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The biomechanical properties of canine skin measured in situ by uniaxial extension

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ABSTRACT

Introduction: A uniaxial extension system was setup to analyze the mechanical properties of dog skin.
Material and methods: Pads were glued onto dog skin with constant reproducible geometrical parameters and the extension force was measured as a function of the extension values. Forty-one sites (82 cycling tests) were investigated in situ on 11 canine cadavers, half of them after surgically isolating the test area from the surrounding skin. Series of loading-unloading cycles of up to 5 N or 10 N or both loads were performed on each site. The elastic properties and the dissipative effects were characterized respectively by evaluating the secant Rigidity at maximum loads and the Fraction of dissipated energy.
Results: A hysteresis phenomenon, implying the need for preconditioning to attain equilibrium cycles, was apparent during mechanical characterization. Polynomial expressions were used to relate the measured Rigidities and the Fractions of dissipated energy with or without sample isolation. The latter were less affected by isolation. The ratios between the Rigidities at 5 N to those at 10 N displayed non-linearity in the investigated extension range in contrary to the Fractions of dissipated energy.
Discussion/conclusion: The parameters confirming the dissipative non-linear elastic behavior of dog skin were identified and the correlation between Rigidity and Fraction of dissipated energy on isolated and non-isolated skin samples was quantitatively determined. This extension setup can now be used as a "true in vivo" mapping tool to determine the mechanical characteristics of the skin during healing processes or during the study of Human skin disease with the dog as an animal model.

1. Introduction

Skin is a complex 3-layered structure, which acts as a physical barrier for the body. It is biomechanically described as a non-linear, pretensioned (Cox, 1941; Langer, 1978), viscoelastic material; and it is known to undergo spontaneous deformation, when harvested from a donor site (Cox, 1941; Delalleau et al., 2006; Diridollou et al., 2000; Dupuytren, 1836; Gąsior-Głogowska et al., 2013; Hendriks, 2001; Hendriks et al., 2003; Langer, 1861). Each layer of the skin is known to have a specific biomechanical role (Gąsior-Głogowska et al., 2013; Bhandal et al., 2012; Delalleau, 2007). The extensibility of the skin (i.e. value of the maximum deformation without rupture) is mainly dictated by the properties of the corneal layer of the epidermis and strongly impacted by its degree of humidity (Leveque and De Rigal, 1985). The anisotropy and tensile strength of the skin are considered to be due to type I collagen fibers, in the deeper layer of the dermis (Agache, 2000; Herbage and Wegrowski, 2004; Manschot et al., 1982; Gąsior-Głogowska et al., 2013; Langer, 1861). Elastin forms a concomitant fibrous network in the dermis and plays an important role in strain recovery of the skin after applying a stress but its contribution to the elasticity modulus is smaller (Delalleau, 2007). The re-orientation of collagen and elastin fibers leads to mechanical relaxation and energy dissipation of the skin after deformation (Silver et al., 2003; Wilkes et al., 1973).

The biomechanical properties of skin have been measured using both in vivo and in vitro methods. The in vitro methods are the simplest because the effect of the sample environment is eliminated and all the applied forces are well controlled. The in vivo tests, in contrast, are challenging because of the roles played by the other layers and underlying structures and because of the preexisting tension and the effect of the 'environment' of the test zone. In vivo test results have tended to be descriptive, to lack reproducibility, and only to provide qualitative estimates

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of the biomechanical properties (Berardesca et al., 1995; Elsner et al., 2002). Current skin measurement devices measure skin properties by applying deformation forces in various ways: traction, tension, suction, torsion or indentation (Agache, 2000; Barbenel and Payne, 1981; Delalleau et al., 2006; Diridollou et al., 2000; Draaijers et al., 2004; Edwards and Marks, 1995; Jansen and Rottier, 1957; Lim et al., 2008; Manschot and Brakkee, 1986a; Paller-Mattei et al., 2008; Reihsner et al., 1995). Some of these tests use multiaxial loads, however, which are difficult to interpret quantitatively and produce very different results from one to another. Uniaxial extension setups can be used to evaluate in-plane directional differences in material properties and can be non-invasive, applicable and easy to use in vivo. A uniaxial extension setup is obtained, in vivo, by attaching two pads to the skin and applying displacements. The force required to displace the pads, as the skin is extended in a test zone between the pads, is measured by a load cell (Baker et al., 1988; Evans and Siesennop, 1967; Gunner et al., 1979; Larrabee, 1986; Manschot and Brakkee, 1986a,b; Vescovo, 2002).

Characterization of the mechanical properties of dog skin would be important to allow its use as an animal model for Human skin disease. In fact, for example, the dog provides a unique spontaneous animal model for Recessive Dystrophic Epidermolysis bullosa (Palazzi et al., 2000) and can be used to test the long-term efficacy of ex vivo therapeutic approaches for this disease. Investigation of the biomechanical properties in such dogs could be beneficial particularly with regard to epidermal grafting and problems encountered during skin graft sampling and wound bed retraction (Gache et al., 2011). Examination of the biomechanical properties of dog skin would also be of interest in Veterinary medicine as regards the healing of canine skin particularly during application of bioactive wound dressings.

Interestingly, dog skin appears to be different from Human skin and from that of other mammals, not in its fundamental composition but in its structure (Scott et al., 2001). The reported average thickness of the skin of dogs, epidermis plus dermis, is 0.5 to 5 mm (millimeters) (Scott et al., 2001); human skin epidermis is on average 0.05 to 0.1 mm and the dermis, 0.5 to 5 mm (MacGrath and Uitto, 2010). Hair follicles are more numerous and different in composition and the subcutis is looser with a different vascularization compared to that of Human skin (MacGrath and Uitto, 2010; Scott et al., 2001). Dog skin seems generally more “movable” than human skin, based for example on visual skin folds, and its biomechanical properties should, in consequence, differ from those of other mammals.

To our knowledge, no ex vivo or in vivo study, characterizing the biomechanical properties of dog skin has been published, whereas numerous studies have been carried out on humans and on other experimental animal models. Thus, this study was designed to preliminary confirm the interest of a uniaxial extension setup to describe the elastic and dissipative mechanical properties of dog skin in situ on cadavers, in the high deformation range, for futures in vivo applications.

2. Materials and methods

2.1. Uniaxial extension setup

A schematic diagram of the system set-up is shown in Fig. 1a and b. It consists of a portable tensile test machine mounted with an extension setup constituted by two aluminum pads, glued onto the skin, guided by a stainless steel rod and a ball bushing. The tensile test micromachine is equipped with a direct current motor (Portscap 28L28-416E.48) powered by a voltage source and associated with a motor-reducer (Portscap R32.0301), which rotates a planetary roller screw (INA—Series RGT) in a mobile crosshead guided by self-aligning linear bearings. The load cell is attached to the immobile pad for the force measurement. The mobile pad is connected to an LVDT sensor (Sensorex®, ref. SX20MR100). The distance between the ridges of the two pads, corresponding to the test zone, is thus

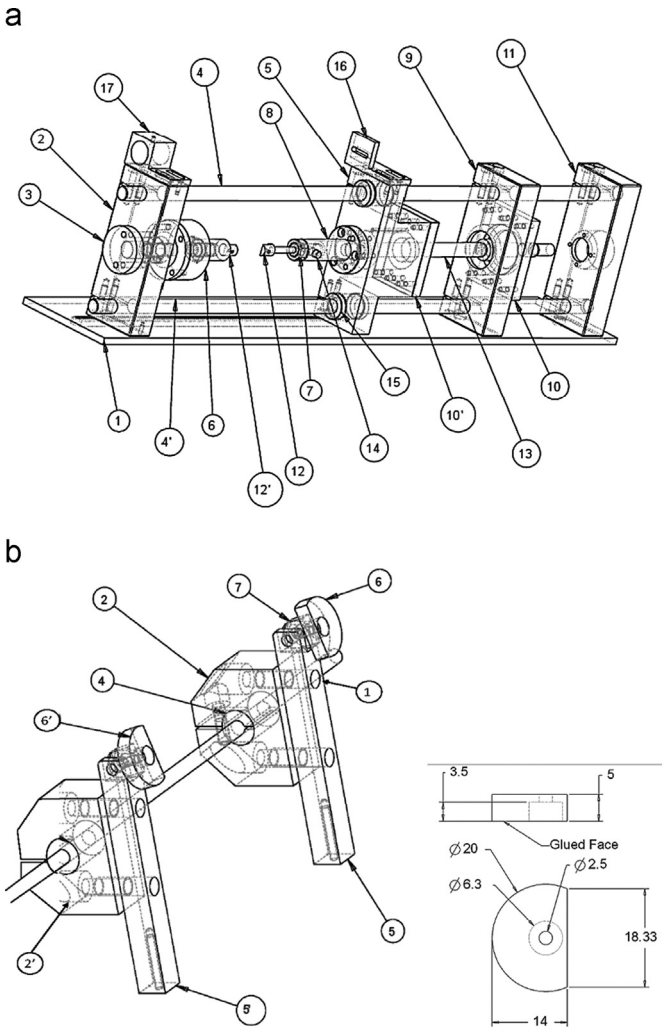

Fig. 1. (a) and (b) Schematic representation of the uniaxial extension set up used in this study. (a) 1: stand plate, 2: fixed rail, 3: flange strength, 4: axis, 5: mobile rail, 6: load cell, 7: counter-washer, 8: ball bushing, 9: fixed rail, 10: hood, 11: rail, 12: self-aligning linear bearings, 13: planetary roller screw, 14: keys, 15: ball socket, 16: LVTD support, 17: LVT support and (b) 1:axis, 2: shaft support, 4: jaws, 5: ring, 6: pad support, 7: pad).

imposed and measured, and the load cell measures the resulting force, disregarding the friction between the guiding rod and the linear ball bushing. A computer is connected to the system and permits the analog to digital conversion and acquisition of the load–displacement curves.

2.2. Test method

All the cadavers (11 dogs: 2 Bichons, 4 cross breeds, 1 cross breed Fox Terrier, 1 cross breed Griffon, 1 Fox Terrier) were frozen to −18 °C except for 2 Beagles which were euthanized for unrelated reasons just before the tests. All the frozen cadavers were thawed to 20 °C before each test.

Preparation of the skin was standardized. The fur was removed from all cadavers with the same clipper and using the same cutting height and the skin was degreased with diethyl ether. The pads were then glued onto the skin using cyanoacrylate glue (loctite® 401, Henkel), at the beginning of each test, and applied with a controlled force of compression (1 N (N) for 30 s (s). The compression force was measured with a force gauge apparatus (Newtonmeter PFI-200N) and the initial distance between the ridges of the two pads was set at 15 mm (mm). After gluing the two pads, the skin was tested to obtain the “non isolated” results. Then, without removing the pads, these results were compared with the results of tests obtained with the same setup after surgical isolation of the skin zone between the pads – tests hereafter labeled “isolated” – to evaluate the impact of the presence of the surrounding skin (‘environment’) on the test zone between the pads (Fig. 2). The act of isolating the skin removes the peripheral forces of the environment before and during measurement, thus partially mimicking an in vitro setting. It was important that the skin remained attached to the body (via the fascia) to allow