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DYNAMICS OF THE NUMBER OF VESSELS DURING THE HEALING PROCESS OF EXCISIONAL WOUNDS IN RATS AFTER THE INFLUENCE OF CHRONIC SOCIAL STRESS

Makyeyeva L. V.[1,2], Frolov O. K.[1], Aliyeva O. G.[2], Tavrog M. L.[2]
1Zaporizhzhia National University, Zaporizhzhia (66 University str, Zaporizhzhia, Ukraine, 69011),
[2]Zaporizhzhia State Medical and Pharmaceutical University, Zaporizhzhia (26 Marii Prymachenko Blvd., Zaporizhzhia, Ukraine, 69035)

Responsible for correspondence:
e-mail: *lyudmylamakyeyeva@gmail.com*

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Annotation. *Optimal vascularization of tissues is a key prerequisite for wound healing and proper tissue function. In this study, we investigated the dynamics of blood vessel density during the healing of excisional wounds in rats exposed to chronic social stress (CSS). The CSS model included social isolation and contact with aggressive conspecifics. Full-thickness wounds were created in male Wistar rats. Histological and morphometric analyses of vessels at the wound edge and periwound area were performed on days 1, 3, 7, 14, and 30 post-injury. The results showed that CSS significantly delayed angiogenesis by reducing vessel density throughout all phases of repair compared to controls. This was accompanied by alterations in mast cell activity - key regulators of loose connective tissue processes, particularly vascular and immune functions. In control animals, the typical progression was observed: initial vessel loss followed by intensive neovascular growth and remodeling. In contrast, the CSS group demonstrated impaired inflammatory response and a reduction in mast cell numbers; however, the increased functional activity of mast cells did not compensate for their reduced morphogenetic regulation of loose connective tissue and angiogenesis, resulting in persistently low vascularization. The findings indicate that chronic social stress negatively affects the cellular and molecular mechanisms of vascular network restoration during wound healing, leading to delayed regeneration and an increased risk of chronic wound pathology. Promising therapeutic strategies may involve correction of the hypothalamic-pituitary-adrenal axis and restoration of diminished local blood flow in the wound area.*
Keywords: *angiogenesis, blood vessels, excisional wound, chronic social stress, wound healing.*

Introduction

Optimal vascularization of tissues and organs is a fundamental condition for the functioning of multicellular organisms, providing one of the essential prerequisites for life. Alongside the development of blood vessels, a supportive structure evolved - loose connective tissue (LCT) – which enables adequate vascular function. Its cellular foundation is the histiogenic cell lineage. The fibrous components of LCT bear the structural load, while the basic substance's acidic and alkaline mucopolysaccharides form tubular and large-cavity structures that utilize capillary movement to transport water-soluble metabolites from blood vessels to cells and vice versa [31, 74].

Numerous studies have shown that mast cells play a key role in maintaining the structural and functional homeostasis of LCT. These cells constitute up to 10% of the total LCT cell population [47]. Mast cell cytoplasmic granules contain a range of pre-synthesized biologically active substances (BAS), and upon activation, mast cells synthesize various mediators, cytokines, and growth factors *de novo* [17, 26]. Mast cells are closely interconnected with other cell types via cytoplasmic transmission channels formed by connexons, emphasizing their regulatory influence on various cells of the internal environment [15, 68]. They also form synaptic connections with sympathetic nerve fibers and engage in receptor-ligand interactions with the central neuroendocrine system [28, 36].

In LCT, mast cells are located along blood vessels and are most densely concentrated in the skin and mucous membranes - systems that are functionally exposed to the external environment [33, 38, 51]. Various skin cells participate in complex interactions and, under reparative activity, can synthesize non-typical substances and structures for their histogenetic type, maintaining the organ's structural-functional homeostasis. For instance, keratinocytes can synthesize mediators of the neurohumoral axis (hypothalamic-pituitary-adrenal, HPA axis) under stress conditions [35, 56, 63].

This study continues our investigation into the effects of chronic social stress (CSS) on the healing of incised wounds in rats. We previously observed that CSS significantly reduces the number of mast cells in the periwound area at all stages of the reparative process (inflammation, proliferation, remodeling) [21]. However, their compensatory functional activity increases through BAS degranulation and *de novo* synthesis, ultimately leading to natural depletion via apoptotic and necrotic pathways [40]. Morphometric analysis showed pronounced thinning of the skin's cellular layers in experimental animals, corresponding with delayed wound healing [41]. In this context, it was of particular interest to study the reparative response of blood vessels, which are responsible for trophic functions and the recirculation of skin cell populations during wound healing under chronic social stress.

Materials and methods

The study involved 50 sexually and socially mature male Wistar rats aged 12-13 months, divided into two groups. The control group consisted of 20 animals (4 rats per each

healing time point), and the experimental group included 30 animals (6 rats per time point), selected for their increased stress sensitivity based on open field test results.

CSS was modeled via three-week social isolation and continuous psycho-emotional influence – placing each subject rat in an environment surrounded by four aggressive conspecifics [42, 53]. Stress presence was verified via open field testing before and after the CSS model was applied [21].

Full-thickness wounds were created in the interscapular region by circular excision of skin and subcutaneous tissue (1.0-1.3 cm diameter), following aseptic preparation and under anesthesia [19].

Tissue samples were collected from the wound bed and adjacent intact skin (within 1 cm from the wound margin) on days 1, 3, 7, 14, and 30. The periwound zone (up to 5 mm from the wound edge) was the primary focus of analysis.

Animals were housed individually in plastic cages under standard vivarium conditions: temperature 20-25°C, humidity up to 55%, natural light cycle, balanced diet. Bedding was changed weekly. No signs of bacterial infection were observed during the experiment.

All experimental procedures were conducted in accordance with international and national ethical guidelines, including Directive 2010/63/EU of the European Parliament and Council (22.09.2010) and the «Common Ethical Principles for Animal Experimentation» (Ukraine, 2001).

For histological analysis, standard procedures were followed. Tissue sections were stained with hematoxylin and eosin. Photodocumentation was performed using a PrimoStar iLED microscope and an Axio Cam ERc5s camera (ZEISS, Germany). Image analysis and quantification of blood vessels were conducted using QuPath software (v0.4.4, UK).

Statistical analysis was performed using IBM SPSS Statistics 26 (IBM Corp., USA). The Kolmogorov-Smirnov test was used to assess the normality of data distribution. Student's t-test was applied to compare intergroup differences. A p-value of <0.05 was considered statistically significant. Results are presented as the mean ± standard deviation (M ± SD).

Results. Discussion

The skin is considered the largest organ in the mammalian body and, according to recent research, is classified as a super-organ, due to its integration of the cutaneous immune-neuro-endocrine system (CINE) [54, 60, 67]. In this complex, cellular and humoral components – although structurally autonomous – are anatomically and functionally interconnected through receptor-ligand interactions and coordinated via multiple signaling molecules, including peptides, neurohormones, neurotransmitters, neurotrophins, cytokines, chemokines, and growth factors. Coordination is further facilitated by cytoplasmic connexon-based communication channels [54].

The systemic regulatory link of CINE is mediated through corticotropin-releasing hormone (CRH), which activates the hypothalamic-pituitary-adrenal (HPA) axis, resulting in the secretion of glucocorticoids and catecholamines. These, in turn, stimulate local regulatory mechanisms within CINE that mirror the central axis: CRH, ACTH, corticosteroids, and catecholamines. This integrated signaling ensures the maintenance of structural and functional homeostasis in both the skin and the organism as a whole [57, 75].

Recent findings indicate that cells derived from both sessile and migratory mesenchymal lineages (e.g., keratinocytes, fibroblasts, endothelial cells, granulocytes, macrophages, dendritic cells, and lymphocytes) can, under stress conditions, express receptors and ligands atypical for their histogenetic origin [75]. This adaptive phenomenon reflects the plasticity of CINE, which enables functional adaptation under stress [3, 16, 54, 58, 59]. These findings have phylogenetic foundations—labile (metaplastic) differentiation is still seen in lower vertebrates, while higher vertebrates, including humans, retain limited capacity for noncanonical product synthesis due to genomic commonality among all cells. Cytokine-driven epigenetic de-reactivation of tissue-specific gene blocks allows such non-traditional expression patterns [7, 80].

The core function of this labile metabolic differentiation is the regulation of internal homeostasis through a balanced interplay of activating and suppressive factors across various stages of physiological or reparative regulation [50, 52].

In light of these insights, we examined restorative vascularization during incised skin wound healing at distinct phases: hemostasis/inflammation, proliferation/differentiation, and remodeling. Researchers indicate that neovascularization in skin wounds occurs through vasculogenesis - recruitment of mesenchymal-derived endothelial progenitors from the bloodstream – and angiogenesis, involving sprouting of new capillaries from existing vessels [32, 55, 66]. The relative contribution of each pathway depends significantly on the stress-reparative context of the wound.

Our and others' findings suggest that mast cells (MCs) play a central role in coordinating both physiological and reparative regeneration in LCT within the skin [4, 69, 71]. In our control samples, the mean vessel count was 3.2±0.5 per mm², aligning with physiological norms reported in the literature [27, 76]. This vascular density results from a balanced synthesis of pro-angiogenic factors – including vascular endothelial growth factor (VEGF), interleukin-8 (IL-8), and basic fibroblast growth factor (bFGF) – alongside anti-angiogenic factors such as thrombospondin, endostatin, and pigment epithelium-derived factor (PEDF) [18, 55, 61, 65].

In these same intact samples, MCs were abundant, most of them showing no signs of degranulation, indicating their regulatory function was maintained through selective transcellular secretion of bioactive mediators.

Wounding in control animals triggers an acute physiological stress, which activates both central and regional regulatory axes (CINE), leading to the release of glucocorticoids, catecholamines, and other mediators [20, 24, 52, 54, 59]. MCs and other CINE cells express receptors for these

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mediators, leading to the initiation of a local inflammatory response – aimed at damage containment, isolation from healthy tissue, and preparation for repair. The vascular and molecular mechanisms underlying this response are well described in the literature [6, 8, 37, 43, 54, 73].

At baseline (day 0), blood vessel densities (Fig. 1) were similar between the control (Fig. 2A) and experimental groups (Fig. 2B) (3.2 ± 0.5 per mm² vs. 3.4 ± 0.4 per mm²), however a reduction in the diameter of superficial vascular plexus vessels was observed in experimental group. Both groups showed significant changes over time of wound healing, with notable differences between groups on several days.

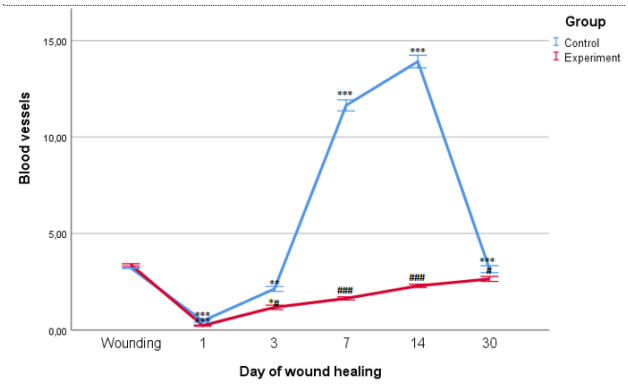

Fig. 1. Dynamics of changes in the amount of blood vessels in periwound skin of rats during healing process in control and after exposure to chronic social stress (M±SD, n=50).
Notes: * - changes are statistically significant, compared to the previous period of wound healing (p≤0.05); ** - changes are statistically significant, compared to the previous period of wound healing (p ≤0.01); *** - changes are statistically significant, compared to the previous period of wound healing (p≤0.001); # - changes are statistically significant, compared to control (p≤0.05) ### - changes are statistically significant, compared to control (p≤0.001)

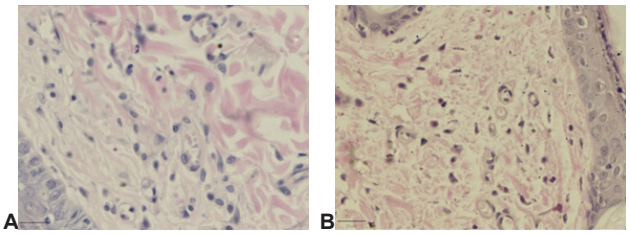

Fig. 2. Microphotographs of a skin flap excised in in control (A) and experimental (B) groups of rats at the day of wounding (H&E staining).

In our study, on day 1 post-injury, the number of blood vessels in the wound margins of control animals dropped to minimal values, reflecting destructive reorganization under the influence of proteolytic enzymes, phagocytosis, and apoptosis. Conversely, the number of MCs in the same samples more than doubled, and most exhibited high functional activity: 55.2% were fully and 19.1% partially degranulated. Their granules contain pre-synthesized mediators such as histamine, serotonin, heparin, tryptase, chymase, and carboxypeptidase, all of which drive the early inflammatory response [5, 38, 45]. MCs also produce cytokines de novo, including IL-1, IL-3, IL-6, IL-8, TNF-α, and TGF-β. Other CINE participants - including keratinocytes, fibroblasts, endothelial cells, and innate lymphoid cells - contribute as well, along with infiltrating immune cells (neutrophils, macrophages, T and B lymphocytes) [48].

The inflammatory reaction in control wounds peaked on day 3, with a sustained high MC density, ~50% of which remained partially degranulated [21, 40].

Studies have shown that hypoxia and nutrient deprivation in wounds serve as potent stimuli for angiogenic signaling, especially via proteolytic enzymes stored in MC granules. Chymase and tryptase are particularly active, exerting destructive and remodeling effects on fibrillar components of LCT. They also activate other enzymes (e.g., collagenases, MMPs), modifying the extracellular matrix and facilitating endothelial migration and proliferation – key events in angiogenesis [25, 46, 64, 79]. Furthermore, these enzymes support recruitment of granulocytes, macrophages, myofibroblasts, lymphocytes, and stem cells to the wound microenvironment [12, 17 25, 37]. These collective actions led to partial vascular restoration, as evidenced by a vessel count of 2.1±0.7 per mm² during this stage.

Effective wound healing also requires phase-appropriate cytokine and growth factor profiles. Inflammation is dominated by pro-inflammatory cytokines (list to be inserted), while the proliferation phase sees an upregulation of anti-inflammatory and pro-angiogenic mediators (e.g., VEGF, bFGF), followed by a shift toward anti-angiogenic factors in the remodeling phase [32, 70, 77].

The progressive thickening of the epidermis and dermis observed in control animals [41] further supports the notion of a well-coordinated inflammatory response, enabling successful transitions to proliferation/differentiation and subsequent remodeling phases.

By days 7 and 14, blood vessel density increased more than fourfold, reflecting intense granulation tissue formation, LCT and dermal remodeling, and keratinocyte basal layer proliferation [41]. These findings align with reports on fibroblast-to-myofibroblast transformation that facilitates wound contraction – a process confirmed in our study. On day 14, the visible wound area had reduced by 82% [41]. These morphogenetic changes were largely driven by MC-derived mediators, as our results show a fivefold increase in MCs during the proliferation phase [21].

Importantly, these changes occurred in parallel with a shift from pro-inflammatory to anti-inflammatory and pro-regenerative signaling, largely produced by the same cell populations involved in earlier phases – demonstrating the labile differentiation capacity of CINE under systemic and local hormonal regulation.

Considering that MCs do not proliferate in the skin [10, 34], their numerical increase is attributed to well-coordinated migration from the bone marrow and redistribution from other body regions. Notably, cell migration, temporary deposition, and recirculation are characteristic features of CINE function during tissue repair. Additionally, the pleiotropic effects of CINE mediators are a hallmark of this regulatory network.

By day 14 of wound healing, both cellular mass and

extracellular matrix production had increased by 4-5 times compared to baseline levels, contributing to the complete closure of the wound defect [41]. Given the intrinsic tendency of cellular systems toward expansive growth and ubiquitous distribution, these newly formed structures are subjected to differentiated restriction and remodeling during the subsequent remodeling phase, returning to the original tissue and organ architecture. This appears to be the primary role of the redundant (or local) regulatory axis within CINE, functioning in coordination with its systemic counterpart.

In our present study, by day 30, the number of blood vessels in the skin of control animals returned to near-baseline levels (3.0±1.2 vessels per mm²), indicating the physiological completion of wound remodeling. At this stage, the synthesis of vasoactive bioactive compounds shifted from pro-angiogenic to anti-angiogenic factors. Restoration of vascular density in the wound margin was primarily achieved via apoptosis of endothelial cells, mediated by macrophages [18, 55, 65]. During this period, remodeling also affected LCT around vessels, the papillary dermis, and subcutaneous adipose tissue, mediated by interactions between MCs and resident skin cells (keratinocytes, endothelial cells, fibroblasts), primarily through vesicular secretion of mediators without overt degranulation [11, 46, 79].

Beyond receptor-ligand interactions, gap junction communication was also detected. Through connexon channels (~2 nm wide), low-molecular-weight molecules, including steroid hormones, can diffuse between cells, influencing cell differentiation processes [16, 32].

These morphogenetic transformations occurred alongside the structural restoration of the skin's layers [41], with the active involvement of MC-derived BASs. The number and metabolic activity of MCs also returned to baseline values [21, 40].

A markedly different vascular response was observed in animals exposed to CSS. Although the initial pre-wound vessel density did not differ significantly from controls, pronounced molecular and cellular alterations were already present. Our previous findings from the same experimental model showed a several-fold increase in MCs, with 36-37% of them partially degranulated [21, 40]. Additionally, there was a decrease in epidermal and dermal thickness [41].

These changes reflect heightened activation of the HPA axis and its local analog in the skin [56]. Physiologically, GCs regulate tissue metabolism via receptors found in nearly all cell types. Under stress, MC-derived mediators play a crucial role in maintaining CINE homeostasis [54]. Notably, serotonin, preformed in MC granules, regulates smooth muscle tone and precise vasomotor control of precapillary vessels in LCT, ensuring tissue perfusion.

In stress conditions, this serotonin-mediated vasomotor control is disrupted, impairing microcirculation, leading to local hypoxia, metabolic stress, and activation of secondary inflammatory responses, often involving vasoconstrictors, stress hormones, and cytokines [9, 29 72]. The cutaneous vascular network enters a spastic state, redirecting blood flow toward vital organs (CNS, excretory system, heart, GI tract, lungs). To prevent overwhelming inflammation in such a large organ, the cytokine profile in the skin adaptively shifts toward anti-inflammatory cytokines such as IL-4, IL-5, IL-10, IL-13, etc [2, 22, 49].

Additionally, chronic stress-induced increases in GCs and catecholamines disrupt blood cell recirculation, promoting cellular sequestration in the skin [1, 23, 30, 39]. Thus, the pre-wound increase in MCs we observed likely reflects an adaptive response to maintain multifunctional metabolic pathways in the skin [21].

In experimental animals, the act of skin wounding itself induced additional acute stress, layered atop chronic stress, producing accentuated hormonal shifts in CINE. Consequently, the Th2-dominated cytokine profile suppressed pro-inflammatory cytokine production (IL-1, IL-2, IL-8, IFN-γ, TNF-α), delaying the onset of inflammation, impairing infiltration of immune cells (neutrophils, eosinophils, lymphocytes, monocytes/macrophages), and reducing immune competence in the wound zone. This delayed inflammatory response increased the risk of chronic wound pathology [13, 24, 44, 78].

Indeed, by day 1, only single vessels were detectable in the wound margins, and by day 3, vessel counts still lagged behind control levels (2.1±0.5 in control vs 1.2±0.4, $p<0.01$ per mm² in experiment). MC numbers were almost halved, though their functional activity was heightened: the cells were larger, and most showed full or partial degranulation [21, 40]. Morphometric analysis revealed nearly twofold delay in epidermal layer restoration, particularly in the basal layer, and dermal thickness was also reduced [41].

These adverse morphogenetic outcomes during the first 3 days were due to suppression of the inflammatory phase under the influence of elevated GCs and catecholamines. Previous studies [17, 52, 62] have shown that while chronic stress enhances MC mobilization and degranulation, it impairs the recruitment and function of macrophages, particularly their phagocytic and cytokine-producing capacity. As a result, key clearance and preparatory mechanisms are compromised, affecting the proliferation and remodeling phases of regeneration [52, 62].

As expected, vascular density in the stressed group remained low during the proliferation and differentiation phases. On days 7 and 14, the number of vessels did not reach pre-injury values, in stark contrast to the several-fold neovascularization increase seen in controls (1.6±0.3 vs 11.6±1.4 per mm² at day 7 and 2.2±0.4 vs 13.7±1.5 per mm²). The low angiogenic response during this time reflects inhibited proliferative and differentiation responses in skin layers. Granulation tissue formation appeared only as small islands by day 7 [41], and the epidermis remained less than half as thick as in controls. As a result, by day 14, only 46.7% of the wound surface was covered with primary matrix, compared to 82,5% in controls [41].

Notably, MC counts during this phase were more than two times lower than in controls. However, their functional

activity was elevated: they were larger, and over 65% were fully or partially degranulated [21, 40]. These findings suggest that, under the influence of stress hormones, MC recruitment from bone marrow persisted, and even local accumulation (deposition**)** occurred. Yet, increased metabolic demands for reparative homeostasis led to quantitative depletion of MCs during active repair [21].

The delayed regeneration observed in inflammatory and proliferative phases continued into remodeling. By day 30, tissue reorganization was still incomplete. We observed deficiencies in cellular and extracellular matrix content [41], and vascular density remained low (2.6±0.2 per mm² compared to 3.4±0.4 per mm² pre-injury). Incomplete epithelialization was evidenced by residual defect areas covered by scabs. The epidermal thickness was significantly reduced, while dermal thickness tended to increase [41], reflecting ongoing remodeling and revascularization processes.

Crucially, by this point we again observed a surge in MC numbers, increasing by 5-fold compared to controls, with 53.8% partially degranulated, and larger cell size [40].

All these findings highlight that reparative processes were still ongoing and that their completion was significantly delayed by chronic social stress.

Conclusion and prospects for further developments

1. Chronic social stress contributes to the chronification of reparative responses at all stages of excisional skin wound healing in experimental animals. This is primarily due to an acute deficit in the recirculation of the full spectrum of cells involved in tissue regeneration within the wound area.

Therefore, a key objective for current and future pharmacological research and therapeutic strategies should be focused on:

- Mitigating the stress impact on the central hypothalamic-pituitary-adrenal (HPA) regulatory axis;
- And enhancing local blood supply within the wound and adjacent tissues.

References

[1] Adameova, A., Abdellatif, Y., & Dhalla, N. S. (2009). Role of the excessive amounts of circulating catecholamines and glucocorticoids in stress-induced heart disease. *Canadian Journal of Physiology and Pharmacology, 87*(7), 493-514. https://doi.org/10.1139/y09-042

[2] Alotiby, A. (2024). Immunology of stress: A review article. *Journal of Clinical Medicine, 13*(21), 6394. https://doi.org/10.3390/jcm13216394

[3] Arck, P. C., Slominski, A., Theoharides, T. C., Peters, E. M. J., & Paus, R. (2006). Neuroimmunology of stress: Skin takes center stage. *Journal of Investigative Dermatology, 126*(8), 1697-1704. https://doi.org/10.1038/sj.jid.5700104

[4] Atiakshin, D., Soboleva, M., Nikityuk, D., Alexeeva, N., Klochkova, S., Kostin, A., … & Tiemann, M. (2023). Mast cells in regeneration of the skin in burn wound with special emphasis on molecular hydrogen effect. *Pharmaceuticals, 16*(3), 348. https://doi.org/10.3390/ph16030348

[5] Azouz, N. P., Hammel, I., & Sagi-Eisenberg, R. (2014). Characterization of mast cell secretory granules and their cell biology. *DNA and Cell Biology, 33*(10), 647-651. https://doi.org/10.1089/dna.2014.2543

[6] Beghdadi, W., Madjene, L. C., Benhamou, M., Charles, N., Gautier, G., Launay, P., & Blank, U. (2011). Mast cells as cellular sensors in inflammation and immunity. *Frontiers in Immunology, (2),* 37. https://doi.org/10.3389/fimmu.2011.00037

[7] Brunet, T., & King, N. (2017). The origin of animal multicellularity and cell differentiation. *Developmental Cell, 43*(2), 124-140. https://doi.org/10.1016/j.devcel.2017.09.016

[8] Cai, C., Cao, Z., Loughran, P. A., Kim, S., Darwiche, S., Korff, S., & Billiar, T. R. (2011). Mast cells play a critical role in the systemic inflammatory response and end-organ injury resulting from trauma. *Journal of the American College of Surgeons, 213*(5), 604-615. https://doi.org/10.1016/j.jamcollsurg.2011.08.009

[9] Cairns, M., Marais, E., Joseph, D., & Essop, M. F. (2025). The role of chronic stress in the pathogenesis of ischemic heart disease in women. *Comprehensive Physiology, 15*(1), e70000. https://doi.org/10.1002/cph4.70000

[10] Chen, H., Xu, Y., Yang, G., Zhang, Q., Huang, X., Yu, L., & Dong, X. (2017). Mast cell chymase promotes hypertrophic scar fibroblast proliferation and collagen synthesis by activating TGF-β1/Smads signaling pathway. *Experimental and Therapeutic Medicine, 14*(6), 4438-4442. https://doi.org/10.3892/etm.2017.5082

[11] Chen, L., Schrementi, M. E., Ranzer, M. J., Wilgus, T. A., & DiPietro, L. A. (2014). Blockade of mast cell activation reduces cutaneous scar formation. *PLoS One, 9*(1), e85226. https://doi.org/10.1371/journal.pone.0085226

[12] Chesko, D. M., & Wilgus, T. A. (2022). Immune cells in cutaneous wound healing: A review of functional data from animal models. *International Journal of Molecular Sciences, 23*(5), 2444. https://doi.org/10.3390/ijms23052444

[13] Christian, L. M., Graham, J. E., Padgett, D. A., Glaser, R., & Kiecolt-Glaser, J. K. (2006). Stress and wound healing. *Neuroimmunomodulation, 13*(5-6), 337-346. https://doi.org/10.1159/000104862

[14] de Almeida, T. F., de Castro Pires, T., & Monte-Alto-Costa, A. (2016). Blockade of glucocorticoid receptors improves cutaneous wound healing in stressed mice. *Experimental Biology and Medicine, 241*(4), 353-358. https://doi.org/10.1177/1535370215612940

[15] de Souza Junior, D. A., Mazucato, V. M., Santana, A. C., Oliver, C., & Jamur, M. C. (2017). Mast cells interact with endothelial cells to accelerate in vitro angiogenesis. *International Journal of Molecular Sciences, 18*(12), 2674. https://doi.org/10.3390/ijms18122674

[16] Dhabhar, F. S. (2013). Psychological stress and immunoprotection versus immunopathology in the skin. *Clinical Dermatology, 31*(1), 18-30. https://doi.org/10.1016/j.clindermatol.2011.11.003

[17] Dileepan, K. N., Raveendran, V. V., Sharma, R., Abraham, H., Barua, R., Singh, V., … & Sharma, M. (2023). Mast cell-mediated immune regulation in health and disease. *Frontiers in Medicine, (10),* 1213320. https://doi.org/10.3389/fmed.2023.1213320

[18] DiPietro, L. A. (2016). Angiogenesis and wound repair: When enough is enough. *Journal of Leukocyte Biology, 100*(5), 979-984. https://doi.org/10.1189/jlb.4MR0316-102R

[19] Dorsett-Martin, W. (2004). Rat models of skin wound healing: A review. *Wound Repair and Regeneration, 12*(6), 591-599. https://doi.org/10.1111/j.1067-1927.2004.12601.x

[20] Doshi, B. M., Perdrizet, G. A., & Hightower, L. E. (2008). Wound healing from a cellular stress response perspective. *Cell Stress and Chaperones, 13*(4), 393-399. https://doi.org/10.1007/s12192-008-0059-8

[21] Frolov, O. K., Makyeyeva, L. V., & Aliyeva, O. G. (2021). Quantitative characteristics of mast cells in the course of

wound healing in rats with chronic social stress. *Acta Biologica Ukrainica, 1*(1), 34-41.

[22] Gabbay, V., Ely, B. A., Vileisis, J. N., Petrovic, Z., Cicvaric, A., Asnis, G., … & Radulovic, J. (2024). Immune and neural response to acute social stress in adolescent humans and rodents. *Translational Psychiatry, (14),* 306. https://doi.org/10.1038/s41398-024-03008-5

[23] Geiger, A. M., Pitts, K. P., Feldkamp, J., Kirschbaum, C., & Wolf, J. M. (2015). Cortisol-dependent stress effects on cell distribution in healthy individuals and individuals suffering from chronic adrenal insufficiency. *Brain, Behavior, and Immunity, (50),* 241-248. https://doi.org/10.1016/j.bbi.2015.07.010

[24] Gouin, J. P., & Kiecolt-Glaser, J. K. (2011). The impact of psychological stress on wound healing: Methods and mechanisms. *Immunology and Allergy Clinics of North America, 31*(1), 81-93. https://doi.org/10.1016/j.iac.2010.09.010

[25] Guth, C., Limjunyawong, N., & Pundir, P. (2024). The evolving role of mast cells in wound healing: Insights from recent research and diverse models. *Immunology and Cell Biology, 102*(10), 878-890. https://doi.org/10.1111/imcb.12824

[26] Hellman, L., Akula, S., Fu, Z., & Wernersson, S. (2022). Mast cell and basophil granule proteases – In vivo targets and function. *Frontiers in Immunology, (13),* 918305. https://doi.org/10.3389/fimmu.2022.918305

[27] Hoshino, T., Cheng, Y., Ninomiya, M., Katsuyama, M., Yamashita, T., Katagiri, C., … & Hara, Y. (2024). Diameter-dependent classification of dermal vasculature using optical coherence tomography angiography. *Quantitative Imaging in Medicine and Surgery, 14*(9), 6238-6249. https://doi.org/10.21037/qims-23-1790

[28] Huang, X., Lan, Z., & Hu, Z. (2024). Role and mechanisms of mast cells in brain disorders. *Frontiers in Immunology, (15),* 1445867. https://doi.org/10.3389/fimmu.2024.1445867

[29] Iadecola, C. (2017). The neurovascular unit coming of age: A journey through neurovascular coupling in health and disease. *Neuron, 96*(1), 17-42. https://doi.org/10.1016/j.neuron.2017.07.030

[30] Ince, L. M., Weber, J., & Scheiermann, C. (2019). Control of leukocyte trafficking by stress-associated hormones. *Frontiers in Immunology, (9),* 3143. https://doi.org/10.3389/fimmu.2018.03143

[31] Janson, K. D., Parkhideh, S., Swain, J. W. R., Weaver, J. D., Hartgerink, J. D., & Veiseh, O. (2025). Strategies for the vascular patterning of engineered tissues for organ repair. *Nature Biomedical Engineering, (9),* 1007-1025. https://doi.org/10.1038/s41551-025-01420-w

[32] Johnson, K. E., & Wilgus, T. A. (2014). Vascular endothelial growth factor and angiogenesis in the regulation of cutaneous wound repair. *Advances in Wound Care, 3*(10), 647-661. https://doi.org/10.1089/wound.2013.0517

[33] Keith, Y. H., Egawa, G., Honda, T., & Kabashima, K. (2023). Mast cells in type 2 skin inflammation: Maintenance and function. *European Journal of Immunology, (53),* e2250359. https://doi.org/10.1002/eji.202250359

[34] Kennelly, R., Conneely, J. B., Bouchier-Hayes, D., & Winter, D. C. (2011). Mast cells in tissue healing: From skin to the gastrointestinal tract. *Current Pharmaceutical Design, 17*(34), 3772-3775. https://doi.org/10.2174/138161211798357854

[35] Kimura, S., Tsuchiya, A., Ogawa, M., Ono, M., Suda, N., Sekimoto, K., … & Tsuji, T. (2020). Tissue-scale tensional homeostasis in skin regulates structure and physiological function. *Communications Biology, (3),* 637. https://doi.org/10.1038/s42003-020-01365-7

[36] Kleij, H. P., & Bienenstock, J. (2005). Significance of conversation between mast cells and nerves. *Allergy, Asthma & Clinical Immunology, 1*(2), 65-80. https://doi.org/10.1186/1710-1492-1-2-65

[37] Komi, D. E. A., Khomtchouk, K., & Santa Maria, P. L. (2020). A review of the contribution of mast cells in wound healing: Involved molecular and cellular mechanisms. *Clinical Reviews in Allergy & Immunology, 58*(3), 298-312. https://doi.org/10.1007/s12016-019-08729-w

[38] Krystel-Whittemore, M., Dileepan, K. N., & Wood, J. G. (2016). Mast cell: A multi-functional master cell. *Frontiers in Immunology, (6)*, 620. https://doi.org/10.3389/fimmu.2015.00620

[39] Kvetnansky, R., Lu, X., & Ziegler, M. G. (2013). Stress-triggered changes in peripheral catecholaminergic systems. *Advances in Pharmacology, (68),* 359-397. https://doi.org/10.1016/B978-0-12-411512-5.00017-8

[40] Makyeyeva, L., Frolov, O., & Aliyeva, O. (2024). Functional changes in skin mast cells during surgical wound healing in rats after the influence of chronic social stress. *Fitoterapiia. Chasopys – Phytotherapy. Journal, (2)*, 36-46. https://doi.org/10.32782/2522-9680-2024-2-36

[41] Makyeyeva, L., Frolov, O., & Aliyeva, O. (2025). Morphometric changes in rat periwound skin during healing of excisional wounds after exposure to chronic social stress. *Innovative Biosystems and Bioengineering, 9*(1), 13-25. URL: http://dspace.zsmu.edu.ua/handle/123456789/22136

[42] Masis-Calvo, M., Schmidtner, A. K., de Moura Oliveira, V. E., Grossmann, C. P., de Jong, T. R., & Neumann, I. D. (2018). Animal models of social stress: The dark side of social interactions. *Stress, 21*(5), 417-432. https://doi.org/10.1080/10253890.2018.1461832

[43] Migalovich-Sheikhet, H., Friedman, S., Mankuta, D., & Levi-Schaffer, F. (2012). Novel identified receptors on mast cells. *Frontiers in Immunology, (3)*, 238. https://doi.org/10.3389/fimmu.2012.00238

[44] Mochel, K., Bronte, J., Kasaba, M., Vempati, A., Tam, C., & Hazany, S. (2025). The impact of psychological stress on wound healing: Implications for neocollagenesis and scar treatment efficacy. *Clinical, Cosmetic and Investigational Dermatology, (18),* 1625-1637. https://doi.org/10.2147/CCID.S528730

[45] Moon, T. C., Befus, A. D., & Kulka, M. (2014). Mast cell mediators: Their differential release and the secretory pathways involved. *Frontiers in Immunology, (5)*, 569. https://doi.org/10.3389/fimmu.2014.00569

[46] Ng, M. F. (2010). The role of mast cells in wound healing. *International Wound Journal, 7*(1), 55-61. https://doi.org/10.1111/j.1742-481X.2009.00651.x

[47] Norrby, K. (2024). On connective tissue mast cells as protectors of life, reproduction, and progeny. *International Journal of Molecular Sciences, 25*(8), 4499. https://doi.org/10.3390/ijms25084499

[48] Oskeritzian, C. A. (2012). Mast cells and wound healing. *Advances in Wound Care, 1*(1), 23-28. https://doi.org/10.1089/wound.2011.0357

[49] Peters, E. M. (2016). Stressed skin? A molecular psychosomatic update on stress-causes and effects in dermatologic diseases. *Journal der Deutschen Dermatologischen Gesellschaft, 14*(3), 233-252. https://doi.org/10.1111/ddg.12957

[50] Pondeljak, N., & Lugović-Mihić, L. (2020). Stress-induced interaction of skin immune cells, hormones, and neurotransmitters. *Clinical Therapeutics, 42*(5), 757-770. https://doi.org/10.1016/j.clinthera.2020.03.008

[51] Ribatti, D. (2024). Mast cells are at the interface between the external environment and the inner organism. *Frontiers in Medicine, (10)*, 1332047. https://doi.org/10.3389/fmed.2023.1332047

[52] Romana-Souza, B., Chen, L., & DiPietro, L. A. (2023). Repeated stress-induced crosstalk between the sympathetic nervous system and mast cells contributes to delayed cutaneous wound healing in mice. *Journal of Neuroimmunology, (379)*, 578104. https://doi.org/10.1016/j.jneuroim.2023.578104

[53] Rygula, R., Abumaria, N., Flügge, G., Fuchs, E., Rüther, E., & Havemann-Reinecke, U. (2005). Anhedonia and motivational deficits in rats: Impact of chronic social stress. *Behavioural Brain Research, 162*(1), 127-134. https://doi.org/10.1016/j.bbr.2005.03.009

[54] Shastri, M., Sharma, M., Sharma, K., Sharma, A., Minz, R. W., Dogra, S., & Chhabra, S. (2024). Cutaneous-immuno-neuroendocrine (CINE) system: A complex enterprise transforming skin into a super organ. *Experimental Dermatology, 33*(3), e15029. https://doi.org/10.1111/exd.15029

[55] Shi, Z., Yao, C., Shui, Y., Li, S., & Yan, H. (2023). Research progress on the mechanism of angiogenesis in wound repair and regeneration. *Frontiers in Physiology, (14)*, 1284981. https://doi.org/10.3389/fphys.2023.1284981

[56] Skobowiat, C., Dowdy, J. C., Sayre, R. M., Tuckey, R. C., & Slominski, A. (2011). Cutaneous hypothalamic-pituitary-adrenal axis homolog: Regulation by ultraviolet radiation. *American Journal of Physiology-Endocrinology and Metabolism, 301*(3), E484-E493. https://doi.org/10.1152/ajpendo.00217.2011

[57] Slominski, A. (2009). On the role of the corticotropin-releasing hormone signalling system in the aetiology of inflammatory skin disorders. *British Journal of Dermatology, 160*(2), 229-232. https://doi.org/10.1111/j.1365-2133.2008.08958.x

[58] Slominski, A. T., Botchkarev, V., Choudhry, M., Fazal, N., Fechner, K., Furkert, J., … & Paus, R. (1999). Cutaneous expression of CRH and CRH-R: Is there a «skin stress response system?» *Annals of the New York Academy of Sciences, (885)*, 287-311. https://doi.org/10.1111/j.1749-6632.1999.tb08686.x

[59] Slominski, A. T., Zmijewski, M. A., Zbytek, B., Tobin, D. J., Theoharides, T. C., & Rivier, J. (2013). Key role of CRF in the skin stress response system. *Endocrine Reviews, 34*(6), 827-884. https://doi.org/10.1210/er.2012-1092

[60] Slominski, A., Wortsman, J., Paus, R., Elias, P. M., Tobin, D. J., & Feingold, K. R. (2008). Skin as an endocrine organ: Implications for its function. *Drug Discovery Today: Disease Mechanisms, 5*(2), 137-144. https://doi.org/10.1016/j.ddmec.2008.04.004

[61] Song, H., Hao, D., Zhou, J., Farmer, D., & Wang, A. (2024). Development of pro-angiogenic skin substitutes for wound healing. *Wound Repair and Regeneration, 32*(3), 208-216. https://doi.org/10.1111/wrr.13154

[62] Theoharides, T. C. (2020). The impact of psychological stress on mast cells. *Annals of Allergy, Asthma & Immunology, 125*(4), 388-392. https://doi.org/10.1016/j.anai.2020.07.007

[63] Tong, R. L., Kahn, U. N., Grafe, L. A., Hitti, F. L., Fried, N. T., & Corbett, B. F. (2023). Stress circuitry: Mechanisms behind nervous and immune system communication that influence behavior. *Frontiers in Psychiatry, (14)*, 1240783. https://doi.org/10.3389/fpsyt.2023.1240783

[64] Tonnesen, M. G., Feng, X., & Clark, R. A. (2000). Angiogenesis in wound healing. *Journal of Investigative Dermatology Symposium Proceedings, 5*(1), 40-46. https://doi.org/10.1046/j.1087-0024.2000.00014.x

[65] Veith, A. P., Henderson, K., Spencer, A., Sligar, A. D., & Baker, A. B. (2019). Therapeutic strategies for enhancing angiogenesis in wound healing. *Advanced Drug Delivery Reviews, (146),* 97-125. https://doi.org/10.1016/j.addr.2018.09.010

[66] Velazquez, O. C. (2007). Angiogenesis and vasculogenesis: Inducing the growth of new blood vessels and wound healing by stimulation of bone marrow-derived progenitor cell mobilization and homing. *Journal of Vascular Surgery, 45*(A), 39-47. https://doi.org/10.1016/j.jvs.2007.02.068

[67] Vidal Yucha, S. E., Tamamoto, K. A., & Kaplan, D. L. (2019). The importance of the neuro-immuno-cutaneous system on human skin equivalent design. *Cell Proliferation, 52*(6), e12677. https://doi.org/10.1111/cpr.12677

[68] Vliagoftis, H., Hutson, A. M., Mahmudi-Azer, S., Kim, H., Rumsaeng, V., Oh, C. K., Moqbel, R., & Metcalfe, D. D. (1999). Mast cells express connexins on their cytoplasmic membrane. *Journal of Allergy and Clinical Immunology, 103*(4), 656-662. https://doi.org/10.1016/s0091-6749(99)70239-3

[69] Voss, M., Kotrba, J., Gaffal, E., Katsoulis-Dimitriou, K., & Dudeck, A. (2021). Mast cells in the skin: Defenders of integrity or offenders in inflammation? *International Journal of Molecular Sciences, (22),* 4589. https://doi.org/10.3390/ijms22094589

[70] Wallace, H. A., Basehore, B. M., & Zito, P. M. (2025). Wound healing phases. In: *StatPearls* [Internet]. StatPearls Publishing. https://www.ncbi.nlm.nih.gov/books/NBK470443/

[71] Wang, R. M., Mesfin, J. M., Karkanitsa, M., Ungerleider, J. L., Zelus, E., Zhang, Y., … & Christman, K. L. (2023). Immunomodulatory contribution of mast cells to the regenerative biomaterial microenvironment. *NPJ Regenerative Medicine, 8*(1), 53. https://doi.org/10.1038/s41536-023-00324-0

[72] Wang, X., & He, B. (2024). Endothelial dysfunction: Molecular mechanisms and clinical implications. *MedComm, 5*(8), e651. https://doi.org/10.1002/mco2.651

[73] Wang, X., Zhang, P., Tang, Y., Chen, Y., Zhou, E., & Gao, K. (2024). Mast cells: A double-edged sword in inflammation and fibrosis. *Frontiers in Cell and Developmental Biology, (12),* 1466491. https://doi.org/10.3389/fcell.2024.1466491

[74] Wang, Y., Keshavarz, M., Barhouse, P., & Smith, Q. (2023). Strategies for regenerative vascular tissue engineering. *Advanced Biology, 2200050.* https://doi.org/10.1002/adbi.202200050

[75] Webster Marketon, J. I., & Glaser, R. (2008). Stress hormones and immune function. *Cellular Immunology, 252*(1-2), 16-26. https://doi.org/10.1016/j.cellimm.2007.09.006

[76] Wells, M. Y., Voute, H., Bellingard, V., Fisch, C., Boulifard, V., George, C., & Picaut, P. (2010). Histomorphology and vascular lesions in dorsal rat skin used as injection sites for a subcutaneous toxicity study. *Toxicologic Pathology, 38*(2), 258-266. https://doi.org/10.1177/0192623309357953

[77] Wong, R. S. Y., Tan, T., Pang, A. S. R., & Srinivasan, D. K. (2025). The role of cytokines in wound healing: From mechanistic insights to therapeutic applications. *Exploration of Immunology, (5),* 1003183. https://doi.org/10.37349/ei.2025.1003183

[78] Woo, K. Y. (2012). Chronic wound-associated pain, psychological stress, and wound healing. *Surgical Technology International, (22),* 57-65.

[79] Wulff, B. C., & Wilgus, T. A. (2013). Mast cell activity in the healing wound: More than meets the eye? *Experimental Dermatology, 22*(8), 507-510. https://doi.org/10.1111/exd.12169

[80] Yang, D., Jones, M. G., Naranjo, S., Rideout, W. M. III, Min, K. H. J., Ho, R., … & Weissman, J. S. (2022). Lineage tracing reveals the phylodynamics, plasticity, and paths of tumor evolution. *Cell, 185*(11), 1905-1923.e25. https://doi.org/10.1016/j.cell.2022.04.015

ДИНАМІКА КІЛЬКОСТІ СУДИН ПІД ЧАС ПРОЦЕСУ ЗАГОЄННЯ РІЗАНИХ РАН У ЩУРІВ ПІД ВПЛИВОМ ХРОНІЧНОГО СОЦІАЛЬНОГО СТРЕСУ

***Макєєва Л. В., Фролов* О. К., Алієва О. Г., *Таврог* М. Л.**

Анотація. *Оптимальна васкуляризація тканин є ключовою умовою процесу загоєння ран і функціонування тканин. У даному дослідженні вивчено динаміку кількості кровоносних судин під час загоєння ексцизійних ран у щурів, які перебували під впливом хронічного соціального стресу (ХСС). Модель ХСС включала соціальну ізоляцію та контакт із агресивними*

одноплемінниками. На самцях щурів лінії Вістар були створені повнотовщинні рани. Гістологічний і морфометричний аналіз судин у зоні краю рани та навколо неї виконували на 1, 3, 7, 14 та 30 добу після нанесення рани. Результати показали, що ХСС суттєво уповільнює ангіогенез, зменшуючи щільність судин у всі фази репарації порівняно з контролем. Це супроводжувалося змінами активності тучних клітин – ключового регулятора всіх процесів пухкої сполучної тканини, зокрема васкулярних та імунних. У контрольних тварин спостерігалось типове прогресування початкової втрати судин із подальшим інтенсивним неоваскулярним ростом і ремоделюванням, тоді як у групі ХСС порушувалась запальна відповідь, кількість тучних клітин знижувалась, однак підвищення їх функціональної активності не компенсувало їх морфогенетичну регуляцію пухкої волокнистої сполучної тканини і ангіогенез, тому васкуляризація залишалась низькою. Отримані дані свідчать, що хронічний соціальний стрес негативно впливає на клітинні та молекулярні механізми відновлення судинної мережі у процесі загоєння, спричиняючи затримку регенерації та підвищений ризик хронічної ранової патології. Перспективними напрямками терапії є корекція гіпоталамо-гіпофізарно-надниркової осі та відновлення зниженого локального кровообігу у рановій зоні.

Ключові слова: *ангіогенез, кровоносні судини, різана рана, хронічний соціальний стрес, загоєння рани.*