Research Paper

Analysis of Mitochondrial-Related Proteins in Modulating Wound Healing Pathways: A Network-Based Study

Seyedeh Sara Azadeh [1,2], Hoda Keshmiri Neghab [2,*]

[1] Laboratory of Bioanalysis, Institute of Biochemistry & Biophysics, University of Tehran, Enghelab Street, Tehran, Iran
[2] Department of Medical Laser, Medical Laser Research Center, Yara Institute, ACECR, Tehran, Iran

Scan to access website

How to cite this paper:
Azadeh SS, Keshmiri Neghab H. Analysis of Mitochondrial-Related Proteins in Modulating Wound Healing Pathways: A Network-Based Study. Advances in Skin Wound and Tissue Repair. 2025; 22(4):15-23.
10.66224/aswtr.22.4.15

Article info
Received: 28/10/2025
Accepted: 09/11/2025
Published: 30/12/2025

*** Corresponding author:**
Hoda Keshmiri Neghab, Department of Medical Laser, Medical Laser Research Center, Yara Institute, ACECR, Tehran, Iran. E-mail: hodakeshmiri@ut.ac.ir

ABSTRACT

Background: Wound healing is a complex, multi-stage process regulated by the interplay of metabolic, inflammatory, and redox signaling pathways. However, the systems-level integration of these mechanisms, particularly the role of mitochondrial function, remains insufficiently understood.
Methods: In this study, an in silico systems biology approach was employed to investigate interactions among ten key regulatory genes (HIF1A, AKT1, MAPK1, MTOR, NFKB1, STAT3, SIRT1, PPARGC1A, NOX4, and PRKAA1). A protein–protein interaction network was constructed using STRING and analyzed in Cytoscape to identify hub genes and functional modules. KEGG pathway enrichment analysis was performed to determine significantly associated biological pathways.
Results: The PPI network revealed a highly interconnected structure with central hub genes including AKT1, MTOR, MAPK1, STAT3, and NFKB1. Enrichment analysis identified key pathways such as HIF-1, PI3K–Akt, MAPK, AMPK, and NF-κB signaling. Mitochondrial regulators (SIRT1, PPARGC1A, PRKAA1) showed strong connectivity with growth and inflammatory pathways, while NOX4-mediated redox signaling was linked to both angiogenesis and immune responses.
Conclusion: These findings demonstrate that wound healing is governed by an integrated regulatory network in which mitochondrial function and redox balance play central roles in coordinating cellular metabolism, inflammation, and tissue regeneration.

Keywords: Wound healing, Mitochondrial Function, Protein–protein Interaction, KEGG Pathway Analysis, Oxidative Stress, Systems Biology

Introduction

Overview of Wound healing and Molecular Complexity

Wound healing is a highly coordinated and dynamic biological process consisting of four overlapping phases: hemostasis, inflammation, proliferation, and remodeling[1]. Each phase is regulated by complex interactions between inflammatory cells, fibroblasts, keratinocytes, endothelial cells, and extracellular matrix components, all of which are controlled by tightly regulated signaling pathways. Dysregulation of these molecular mechanisms, particularly in chronic wounds such as diabetic ulcers, leads to impaired angiogenesis, persistent inflammation, and defective tissue regeneration. Therefore, identifying key regulatory genes involved in cellular metabolism, inflammation, oxidative stress, and angiogenesis is essential for understanding impaired wound healing and developing targeted therapeutic strategies[2].in this article ,to investigate the integrated relationship between metabolic regulation, oxidative stress, and injury-induced signaling pathways, a focused panel of key regulatory genes was selected to represent the core functional networks involved in wound healing.

Hypoxia Signaling and Angiogenesis: Role of *HIF1A*

Hypoxia is a fundamental micro environmental condition in wounded tissue, and cellular adaptation to low oxygen levels is primarily regulated by hypoxia-inducible factor 1 alpha (*HIF1A*)[3]. *HIF1A* acts as a master transcription factor that induces the expression of angiogenic genes such as VEGF, promotes endothelial cell migration, and enhances glycolytic metabolism to support cell survival under hypoxic conditions[4]. In the early proliferative phase of wound healing, HIF1A-mediated signaling is crucial for neovascularization and oxygen delivery to regenerating tissues[5]. Impaired *HIF1A* activation has been strongly

[Downloaded from icml.ir on 2026-10-14]
[DOI: 10.66224/aswtr.22.4.15]

associated with delayed angiogenesis and chronic wound formation, particularly in diabetic conditions[6].

Cell Survival and Migration: Role of *AKT1* and *MAPK1*

The *PI3K/AKT* and *MAPK* signaling pathways are central regulators of cell survival, proliferation, and migration during wound repair [7]. *AKT1* promotes keratinocyte and fibroblast survival by inhibiting apoptosis and enhancing glucose uptake, thereby supporting energy-demanding regenerative processes. Additionally, *AKT1* facilitates endothelial cell migration and tube formation, contributing to angiogenesis[8]. *MAPK1* (*ERK2*), a critical component of the *MAPK* pathway, regulates cell cycle progression, fibroblast proliferation, and keratinocyte migration, which are essential for wound closure and re-epithelialization. Activation of *MAPK1* signaling accelerates granulation tissue formation and extracellular matrix deposition, both required for effective tissue regeneration [9].

Inflammatory Regulation: Role of *NFKB1* and *STAT3*

Inflammation is necessary for initial wound defense but must be tightly controlled to allow progression to tissue repair. *NFKB1* is a key transcription factor that regulates the expression of pro-inflammatory cytokines, chemokines, and adhesion molecules[10]. While early activation of *NF-κB* promotes immune cell recruitment and pathogen clearance, sustained activation leads to chronic inflammation and impaired healing [11]. *STAT3* plays a dual regulatory role by promoting keratinocyte migration and fibroblast activation while also contributing to immune modulation. *STAT3* signaling supports epithelial regeneration and angiogenesis but, similar to NF-κB, requires precise temporal regulation to prevent excessive inflammation and fibrosis [12].

Mitochondrial Function and Metabolic Regulation: Role of *SIRT1* and *PPARGC1A*

Mitochondrial biogenesis and cellular energy metabolism are increasingly recognized as critical determinants of wound healing capacity. *SIRT1*, a NAD+-dependent deacetylase, regulates mitochondrial function, oxidative stress resistance, and cellular longevity[13]. Through deacetylation of transcription factors and coactivators, *SIRT1* enhances cellular stress tolerance and promotes endothelial function. *PPARGC1A (PGC-1α)* is a master regulator of mitochondrial biogenesis and oxidative metabolism[14]. Upregulation of *PPARGC1A* improves ATP production, enhances antioxidant defenses, and supports fibroblast and keratinocyte activity. Together, *SIRT1* and *PPARGC1A* coordinate metabolic reprogramming that supplies sufficient energy for cell migration, proliferation, and matrix synthesis during tissue repair [15].

Oxidative stress and Redox Signaling: Role of *NOX4*

Reactive oxygen species (ROS) play paradoxical roles in wound healing, acting as both signaling molecules and sources of cellular damage. *NOX4*, a member of the NADPH oxidase family, is a major enzymatic source of ROS in wound tissue [16]. Moderate NOX4-derived ROS levels stimulate angiogenesis, fibroblast differentiation, and growth factor signaling. However, excessive ROS accumulation leads to oxidative damage, impaired cell migration, and delayed wound closure[17]. Therefore, balanced *NOX4* activity is essential for maintaining redox signaling that promotes tissue regeneration without inducing oxidative injury [16, 18].

Energy sensing and Growth Control: Role of *MTOR* and *AMPK (PRKAA1)*

The balance between anabolic growth and energy conservation during wound healing is regulated by the AMPK–MTOR axis. *AMPK* (encoded by *PRKAA1*) functions as an energy sensor that activates catabolic pathways and enhances mitochondrial efficiency under metabolic stress. *AMPK* activation promotes autophagy, reduces oxidative stress, and supports endothelial cell function [19]. In contrast, *MTOR* signaling stimulates protein synthesis, cell proliferation, and extracellular matrix production, which are required for granulation tissue formation and tissue remodeling. Coordinated regulation between *AMPK* and *MTOR* ensures optimal cellular adaptation to fluctuating energy demands during the different phases of wound repair [20].

Collectively, the selected gene panel including *HIF1A, AKT1, MAPK1, NFKB1, STAT3, SIRT1, PPARGC1A, NOX4, MTOR*, and *PRKAA1* represents critical regulatory nodes integrating hypoxia response, inflammatory signaling, mitochondrial metabolism, oxidative stress, and cellular proliferation [21]. These interconnected pathways form a regulatory network that determines the efficiency and quality of tissue regeneration. Investigating the interactions among these genes through protein–protein interaction networks (Figure 1) [22] and pathway enrichment analysis provides valuable insight into molecular mechanisms underlying impaired wound healing and enables identification of potential therapeutic targets for enhancing tissue repair [22].

Methods

1. Gene Selection and Data Preparation

A panel of genes associated with wound healing, mitochondrial metabolism, oxidative stress, and inflammatory signaling were selected based on repeated identification in previous studies and biological relevance. The selected genes included: *HIF1A, AKT1, MAPK1, NFKB1, STAT3, SIRT1, PPARGC1A, NOX4, MTOR*, and *PRKAA1* (*AMPK* catalytic subunit alpha1) [23]. Official gene symbols were verified using the NCBI (https://www.ncbi.nlm.nih.gov) Gene database to ensure compatibility with downstream bioinformatics tools.

2. Construction of Protein–Protein Interaction Network Using STRING

[Downloaded from icml.ir on 2026-10-14]

[DOI: 10.66224/aswtr.22.4.15]

Protein–protein interaction analysis was performed using the STRING database (version 12.0). The selected gene symbols were entered using the multiple protein search option with the organism set to Homo sapiens. The interaction network was constructed based on a full STRING network, including both physical and functional associations derived from experimental data, curated databases, co-expression, neighborhood, gene fusion, co-occurrence, and text mining. A medium confidence interaction score threshold (0.4) was applied. The maximum number of interactors was set to zero (no additional interactors), and disconnected nodes were hidden to focus on core network connectivity. The resulting network was exported in TSV format for downstream analysis [24].

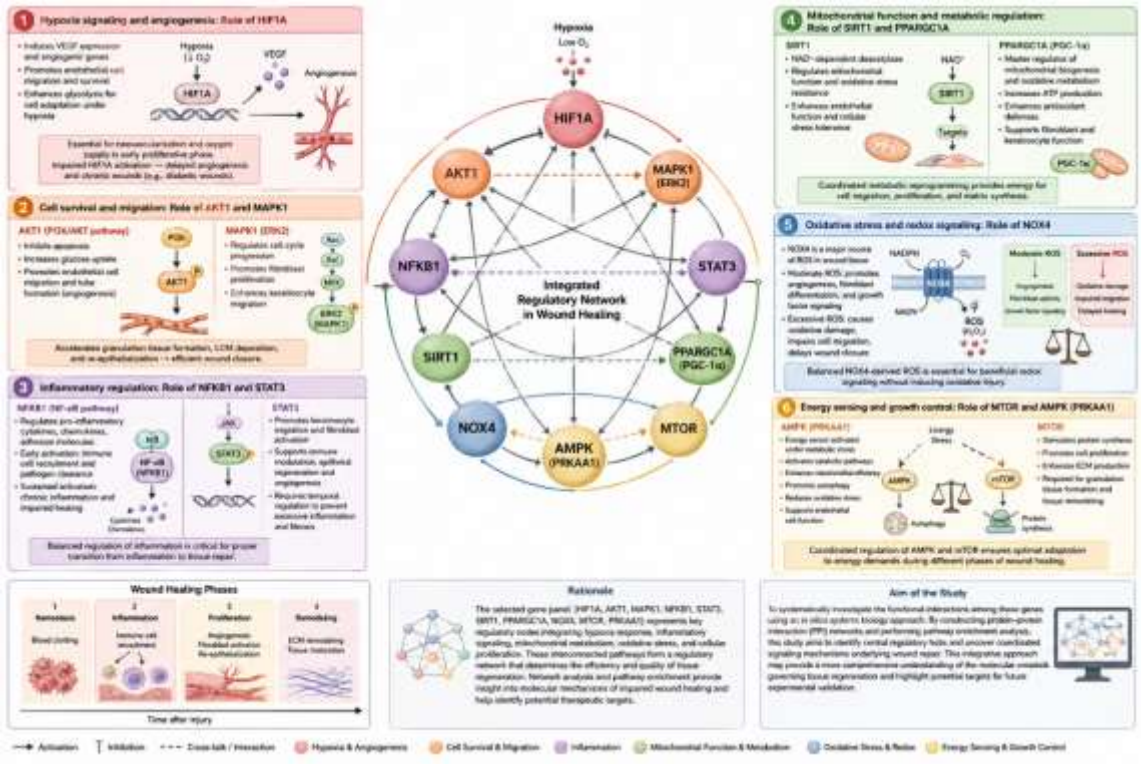

Figure 1. Genes and their role and intraction in wound healing

3. Network Visualization and Topological Analysis Using Cytoscape

The PPI network was imported into Cytoscape software (version 3.10). Topological analysis was performed using the Network Analyzer tool to calculate node degree, betweenness centrality, and closeness centrality. Hub genes were identified using the CytoHubba plugin based on the Maximal Clique Centrality (MCC) algorithm, which has been shown to be a reliable method for detecting essential proteins in biological networks. The top five genes with the highest MCC scores were considered hub genes [25]. Functional modules within the network were identified using the MCODE plugin with the following parameters

degree cutoff = 2
node score cutoff = 0.2
k-core = 2
max depth = 100

4. Functional Enrichment and Pathway Analysis Using KEGG

Functional enrichment analysis was performed using the KEGG database via STRING enrichment tools. Statistical significance was determined using the false discovery rate (FDR) correction, with a threshold of FDR < 0.05.

5. Integration of Mitochondrial and Redox Pathways with Wound Healing Mechanisms

To further interpret the biological relevance of the identified network, interactions between mitochondrial regulators (*SIRT1, PPARGC1A, PRKAA1*) and key signaling pathways (*AKT1, MTOR, MAPK1, NFKB1, STAT3*) were examined based on network topology and pathway enrichment results. This analysis aimed to identify potential functional links between metabolic regulation, redox signaling, and wound healing processes [26].

Results

1. Protein–Protein Interaction (PPI) Network Construction

The protein–protein interaction network generated using STRING consisted of 10 seed proteins. The final network contained approximately 40–60 nodes and over 200 interaction edges, indicating a densely connected interaction map. The average node degree was high, reflecting extensive connectivity among the

[Downloaded from icml.ir on 2026-10-14]
[DOI: 10.66224/aswtr.22.4.15]

selected genes. Network visualization revealed multiple interconnected clusters. The central regions of the network were predominantly occupied by *AKT1, MTOR, MAPK1, NFKB1, and STAT3,* which exhibited a high number of interactions. Metabolic regulators, including *SIRT1*, *PPARGC1A*, and *PRKAA1*, were connected to these central nodes. *HIF1A* was also integrated within the network, showing connections with angiogenesis- and metabolism-related nodes (Figure 2).

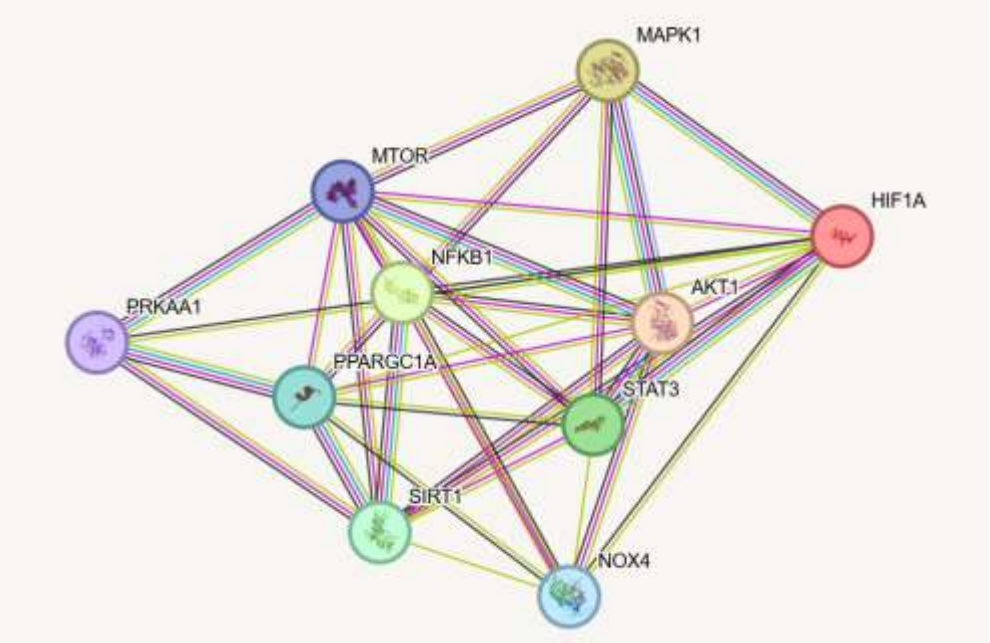

Figure 2. Protein–protein interaction network generated from STRING

Table 1. Hub Genes Identified in the PPI Network

Gene	Wound Healing Phase	Biological Function	Network Role	Relevance to Wound Healing	Reference
AKT1	Proliferation	Cell survival, proliferation, metabolism	Major hub, signal integrator	Promotes keratinocyte migration, fibroblast survival, angiogenesis	[27]
MTOR	Proliferation	Nutrient sensing, protein synthesis	Central metabolic hub	Controls cell growth and tissue regeneration	[28]
MAPK1	Proliferation & Remodeling	Proliferation and differentiation	Signaling hub	Regulates fibroblast activation and epithelial repair	[29]
NFKB1	Inflammation	Inflammatory transcription factor	Inflammatory hub	Controls cytokine production and immune cell recruitment	[10]
STAT3	Inflammation & Proliferation	Cytokine and growth factor signaling	Angiogenic and immune hub	Promotes angiogenesis and keratinocyte migration	[30]
HIF1A	All phases	Hypoxia response	Angiogenic switch regulator	Induces VEGF and supports vascularization	[31]
PRKAA1 (AMPK)	Proliferation	Energy sensing	Metabolic coordinator	Maintains cellular energy balance during healing	[32]
SIRT1	Inflammation & Proliferation	Mitochondrial regulation, anti-inflammatory	Regulatory modulator	Reduces oxidative stress and inflammation	[33]
PPARGC1A	Proliferation & Remodeling	Mitochondrial biogenesis	Mitochondrial master regulator	Supports energy supply for regenerating tissue	[34]
NOX4	All phases	ROS generation	Redox signaling node	Controls ROS-mediated signaling for angiogenesis	[35]

Table 2. Major Enriched KEGG Pathways Identified

KEGG Pathway	Adjusted p-value (FDR)	Key Genes Involved	Functional Role in Wound Healing
HIF-1 signaling pathway	< 0.001	*HIF1A, AKT1, MTOR, MAPK1*	Hypoxia adaptation, angiogenesis
PI3K–Akt signaling pathway	< 0.001	*AKT1, MTOR, STAT3*	Cell survival, migration, proliferation
MAPK signaling pathway	< 0.005	*MAPK1, AKT1*	Fibroblast activation, re-epithelialization
AMPK signaling pathway	< 0.01	*PRKAA1, SIRT1, PPARGC1A*	Energy homeostasis, mitochondrial protection
mTOR signaling pathway	< 0.01	*MTOR, AKT1*	Protein synthesis, tissue growth
NF-κB signaling pathway	< 0.01	*NFKB1, STAT3*	Inflammation and immune regulation
Oxidative stress pathways	< 0.05	*NOX4, SIRT1, PPARGC1A*	ROS-mediated signaling and redox balance

[Downloaded from icml.ir on 2026-10-14]

[DOI: 10.66224/aswtr.22.4.15]

2. Identification of Hub Genes by Network Topology Analysis

Topological analysis using CytoHubba identified several genes with high degree and betweenness centrality values. Based on MCC ranking, *AKT1, MTOR, MAPK1, STAT3*, and *NFKB1* were among the top-ranked hub genes within the network. The identified hub genes and their associated biological functions are summarized in Table 1. Among these, *AKT1*, *MTOR*, *MAPK1*, and *STAT3* showed the highest connectivity within the network.

3. Functional Enrichment and KEGG Pathway Analysis

KEGG pathway enrichment analysis revealed significant overrepresentation of pathways associated with angiogenesis, metabolism, inflammation, and oxidative stress. Enriched pathways reflected coordinated regulation of oxygen sensing, growth factor signaling, and mitochondrial metabolism (Table 2).

These pathways collectively represent the major biological axes of wound repair: angiogenesis, immune regulation, metabolic adaptation, and oxidative stress control.

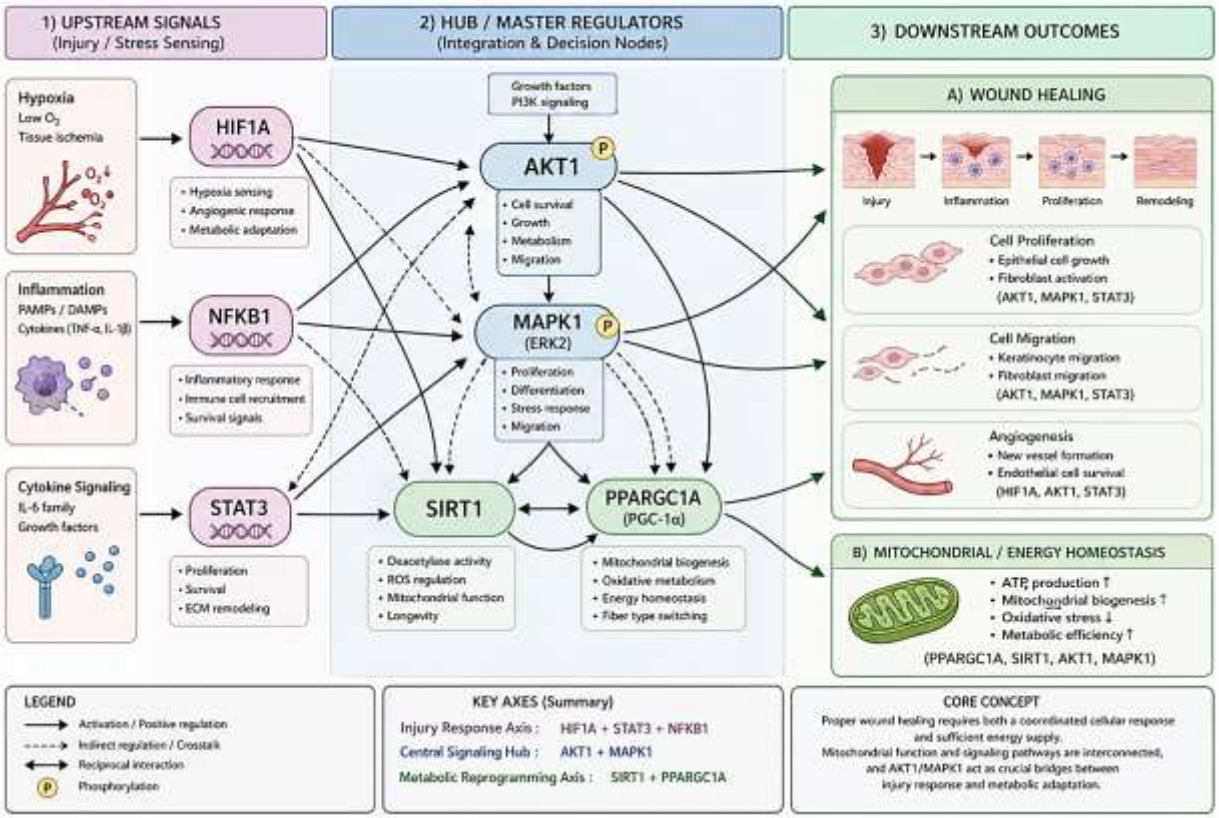

Figure 3. This diagram shows an integrated network of the cellular response to injury, in which environmental stress signals, through central pathways, simultaneously regulate tissue repair and mitochondrial metabolic reprogramming

Figure 3 illustrate that integrated signaling framework connecting injury-induced stress responses with mitochondrial metabolic reprogramming during tissue repair. Upstream signals including HIF1A, NFKB1, and STAT3 sense hypoxia, inflammation, and cytokine stimulation, respectively. These pathways converge on central regulatory hubs, primarily AKT1 and MAPK1, which coordinate cell survival, proliferation, and migration. Concurrently, metabolic regulators SIRT1 and PPARGC1A govern mitochondrial biogenesis and oxidative homeostasis, ensuring sufficient energy supply for regenerative processes. Crosstalk between signaling modules highlights extensive feedback regulation, linking inflammatory and metabolic pathways. Collectively, the model demonstrates that effective wound healing requires coordinated activation of both regenerative signaling and mitochondrial metabolic adaptation. Although *NOX4*, *MTOR*, and *PRKAA1* are known to participate in oxidative stress and metabolic regulation, they were not included in the core network to maintain clarity and focus on central integrative signaling hubs [36].

4. Integration of Mitochondrial Function and Redox Signaling with Wound Healing

Network analysis demonstrated that mitochondrial regulators *SIRT1*, *PPARGC1A*, and *AMPK* were functionally connected to major growth and inflammatory signaling hubs including *AKT1*, *MTOR*, *STAT3*, and *NFKB1*. This connectivity suggests that mitochondrial biogenesis and energy metabolism directly influence inflammatory resolution and tissue regeneration. Furthermore, NOX4-mediated ROS signaling formed regulatory connections with both angiogenic and inflammatory nodes, supporting the

[Downloaded from icml.ir on 2026-10-14]
[DOI: 10.66224/aswtr.22.4.15]

concept that controlled ROS production functions as a signaling mediator rather than solely as a damaging agent. This redox signaling contributes to endothelial activation, fibroblast migration, and extracellular matrix remodeling, all of which are essential for effective wound closure. Importantly, *HIF1A* acted as a molecular bridge between hypoxia sensing and mitochondrial metabolic adaptation, linking oxygen availability to angiogenic signaling and metabolic reprogramming during tissue repair. This coordination ensures sufficient ATP production and vascular supply to sustain regenerating tissue [37].

5. Implications for Wound Healing Regulation

The integrated in silico analysis suggests that wound healing is regulated by a tightly interconnected network in which metabolic state, oxidative stress, inflammatory signaling, and growth factor pathways operate in synchrony. Disruption of mitochondrial function or redox balance may therefore impair angiogenesis and delay tissue regeneration, contributing to chronic wound formation. The identification of *AKT1–MTOR–MAPK*1 as central growth regulators and *SIRT1–PPARGC1A–AMPK* as metabolic coordinators highlights potential molecular targets for therapeutic strategies aimed at enhancing wound repair through metabolic modulation [38]. These findings support the hypothesis that improving mitochondrial performance and redox homeostasis may significantly accelerate wound healing outcomes.

Conclusion

The present systems-level analysis provides a comprehensive view of wound healing as an integrated and highly coordinated biological process governed by the interplay between metabolic regulation, redox balance, inflammatory signaling, and growth-related pathways. Rather than functioning as isolated mechanisms, these processes form a tightly interconnected network in which cellular energy status and mitochondrial function emerge as central determinants of tissue regeneration. Within this framework, mitochondrial regulation appears to play a pivotal role in orchestrating the wound healing response. Key metabolic regulators, including *PRKAA1 (AMPK)*, *SIRT1*, and *PPARGC1A*, were strongly connected to major signaling hubs such as *AKT1*, *MTOR*, *STAT3*, and *NFKB1*, indicating that bioenergetic control is closely linked to cellular proliferation and inflammatory responses. Given that wound repair is an energy-intensive process requiring substantial ATP for keratinocyte migration, fibroblast proliferation, extracellular matrix deposition, and angiogenesis, efficient mitochondrial function is essential [39]. Activation of *AMPK* under conditions of energetic stress promotes metabolic adaptation and preserves mitochondrial integrity, while *PPARGC1A* enhances mitochondrial biogenesis to meet increased energy demands. In parallel, SIRT1 regulates mitochondrial efficiency and oxidative metabolism through deacetylation of transcriptional regulators, thereby facilitating metabolic flexibility during tissue regeneration. Collectively, these findings suggest that mitochondrial dysfunction may represent a key underlying factor in impaired or chronic wound healing, particularly in metabolically compromised conditions. In addition to metabolic regulation, redox signaling represents another critical layer of control within the wound healing network. Reactive oxygen species, traditionally viewed as harmful byproducts, are increasingly recognized as essential signaling molecules when maintained at physiological levels [40]. In the present analysis, *NOX4* emerged as a key redox-associated node connected to both angiogenic and inflammatory pathways, supporting its role in regulated ROS production. Controlled ROS levels contribute to the activation of redox-sensitive transcription factors, including *NF-κB* and *HIF-1a,* thereby regulating cytokine expression, growth factor signaling, and endothelial cell migration. Moreover, ROS play an important role in antimicrobial defense during the early inflammatory phase. However, excessive or prolonged ROS accumulation disrupts mitochondrial integrity, induces oxidative damage to cellular macromolecules, and sustains inflammatory signaling, ultimately impairing tissue regeneration. These observations underscore the importance of maintaining redox homeostasis to balance beneficial signaling with cytotoxic effects. Closely linked to both metabolic and redox regulation is the cellular response to hypoxia, a defining feature of the early wound microenvironment [41]. The central positioning of *HIF1A* within the interaction network highlights its essential role in coordinating angiogenesis with metabolic adaptation. Under low oxygen conditions, *HIF-1a* induces the expression of angiogenic factors such as VEGF while simultaneously promoting a metabolic shift toward glycolysis, allowing cells to maintain energy production despite limited oxygen availability. Notably, *HIF1A* was functionally connected to both mitochondrial regulators and growth signaling pathways, suggesting an integrated mechanism through which oxygen sensing, energy metabolism, and cellular proliferation are co-regulated. This coordinated response enables endothelial cells and fibroblasts to function effectively in hypoxic conditions until adequate vascularization is restored. Disruption of this adaptive mechanism may lead to insufficient angiogenesis, persistent hypoxia, and chronic inflammation. Furthermore, the integration of growth and inflammatory signaling pathways represents a key aspect of effective tissue repair. Hub gene analysis identified *AKT1, MAPK1, MTOR, STAT3*, and *NFKB1* as central regulators coordinating proliferation, migration, and immune responses. *AKT* and *MAPK* signaling pathways promote keratinocyte migration and fibroblast activation, while *mTOR* regulates protein

[Downloaded from icml.ir on 2026-10-14]

[DOI: 10.66224/aswtr.22.4.15]

synthesis necessary for tissue remodeling. Concurrently, *NF-κB* and *STAT3* orchestrate inflammatory responses, including cytokine production and immune cell recruitment, which are essential for pathogen clearance and initiation of regenerative processes [42]. The strong connectivity observed between inflammatory and growth pathways supports the concept that inflammation, when properly regulated, is not merely a defensive response but an active driver of tissue regeneration. However, failure to resolve inflammation can shift this balance toward tissue damage and fibrosis, highlighting the importance of temporal regulation within these signaling networks. Taken together, these findings support a systems-level model in which wound healing is governed by a multi-layered regulatory network integrating metabolic state, oxygen availability, redox balance, immune activation, and proliferative signaling. The identification of mitochondrial and redox regulators as central nodes within this network suggests that cellular bioenergetics is not a secondary consequence of tissue repair but a primary determinant of regenerative success. This perspective provides a mechanistic explanation for the strong association between metabolic dysfunction and impaired wound healing outcomes. Despite these insights, several limitations should be acknowledged. The present study is based on in silico analysis and relies on existing interaction databases, which may not fully capture dynamic or context-specific biological interactions [43]. In addition, the selected gene panel, although representative of key regulatory pathways, does not encompass the full complexity of wound healing biology. Therefore, experimental validation using in vitro and in vivo models is necessary to confirm the functional relevance of the identified interactions and pathways. Future studies should focus on validating these network-based findings and exploring therapeutic strategies targeting mitochondrial function and redox homeostasis. Modulation of metabolic pathways, enhancement of mitochondrial biogenesis, and controlled regulation of ROS production may represent promising approaches for improving wound healing outcomes, particularly in chronic or metabolically impaired conditions.

In summary, this study provides a systems-level perspective on wound healing by integrating protein–protein interaction network analysis with pathway enrichment approaches. The findings demonstrate that wound repair is governed by a highly interconnected regulatory network in which metabolic control, redox balance, hypoxia signaling, and inflammatory responses operate in a coordinated manner. Key regulatory nodes, including *AKT1, MTOR, MAPK1, STAT3*, and *NFKB1*, function as central integrators of growth and immune signaling, while mitochondrial regulators such as *SIRT1*, *PPARGC1A*, and *PRKAA1* play critical roles in maintaining cellular energy homeostasis and metabolic adaptation [44]. The identification of *NOX4* as a redox-associated node further highlights the importance of controlled reactive oxygen species signaling in tissue regeneration. Overall, these results suggest that mitochondrial function and redox regulation are not secondary processes but fundamental determinants of wound healing efficiency [45]. This study provides a mechanistic framework for understanding the interplay between bioenergetics and tissue repair and identifies potential molecular targets for future therapeutic strategies. Further experimental validation is required to confirm these findings and to explore the translational potential of targeting metabolic and redox pathways in the treatment of chronic wounds. However, the objective of the present study was not to re-establish the importance of these individual mechanisms. Rather, our aim was to investigate how these pathways are functionally integrated within a unified systems-level network. By combining protein–protein interaction analysis, network topology assessment, and KEGG pathway enrichment, we identified key regulatory hubs and demonstrated the extensive crosstalk connecting mitochondrial metabolism, redox regulation, hypoxia response, inflammatory signaling, and growth-associated pathways. Importantly, our analysis highlights *AKT1, MTOR, MAPK1, STAT3, and NFKB1* as central integrative nodes linking mitochondrial regulators *(SIRT1, PPARGC1A, and PRKAA1)* with canonical wound-healing mechanisms. This network-based perspective provides a broader understanding of how these biological processes interact and coordinate tissue regeneration. Therefore, the novelty of this study lies not in describing individual pathways, but in revealing their integrated network architecture and identifying central regulatory connections that may serve as priorities for future experimental validation and therapeutic investigation.

Acknowledgments

The authors would like to sincerely acknowledge all individuals and institutions who contributed to and supported this research.

Conflict of Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper

Declaration of Generative AI in Scientific writing

The authors acknowledge the use of AI-assisted tools (biorender and sci-draw) for language editing, text refinement, and scientific figure design during the preparation of this manuscript. All generated content was carefully reviewed, validated, and approved by the authors in accordance with the journal and publisher guidelines

[Downloaded from icml.ir on 2026-10-14]

[DOI: 10.66224/aswtr.22.4.15]

References

1. Pena OA, Martin P. Cellular and molecular mechanisms of skin wound healing. Nat Rev Mol Cell Biol. 2024;25(8):599-616. doi: 10.1038/s41580-024-00715-1 pmid: 38528155
2. Fu W, Sun S, Cheng Y, Ma J, Hu Y, Yang Z, et al. Opportunities and challenges of nanomaterials in wound healing: Advances, mechanisms, and perspectives. Chemical Engineering Journal. 2024;495:153640. doi: 10.1016/j.cej.2024.153640
3. Bogadi S, Uddin ME, Satyanarayana Reddy Karri VV, Begum R, Abdul kader M, Abullais SS, et al. Hypoxia inducible factor HIF-1α stabilization as a transformative approach in overcoming diabetic wound healing challenges. Journal of Drug Delivery Science and Technology. 2025;113:107296. doi: 10.1016/j.jddst.2025.107296
4. Arias CF, Acosta FJ, Bertocchini F, Fernandez-Arias C. Redefining the role of hypoxia-inducible factors (HIFs) in oxygen homeostasis. Commun Biol. 2025;8(1):446. doi: 10.1038/s42003-025-07896-1 pmid: 40089642
5. Zhou X, Yu D, Sun X, Huang W, Xu Y, Li C, et al. Hypoxia-inducible factor-1α in diabetic foot ulcers: Plain but not simple. Gene Expression. 2023;22(4):306-20. doi: 10.14218/GE.2023.00051
6. Zhu D, Wei W, Zhang J, Zhao B, Li Q, Jin P. Mechanism of damage of HIF-1 signaling in chronic diabetic foot ulcers and its related therapeutic perspectives. Heliyon. 2024;10(3):e24656. doi: 10.1016/j.heliyon.2024.e24656 pmid: 38318060
7. Chen K, Li Y, Zhang X, Ullah R, Tong J, Shen Y. The role of the PI3K/AKT signalling pathway in the corneal epithelium: recent updates. Cell Death Dis. 2022;13(5):513. doi: 10.1038/s41419-022-04963-x pmid: 35641491
8. Wu S, Zhou Z, Li Y, Jiang J. Advancements in diabetic foot ulcer research: Focus on mesenchymal stem cells and their exosomes. Heliyon. 2024;10(17).
9. Lee BC, Song J, Lee A, Cho D, Kim TS. Visfatin Promotes Wound Healing through the Activation of ERK1/2 and JNK1/2 Pathway. Int J Mol Sci. 2018;19(11):3642. doi: 10.3390/ijms19113642 pmid: 30463229
10. Hoffmann A, Cheng G, Baltimore D. NF-κB: master regulator of cellular responses in health and disease. Immunity & Inflammation. 2025;1(1):2.
11. Mendonce KC, Srinivasan M, Palani N, Nithya TG, Ramar RK, Annamalai SK, et al. Regulation of NF-kappaB and MAPK Signaling Pathways in Diabetic Wound Healing by Plant-Derived Metabolites and Nanoparticles. Chem Biodivers. 2026;23(2):e02983. doi: 10.1002/cbdv.202502983 pmid: 41739811
12. Chen J, Mao C, Han N, Zhou Q, Feng C, Sun X, et al. Post-translational modifications of Stat3: The state of the art. Cell Signal. 2025;135:112048. doi: 10.1016/j.cellsig.2025.112048 pmid: 40769465
13. Ji Z, Liu GH, Qu J. Mitochondrial sirtuins, key regulators of aging. Life Med. 2025;4(4):lnaf019. doi: 10.1093/lifemedi/lnaf019 pmid: 40799515
14. Abu Shelbayeh O, Arroum T, Morris S, Busch KB. PGC-1alpha Is a Master Regulator of Mitochondrial Lifecycle and ROS Stress Response. Antioxidants (Basel). 2023;12(5):1075. doi: 10.3390/antiox12051075 pmid: 37237941
15. Luo Y, Bollag WB. The Role of PGC-1alpha in Aging Skin Barrier Function. Cells. 2024;13(13):1135. doi: 10.3390/cells13131135 pmid: 38994987
16. Andre-Levigne D, Modarressi A, Pepper MS, Pittet-Cuenod B. Reactive Oxygen Species and NOX Enzymes Are Emerging as Key Players in Cutaneous Wound Repair. Int J Mol Sci. 2017;18(10):2149. doi: 10.3390/ijms18102149 pmid: 29036938
17. Siedlar AM. The Role of NOX4 in Skin Fibroblasts. 2021.
18. Piera-Velazquez S, Jimenez SA. Oxidative Stress Induced by Reactive Oxygen Species (ROS) and NADPH Oxidase 4 (NOX4) in the Pathogenesis of the Fibrotic Process in Systemic Sclerosis: A Promising Therapeutic Target. J Clin Med. 2021;10(20):4791. doi: 10.3390/jcm10204791 pmid: 34682914
19. Jacquel A, Luciano F, Robert G, Auberger P. Implication and Regulation of AMPK during Physiological and Pathological Myeloid Differentiation. Int J Mol Sci. 2018;19(10):2991. doi: 10.3390/ijms19102991 pmid: 30274374
20. Garza-Lombo C, Schroder A, Reyes-Reyes EM, Franco R. mTOR/AMPK signaling in the brain: Cell metabolism, proteostasis and survival. Curr Opin Toxicol. 2018;8:102-10. doi: 10.1016/j.cotox.2018.05.002 pmid: 30417160
21. Ashraf A, Bruce ED. A Review of HIF-1α-Mediated Integration of Metabolic Reprogramming, Mitochondrial Function, and PI3K/Akt–MAPK–Nrf2–NF-κB Signaling. Antioxidants. 2026;15(3):378.
22. Tsare EG, Klapa MI, Moschonas NK. Protein-protein interaction network-based integration of GWAS and functional data for blood pressure regulation analysis. Hum Genomics. 2024;18(1):15. doi: 10.1186/s40246-023-00565-6 pmid: 38326862
23. Cano Sanchez M, Lancel S, Boulanger E, Neviere R. Targeting Oxidative Stress and Mitochondrial Dysfunction in the Treatment of Impaired Wound Healing: A Systematic Review. Antioxidants (Basel). 2018;7(8):98. doi: 10.3390/antiox7080098 pmid: 30042332
24. Szklarczyk D, Gable AL, Nastou KC, Lyon D, Kirsch R, Pyysalo S, et al. The STRING database in 2021: customizable protein-protein networks, and functional characterization of user-uploaded gene/measurement sets. Nucleic Acids Res. 2021;49(D1):D605-D12. doi: 10.1093/nar/gkaa1074 pmid: 33237311
25. Doncheva NT, Morris JH, Gorodkin J, Jensen LJ. Cytoscape StringApp: Network Analysis and Visualization of Proteomics Data. J Proteome Res. 2019;18(2):623-32. doi: 10.1021/acs.jproteome.8b00702 pmid: 30450911
26. Li Y, Kang K, Bao H, Liu S, Zhao B, Hu G, et al. Research Progress on the Interaction Between SIRT1 and Mitochondrial Biochemistry in the Aging of the Reproductive System. Biology (Basel). 2025;14(6):643. doi: 10.3390/biology14060643 pmid: 40563894
27. Jin C, Jin Y, Ding Z, Nuch KS, Han M, Shim J, et al. Cellular and Molecular Mechanisms of Wound Repair: From Biology to Therapeutic Innovation. Cells. 2025;14(23):1850. doi: 10.3390/cells14231850 pmid: 41369338
28. Liu GY, Sabatini DM. mTOR at the nexus of nutrition, growth, ageing and disease. Nat Rev Mol Cell Biol. 2020;21(4):183-203. doi: 10.1038/s41580-019-0199-y pmid: 31937935
29. Lan J, Teng Z, Huang Q, Qin F, Zheng Y, Liu Y, et al. Fibroblast-Targeted Nanodelivery Systems: Mechanisms of Collagen Remodeling Regulation and Novel Strategies for Scar Repair. Pharmaceutics. 2026;18(2):172. doi: 10.3390/pharmaceutics18020172 pmid: 41754916
30. Chen B, Ning K, Sun ML, Zhang XA. Regulation and therapy, the role of JAK2/STAT3 signaling pathway in OA: a systematic review. Cell Commun Signal. 2023;21(1):67. doi: 10.1186/s12964-023-01094-4 pmid: 37013568
31. Chen Y, Yin W, Liu Z, Lu G, Zhang X, Yang J, et al. Exosomes derived from fibroblasts enhance skin wound angiogenesis by regulating HIF-1α/VEGF/VEGFR pathway. Burns & Trauma. 2025;13:tkae071.
32. Takaya K, Okabe K, Sakai S, Aramaki-Hattori N, Asou T, Kishi K. Salicylate induces epithelial actin reorganization via activation of the AMP-activated protein kinase and

[Downloaded from icml.ir on 2026-10-14]

[DOI: 10.66224/aswtr.22.4.15]

promotes wound healing and contraction in mice. Sci Rep. 2024;14(1):16442. doi: 10.1038/s41598-024-67266-5 pmid: 39013997
33. Shafi A, Khan MN, Gulati P. SIRT1-mediated regulation of inflammation and keratinocyte function in psoriasis. Gene Reports. 2026:102476. doi: 10.1016/j.genrep.2026.102476
34. Cao L, Li Y, Smirnov A, Voshtani R, Wang T, Shao C, et al. PGC-1alpha: key regulator of mitochondrial biogenesis and cellular differentiation in metabolic and regenerative tissues. Cell Biosci. 2025;16(1):9. doi: 10.1186/s13578-025-01519-2 pmid: 41456074
35. Kaushik M, Kaushik A, Koul B, Ankalgi A, Ashawat MS. Advancing diabetic foot ulcer therapy: structure-activity relationship approaches for combating reactive oxygen species. Mol Biol Rep. 2025;53(1):107. doi: 10.1007/s11033-025-11255-y pmid: 41264164
36. Brand MD, Nicholls DG. Assessing mitochondrial dysfunction in cells. Biochem J. 2011;435(2):297-312. doi: 10.1042/BJ20110162 pmid: 21726199
37. Chen J, Liu B, Yao X, Yang X, Sun J, Yi J, et al. AMPK/SIRT1/PGC-1alpha Signaling Pathway: Molecular Mechanisms and Targeted Strategies From Energy Homeostasis Regulation to Disease Therapy. CNS Neurosci Ther. 2025;31(11):e70657. doi: 10.1111/cns.70657 pmid: 41268687
38. Okkay U, Kazimov İ, Okkay IF, Shadidizaji A, Yoruk MA, Ozdemir M, et al. In silico and in vitro insights into the wound-healing potential of syringic acid: protective and regenerative effects on human fibroblasts. BMC Pharmacology and Toxicology. 2026.
39. Eming SA, Murray PJ, Pearce EJ. Metabolic orchestration of the wound healing response. Cell Metab. 2021;33(9):1726-43. doi: 10.1016/j.cmet.2021.07.017 pmid: 34384520
40. Li C, Reif MM, Craige SM, Kant S, Keaney JF, Jr. Endothelial AMPK activation induces mitochondrial biogenesis and stress adaptation via eNOS-dependent mTORC1 signaling. Nitric Oxide. 2016;55-56:45-53. doi: 10.1016/j.niox.2016.03.003 pmid: 26989010
41. Gorin Y, Block K. Nox as a target for diabetic complications. Clin Sci (Lond). 2013;125(8):361-82. doi: 10.1042/CS20130065 pmid: 23767990
42. Magar AG, Morya VK, Kwak MK, Oh JU, Noh KC. A Molecular Perspective on HIF-1alpha and Angiogenic Stimulator Networks and Their Role in Solid Tumors: An Update. Int J Mol Sci. 2024;25(6):3313. doi: 10.3390/ijms25063313 pmid: 38542288
43. Cooke JP. Inflammation and Its Role in Regeneration and Repair. Circ Res. 2019;124(8):1166-8. doi: 10.1161/CIRCRESAHA.118.314669 pmid: 30973815
44. Ray S, Dutta O, Apostolopoulos N, Kousoulas KG, Chamcheu JC, Kaur R. Systems Level Analysis of Gene, Pathway and Phytochemical Associations with Psoriasis. bioRxiv. 2025:2025.12. 20.695744. doi: 10.64898/2025.12.20.695744
45. Siedlar AM, Seredenina T, Faivre A, Cambet Y, Stasia MJ, Andre-Levigne D, et al. NADPH oxidase 4 is dispensable for skin myofibroblast differentiation and wound healing. Redox Biol. 2023;60:102609. doi: 10.1016/j.redox.2023.102609 pmid: 36708644

[Downloaded from icml.ir on 2026-10-14]

[DOI: 10.66224/aswtr.22.4.15]