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Method article

Development of tensile strength methodology for murine skin wound healing

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ABSTRACT

In this study, a methodology was evaluated and improved to quickly measure the tensile strength of murine skin in a biomechanical assay for an incisional wound healing model. The aim was to streamline and enhance the wound model, skin specimen preparation, and tensile test so that large numbers of fresh tissue could be tested reliably and rapidly. Linear incisions of 25-mm length were made in the dorsal skin of mice along the spine and metallic staples were used to close the wound. After 20 days, the mice were sacrificed, and a square-shaped section of skin containing the linear incision was excised. Two metallic punches were fabricated and used to punch 15-mm long strips of skin of 2 mm width whose length was orthogonal to the direction of incision. The tensiometer configuration was modified to expedite tensile measurements on fresh skin, and load-to-failure was measured for each strip of skin from the cephalad to the caudal region. We evaluated sources of error in the animal model and the testing protocol and developed procedures to maximize speed and reproducibility in tensile strength measurements. This report provides guidance for efficient and reproducible tensile strength measurement of large numbers of skin specimens from freshly sacrificed animals.

- Tattoo placement to identify the two ends of the healing incisional wound assisted in decreasing error in the position and orientation of tensile strips.
- Custom-made punches to prepare skin strips for tensile testing helped conduct tensile tests of fresh tissue rapidly.
- Alteration of the manual grips of the tensile tester enabled specimens to be gripped rapidly to significantly accelerate testing for each skin strip.

ARTICLE INFO

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Specifications Table

Subject area	*Select one of the following subject areas:* • *Medicine and Dentistry*
More specific subject area	*Healing of skin wound*
Method name	*Tensile Testing for Murine Skin Wound*
Name and reference of original method	Gorodetsky, R., W.H. McBride, and H.R. Withers, *ASSAY OF RADIATION EFFECTS IN MOUSE SKIN AS EXPRESSED IN WOUND-HEALING.* Radiation Research, 1988. **116**(1): p. 135-144. Gorodetsky, R., et al., *EFFECT OF FIBROBLAST IMPLANTS ON WOUND-HEALING OF IRRADIATED SKIN - ASSAY OF WOUND STRENGTH AND QUANTITATIVE IMMUNOHISTOLOGY OF COLLAGEN.* Radiation Research, 1991. **125**(2): p. 181-186. Gorodetsky, R., et al., *RADIATION EFFECT IN MOUSE SKIN - DOSE FRACTIONATION AND WOUND-HEALING.* International Journal of Radiation Oncology Biology Physics, 1990. **18**(5): p. 1077-1081.
Resource availability	*N/A*

Methods detail

Our understanding of the complexity of skin wound healing has increased with the use of various, highly reproducible animal models that can address specific questions with quantitative outcome measures, especially the restoration of biomechanical properties. These complex processes involving inflammatory, fibrobastic, and maturation phases of healing have been extensively investigated in order to develop drugs and devices to treat wounds so that the post-wound skin remodeling results in a morphology that closely resembles that of the intact skin [1–6]. Biomechanical assays serve as useful techniques to determine the extent of skin wound healing and provide insight into the functionality of repaired skin compared with intact skin. Several types of biomechanical assays have been developed that measure the static tensile [7–15], biaxial tensile [16], and rheological properties [17–19] of skin to assess wound healing, aging, drug therapies, and biomaterials used for wound dressings. Among these biomechanical assays, the most common and simplest assay is the tensile test. This test often measures the tensile failure load or stress or the modulus (stiffness) of skin under different conditions. In the absence of standardized tensile test protocols, various investigators use different test specimen geometries and tensile stretching rates to measure tensile properties of skin. These test specimen geometries often depend on the animal model used and the region of the body of the skin being excised.

This study concerns skin incisions with primary closure to serve as an experimental model for controlled clinical surgical settings. This report concerns modifications made to expedite handling a large number of specimens to measure healing of incisional wounds in mice. Pilot studies revealed a lack of feasibility to process large numbers of samples with a standard tensiometer. To solve this limitation, elements of both a mouse skin injury model and of a protocol for measurement of tensile strength were modified.

Materials and methods

Micro-tensiometer

An eXpert 4000 micro-tester (Admet Inc, Norwood MA) with a frame capacity of 45 N and a spatial resolution of 0.0046 mm was used to measure tensile strength of strips of murine skin. The micro-tester was equipped with a 10 N and 50 N load cell and capable of crosshead speeds of up to 500 mm/min and a maximum displacement of 25-mm. The 10 N load cell was used for these studies. The tensiometer has two serrated manual screw-action clamps, each with two screws to secure specimens during tensile deformation. In order to decrease the time required to mount and dismount specimens, the screws of the clamps were set aside and two other clamping approaches were tested: we tried