



# HISTOPATHOLOGICAL CHANGES IN HEALING OF CUTANEOUS WOUNDS TREATED WITH BOVINE COLLAGEN AND BOVINE COLLAGEN-ALGINATE DRESSING IN DOGS

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## Abstract

*Twelve dogs with full thickness cutaneous wounds such as dog bitten wounds, traumatic accident wounds and chronic wounds were selected and randomly divided into two groups. In Group I, a pre-sterilized bovine collagen-alginate sheet and in Group II, a pre-sterilized bovine collagen sheet were applied as wound dressing. Re-application of dressing was continued at 48 hours intervals for a two more times and afterwards weekly bandaging was followed. The animals were observed for a period of 28 days and a cutaneous punch biopsy from the mid-portion of healing wound was taken on day 28 for histopathological examination to evaluate the progress of wound healing. The results of the study revealed that both bovine collagen and bovine collagen-alginate had good graft acceptance, with no adverse reactions and helped in the augmentation of wound healing process.*

**Key words:** Bovine collagen, collagen-alginate, wound healing, dogs.

The launch of natural biopolymers such as collagen, chitin, alginate as wound dressings have transformed the world of regenerative medicine and art of wound care by enhanced healing process. Collagen, being the most abundant protein in the body and a key component of extracellular matrix, play a pivotal role in all phases of wound healing when applied to wound surfaces (Boateng *et al.*, 2008). On the other hand, alginate has additional haemostatic and adherent properties (Sezer and Cevher, 2011). Reports on the use of collagen in clinical veterinary wounds are plenty; however a clinical evaluation with the use of collagen-alginate is scanty. Hence, the present study was undertaken to evaluate the tissue response to full-thickness cutaneous wound healing on application of bovine collagen and bovine collagen-alginate sheets as wound dressing in dogs.

## Materials and Methods

The study was conducted in twelve systemically stable dogs presented to the

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Veterinary hospitals of Kerala Veterinary and Animal Sciences University at Mannuthy and Kokkalai, with full thickness cutaneous wounds. The animals were randomly divided into two groups of six each viz., Group I and Group II. Thorough debridement of the wound, making the wound edges even and irrigation with sterile normal saline were performed under general anaesthesia. In dogs of Group I, pre-sterilised bovine collagen-alginate sheet was applied on the surface of the wound in desired shape and fixed to the edges of the skin using a series of simple interrupted sutures with 1-0 polyglactin 910 suture. Similarly, in Group II, pre-sterilised bovine collagen sheet was applied. Re-application of the material was continued at 48 hours interval twice after which bandaging alone was employed.

The dogs were observed for a period of 28 days. A cutaneous punch biopsy sample of the mid-portion of the biomaterial implanted portion of the healing wound from four dogs in Group I and three dogs in Group II were collected for histopathological studies on 28<sup>th</sup> day. The formalin fixed tissue samples were sectioned and stained with routine Haematoxylin and Eosin stain for histopathological evaluation of wound healing. The parameters studied were necrotic and degenerative changes, extent of neovascularisation, fibrosis, fatty infiltration and the deposition of collagen. The inflammatory response was assessed by observing the presence of polymorphonuclear leucocytes, lymphocytes, macrophages, plasma cells and giant cells.

## Results and Discussion

Complete epithelialization was noticed in three out of four dogs in Group I and one out of three dogs in Group II. The dermis showed fluctuating levels of inflammatory response. The dermis in which there was complete epithelialization, had less inflammatory response with lesser infiltration of lymphocytes and macrophages (Fig. 1) in both the groups. This was in accordance with observations of Lazovic *et al.* (2005), Ophof (2010) and Basha *et al.* (2011). Higher levels of infiltration by neutrophils, lymphocytes,

plasma cells and macrophages were observed in one dog each from Group I and Group II, where epithelialization was not completed (Fig. 2). This was in accordance to Gomez *et al.* (2004) who observed the presence of inflammatory cells on open wounds of horses treated with processed collagen. Only two giant cells were observed in one dog from Group I whereas giant cells were absent in the dermis of other dogs (Fleck and Simman, 2010). But this was converse to the results observed by Truong *et al.* (2005) on using various dermal matrices. Fibroblast proliferation was extensive with marked collagenisation and conspicuous neoangiogenesis in the samples taken from both the groups (Fig. 3). Neovascularization was widespread in the dermis of skin healed by both bovine collagen and bovine collagen-alginate sheet, with no fatty infiltration and fewer areas of necrosis and fibrosis. Regaining the adnexal structures was observed in one dog from each group, where new epithelialization was complete (Fig. 1). Generally these observations were suggestive that the biomaterials elicited least inflammatory reactions as the presence of inflammatory cells and phagocytic cells were less; at the same time the biomaterials promoted the wound healing process as indicated by the neovascularization and fibroblast proliferation, epithelialization and regaining of adnexal structures. (Jothi *et al.*, 2007; Hayashi *et al.*, 2012).

Marked fibroblast proliferation and collagenisation were noticed in the biomaterial treated wounds suggestive of improved wound healing. Most of the wounds healed within 28 days with near normal histomorphology, indicative of possible regenerative process. Overall, both bovine collagen and bovine collagen-alginate sheet treated wounds showed good graft acceptance with excellent wound healing process.

## References

- Boateng, J.S., Matthews, K.H., Stevens, H.N.E. and Eccleston, G.M. 2008. Wound healing dressings and drug delivery systems: a review. *J. Pharm. Sci.* **97**: 2892-2923.

- Basha, S.K., Kumar, R.V.S., Haragopal, V., Sripatha, Sastry, T.P. and Vidyavathi, M. 2011. Effects of fish scales extracted collagen biocases on cutaneous wound healing in dogs. *Res. J. Pharma. Biol. Chem. Sci.* **2**: 36-49.
- Fleck, C.A. and Simman, R. 2010. Modern collagen wound dressings: function and purpose. *J. Am. Col. Certif. Wound Spec.* **2**: 50-54.
- Gomez, J.H., Schumacher, J., Lauten, S.D., Sartin, E. A., Hathcock, T.L. and Swaim, S.F. 2004. Effects of 3 biologic dressings on healing of cutaneous wounds on the limbs of horses. *Can. J. Vet. Res.* **68**: 49-55.
- Hayashi, Y., Yamada, S., Yanagiguchi, K., Koyama, Z. and Ikeda, T. 2012. Chitosan and fish collagen as biomaterials for regenerative medicine. *Adv. Food Nutr. Res.* **65**: 107-120.
- Jothi, N.A., Thilagar, S., Omar, A.R.S., Kamaruddin, M.D. and Shanthi, G., Goh, Y.M. and Sabri, M.Y. 2007. Effects of biomaterials keratin-gelatin and basic fibroblast growth factor-gelatin composite film on wound healing in dogs. *J. Vet. Malaysia.* **18**: 21-26.
- Lazovic, G., Colic, M., Grubor, M. and Jovanovic, M. 2005. The application of collagen sheet in open wound healing. *Ann. Burns Fire Disasters.* **53**: 151-156.
- Ophof, R. 2010. Construction and application of substitutes for oral mucosa in cleft palate repair. *Ph.D. Thesis*, Radboud University, Nijmegen Medical Centre, 193p.
- Sezer, A.D. and Cevher, E. 2011. Biopolymers as wound healing materials: challenges and new strategies. In: Rosario Pignatello (ed.), *Biomaterials Applications for Nanomedicine*. INTECH, pp. 383-414.
- Truong, A-T.N., Kowal-Vern, A., Latenser, B.A., Wiley, D.E. and Walter, R.J. 2005. Comparison of dermal substitutes in wound healing utilizing a nude mouse model. *J. Burns Wounds.* **4**: 72-82.