EUROPEAN JOURNAL OF BIOMEDICAL AND PHARMACEUTICAL SCIENCES
http://www.ejbps.com

ISSN 2349-8870
Volume: 5
Issue: 01
93-99
Year: 2018

EFFECTIVENESS OF *MADHUCA LONGIFOLIA* ROOT BARK POWDER FOR WOUND HEALING ACTIVITY IN WISTAR ALBINO RATS.

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Article Received on 08/11/2017 **Article Revised on 29/11/2017** **Article Accepted on 19/12/2017**

ABSTRACT
Wound healing is the process of repair that follows injury to the skin and other soft tissues. Following injury, an inflammatory response occurs and the cells below the dermis deepest begin to increase collagen production. Later, the epithelial tissue is regenerated. There are three stages to the process of wound healing: inflammation, proliferation, and remodeling. Traditionally, *Madhuca longifolia* is used for wound healing. Since no detailed scientific data are available regarding the wound-healing activity of *Madhuca longifolia*, the present study was designed to explore the same. The aim of this study is to identify the effectiveness of *Madhuca longifolia root bark* powder in wound healing activity on animal model. It is an experimental study on healthy wistar albino rats. It was evaluated in excision wound model. The animals were distributed into 3 groups of 3 each. The animals of group A were left untreated and considered as control. Group B served as standard and received cloxacillin. Group C was considered as test and treated with prepared test drug. Powder of test drug and standard drug were topically applied 500mg every alternative day and bandaged starting from the day of operation, till complete epithelialization up to 14 days. The direct observation of wound size, Exudates type and amount, edges, necrotic tissue type and skin coloration of surrounding wound were records converted into Bates-Jensen Wound assessment Tool. Control group showed continuing recovery due to physiological healing during the experiment up to 14 days. The Test and Standard groups showed considerably minimum duration for complete wound healing. The time duration for complete wound healing of the standard drug was observed 14 days where as Test drug, it was only 12 days. *Madhuca longifolia* root bark powder showed as 60.78% of effectiveness while the standards effectiveness 58.82%. It shows that Test drug is more effectiveness than Standard drug. So it is clearly denotes powder of *Madhuca longifolia* root bark is effective on wound healing.

KEYWORDS: *Madhuca longifolia*, excision wound.

1. INTRODUCTION & LITERATURE REVIEW
Plants are being an effective source of both traditional and modern medicines are genuinely useful for primary healthcare. The medicinal plants produce wide range array of bioactive molecules and rich source of medicines (Agharkar, 1991). Traditional healing systems around the world that utilize herbal remedies are an important source for the discovery of new antibiotics (Okpekon, et al., 2004).

Madhuca longifolia from Sapotaceae family is widely used by the traditional medical practitioners for curing various diseases in their day to day practice. As well as these it composes wound healing property which has not been proved scientifically so far. Therefore, researcher was conducted to this study to prove the wound healing activity in scientific method. *Madhuca longifolia* is enhanced wound healing which is mentioned in the text book Kunapadam Part-1 (Porut Panpiyal) written by Murugesamuthaliyar, it was stated that the plant is applicable for wound. The objective of the study was to identify the effectiveness of *Madhuca longifolia root* bark powder in wound healing activity in Wistar albino rats.

Origin, Distribution and chemical composition
Madhuca, also known as butter tree, is a deciduous tree, 20 meters in height with a spreading top. It has thick leathery leaves and small, fleshy, pale or dull white musk-scented flowers in clusters near the end of branches (Bakhr, 2001). Young parts are pinkish white. Leaves are clustered near the ends of branches, simple, alternate, stipulate, elliptic-oblong, coriaceous, pubescent when young, almost glabrous when mature. Flowers: regular, bisexual, pale yellow, appearing with young leaves and below them, solitary in axils of small

deciduous bracts, pedicles 5-6.2 cm long. Fruits: outer pair nearly glabrous, inner pair finely tormentors. Seeds are large, 4 cm long with moderately hard testa, elliptical, flattened one or two sides, brown and shining when mature. Maximum germination occurs when the seeds are fully mature. Oil: fatty acid composition: palmitic, stearic, oleic and linole (Jayaweera, 2006).

2. MATERIALS AND METHOD

1.1. Introduction of Study

It is comparative animal experimental study conducted at Trincomalee campus, Eastern University.

1.2. Preparation of plant material

Madhuca longifolia was collected at Trincomalee district & taxonomically authenticated by supervisor. Debris and dust were removed from bark. It was washed thoroughly in tap water. After that it was kept in shade for 1 week. Then it was made as powder by using mechanical grinder. The powder were stored in air tight glass container and labeled.

1.3. Adaptation of animal

Albino rats were habitat in well ventilated room fed with pellets twice a day and watered per hour. Female Wistar Albino rats weighing 200 to 250 g were used in the study. Animals were divided into three groups of each with 3 animals. Group A consider as control, group B consider as standard, group C is *Madhuca logifolia* root bark fine powder treated group.

1.4. Excision wound model

The surgical interventions were carried out under the sterile conditions using ketamine anesthesia (30 mg/kg, IP). Hairs were removed from the dorsal thoracic region of the rats. A circular wound of approximately 500 mm^2 was marked on the back of the rat by a standard ring. Full thickness of the marked skin was cut carefully. Then Animals were kept in separate cages.

1.4.1. Treatment for group

The animals were divided into 3 groups of 3 each. The animals of group1 were left untreated and considered as control. Group 2 served as standard and received Cloxacillin powder. Group 3 was considered as test and treated with prepared test drug.

1.4.2. Treatment procedure

Powder of test drug and standard drug were topically applied 500mg every day and bandaged starting from the day of operation, till complete epithelialization up to 14 days.

1.4.3. Recording and observation

Data was collected every three days from each three groups and recorded. The following items were selected under the follow up chart for this study from Bates-Jensen Wound assessment Tool which is easy to obtain the statistical results (Pradhan, 2013).

3. RESULTS

3.1. Wound size

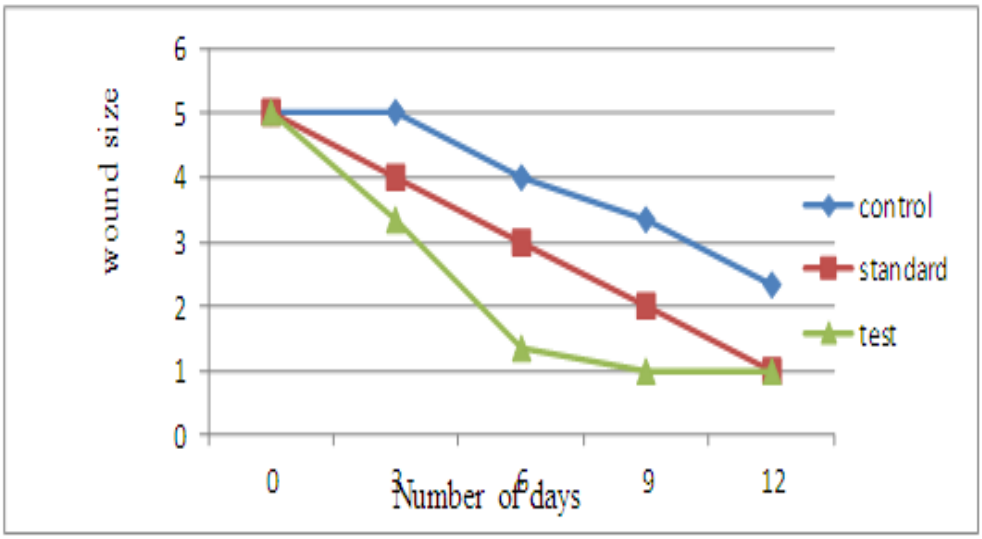

Figure 3-1 wound size variation with time.

Figure 4-1 shows the wound size variation with time in control, standard and test groups. Before treatment all groups have been observed as 5 according to the score.

3.2. Exudates amount.

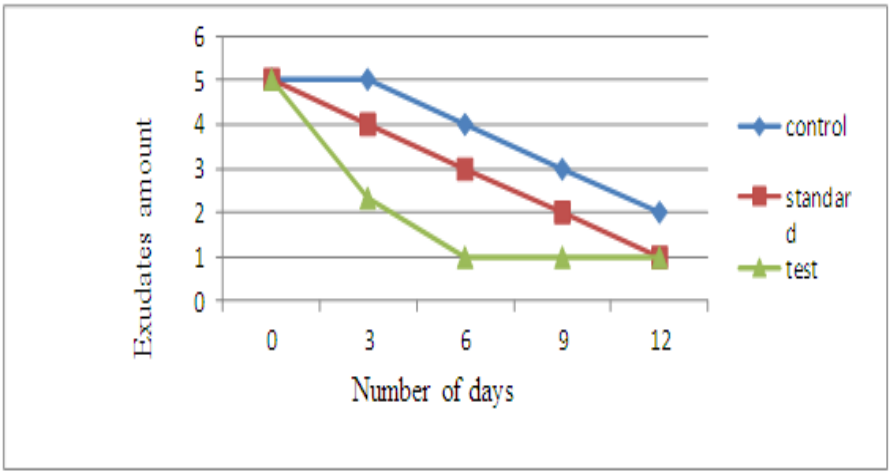

Figure 4-2 Exudates amount variation with time.

Figure 4-2 shows, exudates amount variation with time in control, standard and test groups. Before treatment all groups have been observed as 5 according to score.

3.3. Exudates type

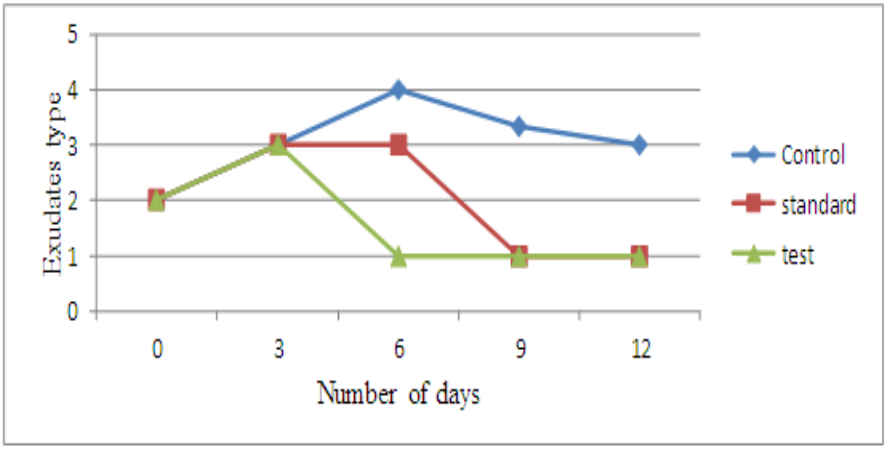

Figure 4-3 Exudates type variation with time.

Figure 4-3 shows the exudates type variation with time in control, standard and test groups. Before treatment all groups have been observed as 2 according to the score.

3.4. Wound edges

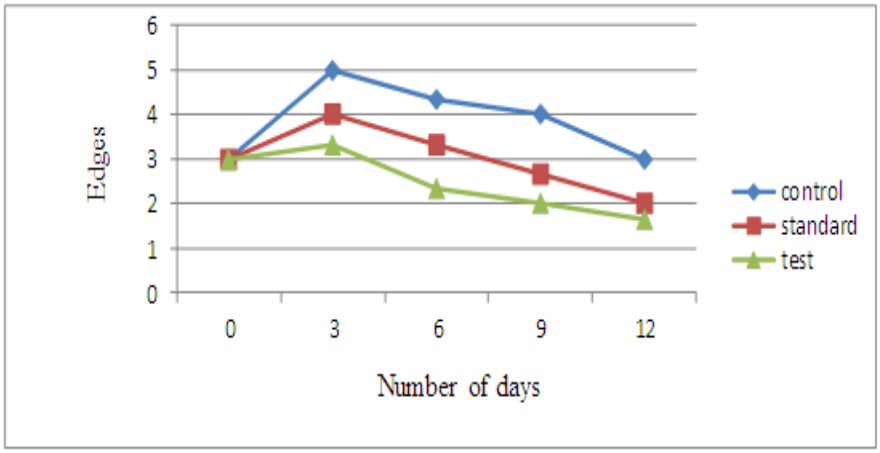

Figure 4-4 wound edges variation with time.

Figure 4-4 shows the wound edges variation with time in control, standard and test groups. Before treatment all groups have been observed as 3 according to the score.

3.5. Necrotic tissue type

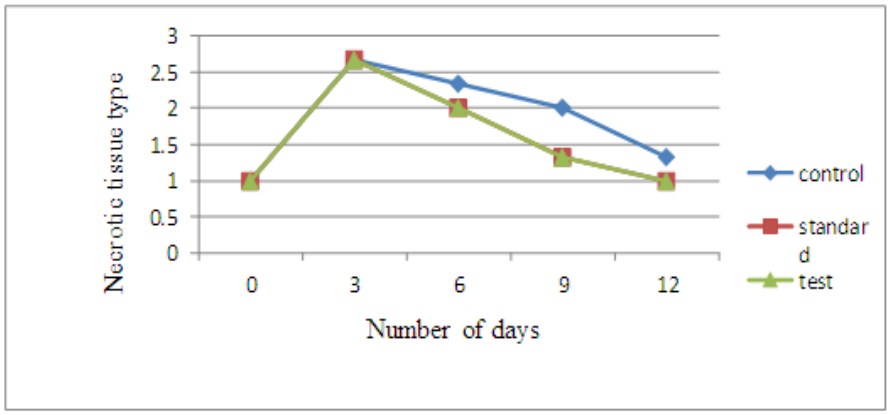

Figure 4-5 Necrotic tissue type variation with time.

Figure 4-5 shows the necrotic tissue type variation with time in control, standard and test groups. Before treatment all groups have been observed as 1 according to the score.

3.6. Skin color surrounding the wound

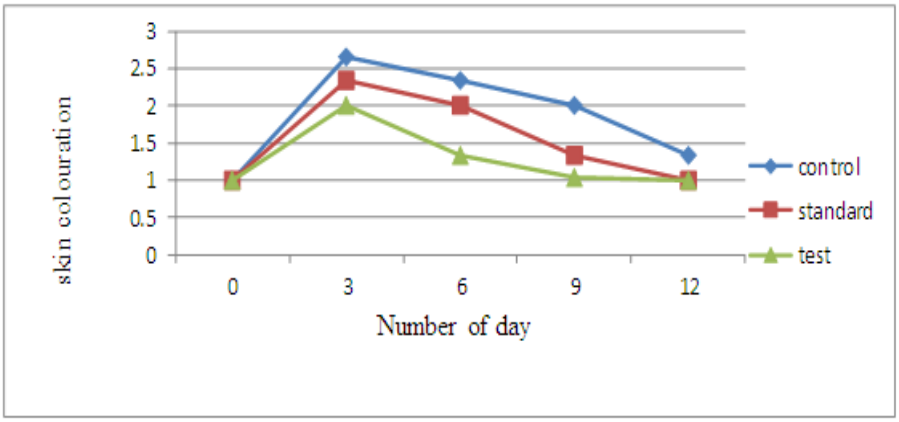

Figure 4-6 skin color variation with time.

On 3rd day test group has been showed 2 while stranded has showed 2.333 and control 2.666 .6th day observation has noted as test group 1.333, stranded is 2 and control is 2.333 according to the score. 9th day test group score is 1.033, stranded score is 1.333 and control group has noted as 2. Final day, both test group and stranded readings could be taken as 1 while control group score is 1.333.

3.7. Overall Therapeutic effect of treatment.

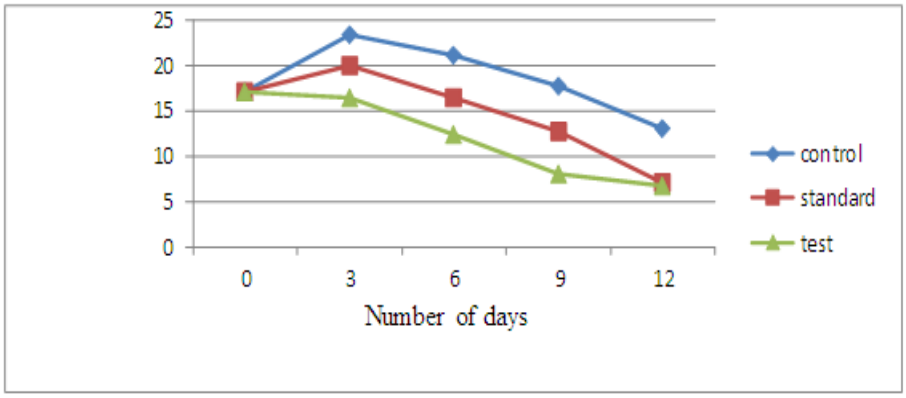

Figure 4-7 overall therapeutic effect with time.

Figure 4-7 shows the overall therapeutic effect with time in control, standard and test groups. Before treatment all groups total mean value have been observed as 17 according to the score.

3.8. Unit healing time

Table 4-1 Unit healing time of standard and test group

	Unit healing time (day/cm^2)
Test group	2.65
Standard group	3.09

Table 4-1 shows unit healing time of standard and test group. In test group UHT is 2.65 and standard group UHT is 3.09.

4. DISCUSSION

The stanza about general character of *Madhuca longifolia* plant indicated that, root bark can cure wound (Murugesamuthaliyar, 2013) therefore I selected root bark of this plant for the research study. Chemically it contains Saponin, steroids, triterpenoids, tannins, flavonoids, alkaloids, edible fats cyanogen and glycosides. (Ramadan, et al., 2006). All parts of *Madhuca longifolia* are medically important in traditional system of medicine.

Control group, wounds were observed mild reduction in size due to physiological healing, even though, there were wounds remained not healed. Gradual healing process was taken place in standard group, but marked reduction in size of wounds was observed in test group. In test group, Size of the wound was gradually decreasing up to third day, afterwards there was a sudden fall in size and it attained minimum level.

In control group, wound showed large amount of discharge up to 3rd day. Then it was gradually decreasing, but wound had not dried completely until last 12th day .There was scanty amount of wound discharge but no considerable discharge. Exudates amount of wound in Standard group, initially wounds showed large amount of discharge then gradually it was decreasing and completely dried up in 12th day. The exudates amount of wound in Test group, it was presented with large amount discharge initially, and then wounds were completely dried in 6th day. It showed that test drug is able to promote dryness of exudates in wounds with minimum duration than standard drug.

In control group, initially wounds showed bloody discharge .On 3rd and 6th day observation there was watery pink color discharge. 12th day observation there was presence of watery pink discharge. In standard group, initially wounds showed bloody discharge. On 3rd and 6th day observation there was watery pink color discharge. On 9th day observation Wounds were completely dried, no discharge. In Test groups, it expresses that the wounds showed bloody discharge initially, on 3rd day pink watery discharge. On 6th day Wounds were completely dried, no discharge.

Before treatment, all groups, the wound edges variation with time in control, standard and test groups have been observed as well defined not attached to wound base. On 3rd day test group has been showed well defined, not attached to wound base while stranded has showed well defined not attached to wound base, rolled under thickened .and control showed well defined, fibrotic scarred . 6th day observation has noted as test group distinct outline clearly visible attached with wound base, stranded showed well defined, not attached to wound base and control showed well defined not attached to base rolled under thickened. 9th day test group showed indistinct diffuse non clearly visible, stranded showed distinct outline clearly visible attached with wound base and control group has noted as well defined not attached to base rolled under thickened. On 12th day test group showed indistinct diffuse non clearly visible while stranded has showed distinct outline clearly visible attached with wound base and control showed well defined not attached to wound base.

Before treatment, all groups, the necrotic tissue type variation with time have been observed as non-visible necrotic tissue. On 3rd day all three groups have been showed loosely adherent yellow slough. On 6th day observation has noted as test and stranded groups showed gray non visible tissue and control showed loosely adherent yellow slough. 9th day test and stranded groups showed gray non visible tissue and control group has noted as no adherent yellow slough. Final day, both test and stranded groups showed non visible necrotic tissue while control group showed gray non visible necrotic tissue.

Before treatment all groups have been observed as normal pink color. On 3rd day test group has been showed bright red in color while stranded has showed hypo pigmented skin and control showed dark red in color. On 6th day observation has noted as test and stranded groups showed bright red in color and control showed dark red in color. 9th day test and stranded groups showed pink color and control group has noted as bright red in color.

The overall therapeutic effect of treatment with time in Control group has been showed continue recovery due to physiological healing during the experiment. The Test and Standard groups have been showed considerable minimum duration for complete wound healing. Test drug effectiveness was calculated as 60.78% while stranded drug effective was 58.82%. It seems more effectiveness in test drug comparing with stranded.

The unit healing time of standard and test group .Wound healing was assessed by UHT (Unit Healing Time) and scoring of signs and symptoms (Pradhan, 2013). Unit healing time of the test drug is 2.65 while stranded was calculated as 3.09. The time duration for complete

wound healing of the standard drug was observed within 14 days, but for test drug, it was only 12 days. It showed that Test drug has faster recovery than Standard drug. The result showed the healing activity improved in wound with days of treatment. Experiment studies showed significant results compared with standard and control group.

In the present study, wound healing activity of *Madhuca longifolia* root bark powder was studied and the results of the present study suggests that powder application of the plant root bark has shown more significant wound healing activity in excision wound model with comparing standard drug.

Taste of Astringent is responsible for wound healing, which are reduce the Exudates amount, promote the dryness of wound and enhance the wound contraction. In Siddha treatment there are so many drugs for wound healing contain Astringent and also hot potency. *Madhuca longifolia* root bark *also* contains *Suwai*-Astringent, *Thanmai*-hot potency, *Pirivu*- Pungent (Murugesamuthaliyar, 2013). So it is clearly denotes the *Madhuca longifolia* root bark powder is effective on wound healing.

5. CONCLUSION

The effectiveness of the test drug was 60.78%. The Unit healing time for the test drug was 2.65. The readings of Drug effectiveness and unit healing time leads to the conclusion of that, the test drug is more effective for wound healing than stranded. The stanza about General character of *Madhuca longifolia* which is mentioned in kunapadam text book (Porut Panpiyal) written by Murugesamuthaliyar was proved by scientific study.

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