



Wound healing profiles of *Ginkgo biloba*

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Abstract

Objective: To evaluate the effects of *Ginkgo biloba* on wound healing. **Materials and methods:** The wound healing effects of dried extract of *Ginkgo Biloba* (GB) were tested at a dose of 50 mg/kg on dead space and excision wound models in male rats. **Results:** GB has significantly ($p < 0.01$) promoted the breaking strength and hydroxyproline content of granulation tissue in dead space wounds. In case of excision wounds while it did not affect the wound contraction, the epithelization period was significantly ($p < 0.01$) shortened. **Conclusion:** GB exhibits a significant pro-healing activity, possibly by influencing the collagenation phase of wound healing.

Key words: *Ginkgo biloba*, Wound healing, Dead space wound, Excision wound, Granulation tissue, Hydroxyproline

1. Introduction

Extracts from the leaves of *Ginkgo biloba* (GB)(Ginkgoaceae) have been used therapeutically for centuries [1]. The GB exhibits a variety of interesting Pharmacological activities such as antioxidant, membrane stabilizing, increase in blood fluidity and improvement in cognition [2,3]. One of the main constituents of the GB is a flavonoid glycoside. The free flavonoids present in *Tridax procumbens* have been reported to have pro-healing activity [4,5]. In view of these the present study has been undertaken to find out if GB also could influence the healing positively.

2. Materials and Methods

The studies were conducted in male Wistar rats (150-200 g) housed individually. The rats were divided into various groups, each containing 8-10 animals.

After overnight starvation, the rats were wounded under light ether anesthesia. Full aseptic measures were not considered necessary and thus, no local or systemic chemotherapy was provided. Animals showing the signs of infection were excluded from the study.

The dried extract of GB (courtesy from M/s. Ranbaxy, Mumbai) standardised to Ginkgo flavone 24% was dissolved in distilled water and administered intraperitoneally at a dose of 50 mg/kg.

2.1 Wound models

2.1.1 Dead space wounds

Dead space wounds were created by implantation of a polypropylene tubes (2.5X0.5 cm) beneath the dorsal paravertebral lumbar skin [6]. On day-10 the granulation tissue harvested was subjected to

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physical as well as biochemical evaluations. First the breaking strength of the tissue was measured by the method of Lee [7]. Then the pieces of the tissue were dried at 60°C for 24h and the dry weight was measured. Later, the hydroxyproline content (measure of collagen) of the tissues was determined [8].

2.1.2 Excision wounds

A circular piece (500 mm² in area) of full thickness skin was excised from the dorsal interscapular region [9]. Wound contraction was monitored by measuring wound area, planimetrically, on the alternate days till the wounds were completely healed. Later the wound closure time 50 (WC₅₀) was calculated by the method of Litchfield and Wilcoxon [10]. Time taken for full-epithelization was measured by recording the days required for fall of scab leaving no raw wound behind.

2.2 Drug administration

All animals of a given group bearing one or the other type of wound received daily either distilled water (0.2 ml i.p.) or GB (50 mg/kg i.p.) from the day of wounding for 10 days in case of dead space wounds and for 21 days in case of excision wounds.

2.3 Statistical analysis

Results were analyzed by Unpaired Student's *t* - test. The minimum level of significance was fixed at $P < 0.05$.

3. Results

3.1 Dead space wounds

Breaking strength of the 10-day old granulation tissue in the control group of animals was 245 ± 16 g.

This was significantly ($p < 0.01$) increased by GB without significantly altering the granulation tissue mass weight. However, it did significantly enhance the content of hydroxyproline in the granulation tissue (see Table 1)

3.2 Excision wounds

As could be seen in the Table 2, GB promoted the epithelization by 3 days, while it did not alter the wound contraction when compared to control.

4. Discussion

Healing a complex process involves a number of phases *viz.*, coagulation, inflammation, collagenation, wound contraction and epithelization. While the phases between coagulation to collagenation are intimately inter-linked, the phases of wound contraction and epithelization are independent to each other and run concurrently. No single wound model, thus, be sufficient to assess the influence of drugs on wound healing.

Therefore, in the present study two wound models *viz.*, dead space (for collagenation phase) and excision wounds (for wound contraction and epithelization phases) were employed.

It is a well-accepted fact that wounds in most tissues heal by repair *i.e.*, by laying down non-specific connective tissue. Further, it is also known that nearly 50% or more of connective tissue is made up of collagen. GB has increased the collagen content of the granulation tissue, as evidenced by significant increase in hydroxyproline content, without significantly altering the granulation weight of the dead space wounds. The granulation weights

Table 1
Effect of *Ginkgo biloba* on dead space wound parameters

Drug (n)	Dose (mg/kg i.p.)	Granulation breaking strength (g)	Dry granulation weight (mg)	Hydroxyproline content (ug)
Control (8)	0.2 ml of distilled water	245 ± 16	123 ± 18	1948 ± 52
GB (9)	50	$323 \pm 19^*$	152 ± 20	$2529 \pm 78^{**}$

Values are in Mean \pm SEM; * $p < 0.01$, ** $p < 0.001$ Vs. control
n = number of animals per group.

Table 2
Effect of *Ginkgo biloba* on excision wound parameters

Drug (n)	Dose (mg/kg i.p.)	Period of epithelization (days)	Wound closure-50 (WC ₅₀) (days)
Control (10)	0.2 ml of distilled water	19.4 ± 0.4	7.8 ± 0.1
GB (9)	50	16 ± 1.15*	7.3 ± 0.3

Values are in Mean ± SEM; * p < 0.01 Vs. control
n = Number of animals in a group.

consist of inflammatory cells, new blood vessels, mucopolysaccharides, collagen etc. May be the GB caused an increase in the collagen amount at the cost of other constituents of the granulation tissue.

The increase in the amounts of collagen perhaps may be responsible for increase in breaking strength. In addition GB also promoted epithelization but without affecting the wound contraction (wound closure time) of the excision wounds. This means that GB without enhancing the activity of the myofibroblasts (force generators for wound contractions), it would still have enhanced the migration and proliferation of epithelial

cells. This kind of differential actions of a drug on the healing is not surprising [4,5], as these two phases run independent to each other.

From these it could be said that GB has prohealing action. Further, it could be assumed that the conferred prohealing action on the GB would be due to the presence of flavanoids (flavanoid glycosides) in GB. The fact that such type of flavanoids enhanced the healing [4,5] strengthens the above argument.

Clinically wound strength is an important aspect in most surgical wounds. A weak scar could lead to wound dehiscence or incisional hernia later. Such an eventuality can be minimized by the administration of GB in a suspected weak scars situations. In case of leg ulcers, extensive burns, and healing of donor area in skin graft surgeries a rapid epithelization with minimum contraction is very handy. Thus, GB could be tested for quicker coverage of epithelial layer.

Further studies are in progress to ascertain if GB could reverse the anti-healing effects of steroids and antiproliferative drugs.

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