

## **EVALUATION OF MODIFIED GELATIN AND AMNIOTIC MEMBRANE AS BIOLOGICAL DRESSINGS FOR WOUND HEALING IN BUFFALO CALVES\***

M. Malakondaiah<sup>1</sup>, N.V. Rao<sup>2</sup> and Makkena Sreenu<sup>3</sup>

A.P. Agricultural University,

College of Veterinary Science

TIRUPATI-517 502, Andhra Pradesh.

Wounds in animals are generally accompanied by haemorrhage, contamination and infection and hence the choice of treatment is to provide good haemostasis for better healing. Several agents like indigenous medicinal preparations (Bhargava *et al.*, 1988 a and b), cartilage powder and tissue extract (Jadon *et al.*, 1985), skin homografts (Artz *et al.*, 1955) and allografts of skin (Brown *et al.*, 1953) have been tried to achieve good healing rate. Recently biological materials like modified gelatin (Ramesh Kumar *et al.*, 1991) and amniotic membranes (Robson and Krizek, 1973) were used as wound dressing agents for various traumatic wounds and burns. The present study was undertaken to evaluate modified gelatin and amniotic membrane as biological dressings on wound healing, in buffaloes.

### **Materials and Methods**

Twenty apparently healthy buffalo calves, aged 2-3 years, weighing 100-150 kgs were used for the study. The animals were treated with triflupromazine\*\* Hcl @ 0.1 mg/kg body weight I/M. Under pocal infiltration using 2% lignocaine, an open wounds of 6 cm x 3 cm were created on thoraco lumbar region on either side of vertebral spine. The wounds were kept open for 24 hours without treatment. The wounds were scraped untill there was haemorrhage and were covered with sterile gauze to prevent contamination. The wounds created on left side of vertebral spine received the treatments as follows.

Group I : Modified gelatin sheet

Group II : Chemically modified amniotic membrane

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\* Siquil : Sarabhai Chemicals, Baroda, India

\* A part of M.V.Sc. thesis approved by A.P. Agricultural University submitted by Senior author

1. Veterinary Asst. Surgeon, AH Department, Kondapuram Nellore District
2. Associate Professor
3. Ph. D. Scholar

The modified gelatin and amniotic membrane were supplied by CLRI. Madras in sealed ampules.

The wounds created on right side were kept as controls in each group and were daily cleaned with 0.85% normal saline. Healing process was judged clinically by observing the appearance of wound, extent of cicatrization and determining the area of wound healing. Wound contraction was measured as per the procedure of Rama Kumar and Tyagi (1972) at 3rd, 6th, 10th 15th and 25th day after wounding. Grossly the wounds were examined for the presence of discharge, granulation tissue and scar formation. Data obtained was subjected to statistical analysis as per Snedecor and Cochran (1967).

## Results and Discussion

Local infiltration produced satisfactory analgesia while inflicting wounds. When modified gelatin sheet and amniotic membrane were applied on the wounds, the material adhered quickly, the wounds remained clear and haemorrhage was arrested. One of the ten wounds treated showed a little pus in amnion treated wounds where as six of the controls showed pus within 2 days and persisted upto 10th day. None of the animals showed pain on palpation of the treated wounds. The mean wound contraction was  $1.20 \pm 0.23$ ,  $3.72 \pm 0.452$  and  $2.75 \pm 0.283$  cm<sup>2</sup> on third,  $2.86 \pm 0.184$ ,  $5.46 \pm 0.285$  and  $4.73 \pm 0.275$  cm<sup>2</sup> on sixth,  $4.85 \pm 0.246$ ,  $7.51 \pm 0.331$  and  $6.09 \pm 0.272$  cm<sup>2</sup> on tenth,  $7.18 \pm 0.322$ ,  $10.94 \pm 0.250$  and  $10.00 \pm 0.249$  cm<sup>2</sup> on fifteenth and

$9.22 \pm 0.095$ ,  $12.55 \pm 0.365$  and  $11.02 \pm 0.358$  cm<sup>2</sup> at 25th post operative day in control, gelatin and amnion treated wounds respectively. The rate of wound contraction was significantly higher ( $P < 0.01$ ) in gelatin and amnion treated wounds when compared to the control at any given period of time. Ramesh Kumar *et al.* (1991) observed absorption of the gelatin film by seventh day of application. In the present study the gelatin sheets adhered well to the wound and was absorbed by 10th day of its application where as the amniotic membrane adhered to the wound within 2 days of application and the membranes remained as a translucent membrane on the wound surface until complete healing had taken place. Modified gelatin sheet was reported to adhere quickly due to an adesive substance glutin present in it (Clause, 1956). Amniotic membrane showed effective adherence to the wound surface, probably due to fibrin bonds between surfaces of the amnion and wound (Jadon *et al.*, 1985). Proper wound adherence prevented oozing of fluids from the wounds. Kanitkar and Tewari (1982) mentioned that the main property of any biological dressing was its adherence to the wound surface.

The wounds were made to bleed following scarification using a sharp sterile scalpel. The normal bleeding time was  $95.50 \pm 2.946$  sec. Following application of modified gelatin sheet and amnion the bleeding time was reduced significantly ( $P < 0.01$ ) to  $67.70 \pm 3.068$  sec. and  $77.50 \pm 1.23$  sec. respectively. Animals showed uneventful recovery without recurrence of haemorrhage from both the treated wounds. None of the treated wounds required

supportive therapy to control haemorrhage and secondary bacterial infection as reported by Ramesh Kumar *et al.* (1991). The normal bleeding time recorded in the present study coincided with the readings of Adval and Gangwar (1971). It was evident from present studies that both materials had haemostatic effects as observed by Clause (1956), Kanitkar and Tewari (1982) and Ramesh Kumar *et al.* (1991). Oedema and seepage of wound exudate were observed upto 3rd POD in control wounds where as no evidence of oedema and seepage were noticed in treated wounds. Control of infection was 100 per cent in treated wounds while it was unsatisfactory in control.

Formation of granulation tissue was evident as early as 6th day in gelatin treated wounds and 7 to 9 days in amniotic membrane treated wounds. The wound cavity was completely filled by 10th day. Granulation tissue formation in control wounds was observed on an average by 12th day. The wound cavity of control group was partially filled by about one third by 14th day and one half by 15th day following wounding. The amniotic membranes formed a dry sheet over the surface of the wound that gradually became detached as reepithelization proceeded beneath the membrane. Despersions of scar were observed on 7th day in control wounds where as no dispersion of scar was observed in treated wounds. The healing rates of control, the gelatin and amniotic membrane treated wounds were  $25.0 \pm 0.360$ ,  $17.90 \pm 0.310$  and  $21.90 \pm 0.230$  days respectively. The treated wounds showed early healing than

control wounds, and gelatin treated wounds healed faster.

Infection in wounds results in tissue destruction, wound dehiscence, delayed healing and increased morbidity and mortality (Singh *et al.*, 1981). Adherence of biological dressing to underlying infected wound bed was seen associated with wound sterilization in the present study. Morris *et al.* (1966) and O'Neill (1970) opined that coverage of contaminated wounds resulted in sterilization of wounds. The wounds treated with gelatin remained clean without pus formation until the healing took place. Control of infection was observed in 90% of the wounds treated with amniotic membrane, the failure due to dispersion of the amniotic membrane and subsequent contamination. Ramesh Kumar *et al.* (1991) observed sepsis in one out of seven cases treated with modified gelatin powder. The scar was more hard in gelatin treated wounds. Neither dispersion of scar nor scar hypertrophy was observed in the treated wounds. Epithelization was clear and the scar was pliable in amniotic membrane treated wounds. It is concluded that both modified gelatin and amniotic membranes enhanced healing process, and modified gelatin was more effective due to the ease of application, quicker adherence, haemostatic and antibacterial effects.

### Summary

The efficacy of the modified gelatin and amniotic membrane as biological dressings in open wounds was evaluated on twenty experimental buffalo calves. Modified

gelatin showed quicker adherence to the wounds than amniotic membrane. The percentage of wound contraction was more in gelatin treated, medium in amniotic membrane treated and least in control wounds. The bleeding time was reduced significantly in treated wounds than control wounds. Early granulation tissue formation, better organisation, and hard scar formation were observed in gelatin treated wounds compared to others.

### Acknowledgement

The authors are thankful to Professor O. Ramakrishna, College of Veterinary Science, Tirupati and Dr. T.P. Sastry and Dr. Muralidhar Rao, Scientists, Bi-products Lab., CLRI, Madras for their valuable suggestions.

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