectroscopy (NMR) and viscoelastic measurements.

Materials & Methods

M. longissimus dorsi (LD) muscles from 22 young bulls of the Swedish Lowland breed were cut out three days *post-mortem*, following the usual chilling regime at the abbatoir. Two groups of ultimate-pH, pH_u, were chosen, one with a normal pH_u (<5.6) and one with an increased pH_u (5.8< pH_u <6.0). The two LD muscles from each animal were cut into six pieces, vacuum-packed and stored following six of the 11 possible ageing procedures (3, 7, 14 and 21 days at 4°C or 7, 14, 21, 35, 49, 63 and 77 days at -1.5°C). After ageing, the meat was cut into slices for sensory evaluations (1.5 cm), Warner-Bratzler shear force (4.5 cm), viscoelasticity (cylindrical: ø; 20 mm, height: 7 mm, cut parallel to the muscle fiber direction) NMR (0.25 g cut perpendicular to the muscle fiber direction) and for myofibrillar length measurements (5 g). The meat for the ***sensory analysis*** was fried in a pan (175°C) to a centre temperature of 70°C, and served to the assessors immediately after cooking. The sensory analysis was performed by a trained expert panel of 15 women and men. Tenderness was judged on a nine-point scale (1=very tough, 9=very tender). ***Warner-Bratzler shear force measurements*** were perfomed on an Instron® Universal testing machine equipped with a modified Warner-Bratzler blade with a square opening of 26x21 mm and a thickness of 1.0 mm. The meat was cooked in a water bath at 74°C for 85 min and chilled to room temperature in ice. The maximum shear force for at least 10 pieces (0.7x1.5 cm), sheared across the fibre direction, was registered. ***Myofibrillar length measurements*** were perfomed as described elsewhere (Olsson & Tornberg, 1992). Five grams of meat was homogenised in an omnimixer for 60 s at 11,000 rpm followed by centrifuging at 2°C for 15 min at 1,000 g. The sediment was resuspended in 25 ml of isolation buffer (100 mM KCl, 20 mM K-phosphate, 1 mM EDTA, 1 mM NaN_3, pH 7.0) and diluted 25 times in the same buffer. The myofibrillar length was measured by using light microscopy (Nikon Optihot,) using video images (Sony 3 CCD) and the image analysis program Image Pro Plus 3.0 (Media Cybernetic, USA). ***NMR measurements*** were performed by measuring the transverse relaxation time (T_2) of the water protons on a Bruker Minispec PC/20 operating at a frequency of 20 Mhz. The T_2:s were measured using a CPMG-sequence with a τ-spacing of 150 µs. The signal amplitude was measured every eight echoes, in total 169 data points, 64 acquisitions were accumulated with a repetition time of 10 s. The FID's were fitted by non-linear regression to a two-component model. ***Viscoelastic measurements*** were performed on a StressTech Rheometer (Reologica Instruments AB, Sweden) using the parallel SF20PC measuring system. All measurements were performed in the linear viscoelastic region using an oscillating sinusoidal shear of 1 Hz and an applied stress of 10 Pa. The viscoelastic behavior was monitored as the storage and loss modulus (G´ & G´´) together with the phase angle (δ).

Results & Discussion

The ageing of LD muscles with a normal pH_u, and an increased pH_u at -1.5 and +4°C, as followed by sensorially determined tenderness, Warner-Bratzler shear force, T_{21} and elasticity, G´, is shown in Figure 1. Both the ageing time*** and temperature*** but not the pH_u have a significant effect on the sensorially determined tenderness. This is in contrast to the shear force measurements in which pH_u*** has a strong influence on the shear force values together with the ageing time*** and the temperature**. This difference could be explained by the fact, that in the W-B force measurements, the shearing force always acts perpendicularly to the muscle fiber direction, whereas during

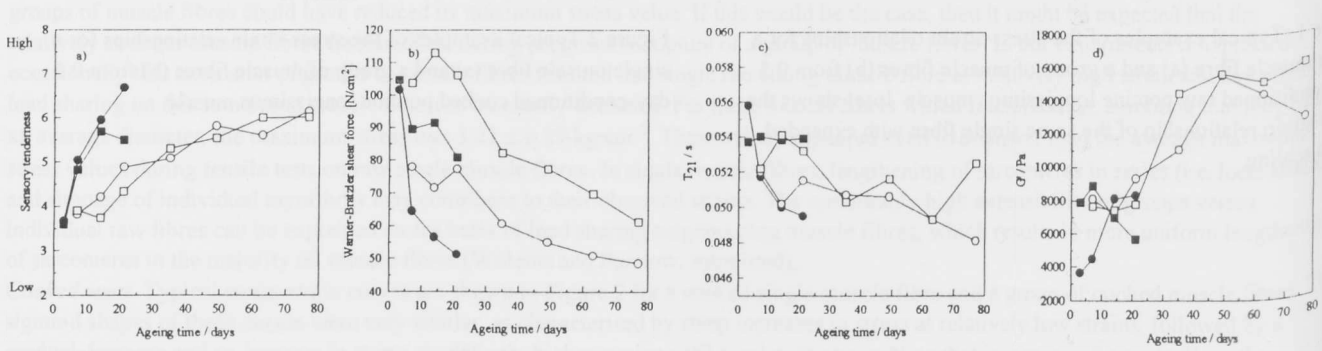

Figure 1. a) Sensorially determined tenderness, b) Warner-Bratzler shear force, c) T_{21} and d) G´ as a function of ageing time, temperature and pH_u for meat with a normal pH_u aged at +4°C; ● and -1.5°C; ○ and for meat with an increased pH_u aged at +4°C; ■ and -1.5°C; □. Every point is a mean of six animals.

chewing the force is more randomly directed against the fibers. The increase in pH_u results in greater muscle shortening, which produces a tougher meat (Olsson *et al.*, 1995). However, the shear force values after three days of ageing are similar for the two pH_u-groups, which does not support this explanation. Futhermore, it can be observed that there is a sharper slope for meat with normal pH_u compared to high pH_u-meat, which suggests that there is a decrease in the proteolytic activity during the ageing of the meat. The decrease is, however, not reflected in longer myofibrils, data not shown, and could therefore be explained by another proteolytic activity, for example the breakdown of costameres (Taylor *et al.*, 1995), which both link adjacent myofibrils and the myofibrils to the sarcolemma.

Both the T_{21}, which is the fastest relaxation time for the water present in the meat, arising from the water inside the myofibrils (Tornberg & Larsson, 1986), and the elastic modulus G´ are significantly influenced by the ageing time ($p<0.05$ and $p<0.001$, respectively), Figure 1. T_{21}, which in its simplest form describes the average distance for a water molecule to a protein surface, decreases with prolonged ageing time. We suggest that this observation reflects a disordering of the myofibrillar structure resulting in a shorter average distance for water to hit a protein surface. Hence a shorter T_{21}. The constant time dependence of T_{21} for meat with an enhanced pH_u aged at 4°C could be explained by a pH-induced myofibrillar swelling. **The elasticity** of the meat increases with prolonged ageing. This observation is, perhaps, unexpected but might be explained by the increase in ruptures obtained in meat during ageing, thereby causing larger numbers of contact points per unit volume and hence increasing the elasticity of the meat.

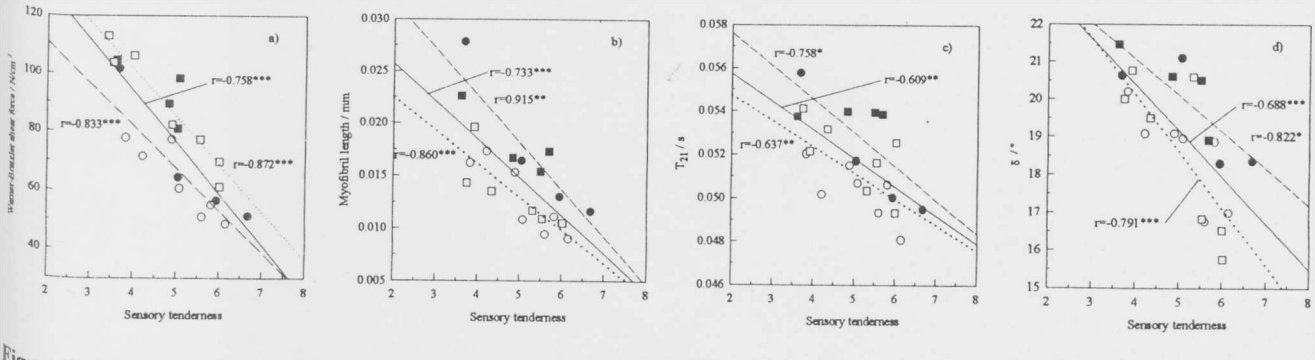

Figure 2. a) Warner-Bratzler shear force b) myofibillar length, c) T_{21} and d) phase angle, δ, as a function of sensory tenderness. Description of symbols as in Figure 1.

If we compare the different instrumental parameters measured in this study with the sensorially determined tenderness we find that there are quite good correlations between the different methods, Figure 2. These correlations are, according to the figures significantly dependent on the pH_u of the meat, or on ageing temperature, or both. Meat with the same sensory tenderness but with an enhanced pH_u has a significantly higher shear force*** than meat with normal pH_u. Again, this can be explained by the differences in the directions of the shearing forces against the muscle fiber directions for the two methods. Warner-Bratzler shear force measurments seem, therefore, to be more discriminating than sensory analyses when measuring the toughness of beef, especially if the variance is due to an increased pH_u. Meat aged at -1.5°C needs significantly shorter*** myofibrils than meat aged at 4°C to achieve the same sensory tenderness, independent of the pH_u. This indicates that there might be two different ageing processes at the two temperatures studied. The relationship between sensory tenderness and T_{21} and phase angle, respectively, is not as good as it was for the sensory tenderness related to Warner-Bratzler shear force and myofibril length. However, both pH_u* and ageing temperature** have a significant effect on the correlation of T_{21} to tenderness, whereas only ageing temperature** significantly affects the correlation between the phase angle and the sensory tenderness. In spite of the lower linear correlations there is a significant relationship between T_{21} and the sensorially determined tenderness. A faster relaxation time is linked to a higher degree of tenderness. This might structurally be explained by the increase in unordered structure on the myofibrilar level that is obtained during ageing. The water molecules cannot diffuse as far in an unordered myofibrill before it meets a protein surface and exchanges the excited protons for relaxed protein protons. Similar to the T_{21} - sensory tenderness behaviour, the phase angle decreases with increasing sensory tenderness. This means that the meat becomes more elastic when tenderness increases. This is in correspondance with the results obtained in the heat treatment of meat up to 65°C (Tornberg & Persson, 1988; Josell & Tornberg, 1994) where increased elasticity gave rise to enhanced tenderness.

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