

THE RELATIONSHIP BETWEEN EARLY *POSTMORTEM* pH AND THE TENDERISATION OF BEEF MUSCLES.

G.R. O'HALLORAN¹, D.J. TROY¹ & D.J. BUCKLEY².

¹Teagasc, The National Food Centre, Dunsinea, Castleknock, Dublin 15, Ireland.

²Department of Food Technology, University College Cork, Ireland.

KEYWORDS Tenderness, beef, pH, glycolysis, proteolysis, calpains, cathepsins.

BACKGROUND

The properties of meat that are of most interest to the consumer, i.e. tenderness, are strongly affected by perimortal treatments: the physical conditions that are imposed on the animal in the last few days of life and on the carcass in the first few hours *postmortem*. The effects of these treatments are exerted through anaerobic glycolysis, the *postmortem* breakdown of glycogen to lactic acid. According to Tarrant *et al.* (1977), considerable variations in glycolysis may occur between animals and also between muscles within a beef carcass. The rate of glycolysis is a major determinant of meat tenderness through its effect on pH and hence its influence on proteolytic enzyme activity (Marsh, 1993). There is general agreement that tenderisation during the storage of meat occurs by proteolysis of myofibrillar and cytoskeletal proteins. Two endogenous proteolytic systems have been implicated. Neutral calpains degrade myofibrillar and cytoskeletal proteins (Dayton *et al.*, 1981) and mimic *postmortem* histological changes in myofibrils (Penny *et al.*, 1984). Lysosomal acidic cathepsins also degrade myofibrils and isolated proteins (Ouali *et al.*, 1987). The relative contributions of these enzymes to tenderisation is still only speculative and a co-operative mechanism involving both groups has been suggested (Alarcon-Rojo, 1990). The rate of *postmortem* glycolysis is influenced by a number of factors including temperature and electrical stimulation (ES). Previous studies have examined the effect of the rate of glycolysis in carcasses by altering the chilling rates (Marsh *et al.*, 1987), applying electrical current (Pike *et al.*, 1983), or by varying the degree of fat cover (Shackelford *et al.*, 1994).

OBJECTIVE

Ultimate pH and its important effects on tenderness has previously been studied (O'Halloran *et al.*, 1994). The present study was designed in order to evaluate the influence of early *postmortem* pH on the tenderness of beef *m. longissimus dorsi* (LD), as measured physically, mechanically and biochemically. Carcasses with naturally different glycolytic rates *postmortem* were chosen, without the confounding effects of temperature (different chilling regimes, animal size and grade) and ES treatments.

METHODS

127 Hereford X heifers (similar grade, size and age) were slaughtered in a local beef processing plant. The pH of LD muscles were taken using a portable pH meter (Orion) and electrode (Amagross) every hour for 8 hours and again at 24 hours *postmortem*. The temperature of three carcasses and ambient temperature was measured every half hour with a data logger (Grant squirrel). 24 LD muscles were grouped according to their pH fall *postmortem*; slow, intermediate and fast (Fig.1). The muscles were excised 24 hours post-slaughter and were sampled and extracted for SDS-PAGE. The muscles were then stored at 4°C and sampled at 2, 7 and 14 days *postmortem*.

Intramuscular fat and moisture was determined using a CEM analysis system (Bostian *et al.*, 1985). Sarcomere lengths were measured by diffraction of a laser beam according to the procedure described by Cross *et al.* (1980). Sensory analysis was performed by an eight-membered in-house trained taste panel on steaks grilled to an internal temperature of 70°C, according to American Meat Science Association Guidelines (1978). Panelists ranked the steaks for tenderness, juiciness and overall acceptability using a hedonic scale of 1-8. Warner Bratzler Shear force values were taken on 1.25cm diameter cores, cut from 2.5cm steaks parallel to the fibres and cooked to an internal temperature of 60°C and 80°C, using an Instron Universal testing machine. The percentage cook loss was determined at 60°C and 80°C. Myofibrils were extracted and SDS-PAGE (sodium dodecyl sulphate polyacrylamide gel electrophoresis) was performed according to a procedure by Greaser (1993).

The results were statistically analysed using a one way classification design (Genstat. Numerical Algorithms Group Ltd. Oxford).

RESULTS AND DISCUSSION

Glycolytic rate in the few hours following slaughter is a major determinant of quality, and is known to vary widely and unaccountably; the nature of the neurological damage done at the time of stunning is a possible cause of this variability (Kauffman *et al.*, 1986). In this study a significant difference in pH fall was found between the slow, intermediate and fast glycolysing groups (Fig.1). Fast glycolysing muscle was rated significantly more tender and acceptable by trained sensory analysts. The slow group was considered significantly tougher ($p < 0.001$) at 2 and 7 days *postmortem*. The fast group had a significantly lower shear force ($p < 0.001$) at each stage of ageing. The slow group had the highest shear force. In texture assessment the fast glycolysing muscle cooked to 60°C and 80°C was rated significantly more tender ($p < 0.01$). No significant difference was found between the groups in cook loss at either temperature, juiciness as assessed by panelists, or intramuscular moisture content, therefore perceived juiciness did not contribute significantly to the differences in tenderness. Sarcomere lengths were shorter in the slow group at 2 days *postmortem*. Salm *et al.* (1983) found that unstimulated muscles had shorter sarcomeres than stimulated muscles, in which the pH fell rapidly *postmortem*. No significant difference was found at 7 and 14 days *postmortem*.

SDS-PAGE patterns indicated a more rapid and greater degree of proteolysis in the fast glycolysing muscle as evidenced by the

appearance of the 30kDa band even after 1 day *postmortem*. A lower degree of proteolytic action, as shown by the presence of the cytoskeletal protein desmin at 7 days *postmortem*, was observed in the slow glycolysing muscle. Degradation of desmin during ageing has been shown previously (Penny *et al.*, 1984). Nebulin, another important cytoskeletal protein, was degraded in the fast glycolysing muscle by 1 day *postmortem* in both muscles. It is known that nebulin is highly prone to proteolysis by endogenous muscle proteases such as cathepsin L (Penny *et al.*, 1984). It has been suggested that degradation of nebulin could trigger subsequent alterations in the myofibril during meat ageing (Fritz and Greaser, 1991). Creatine kinase appearance was stronger in fast glycolysing muscle. This is a sarcoplasmic protein, which may have denatured and precipitated onto the surface of the myofibrils due to low pH and high temperature conditions. According to Lawrie (1985), the denaturation and inactivation of creatine kinase would indicate a faster rate of *postmortem* glycolysis by causing an excess of ADP and inorganic phosphate in the muscle.

Marsh *et al.* (1987) used different forms of ES and variable cooling rates to produce wide ranges of early *postmortem* glycolytic rates and concluded that tenderness was highest when glycolysis proceeded at a rate which produced a pH of 6.1 at 3 hours *postmortem*. Martin *et al.* (1983) concluded, with consistent positive correlations between rate of pH decline and tenderness, that faster glycolysing muscles, whether stimulated or not, were the most tender. The findings agree with the belief that tenderness is promoted by the rapid attainment of a low pH, high temperature condition - a combination that supposedly releases and/or activates proteolytic enzymes of the lysosome (Dutson *et al.*, 1980). According to Moeller *et al.* (1977), low pH and high temperature conditions in muscle early *postmortem* disrupt the lysosomal membrane freeing lysosomal enzymes into the cytoplasm.

CONCLUSION

The results show that early *postmortem* pH plays an important role in proteolysis and tenderisation. This is thought to be through the effect of pH on the endogenous enzyme systems, thereby optimising their action in the fast glycolysing muscle. This may explain the wide variability in the tenderness of beef within and between similar animals.

REFERENCES

- Alarcon-Rojas, A.D. (1990). Changes in texture, structure and enzyme levels in conditioning of meat and their relationship with tenderisation. PhD Thesis, University of Bristol.
- AMSA, (1978). *Guidelines for cookery and sensory evaluation of meat*.
- Bostian, M.L., Fish, D.L. and Webb, N.B. (1985). *J.Assoc. of Official Analytical Chemists*, **5**,876.
- Cross, H.R., West, R.L. and Dutson, T.R. (1980). *Meat Sci.*, **5**,261.
- Dayton, W.R., Lepley, R.S.A. and Schollmeyer, J.V. (1981). Proc. 34th Annual Recip. Meat Conf. Oregon State University, OR, p.17.
- Dutson, T.R., Smith, G.C. and Carpenter, Z.L. (1980). *J.Fd.Sci.*, **45**:1097.
- Fritz, J.D. and Greaser, M.L. (1991). *J.Fd.Sci.*, **56**:607.
- Greaser, M.L. (1993). *Electrophoretic methods for analysis of muscle proteins*. Personal communication.
- Kauffman, R.G. and Marsh, B.B. (1986). In: *The Science of Meat and Meat Products*. 3rd Ed. Food and Nutrition Press, USA.
- Lawrie, R.A. (1985). In: *Meat Science*. 4th Ed. Pergamon Press, Oxford.
- Marsh, B.B., Ringkob, T.P., Russell, R.L. Swartz, D.R. and Pagel, L.A. (1987). *Meat Sci.*, **21**,241.
- Marsh, B.B. (1993). ICoMST Review paper, Session 3.
- Martin, A.H., Murray, A.C., Jeremiah, L.E. and Dutson, P.J. (1983). *J.Anim.Sci.*, **57**,1456.
- Moeller, P.W., Fields, P.A., Dutson, T.R., Landmann, W.A. and Carpenter, Z.L. (1977). *J.Fd. Sci.*, **42**:510.
- O'Halloran, G.R., Troy, D.J. and Buckley, D.J. Proc. 40th ICoMST, The Hague, The Netherlands. S-IVB.16.
- Ouali, A., Garrel, N., Obled, A., Deval, C., Valin, C. and Penny, I.F. (1987). *Meat Sci.*, **19**,83.
- Pike, M.M., Rinkbob, T.P., Beekman, D.D., Koh, Y.O. and Gerthoffer, W.T. (1993). *Meat Sci.*, **34**,13.
- Penny, I.F., Etherington, D.J., Reeves, J.L. and Taylor, M.A.J. (1984). Proc.30th Eur. Meet. Meat Res. Workers, Bristol. **3**,19,133.
- Salm, C.P., Forrest, J.C., Aberle, E.D., Mills, E.W., Snyder, A.C. and Judge, M.D. (1983). *Meat Sci.*, **8**,163.
- Shackelford, S.D., Koohmaraie, M., Savell, J.W. *Meat Sci.*, **37**, 195.
- Tarrant, P.V. and Mothersill, C. (1977). *J.Sci.Fd.Agric.*, **28**,739.

