INVESTIGATION OF WOUND HEALING ACTIVITY OF CRESSA CRETICA BY EXCISION AND INCISION METHOD

*1Pragati Khare, 2Noopur Khare and 1Bharat Gupta
1Department of Pharmacy, Shri Ram Murti Smarak (C.E.T), Bareilly, U.P., India.
2Department of Biotechnology, Shri Ram Swaroop Memorial University, Lucknow, U.P, India.

*Corresponding Author: Pragati Khare
Department of Pharmacy, Shri Ram Murti Smarak (C.E.T), Bareilly, U.P., India.

ABSTRACT
Objective: Cressa cretica (Convolvulaceae) is a traditional medicinal plant known as Rudanti. This plant has been used for the treatment of a variety of diseases. The plant of Cressa cretica showed antibilious, antituberculosis and expectorant, nootropics activities. Methods: This study was done to investigate the possible wound healing effects of Cressa cretica plant extract (CCE) using Excision and incision wound models. 24 swiss albino mice of either sex weighing between 25-30gm were randomly selected and divided into 4 equal groups. Group-I (control) received Control (7% HPMC gel), Group-II received Aloe vera gel (90%), III received Cressa cretica (4% HPMC gel) and Group IV received Cressa cretica (8% HPMC gel). Drug treatment was given for ten days. Results: CCE produced significant wound healing effect at dose of 4% HPMC gel & 8% HPMC gel administered for 10 days. The efficacy of CCE was found to be comparable to that of Aloe vera. Conclusion: The results of the present study indicate that CCE possesses significant wound healing activity compared to that of Aloe vera.

KEYWORDS: Cressa cretica, excision test, incision test, Aloe vera.

INTRODUCTION
Cressa cretica L. (Convolvulaceae), popularly known as ‘Rudanti’ in Hindi is a useful medicinal plant. Different parts of the plant have been claimed to be valuable in a wide spectrum of diseases.[1] In earlier studies Cressa cretica Linn flowers exhibited cytotoxic and anti-inflammatory activity in vitro. Cressa cretica is reported to be antibilious, antituberculosis and expectorant.[2] It has been reported that five flavonoids (quercetin, quercetin-3-O-glucoside, kaempferol-3-Orhamnoglucoside and rutin) are present in the aerial parts of Cressa cretica.[3]

Wound healing is an important area of research. Nowadays, scientists are targeting at the cell signaling which is included in generation of new tissues and repairing the wound.[4] The use of herbal medicines for the treatment of human ailments has been a natural approach to the health care. In the search for new therapeutic products for the treatment of neurological disorders, medicinal plants have proven to exhibit pharmacological effectiveness in variety of animal models.[5] Thus the present study has been done to investigate the wound healing activity of Cressa cretica plant extract (CCE) in mice by using excision and incision wound model.

MATERIALS AND METHODS
Preparation of Cressa cretica plant extract (CCE)
The plant of Cressa cretica was washed thoroughly in tap water, shade dried and powdered. This powder was packed into Soxhlet column and extracted with petroleum ether (60-80°C) for 24 h. The same marc was successively extracted with chloroform (50-60°C) and later with ethanol (68- 78°C) for 24 h. The extracts were concentrated on water bath (50°C). After concentrated preparation, the dried powder extract was stored at room temperature. The yield of the petroleum extract, chloroform extract and ethanolic extract were found to be 0.8% (w/w), 0.8% (w/w) and 1.0% (w/w) respectively. Ethanolic extract was used for the experimental study.

Plan of study
Animals
Animals were procured from Central Animal House, SRMS, Bareilly. Animals were approved by Institutional Animal Ethic Committee (IAEC) of SRMS, Bareilly. Approval number (715/02/a/CPCSEA) was given for this work. The preferred rodent species included mice. Swiss albino strains of young healthy adult of either sex animals in equal numbers per group (n= 6) were taken. At the commencement of the study the weight variations of animals used was kept minimal and not exceeded ±
20% of the mean weight of each animal. Normal weight of mice was 25-30 gm.

The temperature of the experimental animal room was maintained to be 22°C (±3°C). Relative humidity was maintained between 50–60%. Lighting was artificial, the sequence being 12 hours light, 12 hours dark. For feeding, conventional laboratory diets were used with drinking aqueous supplied *ad libitum*. Animals of same group were caged together. Healthy young adult of either sex mice were randomly assigned to the control, standard and treatment groups. The animals were identified uniquely (i.e., by marking at the base of the tail) and acclimatized for not less than 5 days in their cages prior to the start of the study.

**Study Design**
The mice were divided into 4 groups:

**Table 1: Study groups and their treatments**

<table>
<thead>
<tr>
<th>GROUPS</th>
<th>TREATMENT</th>
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</thead>
<tbody>
<tr>
<td>Group I</td>
<td>Control (7% HPMC gel)</td>
</tr>
<tr>
<td>Group II</td>
<td>Aloe vera gel (90%)</td>
</tr>
<tr>
<td>Group III</td>
<td><em>Cressa cretica</em> (4% HPMC gel)</td>
</tr>
<tr>
<td>Group IV</td>
<td><em>Cressa cretica</em> (8% HPMC gel)</td>
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</table>

**Laboratory models for testing wound healing activity**

**Effect on excision wound**: Pentobarbitone (30 mg/kg i.p.) was used to anaesthetize the mice. An impression was made on the dorsal thoracic region 1 cm away from vertebral column and 5 cm away from ear on the anaesthetized mice. This area was shaved one day before the experiment. The skin of the impressed area was excised to get a wound area of 500 mm². Haemostasis was achieved by blotting the wound with cotton dipped in normal saline.

Wound area was measured by measuring the area on a millimeter scale graph paper. The percentage wound healing was calculated of original wound size for each mice on 2, 4, 8, 10, 12, 14, 16, 18, 22 days post wounding for the final analysis of results. Removal of scar leaving no wound behind was considered as end point of complete epithelization and the days required for this was considered as period of epithelization.[6]

**Effect on incision wound**: Para vertebral straight incision of about 6 cm was made through the skin, on either side of the vertebral column by sharp scalpel. After haemostasis the wound was closed by sutures placed 1 cm apart. Mice were treated with drugs as mentioned in excision model from 0th day to 9th post-wounding day. The wound breaking strength was estimated was estimated on 10th day by continuous constant water flow technique.[6]

**Statistical Analysis**
Results are expressed as MEAN ± SEM. The difference between experimental groups were compared by one-way ANOVA followed by Dunnet’s test. The results were considered statistically significant when P<0.05.

**RESULTS**

**Fig. 1**: Epithelization period of various groups.

**Fig. 2**: Breaking strength of various groups in excision wound.

**Fig. 3**: Breaking strength of various groups in incision wound.
DISCUSSION
This research study was conducted to investigate the wound healing activity of *Cressa cretica* leaf extract in experimentally induced wounds in mice. The results of this study revealed the wound healing property of *Cressa cretica* leaf extract. The breaking strength, wound contraction and period of epithelization were elevated by the use of *Cressa cretica* leaf extract if used orally or topically.

CONCLUSION
It is concluded from the study that the ethanolic leaf extract of *Cressa cretica* possesses wound healing activity as it was reported that *Cressa cretica* extract was much potent in excision and incision models of wound healing activity. Further studies are needed to prove the wound healing property of ethanolic extract of leaves of *Cressa cretica* (4% and 8%) in other animal models. Since *Cressa cretica* extract proved to be more potent than Aloe vera in incision and excision wound healing models, isolation of the active principles from the extract may cause development of wound healing drugs.

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REFERENCES