METHODS FOR DETERMINING ANIMAL AGE

D.J. Horgan, N.L. King and L. Kurth, CSIRO Division of Food Processing, Meat Research Laboratory, P.O. Box 12, Cannon Hill, Queensland 4170, Australia

SUMMARY

Three measures of the thermal stability of collagen (thermal transition temperature, isometric tension and hydroxyproline solubilised on heating) have been studied as a function of animal age, with a view to evaluating their potential for estimating the age of an animal from which a sample of meat originated.

The variation in thermal transition temperature between animals of similar age was too large for reliable estimation of animal age by this method. Hydroxyproline solubilised on heating decreased sharply with age up to one year but changed little with age in older animals. The peak force developed during isometric heating of goat LD tendon was found to increase linearly with age up to 12 years, and to provide an estimate of age with 95% confidence limits ranging from ± 6 months at a mean age of 1 year to ± 18 months at a mean age of 8 years.

INTRODUCTION

Many contracts for the supply of meat stipulate that it should originate from animals within a certain age range. Although animal age is traditionally estimated by dentition (e.g. Franklin 1950), alternative methods are required when meat has been removed from a carcass, so that purchasers of meat cuts may check for compliance with their specifications.

Most proteins in a living animal are turned over rapidly, but some of the collagen appears to survive, and undergoes changes in chemical and physical properties which may serve as a basis for determining animal age. The purpose of this paper is to examine the manner in which several properties of collagen change with age, with a view to evaluating their potential for estimating the age of meat-producing animals. Methods reported to yield values related to age are: isometric tension of rat-tail (Brocas and Verzár 1961a) and frog-finger (Brocas and Verzár 1961b) tendons, as well as hydroxyproline solubilised on heating cattle corium (Verzár 1960), rat-tail tendon (Verzár and Meyer 1961), human skin (Verzár 1962) and rat muscle (Schaub 1963). In the present study these two methods, as well as differential scanning calorimetry, were applied to goat tendon and intramuscular connective tissue.

EXPERIMENTAL METHODS

Fifty-seven goats of known birthdays were acquired from a number of properties in South-East Queensland. After stunning by captive bolt pistol, the animals were exsanguinated. The dressed carcasses were pelvic hung in a chiller at 2°C for 48 h. The following muscles were then removed and stored frozen for future use: *longissimus dorsi* (LD), *semimembranosus* (SM), *semitendinosus* (ST), *biceps femoris* (BF), *psoas major* (PM) and *vastus lateralis* (VL).

Samples of tendon were obtained by dissection and suspended overnight in 50 mM sodium citrate, 10 mM ethylene diamine tetra-acetic acid, pH 6.0 (buffer A) prior to measurement of thermal properties. Intramuscular connective tissue preparations were carried out as described by King (1987) except that buffer A was used throughout. Hydroxyproline solubilised on heating connective tissue at 90°C for 20 min in buffer A was determined as described by Schaub (1963). Samples of connective tissue were suspended in buffer A for measurements of thermal transition temperature by differential scanning calorimetry (King 1987) and isometric tension (Snowden et al. 1977).

FIGURE 1

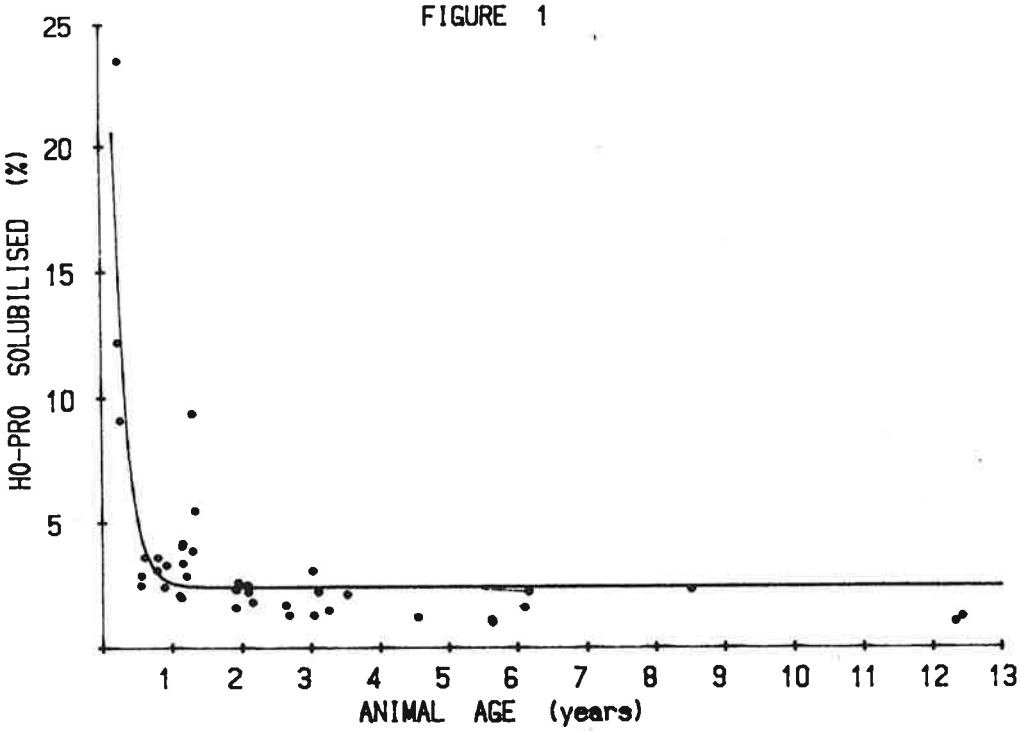

Figure 1: Hydroxyproline solubilised on heating. Goat LD intramuscular connective tissue was heated at 90°C for 20 min in buffer A. Each point represents a single determination. Hydroxyproline solubilised is expressed as a percentage of the total hydroxyproline in the sample of connective tissue. The curve was obtained by fitting an equation of type $y = A + B\exp(Kt)$ where t is animal age and A, B, K are constants.

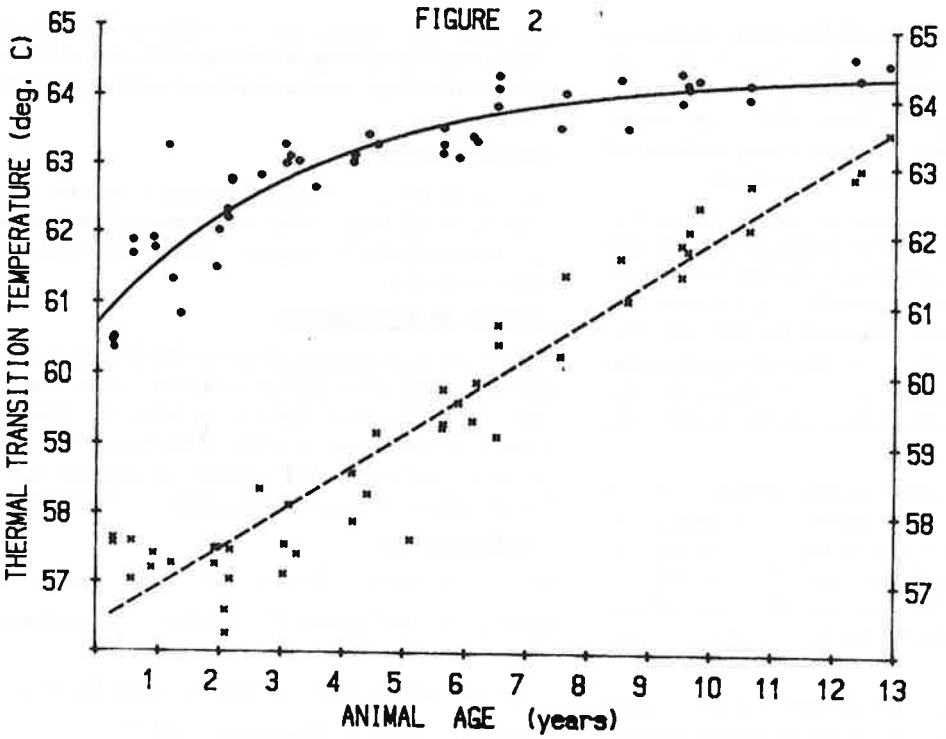

Figure 2:Thermal transition temperature as a function of animal age. Each point is the mean of two determinations. 0____0 Goat LD intramuscular connective tissue in buffer A. The curve was obtained by fitting an equation of the type $y = A + B \exp(Kt)$ where t is animal age and A, B, K are constants. X___X Goat LD tendon in buffer A.

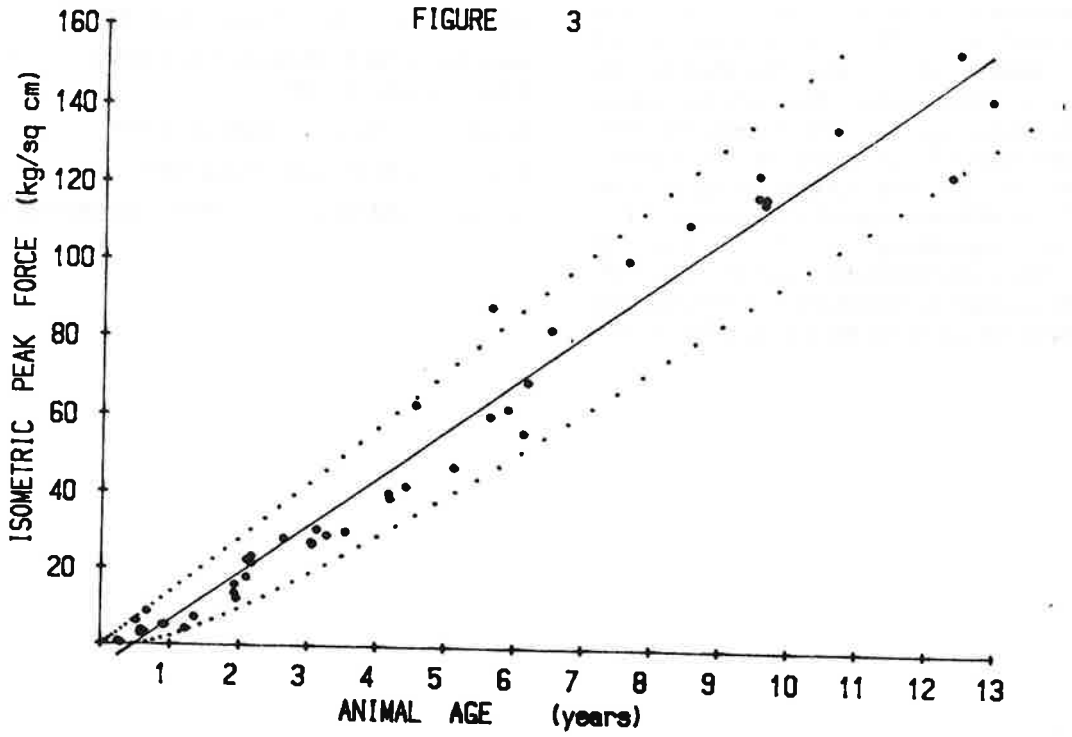

Figure 3: Peak force developed during isometric heating. Goat LD tendon was immersed in buffer A and the temperature raised at 2°C/min. Each point is the mean of at least 3 determinations. ..... 95% confidence limits obtained from square root (isometric tension) vs square root (animal age).

RESULTS AND DISCUSSION

Dentition of the goats used in this study was assessed at slaughter. The age range of animals exhibiting no permanent incisors was 0-16 months; 1-2 incisors, 14-26 months; 3-4 incisors, 23-25 months; 5-6 incisors, 23-53 months; and 7-8 incisors, 32 months upwards. Most of the older animals had lost some of their incisors. The wide age range at each stage (except 3-4 tooth) and the overlap between ranges limit the precision with which animal age can be estimated from dentition. Furthermore, dentition does not distinguish between ages in the later years of an animal's life, i.e. beyond the full-mouth stage. As diet has been reported to influence dental development of sheep (Franklin 1950) the diverse sources and nutritional backgrounds of the goats used in this study may have expanded the above ranges beyond those expected for a group of animals on a uniform diet.

Hydroxyproline solubilised on heating LD intramuscular connective tissue is shown as a function of animal age in Fig.1. Similar results were obtained for SM, ST, BF and PM intramuscular connective tissue. As reported previously for rat muscle (Schaub 1963) there is a sharp

decline with age for young animals but little change in older ones. Although older age groups are not differentiated, this method may be of use for distinguishing very young kid from adult goat meat. However, the large variation between young animals of similar age precludes accurate age determination.

The thermal transition temperature of intramuscular collagen also exhibits a curvilinear relationship with age (as shown for LD in Fig. 2) but the curvature is not as sharp as for hydroxyproline solubilised on heating. Curves of similar shape were obtained for BF, SM, ST, PM and VL. For each muscle, the scatter of experimental values about the fitted line is such that only an approximate estimate of animal age could be obtained by this method.

Figure 2 also shows thermal transition temperature of LD tendon as a function of age. In contrast to intramuscular collagen, there is a linear relationship. The scatter of experimental points is such that the 95% confidence limits are approximately 2.4 years on either side of the regression line - too imprecise to be of value for age estimation.

Isometric peak force developed during heating of goat LD tendon is shown in Fig. 3. In common with the tendon thermal transition temperatures, there is a linear relationship with age. There is less scatter of experimental points about the fitted line than for the thermal transition temperatures. However, the scatter increases with animal age. For this reason, the 95% confidence limits shown in Fig. 3 were calculated from a plot of square root (isometric tension) vs square root (age). The 95% confidence limits for mean ages of 1, 2, 4 and 8 years were respectively ± 6, ± 8, ± 11 and ± 18 months. Isometric tension therefore appears to provide a more reliable estimate of animal age than does thermal transition temperature, and the peak force is linear over the full age range, in contrast to hydroxyproline solubilised on heating. Unfortunately, not all tendons are sufficiently large and uniform to yield consistent peak force values.

CONCLUSION

The peak force developed during isometric heating of goat LD tendon provides an estimate of animal age, the precision of which compares favourably with estimates from dentition.

ACKNOWLEDGEMENT

This work was supported in part by the Australian Meat and Livestock Research and Development Corporation. The authors are indebted to Mrs. C. Kennedy for technical assistance, to Mr. P.N. Jones for advice on statistics and to Mr. R.F. Dickinson and Mr. R.J. Logue for slaughter of animals and dressing of carcasses.

REFERENCES

Brocas, J. and Verzár, F. (1961a). *Gerontologia* **5**:223.

Brocas, J. and Verzár, F. (1961b). *Gerontologia* **5**:228; *Experientia* **17**:421.

Franklin, M.C. (1950). *CSIRO Bulletin No.252.*

King, N.L. (1987). *Meat Science* **20**:25.

Schaub, M.C. (1963). *Gerontologia* **8**:16.

Snowden, J. McK., Bouton, P.E. and Harris, P.V. (1977). *J. Food Science* **42**:890.

Verzár, F. (1960). *Gerontologia* **4**:104.

Verzár, F. (1962). *Experientia* **18**:473.

Verzár, F. and Meyer, A. (1961). *Gerontologia* **5**:163.