ABSTRACT
The wound healing activity of ethanolic extract of dried flowers of Mesua ferrea evaluated on excision and incision model, in albino rats, in the form of an ointment with two concentrations (5% and 10% w/w ointment of bark extract in simple ointment base). Both concentrations of the ethanolic extract showed significant response in both the wound types tested when compared with the control group. Nitrofurazone ointment (0.2%w/w) used as standard.

I. INTRODUCTION
Wound healing process involves several steps, which involves coagulation, formation of granulation tissue, collagenation and aquisation of wound strength. During the formation of new tissue, endothelial cells proliferate and form new blood vessels.

The volatile oil extracted from the flowers has antibacterial, antifungal and anthelmintic properties and shows excellent anti-inflammatory and styptic activity. The root of this herb is used as an antidote for snake poison, the dried flowers are used for bleeding hemorrhoids and dysentery with mucus while the fresh flowers are useful for excessive thirst, excessive perspiration, cough, and indigestion, the seed oil obtained from the plant is considered to be very useful in vata disorders and skin diseases. Thus almost every part of plant has therapeutic qualities and proves useful in treating various ailments. The seed oil is considered to be very useful in conditions like vata and skin diseases. Dried flowers are used for bleeding hemorrhoids and dysentery with mucus. Fresh flowers are useful remedy for itching, nausea, erysipelas, bleeding piles, metorrhagea, menorrhagea, excessive thirst, and sweating. Oil from the seeds is used for sores, scabies, wounds, and rheumatism. The stamens which yield the drug Nagakesara contain mesuferrone-A and B, mesuferrol, mesuanic acid, a- and b-amyrin, and beta-sitosterol. The seeds and heartwood contain a number of xanthones and coumarins. Xanthones isolated from the plant given orally or intraperitoneally have been shown to exhibit significant anti-inflammatory activity in normal as well as adrenolecetomised rats. The Ethanolic extract of the plant showed diuretic and hypotensive activity. Volatile oil from the flowers showed antibacterial, antifungal and anthelmintic activities. Mesua ferrea has been used in folk medicine for the treatment of skin diseases and wound. A survey of literature revealed that no systematic approach has been made to study the wound healing activity of this plant. Thus the present study was undertaken to assess the effect of this indigenous plant on different parameters related to wound healing in rats.

II. MATERIALS AND METHODS
Plant material
The dried aerial parts of Mesua ferrea was purchased from Dravid Herbs World, Pondicherry, India.

Extraction
Mesua ferrea aerial parts extracted with 90% ethanol in a soxhlet extractor. The extract was concentrated under reduced pressure at a temperature below 60°C to yield a syrupy mass (Yield ~8.68%), which was used for the present investigation.

Preliminary phytochemical investigation
Preliminary phytochemical analysis shows the presence of glycosides, tannins, phenolic compounds and flavonoids.

Preparation of drug formulation
Two types of ointment formulations with different concentration of the extract were prepared viz. 5%(w/w) ointment, where 5g. of extract was incorporated in 100g. of simple ointment
base; 10% (w/w) ointment where 10g of extract incorporated in 100g of simple ointment base. Nitrofurazone ointment (0.2% w/w) was used as standard drug for comparing the wound healing potential of the extract in different animal model.

Animals
Healthy Wistar albino rats of either sex weighing 150-200gm were used. They were kept in a standard environment condition and fed with rodent diet and water ad libitum. The experimental protocol have been approved by institutional animal ethical committee.

vi. Wound healing activity
The wound healing studies were carried out using ether anaesthetized rats in two different wound model at two different concentrations (5% and 10% w/w).

a. Incision wound - In incision wound model four groups (The group I was considered as control, the group II served as the reference standard and treated with 0.2% w/w Nitrofurazone ointment. The group III animals were treated with the 5% w/w ethanolic extract and the group IV animals were treated with 10% w/w ethanolic extract of Mesua ferrea of animals containing six in each group. Paravertebral incision of 6cm. long were made on either side of the vertebral column of the rat. Care was taken to see that incision was at least 1cm. lateral to vertebral column. The wounds were closed with interrupted sutures of 1cm. apart. The animals were caged individually. The sutures were removed on 8th post wounding day. The tensile strength of the wound was measured on 10th post wounding day.

b. Excision wound - In excision wound four groups (The group I was considered as control, the group II served as the reference standard and treated with 0.2% w/w Nitrofurazone ointment. The group III animals were treated with the 5% w/w ethanolic extract and the group IV animals were treated with 10% w/w ethanolic extract of Mesua ferrea of animals containing six in each group. A circular piece of full thickness (approx. 500 mm²) was cut off from a predetermined area on the back of the rat. Wounds were traced on 1mm² graph paper on the day of wounding and subsequently on alternate days until healing was complete. Changes in wound area were calculated, giving an indication of the rate of wound contraction. Number of days required for falling of the eschar without any residual raw wound gave the period of epithelization. The ointment of the fruit extract, standard drug and simple ointment was applied to the wound twice daily, until recovery to the respective groups of animals.

Statistical analysis - The results are expressed as mean ± SE of six animals in each group. The data were evaluated by students t-test and the values of P ≤ 0.001 were considered statistically significant.

III. RESULTS
It was observed that the wound healing contracting ability of the extract ointment in different concentrations was significantly greater than that of the control (i.e. simple ointment treated group). The 10% (w/w) extract ointment treated groups showed significant wound healing from the fourth day onwards, which was comparable to that of the standard drug, i.e. nitrofurazone ointment treated group of animals. The wound closure time was lesser, as well as the percentage of wound contraction was much more with the 10% w/w extract ointment treated group (18 ±1 days for 100% contraction which was almost similar to that of the nitrofurazone treated group). The 5% (w/w) extract ointment treated group of animals showed significant wound contraction from the eighth day onwards and achieved 100% with the wound closure time of 20 ±2 days.

In incision wound model the measurement of the effect of the extract and standard drug on the tensile strength is shown in Table II. The tensile strength of the 10% extract treated group and the nitrofurazone ointment treated group were comparable to each other. The 5% extract ointment treated group showed a lesser but significant increase in the tensile strength compared to the control group. Thus both concentrations of the extract as well as the standard drug showed a significant increase in tensile strength in the 10 days old wound. The results of the present study revealed that both concentration (5% and 10% w/w) of ethanolic extract of Mesua ferrea flower have significant wound healing activity in both incision as well as excision wound models.

IV. DISCUSSION
Wound healing is a fundamental response to tissue injury that results in restoration of tissue integrity. This is mainly achieved by the synthesis of the connective tissue matrix. Collagen is a major protein of the extracellular matrix and is the component that ultimately contributes to wound strength. Tannins promote the wound healing through several cellular mechanisms; chelation of the free radicals and reactive species of oxygen, promoting contraction of the wound and increasing the formation of capillary vessels and fibroblasts and including keratinocyte proliferation, but do not act on the differentiation towards cornified cells. Similar findings have been reported with the extracts of the plants containing tannins by earlier
However, our results revealed that tannins are one of the important phytoconstituents responsible for wound healing mainly due to their astringent and antimicrobial property. Hence, it can be inferred that the wound healing activity of the *Mesua ferrea* is due to its tannin content, which seems to be responsible for wound contraction and increased rate of epithelization.

Table 1: Effect of ethanolic extract of *Mesua ferrea* on % wound closure of excision wound model

<table>
<thead>
<tr>
<th>Group</th>
<th>4th Day</th>
<th>8th Day</th>
<th>12th Day</th>
<th>16th Day</th>
<th>Period of epithelization in days</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>18.52±0.68</td>
<td>27.21±1.02</td>
<td>48.21±1.80</td>
<td>68.53±2.60</td>
<td>24</td>
</tr>
<tr>
<td>Nitrofurazone</td>
<td>35.28±0.15</td>
<td>76.80±0.19</td>
<td>89.81±0.58</td>
<td>97.11±0.48</td>
<td>18</td>
</tr>
<tr>
<td>Extract(10%)</td>
<td>33.44±1.01</td>
<td>70.24±1.24</td>
<td>82.38±2.36</td>
<td>90.24±2.10</td>
<td>18*</td>
</tr>
<tr>
<td>Extract(5%)</td>
<td>19.82±1.02</td>
<td>35.46±1.82</td>
<td>62.56±2.78</td>
<td>76.56±2.32</td>
<td>20</td>
</tr>
</tbody>
</table>

Values are mean ±SE
*P<0.001 vs control
n=6 animals in each group

Table 2: Effect of ethanolic extract of *Mesua ferrea* on tensile strength of incision wound

<table>
<thead>
<tr>
<th>Treatment groups</th>
<th>Tensile strength(g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>310±4.6</td>
</tr>
<tr>
<td>Nitrofurazone</td>
<td>564±1.8*</td>
</tr>
<tr>
<td>Extract(10%)</td>
<td>524±4.2*</td>
</tr>
<tr>
<td>Extract(5%)</td>
<td>448±5.8*</td>
</tr>
</tbody>
</table>

Values are mean ±SE
*P<0.001 vs control
n=6 animals in each group

REFERENCES