Comparative histophysiological study the effective treatment of induced eczema in rats that treated with Lanolin and glycerine with Vaseline in rats

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Abstract

The present study has been designed to evaluate the potent role of healing efficacy of lanolin (which is also called wool wax), is an oily product secreted by the sebaceous glands beneath the skin of sheep which is have anti-inflammatory, anti-microbial, skin protecting properties, a forty-eight mature albino rats (weighted 250-350 g) were randomly assigned to two groups; control group and eczema group which is induced by 99.9% pure acetone. By squad cotton and put it on shaved skin; after 3 days of acetone application the eczema group were divided randomly into two equal groups each group 16 rats, treat these 32 rats with glycerin and lanolin, after 24h five rats from each group were sacrificed, and remaining rats were treated with glycerin and lanolin for 72h and then sacrificed at the third day, the histopathology there is a lot of histopathological changes occur in tissue section in eczematous skin in comparison with control section; there were obvious thickness and enlargement in the length of outermost layer of skin, also there is fissures and cracks in the epidermal layer due to the dryness of skin and aggregation of inflammatory cells. And as well as keratinization of epithelial layer and changes in skin color, the results indicate there were optimum healing effect of lanolin group in compare with glycerin group after 72h, Showed back the outer most layer of the skin to its normal length, also decrease in inflammatory cells number.

Key words: - Lanolin, Eczema, Glycerin

دراسة نسجية فسلجية مقارنة للعلاج الاكزيما المستحدثة بالالولين و مزيج الكليسرين مع الفازلين في الجرذان

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الخلاصة

صممت هذه الدراسة ليبيان مدى التاثير القوي لللانولين (الذي يسمى أيضا بشعير الصوف أو دهن الصوف و يفرز من الخندقية تحت جلد الاشجار الالوفيات (الذي يملؤها صمعية مضادة للالتهاب و مضادة للميكروبات بالإضافة إلى خصائص وقاية الجلد و يعمل على ترطيب الجلد الجاف بشكل فعال جدا ) بالمقارنة مع الكليسرين في علاج حالات الاكزيما المستحدثة بواسطة الاستيتو في الجرذان). استخدم في هذه التجربة 48 جرذ ناضج تتراوح أوزانهم من 250 غم الى 350 غم و تم توزيعها بشكل عشوائي في مجموعتين، مجموعة السيطرة و مجموعتان مستخدمة لعلاج حالات الاكزيما المستحدثة بواسطة الاستيتو في الجرذان، استخدام في هذه التجربة 48 جرذ ناضج تتراوح أوزانهم من 250 غم الى 350 غم و تم توزيعها بشكل عشوائي إلى مجموعتين، مجموعة السيطرة و المجموعة المصابة بالاكزيما في علاج حالات الاكزيما المستحدثة بواسطة الاستيتو، وضع عليه الجلد بعد حلق الشعر، بعد 3 أيام من استخدام الاستيتو يتم عزل المجموعة المعالمة (المقاومة للاكزيما) تقسم عشوائيا إلى مجموعتين كل مجموعة 16 جرذ، ثم علاج كل ال 32 جرذا، 16 جرذ بواسطة الاستيتو و 16 جرذ بواسطة الكليسرين، تمت التضحية بثمانية جرذان من كل مجموعة معدلة بعد مرور 24 ساعة، تم الاستمرار بعلاج البقية لمدة 72 ساعة ثم تمت التضحية في اليوم الثالث.

الخليط النسيجي أظهر أن هناك الكثير من التغييرات النسيجية حصلت في مجموعة الاكزيما بالمقارنة مع مجموعة السيطرة حيث هناك نقص واضح في الطبقة الخارجية للجلد و وجود تشققات و تقريض في الطبقة الظهارية و تغير لون الجلد، النتائج اظهرت عودة النسيج إلى حالته الطبيعية في مجموعة الالولين 72 ساعة بالمقارنة مع الكليسرين 72 ساعة.
**Introduction**

Lanolin, which is also called wool wax, wool yolk, or wool grease, is an oily product secreted by the sebaceous glands beneath the skin of sheep. The lanolin is an agricultural complex obtained from sheep wool. It has been recognized two millennia ago. It is a yellow to pale brown color with an oily / creamy texture [1].

The lanolin can be used in several technical applications. It can be used by the pharmaceutical industry and in cosmetic products specifically lanolin derivatives as well as in personal care and industrial section. The lanolin is for topical uses only [2]. It can absorb more than 200% of its weight in water (WW). (Sengupta, and Behera, 2014).

The Lanolin, which is have emollient, anti-inflammatory, anti-microbial, barrier-repair, skin protecting properties. It has an effective moisturizing property with a long history of beneficial use and safe in topical preparations and Moreover the lanolin has been shown to creates an air permeable barrier and promotes moist wound healing when applied to an injured skin, and promotes moist wound healing and creates an air-permeable barrier when applied to eczematous hand. It is certainly having antimicrobial, anti-inflammatory, barrier repair and skin-protecting properties [3].

**Material and methods**

A 48 mature male albino rats weighing (250 – 350) g, were obtained from vet. Medicine Laboratories. (department of physiology and pharmacology, faculty of veterinary medicine kufa - Iraq). The animals were housed in clean plastic cages (4 animals per cage) and kept in well ventilated under controlled temperature (between 23 ºC and 25 C) and 12 h light/dark cycle (light between 07:00 and 19:00). Food and water were freely available. Animals were fed with commercial food from the manufacturer green world company.

During the lab work in the lab animal house use lab coat, cloves and face mask which is surgical disposable, the rats in sterile surgical area, the rats were anesthetized with ketamine – xylazine mixture (0.1 ml) for each rat by intramuscular injection with insulin syringe, after 5minute use the permanent marker to border the shaving area, and then the hair on their dorsal area was shaved. Shaving with (braun) shaving machine and then with (venous) shaving Razor followed by a soft hair removing cream.

Squad the cotton with 99.9% pure acetone and put it on skin for 5 min. for 3 days subsequently to induce eczema according to (Zhou, & Sui, 2019). the use of acetone because of the acetone principle work on skin is alteration the cutaneous barrier of the skin.

Apply lanolin on third group of rats after 72h, and also apply glycerin on fourth group after 72h. They take constant weight of jelly Vaseline from (unilever company) and pure fluid glycerin with electronic balance then dissolve the Vaseline in water bath in flask, 50 c for 30 min. we use a spoon to mix the mixture, put the mix. In large container 400ml and cover it with parafum to prevent the oxidation, until use it the next day to treat the induced eczema. (Rohmani, et al., 2019).

Histopathological study of organ (skin) was fixed in 10% formalin directly until study. The of experiment rats were sacrificed and specimens from organ were enucleate immediately and tissue samples were taken from organ and fixed in 10% buffered formalin for 48 hours at room temperature. After fixation the tissues were dehydrated in a graded of alcohol concentration then clearing in two stages of xyline and embedded in liquid paraffin for 2 hours at 56Cº. The sectioning of tissue was done at 5 micrometers by microtome. Finally, dewaxed and stained with Harris Haematoxylin and Eosin (H&E). The section tissues were examined using X10, and X40 objective of light microscopy [4].

**Results**

The histological of the skin show the changes that occur in the dermal tissue of rat that have induced eczema by acetone as show keratinized, dry and inflamed skin. There is edema space in tissue parenchyma with blood vessel congestion and aggregation of inflammatory cells with hyperemia of tissue parenchyma. As well as the histological will
show the changes that occur after treatment application of four groups of rats.

Figure 4 – 7. The histopathological section of skin in normal adult rat (control group) X10. Show the normal squamous epithelium (epidermis) and a fibroelastic connective tissue layer (dermis). The hair follicle also shows up in this section. The tissue is stained with H & E stain.

Figure 4 – 8. The histopathological section of skin in rat has eczema which is induced by acetone (eczema group). Show the apparent thickness and enlargement in the length of outermost layer of skin (green arrow). Also, there is fissures and cracks in the epidermal layer due to the dryness of
skin (red arrow). As well as edema beneath dermal layer (white arrow). The section also shows hemorrhagic area (yellow arrow) and aggregation of inflammatory cells (blue arrow). And keratinization of epithelial layer (black arrow). As well as changes in skin color to be brownish. The tissue is stained with H & E stain (X10).

Figure 4 – 9. The histopathological section of skin in adult rat that treated with glycerin after 24h of eczema. Showed there is a little area of peeling of the keratinization layer by activity of glycerin (red arrow). And some droplet of glycerin fluid that has been absorbed by the skin (green arrows). But there is no significant changes with eczema group. The tissue is stained with H & E stain (X10).

Figure 4 – 10. The histopathological section of skin in adult rat that treated with lanolin after 24h of eczema. Showed there is a slight change in the epidermal layer as decreasing in the length of layer (black arrows). As well as slight peeling of the keratinization layer (red arrow). But there is still edema fond (yellow arrow). The tissue is stained with H & E stain (X10).
Figure 4 – 11. The histopathological section of skin in adult rat that treated with glycerin after 72h of eczema. Showed very thin layer in the epidermis thinner than the normal because of the peeling effect of glycerin (blue arrow). And there is a lot of peeling area (black arrow). But still not treated well. The tissue is stained with H & E stain (X10).

Figure 4 – 12. The histopathological section of skin in adult rat that treated with lanolin after 72h of eczema. Showed back the outer most layer of the skin to its normal length (black arrow). Also decrease in inflammatory cell number (yellow arrow). The skin in general almost return to its normal situation. The tissue is stained with H & E stain (X10).
Discussion
Histopathological control group analysis revealed normal skin with normal squamous epithelium and a fibroelastic connective tissue layer, according to histopathological evaluation [5]. The eczematous skin indicates acetone-induced damage. Show the apparent thickness and enlargement in the length of outermost layer of skin because of the effect of acetone which is destroy the profilaggrin protein leading to defect in skin barrier and increase in trans-epidermal water loss leading to dryness, as well as aggregation of inflammatory cells [6].

Also, there is fissures and cracks in the epidermal layer due to the dryness of skin. As well as edema beneath dermal layer. And keratinization of epithelial layer. As well as changes in skin color to be brownish color [7].

The histopathological sections stained with H&E of skin in adult rat that treated with glycerin after 24h of eczema. Showed there is a little area of peeling of the keratinization layer by activity of glycerin [8], the histopathological section of skin treated with glycerin after 72h of eczema. Showed very thin layer in the epidermis thinner than the normal because of the peeling effect of glycerin. The results showed that the glycerin has peeling effect on epidermal layer of skin [9].

Histopathological section of skin in adult rat that treated with lanolin after 24h of eczema. Showed there is a slight change in the epidermal layer as decreasing in the length of layer, because the anti-inflammatory effect of lanolin [2].

As well as slight peeling of the keratinization layer. The tissue is stained with H & E stain. [10], the histopathological section of skin in adult rat that treated with lanolin after 72h of eczema. Showed back the outer most layer of the skin to its normal length. Also decrease in inflammatory cell number, by effect of anti-inflammatory lanolin as well as skin reconstructive and repair of skin fissures and cracks [11], also, there was decrease in trans-epidermal water loss because of the moisturizing effect of lanolin [12], the skin in general almost return to its normal situation [13].

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