

Histological Evaluation of the Effect of Topical Application of Honey on Healing of Chemically Induced Gingival Ulcer (Experimental Study on Rabbits)

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ABSTRACT

Background: Wound healing is a complex process, which is influenced by several factors. The use of natural products as an alternative treatment has been on the rise in the last few decades. Honey is a natural product that has been recently introduced in new medical practice. The aim of this study was to evaluate the effect of topical honey dressing on healing of chemically induced oral gingival ulcer by histological analysis.

Materials and methods: Twenty-six New Zealand rabbits were used and divided into control and treated groups, wound was created on the rabbit oral mucosa by applying a piece of filter paper ($\phi 5$ mm) soaked with 50% acetic acid for 2 minutes to the mandibular gingival mucosa and then experimental groups treated by daily application of natural honey on the ulcer site for 4 days then the sites of the ulcer were examined histologically .

Results: Honey showed marked active anti-inflammatory effect as a natural agent on a chemically induced oral ulcer in treated animals, histological examination of biopsies from ulcers after four days of treatment showed marked difference in the rate of healing process and epithelialization of ulcers site between the two groups.

Conclusions: honey has a clear effect on the healing process of oral gingival ulcer.

KEY WORDS

honey, gingiva, chemical ulcer

المستخلص

المقدمة: التئام الجروح هي عملية معقدة، والتي تتأثر بعدة عوامل. إن استخدام المنتجات الطبيعية كعلاج بديل أخذ في الازدياد في العقود القليلة الماضية. العسل هو المنتج الطبيعي الذي تم عرضه مؤخرًا في الممارسة الطبية الجديدة. الهدف من هذه الدراسة هو تقييم تأثير العسل الطبيعي على شفاء قرحة اللثة الفموية الناجمة كيميائياً عن طريق التحليل النسيجي.

المواد والطرق: استخدمت ست وعشرون من الأرانب النيوزيلندية وتم تقسيمها إلى مجموعة السيطرة ومجموعة المعالجة، وتم إنشاء الجروح الكيميائية على الغشاء المخاطي للفم في الأرانب عن طريق تطبيق قطعة من ورق الترشيح غارقة مع حمض الخليك 50٪ لمدة 2 دقيقة على اللثة السفلية ثم تم معالجة مجموعة المعالجة من خلال التطبيق اليومي للعسل على موقع القرحة لمدة 4 أيام. وبعد ذلك تم فحص موقع القرحة نسيجياً.

النتائج: أظهرت نتائج الفحص النسيجي بان العسل مضاد فعال للالتهابات لعلاج قرحة الفم في الحيوانات في مجموعة المعالجة وأوضحت النتائج الفرق في معدل الاندمال وتشكل النسيج الظهاري وعملية الشفاء من القرحة بين المجموعتين. الاستنتاجات: العسل له تأثير واضح على معدل عملية الشفاء من قرحة الفم.

الكلمات الرئيسية:

العسل الطبيعي، التقرح الكيميائي، اللثة.

INTRODUCTION

The oral cavity is covered by oral mucosa. The oral mucosa consists of two components: (the oral epithelium) which is stratified squamous epithelium and an underlying connective tissue layer that is called (the lamina propria). Functions of the oral mucosa include protection, sensation and secretion (Antonio. Nanci,2008). Maintaining the healthily of the oral mucosa is very important not only for oral function but also for general health.

Wound healing is a physiological process initiated and influenced by many factors. This process can be organized into four stages:

1-haemostasis,2-inflammation,3- proliferation (consisting of granulation, and epithelization), and 4- finally remodeling (Diegelmann and Evans, 2004).

Honey has been a combination of low water activity, low pH, and the generation of hydrogen peroxide(Mundo et al., 2004), these give it the antimicrobial effectiveness (Cooper and Molan, 2001)

It is now understood that honey is not just sugar syrup with certain physical properties that make it suitable as a wound dressing material, but it is a biological wound dressing with multiple bioactive components that can accelerate the healing processes. (Molan,2011)

MATERIALS AND METHODS

Twenty- six New Zealand white rabbits (2.5–3.0 kg, male) fed a normal diet and maintained under a 12-hour-light/12-hour-dark cycle at 22°C, were divided into(thirteen rabbits as experimental and thirteen as control). Chemical wound was created on the oral mucosa of the rabbit by applying a piece of filter paper ($\phi 5$ mm) soaked with 50% acetic acid for 2 minutes to the mandibular gingiva. Ulcer formation was clear after 24 hours, and then natural honey dressing topically applied to the wound daily. The size of ulcer was measured on day 4 after wound formation, and the tissues around the wound were obtained for histological examination. Excised tissue

was fixed in formalin and embedded in paraffin. Sections were cut at 5 µm thickness, deparaffinized and stained with hematoxylin and eosin.

RESULTS

The clinical manifestations:

Twenty-four hours after acetic acid application, the animals develop erythematous swelling about 1cm in diameter, ulcer of the gingival surface was seen in all animals. The animals were suffering from pain during examination; redness, and pus discharge without signs of improvement until the end of study this condition observed for the following 4 days in control groups throughout the period of the study. While the experimental groups, the animals remained healthy, without any evidence of infections.(as a result as honey effect).

Histological manifestations

Control groups:

At 4 days: The microscopic examination showed the following characteristics:

At the epithelial level: areas of mucosa without epithelium, covered by ulcerations, exudate with cell necrosis and infiltration by inflammatory cells (Figures-1& 2).

Experimental groups

After 4 days of daily honey dressing the histological section shows the reepithelization process, and in this period we showed a typical histological picture of proliferative phase with present of layers of fibroblasts and new vessels, with marked epithelial-connective tissues interface, and at the level of lamina propria we showed newly formed collagen bundle (Figures-3&4).

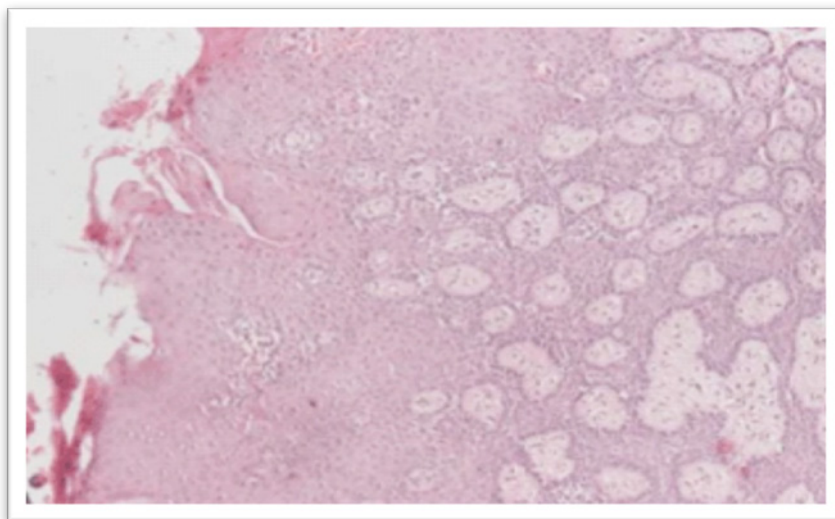


Figure 1 – Free gingival mucosa with area of ulceration, exudate with cell necrosis (the arrow) (H&E stain, ob. ×10). (Control groups).

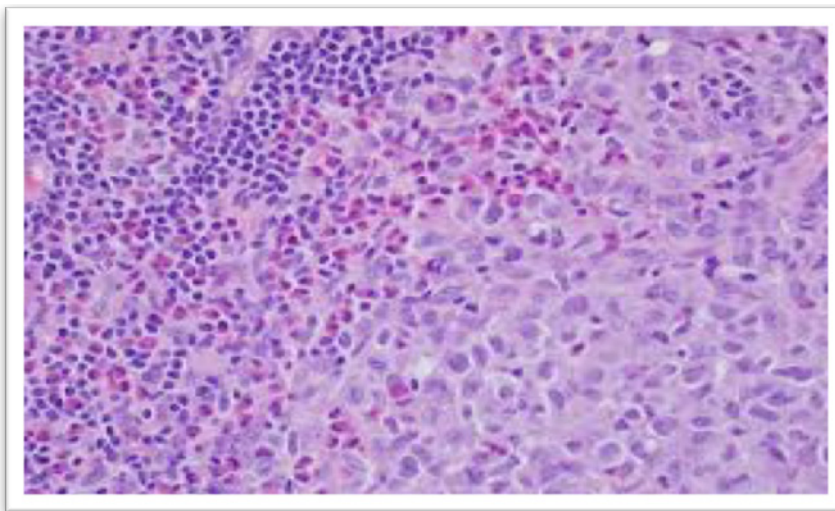
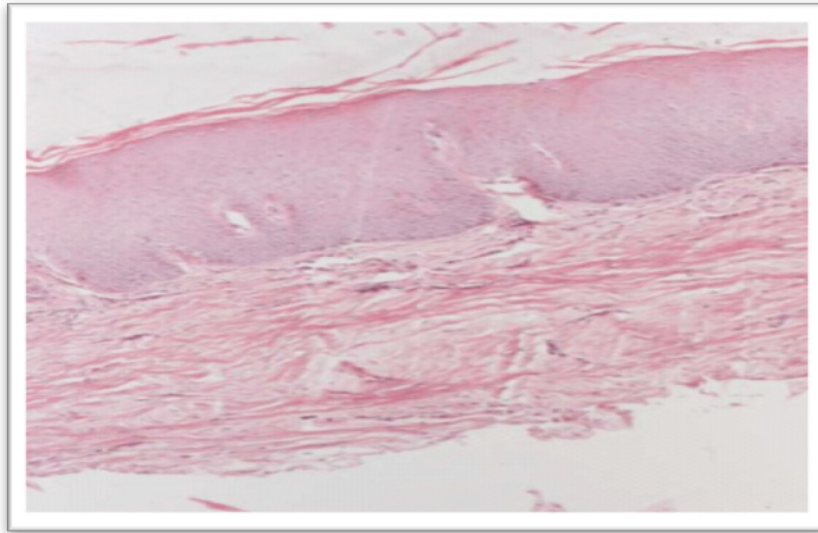
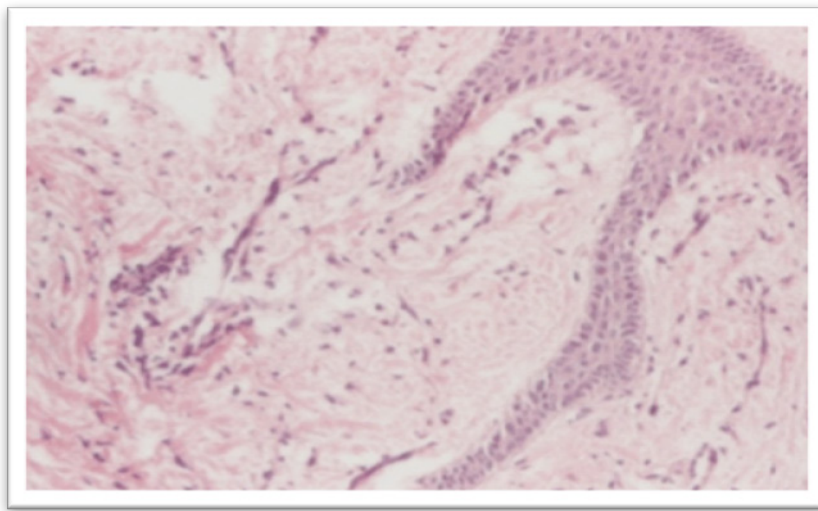


Figure 2- H&E stained section of control group, notice site of ulceration and notice huge of inflammatory cells (the arrow). (Ob. X20).



Figures 3 . Healed epithelial of gingiva after complete treatment show keratinized layer of gingiva (H&E stain, ob. $\times 10$). (Experimental groups)



(Figures 4): layers of fibroblasts cells (black arrow) and new collagen fiber (blue arrow) (H&E stain, ob. $\times 20$). (Experimental groups).

DISCUSSION

The honey has a therapeutic effect on the treatment of gingival ulcer in agreement with the finding of (Iftikhar I. et al 2010). The results show that the healing properties of the honey include stimulation of tissue growth, and enhanced epithelialization, these findings confirm the studies of (Noori et al 2011).

In honey dressed ulcers, there is early subsidence of acute inflammatory changes, good control of infection with accelerated ulcer healing was observed, these findings confirm the studies of (Liza G, and Ovington, 1999) and (Molan, 2001). Honey has been found to improve healing time as in mild to moderate superficial burns and ulcers, (Evans and Flavin, 2008;).

The pus and inflammatory exudates become relieved by the honey thus protecting the underlying

tissue and improved normal healing and the epithelialization and no toxic effects have been reported with honey usage this is in agreement with (Aldouri, 2003).

The result of our study that the honey was an excellent accelerator of ulcer healing and has a better effect in the treatment of mucosal ulcers was in agreement with the finding of (Al-Waili, 1999). The honey is a sticky viscous material that makes it cover and adherent to the ulcer, so that it was preventing the secondary infection as carob (Altaie, 1998).

CONCLUSION

The anti-bacterial, anti-inflammatory and anti-oxidant, with good physical properties of natural honey, makes it an accepted natural agent for healing of oral chemical ulcers without any side effects.

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