Comparison of Human, Porcine and Rodent Wound Healing With New Miniature Swine Study Data

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ABSTRACT

Accidental wounds are one of the most common reasons for human visits to hospital emergency services. Healthy individuals normally heal induced skin defects in a reasonable fashion if complications, such as infections, can be avoided. Aged individuals or diabetics frequently suffer delayed wound healing. Wound researchers require efficient animal models which are predictive of human responses. Pig skin is anatomically, physiologically, biochemically and immunologically similar to human skin, and the skin is 'fixed skin' like humans and unlike rodents or rabbits. Loose skin allows accelerated closure of surgically induced rodent wounds by primary contraction unlike the normal primary response of re-epithelialization in swine and humans. Pig skin mirrors human skin in having a sparse haircoat, a relatively thick epidermis, similar epidermal turnover kinetics, lipid composition and carbohydrate biochemistry, lipid biophysical properties, and a similar arrangement of dermal collagen and elastic fibers. In our studies, healthy juvenile miniature swine wound re-epithelialization progressed relatively quickly (average 0.109 mm/hr at d19) while geriatrics progressed more slowly (average 0.048 mm/hr at d32). Untreated adult diabetic Yucatan miniature swine wound re-epithelialization rates were 88% of conventional non-diabetic (control) Yucatan models on d29 post wounding. Postulations for this result will be offered. Control juvenile or young adult Sinclair or Yucatan miniature swine dermal wounds heal at faster rates (re-epithelialization rate) than those of geriatric or diabetic animals. The healing rate differential can be as high as 2:1 for juveniles over geriatrics. Porcine or miniature swine models offer significant advantages and have a record of predicting treatment modalities in human over models with loose skin. Miniature swine models can provide useful efficacy data for novel cutaneous therapy products.

OVERALL RESEARCH OBJECTIVES

The purposes of this study were to:

1. Compare human, porcine and rodent wound healing looking for unique traits which could influence healing rates in an animal model system.
2. Measure Yucatan and Sinclair miniature swine full-thickness dermal healing rates following surgical wounding.

HYPOTHESIS

1. That apparent advantages and disadvantages or unique traits exists for various animal models used in wound healing studies.
2. That diabetic miniature swine full-thickness dermal wound healing rates will be considerably less than non-diabetic and geriatric rates will be less than juvenile animals.

SPECIFIC AIMS

1. Perform literature search on wound healing in humans and animal models of interest.
2. Perform wound healing studies on normal vs diabetic Yucatan and on juvenile vs geriatric Sinclair miniature swine. Assess re-epithelialization rates in each group.

EXPERIMENTAL DESIGN

1. Literature references, papers and databases were screened for appropriate comparison parameters and a summary table constructed (Table 1). Parameters of interest included: skin thickness, strateum corneum thickness, presence of fixed or loose skin, haircoat density, melanin content of epidermis, presence and distribution of eccrine or apocrine glands in skin, epidermal cellular turnover rate, dermal vascularization, skin blood flow, skin pH, presence of delayed healing for diabetic wounds, adult body mass, skin surface area/unit mass, primary wound healing pattern, and wound healing time-course. Porcine re-epitheliazation rates were determined from the literature (Table 2).
2. Full-thickness wound studies were performed in juvenile and geriatric Sinclair miniature swine and in diabetic and control adult Yucatan miniature swine and rates of re-epithelialization calculated and compared (Table 3).

EXPERIMENTAL RESULTS

Table 1: Dermatology and Wound Healing Models: Human vs Porcine vs Rodent

Parameter	Human	Porcine (Domestic)	Std. Yucatan MS	Rat	Mouse
Skin Thickness	2-3 mm	1.5-2 mm	1.5-2 mm	1-2 mm	Very thin (0.4-1 mm)
Epidermal thickness	Relatively thick 70 (50-100) µm, 2.67 layers	Relatively thick 52-100 µm, 3.94 layers	Relatively thick: 50-100 µm, 4-5 epithelial sub layers	21.7 µm, 1.83 layers	9.4-13.3 µm, 1.75 layers
Stratum corneum thickness	10-12.05 µm	12.28 µm	10-20 µm	5 µm	2.9 µm
Fixed Skin	Yes	Yes	Yes	No	No
Hair Coat	Sparse, 11 hairs/cm²	Sparse, 11 hairs/cm²	Sparse 'hairless', 11-25 hairs/cm²	Thick, 289 hairs/cm²	Thick, 658 hairs/cm²
Substantial Melanin in Epidermis	Yes	No for Yorkshire	Yes some, Grey color skin	Only in darkly pigmented strains	Only in darkly pigmented strains
Eccrine Sweat Glands	Yes	Yes, snout, lips, carpal gland only	Yes, snout, lips, carpal gland only	Yes, paws	Yes, paws
Aprocrine Sweat glands	Yes	Yes	Yes	ua*	ua
Epithelial Cellular turnover rate	28d	28d	28-30d	ua	ua
Dermal vascularization	ua	Less than human	Less than human	ua	ua
Skin blood flow rate, back (ml/min/100g)	3.12	3.0	3.0	9.6	20.6
pH of skin	5	6-7	6-7	ua	ua
Delayed diabetic wounds	Yes	Yes	Yes	ua	ua
Adult Body mass	70 kg	150 kg	50-70 kg	250-400g	25-35g
Skin surface area/unit mass	ua	Comparable to man	Comparable to man	High	Highest
Primary Wound Healing Pattern	Re-epithelialization	Re-epithelialization	Re-epithelialization	Contraction	Contraction
Wound Healing Timecourse	7-14d or longer depending upon wound size	12-14d or longer depending upon wound size	12-14d or longer depending upon wound size	Closes in just few days due to contraction of skin	Closes in just few days due to contraction of skin

**ua = unavailable or not found*

Table 2: Literature Cited Studies: Miniature Swine & Domestic Pig Re-epithelialization Rates* by Microscopic Observation

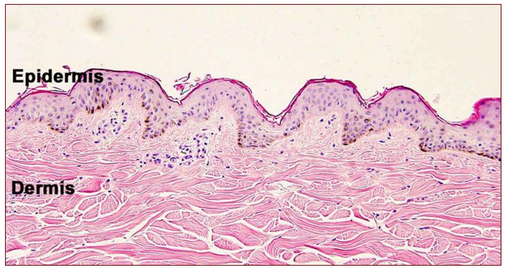
Strain Pig	Status / Sex / N Animals	Age / Weight	Type Dermal Wound	Post-Wounding Measurement Timepoint	Re-epithelialization Rate (mm/hr)	Reference
Yucatan Minitaure Swine	Normal / Female/16	6 mths/20-25 kg	Split-thickness	96 hr	0.057	Chvapil & Chvapil[1], 1992
Yorkshire	Normal / Female/16	2-3 mths/30-45 lb	Split-thickness	54 hr	0.101	Chvapil & Chvapil[1], 1992
Yorkshire	Normal / Female/4	60 kg	Full-thickness	d12	0.050	Velander et al.[2], 2008
Yorkshire	Diabetic/ Female/3	60 kg	Full-thickness	d12**	0.021	Velander et al.[2], 2008

**Rates per hour calculated from data presented; **Full re-epithelialization was delayed in diabetic pigs until d18, while non-diabetics completed this phase in 12-14d*

Table 3: Sinclair Research Center Studies: Miniature swine Re-epithelialization Rates* by Microscopic and Macroscopic Observation

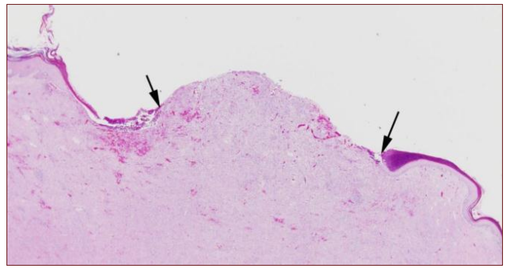
Strain MS	Status / Sex / N Animals	Age / Weight	Type Dermal Wound	Post-Wounding Measurement Timepoint	Re-epithelialization Rate (mm/hr)	Reference
Yucatan	Normal Castrate/ male/2	1-2 yr, 54.3-57.5 kg	Full-thickness	d29	**0.072****	Internal Data, SRC 2008
Yucatan	Diabetic Castrate/ Male/4	1-2 yr, 41.9-53.1 kg	Full-thickness	d29	**0.064****	Internal Data, SRC 2008
Sinclair	Normal/ Female/5	9-10.8 yr Geriatric adult, 76.9-93.5 kg	Full-thickness	d32	**0.048*****	Internal Data, SRC 2008
Sinclair	Normal/ Female/5	0.5 yr Juvenile/Young Adult, 14.8-20.9 kg	Full-thickness	d19	**0.109*****	Internal Data, SRC 2008

**Rates per hour calculated from SRC research data; **microscopic observations; ***macroscopic observations*



◀ **Figure 1**
Histological section of normal Yucatan miniature swine skin (H&E X 100)

Figure 2 ▶
Histological section of chronically wounded diabetic Yucatan pig skin with loss of epithelial integrity (between arrows), extensive fibrosis and lack of chronic active inflammation (H&E X 100)



DISCUSSION

Literature Review on Models

In 2001, Sullivan et al. evaluated 25 wound treatments and showed that the porcine model results was predictive with human study results 78% of the time.[3] The anatomy and physiology of the cutaneous blood supply and the wound healing characteristics have made the miniature pig a standard model for plastic surgical and wound healing studies.[4,5] Besides the anatomic similarities, swine are equivalent to primates for percutaneous absorption studies and have similar lipid biophysical properties, epidermal turnover kinetics and carbohydrate metabolism in the skin. [6] Pigs have fixed skin like humans and heal primarily by re-epithelialization rather than contraction.[12] For our research, Yucatan (Mini or Micro) or Sinclair miniature swine have found the greatest applications for wound healing. Fewer animals are required when miniature swine models are used for wound healing experiments because of the potential number of wound treatment sites per animal. Each animal can serve as its own internal control, as long as the drug effect is localized to dermis and not systemic. Many 1.5 cm square wounds (16-20, up to 24) can be placed on an adult MS dorsum without difficulty. Swine tolerate the wound modeling incredibly well with the use of central acting (opioids) based on food intake, behavior, and tolerance of wound handling. The ability to perform paired wound healing measurements reduces the effect of inter-animal variability and permits more robust paired statistical tests.[7] Unlike rodents, pigs possess sufficient body weight to tolerate prolonged topical administration of potent drugs.[8] Bellinger et al. (2006) have stated "Pigs have great potential as a relevant animal; model of insulin-resistant type 2 DM to identify mechanisms that lead to the development of diabetic complications and to develop and test novel therapeutic compounds."[9] Pigs can serve as models of delayed wound healing associated with the diabetic state. Although dorsal pig skin does not contain eccrine sweat glands like human skin, the porcine model of partial-thickness skin wound healing has been validated and appears to predict effectively the effects of novel treatments in humans.[7,10] Hartwell (1955) determined that porcine skin is anatomically and physiologically more similar to human skin than is skin from small laboratory animals.[11]

Why So Little Difference Between SRC Diabetic and Control Re-epithelialization Rates?

1.Insulin therapy level was not restricted leading to nearly normal (88% of normal) wound healing in diabetic miniature swine.

2.Control conventional miniature swine rate of epithelial closure was delayed by low grade infections.

3.The diabetic status of the pigs had not become chronic and/or the age of the pig was too young to fully model geriatric diabetic or chronic diabetic wounds.

CONCLUSION

1.The miniature swine model is a predictive wound healing surrogate for human wound healing research. Putative treatment drugs or modalities can be screened in these models.

2.Control juvenile or young adult Sinclair or Yucatan miniature swine dermal wounds heal at faster rates (re-epithelialization rate) than those of geriatric or diabetic animals. The healing rate differential can be as high as 2:1 for juveniles over geriatrics. The re-epithelialization rate of wounds on adult diabetic Yucatan miniature swine was 88% of that of wounds on normal Yucatans.

3.Porcine or miniature swine models offer significant advantages and have a record of predicting treatment modalities in human over models with loose skin.

4.Re-epithelialization is one of four major parts of wound healing and is a critical phase.

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