Research Article **ISSN: 2393 – 9532**

International Journal of Arts and Science Research

Journal home page: www.ijasrjournal.com

https://doi.org/10.36673/IJASR.2021.v08.i01.A03

COMBINED WOUND HEALING ACTIVITY OF *CALENDULA OFFICINALIS* AND B-GLUCANS

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ABSTRACT

In this study, albino mice were used to test the wound healing ability of a gel made from hydro alcoholic extracts of *Calendula officinalis* (CE) and β-glucans in an excision wound model. The treated animals showed a considerable reduction in the time it took for epithelization and wound contraction in excision wound models, as well as rapid wound healing activity in a mixed gel. It's possible that this is related to synergism. The improved wound healing activity of hydro alcoholic extracts may be related to free radical scavenging action and phytoconstituents found in it, which, either alone or in combination, speed up the wound healing process.

KEYWORDS

β-glucans, Wound healing properties and *Calendula officinalis*.

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INTRODUCTION

A wound is a break in the tissue's integrity that causes harm and usually leads in loss of function. Wound healing is a biological process that is started by trauma and usually ends with scar development. Coagulation, epithelization, granulation and tissue re-modelling are all aspects of the wound healing process. Wound healing frequently occurs in a direction that is different from its normal course, and underhealing, overhealing, or no healing of the wound is typical[1]. Many ayurveda medicinal herbs have a vital part in the treatment of wounds. Plants

are more effective healers because they support natural mending mechanisms.

Calendula officinalis, sometimes known as pot marigold, is a common garden plant in the Compositae family[2-4]. The blossoms are the part of the herb that is used medicinally[5], either as infusions, tinctures, liquid extracts, lotions or ointments, or in one of a multitude of over-the-counter skin and hair products available around the world. The activation of immunological and cutaneous cells by β-glucans (BG) molecules promotes moist wound healing and repair. Homeostasis, re-epithelization, granulation, tissue creation, and extracellular matrix remodeling are all part of the wound healing process[6]. As a result, a multi-modal therapeutic method may help the wound heal faster. The goal of this study was to see how the gel of *Calendula officinalis* (CE) and β-glucans worked together to treat experimentally produced lesions in mice.

MATERIAL AND METHODS

Preparation of extract

Calendula officinalis leaves (100g) were coarsely pulverized. In a Soxhlet extractor, the powdered materials were loaded and defatted using petroleum ether (40-60ºC). The marc was dried and extracted three times with ethanol (50 percent v/v) in the same extractor. Finally, using a rotary evaporator under vacuum, the extracts were condensed to a semi-solid mass. The solvent was removed from the dried extract by placing it in a desiccator. The extract of *Calendula officinalis* was designated as CEE**.**

Animals

Wistar albino mice of more ever sex weighing 25-40g were kept in plastic cages at a temperature of 252°C, a relative humidity of 50±15°C and a standard photoperiod of 12 hours dark/12 hours light. The animals were given a conventional pellet diet and free access to water. Following wounding, the animals were placed in individual cages for treatment until the wounds had healed completely. Six animals were employed in each group of different models. The King Khalid University Institutional Animal Ethical Committee approved the animal experiments, and the research work was carried out as per the guidelines of the US National Institute of Health's standards for the care and use of laboratory animals (NIH Publication No.85-23, amended 1996).

Evaluation of Wound Healing Activity[7]

Excision model

Mice of both sexes, weighing 25-40g, were randomly collected. They were divided into four groups of six and placed in various cages.

Treatment groups

Group I: carbopol gel, Group II: CEE gel, Group III: CEE gel + BG, Group IV: Standard (metrozyl gel) for the excision wound study.A total of six animals were chosen for each group. Under light ether anaesthesia in aseptic setting, a circular wound of about 10mm diameter was created on the depilated dorsal thoracic area of mice and examined throughout the investigation. Individual cages were utilised to house the animals. 2.5% carbopol gel is applied to Group-I animals. With a CEE of 2.5 percent, Group II is used. Group III receives a thin layer of 2.5 percent CEE + BG (80mg/kg, po). Metrozyl gel is administered twice daily as a thin layer to Group IV animals. The treatment gel was applied twice daily and the BG was given once daily, po. On the 2, 4, 6, 8, 10, 12, 14, 16, 18, 20, 22nd post-wounding days, the wound area can be assessed. The percent of wound contraction was computed from the day the wound area was measured, as well as the epithelization time.

RESULTS AND DISCUSSION

Animals treated with CEE, CEE + BG showed a considerable increase in wound-healing activity when compared to those that received control treatments. The effects of *Calendula officinalis* hydro alcoholic extracts and a combined extract of both the plant and BG on wound healing activities in mice with excision wounds were shown in Table No.1. When comparing groups treated with CEE and CEE + BG to control groups, there is a significant increase in percent wound contraction in the CEE and CEE + BG groups.

Discussion

Myofibroblast activity is thought to be responsible for wound healing[8]. Because CEE + BG improved wound contraction, myofibroblasts' contractile

properties would have been boosted, or the quantity of myofibroblasts recruited into the wound area would have increased. CEE dramatically accelerated epithelialization in an excision wound model, while CEE in combination with BG accelerated epithelialization. On the 15th day, concomitant administration of CEE and BG considerably accelerated wound contraction. It also appears that CEE was able to promote epithelialization either by facilitating the proliferation of epithelial cells or by increasing the viability of epithelial cells.

β-glucans possess a broad spectrum of biological activities that enhance immunity in humans. Topical applications of β-glucans are increasing, especially due to their pluripotent properties. Macrophages, keratinocytes and fibroblasts are considered the main target cells of β-glucans during wound healing. β-glucans enhance wound repair by increasing the infiltration of macrophages, which stimulates tissue granulation, collagen deposition and re-epithelialization. β-glucans wound dressings represent a suitable wound healing agent, with great stability and resistance to wound proteases[6].

β-glucan wound dressings are a good wound healer because they are stable and resistant to wound proteases[6]. Based on the aforesaid individual properties of CEE and β-glucans, combining the two in a multi-modal treatment method considerably improved wound healing. CEE's improved wound contraction impact and epithelization could be used in the treatment of open wounds in the future. However, a well-designed clinical assessment will be required to confirm this suggestion.

Table No.1: Excision wound model

S.No	Groups	% Wound contraction					Epithelialization Period
		2nd day	4th day	6th day	8th day	10th day	
1	I	42.1+3.27	63.63+4.52	72.9+6.07	79.55+6.62	79.55+6.62	18.58
2	II	34+ 0	66.30+5.83	77.84+6.7	89.51+ 9.9	89.51 + 9.9	14.33
3	III	41.84+2.27	72.69 +6.4	81.79 +6.39	91.63 +6.8	91.63 +6.8	11.15
4	IV	43.28+3.7	52.36 +5.6	75.17+5.34	81.51+5.94	82.51+5.9	16.67
5	V	42.1+3.27	63.63+4.52	72.9+6.07	79.55+6.62	79.55+6.62	18.58

CONCLUSION

The current investigation found that an ethanol extract of CEE+β-glucans possesses features that allow it to promote faster wound healing activity when compared to placebo controls. Further research into the topical treatment and management of wounds with CEE+β-glucans is needed due to wound contraction and enhanced tensile strength.

ACKNOWLEDGEMENT

The authors are grateful to the Deanship of Scientific Research at King Khalid University for funding this study through the Small Research Group Project, under grant number GRP/355/42.

CONFLICT OF INTEREST

We declare that we have no conflict of interest.

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Please cite this article in press as: Krishnaraju Venkatesan *et al*. Combined wound healing activity of *calendula officinalis* and β-glucans, *International Journal of Arts and Science Research,* 8(1), 2021, 16-19.