A Comparative Histological Study to the Effect of Adipose Derived Mesenchymal Stem Cells Versus Virgin Coconut Oil in Skin Wound Healing of Adult Male Albino Rat

Original Article

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ABSTRACT

Background: Skin wounds comprise a clinical load for many patients. Owing to their availability and ability for differentiation, adipose-derived stem cells (ADSCs) are considered a promising line in the tissue regenerative medicine. Virgin coconut oil (VCO) has been also reported to help in wound healing by decreasing time of complete re-epithelization. However, there was lack in literature about histological details.

Aim: The study aimed to compare the effect of ADSCs versus natural VCO in wound healing.

Material and Methods: Thirty adult male rats were used in three experimental groups, ten rats in each. All animals were exposed to excisional skin wound injury. Group I was used as wounded untreated group. Group II was injected intra-dermally with 1x106 ADSCs in four injection sites around the wound, while group III was treated with topical application of 0.5 gm VCO ointment. After 2 weeks, all rats were sacrificed. Macroscopic shrinkage of wounds was recorded and compared. Skin samples were harvested and processed for light microscopic examination using H. & E. & Masson's trichrome stains. Morphometric measurements were done for both epidermal thickness & collagen fibers area percentage. Data was analyzed statistically.

Results: Histological examination of group II revealed regenerated epidermis and dermal skin appendages. Masson's trichrome stained sections revealed thick collagen bundles running in different directions. In Group III, epidermis was thin with few developing dermal skin appendages. Masson's trichrome stained sections showed dense parallel bundles of collagen fibers. **Conclusion:** Virgin coconut oil enhanced skin wound healing. However, ADSCs proved to be more efficient in regaining mature skin structure.

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Key Words: Coconut oil, skin, stem cells, wound.

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INTRODUCTION

Skin wound patients are usually burdened with pain and sometimes with imperative hospitalization with subsequent high economic costs^[1]. Wound healing occurs through three overlapping stages: inflammatory stage; with hemostasis (first three days), proliferative stage; with angiogenesis & collagen fibers deposition (initiates from day 1 & reaches maximum by day 5 or 6) and remodeling stage, initiates 6 days after skin wound and can continue for long time^[2,3].

Bone marrow mesenchymal stem cells (BMSCs) were reported to acquire the ability of proliferation and differentiation, and increasing angiogenesis, neovascularization, so they might be considered as appropriate tool for wound healing^[4].

Nevertheless, it has been recorded that adipose derived mesenchymal stem cells (ADSCs) have a more promising future in the field of tissue regeneration. They have many advantages over the BMSCs due to their greater availability, less invasive harvesting techniques and higher cell yield, hence, they are considered as an ideal alternative^[5].

On the other hand, wound healing might be also facilitated by natural products^[6]. Virgin coconut oil, as one of these natural products, has been found to help in the wound healing process. It has been reported to decrease the time needed for full epithelization, stimulate production of collagen and increase collagen cross linking which was indicated by the notable increase in pepsin-soluble collagen^[7,8]. Although some studies have declared the effectiveness of natural components in wound healing, yet further detailed histological studies is still needed to document effectiveness in application^[6].

AIM

This study targeted to compare the histological effects of ADSCs versus a natural treatment as VCO in wound healing.

MATERIAL AND METHODS

Animals

Thirty male adult albino rats weighing 200 - 220 gm were obtained from the animal house of the Faculty of Medicine Ain Shams Research Institute (MASRI).

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Rats were kept in ordinary wire-mesh cages in a room temperature $(21\pm3 \text{ °C})$. They were provided standard rat diet and granted free access to water. They were housed for a week preceding the experiment in order to accommodate to the experimental conditions.

Preparing adipose tissue cell suspension and isolation of ADSCs

Adipose derived mesenchymal stem cells were obtained from stem cell unit, Histology Department, Faculty of Medicine, Ain Shams University. They were obtained from the inguinal groove adipose tissue of 6 wks old male rats. Then, they were washed using saline solution and incubated in Dulbecco's modified Eagle's medium. Centrifugation of the cell suspension with subsequent collection of the sediments of stromal vascular fraction was performed. Resuspension of isolated nucleated cells in complete culture medium supplemented with 1% penicillin-streptomycin was done. Incubation of cells was then done for 12-14 days as primary culture or upon formation of large colonies. On development of large colonies (80-90% confluence), cultures were washed two times by phosphate buffer saline. Cells were then trypsinized using 0.25% trypsin in 1mM of Ethylene diamine tetra acetic acid (EDTA) for 5 minutes at 37°C. Then centrifugation was done with subsequent re-suspension using serum-supplemented medium and incubated in culture flasks (50 cm2). On the 14th day, the adherent cell colonies were trypsinized and counted^[9].

Virgin Coconut oil was obtained from Emtinan ® Company, Cairo, Egypt. Coconut oil ointment was prepared by admix of coconut oil with Vaseline base in a ratio of 1:3 respectively^[10].

Induction of sterile excisional skin wound

Rats were anesthetized using an intramuscular injection of ketamine 40 mg/kg and xylazine 5 mg/kg. Shaving of the surgical area was done by an electric razor with subsequent disinfection by 70% ethanol. Then, a full thickness circular wound (1.5 cm in diameter) was done on the middle of the right dorsal side where removal of the epidermal together with the dermal layers down to the subcutaneous connective tissue was done. This was performed under standard sterile conditions^[11]. Topical application of betadine to the wound site followed by rinsing with normal saline was done^[12]. After that, every rat was kept in a separate cage.

Experimental protocol and design

This experiment followed the guidelines of the Committee of the Animal Research Ethics (CARE) at Faculty of Medicine, Ain Shams University.

Animals were divided into three groups, ten rats in each

Group I (wounded untreated group)

Ten adult rats that were exposed to excisional skin wound injury. They were then left untreated for 2 weeks.

Group II (ADSCs treated group)

Ten adult rats were exposed to excisional wound injury, then intradermal injection of 1mL of final product containing 1×10^6 ADSCs (500 µL Cytocare 532 containing 0.5×10^6 cells) was done once into four injection sites around each wound^[13].

Group III (Coconut oil treated group)

Ten adult rats were exposed to excisional wound, then each wound was treated with single topical application of 0.5 gm coconut oil ointment, which was prepared by admix the coconut oil extract with Vaseline base with ratio 1:3 respectively^[10].

After 2 weeks, all rats were anesthetized using ether inhalation and then sacrificed. Skin samples, including the wound + 3 mm from the nearby normal skin, were harvested. Specimens were fixed in 10% neutral-buffered formalin, dehydrated, then embedded in paraffin blocks. Sections of 5μ m thickness were subsequently cut and stained with H&E^[14]. Masson's trichrome staining was also done to determine the degree of collagen fibers synthesis^[15].

Other specimens, 1mm3 in size, were fixed in 2.5% glutaraldehyde from which semi-thin sections (1 μ m thick) were prepared and stained with toluidine blue then examined by light microscope^[14].

Wound area measurement

Starting from day zero (day of wound induction) and every four days, measurement of the wound area was performed and calculation of the percentage of wound healing was done using the following equation:

Area of original wound – Area of actual wound/ Area of original wound x100%

Measuring of the progressive decrease in wound area was done by tracking the margin of the wound on a paper then placing this paper on a graph sheet paper of 1 mm² and counting the numbers of squares. The area of wound contraction was calculated by subtracting the area of total open wound from that of the initial tracing^[16,17].

Morphometric measurements

Ten non-overlapping fields from different sections of each group were used to measure the following parameters: Total epidermal thickness was measured in H&E stained sections at magnification of x400 and total area percentage of collagen fibers was measured in Masson's trichrome stained ones at magnification of x400. This was done using Leica LAS V3.8 image analyzer computer system (Switzerland).

Statistical analysis

The data was analyzed by Graph pad prism, software program, version 5.0(2007) (Inc., CA, and USA). Statistical difference among groups was determined using ANOVA followed by post hoc "Tukey Test" and "Bonferroni Test". The *P values* were obtained and interpreted where; *p*>0.05

was considered statistically insignificant, p < 0.05 was statistically significant and p < 0.001 was considered highly significant.

RESULTS

Macroscopic results (Figure 1, chart 1, graph 1)

The untreated wound area in rats of group I showed 0% reduction on 4th day, 15% on 8th day and 55% reduction on the 12th day). After local treatment by ADSCs, the wound area revealed 25% reduction on 4th day, 60% on 8th day and 92% on 12th day. The percent of reduction was significantly increased (P< 0.05) in comparison to the wounded untreated group at day 8. Regarding treatment by coconut oil, the wound area showed 15% reduction on 4th day, 36% on 8th day and 70% on 12th day.

Microscopic results

Wounded untreated group (group I)

Histological examination of H&E-stained sections after 2 weeks revealed re-epithelization by a relatively thin intact epidermis, with discontinuous keratin. The epidermal thickness showed highly significantly deccrease (P<0.001) as compared to normal epidermal thickness (Chart 2). Papillary dermis showed thick, disorganized collagen fibers, widely separated with empty areas (Figures 2,3). The reticular dermis appeared more condensed with collagen fibers. Disturbed hair follicles and few sebaceous glands were observed (Figure 3).

Masson's trichrome sections showed condensation of the collagen fibers underneath the epidermis at the wound site. Few thick bundles of collagen fibers were observed in the other areas of the papillary and reticular dermis (Figure 4). Statistically, the collagen area percentage was found to be highly significantly decreased (P<0.001) as compared to normal skin (Chart 3).

Semithin sections showed the apparenty thin epidermis. Most of the cells has pale ghost like nuclei. Some cells were closely packed while other dividing cells were separated from each other with large intercellular spacing (Figure 5).

ADSCs treated group (group II)

Histological examination of sections stained by H&E demonstated keratinised continuous thick epidermis with multiple cell layers at the wound area (Figure 6). The epidermal thickness was highly significantly increased (P<0.001) as compared to group I (Chart 2). Large amount of granulation tissue is formed in the papillary dermis

under the re-epithelised epidermis with parallel collagen fibers. Thich collagen bundles appeared underneath the granualtion tissue. The reticular dermis showed marked increase in collagen fibers as compared to group I. Multiple hair follicles & noticable sebaceous glands were seen (Figure 7). Sections stained by Masson's trichrome showed regular parallel collagen fibers underneath the re-epithelized epidermis (Figure 8), which was apprently increased as compared to group I. Dense irregularly arranged collagen fibers surrounding skin appendages (Figure 9). Statistically, the collagen area percentage was found to be highly significantly increased (P<0.001) as compared to group I (Chart 3).

Semi-thin sections showed multiple layers of epidermal cells with narrow intercellular spaces. Most of the cells had vesicular nuclei with prominent nucleoli. Mitotic figures were seen in some sections denoting cell division for regeneration (Figure 10).

Coconut oil treated group (group III)

Histological examination of sections stained by H&E showed the wound area with intact epidermis in most sections with disrupted keratin over the wound site. The epidermis appeared thick and hypertrophied in some areas especially at the wound margin, and thin in other areas, with a statistically significant increase (P < 0.05) in epidermal thickness was detected as compared to group I (Chart 2). Very few and shallow dermal papillae were encountered in the healed wound area in some sections (Figure 11), and were numerous and deep in other sections (Figure 12). Papillary dermis showed numerous thin collagen bundles with abnormal organization that extended also to the reticular dermis (Figures 11,12). The reticular dermis showed dense aggragated collagen replacing the hair follices in some sections (Figure 12). The reticular dermis appeared almost devoid of hair follicles and their sebaceous glands (Figures 11,12). Masson 's trichrome sections showed deposition of thin bundles of collagen fibers under the wound site parallel to the epidermis and were seen apprently few in the reticular dermis (Figure 13). In other sections, dense collagen fibers were seen in both papillary & reticular dermis. Collagen fibers appear parallel in some areas and interlacing in other areas (Figure 14). A staistically significant increase in collagen area percentage (P < 0.05) was detected compared to group I (Chart 3). Semithin sections showed few layers of epidermal cells with some intercellular spaces. The epidermis appeared thinner as compred to that of group II. Most of the cells had pale nuclei (Figure 15).



Fig. 1: A photographic follow up of the gross size of the skin wound of different groups every 4 days.



Fig. 2: A photomicrograph of a section of a rat 's skin of wounded untreated group (group I) showing relatively thin epidermis (E). Papillary dermis (PD) contains irregular widely separated collagen fibers (c) & empty areas ($\uparrow\uparrow$). (H&E X200).

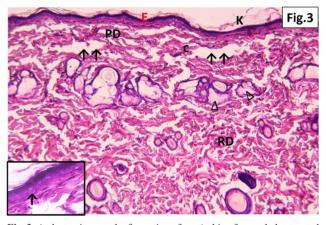


Fig. 3: A photomicrograph of a section of a rat's skin of wounded untreated group (group I) showing relatively thin epidermis (E) and discontinuous keratin (K). Papillary dermis (PD) contains irregular widely separated collagen fibers (c) & empty areas ($\uparrow\uparrow$). Reticular dermis (RD) is more condensed with collagen fibers. Few sebaceous glands can be noticed (Δ) Inset: Epidermis with few cell layers (\uparrow). (H&E X400, inset X1000).

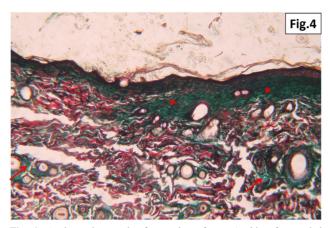


Fig. 4: A photomicrograph of a section of a rat 's skin of wounded untreated group (group I) showing few thick bundles of collagen fibers, most of them are parallel to epidermis (*). Notice few collagen fibers in the dermis (\uparrow). (Masson's trichrome X 400)

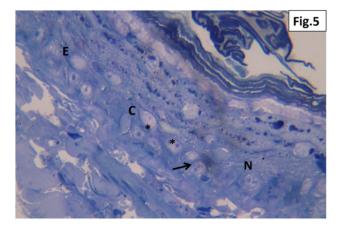


Fig. 5: A photomicrograph of a semi-thin section of a rat 's skin of wounded untreated group (group I) showing epidermis (E), with few cell layers (C). Most of the cells has pale ghost like nuclei (N). Note some cells are packed (\uparrow) while other dividing cells are separated from each other (*). (Toluidine blue x1000)

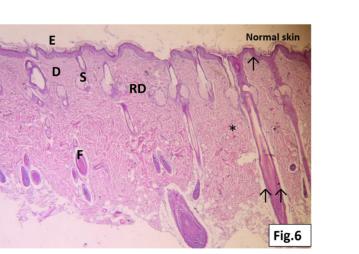


Fig. 6: A photomicrograph of a section of a rat's skin of ADSCs treated group (group II) showing wound area, with continuous multilayered epidermis (E). Reticular dermis (RD) containing hair follicles (F) & sebaceous glands (S). Notice the normal skin epidermis adjacent to the wound area (\uparrow), with collagen fibers (*) & numerous hair follicles ($\uparrow\uparrow$). (H&E x 200)

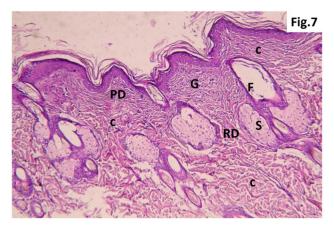


Fig. 7: A photomicrograph of a section of a rat's skin of ADSCs treated group (group II) showing large amount of granulation tissue (G) with thick collagen fibers underneath it (c) in the papillary dermis (PD). The reticular dermis (RD) appears with regenerated hair follicles (F) & numerous sebaceous glands (S). (H&E X400)

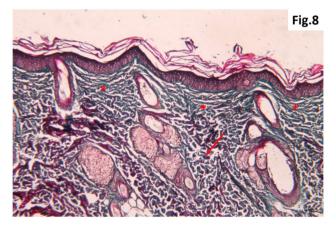


Fig. 8: A photomicrograph of a section of a rat's skin of ADSCs treated group (group II) showing many collagen fibers (\uparrow). Many fibers appear parallel to epidermis (*). (Masson's trichrome X 400)

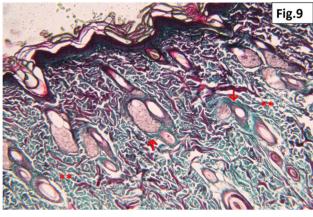


Fig. 9: A photomicrograph of a section of a rat's skin of ADSCs treated group (group II) showing dense irregularly arranged collagen fibers (**) surrounding the skin appendages ([†]). (Masson's trichrome X 400)

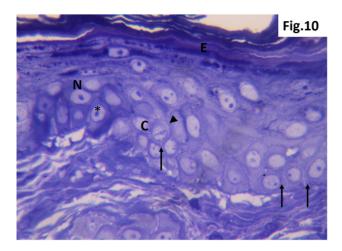


Fig. 10: A photomicrograph of a semi-thin section of a rat's skin of ADSCs treated group (group II) showing epidermis (E) having multiple cell layers (C) with narrow intercellular spaces (\uparrow). Most of the cells have vesicular nuclei (N) with prominent nucleoli (*). Notice the mitotic figure (\blacktriangle). (Toluidine blue x 1000).

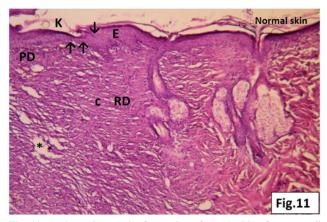


Fig. 11: A photomicrograph of a section of a rat's skin of coconut oil treated group (group III) showing discontinuous keratin (K) on the surface of the epidermis. The epidermis appears hypertrophied (\uparrow) at the wound edge, and very thin ($\uparrow\uparrow$) in other areas. Condensed collagen fibers (c) are observed in the papillary (PD) & reticular dermis (RD) with empty spaces (*) in reticular dermis. No skin appendages can be observed. Note area of normal skin (Normal skin). (H&E X 400)

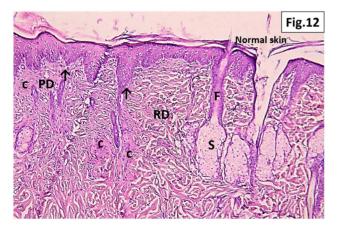


Fig. 12: A photomicrograph of a section of a rat's skin of coconut oil treated group (group III) showing the epidermis with increase in dermal papillae (\uparrow). Condensed collagen fibers (c) are seen in both papillary (PD) & reticular dermis (RD) with no skin appendages. Note area of normal skin (Normal skin) with well developed hair follicle (F) & sebaceous gland (S). (H&E X 400)

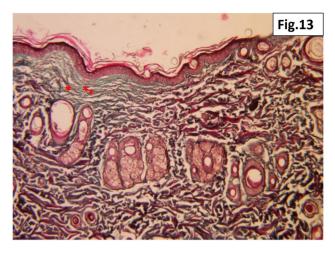


Fig. 13: A photomicrograph of a section of a rat's skin of coconut oil treated group (group III) showing deposition of thin collagen fibers, mostly parallel to the epidermis (*). (Masson's trichrome X 400)

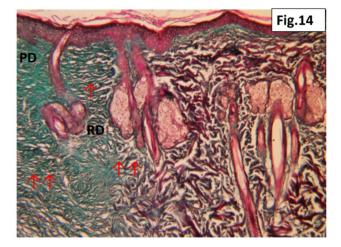


Fig. 14: A photomicrograph of a section of a rat's skin of coconut oil treated group (group III) showing areas of dense collagen fibers in both papillary (PD) & reticular dermis (RD). Collagen fibers appear parallel (\uparrow) in some areas and interlacing in other areas ($\uparrow\uparrow$).(Masson's trichrome X 400)

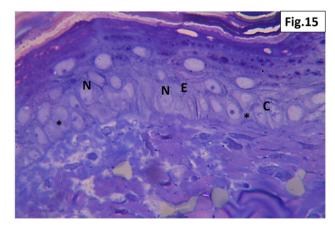
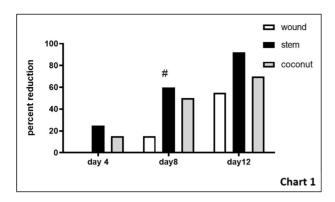
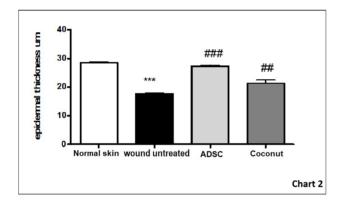


Fig. 15: A photomicrograph of a semi-thin section of a rat's skin of coconut oil treated group (group III) showing epidermis (E) having few cell layers (C) with some intercellular spaces (*). Most of the cells have pale nuclei (N). (Toluidine blue X 1000)



Column chart 1: Data are mean \pm SD (n=10). # (P<0.05), in comparison to wounded untreated group, by two-way ANOVA with Bonferroni posthoc test.



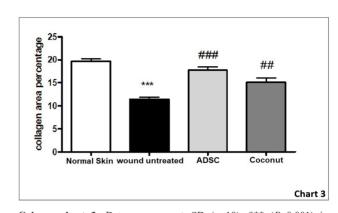
Column chart 2: Data are mean \pm SD (n=10). *** (P<0.001) in comparison to normal skin ## (P<0.05), ### (P<0.001), in comparison to wounded untreated group, by one-way ANOVA with Tukey's post-hoc test.

DISCUSSION

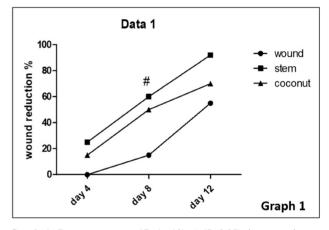
Despite the availability of many therapies, skin wound is still considered one of the persistent health care problems. Sometimes they heal slowly as in deep or chronic wounds. Thus, the current study aimed to compare the effect of ADSCs as a promising method of regenerative medicine and that of the coconut oil as a natural product in the process of wound healing.

In the present work, the ADSCs treated group macroscopically and statistically showed a great increase in the rate of healing of the wound area. The skin almost regained its normal appearance after two weeks. In accordance with this finding, it was previously declared that ADSCs have been used with success in cases of soft tissue regeneration and reconstruction after mastectomy and soft tissue repair^[5].

In the present study, histological examination of sections of the ADSCs treated group stained by H&E demonstrated almost normal epidermis with multiple cellular layers was observed. Semi-thin sections showed multiple layers of epidermal cells with narrow intercellular spaces. Most of the cells had vesicular nuclei. This was in accordance



Column chart 3: Data are mean \pm SD (n=10). *** (P<0.001) in comparison to normal skin ## (P<0.05), ### (P<0.001), in comparison to wounded untreated group, by one-way ANOVA with Tukey's posthoc test.



Graph 1: Data are mean \pm SD (n=10). # (P<0.05), in comparison to wounded untreated group, by two-way ANOVA with Bonferroni post-hoc test. Stem cell group showed highest wound reduction percent.

with a previous research which reported thick regenerated epithelium after three weeks of stem cell treatment^[18].

In the present study granulation tissue was observed under the re-epithelized epidermis. In addition, multiple well developed hair follicles & sebaceous glands in the dermis were noticed. Going with these observations, healthy granulation tissue deposition, complete healing and wound covering with regenerated normal hairy skin were previously reported after three weeks of stem cells treatment^[19,20]. On the other hand, failure of skin appendages to regenerate completely was also reported previously^[18].

Zhao *et al.*, 2015 explained that ADSCs enhancement of wound healing was via macrophage stimulating protein release^[9]. Moreover, studies found that ADSCs secreted high quantities of angiogenic growth factors including; vascular endothelial growth factor and Insulin growth factor-1 when injected intradermally^[21]. Additionally, some studies declared that ADSCs can be differentiated into endothelial & epithelial cells in injured tissues^[22].

Other studies declared that ADSCs can induce differentiation of fibroblasts in injured tissues, which help in wound healing, on the other hand it prevent excessive proliferation and migration of fibroblast, which are responsible for hypertrophic scars^[23]. ADSCs are nowadays considered to be a new line for regenerative hair therapy^[24].

In the coconut oil treated group of the present work, moderate increase in the healing rate was macroscopically and statistically documented. In agreement, coconut oil was confirmed previously to fasten wound healing^[25].

Histological examination of sections of coconut oil treated group stained by H&E revealed thin irregular continuous epidermis, with hypertrophy at the junction with normal skin. Dermis showed widely spaced collagen fibers with almost no skin appendages. Previous investigations showed that VCO promoted healing of diabetic wounds and it showed better results compared to topically used silver sulfadiazine^[25]. In the present study munerous deep dermal papillae were encountered in some sections. Going with this finding, a study declared that dermal papillae are essential for proper skin repair and for generation of hair bud like structures^[26]. Another study declared that topical vitamin C application in aged skin leads to an increase in the dermal papillae density through angiogenesis. The authors added that these papillae contain nutritive capillaries for the skin & they decrease with age^[27].

In the current study, continuous keratin was observed in the epithelium of sections of ADSCs treated group & discontinuous keratin was observed in sections of both; wound group & coconut oil treated group. Keratinocytes are considered as the major cellular component of the outermost layer of skin^[28]. It was previously observed that in wound healing, keratinocytes at the edges of the wound proliferate and migrate to cover the surface of the epidermal defects^[29].

In the present study, sections of ADSCs treated group stained by Masson's trichrome revealed increase in collagen fibers in a meshwork pattern, and some collagen bundles appeared parallel to the epidermis. In accordance, a previous study observed a dense network of multidirectional thick collagen fibers 3 weeks following stem cell treatment^[18]. Another study also revealed the presence of multidirectional collagen bundles in fully regenerated skin^[30]. Collagen is an important extracellular matrix protein; it is responsible for the tissue matrix integrity and also is involved in tissue homeostasis and epithelization in wound healing process^[28]. It is synthesized by fibroblasts, they give strength to all tissues and play the main role, especially in the proliferative and remodelling phases of regeneration^[30]. Other study documented parallel collagen bundles in regenerating skin^[31]. Moreover, ADSCs was proved to promote the proliferation of human dermal fibroblast in the reepithelialization stage of wound healing, by either directly contacting cells or by paracrine activation^[32]. Hence, fibroblasts' activation might explain the increased collagen fibers deposition in group II of the present study.

On the other hand, sections of coconut oil treated group stained by Masson's trichrome in the present study showed collagen bundles parallel to epidermis in some sections, or condensed in both papillary & reticular dermis in others. It has been reported that VCO-treated wounds revealed increased neovascularization and fibroblast proliferation that was further confirmed by a notable increase in Pepsinsoluble collagen thus indicating an increase in collagen cross-linking^[33]. Furthermore, other authors added that coconut oil effect on wound contraction and healing could be attributed to its antioxidant and antibacterial activity^[34].

CONCLUSION

After 2 weeks of excisional wound injury, Virgin Coconut Oil enhanced re-epithelization and collagen deposition, with almost no observed skin appendages in the wound area. On the other hand, Adipose derived mesenchymal stem cells were more efficient as regards re-epithelization and regaining of skin to its mature structure and appendages more rapidly. However, virgin coconut oil can be used in places where adipose stem cells are not available, yet it may require more time for healing.

DATA AVAILABILITY STATEMENT

The data supporting the findings of this study are available from the corresponding author upon reasonable request.

CONFLICT OF INTERESTS

There are no conflicts of interest.

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الملخص العربى

دراسه نسيجيه مقارنه لتاثير الخلايا الجذعية المشتقة من النسيج الدهني مقابل زيت جوز الهند البكر في التئام الجروح الجلدية للذكور البالغين من الجرذان البيضاء

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الخلفية: الجروح تمثل عبء سريري على العديد من المرضى. توفر الخلايا الجذعية المستخلصة من النسيج الدهني (ADSCs) مستقبلًا واعدًا في مجال الطب التجديدي للأنسجة نظرًا لتوافر ها الواسع وقدرتها على التمايز. كما تناولت بعض الدراسات تأثير زيت جوز الهند البكر (VCO) في التئام الجروح عن طريق تقليل وقت إعادة بناء النسيج الظاهري. ولكن هناك نقص في المراجع بخصوص التفاصيل التركيبية النسيجية.

الهدف: مقارنة تأثير الخلايا الجذعية المستخلصة من النسيج الدهني وزيت جوز الهند البكر في التئام الجروح. **المواد والطرق المستخدمه**: تم استخدام ثلاثين جرذا في ثلاث مجموعات تجريبية، عشرة جرذان لكل منها. وقد تم تعرضهم جميعهم لجرح استئصالي في الجلد. المجموعة الأولى (مجموعة الجرح الضابطة الغير معالجة). في المجموعة الثانية (مجموعة المعالجة بالخلايا الجذعية) تعرضوا لجرح استئصالي في الجلد، ثم تم حقنهم داخل الجلد ١ معالجة الثانية (مجموعة المعالجة بالخلايا الجذعية) تعرضوا لجرح استئصالي في الجلد، ثم تم حقنهم داخل الجلد ١ معالجة الثانية (مجموعة المعالجة بالخلايا الجذعية) تعرضوا لجرح استئصالي في الجلد، ثم تم حقنهم داخل الجلد ١ معالجة مناها الثانية (مجموعة المعالجة بالخلايا الجذعية) تعرضوا لجرح مواقع حقن ، في حين أن المجموعة الثالثة (المجموعة المعالجة بزيت جوز الهند) تعرضوا لجرح استئصالي في الجلد ثم تم علاجهم بواسطة مر هم زيت جوز الهند ٥، جم . وقد تم التضحية بجميع الجرذان بعد أسبوعين. تم تجمع عينات الجلد، وإعدادها لفحص الأنسجة بالميكر وسكوب الضوئي. تم تسجيل انكماش الجروح ومقارنتها. تم إجراء القياسات المور فومترية لكل من سماكة البشرة ونسبة الكولاجين، وتم تحليل البيانات إحصائياً.

النتائج: أظهر الفحص النسيجي لمجموعة الخلايا الجذعية بشرة متجددة مع زوائد جلدية في dermis. أظهرت الشرائح المصبوغة بصبغة ماسون ثلاثية الألوان حزم الكولاجين السميكة في اتجاهات مختلفة. في المجموعة المعالجة بزيت جوز الهند، كانت البشرة رقيقة دون ظهور الزوائد الجلدية في الأدمة. أظهرت الشرائح المصبوغة بصبغة ماسون ثلاثية الألوان حزم الكولاجين المتوازية الكثيفة.

الخلاصة: زيت جوز الهند البكر يعزز شفاء الجروح الجلدية، وعلى الرغم من ذلك، أثبتت الخلايا الجذعية المستخلصة من النسيج الدهني أنها أكثر كفاءة في استعادة البنية الناضجة والتئام الجلد بشكل أسرع.