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Original Article

Pro-inflammatory 'M1 macrophage' vs anti-inflammatory 'Hydrocortisone' a new approach to wound healing in HaCaT cells

Selin Engur Ozturk[1]

[1]Pamukkale University, Department of Pharmacy Services, Denizli, Turkiye

ABSTRACT

Background and Aims: Wound healing is a process of repairing the skin that has lost its integrity through inflammation, proliferation, and remodeling. Macrophages exhibit adaptability, transitioning from a pro-inflammatory "M1" to an anti-inflammatory "M2" phenotype throughout wound healing for optimal outcomes. Hydrocortisone's M2c polarization makes it a key agent for balancing M1/M2 polarization. In this study, we specifically explored the effects of M1 macrophages and hydrocortisone on cell migration and wound healing in HaCaT keratinocytes.

Methods: To better understand how macrophages contribute to wound healing, we created a co-culture scratch assay model of HaCaT cells using M1-polarized macrophages derived from THP-1 cells. In addition, we administered hydrocortisone, 'an anti-inflammatory drug', to our experimental groups to compare the effects. We determined the proliferation effects of different concentrations of hydrocortisone and PMA on HaCaT cells. Then, we evaluated the effects of polarized M1 macrophages and hydrocortisone on the wound healing of HaCaT cells by scratch assay and COL1A1 mRNA gene expression levels.

Results: As a result, it was determined that 100 µM hydrocortisone increased HaCaT cell migration and *COL1A1* mRNA gene expression compared to control, while M1 polarized macrophages decreased these effects negatively.

Conclusion: To understand the macrophages responsible for the mechanisms of wound healing, much more study is required. Macrophages are a vital component in the healing process for wounds, and the shifting of M1/M2 in the treatment of wounds can potentially lead to the enlargement of novel treatment methods.

Keywords: HaCaT, hydrocortisone, macrophage, wound healing

INTRODUCTION

The process of wound healing holds significant importance in enhancing the quality of life of patients as it facilitates the restoration of skin integrity, the preservation of underlying tissues, and safeguarding against potential risks of infection and dehydration (Rahmannia, Amini, Chien, & Bayat, 2022). Collagen type I, which is responsible for fiber formation, is predominantly present in the skin and is encoded by the COL1A1 gene. It constitutes approximately 90% of the total collagen in the skin (Öztürk, Çevikelli, Tilki, Güven, & Kıyan, 2023).

The pivotal role of macrophages in the regulation of wound healing is widely acknowledged. The cells demonstrate notable adaptability and an altering phenotype, shifting from a pro-inflammatory or "M1" phenotype to an anti-inflammatory "M2" phenotype during the distinct phases of the wound healing process to facilitate ideal healing outcomes (Sharifiaghdam et al., 2022). M1 macrophages, along with other cells in the microenvironment, play a role in wound healing. This has a negative impact on wound closure, migration, and collagen expression, particularly in diabetic wounds (Miao et al., 2012). Limiting the M1 macrophage to the processes of bacteriophage, necrotic cell removal, and proinflammatory cytokine secretion in the wound microenvironment might be helpful in the wound healing process (Basu Mallik, Jayashree, & Shenoy, 2018). Establishing the M1/M2 polarization balance in the wound microenvironment will contribute to wound healing studies because the effects of M2 macrophages on preventing tissue damage are known (Louiselle, Niemiec, Zgheib, & Liechty, 2021).

Studies on macrophages are divided into different functional categories: proinflammatory and anti-inflammatory responses; M1 and M2 macrophages, respectively (Bashir, Sharma, Elahi, & Khan, 2016). M1 macrophages can become polarized in the combination of lipopolysaccharide (LPS) and interferon-gamma (IFN-γ) (Engür-Öztürk & Dikmen, 2022; Orecchioni,

Corresponding Author: Selin Engür Öztürk **E-mail:** selino@pau.edu.tr
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Ghosheh, Pramod, & Ley, 2019; Chanput, Mes, Savelkoul, & Wichers, 2013). Although M1 macrophages have potent antimicrobial activity, they can also exert the effect of mediating ROS-induced tissue damage and inhibiting tissue regeneration and wound healing (Huang, Li, Fu, & Xin, 2018). The anti-inflammatory activity of M2 macrophages inhibits the chronic inflammatory response to prevent this tissue damage (Shapouri-Moghaddam, 2018). M2 macrophages, particularly M2c macrophages, are polarized by glucocorticoids, which are anti-inflammatory agents (Engür-Öztürk & Dikmen, 2022; Foey, 2014; Tu et al., 2017; Huang et al., 2018).

Wound healing is a process of tissue regeneration that includes inflammation as an initial step. If the physiological inflammatory response during wound healing is prolonged or intensified, it results in a delay in the subsequent stages of appropriate wound healing (Öztürk et al., 2023). For these reasons as well as to promote M2c macrophage polarization, anti-inflammatory agents are required.

In this scope of study, to better understand how macrophages contribute to wound healing, we created a co-culture assay model of HaCaT cells using pro-inflammatory M1-polarized macrophages derived from THP-1 cells. In addition, we administered hydrocortisone, 'an anti-inflammatory drug' provides M2c polarization. We determined the proliferation effects of different concentrations of hydrocortisone and PMA on HaCaT cells. Then, we evaluated the effects of polarized M1 macrophages and hydrocortisone on the wound healing of HaCaT cells by scratch assay and *COL1A1* mRNA gene expression levels.

MATERIALS AND METHODS

Cell culture and treatment

Human skin keratinocytes HaCaT cells (CLS No: 300493, Germany) and human monocyte THP-1 cells (ATCC®TIB-202™, USA) were grown in RPMI-1640 medium supplemented with 10 % fetal bovine serum (FBS) and 1% penicillin/streptomycin at 37ºC in a humidified incubator with a 5% CO2 atmosphere.Hydrocortisone and phorbol-12-myristate-13-acetate (PMA) (Sigma Aldrich, USA) were dissolved in dimethyl sulfoxide (DMSO) as a stock solution.

Cell proliferation assay

HaCaT keratinocyte cell viability and proliferation were evaluated by the MTT (3,4,5-dimethylthiazol-2-yl)-2-diphenyltetrazolium bromide) method. HaCaT cells were inoculated into 96-well plates at densities of 5 x 10^3 cells/well. After 24h, they were treated with various concentrations of PMA and hydrocortisone for 48h. After incubation, MTT solution was added to reach a final concentration of 0.5 mg/mL and incubated for 3 hours in the incubator. Then, crystals of MTT-formazan were dissolved by adding 100 µL of DMSO to each well. At 540 nm, absorbances were measured using a Cytation 3 cell imaging multi-mode reader (Bio-Tek).

Polarization of M1 subtype macrophages

In our previous study, differentiation of THP-1 monocyte cells to M0 macrophages and then polarization into M1 macrophages were described (Engür-Öztürk & Dikmen, 2022). Briefly, PMA was used to induce macrophage-like (M0) differentiation in THP-1 cells for 24 hours. M0 macrophages were polarized into M1 macrophages after 24 hours of exposure to 20 ng/mL LPS + IFN-γ.

Wound healing with scratch assay in a co-culture model

A co-culture model was established to investigate the effects of M1-polarized macrophages on HaCaT cell proliferation and wound healing. Using transwell inserts, M1-polarized macrophages were co-cultured with HaCaT cells. The HaCaT cells and M1 macrophages were co-cultured using six-well plate cell culture inserts with a 0.4-µm porous membrane dividing the upper and lower chambers. Briefly, THP-1 monocytes were seeded in the transwell apparatus' upper chamber and stimulated to differentiate into M1 polarized macrophages with PMA, IFN-*gamma* and LPS. HaCaT cells were seeded 1×10^6 cells per well 24 h before M1 macrophage polarization ended. Before the lower and upper chambers were assembled, a scratch assay was performed to determine how the cytokines released by M1 macrophages affected the ability of HaCaT cells to proliferate and migrate (Engür-Öztürk & Dikmen, 2022). After removing the medium, a 100 µL sterile plastic pipette tip was used to create a linear wound in the monolayer. The upper chambers, which contained M1 macrophages, were then positioned directly on cover of the HaCaT cells in the plates. For 48 h, HaCaT and M1 macrophage cells were incubated together. In addition, HaCaT cells were incubated for 48 h at 100 µM hydrocortisone to determine its effect on HaCaT cell proliferation during the wound healing experiment. At the end of the incubation periods, the wound was visualized with a Leica DM 300 light microscope for analysis of diameter change(Yuksel, Dikmen, & Canturk, 2021).

RT-PCR analysis

RNA was isolated from HaCaT cells treated with hydrocortisone or co-cultured with M1 polarized macrophages. Total RNA isolation was performed on the MagNA Pure LC 2.0 system (Roche, Germany), 500 ng total RNA was used for cDNA synthesis from each RNA population and cDNA synthesis was performed with Transcriptor High Fidelity cDNA Synthesis Kit (Catalog no: 05091284001, Roche, Germany) according to the manufacturer's instructions. The total mRNA amounts of the

samples were measured at 260 and 280 nm in the NanoDrop 2000® (Thermo Fisher, USA) spectrophotometer.

Quantitative real-time polymerase chain reaction (qRT-PCR) was used to assess the mRNA levels of collagen type I alpha 1 chain (*COL1A1*) gene expression in relation to wound healing. As an internal positive control, glyceraldehyde-3-phosphate dehydrogenase (GAPDH) mRNA levels were used. Probe–primer pairs for target genes were supplied from Roche Diagnostic as real-time ready catalog assays. The primer sequences were *COL1A1* forward: 5-GCA AGA CAG TGA TTG AAT ACA AAA CCA-3, reverse: 5- ATC AAA GGA GCG GAT CGA GTG GTC-3 and GAPDH forward: 5-CTCTGCTCCTC CTGTTCGAC-3, reverse: 5- ACGACCAAATCCGTTGACTC-3. The real time PCR mix kit (LightCycler® 480 Probes Master, Catalog no: 04707494001, Roche, Germany), containing 10 µL 2x LightCycler® 480 Probes Master, 1 µL of each primer (Real Time Ready Assay, Roche), 4 µL PCR grade water, and 5 µL of cDNA were prepared. The cycling conditions included an initial incubation step at 95°C for 10 min, followed by 45 cycles of amplification with 10 s at 95°C, 30 s at 60°C and 1s at 72°C. The final cooling step was holding at 40°C for 30s. Results were analyzed by advanced relative quantification with the LightCycler® 480 System's software (version 1.5.0.39) (Öztürk et al., 2023).

Statistical analysis

GraphPad Prism 8.0 software was used for one-way ANOVA and Tukey's post hoc test. P values represent the significance of the results compared to the control group (P<0.0001****, P<0.001***, P<0.01**, P<0.05* and P>0.05 n.s) (± standard deviation).

RESULTS

Proliferative effects of hydrocortisone were assessed using the MTT assay

HaCaT cells were exposed to various concentrations of PMA and hydrocortisone for 48h, and the proliferative effect of the cells was determined by the MTT method. As a result, in comparison to the control group, PMA concentrations had no effect on cell proliferation (**Figure 1**). However, 100 µM hydrocortisone showed significant proliferative effects on the cells (****p<0.0001) (**Figure 2**).

M1 subtype polarization of THP-1-derived macrophages

In this study, THP-1 cells were exposed to PMA concentrations for 24h in order to differentiate them into M0 macrophages. These M0 cells were then polarized to M1 macrophages by incubating them for 24 h with 20 ng/mL LPS+IFN-γ. As a result,

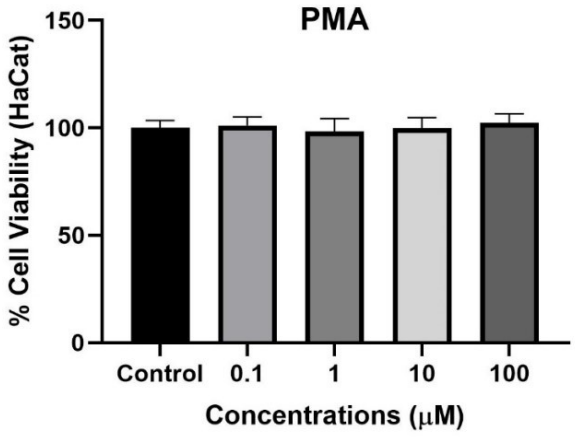

Figure 1. The HaCaT cells were treated with different concentrations of PMA for 48 h and percentage cell viability was determined from MTT results.

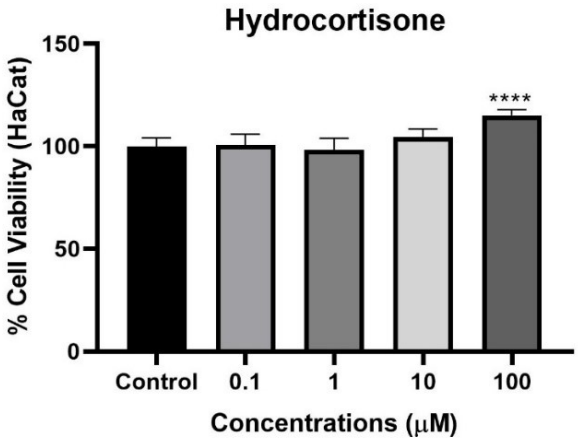

Figure 2. The HaCaT cells were treated with different concentrations of hydrocortisone for 48 h and percentage cell viability was determined from MTT results.

THP-1 cells, M0 macrophages, and M1 polarized macrophage cells were photographed with a microscope (Leica DM 300) (**Figure 3**).

Assessment of wound healing

Using a wound healing assay that measures cell population growth, the spread and migratory capacities of HaCaT cells were examined. The wound diameter change data, expressed in graphics, was obtained with the measurement program (The LAS Image Analysis Application) of the microscope (Leica DM) on the photographs, an example of which is shown in **Figure 4**. Each of the 48-h control and hydrocortisone groups had smaller wounds compared to the 0h control group. In addition, when a comparison was made with the control group at 48h, no significant difference was found between the hydrocortisone

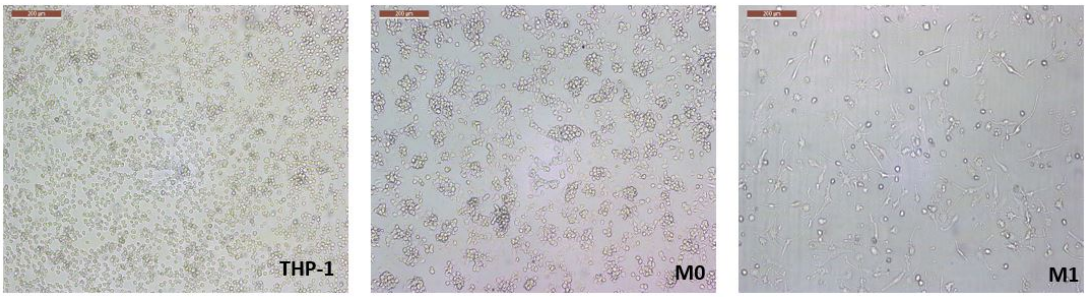

Figure 3. Microscope image of THP-1 cells (A), THP-1 derived macrophages (M0) and M1 polarized macrophage 10X objective.

and the control. As a result, the presence of M1 macrophages negatively affected the migration of HaCaT cells and therefore wound closure, while 100 µM hydrocortisone was effective in cell proliferation but did not show a significant effect in terms of wound closure (Figure 5).

Evaluation of *COL1A1* mRNA expression levels by RT-PCR analysis

HaCaT cells were treated with hydrocortisone (100 µM) and co-cultured with M1 macrophages, and mRNA expression levels of the *COL1A1* gene were determined using RT-PCR. Expression levels of the *COL1A1* gene were decreased in response to M1 macrophage co-culture ($P<0.05$*). Contrarily, in HaCaT cells treated with 100 µM hydrocortisone, the expression levels of the *COL1A1* gene increased approximately 1.5-fold ($P<0.01$**) (**Figure 6**).

DISCUSSION

The complex multicellular process of wound healing involves keratinocytes, fibroblasts, endothelial cells, and inflammatory cells (Leibovich & Ross, 1975; Loots et al., 1998; Huang et al., 2019). For a very long time, fibroblasts and keratinocytes have been the focus of studies on the repair of skin wounds. Therefore, current research is aimed at evaluating intracellular wound healing functions, and the focus of this evaluation is on these cell types (Calabrese, Dhawan, Kapoor, Agathokleous, & Calabrese, 2022). Therefore, the HaCaT keratinocyte cells used in our study were a viable alternative for a modeling of the process of healing wounds. During wound healing, macrophages play important roles, and a delayed healing period is associated with an ongoing inflammatory response (Huang et al., 2019). Recent research indicates that wound-healing macrophages exist in a variety of phenotypic states and may have a significant influence on the healing of wounds (Koh & DiPietro, 2011). The classically M1 and alternatively M2 polarized macrophages are defined (Engür-Öztürk & Dikmen, 2022). M1 macrophages are pro-inflammatory and eliminate damaged tissue, and they are crucial in the eradication of necrotic cells from the damage and other debris during the initial periods of the inflammatory phase (Calabrese et al., 2022;Delavary, van der Veer, van Egmond, Niessen, & Beelen, 2011) Similar to the findings that we obtained, Huang et al. demonstrated that the ability of cultured keratinocytes to migrate was inhibited when M1 macrophages were present in the environment (Huang et al., 2019).

In all wounds, macrophages are crucial, from contributing to inflammation to killing pathogens to resolving inflammation and initiating tissue remodelling and regeneration M2 macrophages are important players in tissue repair (Kim & Nair, 2019). M2 macrophages, particularly M2c macrophages, are polarized by the presence of glucocorticoids (Engür-Öztürk & Dikmen, 2022). It is widely known for its ability to reduce inflammation; a glucocorticoid (e.g., hydrocortisone) controls the increase of keratinocyte cells and regulates the dermal process (Terao & Katayama, 2016). Also, topical applications with anti-inflammatory drugs (such as corticosteroids) are one of numerous treatments used to promote gingival wound healing (Kongkadee, Wisuitiprot, Ingkaninan, & Waranuch, 2022).

Collagen I is one of the dermal extracellular matrix proteins in the skin (Krieg & Aumailley, 2011). Specifically, collagen type I, which is the product of the *COL1A1* gene, represents the predominant form of collagen within the skin, comprising approximately 90% of the total collagen content (Gelse, Pöschl, Aigner, 2003). Therefore, the effects on *COL1A1* gene expression levels related to wound healing have been tested in our study. The increased *COL1A1* mRNA expression levels detected in HaCAT cells treated to hydrocortisone can be attributed to pharmacologically induced anti-inflammatory properties, which is consistent with previous research findings (Wu et al., 2017).

Our study findings indicate that the utilization of hydrocortisone leads to an elevation in keratinocyte cell proliferation, facilitates cell migration, and enhances collagen expression levels in the environment of wound healing. Further research may establish hydrocortisone as a valuable therapeutic agent for M1/M2c conversion in the scope of wound healing treatment.

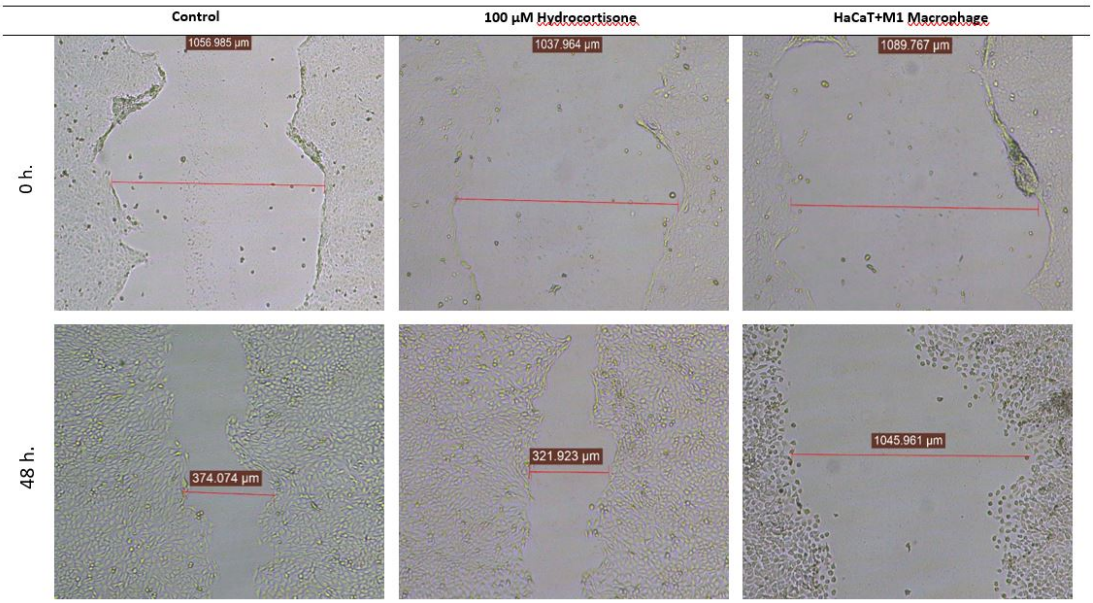

Figure 4. Scratch assay, photographs of HaCaT cells at 0 and 48. hours and wound diameter (A representative result for each group from two independent replicates is shown, 10X objective).

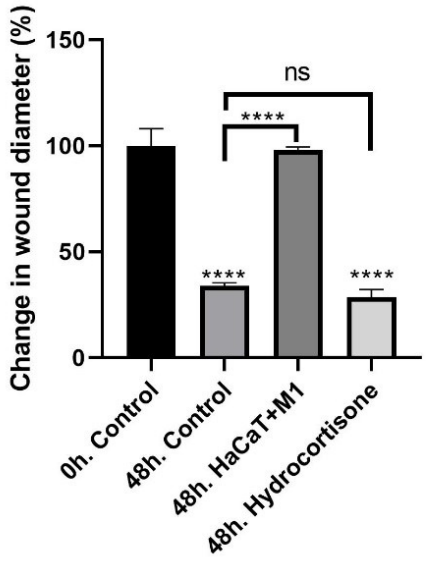

Figure 5. Effects of wound healing effects of M1 macrophage co-culture and 100 µM hydrocortisone on HaCaT cells on wound diameter change at 0 and 48 hours (±Sd., n=3, ns: not significant, p<0.0001**** compared to the 0 h. control group).

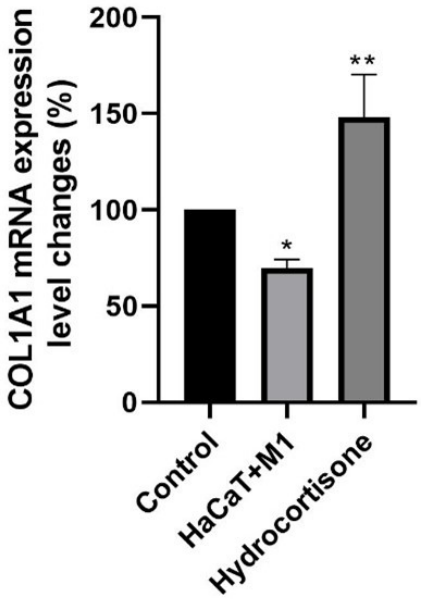

Figure 6. Changes in the mRNA expression levels (%) of wound healing related *COL1A1* gene. The error bars represent the standard deviations (n = 4, P<0.05*, P<0.01**).

CONCLUSION

Understanding the role of macrophages in the mechanisms of wound healing will require extensive research. Macrophages are a critical component of this dysregulation, and studies have revealed that macrophage polarization and timing are becoming increasingly important for both knowledge of disease progression and potential novel treatment methods.

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ORCID IDs of the authors

Selin Engur Ozturk 0000-0003-1534-8117

REFERENCES

Bashir, S., Sharma, Y., Elahi, A., & Khan, F. (2016). Macrophage polarization: the link between inflammation and related diseases. *Inflammation Research*, 65(1), 1–11. https://doi.org/10.1007/s00011-015-0874-1

Basu Mallik, S., Jayashree, B. S., & Shenoy, R. R. (2018). Epigenetic modulation of macrophage polarization- perspectives in diabetic wounds. *Journal of Diabetes and Itscomplications*, 32(5), 524–530. https://doi.org/10.1016/j.jdiacomp.2018.01.015

Calabrese, E. J., Dhawan, G., Kapoor, R., Agathokleous, E., & Calabrese, V. (2022). Hormesis: Wound healing and keratinocytes. *Pharmacological Research*, 183, 106393. https://doi.org/10.1016/j.phrs.2022.106393

Chanput, W., Mes, J. J., Savelkoul, H. F., & Wichers, H. J. (2013). Characterization of polarized THP-1 macrophages and polarizing ability of LPS and food compounds. *Food & Function,* 4(2), 266–276. https://doi.org/10.1039/c2fo30156c

Delavary, B. M., van der Veer, W. M., van Egmond, M., Niessen, F. B., & Beelen, R. H. (2011). Macrophages in skin injury and repair. *Immunobiology*, 216(7), 753–762. https://doi.org/10.1016/j.imbio.2011.01.001

Engür-Öztürk, S., & Dikmen, M. (2022). Proteasome inhibitor immunotherapy for the epithelial to mesenchymal transition: assessing the A549 lung cancer cell microenvironment and the role of M1, M2a and M2c 'hydrocortisone-polarised' macrophages. *Molecular Biology Reports*, 49(6), 4777–4793. https://doi.org/10.1007/s11033-022-07329-w

Foey, A. D. (2014). Immune Response Activation. Guy Huynh Thien Duc (Eds.) Macrophages—masters of immune activation, suppression and deviation (pp. 121-149). BoD – Books on Demand Press.

Gelse, K., Pöschl, E., & Aigner, T. (2003). Collagens–structure, function, and biosynthesis. *Advanced Drug Delivery Reviews, 55*(12), 1531–1546. https://doi.org/10.1016/j.addr.2003.08.002

Huang, S. M., Wu, C. S., Chiu, M. H., Wu, C. H., Chang, Y. T., Chen, G. S., & Lan, C. E. (2019). High glucose environment induces M1 macrophage polarization that impairs keratinocyte migration via TNF-α: An important mechanism to delay the diabetic wound healing. *Journal of Dermatological Science,* 96(3), 159–167. https://doi.org/10.1016/j.jdermsci.2019.11.004

Huang, X., Li, Y., Fu, M., & Xin, H. B. (2018). Polarizing Macrophages In Vitro. *Methods in Molecular Biology*, 1784, 119–126. https://doi.org/10.1007/978-1-4939-7837-3_12

Kim, S. Y., & Nair, M. G. (2019). Macrophages in wound healing: activation and plasticity. *Immunology and Cell Biology, 97*(3), 258–267. https://doi.org/10.1111/imcb.12236

Koh, T. J., & DiPietro, L. A. (2011). Inflammation and wound healing: the role of the macrophage. *Expert reviews in Molecular Medicine,* 13, e23. https://doi.org/10.1017/S1462399411001943

Kongkadee, K., Wisuitiprot, W., Ingkaninan, K., & Waranuch, N. (2022). Anti-inflammation and gingival wound healing activities of Cannabis sativa L. subsp. sativa (hemp) extract and cannabidiol: An in vitro study. *Archives of Oral Biology*, 140, 105464. https://doi.org/10.1016/j.archoralbio.2022.105464

Krieg, T., & Aumailley, M. (2011). The extracellular matrix of the dermis: flexible structures with dynamic functions. *Experimental Dermatology*, 20(8), 689–695. https://doi.org/10.1111/j.1600-0625.2011.01313.x

Leibovich, S. J., & Ross, R. (1975). The role of the macrophage in wound repair. A study with hydrocortisone and antimacrophage serum. *The American Journal of Pathology*, 78(1), 71.

Loots, M. A., Lamme, E. N., Zeegelaar, J., Mekkes, J. R., Bos, J. D., & Middelkoop, E. (1998). Differences in cellular infiltrate and extracellular matrix of chronic diabetic and venous ulcers versus acute wounds. *Journal of Investigative Dermatology*, 111(5), 850-857.

Louiselle, A. E., Niemiec, S. M., Zgheib, C., & Liechty, K. W. (2021). Macrophage polarization and diabetic wound healing. *Translational Research: The Journal of Laboratory and Clinical Medicine*, 236, 109–116. https://doi.org/10.1016/j.trsl.2021.05.006

Miao, M., Niu, Y., Xie, T., Yuan, B., Qing, C., & Lu, S. (2012). Diabetes-impaired wound healing and altered macrophage activation: a possible pathophysiologic correlation. *Wound repair and regeneration: official publication of the Wound Healing Society [and] the European Tissue Repair Society,* 20(2), 203–213. https://doi.org/10.1111/j.1524-475X.2012.00772.x

Orecchioni, M., Ghosheh, Y., Pramod, A. B., & Ley, K. (2019). Macrophage Polarization: Different Gene Signatures in M1(LPS+) vs. Classically and M2(LPS-) vs. Alternatively Activated Macrophages. *Frontiers in Immunology*, 10, 1084. https://doi.org/10.3389/fimmu.2019.01084

Öztürk, A. A., Çevikelli, T., Tilki, E. K., Güven, U. M., & Kıyan, H. T. (2023). Ketorolac Tromethamine Loaded Nano-Spray Dried Nanoparticles: Preparation, Characterization, Cell Viability, COL1A1 Gene Simulation and Determination of Anti-inflammatory Activity by In vivo HET-CAM Assay. *Current Drug Delivery*, 20(6), 830–840. https://doi.org/10.2174/1567201820666230125144133

Rahmannia, M., Amini, A., Chien, S., & Bayat, M. (2022). Impact of photobiomodulation on macrophages and their polarization during diabetic wound healing: a systematic review. *Lasers in Medical Science*, 37(7), 2805–2815. https://doi.org/10.1007/s10103-022-03581-5

Shapouri-Moghaddam, A., Mohammadian, S., Vazini, H., Taghadosi, M., Esmaeili, S. A., Mardani, F., Seifi, B., Mohammadi, A., Afshari, J. T., & Sahebkar, A. (2018). Macrophage plasticity, polarization, and function in health and disease. *Journal of Cellular Physiology*, 233(9), 6425–6440. https://doi.org/10.1002/jcp.26429

Sharifiaghdam, M., Shaabani, E., Faridi-Majidi, R., De Smedt, S. C., Braeckmans, K., & Fraire, J. C. (2022). Macrophages as a therapeutic target to promote diabetic wound healing. *Molecular therapy: the journal of the American Society of Gene Therapy*, 30(9), 2891–2908. https://doi.org/10.1016/j.ymthe.2022.07.016

Terao, M., & Katayama, I. (2016). Local cortisol/corticosterone activation in skin physiology and pathology. *Journal of Dermatological Science*, 84(1), 11–16. https://doi.org/10.1016/j.jdermsci.2016.06.014

Tu, G. W., Shi, Y., Zheng, Y. J., Ju, M. J., He, H. Y., Ma, G. G., Hao, G. W., & Luo, Z. (2017). Glucocorticoid attenuates acute lung injury through induction of type 2 macrophage. *Journal of Translational Medicine* , 15(1), 181. https://doi.org/10.1186/s12967-017-1284-7

Wu, M. H., Shih, M. H., Hsu, W. B., Dubey, N. K., Lee, W. F., Lin, T. Y., Hsieh, M. Y., Chen, C. F., Peng, K. T., Huang, T. J., Shi, C. S., Guo, R. S., Cai, C. J., Chung, C. Y., & Wong, C. H. (2017). Evaluation of a novel biodegradable thermosensitive ketohydrogel for improving postoperative pain in a rat model. *PloS one*,

12(10), e0186784. https://doi.org/10.1371/journal.pone.0186784

Yuksel, S. N., Dikmen, M., & Canturk, Z. (2021). Evaluation of Real Time Cell Proliferation, Anti-Inflammatory and Wound Healing Potential of Helenalin on HaCaT Keratinocytes Treated with Lipopolysaccharide Stimulated Monocytes. *Indian Journal of Pharmaceutical Sciences*, 83(2), 219-229.

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