Developing new technology for measuring camelid meat quality

Howard J. Swatland*

University of Guelph, Guelph, Ontario N1G 2W1, Canada

Abstract

Objective measurements of meat yield and quality are useful in improving meat production scientifically and grading meat products commercially. Rapid, nondestructive, on-line methods are based on the biophysical properties of meat. This paper considers how on-line methods might be adapted for camelid meat. A survey of biophysical properties of camelid tissues shows concepts developed for meats such as beef, pork and chicken are equally applicable to camelid meat. Polarizedlight interferometry revealed how pH affected myofibrillar refraction in camelid muscle fibres, with implications in numerous aspects of meat spectrophotometry. The fluorescence of camelid connective tissues was strong, and similar in emission spectrum to that of beef. The paper concludes with suggestions for future research.

Keywords: Meat quality, polarized light, reflectance, fluorescence

* 33 Robinson Avenue, Guelph, Ontario N1H2Y8, Canada. Tel: 1-519-821-7513. Email: swatland@uoguelph.ca

Introduction

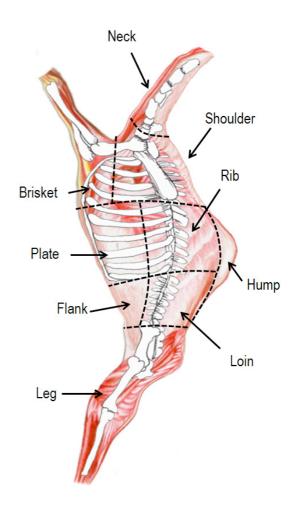
Subjective evaluation is used widely for grading the quality of many types of meat, but only works effectively where there is vertical integration from source to consumer. A lack of objective criteria for meat quality hinders the feed-back of information to breeders and feeders, as well as the feed-forward of information retailers to and consumers. The challenge to meat scientists and technologists is to develop rapid, non-destructive and microbiologically safe methods to measure meat quality objectively, and to demonstrate the commercial

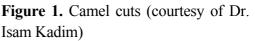
advantage to be gained. With this goal in mind, the main features of camelid meat are reviewed and preliminary data are presented to show aspects of camelid meat quality that might be measured on-line using existing methods (Swatland, 1995).

Camelid meat

Camelus dromedarius, the Arabian camel or dromedary, has one hump, long slender limbs, runs swiftly and may be used for racing. *Camelus bactrianus*, the Bactrian camel of the cold deserts of Asia, has two humps and thick limbs. Camel meat tastes rather like beef and has a similar nutrient value, although usually lower in fat content (Kadim et al., 2008a) and vitamin E (Soltanizadeh et al., 2010). Prime meat from young camels may be cooked rapidly with dry heat, while meat from the extremities of young animals and all the meat from older animals requires cooking with moist heat. Thus, both the toughness and fat content of camel meat increase with age (Kadim and Mahgoub, 2008). In a zoological context, camelids are intermediate between pigs (which have upper incisors) and cattle and sheep (which have a horny pad in place of upper incisors). Camels have vestigial upper incisors, plus well developed canine teeth which may be tusk-like (canine teeth are present in pigs but not in cattle and sheep).

The shape of a camel carcass (Figure 1) differs radically from the shape of a beef, lamb or pork carcass. Apart from the obvious shape of the dorsal hump, the most notable feature is the restriction of hind-limb muscles near the pelvis: they do not overlap with the abdominal muscles. Thus, regardless of how the carcass is suspended, there is an indentation ventral to the ilium (i.e., between the flank and leg cuts shown in Figure 1). This is because the camel has long limbs capable of considerable rotation relative to the vertebral axis. In a sitting camel, the distal end of the femur projects downwards towards the ground. Whereas, in sitting cattle and sheep, the distal end of the femur projects upwards. The camel also has a broad cutaneous pad on each foot instead of two hooves. Camels are ruminants with four stomach compartments, but the omasum is indistinct, not hard and round as in cattle (Clutton-Brock, 1987; Elgasim and Alkanhal, 1992).





The llama (*Lama glama*) is multipurpose domesticated camelid of the Andes. Most meat is used domestically, although in Bolivia and Peru there are commercial markets. Llama meat is similar in taste to mutton and has technological characteristics similar to other meats (Salva et al., 2009). A llama produces about 12 to 15 kg of charqui (Calle Escobar, 1984; Iniguez et al., 1998). Charqui is an intermediate moisture (45%) meat product with a high (15%) sodium chloride content. Typically it has some protein denaturation in the A-band and M-line, together with empty fluid channels created during dehydration. For alpaca chargui, the meat is cut into slices 0.5 to 1 cm thick, treated with salt, and then soaked in brine for two or three days. Two to three weeks open air drying in the Andes resembles freezedrying. Final drying is done under a roof (Calle Escobar, 1984; Biscontini et al., 1996). The alpaca, Lama pacos, is famous for its long, fine wool. The meat is consumed domestically when only a few animals are kept, but the surplus from larger ranches may be used for charqui. In Australia, alpaca meat is sold as La Viandé (Davison, 2012).

Technology for meat quality assessment

Table 1 gives a summary of the methods available to assess meat quality on-line (Swatland, 1995). Carcass quality usually involves a measure of the fatness of the carcass. This is included at the top of the table. Meat yield is important and the technology overlaps with meat quality and with fat quality.

Physical properties of camelid meat

The methods in Table 1 are based on the basic biophysical properties of common meats such as beef and pork. These methods may be applicable to camelid meat, but research is required to prove this. As a preliminary study, results are presented here from a small number of previously frozen samples of Australian camel meat purchased in Canada. Future studies are required to characterize biological variation in fresh meat from known sources. The preliminary results reported here merely show what measurements are possible.

Muscle reflectance

Reflectance from meat is far more complex than is realized by routine researchers using а commercial colorimeter on a meat surface to record its chromaticity coordinates. First we need to know the emission spectrum of the illuminator - it may look like bright, white light to the casual observer, but every light source is different, largely depending on its temperature. Some light is reflected directly from the meat surface without entering the meat (specular or mirrorlike reflectance following Fresnel Specular reflectance is equations). and polarized, may be partly extinguished by rotating a polarizer (as Polaroid sunglasses) in to the appropriate angle. Thus, a polarizer should be used in any system quantify flecks of attempting to marbling far using video image analysis, otherwise it is difficult to separate the fat flecks from the bright flecks of specular reflectance (which are a function of surface irregularity). Light entering the meat is scattered. Some of it scatters back to the meat surface to appear as diffuse or Lambertian reflectance. Lambertian

Basis	Methods	Prediction	
Subcutaneous fat depth and muscle cross sectional area (usually <i>Longissimus thoracis</i>)	Optical probes using diodes, or ultrasonics, or video image analysis (VIA) of cut surfaces	Meat yield. Assuming bone content is constant (which is not always true), subtract an estimate of fat content from total mass and the remainder is an estimate of the meat content.	
Acidity, pH	Glass (calomel half-cell) or ion-sensitive field-effect transistor (ISFET) electrode	sitive field-effect softness	
Electrical impedance	2 or 4 electrodes, conductivity, capacitance, phase anglePaleness-darkness, fluid exudat softness		
Muscle internal reflectance	Fibre-optic spectrophotometry	Myoglobin concentration, paleness- darkness	
Fat internal reflectance	Fibre-optic spectrophotometry	Carotene yellowness, short-chain triglyceride translucency	
Connective tissue	Subcutaneous fat -depth probe adapted for UV fluorescence	Amount and distribution of collagen and elastin, and pyridinoline cross- linking of collagen	
Rheology	Electromechanical probes using compression or rotation, and elastic deformation detected ultrasonically	Toughness	
Surface appearance	Video image analysis	Carcass shape (muscularity), rib-eye area and marbling, subcutaneous fat colour	
Near infrared reflectance	Fibre-optic and surface reflectometers	d surface Triglyceride content, collagen content	

Table 1	. Summary	of existing	on-line methods.
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reflectance appears similar at all angles, whereas specular reflectance, like a mirror, has a strong angular effect. Fortunately, randomization of numerous, small reflective surface on a typical meat sample tends to obscure angular effects. Far more important for meat reflectance is the intrinsic anisotropy of meat which makes meat appear dark when muscle fibres are cut perpendicularly and pale when they are cut longitudinally (Elliott, 1967).

Meat is composed of microscopic muscle fibres. Their size depends on

the size to which an animal has grown, but typically they are about 0.1 mm in diameter and hundreds of millimetres in length. Muscle fibres conduct light by a series of total internal reflections, just like optical fibres, because their myofibrillar cores have a higher refractive index than the surrounding intercellular fluid. So, if muscle fibres are cut perpendicularly to the measured surface, they will conduct light deep into the meat and the meat will appear But if the muscle fibres are dark. parallel to the measured surface they will scatter rather than conduct light and the meat will appear pale. Measuring meat reflectance with an uncontrolled angle of muscle fibres to the measured surface may be a noticeable source of error, as may bulging of a meat surface into the measuring aperture of the apparatus. To make measurements which are repeatable by other researchers, the optical geometry of the apparatus must be known, particularly the angles of the illuminator and the photometer relative to the sample.

How is light scattered in meat? At extremely low pH values, sarcoplasmic proteins may be precipitated between myofibrils to create the extremely high scattering observed in severely pale, soft, exudative (PSE) meat. Kadim et al. (2008a) found Arabian camel meat (*longissimus thoracis*) to have a mean pH of 5.89, with a range from 5.56 to 6.61. Rates of post-mortem glycolysis may be relatively slow in camel meat (Soltanizadeh et al., 2008). Low ultimate pH values been reported in some camel muscles (Gheisari et al., 2009) and in fermented products (El Malti and Amarouch, 2009). Postmortem electrical stimulation of camel meat also may lower pH levels (Kadim et al., 2009), but sarcoplasmic proteins are unlikely to be a noticeable source of light scattering in this pH range, thus leaving a clear optical pathway between the myofibrils. Myoglobin is dissolved in this clear pathway, creating the redness of camel meat by strongly absorbing violet and green light before it can escape from the meat surface as diffuse reflectance (Figure 2). The optimum optical pathlength through camel meat has not vet been systematically investigated but this has also been neglected for other species of meat animals.

The features seen in Figure 2 are similar to those of beef, and indicate strong absorbance by haemoproteins in the Soret band (420 nm) and a secondary absorbance band with a dimple at 560 nm. The presence of the dimple shows oxidation of myoglobin to metmyoglobin, as would be expected in a frozen sample slowly thawed. Metmyoglobin formation may be for responsible the subjective appearance of camel meat being described as from raspberry red to dark brown (Kadim et al.. 2008b).

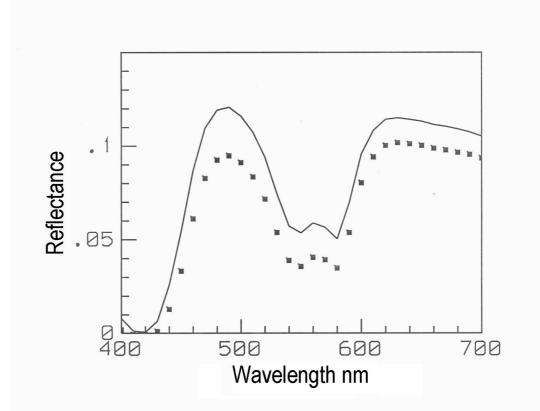


Figure 2. Fibre-optic reflectance of camel meat (*longissimus thoracis*) showing mean (solid line) and standard deviation (n = 5) subtracted from mean (solid squares).

Myofibrillar refraction

Although camelid meat has not yet been widely reported as suffering from pH-related problems such as PSE and dark, firm, dry (DFD) meat, the basic sources of light scattering in camelid meat are still important because they underlie every aspect of meat reflectance and colour. For example, reflectance (especially near infrared) is becoming a popular method in attempts to predict meat toughness (Monin, 1998), and toughness is important, especially in meat from older camels (Kadim et al., 2008b). Without a scientific understanding of light scattering in meat, the best that can be achieved is an empirical correlation -

highly dependent on the apparatus and a particular set of samples, and hence, not easily transferable or repeatable for on-line use.

The only source of light scattering in which verified meat has been experimentally is myofibrillar refraction. Other sources of scattering are possible, such as reflectance from cell membranes forming refractive index boundaries with intra- and extracellular fluids, but they require verification. Beef, pork and chicken all exhibit myofibrillar refraction (Swatland, 2008), and the preliminary data reported here show the existence of myofibrillar refraction in camelid meat. To explain how refraction affects reflectance, consider the example of white paint. White paint contains refractive granules, and very white, expensive paints have granules with the highest refractive index (Williamson and Cummins, 1983). Refractive inclusions in paint cause light scattering to return light to the observer, making the paint appear white when white light. The illuminated by dominant refractive components of meat are the contractile myofibrils almost filling every muscle fibre (Figure 3). Myofibrils are sensitive to pH. When the pH is relatively high, as in living muscle and in meat with minimal post-mortem glycolysis, negative electrostatic repulsion between myofilaments keeps the

myofilaments relatively far apart separated by water. When the pH decreases in muscle post-mortem, the myofilaments move closer together with two important consequences water is released to contribute to drip losses from meat (purge in packaged meat), and the refractive index of myofibrils is increased. Increased refractive index increases refractive scattering and meat becomes more pale as pH decreases. To measure these refractive changes we exploit the fact that myofibrils are birefringent – they have two refractive indices The familiar names of myofibrillar striations – A-band (anisotropic band) and I-band (isotropic band) derive from this feature discovered many years ago.

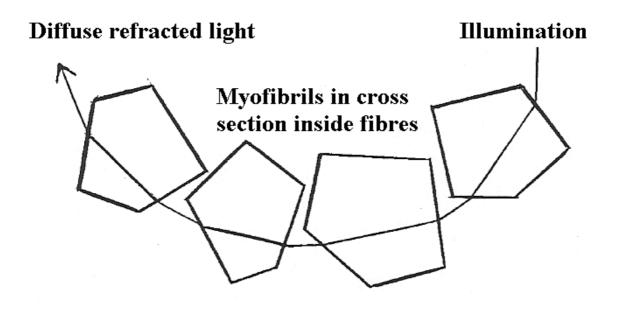


Figure 3. Light from above meat returned to the observer by myofibrillar refraction (from Swatland, 2004b).

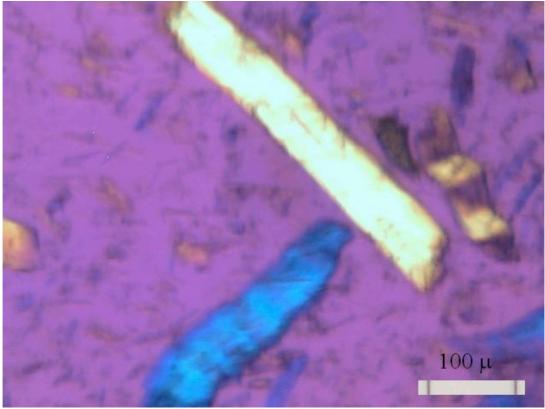


Figure 4. Two camel muscle fibre fragments seen against the interference background created by a first-order red retardation (λ) plate. The long fibre fragment perpendicular to the slow axis of the compensator has decreased the interference to appear first-order bright yellow (less than the background red). The short fibre fragment whose slow axis is parallel to that of the compensator has increased the interference to appear second-order blue (higher than the background red).

Interferometry is the measurement of the interference between waves. One set is superimposed on a second set, either to reduce wave amplitude (destructive interference) or to increase amplitude the (constructive interference). **Myofibrils** are birefringent, so that polarized transmitted light splits into two pathways _ the ordinary and extraordinary ray paths. When recombined after being transmitted through the myofibrils, the ravs combine to produce the first-order white interference commonly seen

when a muscle fibre is examined with a polarizing microscope. However, it is possible to increase the interference order by using a compensator in the polarizing microscope. А fixed compensator such as a first-order (λ) red plate produces a visible background interference colour (red) against which a muscle fibre may be viewed and measured. In one orientation the muscle fibre adds to the background interference, while perpendicular to the first orientation it subtracts from the background interference (Figures 4 and 5). In other words, when the slow-axis Swatland/ Journal of Camelid Science 2012, 5:25-40 http://www.isocard.org

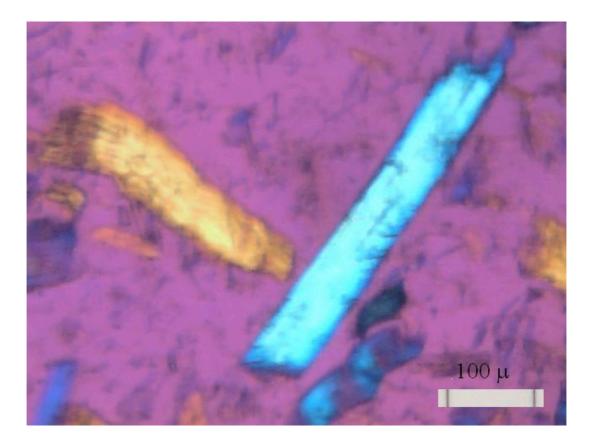


Figure 5. The same two camel muscle fibre fragments of Fig. 3 seen against the same interference background (first-order red) but after clockwise rotation of the fragments by 90° . The long fibre fragment has now increased the interference order to appear as second-order blue, while the short fibre fragment has decreased the interference to appear first-order bright yellow.

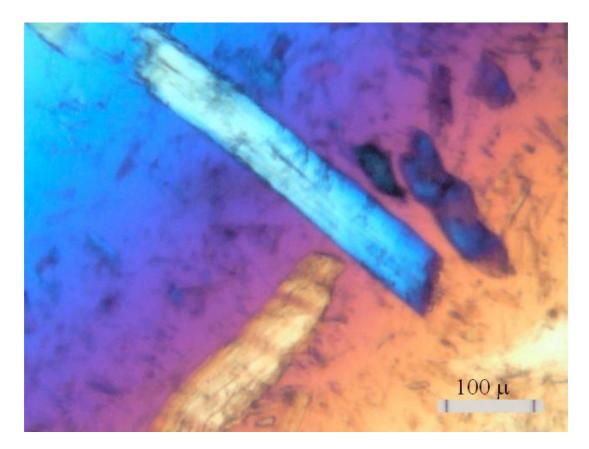


Figure 6. The same two fibre fragments seen in Figs. 3 and 4, now viewed against the background created by a rotary compensator. First-order bright yellow is the background at the bottom right corner. Second-order blue is the background at the top left corner. The long fibre fragment whose slow axis is parallel to the slow axis of the rotary compensator has increased the interference order. The short fibre fragment whose slow axis of the rotary compensator has decreased the interference order.

 (γ) of the compensator is parallel to the slow axis of the muscle fibre, the interference order is advanced, and vice versa. A rotary compensator produces the whole range of background interferences so that, in the optical axis, measurements may be made at any selected background interference (Figure 6). This greatly simplifies interferometry when the interference colour is measured with а monochromator, as explained in detail elsewhere (Swatland, 2009). So how do muscle fibres from camelid meat behave – can we detect any effect of pH on myofibrillar refraction? As shown in Figure 7, adjusting the pH of camelid muscle fibres using 0.2 M phosphate buffer causes a strong change in refraction. Thus, we can expect that as the pH declines as a consequence of post-mortem glycolysis, myofibrillar refraction is increased to scatter more light and increase the paleness of the meat.

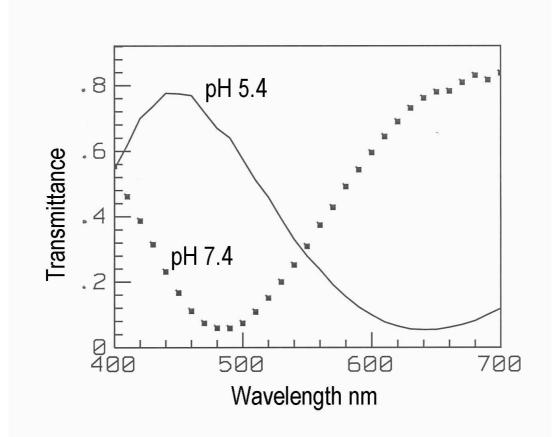


Figure 7. Polarized-light interferometry of two camel muscle fibre fragments of equal diameter orientated with their slow axis parallel to that of the compensator at tilt 5.5° .

Connective tissue fluorescence

Connective tissue contains two predominant proteins, collagen and elastin, and both proteins are well known sources of meat toughness. The amount, tensile strength and heatstability of collagen increase as animals grow older – especially if they use their muscles for locomotion. Elastin is completely resistant to cooking and tends to follow the physiological pattern of muscle activity, being abundant in postural particularly muscles. Both collagen and elastin exhibit a blue-white fluorescence when illuminated by UV light. Pyridinoline linkages in collagen increase both tensile strength and fluorescence. Thus, UV fluorescence may be used to identify meat from older animals which is likely to be tough when cooked. Meat fluorescence may be detected using laboratory apparatus, but the apparatus is expensive, delicate and unsuitable for use in an abattoir. Fortunately, however, the fluorescence of major seams of connective tissue in meat is so strong that it can be detected using a fibre-optic probe. The main question is - do seams of connective tissue in camelid meat fluoresce as strongly as they do in beef? Figure 8

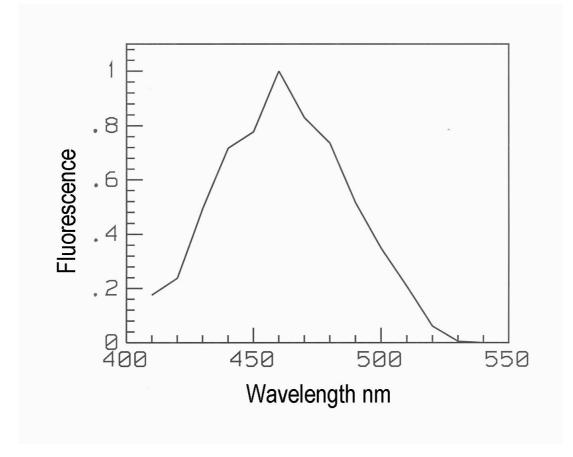


Figure 8. Relative fluorescence of the aponeurosis over camel longissimus thoracis.

shows the fluorescence of the aponeurosis (epimysium) over the *longissimus thoracis* in a camel steak. The fluorescence is as strong as that of beef and, hence, we can expect that technology developed for the detection of connective toughness in beef will also work for camelid meat.

Adipose tissue reflectance

Reflectance of adipose tissue is important for two reasons. Firstly, there may be quality concerns with the nature of the fat. For example, pork fat is sometimes dark and oily, while beef fat may be coloured yellow by dietary carotene. Both traits are judged as undesirable in many countries. The second reason for the importance of adipose reflectance is less direct. Meat grading usually includes a measure of meat yield – the percentage of saleable lean meat on a carcass. This is difficult to predict without information on the fatness of the carcass. Thus, numerous methods such as ultrasonics and optical probes have been developed to assess fatness. Optical probes are pushed in to the carcass and detect fat depth by detecting the boundary between muscle and fat. If the boundary is not distinct, a probe may give erroneous predictions of fatness. This may be a serious commercial problem if a producer receives a premium payment for a lean carcass and a penalty for a fat carcass.

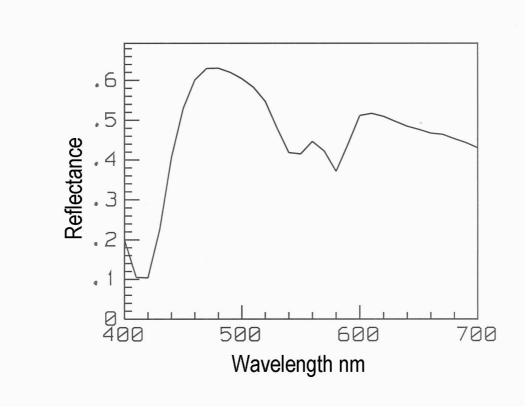


Figure 9. Fibre-optic reflectance spectrum of camel subcutaneous adipose tissue.

If this approach is to be used for camelid carcasses, then Figure 9 is of interest. It shows, as expected, that fat has a much higher reflectance than lean muscle (Figure 2) but, in the sample shown in Figure 9, there is a problem. The reflectance spectrum of adipose tissue is normally flat, with little indication of selective absorbance by haemoproteins. But, in Figure 9, there is evidence of selective absorbance most likely from haemoglobin in adipose tissue capillaries, rather than from myoglobin as in Figure 2. This could be due to chance, but might also be an indication that camelid adipose tissue may retain erythrocytes more readily than the fat of other species. A scientific study is required to test this possibility.

Future research

Very few of the methods used routinely in a meat science laboratory can be applied on-line in the meat industry, and this calls for a serious attempt to develop new technology. The apparatus must be rugged, nondestructive and easy to operate in the difficult working conditions found in the meat industry. Although this is a great challenge, the advantages to be gained from successful methods are enormous. Biophysical methods offer the greatest opportunities for on-line delivery, but meat biophysics is a poorly developed subject. Another problem is that commercial production of new technology requires either a high profit from manufacturing a small number of instruments, or a low profit from a large number. The best hope of success in this area is to search for novel biophysical indicators of meat quality. With this in mind, let us consider a possible breakthrough target.

Methods are available already for estimating quality parameters such meat yield, muscle colour, marbling, pH, fat colour and translucency (Table 1). But meat toughness is the most important and most difficult. There are many causes of toughness, but the dominant three are: (1) inadequate aging, (2) connective tissue, and (3) sarcomere length. The first can be solved by ensuring adequate postmortem aging, although this may fail if endogenous autolytic enzymes are inactive. Possible detection methods include measurements of electrical impedance (capacitance and resistance) and light scattering as an indicator of pH affecting autolytic enzymes. The second source of toughness, connective tissue, can be detected by fluorescence which is sensitive enough to detect even seasonal variation in well-aged beef roasts (Swatland. 2003). Sarcomere length is the final obstacle. Meat from any source may be made intolerably tough if muscles contract as rigor mortis develops. A typical cause is rapid refrigeration (cold shortening). The effects are readily visible under the microscope where the contractile units (sarcomeres) along muscle fibres are unusually short. But the methods we

use at present, such as phase contrast, differential interference contrast or polarized light, cannot withstand the scattering of light which occurs in bulk meat. Polarized light offers some interesting possibilities (hence the potential importance of data in Figures 4 to 7). Also, we must not pre-judge the importance of a signal versus its background noise. Thus, when scattering is the background noise, the scattering also contains information on pH (which affects autolytic enzymes) and sarcomere length (Swatland, 2005). There are many possibilities for future research in this area, exploring the biophysical properties of camelid meat and adapting them for commercial use. As the problems of global warming and the expansion of arid areas become more severe (Lutz, 1998; Makoto et al., 2006), the contribution of camelid meat to World protein production may increase (McCloy and Rowe, 2000), thus justifying the development of new technology for quality control and grading of premium exports.

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