



## Short communication

## The toughness of adipose tissue: measurements and physical basis

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## ABSTRACT

The measured toughness  $J_C$  of adipose and dermal porcine tissues are 4.1 and 17  $\text{kJ m}^{-2}$ , respectively, via a trouser tear test. An assessment is made of the contribution to overall toughness from the microstructural elements. The analysis suggests that the toughness of adipose tissue is determined by the collagen network that surrounds the adipocytes. The volume fraction of the interlobular septa is sufficiently low for it to make a negligible contribution to the macroscopic toughness.

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## 1. Introduction

Subcutaneous adipose tissue is a soft connective tissue that resides under the dermal layer of skin (Goldberg and Rabinovitch 1983). In this study, the toughness of adipose tissue is measured using a trouser tear test (Rivlin and Thomas 1953), as recommended by Purslow (1983). Additionally, the crack tip zone of adipose tissue is examined in a scanning electron microscope (SEM). A limited set of toughness measurements are also reported for porcine dermal tissue, for comparison purposes.

Adipose tissue is comprised of lipid-filled cells called *adipocytes*, of diameter approximately 80  $\mu\text{m}$  (Sheldon 1965; Björntorp and Martinsson 1966). Adipocytes are enclosed within an extra cellular matrix constructed from two distinct collagen-based structures: *reinforced basement membrane* and *interlobular septa*, see Fig. 1. The reinforced basement membrane, of thickness 2  $\mu\text{m}$ , is comprised of various types of fibrous collagen and resembles the walls of a 3D closed-cell foam (Abrahamson 1986; Nakajima et al., 1998), with each adipocyte occupying a pore cavity of the foam. The interlobular septa form a type I collagen 3D network, resembling a 3D open-cell foam (Urmacher 1997). The two collagen structures are inter-penetrating: it is argued that the septa fibres act as reinforcement fibres of the closed-cell reinforced basement membrane, similar to fibre reinforcement of an engineering closed-cell foam. The dominant contribution to toughness from these two collagen structures is assessed herein.

At low strain rates adipose tissue is a viscoelastic solid with a storage modulus  $E^* \approx 1 \text{ kPa}$  (Miller-Young et al., 2002; Patel et al., 2005; Gefen and Haberman 2007; Geerligs et al., 2008a; Comley and Fleck 2009). No measurements of the toughness of adipose tissue have been found in the existing literature.

## 2. Materials and method

Samples of fresh porcine adipose tissue from the jowl and dermal tissue from the foreleg were obtained<sup>1</sup> and stored for less than four hours in phosphate buffered saline (PBS) prior to testing. Trouser tear specimens (Purslow, 1983; Chin-Purcell and Lewis, 1996) of length  $L=30 \text{ mm}$  and width  $W=20 \text{ mm}$ , and a ligament length of approximately  $D=L/3$  were prepared (thickness  $B=5 \text{ mm}$  for adipose and  $B=3 \text{ mm}$  for dermis). The legs of each specimen were pulled apart by a screw-driven tensile test machine at a constant displacement rate  $\dot{u}$  in the range 0.01–10  $\text{mm s}^{-1}$ , and the force  $F$  versus displacement  $u$  was recorded.

## 3. Results

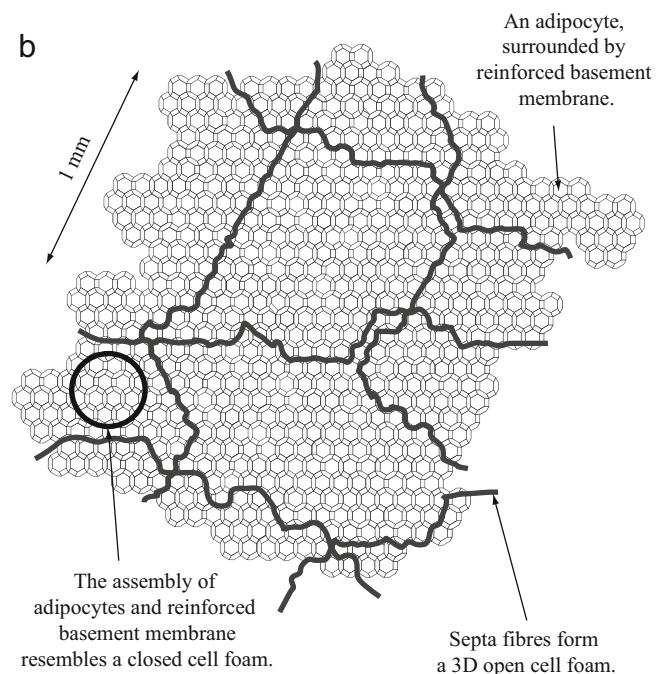
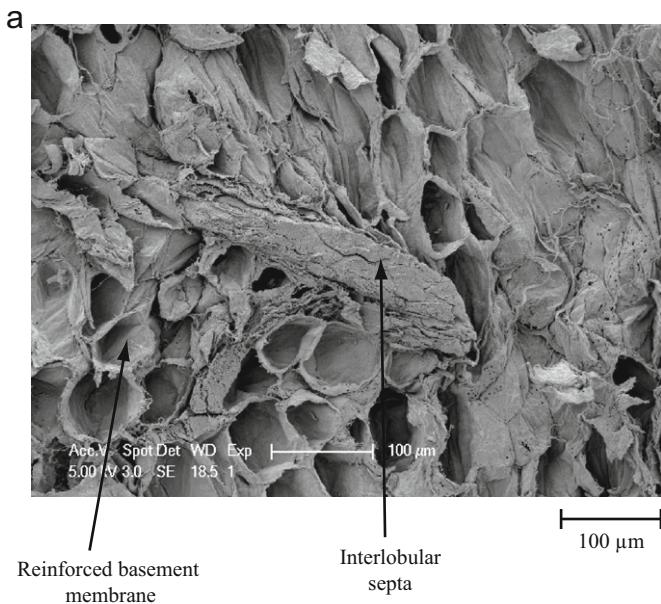
The  $F-u$  curves from the trouser tear tests are shown in Fig. 2 for selected values of  $\dot{u}$ . In each case  $F$  increased with  $u$  and then oscillated about a mean value  $\bar{F}$  ( $\bar{F}$  is calculated as the average force measured following onset of cracking until the end of the test). Video monitoring revealed that crack growth initiated when the load achieved the plateau value, and thereafter, the crack growth rate  $\dot{a}$  was constant. Thus, it was adequate to deduce  $\dot{a}$  from the total crack extension. The toughness  $J_C$  is given by (Rivlin and Thomas 1953; Purslow 1989; Chin-Purcell and Lewis 1996)

$$J_C = \frac{2\bar{F}}{B} \quad (1)$$

and is plotted as a function of crack velocity  $\dot{a}$  in Fig. 3 for both tissues. The average toughness is  $J_C=4.1 \text{ kJ m}^{-2} \pm 1.2 \text{ kJ m}^{-2}$  (1 s.d.) for porcine adipose tissue and  $J_C=17 \text{ kJ m}^{-2} \pm 4 \text{ kJ m}^{-2}$  (1 s.d.) for porcine dermis, independent of crack velocity  $\dot{a}$  suggesting that viscoelastic effects are minor at these low crack velocities. The toughness value measured here for the dermis is of a similar magnitude to the values reported by Purslow (1983),

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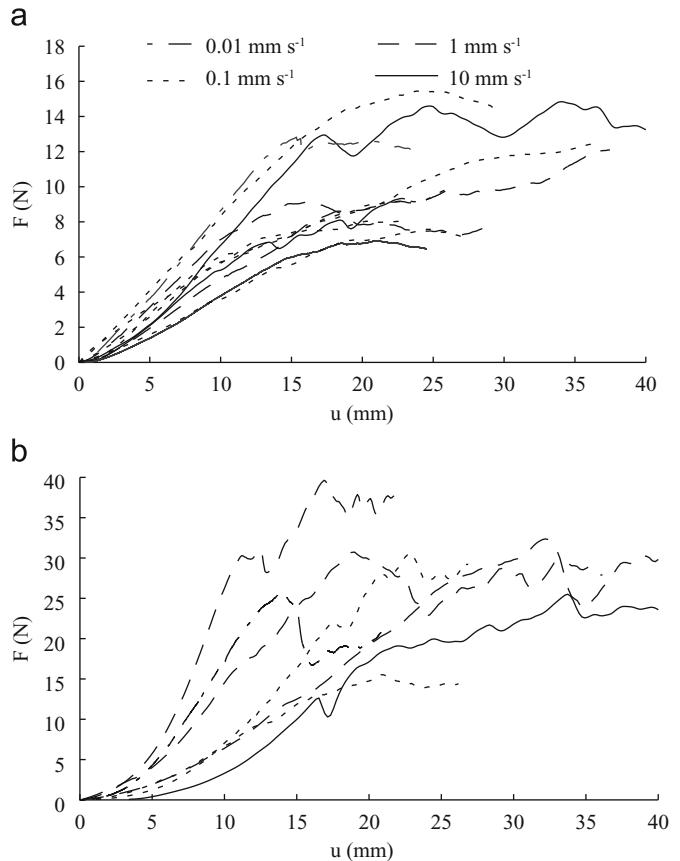


**Fig. 1.** (a) Scanning electron microscope (SEM) image, showing the reinforced basement membrane that surrounds each adipocyte and an interpenetrating septa fibre, (b) a sketch of a lobule of adipose tissue.

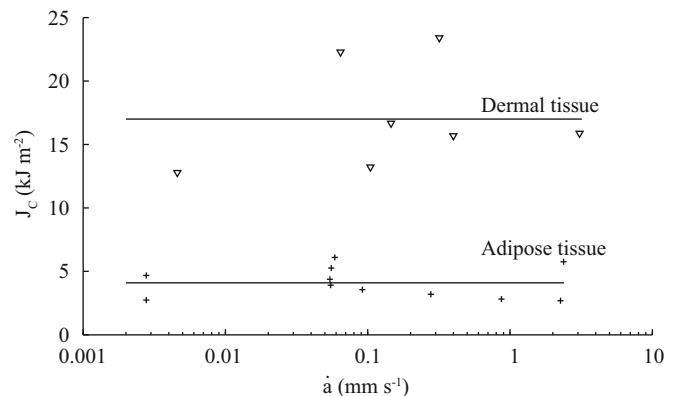
although he observed a slight increase in value (by a factor of 2) when  $\dot{a}$  is increased from  $0.08$  to  $8 \text{ mm s}^{-1}$ .

In order to determine the micromechanism of crack advance a test at  $\dot{u} = 0.1 \text{ mm s}^{-1}$  was interrupted after a crack extension of  $3.8 \text{ mm}$  and the crack tip was examined in an SEM, as follows. To maintain the deformed configuration the specimen was clamped by cotton thread to a glass slide, and then chemically fixed, see Fig. 4a. The glass slide was then removed and the crack tip zone was examined in a SEM.

Scanning electron micrographs of the crack front are shown in Fig. 4. Three distinct features of the collagen network are observed: (i) the reinforced basement membrane surrounding the adipocytes has ruptured, allowing the lipid to leak out, (ii) the crack path follows the boundaries of the reinforced basement



**Fig. 2.** Results from trouser tear tests at selected displacement rates for (a) porcine adipose tissue and (b) porcine dermis.

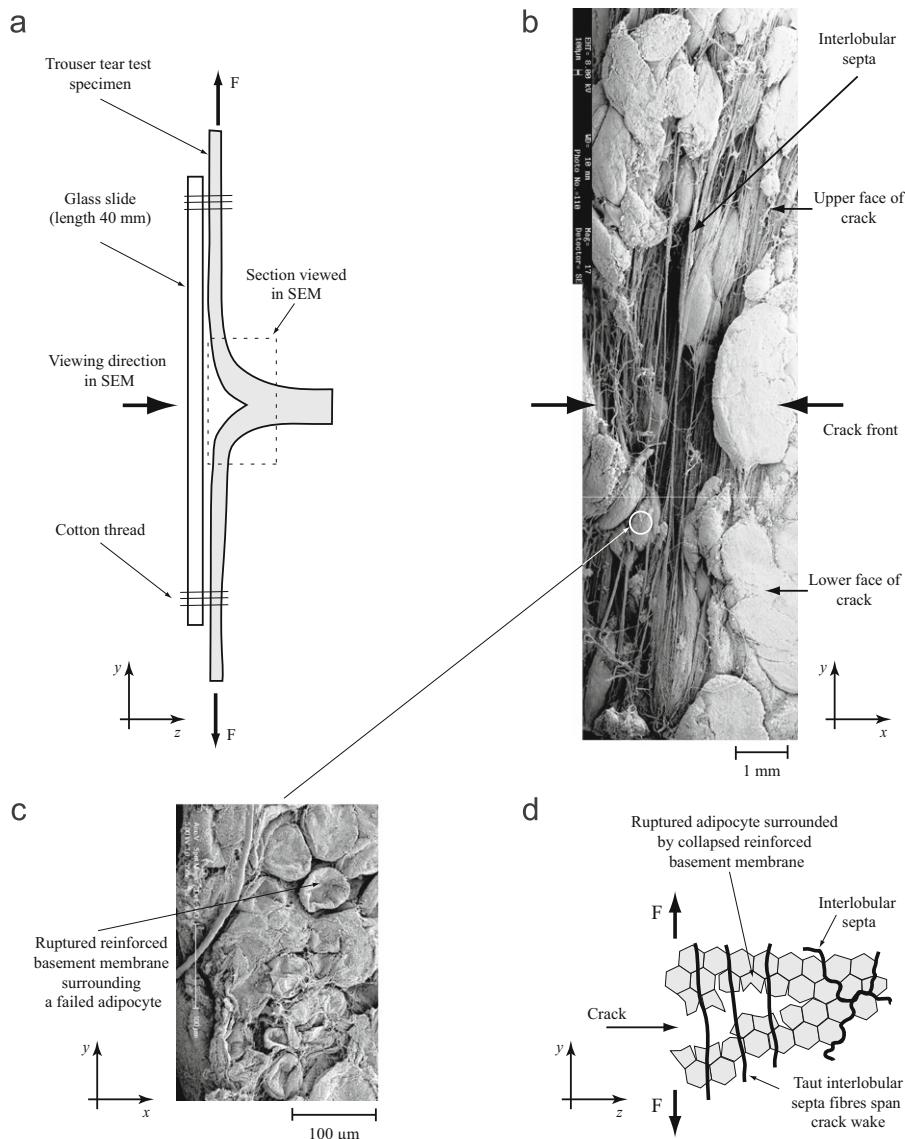


**Fig. 3.** Toughness  $J_c$  versus crack growth rate  $\dot{a}$  for adipose and dermal tissue. The average values of  $J_c$  are shown by solid lines.

membrane, reminiscent of inter-granular fracture of a polycrystalline solid (see Fig. 4d) and (iii) fractured septa fibres are loose and wavy whereas the intact fibres span the crack wake, obscuring the view of the crack tip.

#### 4. Discussion

The individual contributions to toughness from the septa fibres, acting as an open-cell foam and the reinforced basement membrane, acting as a closed-cell foam is now assessed. While it is recognised that there is a degree of interaction between these



**Fig. 4.** (a) The clamping method used during specimen fixing, (b) and (c) scanning electron micrographs, at two magnifications, of the crack front of adipose tissue fractured during a trouser tear test, (d) a sketch of the crack propagation through the reinforced basement membrane.

two collagen structures, the dominant contribution to toughness is assessed by considering the two limiting cases in turn.

It is recognised that the response of collagen fibres is non-linear viscoelastic (Geerligs et al., 2008b). However, Gutierrez-Lemini (2002) shows that the methods developed for calculating J-integral for an elastic body can be applied to viscoelastic solids since  $J_C$  is not dependent upon the heredity of the solid. Therefore, a first order estimate of the relative significance of these foams to the macroscopic toughness can be obtained from elastic fracture models. It will be shown that the main contribution to toughness is from the reinforced basement membrane, with a negligible contribution from the septa fibres.

First assume that the septa fibres resemble an open-cell foam of volume fraction  $\bar{\rho}_s$  made from walls of cell length  $l$  and fracture strength  $\sigma_f$ . The fracture toughness  $K_{IC}$  of a foam subjected to mode I loading is given by (Gibson and Ashby, 1997)

$$K_{IC} = 0.65\bar{\rho}_s^{3/2}\sigma_f\sqrt{\pi l} \quad (2)$$

Application of Eq. (2) to the mixed mode I & III trouser tear test would require a small adjustment to the pre-multiplier, which we

have ignored herein. The toughness of the septa follows immediately as

$$J_C = (K_{IC}^2/E_s^*) \quad (3)$$

Data for the tensile strength  $\sigma_f$  of individual septa fibres are not available. However, the tensile strength of the dermis provides a suitable estimate for the tensile strength of the septa fibres,  $\sigma_f \approx 10 \text{ MPa}$  (Ankersen et al., 1999). Upon taking  $\bar{\rho}_s = 3 \times 10^{-4}$  and  $l = 1 \text{ mm}$  the fracture toughness of the septa network is estimated to be  $K_{IC} = 1.89 \text{ N m}^{-3/2}$  via Eq. (2). The Young's modulus of a septa fibre is taken to equal that of a collagen fibre bundle in a tendon,  $E_s \approx 1 \text{ GPa}$  (Haut, 1983). This leads to an estimate of  $E_s^* = \bar{\rho}_s^2 E_s = 90 \text{ Pa}$  (Gibson and Ashby, 1997) for the Young's modulus of the septa open-cell foam. The toughness of the septa is then estimated to be  $J_C = 40 \text{ mJ m}^{-2}$ , via Eq. (3). This is substantially lower than the measured toughness of adipose tissue,  $J_C = 4.1 \text{ kJ m}^{-2}$ .

Second, we estimate the toughness contribution from the reinforced basement membrane, acting as a closed-cell foam. The toughness of a closed-cell foam  $J_C$  scales with the toughness

of the wall material  $J_{Cm}$ , according to the rule-of-mixtures formula  $J_c = \bar{\rho}_m J_{Cm}$ , where  $\bar{\rho}_m$  is the volume fraction of reinforced basement membrane. No independent measurements exist for  $J_{Cm}$ , and so the value  $J_{Cm} = 17 \text{ kJ m}^{-2}$  as recorded in Fig. 3 for the dermal tissue is adopted: the dermis is a dense mesh of type I collagen. An estimate for  $\bar{\rho}_m = 0.1$  is taken from Fig. 1a and similar unpublished images giving  $J_c = 1.8 \text{ kJ m}^{-2}$ . This is of similar magnitude to the measurement of toughness for adipose tissue,  $J_c = 4.1 \text{ kJ m}^{-2}$ .

## 5. Conclusions

The toughness of porcine adipose tissue and dermal tissue has been successfully measured using the trouser tear test. It is argued that the toughness of adipose tissue derives from that of the reinforced basement membrane, with a minor contribution from the interlobular septa fibres.

## Conflict of interest statement

All authors deny having any financial and personal relationships with other people or organizations that could inappropriately influence our work.

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