Acta Biomaterialia 48 (2017) 341–356

Contents lists available at ScienceDirect

Acta Biomaterialia

journal homepage: www.elsevier.com/locate/actabiomat

Full length article

The hyperelastic and failure behaviors of skin in relation to the dynamic application of microscopic penetrators in a murine model

Stefano C. Meliga [a], Jacob W. Coffey [a,b], Michael L. Crichton [a,b], Christopher Flaim [c], Martin Veidt [d], Mark A.F. Kendall [a,b,e,*]

[a] The University of Queensland, Delivery of Drugs and Genes Group (D2G2), The Australian Institute for Bioengineering and Nanotechnology, St Lucia, QLD 4072, Australia
[b] ARC Centre of Excellence in Convergent Bio-Nano Science and Technology, The University of Queensland, Australia
[c] Vaxxas Pty Ltd, TRI, 37 Kent St, Woolloongabba QLD 4102, Australia
[d] School of Mechanical and Mining Engineering, Frank White Building 43, The University of Queensland, Brisbane, QLD 4072, Australia
[e] The University of Queensland, Faculty of Medicine and Biomedical Sciences, Royal Brisbane and Women's Hospital, Herston, Queensland 4006, Australia

ARTICLE INFO

Article history:
Received 8 April 2016
Received in revised form 25 September 2016
Accepted 12 October 2016
Available online 13 October 2016

Keywords:
Skin
Penetration
Microprojection arrays
Nanopatch
Subcutaneous backing
Strain-rate
Hyperelastic

ABSTRACT

In-depth understanding of skin elastic and rupture behavior is fundamental to enable next-generation biomedical devices to directly access areas rich in cells and biomolecules. However, the paucity of skin mechanical characterization and lack of established fracture models limits their rational design. We present an experimental and numerical study of skin mechanics during dynamic interaction with individual and arrays of micro-penetrators. Initially, micro-indentation of individual skin strata revealed hyperelastic moduli were dramatically rate-dependent, enabling extrapolation of stiffness properties at high velocity regimes (>1 ms^{-1}). A layered finite-element model satisfactorily predicted the penetration of micro-penetrators using characteristic fracture energies (~10 pJ μm^{-2}) significantly lower than previously reported (≫100 pJ μm^{-2}). Interestingly, with our standard application conditions (~2 ms^{-1}, 35 g piston mass), ~95% of the application kinetic energy was transferred to the backing support rather than the skin ~5% (murine ear model). At higher velocities (~10 ms^{-1}) strain energy accumulated in the top skin layers, initiating fracture before stress waves transmitted deformation to the backing material, increasing energy transfer efficiency to 55%. Thus, the tools developed provide guidelines to rationally engineer skin penetrators to increase depth targeting consistency and payload delivery across patients whilst minimizing penetration energy to control skin inflammation, tolerability and acceptability.

Statement of Significance

The mechanics of skin penetration by dynamically-applied microscopic tips is investigated using a combined experimental-computational approach. A FE model of skin is parameterized using indentation tests and a ductile-failure implementation validated against penetration assays. The simulations shed light on skin elastic and fracture properties, and elucidate the interaction with microprojection arrays for vaccine delivery allowing rational design of next-generation devices.

1. Introduction

Next-generation healthcare increasingly relies on minimally-invasive biomedical devices capable of negotiating skin mechanical properties to mediate intracutaneous and transcutaneous tasks like administering therapeutics [1,2], extracting diagnostic biomarkers [3–5], and performing surgical procedures [6,7]. For instance, epidermal and dermal targeted delivery of vaccines is a promising approach for increasing global vaccine coverage, due to the ease of access and unique immunological properties of the skin [8–11]. Passive permeation of the antigen is impractical due to the large molecular size of most antigens [12,13]; hence, the payload is actively transported to the viable-cell strata by mechanically breaching through the skin's outer barriers (Fig. 1a). This is typically achieved by either: 1) high-pressure jet injectors that fire

* Corresponding author at: The University of Queensland, Delivery of Drugs and Genes Group (D2G2), The Australian Institute for Bioengineering and Nanotechnology, St Lucia, QLD 4072, Australia.
E-mail address: m.kendall@uq.edu.au (M.A.F. Kendall).

http://dx.doi.org/10.1016/j.actbio.2016.10.021
1742-7061/

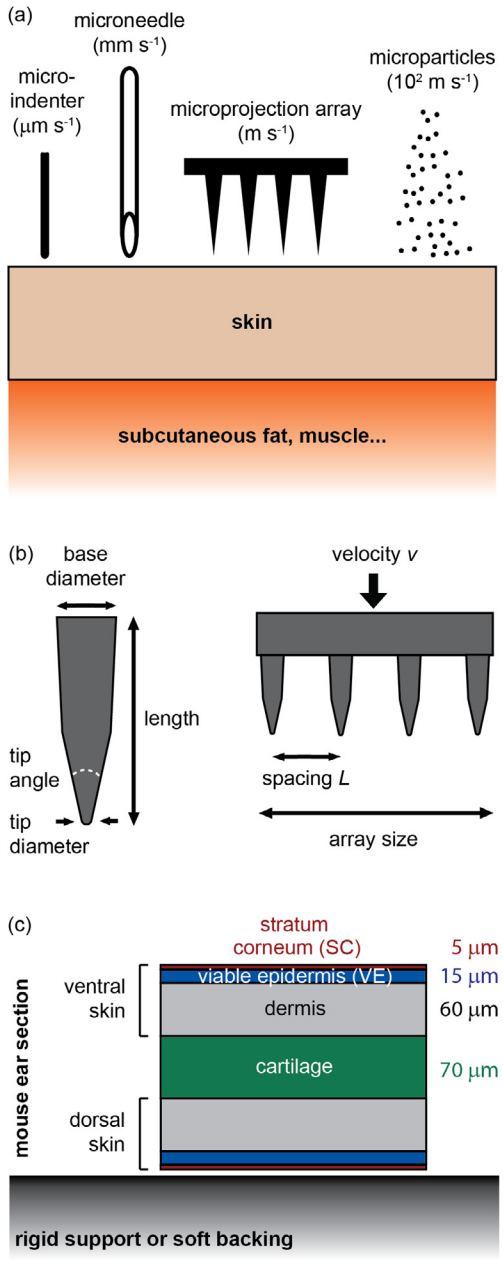

Fig. 1. a) Out-of-plane mechanical properties of skin at the microscale are typically measured *ex vivo* using indentation (e.g. AFM) at velocities up to ~100 μm s^{-1}; however, vaccines are delivered *in vivo* across the skin superficial barriers using penetrators i.e. microneedles or microprojections arrays applied (by hand or impact applicators) at velocities ≫mm s^{-1}; strain-rate effects and subcutaneous layers play and important mechanical role during skin penetration. b) Schematics of design specifications for individual and arrays of penetrators (e.g. microneedles/microprojections). c) Mouse ear section and skin layer thicknesses considered in this work.

the payload in liquid or powder form (microparticles) [14–16]; or 2) penetrator tips that deposit payload through a channel in the skin (e.g. intradermal needles and hollow microneedles) [1,16–19], or embed the payload in a matrix/coating that dissolves in the skin (e.g. dissolvable/coated microneedle and microprojection arrays [1,16,18,20–24]). Studies have reported improved immune responses compared to standard syringe injection [2,11,16,25,26]. The mechanisms underlying the low-dose efficacy or increased potency however, are not yet fully understood [27,28], which limits our ability to fully realize the potential of cutaneous vaccination.

Precise penetration to the desired depth for vaccine uptake by sitespecific cells has led to potent immune responses [2,22,29]. Achieving this relies upon negotiating the unique elastic and failure properties of the skin – a multilayer composite material. However, while there have been some mechanical characterizations [30–35] and underlying linear and non-linear elastic models [36–39], there is still a paucity of investigations focusing on skin elastic and failure behaviors in mechanical conditions relevant for epidermal and dermal delivery of vaccines. In addition to the skin's intrinsic structural complexity, complicating factors include variation between species (e.g. mouse vs. human [40,41]), individuals (ethnicity [42], gender [41,43]) and within individuals (age [41,44,45], body site [45,46]). Firstly, assumptions of skin homogeneity and isotropicity result in different elastic moduli depending on the loading mode. In this case, out-of-plane indentation tests should only be considered because they best capture the skin response during penetration. Such tests have yielded Young's moduli between 3 kPa and 30 MPa [39,47–50], in contrast to 2–100 MPa for in-plane dilatation tests [41,43,44,46,51,52] and 4850 kPa for shear tests [32,33,53,54]. Secondly, the Young's moduli extrapolated from indentations show a marked inverse dependence with the probe diameter [50]. For example, microscopic tips of 1–40 μm in diameter (i.e. relevant for microdevice penetration) have yielded 1–30 MPa [37,50], in contrast to 3–25 kPa for millimeter-sized tips [47–49]. Thirdly, although literature on skin viscoelasticity [34,37,40,44,49,50,55–57] demonstrates the rate-dependence of skin elasticity, we could not find any published out-ofplane tests where the load was applied at velocities >1 ms^{-1} or strain rates >1 μs^{-1}, most relevant for microdevice penetration.

On the other hand, while underlying linearelastic and hyperelastic descriptions are corroborated by empirical data [37–39], the skin lacks established constitutive models of failure. Skin penetration by individual needles has typically been described using either of the following two approaches: 1) *stress-based failure criteria* which extend the traditional yield criteria (e.g. von Mises yield criterion [58,59]) such that the skin fails when the stress (typically the von Mises component) exceeds a threshold strength [60–63] (this does not account for the irrecoverable energy dissipated into material damage and, thus, is not well suited to model the penetration achieved from a given velocity); or 2) *energybased fracture propagation* which extends the concept of fracture toughness [64,65] to ductile materials, i.e. an energy per unit area representing the cost to create crack interfaces [43,66–69]; this model, though, does not specify if an initial notch forms at all (failure initiation), how the crack propagates (e.g. direction and speed), and what fraction of the penetrator energy is utilized in the fracture (as opposed to being elastically stored or dissipated in viscous or plastic phenomena). The prediction of skin penetration thus requires a complete description of the spatial stress-strain distributions to detect the instant and coordinates of failure initiation, and the energy repartition among various reversible and irreversible phenomena.

In summary, the limited understanding of skin elastic response to high strain rates, mechanisms of failure and fracture, and interaction with multiple penetrators have limited the scope of devices for achieving precise epidermal and dermal drug/vaccine targeting. Our case study is the Nanopatch™, a densely-arranged array (≫1000 cm^{-2}) of microprojections, (solid cone-like structures measuring ~100 μm in length [70], Fig. 1b). Precise and consistent targeting of specific strata is crucial to elicit immune responses and is achieved by applying the array against the skin at controlled velocities (~1 ms^{-1}) [71,72]. An indepth understanding of the skin mechanical interaction with microneedles/microprojections will allow further tailoring of the array design and optimization of application conditions to achieve customized antigen placement and highly-immunogenic configurations. This knowledge is critical