Characterization of Thermomechanical Behaviour of Skin Tissue II. Viscoelastic Behaviour

F. Xu, T. Wen, K. A. Seffen, and T. J. Lu

***Abstract*—This paper presents the viscoelastic behaviour of skin tissue under different thermal loadings and discusses the effect of temperature and corresponding dermal collagen denaturation on the mechanical viscoelastic properties of skin tissue. Differential scanning calorimetry (DSC) has been used to detect dermal collagen denaturation and to assess its thermal stability. The DSC results under various heating rates are used to derive the Arrhenius parameters (E_a , A) in the burn damage integration, which are then used to calculate the degree of denatured collagen in the skin. Temperature tests have been performed using a dynamic mechanical analyzer (DMA) to evaluate changes in the skin viscoelastic properties as a function of collagen damage, specifically, to assess the changes in the storage modulus (E'), and loss factor ($\tan\delta$). The results show remarkable changes in E' , which is maybe due to the release of water, but there is no significant effect from $\tan\delta$. These results suggest that at a constant frequency the denaturation of collagen molecules has little effect on the viscoelasticity.**

***Index Terms*—Skin tissue, thermomechanical behaviour, thermal denaturation, viscoelasticity.**

I. Introduction

Skin is a viscoelastic material. Previous studies of the viscoelasticity of skin tissue reveal that its stress-strain relationship depends on strain rate, loading rate, the period of loading and on the preconditioning stress history, and that it exhibits considerable hysteresis in cyclic tests, as well as stress relaxation under constant strain [1]-[3]. Most of these studies were performed under quasi-static experimental conditions; however, few researchers have described the dynamic viscoelastic properties of skin tissue [4], [5].

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As the skin temperature increases, denaturation of collagen takes place. There are not only structure changes, but the hydration of collagen also changes with denaturation, which may involve a liberation of water initially and, an absorption of water later. Not surprisingly, thermal denaturation of a collagenous tissue can lead to marked changes in its viscoelastic properties. To the authors' knowledge, there is no published study on temperature-dependent skin viscoelasticity, although other collagenous tissues have been studied, for example, cartilage [6], [7] and bone [8], [9].

The objective of this investigation is to test the hypothesis that collagen is a significant determinant of skin viscoelasticity, and particular attention has been paid here to the thermomechanical properties of skin under dynamic loading. For this purpose, collagen in skin tissue is thermally denatured to different degrees by applying heat. Differential scanning calorimetry (DSC) measurement is used to detect the denaturation of collagen and to measure its thermal stability. The integrity of the collagen network is then analyzed using the thermal damage integration. In addition, a dynamic mechanical analysis (DMA) is carried out to characterize the viscoelastic properties of skin tissue as a function of temperature and collagen damage.

II. Quantification of Thermal Denaturation and Thermal Damage

Thermal damage and thermal denaturation can be quantified by the following calculations:

$$\Omega(t) = \ln\left(\frac{C(0)}{C(t)}\right) = \int_0^t A\exp\left(-\frac{E_a}{RT}\right)dt \tag{1}$$

$$Deg(t) = \frac{C(0)-C(t)}{C(0)} = 1-\exp\left(-\Omega(t)\right) \tag{2}$$

where t is the time after starting, Ω is the dimensionless indicator of thermal damage, "*Deg*" is the degree of thermal damage (concentration of denatured collagen), $C(0)$ and $C(t)$ are the initial and current concentrations of un-denatured collagen, A is a material parameter (frequency factor), E_a is

the activation energy, and $R = 8.314$ J/mol K is the universal gas constant. The Arrhenius parameters (E_a , A) are derived experimentally from the DSC results in Section 4.1.

III. Materials and Methods

A. Sample and sample preparation

The sample and its preparation have been described in detail in our first paper, see [10].

B. Experimental procedure

Differential Scanning Calorimetry (DSC)

Differential scanning calorimetry has been used extensively to characterize the thermal behaviour of collagenous tissues [11], [12]. DSC detects thermodynamic changes by measuring the flow of heat between a sample and a reference, from which the effect of thermal history on the heat capacity can be determined, enabling different parameters to be interrogated, such as the thermal denaturation temperatures of skin collagen, reaction kinetics, etc. In this study, the thermal stability of collagen in skin tissue was assessed with a TA Instruments DSC of type Q1000 T_{zero}, scanning from 20 to 100 °C at four different heating rates (2, 5, 10, 20 °C/min).

Dynamic Mechanical Analysis (DMA)

The viscoelastic properties of the skin tissue as a function of temperature were measured with a Dynamic Mechanical Analyzer (TA Instruments DMA Q800). The measurements are carried out by bending a single cantilever at a frequency of 1 Hz, over a temperature range from 25 to 100 °C, subject to different heating rates (2, 5, 10, 20 °C/min). The flexural moduli are measured within the linear elastic region. The deformation mode produces a combination of both flexural and shear forces where, in short thick specimen, shear forces are dominant and vice versa in long thin specimens. In our tests, the length to thickness ratio of skin samples is greater than 10, which favors flexure [7]. Note that all the tests are performed in a test chamber filled with air.

IV. Results and Discussion

A. DSC results

A typical DSC thermogram of a pig flank skin sample is shown in Fig. 1. The measured specific heat capacity, c_p , compares well with published results [11], [13]. For a stress-free skin sample, denaturation is characterized by a sudden increase in the energy absorption endotherm, starting at about 60 °C: with further heating, the endotherm reaches its maximum value at the denaturation temperature, 66.83 °C and then decreases. It is also observed that the denaturation endotherm characteristics are heating rate dependent but are not shown here for they offer a similar trend.

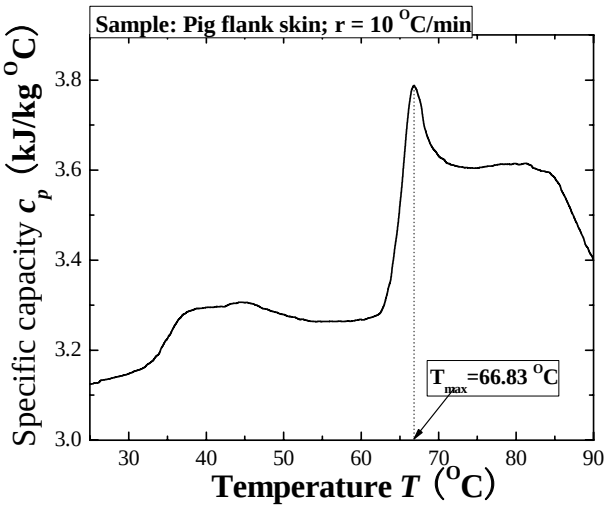

Fig. 1 Characteristic DSC thermogram of a skin tissue sample

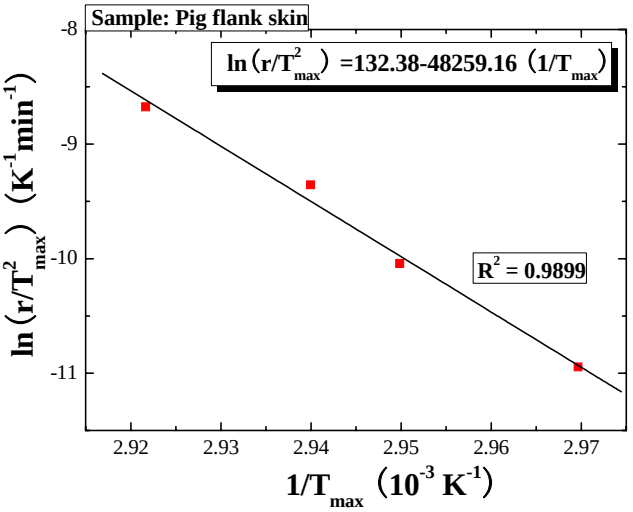

Fig. 2 Characteristic plot of $\ln\left(r/T^2_{\max}\right)$ vs. $1/T_{\max}$ for skin tissue

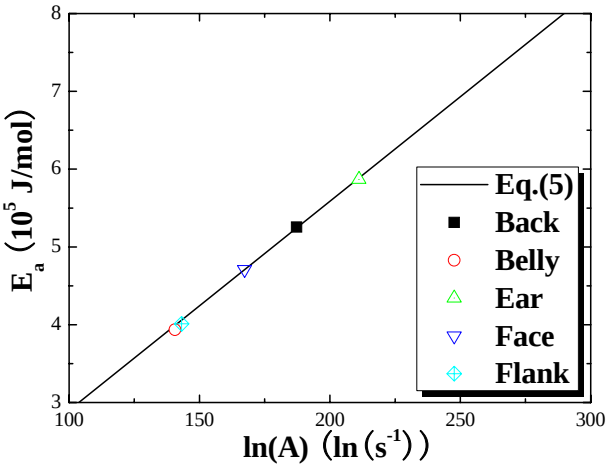

Fig. 3 Comparison of our results of Arrhenius parameters (A , E_a) with literature results

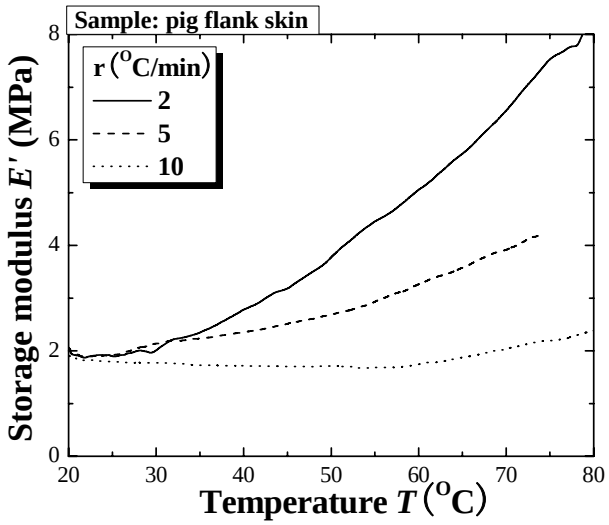

Fig. 4 Variation of storage modulus with temperature

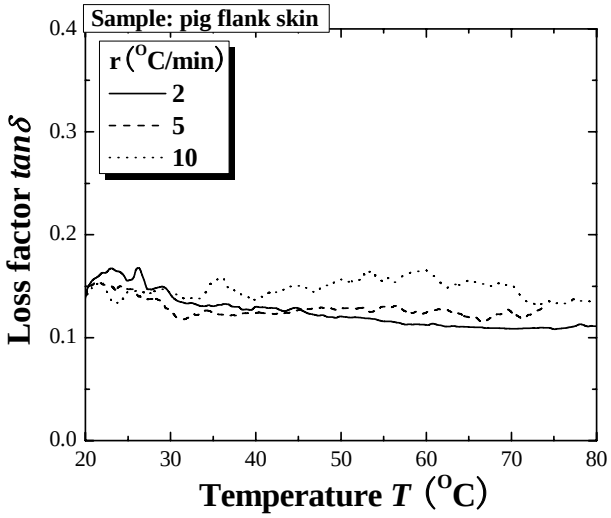

Fig. 5 Variation of loss factor with temperature

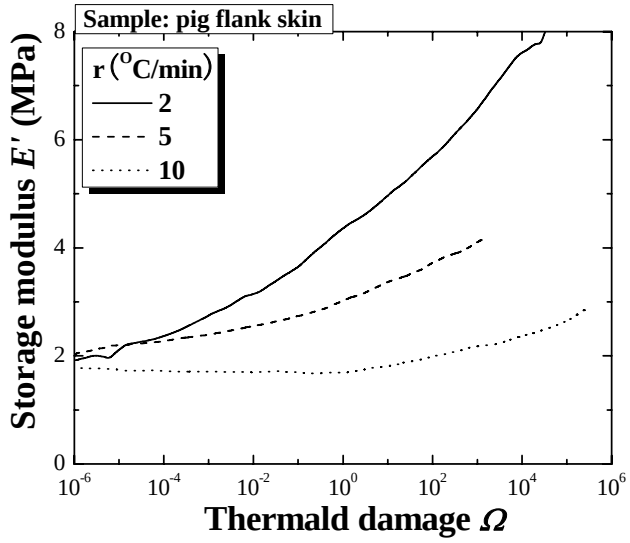

Fig.6 Variation of storage modulus with thermal damage

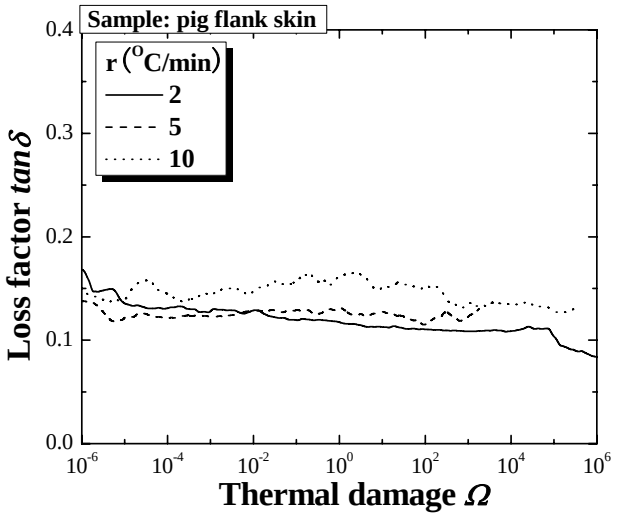

Fig. 7 Variation of loss factor with thermal damage

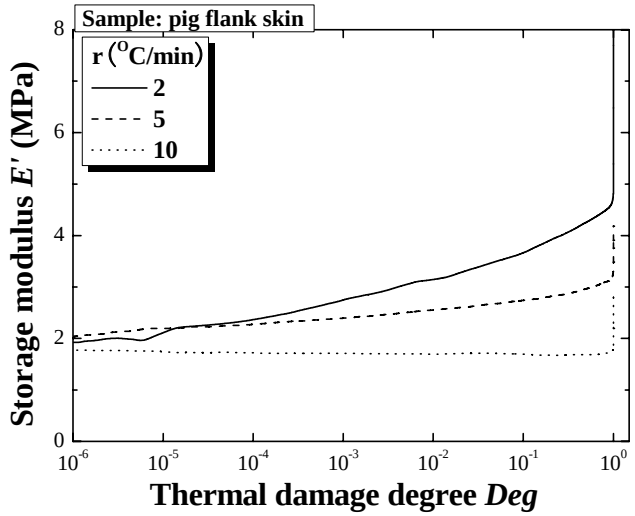

Fig. 8 Variation of storage modulus with thermal damage degree

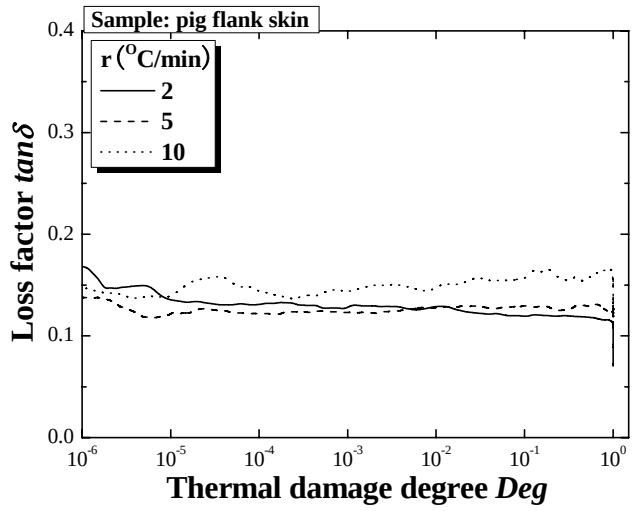

Fig. 9 Variation of loss factor with thermal damage degree

Table I Experimental results of Arrhenius parameters (E_a , A)

Sample	Slope	Intercept	E_a $\left(10^5\ \mathrm{J/mol}\right)$	A $\left(\mathrm{s}^{-1}\right)$
Back skin	-63200.78	176.21	5.255	2.126×10^{81}
Belly skin	-47323.37	129.83	3.935	1.151×10^{61}
Ear skin	-70568.67	200.03	5.867	5.240×10^{91}
Face skin	-56647.50	156.36	4.710	4.575×10^{72}
Flank skin	-48259.16	132.38	4.012	1.501×10^{61}

In this study, the collagenous tissue is heated from an initial value at a constant heating rate, r . According to the thermodynamic and Arrhenius equation, the following relation between the parameters, E_a and A , and the peak temperature of thermal denaturation, $T_{\max}$, can be obtained as [14]-[16]:

$$\frac{rE_a}{ART_{\max}^2} = \exp\left(-\frac{E_a}{RT_{\max}}\right) \tag{3}$$

Thus, the following equation becomes apparent,

$$\begin{aligned} &\ln\left(\frac{rE_a}{ART_{\max}^2}\right) = \ln\left(\exp\left(-\frac{E_a}{RT_{\max}}\right)\right) \\ &\Rightarrow \ln\left(\frac{r}{T_{\max}^2}\right) = -\frac{E_a}{R}\frac{1}{T_{\max}} - \ln\left(\frac{E_a}{R}\right) + \ln(A) \end{aligned} \tag{4}$$

The value of activation energy E_a can be obtained from the slope of the plot of $\ln\left(r/T_{\max}^2\right)$ versus $1/T_{\max}$, while the value of A can be derived from the intercept value. A characteristic plot for flank skin sample is given in Fig. 2 and for separate tests with belly, back, ear and face skin, the Arrhenius parameters are presented in Table I. As shown in our early study [17], there is a linear relationship between $\ln(A)$ and E_a given by, after curve fitting:

$$E_a = 21149.324 + 2688.367\ln(A) \tag{5}$$

The present results compare favourably with this equation. Accordingly, the thermal damage and the degree of thermal denaturation of skin collagen for a given heating history can be found using Eq.(1) and Eq.(2).

B. DMA results

The viscoelastic properties of skin are obtained from the DMA tests. During each DMA experiment, the storage modulus (E'), loss modulus (E''), and the loss factor ($\tan\delta$), are measured as a function of the temperature history. The storage modulus is related to the stored elastic energy in the viscoelastic cycle, and can be considered analogous to a stiffness measurement of a monotonic test; loss factor yields the ratio between the mechanical energy lost and stored during a given cycle, $\tan\delta = E''/E'$, effectively measuring the damping performance of the sample.

The storage modulus and the loss factor are given in Fig. 4, and Fig. 5, respectively as a function of temperature. The storage modulus increases with increasing temperature: the skin gets stiffer at higher temperatures and the increasing rate increases with heating rates. The loss factor, however, is independent of temperature but increases marginally with the rates of heating: temperature has little effect on the viscous property of skin or the damping of the sample is not affected by temperature. The value of loss factor in our results is about 0.14, similar to the result measured by the method of surface wave propagation at room temperature [18].

It has been generally accepted that, under quasi-static conditions, collagen is responsible for the viscoelastic properties and the stiffness of skin tissue [19] , suggesting that collagen plays an important role in determining the overall mechanical properties of skin tissue. Thus, it seems reasonable to assume that collagen may play a major role in the viscoelastic nature of skin tissue under thermal loadings. In order to investigate this hypothesis, the storage modulus and loss factor are plotted as a function of thermal damage and thermal denaturation degree, as given in Fig. 6-9. These results show that the effects of thermal damage and thermal denaturation degree are similar to that of temperature above; in other words, the change of the interval structure of collagen during thermal denaturation does not affect the storage modulus and the loss factor at the test frequency used in this study. It has been shown elsewhere that the mechanical changes observed using dynamic mechanical analysis are the result of water loss and of changes in the intrinsic molecular structure of the material with heating [7]. Furthermore, weight loss of skin samples has also been observed during the test. Thus, the variations in our tests may be due to due to the release of water by the denaturation of collagen molecular during heating.

V. Conclusion

To our knowledge, this study first characterizes the effect of temperature and corresponding thermal denaturation of skin collagen on skin viscoelastic properties. The results show that the storage modulus is temperature sensitive, which is possibly due to dehydration. However, the loss factor does not show a significant dependency on either temperature or thermal denaturation. These results suggest that at a constant frequency, the denaturation of collagen has little effect on skin viscoelasticity.

Further study should focus on identifying the relative

contribution of water loss and thermal denaturation. Although not explored in this study, it is expected that the test frequency plays a role. In future, more extensive studies using a wide range of frequencies should be performed.

REFERENCES

[1] Y. C. Fung, *Biomechanics: Mechanical Properties of Living Tissues*. New York: Springer-Verlag, 1993,

[2] Y. Lanir, "Biaxial stress relaxation in skin," *Ann. Biomed. Eng.*, vol. 4, 1976, pp. 250-270.

[3] T. Hermanns-Le, I. Uhoda, S. Smitz, and G. E. Pierard, "Skin tensile properties revisited during ageing. Where now, where next?," *J. Cosmet. Dermatol.*, vol. 3, 2004, pp. 35-40.

[4] E. K. Dawes-Higgs, M. V. Swain, R. J. E. D. Higgs, R. C. Appleyard, and S. Kossard, "Accuracy and reliability of a dynamic biomechanical skin measurement probe for the analysis of stiffness and viscoelasticity," *Physiol. Meas.*, vol. 25, 2004, pp. 97-105.

[5] C. Jacquemoud, K. Bruyere-Garnier, and M. Coret, "Methodology to determine failure characteristics of planar soft tissues using a dynamic tensile test," *Journal of Biomechanics*, vol. 40, 2007, pp. 468-475.

[6] K. K. H. Chao, M. A. Burden, and B. J. F. Wong, "Dynamic changes in the elastic modulus of lagomorph nasal septal cartilage during Nd:YAG (λ=1.32 μm) laser irradiation," San Jose, CA, 2001, pp. 247-254.

[7] Y. Chae, G. Aguilar, E. J. Lavernia, and B. J. Wong, "Characterization of temperature dependent mechanical behaviour of cartilage," *Lasers Surg. Med.*, vol. 32, 2003, pp. 271-278.

[8] J. Yamashita, B. R. Furman, H. R. Rawls, X. Wang, and C. M. Agrawal, "Collagen and bone ciscoelasticity: a dynamic mechanical analysis," *J. Biomed. Mater. Res. (Appl. Biomater.)*, vol. 63, 2002, pp. 31-36.

[9] J. F. Mano, "Viscoelastic properties of bone: Mechanical spectroscopy studies on a chicken model," *Materials Science and Engineering C*, vol. 25, 2005, pp. 145-152.

[10] F. Xu, T. Wen, K. A. Seffen, and T. J. Lu, "Characterization of Thermomechanical Behaviour of Skin Tissue I. Tensile and Compressive behaviours," in *World Conference on Engineering* London, UK, 2007.

[11] G. S. Young, "Thermodynamic Characterization of Skin, Hide and Similar Materials Composed of Fibrous Collagen," *Studies in Conservation*, vol. 43, 1998, pp. 65-79.

[12] R. Schiller, A. P. Funke, and C. Gunther, "DSC measurements on full thickness mice skin: An additional tool to investigate permeation enhancement of highly lipophilic drugs," *Journal of Thermal Analysis and Calorimetry*, vol. 77, 2004, pp. 497-510.

[13] F. C. Henriques and A. R. Moritz, "Studies of thermal injury, 1. The conduction of heat to and through skin and the temperatures attained therein. A theoretical and an experimental investigation," *A. J. Pathol.*, vol. 23, 1947, pp. 531-549.

[14] T. Ozawa, "Kinetic analysis of derivatives curves in thermal analysis," *Journal of Thermal Analysis and Calorimetry*, vol. 2, 1970, pp. 301-324.

[15] C. A. Miles, "Kinetics of collagen denaturation in mammalian lens capsules studied by differential scanning calorimetry," *Int. J. Biol. Macromol.*, vol. 15, 1993, pp. 265-271.

[16] C. A. Miles, T. V. Burjanadze, and A. J. Bailey, "The kinetics of the thermal denaturation of collagen in unrestrained rat tail tendon determined by differential scanning calorimetry," *J.Mol.Biol.*, vol. 245, 1995, pp. 437-446.

[17] F. Xu, T. Wen, K. A. Seffen, and T. J. Lu, "Biothermomechanics of Skin Tissue," *Journal of the Mechanics and Physics of Solids*, 2007,

[18] S. J. Kirkpatrick, D. D. Duncan, and L. Fang, "Low-frequency surface wave propagation and the viscoelastic behaviour of porcine skin," *Proceedings of SPIE* vol. 5474, 2004, p. 127

[19] F. H. Silver, J. W. Freeman, and D. DeVore, "Viscoelastic properties of human skin and processed dermis," *Skin Res.Technol.*, vol. 7, 2001, pp. 18-23.