## Meat tenderness: from observation to prediction How did we get here? Where are we going?

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Some time ago we reached the point where we would like to be able to predict from raw meat how tough or tender the cooked meat will be, particularly beef since pork is rarely found to be tough unless it has been overcooked. There has been sufficient knowledge gathered over the years concerning factors affecting tenderness, the properties of meat/muscle components, and ways of measuring variations in those properties, that reliable prediction is almost within our grasp. But how did we get here, and what are some of the new technologies available that could help us reach the goal?

We got here slowly, through decades of observations, followed by exploring, recording, and analyzing cause and effect, while all along applying the most current developments in technology to either advance or explore detailed knowledge of various physical and chemical properties of the primary contributors to toughness in meat. In a way, this could be thought of as working while looking back over our shoulders. That is, looking at what had already occurred in order to determine relationships and their relative strengths, this is essential to gain understanding. A typical progression can be traced through the following work on tenderness (see reference list for titles): Hiner & Hankins (1950), Wierbicki et al. (1956), Pearson (1963), Bouton et al. (1973), Dutson et al. (1976). Our knowledge has expanded sufficiently that now we can use it to try to look forward. That is, based on these now-established relationships, attempt to predict what will happen under certain sets of conditions. This is part of the natural progression in the collection and application of knowledge. In meat research it is an approach that started over 20 years ago and which has grown by leaps and bounds, particularly over the past decade. Some of the early tenderness predictions were attempted using mechanical shear force measurements on raw meat to predict the tenderness of cooked meat (Shorthose et al., 1988), then biochemical and histological traits were used (Whipple et al., 1990), pH (Purchas, 1994), colour in general (Wulf et al., 1997) and resistance of raw intact muscle to the controlled insertion of a probe (George et al., 1997). In the early 1990's, as the interactions of light with biological tissues were becoming better known, the response of raw meat to various wavelengths of light were reported (Mitsumoto et al, 1991; Hildrum et al., 1995; Swatland, 1996), and since 2000 there have been an increasing number of papers yearly.

In early research, tenderness was almost always evaluated after the full conversion of muscle to meat was complete, and in beef, often after an additional ageing period was applied to take advantage of the tenderizing effects of proteolysis. In this way, the accumulated effects of genetic makeup, environment and feeding, slaughter,

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cooling and cooking method were included in the final evaluations. Scientifically, if one or more of these influences can be kept constant, it (or they) can essentially be disregarded in the final evaluation, leaving freedom to isolate and study the other influences. That has been done, and has led us to the knowledge that breed, gender, age, growth conditions (including hormone implants) all affect the primary components within muscle contributing to tenderness. These components are collagen (types and the proportions present, degree of permanent cross-linking – increased permanent cross-links is negatively related to tenderness), fibers (type and size), and amount of marbling. Slaughter and cooling conditions have been found to affect the contraction state of fibers (sarcomere length) at rigor, chemical processes such as the rate of pH drop and therefore the water holding capacity of the meat (which is also affected by pre-slaughter handling), and the rate at which calcium-dependent protein breakdown occurs (calpain/calpastatin system) leading up to and during the ageing process. The degree and type of cooking can also affect tenderness.

Apart from the influence of aging (manner, packaging, and length of time) and the impact of cooking on tenderness, which is almost entirely in the consumer's hands, almost everything that affects tenderness has done so by the time a beef carcass is graded so this appears to be a logical point at which to attempt a prediction. Predictions can be made using measurements directly from one or more of the primary muscle components known to contribute to tenderness, from the effects the state of these components result in, or from a combination of both.

An example of a direct measurement on a strongly contributing primary component is assessment of the amount of connective tissue and the number of permanent crosslinks present, through the use of light. Since one of the properties of connective tissue crosslinks is that they fluorescence in the visible light range when exposed to long wave ultraviolet light (shorter wavelengths than most human eyes can see), information on the frequency and intensity of fluorescence from a probing pathway perpendicular to the known connective tissue orientation in a muscle could be used as an indicator of tenderness or toughness. This has met with limited success in the past (Swatland and Findlay, 1997; Swatland, 2000), possibly partially contributed to by the difficulty of distinguishing connective tissue from fat-stored Vitamin A which is excited by and fluoresces at wavelengths very close to those of connective tissue. The increased ability of some of the newest equipment to isolate neighboring wavelengths could make revisiting the contribution of connective tissue by way of its fluorescence again worth pursuing.

Examples of effects due to the state of primary components are the time to rigor (measured on the carcass in the past with various types of rigorometers), pH, and sarcomere length. These are all inter-related, therefore measurement of one gives information on the others. A number of approaches to measuring these on the carcass have been made, with varying success. Rigorometers and tenderometers have been used to measure first the speed with which a muscle goes into rigor (increased speed -> increase toughness), and then how much flexibility the muscle has after rigor has been reached (increased flexibility -> increased tenderness) (Swatland, 1995). When post-rigor muscle pH is lower than normal or has dropped more quickly than normal, meat tends to be tough because the water holding capacity has dropped. Sarcomere length is commonly measured on a sample of meat removed

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from the carcass, put into suspension, and viewed under a microscope with polarized light. Longer sarcomeres are associated with more tender meat. Soon it may be possible to use ultraviolet light to take advantage of both the autofluorescence and anisotropic nature of the tryptophan in meat proteins to measure sarcomere length in place, although to date it has only been possible to isolate cold-shortened meat (Luc et al., 2008) and detect a certain degree of ageing (Clerjon et al, 2011).

An example of a combination of primary component and the effect created due to the state of the component is the surface appearance and colour of a steak. The surface appearance and texture is contributed to by the type and diameter of fibers, how much water they hold, and the architecture of the connective tissue. These and the meat colour are associated with the pH. Viewed under raking light (light hitting the surface at a very shallow angle), the surface may appear very smooth and hard, soft and spongy, or covered in bumps. Using light reflectance properties, the information held in this texture has been successfully analysed with wavelet transformation of images to predict a number of important eating qualities of meat, although tenderness has been more difficult to predict well than flavour or juiciness has (Jackman et al., 2009).

Near infra-red (NIR) light has become one particular light-tool of interest to meat researchers. The light that humans can see runs from wavelengths about 400 nm long that look violet, to wavelengths about 700 nm long that look deep red. As light waves increase further in length, the human eye can no longer see them, although our skin can begin to feel the heat of them. Light is considered to be NIR if it falls between approximately 700 nm or 780 nm and 3000 to 5000 nm. As waves increase in length they are called mid-infrared, then long-infrared (the region in which thermal imaging work is conducted), and finally, by the time they reach 30,000 to 50,000 nm they are called far infra-red. The ranges and names are somewhat variable depending on the division scheme that is being used. When light contacts an object some of the light is reflected, some is absorbed, and in some cases some is transmitted right through the object. If this is in the visible spectrum, the reflected and transmitted light are the parts that we can see and they tell us the colour, surface characteristics, and translucency of the object. The absorbed light affects the molecular structure of the object and is a contributor, for example, to increased metabolic activity in plant cells, or the rate at which metmyoglobin forms from oxymyoglobin in a raw steak. We instinctively use this information from visible light to identify everything we see around us as well as the state it is in. Although we cannot see reflections or transmissions of NIR wavelengths, we can measure them. We can also measure the molecule-unique responses of absorbed NIR light. As in the visible spectrum, it is the uniqueness of the reflectance and absorbance responses which enable us to make very accurate identifications.

NIR reflectance has been used to identify the character of meat at least since the early 1980's. Mitsumoto et al. (1991) was one of the first reports of NIR being used in relation to tenderness. By 1994 (Hildrum et al.) it was being used for tenderness prediction from intact beef samples in the lab. Since then, more accurate equipment has been pressed into service as technology has advanced, methods have been

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undergoing refinement, ways of using sensitive equipment on intact carcasses in the harsh kill floor and cooler environments are being explored, and non-contact methods of recording readings (eg. hyperspectral imaging) are being tried (Naganathan et al., 2008). The continued interest and improvements in this method of tenderness prediction bodes well for its eventual success.

With the advances in understanding of light beyond the ends of the visible spectrum, the increased sensitivity and decreased cost of related equipment, and ever-increasing computing power available, it has become increasingly possible (and easy) to 'read' the state of meat at the time of grading. Meshing that knowledge with the cause-and-effect knowledge about meat tenderness that we already have and that we continue to discover, makes predicting tenderness with reasonable accuracy ever more likely.

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