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TGα2β-modified adipose-derived stem cell exosomes regulate collagen synthesis in fibroblasts and enhance chronic ulcerative wound repair through the ILK/AKT signaling pathway

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Abstract: **Background**: Chronic ulcerative wounds, particularly diabetic foot ulcers, present a significant clinical challenge owing to their complex pathophysiology and limited response to conventional therapies. Although exosomes derived from adipose-derived mesenchymal stem cells (ADSC-exos) have shown promise in promoting tissue repair, the specific mechanisms underlying their effects, particularly their involvement in the integrin-linked kinase (ILK)/AKT signaling pathway, remain poorly elucidated. **Objectives**: This study aimed to elucidate whether exosomes from ADSCs modified with ITGα2β (ITGα2β-ADSC-exos) enhance chronic ulcerative wound healing by modulating fibroblast collagen synthesis via the ILK/AKT signaling axis. **Methods**: ITGα2β-ADSC-exos were characterized using transmission electron microscopy, nanoparticle tracking analysis and Western blotting. Their effects on ILK/AKT pathway activation, migration and collagen production in fibroblasts were assessed *in-vitro*. For *in-vivo* validation, a diabetic rat model with full-thickness skin wounds was established and randomly allocated into five groups: Control (PBS), unmodified exosomes, ITGα2β-Modified exosomes (ITG-Exo), ITG-Exo co-administered with the ILK inhibitor QLT0267 and ITG-Exo co-administered with the AKT inhibitor MK-2206. Wound healing progression, histological changes, microvessel density (via CD31 immunohistochemistry) and the expression of pathway-related proteins (p-AKT, COL1A1, COL3A1) were evaluated. **Results**: ITGα2β modification enhanced the levels of characteristic markers and the yield of ADSC-exos. These modified exosomes potently activated the ILK/AKT pathway in fibroblasts, subsequently promoting fibroblast proliferation, migration and the synthesis of type I and III collagen. In diabetic rats, ITGα2β-ADSC-exos significantly accelerated wound closure, improved granulation tissue formation and increased microvessel density. Crucially, these therapeutic effects were entirely abrogated by co-administration of either the ILK inhibitor QLT0267 or the AKT inhibitor MK-2206, which instead exacerbated tissue vacuolization and inflammation. Western blot analysis confirmed that the pro-healing effects correlated with upregulation of p-AKT and collagen proteins. **Conclusion**: ITGα2β-modified ADSC-exos regulate collagen synthesis in fibroblasts and facilitate chronic ulcerative wound healing via the ILK/AKT signaling pathway.

Keywords: ADSCs; Chronic ulcerative wounds; Exosomes; ILK/AKT

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INTRODUCTION

The rising prevalence of diabetes in China has led to an increase in chronic ulcerative wounds, particularly diabetic foot ulcers (DFUs). It is projected that the number of adults with diabetes will reach 439 million (Deng *et al*., 2024; McDermott *et al*., 2023). Hyperglycemia-induced vascular damage is a major contributor to severe diabetic complications such as DFUs. Reports indicate that 10-25% of diabetic patients develop DFUs, imposing a substantial burden on both patients and healthcare systems (Taki *et al*., 2022). Conventional management of chronic ulcerative wounds includes debridement, pressure offloading, infection control and stringent glycemic regulation. However, these approaches often yield suboptimal outcomes (Wu *et al*., 2023). Notably, DFUs-related amputations account for 84% of all lower-limb amputations, leading to severe psychological distress and elevated mortality, thereby posing a significant societal burden.

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Chronic ulcerative wounds are defined as those that fail to progress through the normal healing trajectory within an expected timeframe despite standard care. The pathogenesis of such wounds is multifactorial and their treatment is often protracted. Under physiological conditions, cutaneous wound healing progresses through four overlapping phases: hemostasis, inflammation, proliferation and remodeling (Holl *et al*., 2021; Singer, 2022). However, due to internal and external factors, some wounds are arrested at a particular phase or fail to complete repair, ultimately becoming chronic. Current clinical strategies primarily involve wound dressings and surgical debridement to remove necrotic tissue and control localized infection, yet they often fall short of promoting adequate revascularization, growth factor activity, or modulation of cellular apoptosis.

Exosomes have emerged as promising candidates for accelerating wound repair by promoting re-epithelialization, angiogenesis, inflammation regulation and ECM remodeling. Traditionally, cell-based therapies involve the local injection of mesenchymal stem cells

(MSCs) into or around the wound bed. However, the therapeutic efficacy of such approaches is limited by poor cell retention and an unfavorable microenvironment for stem cells in diabetic wounds. As exogenous stem cells have a short lifespan under diabetic conditions, exosomes—which carry bioactive molecules, including nucleic acids, microRNAs and proteins—have gained attention for their role in mediating both local and systemic intercellular communication. Notably, MSC-exos exhibit reparative capacities comparable to, or even greater than, those of the parent cells (Yu *et al*., 2024; Chen *et al*., 2026). Moreover, exosome-mediated cell-free therapy offers advantages over cell transplantation, including better stability, easier storage and lower immunogenicity. For instance, fibroblast-derived exosomes have been shown to enhance angiogenesis and facilitate skin wound healing (Allegra *et al*., 2022; Talbott *et al*., 2022). Despite these advances, rapid clearance of exosomes from the wound site remains a challenge. The integration of exosomes with biomaterials to prolong their residence time without compromising bioactivity represents an active area of investigation.

Integrin-linked kinase (ILK) plays a crucial role in wound contraction and re-epithelialization. Studies in rat models have demonstrated that ILK participates in skin wound healing and its inhibition—along with that of the PI3K/AKT pathway—impedes wound contraction and re-epithelialization, thereby stalling the repair process (Alobaid *et al*., 2024; Xin *et al*., 2026; Bildyug, 2024). *In-vitro* evidence further indicates that ILK and PI3K/AKT inhibitors suppress fibroblast contractility, suggesting that ILK also influences angiogenesis and wound healing (Chu *et al*., 2024). Nevertheless, the potential role of ILK in wound vascularization remains underexplored. Elevated ILK levels correlate positively with microvessel density in chronic wound tissues and fibroblasts overexpressing ILK promote the migration of HUVECs. Fibroblasts are fundamental to skin homeostasis and repair and their functional transition is crucial for successful healing. Emerging data suggest that the ECM may regulate this process, as transmembrane integrin receptors mediate cell-ECM interactions (Jiang *et al*., 2023; Song *et al*., 2023).

Against this background, the present study aims to elucidate whether ITGα2β-modified exosomes derived from ADSC-exos promote chronic ulcerative wound repair by modulating collagen synthesis in fibroblasts through the ILK/AKT signaling pathway.

MATERIALS AND METHODS

Cells source

The 3T3 fibroblast cell line (purchased from Guangzhou Stem Cell Co., Ltd., China; Catalog No. GZ-MSC-3T3) was provided by Guangzhou Stem Cell Co., Ltd. Cells were cultured in DMEM/F12 medium supplemented with 10% exosome-free fetal bovine serum and maintained in a humidified incubator at 37°C with 5% CO_2. Cells exhibited robust proliferation and differentiation potential under these conditions. Cells at passages 3–6 were used for all experiments.

Exosomes isolation

Exosomes were isolated from the supernatant of ADSC cultures (second passage) using the ExoQuick-TC kit (System Biosciences, USA; Catalog No. EXOTC50A-1) following the manufacturer's instructions. Briefly, the collected supernatant was centrifuged at 300 × g for 10 minutes at 4°C to remove cellular debris, then centrifuged at 2,000 × g for 30 minutes. The clarified supernatant was mixed with ExoQuick-TC precipitation reagent at the recommended ratio and incubated overnight at 4°C. The mixture was then centrifuged at 15,000 × g for 30 minutes to pellet the exosomes, which were subsequently resuspended in phosphate-buffered saline (PBS). Protein concentration was determined using the bicinchoninic acid (BCA) assay (Beyotime Biotechnology, China; Catalog No. P0012).

Exosome characterization

Isolated exosomes were characterized by transmission electron microscopy (TEM) for morphological assessment, nanoparticle tracking analysis (NanoSight LM10, Malvern Panalytical, UK) for size distribution and Western blotting for detection of exosomal markers CD63 and CD81 (antibodies from Abcam, USA; CD63: ab134045; CD81: ab109201).

Functional assessment of exosomes

To evaluate the functional effects of ITGα2β modification, 3T3 fibroblasts were divided into three groups: PBS control, unmodified exosome-treated and ITGα2β-modified exosome-treated groups. Exosomes were added at a concentration of 100 μg/mL and co-cultured with fibroblasts for 48 hours. Cells were then harvested for subsequent analyses. The method for ITGα2β modification of ADSC-exos was adopted from a previously described protocol (Feng *et al*., 2025), with minor modifications.

Chronic ulcerative wound model

Male Sprague-Dawley rats (weight 210 ± 25 g, approximately 10 weeks old) were obtained from the SPF Animal Experiment Center of The Third People's Hospital of Gansu Province. Animals were housed under controlled temperature (25°C), humidity and a 12-hour light/dark cycle. Diabetes was induced by intraperitoneal injection of streptozotocin (STZ) (Sigma-Aldrich, USA; Catalog No. S0130) after an 18-hour fast, following the established protocol described in a previous study (Rai *et al*., 2022). Blood glucose levels were monitored on days 3, 7, 10 and 14 post-injection using a glucometer (Accu-Chek Active, Roche, Germany). Diabetic status was maintained for three weeks prior to wounding. All procedures adhered to the Consensus Author Guidelines on Animal Ethics and Welfare (Percie du Sert *et al*., 2020) and were approved by the Institutional Gansu Provincial Hospital of Traditional

Chinese Medicine Ethics Committee (Approval No. GS20250895).

Experimental grouping and interventions

Rats with successful model establishment were randomly divided into five groups (n=8 per group) using a random number table: Blank control group, Unmodified exosomes group, ITGα2β-Modified exosomes (ITG-Exo) group, ITG-Exo+QLT group and ITG-Exo+AKTi group. The Blank Control group received a local wound injection of 100 μL PBS solution. The Unmodified Exosomes group was injected with 100 μg unmodified ADSC-derived exosomes suspended in PBS. The ITG-Exo group received 100 μg ITGα2β-modified ADSC-exos in PBS. The ITG-Exo+QLT group was co-administered with 100 μg ITGα2β-modified exosomes and the ILK inhibitor QLT0267 (dissolved in PBS at a dosage of 5 mg/kg; Selleck Chemicals, USA; Catalog No. S1092). The ITG-Exo+AKTi group received a combination of the same dose of ITGα2β-modified exosomes (100 μg) and the AKT inhibitor MK-2206 (dissolved in PBS at a concentration of 10 mg/kg; Selleck Chemicals, USA; Catalog No. S1078) . The dosages of QLT0267 and MK-2206 were selected based on a previous *in-vivo* study). (Pérez-Núñez *et al*., 2023). Interventions were administered locally once daily for seven consecutive days, starting on the first day post-wounding. All assessments were performed by investigators blinded to group assignments.

Histopathological analysis

Wound tissues were collected on day 20, fixed with 4% paraformaldehyde, paraffin-embedded and sectioned at 5 μm thickness. Sections were stained with hematoxylin and eosin (H and E) and examined under a light microscope (Olympus BX53, Japan) to evaluate granulation tissue formation, collagen arrangement, inflammatory infiltration and re-epithelialization.

Immunohistochemistry and microvessel density analysis

Paraffin sections were deparaffinized, rehydrated and subjected to antigen retrieval using sodium citrate buffer (pH 6.0) under high-pressure heat. Endogenous peroxidase activity was blocked with 3% H_2O_2 for 30 minutes. After blocking with 10% goat serum, sections were incubated overnight at 4°C with anti-CD31 antibody (1:200; Abcam, USA; Catalog No. ab28364). HRP-conjugated secondary antibody (1:500; Zhongshan Golden Bridge Biotechnology, China; Catalog No. PV-6001) was applied for 1 hour at room temperature, followed by DAB development and hematoxylin counterstaining. Microvessel density was quantified by counting CD31-positive vessels with clear lumens in five randomly selected fields per slide (200× magnification).

Western blotting

Cells treated under different conditions or wound tissues collected on day 20 were lysed using RIPA lysis buffer (Beyotime Biotechnology, China; Catalog No. P0013B) to extract total protein. Protein concentration was quantified using the BCA assay. Equal amounts of protein samples (typically 30 μg) were separated by SDS-PAGE electrophoresis and subsequently transferred onto PVDF membranes (Millipore, USA; Catalog No. IPVH00010) . After transfer, the membranes were blocked at room temperature for 1 hour with TBST containing 5% skim milk. The membranes were then incubated overnight at 4°C with the following specific primary antibodies (all diluted at 1:1000): phosphorylated AKT (p-AKT), total AKT (t-AKT; Cell Signaling Technology, USA; Catalog No. 4060), total AKT (t-AKT; Cell Signaling Technology, USA; Catalog No. 4691), collagen type I (COL1A1; Abcam, USA; Catalog No. ab138492) and collagen type III (COL3A1; Abcam, USA; Catalog No. ab7778). GAPDH (1:2000; Proteintech, China; Catalog No. 60004-1-Ig) was used as the loading control. After washing, membranes were incubated with HRP-conjugated secondary antibodies (1:5000; Zhongshan Golden Bridge Biotechnology, China; Catalog No. ZB-2301) for 1 hour and developed using an ECL substrate (Millipore, USA; Catalog No. WBKLS0500). Band intensities were analyzed with ImageJ software (National Institutes of Health, USA, version 1.53).

Migration and proliferation assay

The cells were inoculated and a 200 μL pipette tip was used to make 5 parallel scratches in each well, with the initial scratch width measured. The scratch width was measured using an optical microscope (Leica® DMi8, Germany) and ImageJ software to simulate the isolation zone experiment. The scratch assay was performed according to the method described in a previous study (Xia *et al*., 2025). For proliferation assays, a Transwell system was used to assess the effects on cell proliferation.

Statistical analysis

Data were analyzed using SPSS 19.0 software (IBM, USA). Measurement data were presented as mean ± standard deviation and one-way ANOVA was used for analysis of group means. Correlation analysis was performed using the Pearson correlation coefficient. A P value < 0.05 indicated statistical significance.

RESULTS

ITGα2β modified ADSC-exosomes

Approximately 3 mL of ADSC-exosomes were obtained from 50 mL of ADSC supernatant, with a final protein concentration of 1617.2 μg/mL. Transmission electron microscopy (TEM) revealed typical cup-shaped vesicular structures (Fig. 1A). NanoSight LM10 showed an exosome concentration of 4.19×10^{11} particles/mL, of which 44.4% fell within the expected size range of 30–150 nm (Fig. 1B). Western blotting confirmed the enrichment of exosomal markers CD63 and CD81 in ADSC-exos compared with the parental ADSCs (Fig. 1C).

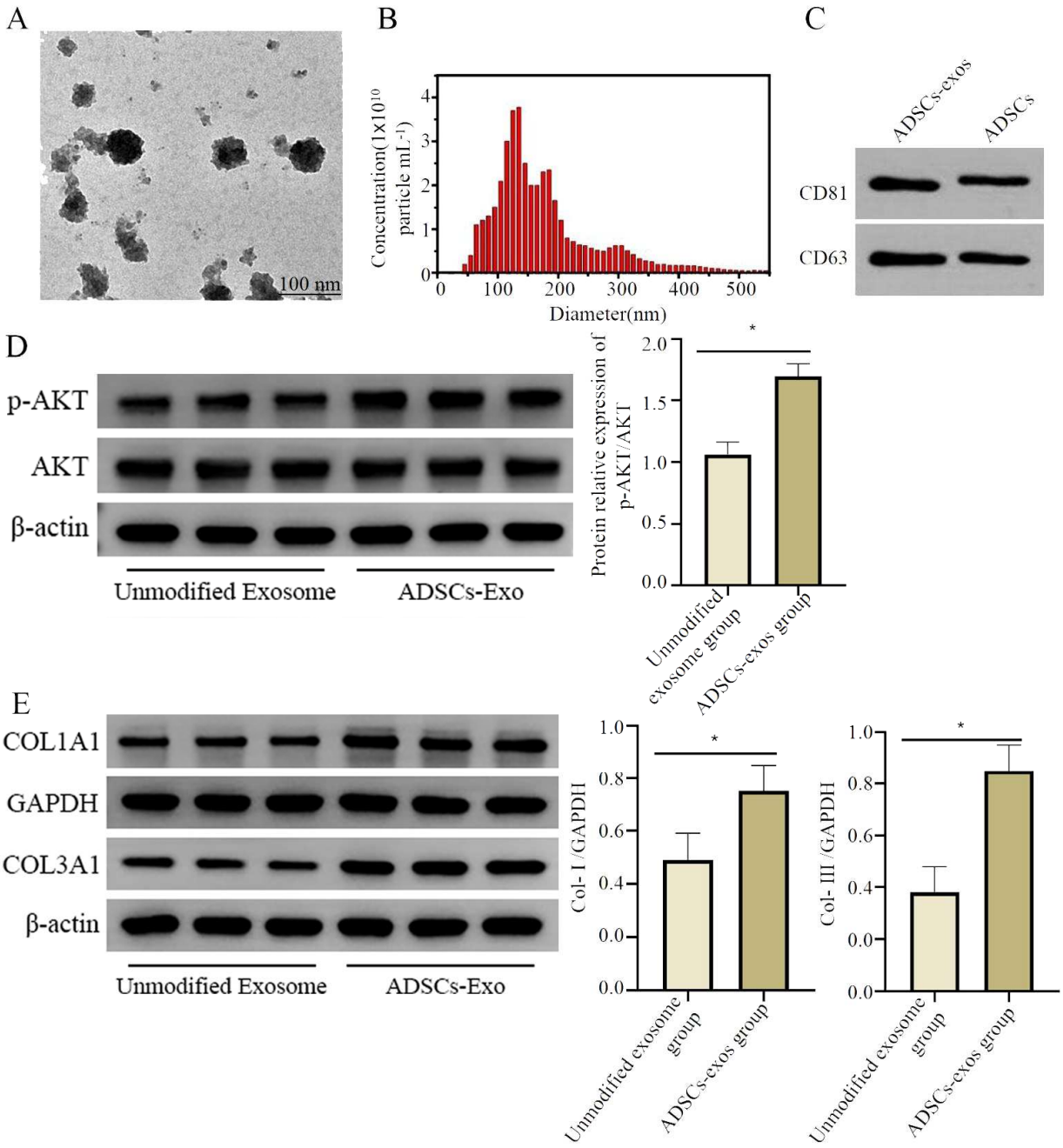

Fig. 1: Characterization and functional analysis of ITGα2β-modified ADSC exosomes
(A) Transmission electron microscopy image of ITGα2β-modified ADSC-derived exosomes, showing the typical cup-shaped morphology (scale bar: 100 nm); (B) Transmission electron microscopy images of ITGα2β-modified ADSC exosomes reveal the typical cup-shaped morphology (scale bar: 100 nm); (C) Western blotting analysis was performed to detect the expression of the marker proteins CD63 and CD81 in ADSC cells and their exosomes; (D) Western blot analysis revealed that, compared to unmodified exosomes, treatment with ITGα2β-modified ADSC exosomes significantly elevated the protein levels of p-AKT in 3T3 fibroblasts; (E) Western blottingdemonstrated that ITGα2β-modified ADSC exosomes upregulated the expression of both collagen I and collagen III in 3T3 cells. * $P < 0.05$.

Treatment of 3T3 fibroblasts with ITGα2β-modified ADSC-exosomes significantly increased the p-AKT/AKT ratio compared with unmodified exosomes (Fig. 1D). Concurrently, expression levels of COL1A1 and COL3A1 were markedly upregulated in cells treated with ITGα2β-modified exosomes (Fig. 1E). Together, these data indicate that ITGα2β modification enhances the ability of ADSC-exosomes to activate the downstream ILK/AKT signaling pathway and promote collagen synthesis.

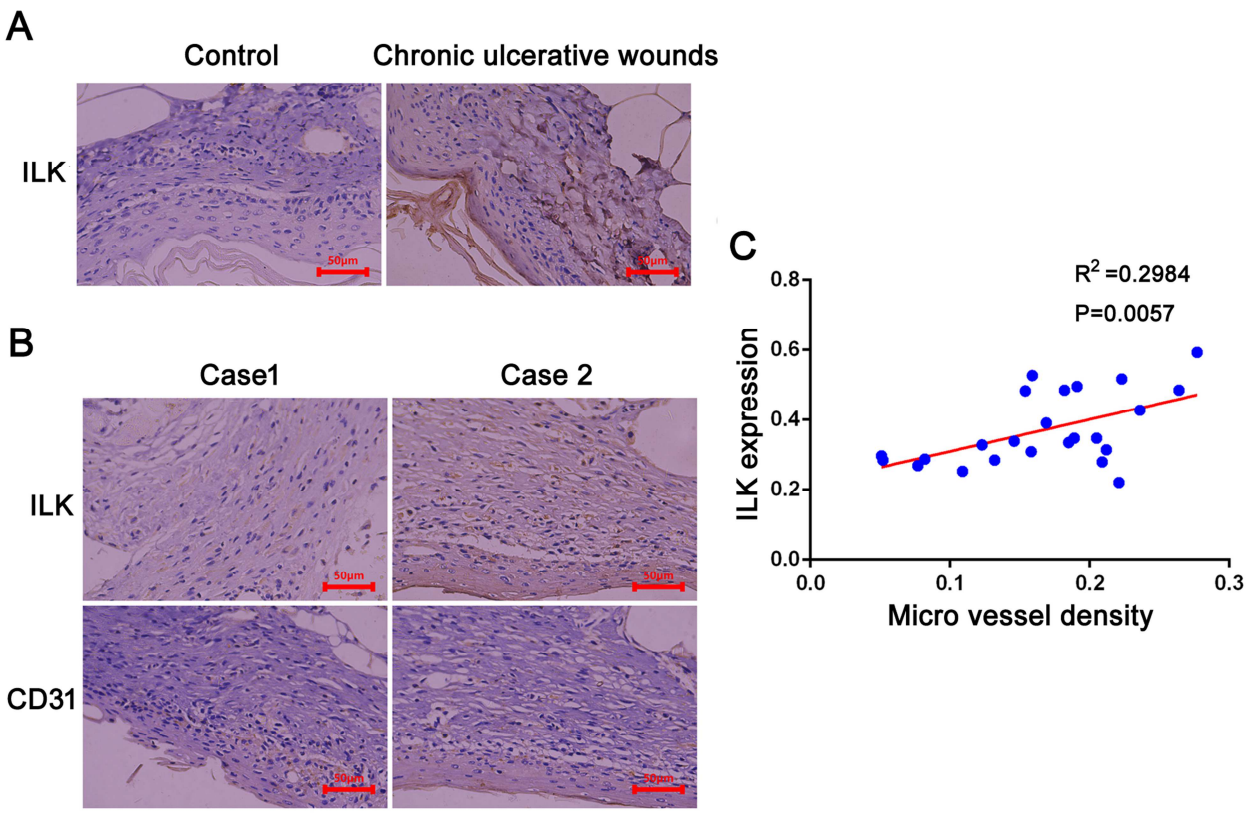

Fig. 2: Increased ILK expression is positively correlated with vascularization in chronic wound tissue (×400). (A) Representative images of immunohistochemical staining for ILK in chronic wound tissue and adjacent normal skin (×400); (B) A bar chart presenting the semi-quantitative analysis of immunohistochemical staining for ILK and CD31 expression; (C) A linear regression analysis reveals that ILK expression levels are significantly positively correlated with microvascular density. * P < 0.05.

Increased expression of ILK is positively correlated with vascularization in chronic wound tissue

To identify the potential role of ILK in angiogenesis and wound healing, immunohistochemical staining was performed on chronic wound tissues and adjacent normal skin. ILK expression was markedly elevated in chronic wound tissues compared to normal controls (Fig. 2A). Notably, high ILK immunoreactivity was also observed in regions of increased vascular density. Statistical analysis revealed a significant positive correlation between ILK expression and microvessel density (r=0.6859, $P < 0.001$; Figs. 2B and 2C), suggesting that ILK may promote vascularization during chronic wound healing.

Overexpression of ILK promotes angiogenesis

To investigate the functional role of ILK in angiogenesis, ILK-overexpressing fibroblasts (ILK-OV) and their corresponding negative controls (NC) were established (Fig. 3A). Conditioned medium (CM) collected from ILK-OV and ILK-NC fibroblasts was applied to 3T3 fibroblasts. Cell viability assays revealed a significant increase in 3T3 cell proliferation following treatment with ILK-OV CM compared to ILK-NC CM (Fig. 3B). Flow cytometry indicated a reduction in apoptotic cells upon treatment with ILK-OV fibroblasts (Fig. 3C). Furthermore, wound healing experiments demonstrated that 3T3 cell migration was accelerated following CM treatment with CM (Fig. 3D), which was supported by the results of Transwell experiment confirming the enhanced migration of 3T3 cells (Fig. 3E). Taken together, these observations indicate that ILK-upregulated fibroblasts effectively mediate interactions with 3T3 cells, thereby promoting angiogenesis.

ITGα2β-modified ADSCs exosomes mediate chronic ulcerative wound repair through ILK/AKT signaling

The ILK inhibitor QLT0267 and the AKT inhibitor MK-2206 delayed wound healing. To investigate the roles of ILK and AKT in promoting wound repair, full-thickness wounds were created on the depilated backs of 24 mice. From day 10 onward, the wound healing rate in the ITG-Exo group was significantly higher than that in the Exo group, while the ITG-Exo+QLT and ITG-Exo+AKTi groups exhibited significantly lower healing rates at all time points compared to the ITG-Exo group (Fig. 4A). Histologically, the ITG-Exo+QLT and ITG-Exo+AKTi groups displayed severe porous and vacuolated fibrous tissue accompanied by inflammatory cell infiltration, whereas the ITG-Exo group showed improved wound recovery and marked dermal hyperplasia (Fig. 4B).

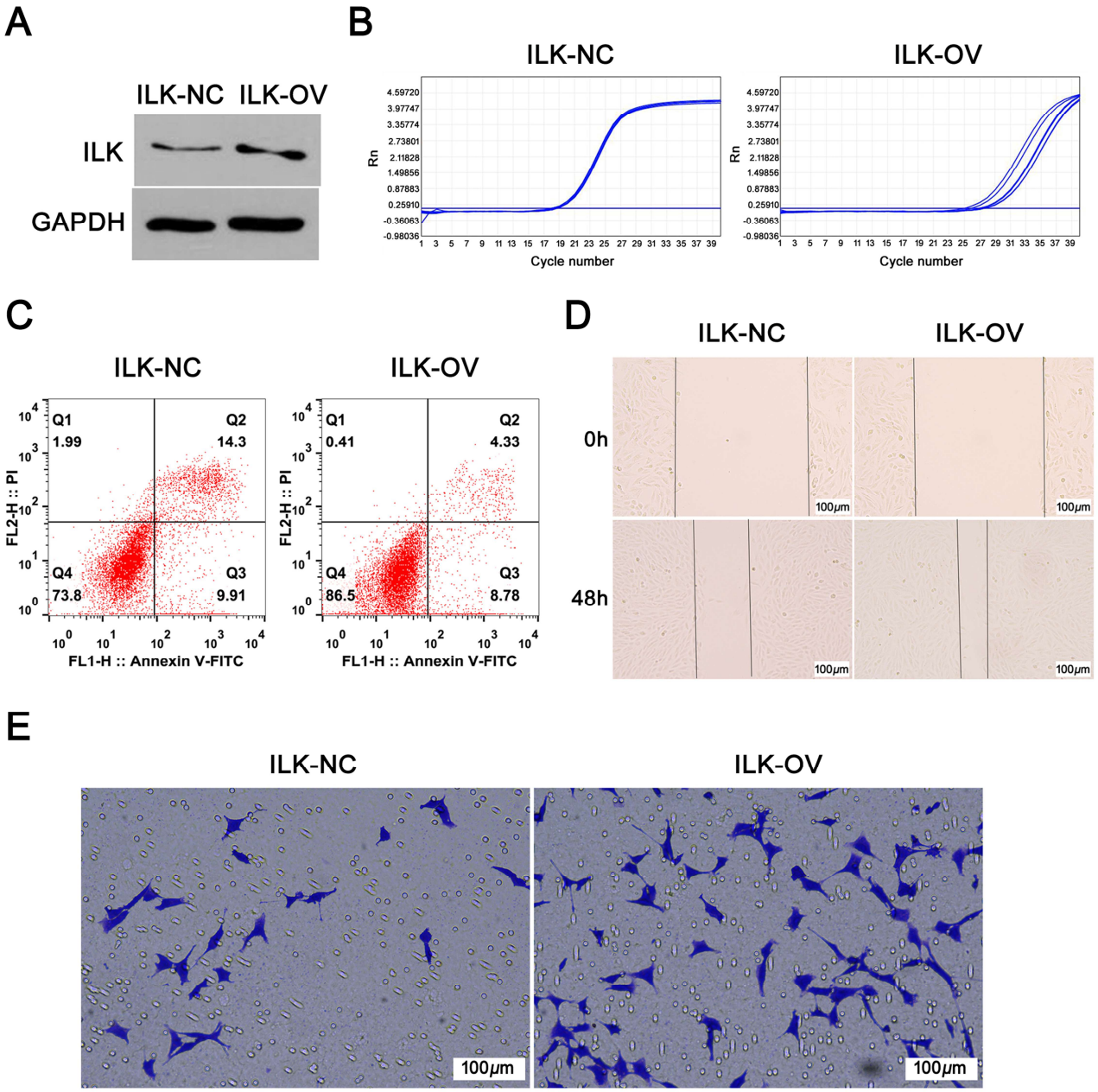

Fig. 3: ILK-upregulated fibroblasts promote angiogenesis.
(A) Western blot quantification of ILK protein levels in fibroblasts transfected with lentivirus overexpressing ILK (ILK-OV) or negative control (NC); (B) CCK-8 assay showed that the conditioned medium (CM) from ILK-overexpressing fibroblasts significantly promoted the proliferation of 3T3 cells; (C) Flow cytometry analysis indicated that treatment with ILK-OV conditioned medium (CM) reduced the apoptosis rate in 3T3 cells; (D) The scratch assay demonstrated that ILK-OV conditioned medium (CM) significantly accelerated the migration of 3T3 cells; (E) The Transwell migration assay confirmed that ILK-OV conditioned medium (CM) enhanced the migratory capacity of 3T3 cells, indicating that fibroblasts with upregulated ILK expression can effectively promote the migration of endothelial-like cells through paracrine mechanisms. * $P < 0.05$.

IHC staining for CD31 indicated that microvessel density in the ITG-Exo group was significantly increased compared to the blank control and Exo groups, whereas the ITG-Exo+QLT and ITG-Exo+AKTi groups showed significantly reduced density compared to the ITG-Exo group (Fig. 4C). On day 20, the levels of ADSC-exos, ILK expression and microvessel density were evaluated. Correlation analysis revealed a negative correlation between ADSC-exos and ILK expression, a positive correlation between ILK expression and microvessel density and a negative correlation between ADSC-exos and microvessel density (Fig. 4D).

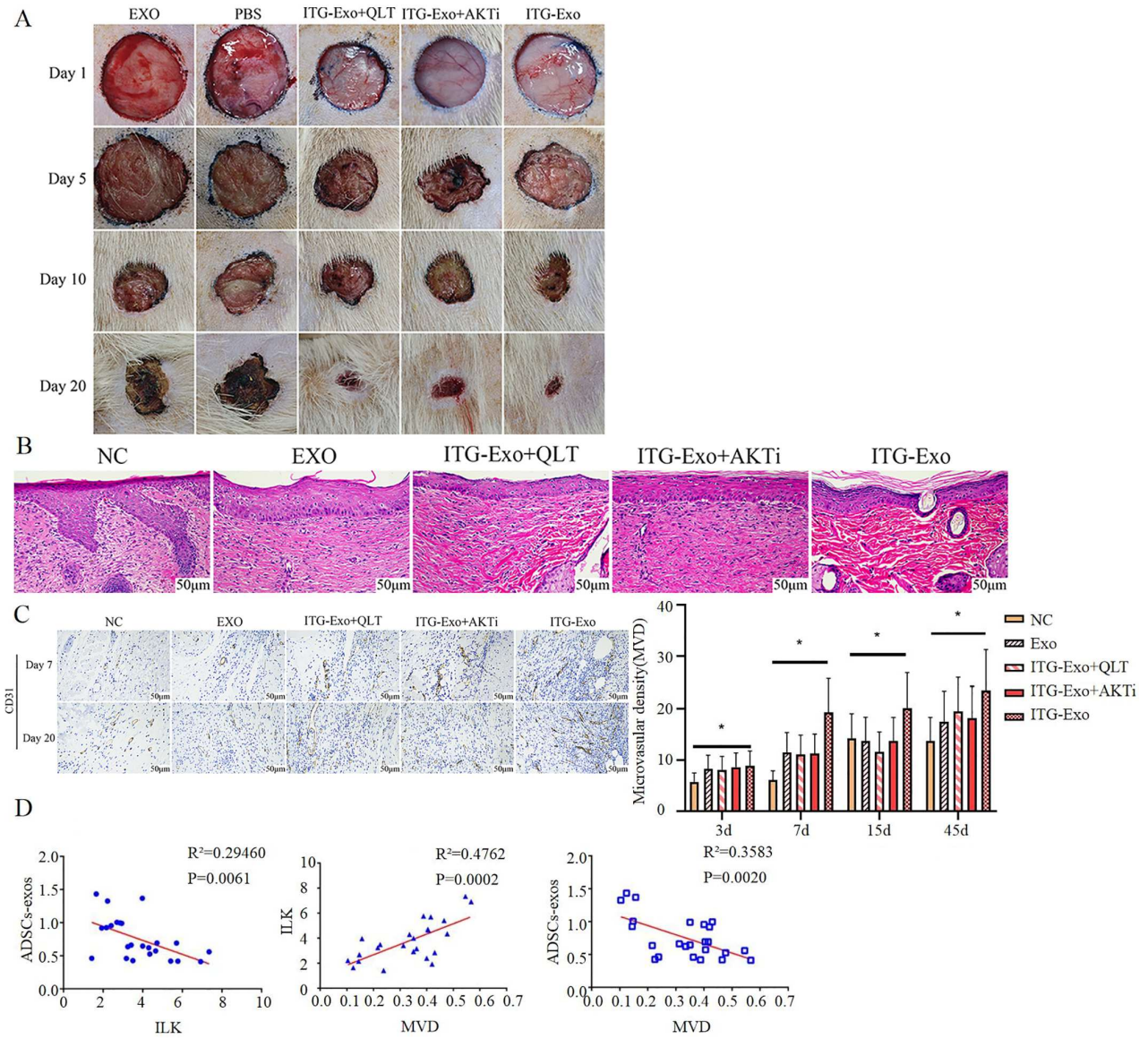

Fig. 4: ITGα2β-modified ADSCs exosomes mediate chronic ulcerative wound repair through ILK/AKT signaling. (A) Representative wound images of rats in each group on days 1, 5, 10, and 20 post-modeling; (B) H and E-stained sections of wound tissue on day 20 (scale bar: 200 μm). The ITG-Exo group exhibited favorable granulation tissue formation and re-epithelialization, whereas the ITG-Exo+QLT and ITG-Exo+AKTi groups showed delayed repair; (C) CD31 immunohistochemical staining (scale bar: 100 μm) and quantitative statistics of microvessel density. Angiogenesis was significantly enhanced in the ITG-Exo group; (D) Pearson correlation analysis revealed that the enrichment level of ADSC-exos was negatively correlated with ILK expression and microvessel density, while ILK expression was positively correlated with microvessel density. * $P < 0.05$.

These results suggest that during wound repair, changes in ILK expression are closely associated with the extent of angiogenesis, while the level of exosome presence may also be related to this pathophysiological process.

ITGα2β-modified exosomes regulate collagen synthesis in fibroblasts via the ILK/AKT pathway

Western blot analysis was performed to evaluate the regulatory effects. The results demonstrated that, compared with the blank control and Exo groups, the expression levels of p-AKT, COL1A1 and COL3A1 were significantly upregulated in the ITG-Exo group. In contrast, both the ITG-Exo+QLT and ITG-Exo+AKTi groups exhibited markedly reduced p-AKT levels and significantly suppressed collagen expression. These findings further confirm that ITGα2β-modified exosomes regulate collagen synthesis through the ILK/AKT signaling axis (Figs. 5A and 5B).

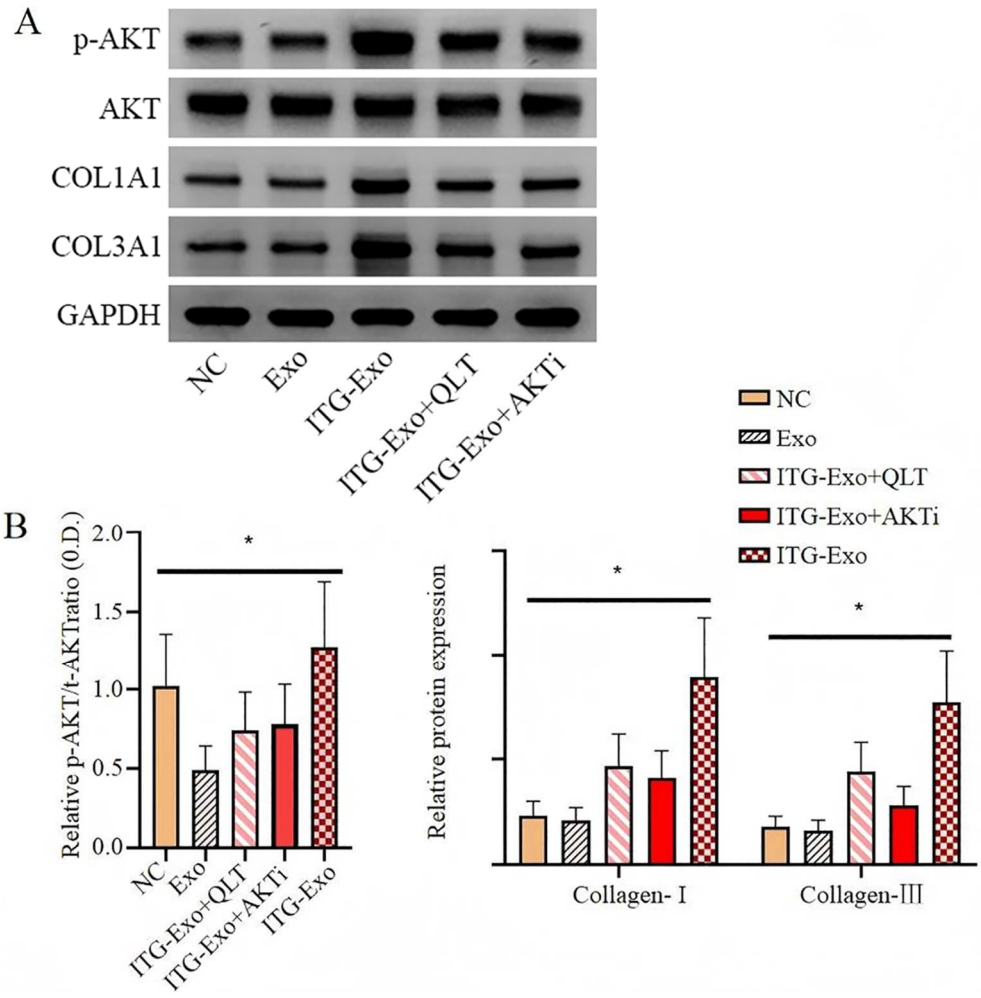

Fig. 5: ITGα2β-modified exosomes regulate collagen synthesis in fibroblasts via the ILK/AKT pathway.
(A) Western blot representative band images showing the expression of key proteins in wound tissues from each group; (B) A bar chart presenting the semi-quantitative analysis of band grayscale values in figure A. The results show that the expression levels of p-AKT, COL1A1 and COL3A1 in the ITG-Exo group were significantly higher than those in the Exo group. However, co-administration of QLT0267 or MK-2206 reversed the upregulation of these proteins induced by ITGα2β-exos. * $P < 0.05$.

DISCUSSION

Chronic ulcerative wounds represent a complex and challenging clinical condition, often associated with poor outcomes, high rates of disability and significant mortality. At present, the precise molecular and cellular mechanisms underlying the pathophysiology of these wounds remain incompletely understood. In patients with chronic ulcerative wounds, healing is frequently stalled during a prolonged inflammatory phase, driven by elevated levels of pro-inflammatory mediators and diminished expression of key growth factors (Zhou *et al*., 2023; Mu *et al*., 2022; Wang *et al*., 2025a). Therefore, this study aimed to elucidate the mechanistic basis of impaired wound repair by identifying the fundamental drivers of healing dysregulation. The insights gained from this work may contribute to a clearer understanding of the etiology of chronic ulcerative wounds and inform the development of more effective therapeutic strategies.

Therapeutic strategies involving fibroblasts and their derivatives hold promise for reducing amputation risk and alleviating the social and medical burden associated with chronic wound management. In the present study, ITGα2β-ADSC-exosomes were generated. Notably, ITGα2β-modified ADSC-exosomes were shown to retain potent biological activity and to significantly enhance diabetic wound healing. The potential of stem cell-based therapies and biotechnological approaches in wound repair has garnered increasing attention in recent years (Liu *et al*., 2023). Among various stem cell types, ADSCs exhibit distinct therapeutic profiles characterized by their ability to promote neovascularization, stimulate collagen synthesis and exert immunomodulatory effects, thereby facilitating

wound repair in both human and animal models (Nazbar *et al*., 2022). Fibroblasts similarly contribute to wound healing by supporting re-epithelialization, promoting angiogenesis and uniquely enhancing hair follicle regeneration. Upon injury, fibroblasts migrate to the wound site and undergo functional switching; although the precise regulatory mechanisms remain incompletely understood, emerging evidence suggests the involvement of several signaling pathways (Wang *et al*., 2025b; Yu *et al*., 2025; Yi *et al*., 2026).

Although ITGα2β-modified ADSC-derived exosomes demonstrate considerable therapeutic promise, several challenges hinder their clinical translation. Currently, the production process lacks standardized protocols, resulting in batch-to-batch variability that compromises reproducibility. In addition, exosome stability during storage and transport, as well as their behavior within the complex wound microenvironment, including targeting efficiency and retention, requires systematic evaluation. Long-term safety profiles, including potential immune responses and other biosafety considerations, also warrant thorough investigation. To facilitate the clinical advancement of this approach, future efforts should focus on establishing robust quality-control systems, optimizing scalable production methods and conducting comprehensive pharmacological and toxicological studies.

CONCLUSION

ILK binds to integrins and IPP complexes to influence various cell activities. The ILK/AKT axis exerts its biological effects by mediating several signaling cascades, notably the PI3K/Akt signaling pathway (Hu *et al*., 2021). Previous studies have shown that ILK deficiency impairs wound healing, a process closely linked to ILK/PI3K/AKT signaling. Moreover, ILK is recognized for its roles in enhancing cell migration and adhesion and facilitating tissue repair in contexts such as liver injury (Martin *et al*., 2022; Campillo *et al*., 2022; Rajesh *et al*., 2022; Psaraki *et al*., 2022). In the present study, ITGα2β-modified ADSC-exosomes were observed to activate the downstream ILK/AKT pathway in fibroblasts, thereby promoting fibroblast recruitment and angiogenesis, which are critical determinants of successful wound healing. Furthermore, treatment with ITGα2β-exosomes significantly upregulated phosphorylated AKT (p-AKT) and increased the expression of collagen types I and III in fibroblasts. This pro-collagen synthesis effect was abolished upon co-administration of either the ILK inhibitor QLT0267 or the AKT inhibitor MK-2206, indicating that the induction of collagen synthesis by ITGα2β-exosomes is strictly dependent on ILK/AKT pathway activity. Collectively, integrating functional assays, inhibitor-based intervention studies and correlation analyses, it is concluded that ITGα2β-modified ADSC-exosomes enhance chronic ulcerative wound repair primarily through activation of the ILK/AKT signaling pathway, which in turn modulates collagen synthesis and functional behavior of fibroblasts.

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Authors' contributions
Qin Qi: Conceptualized the study, designed the experimental protocol, supervised data interpretation and manuscript writing and assumed overall responsibility for the research direction and content. Established animal models, performed *in-vivo* experiments and conducted data collection and statistical analysis; Lifang Lu: Conducted cell culture, exosome extraction and characterization, Western blot and immunohistochemical experiments and participated in experimental procedures, data organization, figure preparation and initial draft writing; Xueyan Nan: Assisted with experimental material preparation, literature review and manuscript formatting and proofreading. All authors participated in reviewing and revising the manuscript and approved the final version for publication.

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Data availability statement
The datasets generated and/or analyzed during the current study are available from the corresponding author on reasonable request.

Ethical approval
This study has been approved by the Ethics Committee of The Third People's Hospital of Gansu Province (Approval No. GS20250895). This study was performed in adherence with the ARRIVE guidelines. See supplementary file for the ARRIVE checklist.

Conflict of interest
The authors declare no conflict of interest.

Supplementary data
https://pjps.pk/uploads/2026/08/SUP1787211554.pdf

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