ABSTRACT

Introduction: Untreated cuts in some period of time, caused by various reasons, can be a problem. Alternative therapy used to speed up the healing of the cut is Nigella Sativa (NS). Objective: to investigate the number of fibroblast and collagen by giving 40% and 60% NS seed extract on the cut.
Methods: post test only control group design, 48 male balb/c strain rats were divided into 2 large groups, with each cage contained 24 rats. Each group then divided to 4. Each group is subdivided into 4. Nigella sativa seed extract ointment with a concentration of 40% (NS-40) and 60% (NS-60) smeared 3x daily for 7 days (fibroblasts) and 14 days (collagen). Evaluation derived from preparations using HE staining and Masson's Trichrome respectively.
Results: Mann Whitney analysis shows that the number of fibroblasts and collagen in the CG group, NS-40, and NS-60 Higher than CN, p < 0.05. The number of fibroblasts in the NS-40 and NS-60 was not significantly different compared to the CG, as well as CN-40 with CN-60, p > 0.05. Sedangkan jumlah kollagen pada CN-60 lebih tinggi bermakna dibanding CN-40, p < 0.05.
Conclusion: administration of Nigella sativa seed extract ointment with a concentration of 40% and 60% in cuts for 7-14 days increase the number of fibroblasts and collagen, resulting in a shorter healing period.

Keywords: black cumin seed extract ointment (Nigella sativa), fibroblasts, collagen, cuts.
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million cases, amputation 0.2 million per year, 0.6 million carcinoma per year, melanoma 0.1 million, skin cancer complications in 0.1 million cases (Med Market Diligence, 2009).

The wider community povidone iodine is usually used to treat cuts, but the use of povidone iodine can cause allergies and hinder the process of wound healing and scarring, resulting in the need of other treatment alternatives that better (Henry, 2007). Nigella sativa (NS) or have a popular name of black seed (black seed) or Seed (Habbat al baraka) turned merupak an plant that has many benefits. One of the benefit is anti-inflammatory role in the wound healing process (Parakh, 2010). Nigella sativa can stimulate the activity of cytokines Macrofag Activating Factor (MAF) thereby improving the function of the macrophages plays a role in cellular immune system. Nigella sativa also contains saponins that play a role in helping the process of wound healing (angiogenesis) via Vascular Endothelial Growth Factor (VEGF). In addition, the content of zinc in the NS can also help the cut healing in experimental animals. Zinc is known to have the ability to help accelerate the healing of wounds, both as an important activator of enzymes in the formation of protein as well as the bodily defense. Zinc function in replication of fibroblasts, the synthesis of collagen, and also collagen crosslinking in tissue injuries (Niluh, 2012). Zinc deficiency can result in decreased Tensile Strength and delays in wound closure, therefore lubrication NS seed extract is expected to prevent the possibility (Dharma et al., 2010).

This study was conducted to prove the increased number of fibroblasts and collagen in mice balb/c with cuts after administration seed extract ointment NS in dosage by 40% and 60%.

METHODS

In experimental research with draft “post test only control group design”, as much 48 strain male mice balb/c, are divided into two major groups, each consisting of 24 male for checking the number of fibroblasts and collagen. A total of 2 4 individuals in each group were then divided into 4 groups randomly, each consisting of 6 rats. Negative control group (CN), only distilled water smeared. Garamycin group (GG) smeared with ointment garamycin. Group N igela s Ativa (NS-40) ointment smeared NS 40%. Group N igela sativa (NS-60) smeared with ointment NS 60%. Before getting ointment, The backs s ach Mice Made cuts Along 2 cm?. Ointment smeared 3x daily for 7 days (fibroblasts) and for 14 days (collagen). Day 8 and 14 skin tissue sampling was conducted to make preparations, then on each of the preparations stainings with HE and T richrome were carried out. A statistical analysis made considering the data obtained is not normal and homogeneous. Statistical analysis was considered significant when p-value <0.05. This research was conducted in the laboratory of Chemistry and Biology, Faculty of Medicine, Sultan Agung Islamic University and the Laboratory...
RESULTS

After 7 and 14 days of treatment, the results and means were obtained as shown in figure and table 1. The statistical analysis showed that the highest amount of fibroblasts were found in the NS-40 group followed by the NS-60, CG, and the lowest group in the CN group. The highest amount of collagen was found in the NS-60 group followed by the NS-40 group, CG, and the lowest was the CN group. The result of Kurkall Wallis statistic analysis showed that there was a significant difference between the groups, p <0.05.

Number of Fibroblasts

Mann Whitney statistical analysis showed that the number of fibroblasts in the NS-60 and CG groups was significantly higher than in the CN group, p <0.05. While the number of fibroblasts in the NS-40 group tended to be significantly higher than CN, p = 0.06. The number of fibroblasts in the group of NS-60 and NS-40 compared to the CG group, although higher,
but not significant, p > 0.05. Demian did the number of fibroblasts in the group of NS-40 NS-60, although higher, but not significant, p > 0.05 (figure 4).

**Number of Collagen**

In accordance with the results of statistical analysis Mann Whitney, the amount of collagen in the group of NS-60, NS-40, and CG were significantly higher than the CN group, P <0.05. The amount of collagen in the NS-60 and NS-40 groups were significantly higher than in the CG group, p <0.05. The same thing happened in the NS-60 group compared to the NS-40 group, p <0.05 (figure 5).

**DISCUSSION**

These results indicate that administration of NS seed extract at a concentration of 40% proved to increase the number of fibroblasts in mice suffered from cuts and healing. The results of this study are supported by an invitro study conducted by Rachman et al. showed that Nigella sativa in vitro can increase the proliferation Human cells gingival fibroblasts (HGF) monolayer slices are created so that the closing wedge (Rahman et al., 2014).

Healing effect caused by seed extract NS allegedly associated with the saponin contained in NS. Various studies have shown that saponins can help the process of wound yembuhan pen, so that the healing of wounds shorter duration. This is because saponin have an effect in stimulating the Vascular Endothelial Growth Factor (VEGF), which can then be Activate Transforming Growth Factor beta (TGF- B) and Platelet Derived Growth Factor (PDGF) that resulted in the proliferation of fibroblasts (Scultz, 2003). Saponins may also act as immunomodulators that can induce macrophage cell activity, limfosit T, interleukin-1h, and Tumor Necrosis Factor [(TNF-α) (Suryadi, 2012)]. Furthermore, various s el in the immune system secrete growth factors and cytokines to accelerate wound healing (Surkhai et al., 2011). Moreover, saponin is also proved able to stimulate the synthesis of fibroblasts by fibronectin (Froschle, 2004). This is in line with studies reported by Nova and colleagues. Studies conducted by Nova indicated that administration of nigela satin extract proved to increase the number of fibroblasts and macrophages activity in healing wounds from rat tooth extraction (Nova et al., 2013). In addition, N S also contains essential fatty acids such as linoleic acid and oleic acid to maintain the integrity of the epidermis as a barrier on the skin, maintaining the water content, and accelerate wound healing by enhancing the migration of immune cells to the wound tissue. Additionally, NS containing flavonoid known to increase the amount of collagen and stimulate macrophage activity to trigger epitelialisation process, increase the production of extracellular matrix, growth factors, cytokines, and angiogenesis through the release of growth factors such as keratinocytes Growth Factor [(KGF) (Ambiga et al., 2007)]. However, this study has a limitation that is not decisive where the active compound contained in the seed extract Nigella sativa that stimulates healing of cuts in mice faster. Therefore it is still require further research to determine the content.

In this study, the number of fibroblasts in the group of 40 tend to be more abundant NS- compared to the NS-60 group on day 7. It can be deduced that the number of fibroblasts in the NS-60 group were less a result of the wound healing process that lasts shorter, so many fibroblasts that have been converted into fibrosit then collagen. The statement is in accordance with the results of the examination of collagen at day 14 in the study, which showed that the amount of collagen in the NS-60 group were significantly higher than those in NS-40 and the group garamycin. Referring to the results of this study, then indirectly it can be said that the total collagen starts to increase at day 7, and gradually reached its peak at day 14.

**CONCLUSION**

Based on the results of this study as a whole it can be concluded that administration of black cumin seed extract ointment or Nigella sativa with a concentration of 40% and 60% in cuts for 7-14 days can increase the number of fibroblasts and collagen, resulting in a shorter healing cuts. Nigella sativa With a 60% better concentration in increasing the amount of fibroblasts and collagen than NS with concentrations of 40% and garamycin.

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