



Efficacy of Gamat Extract in Wound Healing in Albino Wistar Rats

Barathi S.Subramaniam^{1*}, Arul Amuthan², Prathibha M. D' Almeida¹, Arunkumar HD³

¹Department of Physiology, Melaka Manipal Medical College, Manipal University, India.

²Department of Pharmacology, Melaka Manipal Medical College, Manipal University, India.

³Department of Anesthesiology, Kasturba Medical College, Manipal University, India.

*Corresponding author's E-mail: barathi2021@gmail.com

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ABSTRACT

Stichopus horrens (Gamat: Malay word for sea cucumber) belongs to the family Holothuroidea. It is very abundant in coastal regions of Malaysia and Indonesia. It is a Malay traditional medicine and has been used to cure various ailments for 300 years. Anecdotes claim that, people have used Gamat as a medicine after childbirth and surgery, wound healing, ulcer, chest pain, pneumonia and asthma. However there are no scientific reports on wound healing activity of the sea cucumber, Stichopus horrens (Gamat). An experimental study was carried out to evaluate the wound healing potential of Stichopus horrens (S. horrens) in Rattus norvegicus. All experiments were conducted following standard procedures. The extract of S. horrens in the form of oil was used for evaluating the wound healing potential in excision wound model. Percentage of wound contraction and period of epithelization was studied. Stichopus horrens processed oil demonstrated higher percentage of wound contraction compared to the standard (Povidone iodine ointment) and the control. Period of epithelization was not hastened. The wound contraction elicited by Stichopus horrens in this investigation following topical administration strongly supports the verbal claim of traditional doctors on the use of this processed oil.

Keywords: Stichopus horrens, wound contraction, folklore, Gamat processed oil.

INTRODUCTION

Stichopus known as gamat in Malaysia, Teri pang in Indonesia and bat or balat among the ethnics of Sabah are sea cucumbers of Holothuroidea family (figure 1). It is very abundant in coastal regions of Malaysia and Indonesia.

Sea cucumbers are dried and powdered to make lotion, oil, cream, or cosmetics. It is mixed with clay and applied as a facial mask. A high dose (100 mg/kg) of sea cucumber extract administered intraperitoneally in mice has shown that it is effective against internal pain, but ineffective against externally induced pain¹. It is also suggested that the fatty acids necessary in tissue repair is present in sea cucumber².

Traditional medicines provide moist environment which facilitates establishment of suitable environment, hence more than 80% of the world population still depends upon traditional medicines for their ailments³. Various tribes have applied the gamat over fresh cuts and wounds and claim for its promising wound healing activity⁴.

The present study was conducted to verify the excision wound healing capacity of Stichopus horrens processed oil in experimentally induced excision wounds in Rattus norvegicus.

MATERIALS AND METHODS

Experimental animals

Wistar albino rats (Rattus norvegicus), four weeks old, of either gender weighing between 100 and 150 g were obtained from the Manipal University Central Animal

House and used for wound healing investigation. The animals were housed in standard environmental conditions of temperature, humidity and a 12 h light and 12 h dark cycle. Rats were fed with standard rodent diet and tap water ad libitum. Ethical clearance (IAEC/KMC/67/2011-2012) was obtained from the Institutional ethics committee for conduct of the study. The study was carried out following the guidelines of the principles of laboratory animal care⁵.

Chemicals used

Povidone iodine was procured from hospital pharmacy, Kasturba Hospital Manipal, India.

Sea Cucumber oil (Minyak Gamat Plus) manufactured by Shaari, Malaysia was purchased from drug store.

Excision wound model^{6,7}

Two weeks after procurement, a round seal of 2.5cm diameter was impressed on the dorsal thoracic region 5 cm away from the ears and 1 cm away from the vertebral column of the anaesthetized rat. This area was shaved and sterilized with 70% ethanol. Full thickness skin from demarked area was excised to get a wound measuring 500 sqmm^{8,9}.

After achieving full hemostasis by mopping the wound with cotton swab soaked in warm saline, all wounds were left open and were placed in individual cages. The animals were randomly divided into three groups of six rats each.

In the control group (group 1), the wound was left undressed to the open environment and no local or



systemic antimicrobial agents were used¹⁰. This model was used to monitor the rate of wound contraction and compared with the treatment groups.

Povidone iodine was applied topically to the wounds of standard animal group (group 2).

The animals in the experimental group (group 3) were topically applied with the gamat processed oil extract twice daily for consecutive 21 days.

In this wound model, two physical attributes of healing namely; percentage of wound contraction rate and epithelisation period were studied.

From the day of wounding, the wound contractions were measured by a tracing paper on the wounded margin on every fourth day until the wound completely healed, restraining the animals in proper position during the process. The measured wound contraction was converted into percentage taking the size of the wound at time of wounding as 100 %.

Restraining the animals in proper position, the wound contractions were measured using a tracing paper on the wounded margin.

$$\% \text{ of wound contraction} = \frac{\text{Initial wound size} - \text{specific day wound size}}{\text{Initial wound size}} \times 100$$

Epithelization period was monitored by noting the number of days required for the scar to fall off leaving no raw wound behind^{11, 12}. The wound contraction and period of epithelization were measured in all groups.

Statistical analysis

Data was analyzed using one- way Analysis of Variance (ANOVA) followed by Scheffe's test. P value of 0.05 was considered as significant.

RESULTS AND DISCUSSION

The results shown in Table 1 reveal the following:

At day 4, the group 3 exhibited significantly smaller wounds when compared to the group 1 (P<0.001) and similar changes were observed between Group 1 and 2 (P<0.001). There was no significant difference between Group 2 and Group 3. On days 8, 12, 16 and 21; there was no significant change in the wound contraction amongst the groups (p >0.05 each group) except a significant change between group 1 and group 2 (P<0.05) on day 8.

All the animals studied showed similar period of epithelization of 21days. Thus, gamat in topical applications, enhanced wound contraction in this model only in the initial period of wound healing.

All of the wounds in this study exhibited the typical pattern of wound healing, with minimal wound contraction noted during the initial lag phase followed by a rapid increase in wound contraction during the proliferative phase (day 6 to day 21).

Table 1: Mean Values of Percentage of Wound Contraction Expressed As Mean \pm SEM

Day	Group I (Normal Saline)	Group II (Povidone Iodine)	Group III (Experimental Group)
4	45.83 \pm 4.53	13.87 \pm 5.04	10.6 \pm 2.51
8	73.57 \pm 3.31	55.5 \pm 8.08	71.27 \pm 5.36
12	86.93 \pm 1.64	79.87 \pm 2.63	86.1 \pm 2.32
16	96.17 \pm 0.97	91 \pm 2.26	92.47 \pm 1.96
21	99.07 \pm 0.42	96.9 \pm 1.43	99 \pm 0.48

Wound healing is a natural and spontaneous phenomenon which occurs irrespective of (and sometimes despite) the surgeon. Although the basic events have been observed for many years, the factors which initiate and control the process remain incompletely understood. The pattern of wound healing may be affected by cytokine, endocrine or pharmacological manipulation of the wound's environment. Some wounds heal entirely without surgical intervention. Even some larger tissue craters contract amazingly rapidly, with local treatment¹³.

Healing begins the moment a wound is sustained. However, in a contaminated wound, particularly if there is dead or damaged tissue, bacteria also multiply and increase in number. In most wounds, the first 4-6 hours is a period when bacteria are present but not in sufficient numbers to influence the healing cascade greatly; if they can be eliminated or their environment made unattractive during this time then healing proceeds normally. If this fails to occur, the balance from 6 to 18 hours tends progressively to tip in favor of the proliferating bacteria. By this time they are themselves influencing the inflammatory process, so that the wound must usually be regarded as infected.

Collagenation, wound contraction and epithelization are crucial phases of wound healing. The phases of inflammation, macrophasia, fibroplasia and collagenation are intimately interlinked. Thus an intervention into any one of these phases by drugs could eventually either promote or depress one other or all phases of healing¹⁴.



Figure 1: Gamat



Figure 2: Day 0 Excision wound



Figure 6: Normal Saline Applied Wound on day 18



Figure 3: Gamat applied wound on day 8



Figure 7: Povidone Iodine Applied Wound on day 18



Figure 4: Povidone Iodine applied wound on day 8



Figure 8: Gamat applied wound on day 18



Figure 5: Normal Saline Applied Wound on day 8

CONCLUSION

The present study was undertaken to evaluate whether gamat promote wound healing in experimentally induced excision wounds in rats. It has shown remarkable healing property in initial phase of wound healing. The healing or wound contraction elicited by *Stichopus horrens* processed oil in this investigation following topical administration strongly supports the verbal claim of folklore medicine. Further studies needs to be done to explore the exact mechanism by which gamat processed oil extract facilitates wound healing and its healing effect on other type of wounds such as burns.

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