A COMPARATIVE EVALUATION OF PORCINE AND BUFFALO SKIN AS POTENTIAL SOURCE TISSUES FOR DEVELOPMENT OF DERMAL EXTRACELLUALR MATRIX SCAFFOLDS

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Abstract

Raw porcine skin from 6-8 months Large White Yorkshire (LWY) pigs and 18-24 month old buffaloes were evaluated. The buffalo dermis from 18-24 months age group may undergo rapid enzymatic degradation when implanted *in vivo.* Unless exogenously stabilised, the buffalo skin may be a less desirable material for scaffold development. Though the total collagen content and thickness of buffalo skin was significantly higher than porcine kin, buffalo skin had asignificantly higher ASC and NSC contents which are indicative of fewer natural cross-links and susceptibility to enzymatic degradation. But these observations should be judged with caution because the properties of the scaffold can alter with subsequent treatments like cross-linking and sterilization.

Key words: *Extracellular matrix scaffolds, pig skin, buffalo skin*

Biological scaffold materials composed of extracellular matrix (ECM) have been developed to facilitate the constructive remodeling of many different tissues in preclinical animal studies and human clinical applications. A number of collagen-based bioscaffolds for use as dermal substitutes are available in the market either in the form of ECM scaffolds or reconstituted collagen scaffolds. These products are relatively expensive and are not affordable to the common man. Many of the ECM scaffolds are prepared from xenogenic source tissues, and most of these tissues are abattoir co-products which are currently underutilized.

Reing*et al.* (2010) stated that the use of mammalian extracellular matrix (ECM) as surgical mesh materials and as scaffolds for regenerative medicine applications was in common place. These tissues included dermis, pericardium, small intestine and urinary bladder. These tissues were harvested from different species including pig, cow, horse and human.

Rodrigues *et al.* (2010) stated that tissues of animal origin have been used for thousands of years to cover extensive skin wounds. Porcine skin was one of the heterologous materials under study, mainly on account of its high similarity to human skin and its biocompatibility, mechanical resistance and low antigenicity. Buffalo hide is an abundant byproduct in the slaughter industry which

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can be effectively utilized for the scaffold preparation. The scaffold preparation from porcine skin has been well studied. Hence the objective of the study was to compare various physico-biochemical properties of buffalo hide and porcine skin to assess their suitability for scaffold preparation.

Materials and Methods

Six Large White Yorkshire pigs (LWY) of 6-8 moths age and six male buffaloes of 18-24 months were randomly selected and brought from farms of KVASU and slaughtered hygienically at Meat Technology Unit. Skinfrom the dorso lateral region of slaughtered pigs and buffaloes was collected as approximately 12 X 15 cm pieces. The hairs were shaved off and the subcutaneous fat and muscular tissue were removed. The total yield of the skin was also noted for both pigs and buffaloes. Various attributes of porcine and buffalo skin were analysed as mentioned below.

1. Thickness of skin/hide
2. Total yield of skin as percentage of live weight
3. Chemical composition, viz. moisture, fat and total protein(AOAC, 2010 and Starcher, 2001)
4. Total collagen content(Stegman and Stadler, 1967)
5. Neutral salt soluble and acid soluble collagen (Reddy et al., 2002)
6. Resistance to *in vitro* enzymatic degradation

Thin strips of dermal layer, approximately measuring 0.5 cm x 0.5 cm, were separated from porcine and buffalo skin, using a BP blade and these dermal strips were used.

Results and Discussion

The skin samples from the DL location of 6-8 months old LWY and 18-24 months old buffaloes were compared to evaluate the differences between these animal groups with respect to different characteristics in order to select the best raw material on the basis of the characteristics studied. The mean thickness of different layers of skin from LWY pigs and buffaloes is presented in Table 1. The total skin thickness of buffalo skin was 6060 ± 0.95 µm which was significantly ($p<0.05$) higher than LWY skin. The total yield of skin/hide from LWY pigs and buffaloes is presented in Table 1. Hide yield from buffaloes (12.72 ± 1.87 per cent) was significantly lower than the thickness of LWY porcine skin.

Thickness of different layers of skin

The thickness of different layers of LWY and buffalo skin (Table 1) was measured using histological sections and measuring microscopically with an ocular micrometer. The buffalo skin had a significantly thicker reticular dermal layer (4334.0 ± 134.0 µm) whereas papillary layer was significantly higher ($p<0.05$) for LWY pigs.

Chemical Composition

The chemical composition of skin from LWY and buffalo is presented in Table 2.LWY pig skin had significantly lower moisture content (46.39 ± 1.26 per cent) thanbuffalo skin. Fat percentage was significantly higher in the LWY pig skin when compared to the buffalo skin.

Total Collagen Content

The total collagen content of full thickness LWY and buffalo skin is presented in Table 3. The collagen content was significantly different between the two animal groups. The buffalo skin had significantly higher total collagen content (87.81 ± 1.43 per cent).

Neutral Salt Soluble and Acid Soluble Collagen

The neutral salt soluble collagen (NSC) and acid soluble collagen (ASC) content of LWY and buffalo are presented in Table 3. Buffalo skin has the highest NSC and ASC.

Enzymatic Sensitivity

The enzymatic sensitivity of LWY and buffalo dermis measured as per cent weight loss after collagenase degradation is presented in Table 6. There was a significantly ($p<0.05$) higher weight loss in buffalo dermis compared to the LWY dermis and there was almost 100 per cent degradation of buffalo dermis after 48 hours of collagenase digestion.

Table 1. Yield of skin/hide and thickness of different layers of skin of LWY and Buffalo, µm

Animal Group	Yield	Total skin thickness	Epidermis	Papillary layer	Reticular layer
LWY	15.25±2.46[a]	3860±0.29b	43.860±7.74[b]	167.700±38.70a	2030.460±424.70[b]
Buffalo	12.72±1.87[b]	6060±0.95a	87.000±9.00[a]	97.000±11.00[ab]	4334.000±134.00[a]

Means bearing same superscripts in the column do not indicate significant difference (P<0.05)
LWY-Large White Yorkshire pig

Table 2. Chemical composition of LWY and Buffalo skin

Animal Group	% chemical composition		
	Moisture	Fat	Total Protein (% dry fat free basis)
LWY	46.39±1.26[b]	9.54±1.15[a]	85.650.84[b]
Buffalo	62.37±1.0[a]	2.620.38[c]	89.551.02[b]

Means bearing same superscripts in the column do not indicate significant difference (P<0.05)LWY-Large White Yorkshire pig

Table 3. Collagen characteristics LWY and Buffalo skin

Animal group	Total Collagen (% dry fat free basis)	Neutral salt soluble collagen	Acid soluble collagen
		(% of total collagen)	
LWY	82.26±1.43[b]	.00333±0.0003[b]	.00847±0.0003[b]
Buffalo	87.81±1.1[a]	.07400±0.0035[a]	.06280±0.0093[a]

Means bearing same superscripts in the column do not indicate significant difference (P<0.05)
LWY-Large White Yorkshire pig

Table 4 . Weight loss of LWY and Buffalo dermis after enzymatic degradation

Animal group	Percent weight loss after incubation for		
	24 hrs	48 hrs	72 hrs
LWY	28.77±4.51[f]	56.33±1.23[d]	68.67±2.93[b]
Buffalo	99.75±0.04[a]	100±0.00[a]	100±0.00[a]

Means bearing same superscripts in the column do not indicate significant difference (P<0.05)
LWY-Large White Yorkshire pig

From the results, it is inferred that the buffalo dermis from 18-24 months age group may undergo rapid enzymatic degradation when implanted *in vivo.* Unless exogenously stabilised, the buffalo skin may be a less desirable material for scaffold development. Though the total collagen content and thickness of buffalo skin was significantly higher than the other group, buffalo skin had a significantly higher ASC and NSC contents which are indicative of fewer natural cross links in the buffalo skin. The final quality of the finished material could change depending on subsequent processing treatments.

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