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Supporting Information

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Polydopamine Decorated Microneedles with Fe-MSC-Derived Nanovesicles Encapsulation for Wound Healing

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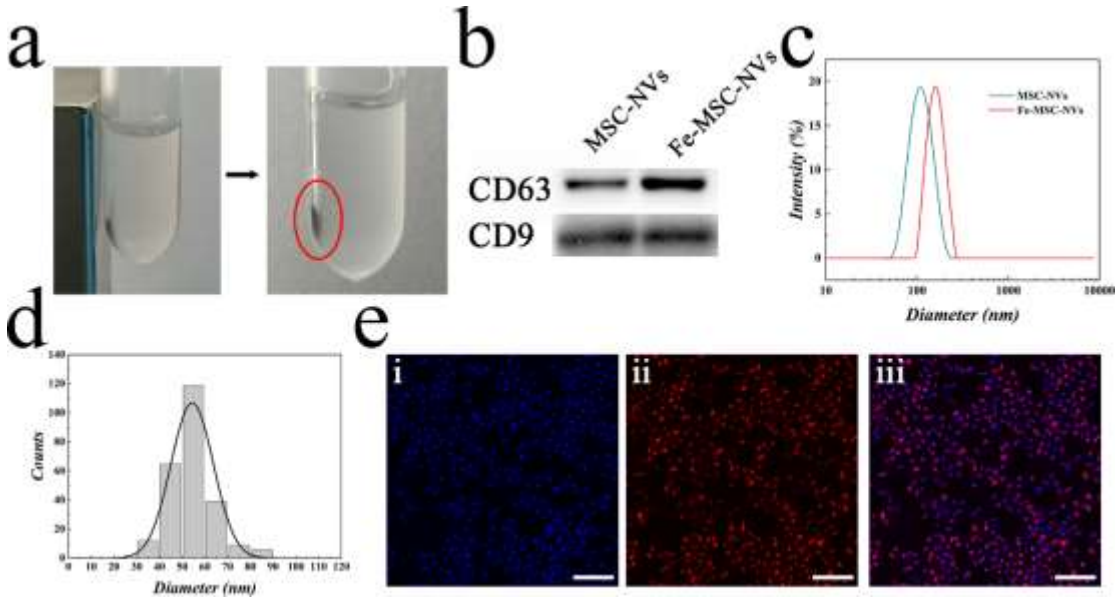

Figure S 1 (a) Magnetic attraction of Fe-MSCs. (b) The exosomal markers (CD9 and CD63) in MSC-NVs and Fe-MSC-NVs were analyzed by Western blot. (c) The particle size of MSC-NVs and Fe-MSC-NVs. (d) Corresponding size distribution histogram of PDA NPs derived from TEM images. (e) Evaluation of cellular uptake of DiD labeled Fe-MSC-NVs in HUVEC: (i) DAPI, (ii) DiD labeled Fe-MSC-NVs, (iii) Merge. Scale bars: 200 μm.

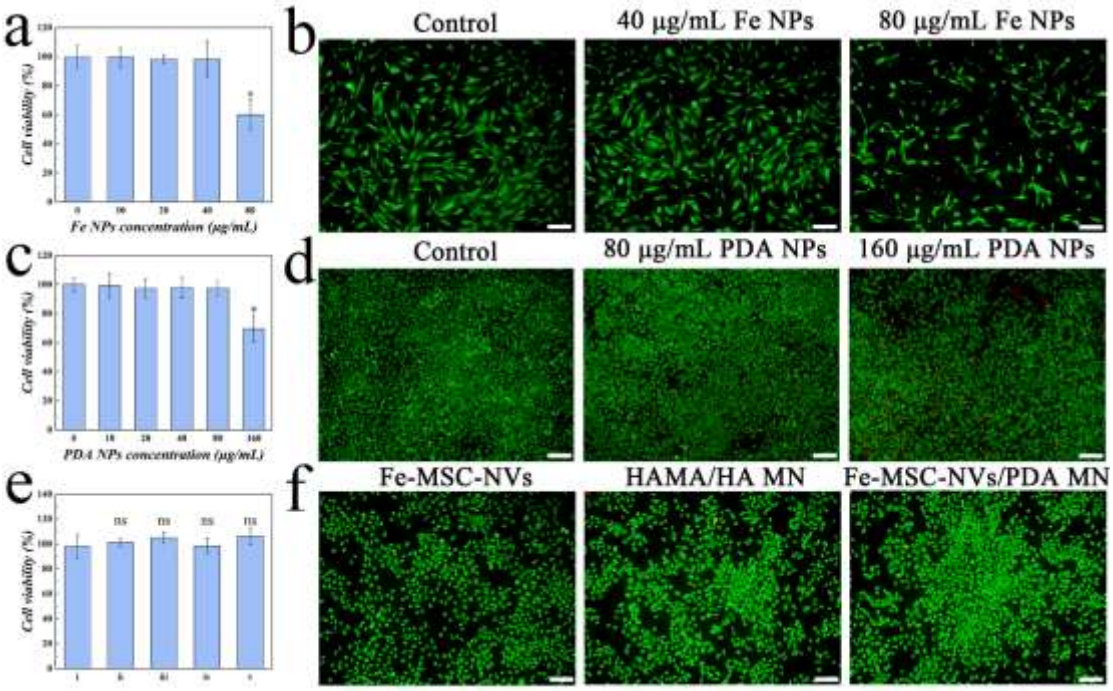

Figure S 2 (a) The viabilities of MSCs incubated with various amounts of Fe NPs for 24 h. n = 5,* $p < 0.05$ versus control. (b) Calcein AM/PI staining of MSCs in PBS (Control), 40, 80 μg/mL Fe NPs. (c) The viabilities of HUVEC incubated with various amount of PDA NPs for 24 h. n = 5, * $p < 0.05$ versus control. (d) Calcein AM/PI staining of HUVEC in PBS (Control), 80, 160 μg/mL PDA NPs. (e) The viabilities of HUVEC incubated for 24 h with different groups of (i) PBS (Control), (ii) 50 μg/mL MSC-NVs, (iii) 50 μg/mL Fe-MSC-NVs, (iv) HAMA/HA MN, (v) Fe-MSC-NVs/PDA MN. n = 5, ns $p > 0.05$. (f) Calcein AM/PI staining of HUVEC in 50 μg/mL Fe-MSC-NVs, HAMA/HA MN, Fe-MSC-NVs/PDA MN. Scale bars: 100 μm.

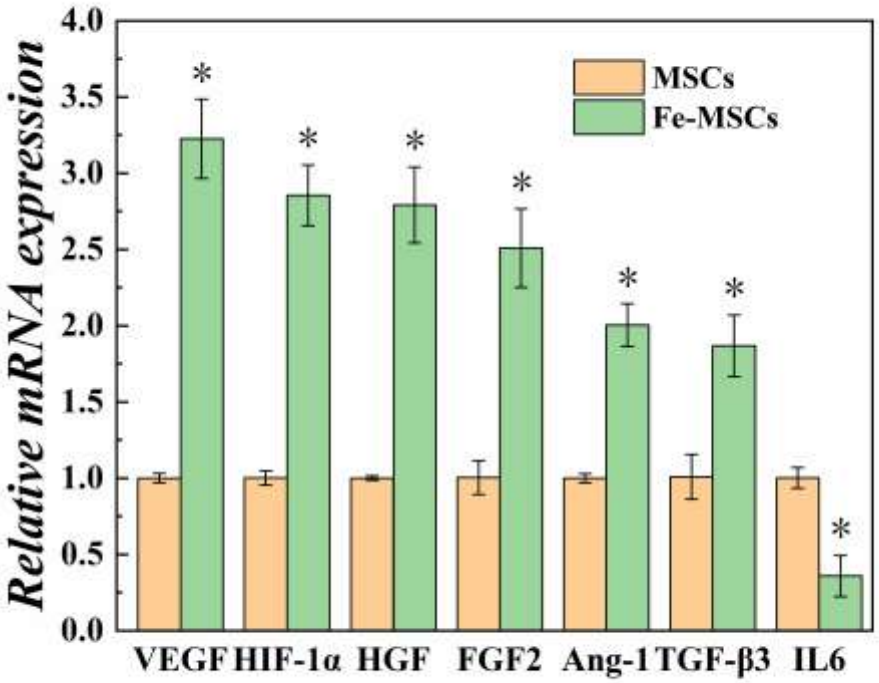

Figure S 3 Relative mRNA expression of VEGF, HIF-1α, HGF, FGF2, Ang-1, TGF-β3, and IL-6 in MSCs and Fe-MSCs. n = 3, * $p < 0.05$ versus MSCs.

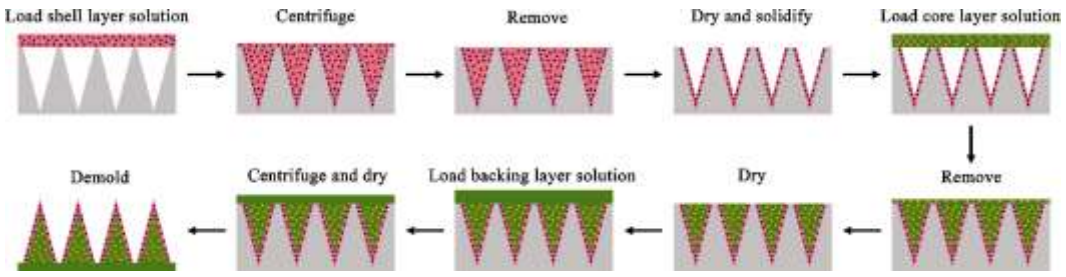

Figure S 4 Schematic illustration of the fabrication of the core-shell MNs.

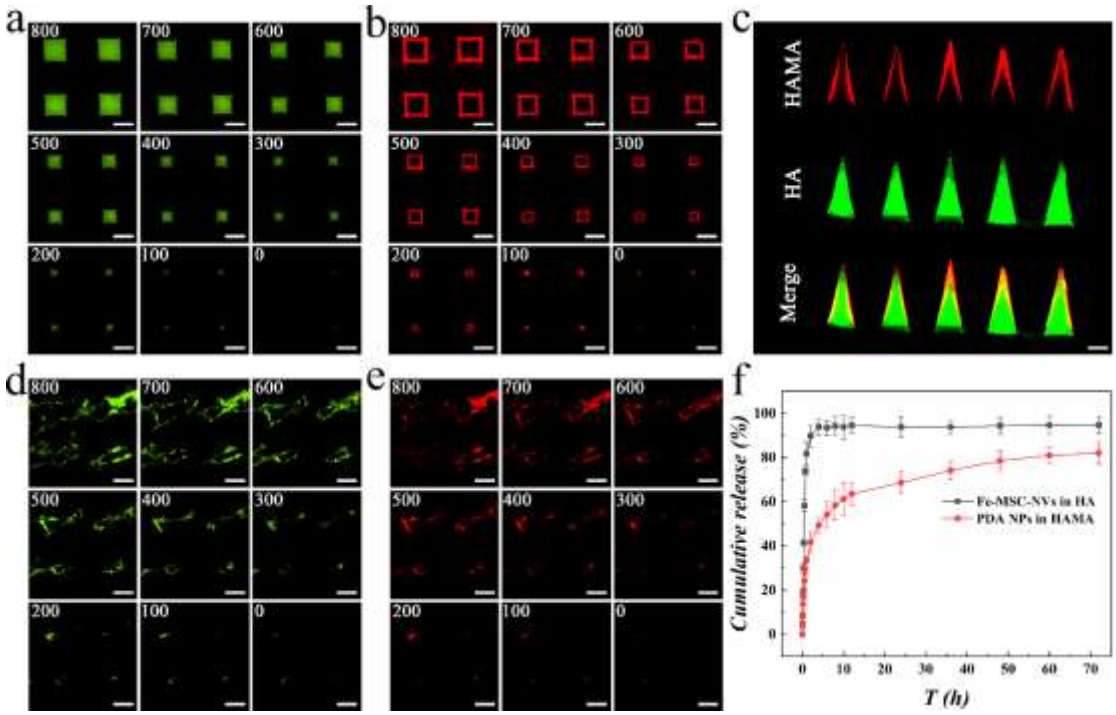

Figure S 5 CLSM images of (a) DiO-labled Fe-MSC-NVs layer and (b) red-microspheres layer at varying depths: 0, 100, 200, 300, 400, 500, 600, 700, 800 μm. Scale bars: 200 μm. (c) CLSM images of core-shell MN array. Scale bar: 200 μm. (d, e) *Ex vivo* transdermal delivery of DiO-labled Fe-MSC-NVs in HA hydrogel and red-microspheres in HAMA hydrogel through the designed MN patch. CLSM images of (d) DiO-labled Fe-MSC-NVs, (e) red-microspheres after the MN patch applied on the rat skin. Scale bars: 200 μm. (f) Stimulated release profile of DiO-labled Fe-MSC-NVs in HA hydrogel and red-microspheres in HAMA hydrogel from the core-shell MN patch, respectively (n=3).

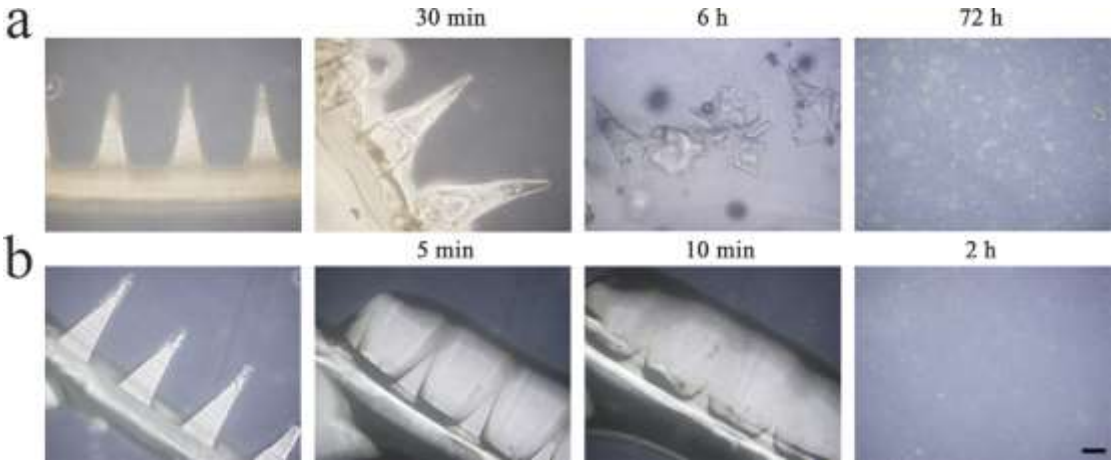

Figure S 6 The degradation of (a) HAMA/HA MN patch and (b) only HA MN patch in the PBS containing 100 units of hyaluronidase at 37°C at different times. Scale bar: 200 μm.

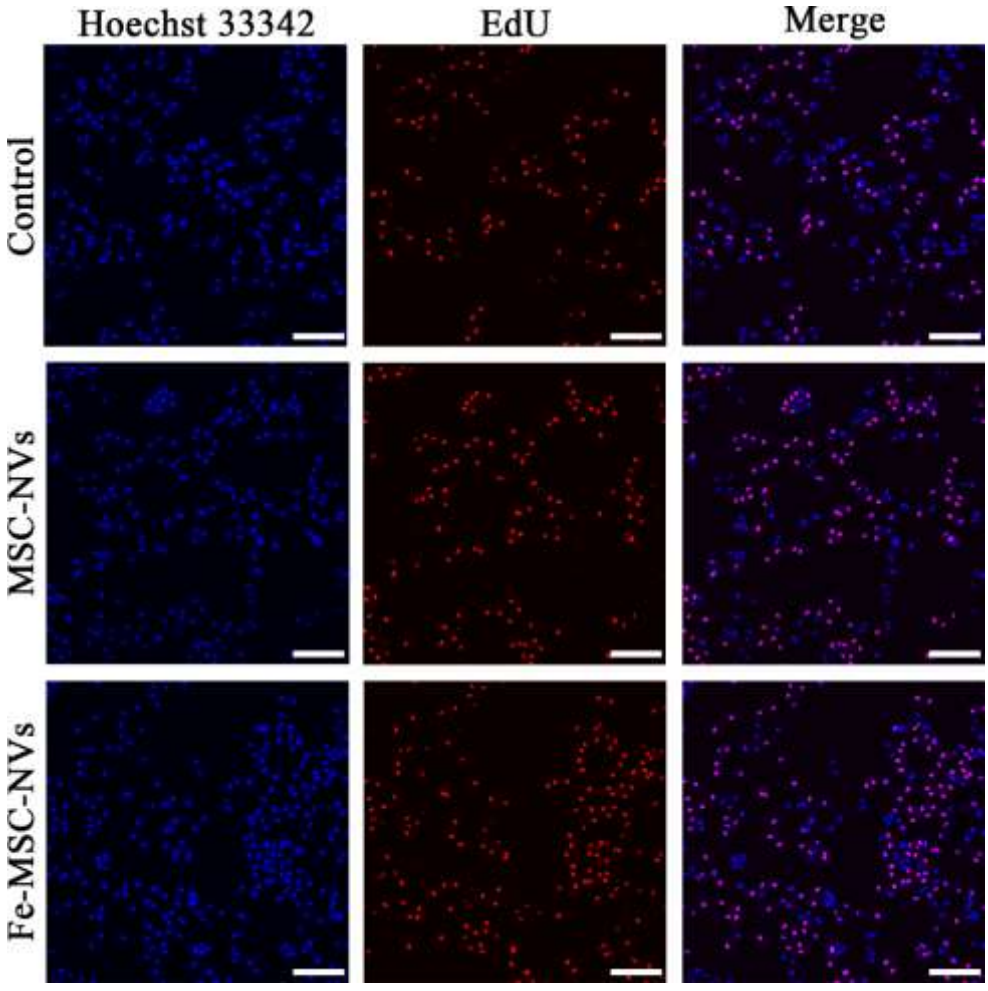

Figure S 7 Fluorescence images of HUVEC cultured in the medium supplementary with PBS (Control), MSC-NVs, or Fe-MSC-NVs for 24 h. Red: 5-Ethynyl-2'-deoxyuridine (EdU); blue: nucleus. Scale bars: 200μm.

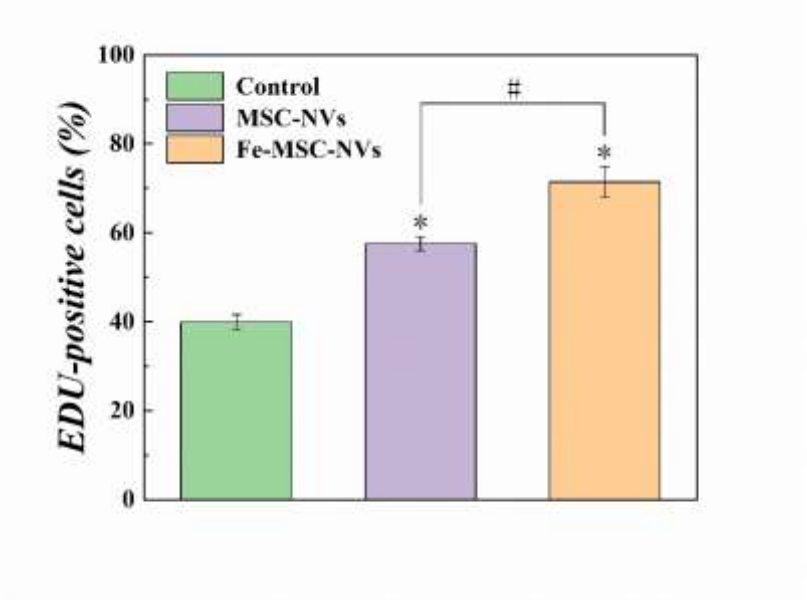

Figure S 8 Corresponding quantitative analysis of the proliferation assay in HUVEC.

n = 5, * $p < 0.05$ versus control. # $p < 0.05$.

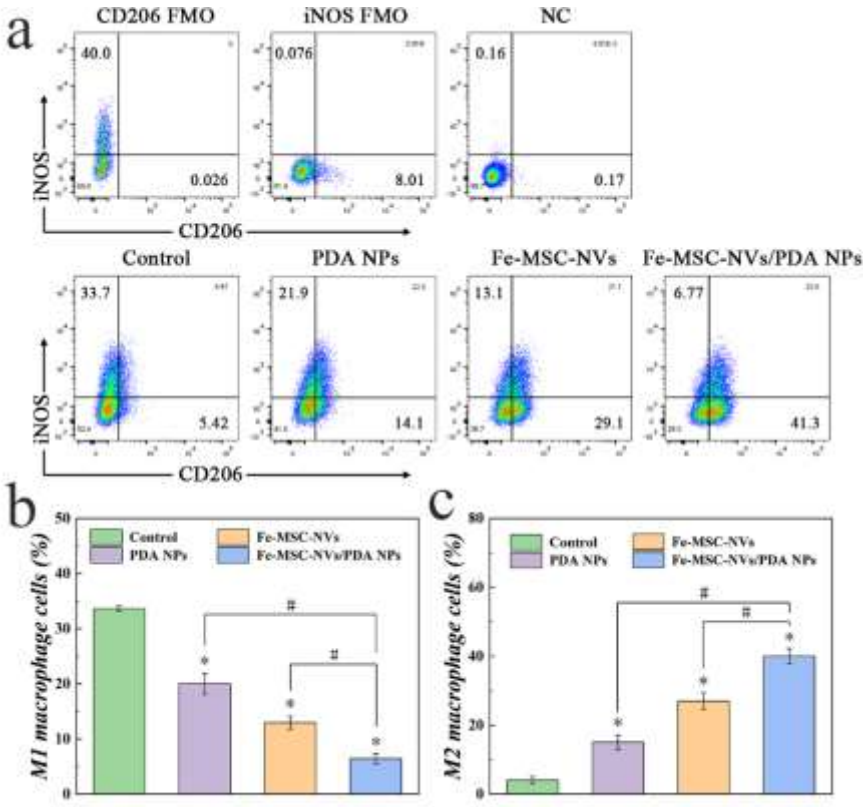

Figure S 9 (a) The CD206 FMO control and iNOS FMO control were used for gating different cell populations during FACS analysis; FACS analysis of iNOS and CD206 in $F40/80^{+}CD11b^{+}$ cells after being cocultured with PBS (negative control), LPS (control group), LPS/PDA NPs, LPS/Fe-MSC-NVs, or LPS/ Fe-MSC-NVs/PDA NPs. (b, c) Corresponding quantitative analysis of the positive expression percentage of (b) M1 macrophage cells ($iNOS^{+}CD206^{-}$) or (c) M2 macrophage cells ($CD206^{+}iNOS^{-}$) by flow cytometry. n = 5, * $p < 0.05$ versus control. # $p < 0.05$.

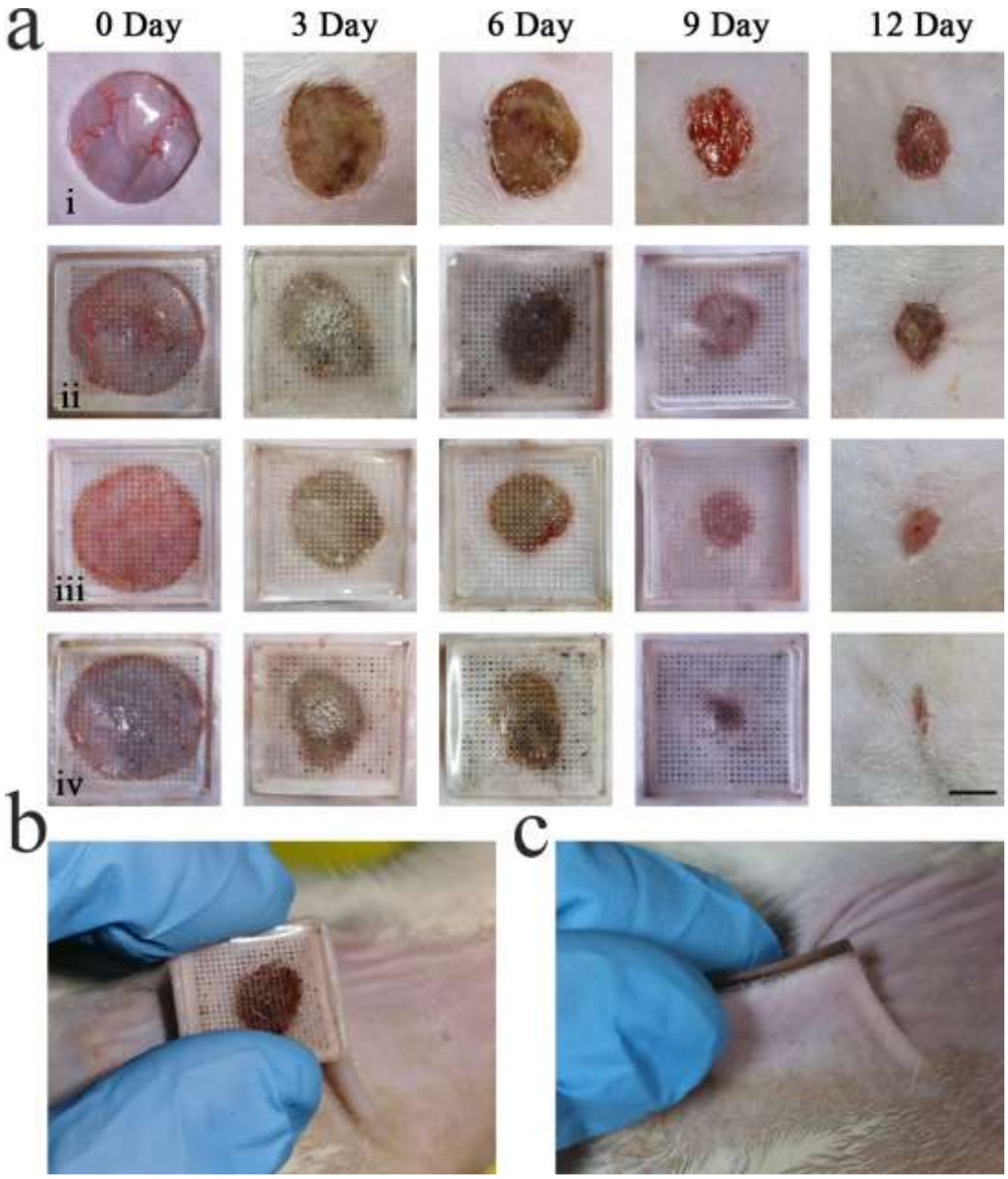

Figure S 10 (a) The photographic images of the patches applied on wound site in different treatment groups: control group, PDA MN, Fe-MSC-NVs MN, and Fe-MSC-NVs/PDA MN on days 0, 3, 6, 9, 12. Scale bar: 5 mm. The photographic images of the patches applied on wound site: (b) the front side and (c) the side view

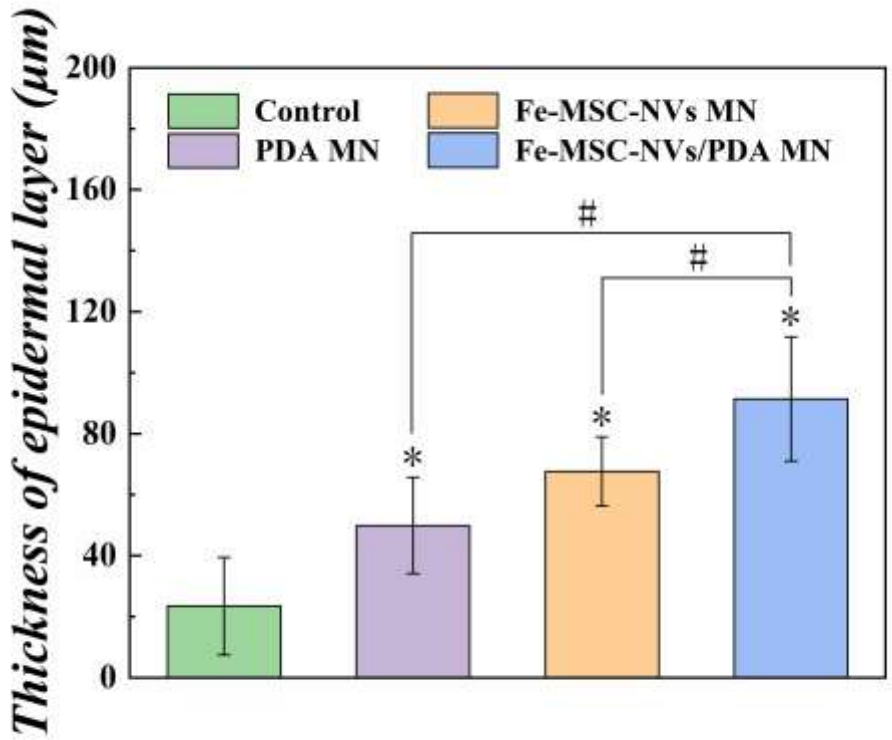

Figure S 11 Thickness analysis of epidermal layer in different treatment groups: control group, PDA MN, Fe-MSC-NVs MN, and Fe-MSC-NVs/PDA MN on day 12. n = 6, * $p < 0.05$ versus each control group. # $p < 0.05$.

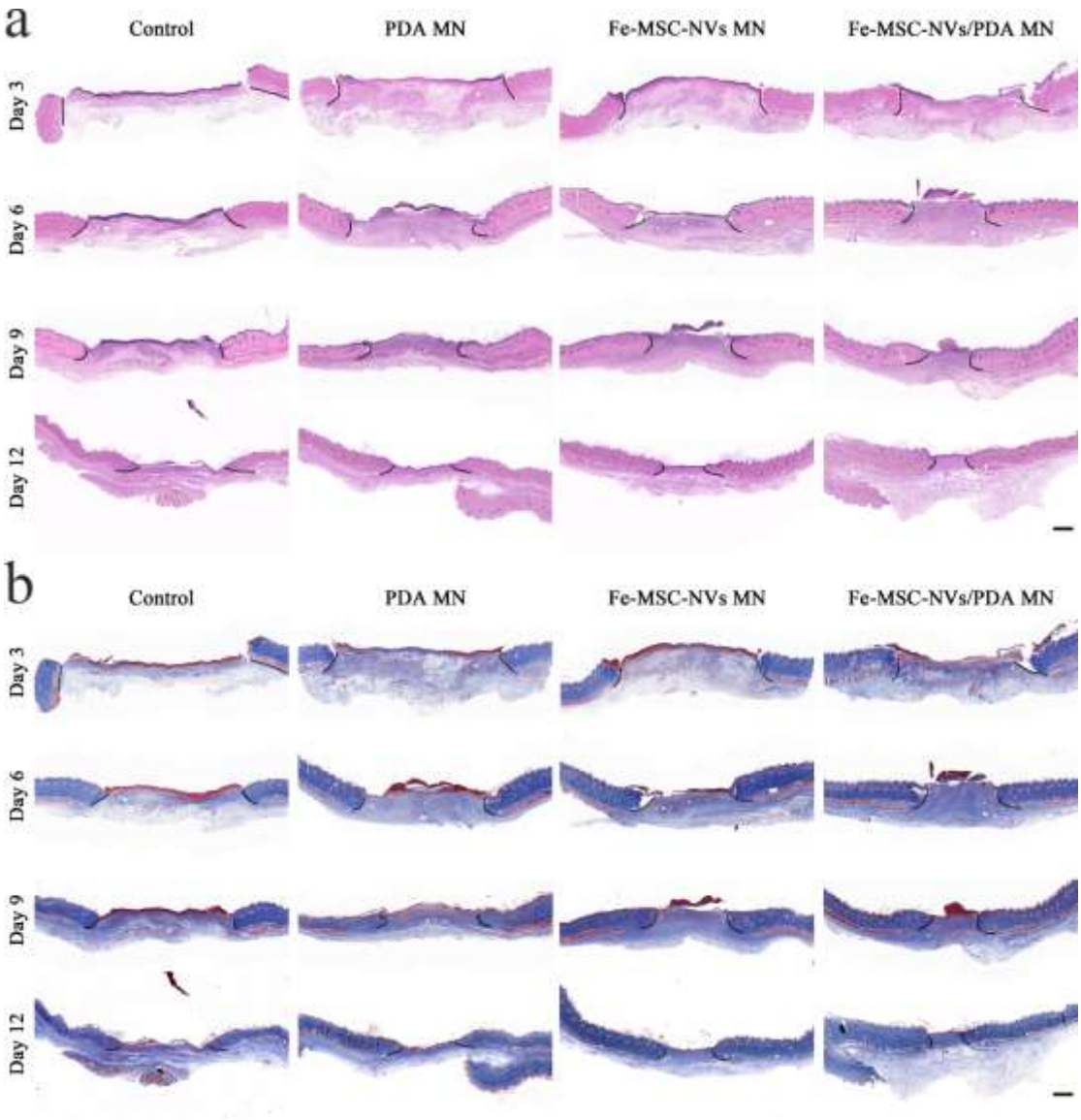

Figure S 12 Representative images of (a) hematoxylin and eosin (H&E) staining and (b) Masson's trichrome staining in different treatment groups: control group, PDA MN, Fe-MSC-NVs MN, and Fe-MSC-NVs/PDA MN on days 3, 6, 9, 12. Scale bars: 1 mm.

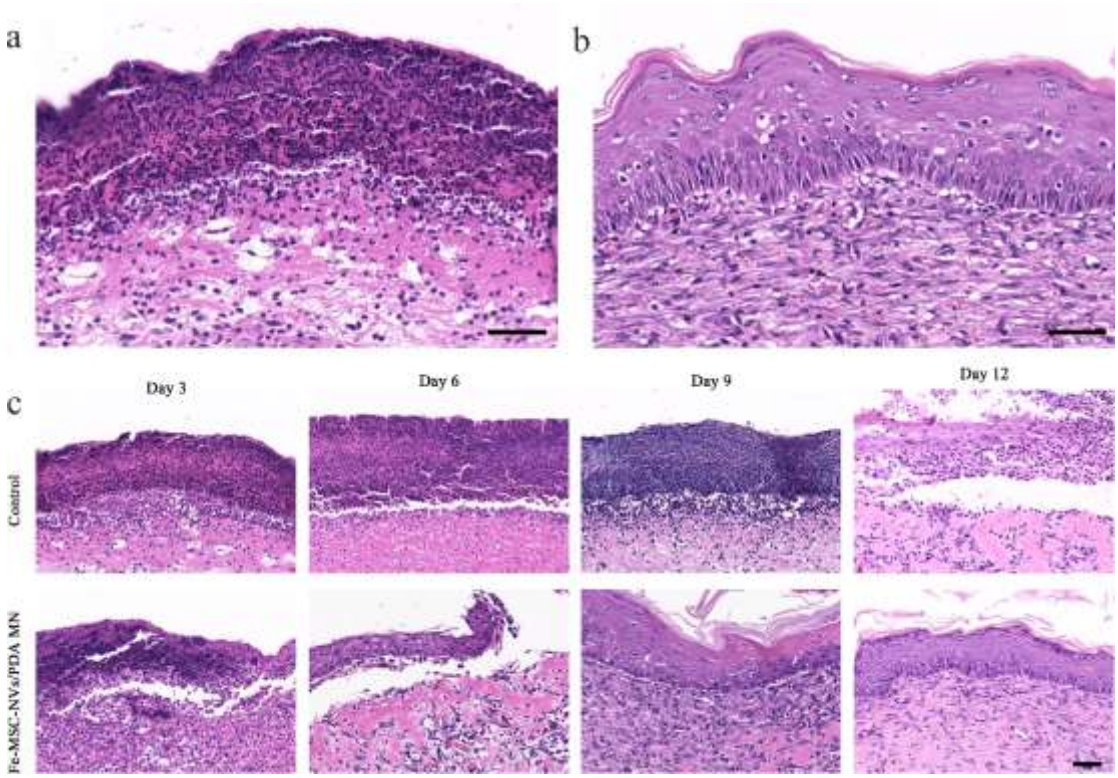

Figure S 13 Representative images of (a) wound crust and (b) epidermal layer in skin wounds. Scale bars: 50 μm. (c) Representative images of H&E staining in control group and Fe-MSC-NVs/PDA MN group. Scale bar: 50 μm.

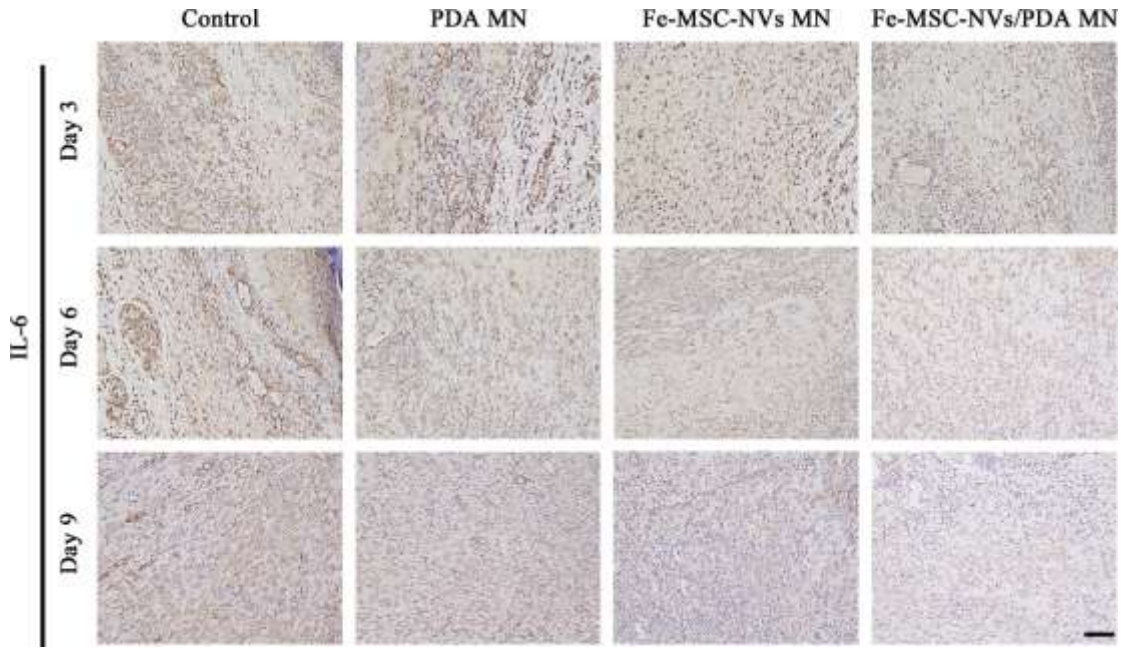

Figure S 14 Immunohistochemical staining of IL-6 of granulation tissues in different treatment groups: control group, PDA MN, Fe-MSC-NVs MN, and Fe-MSC-NVs/PDA MN on days 3, 6, 9. Scale bar: 100 μm.

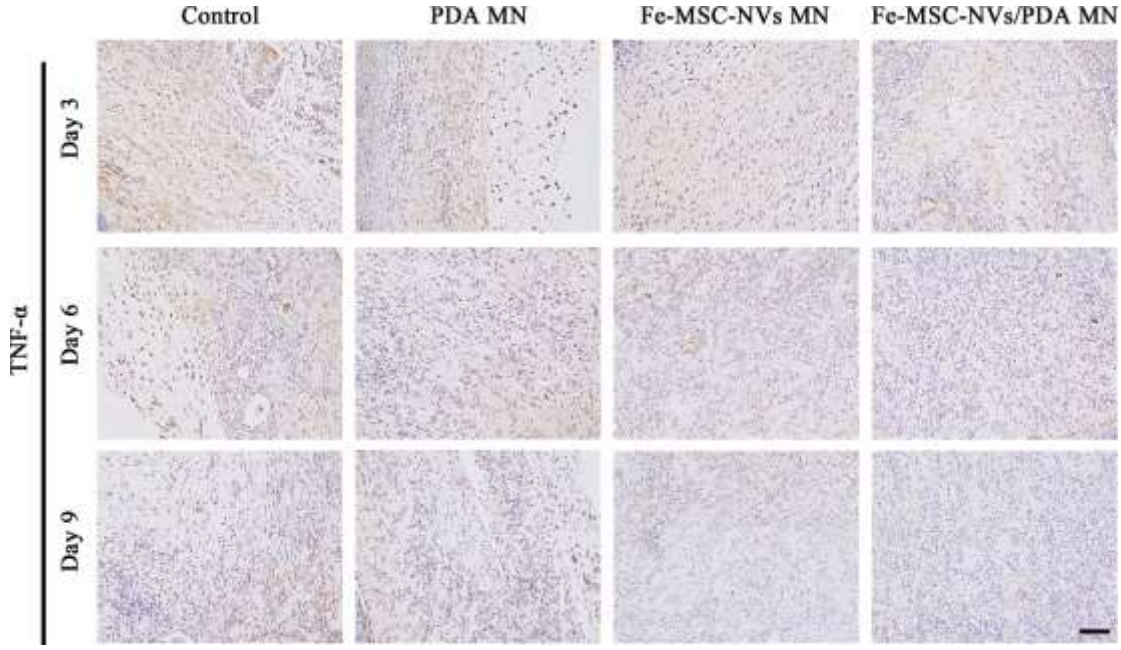

Figure S 15 Immunohistochemical staining of TNF-α of granulation tissues in different treatment groups: control group, PDA MN, Fe-MSC-NVs MN, and Fe-MSC-NVs/PDA MN on days 3, 6, 9. Scale bar: 100 μm.

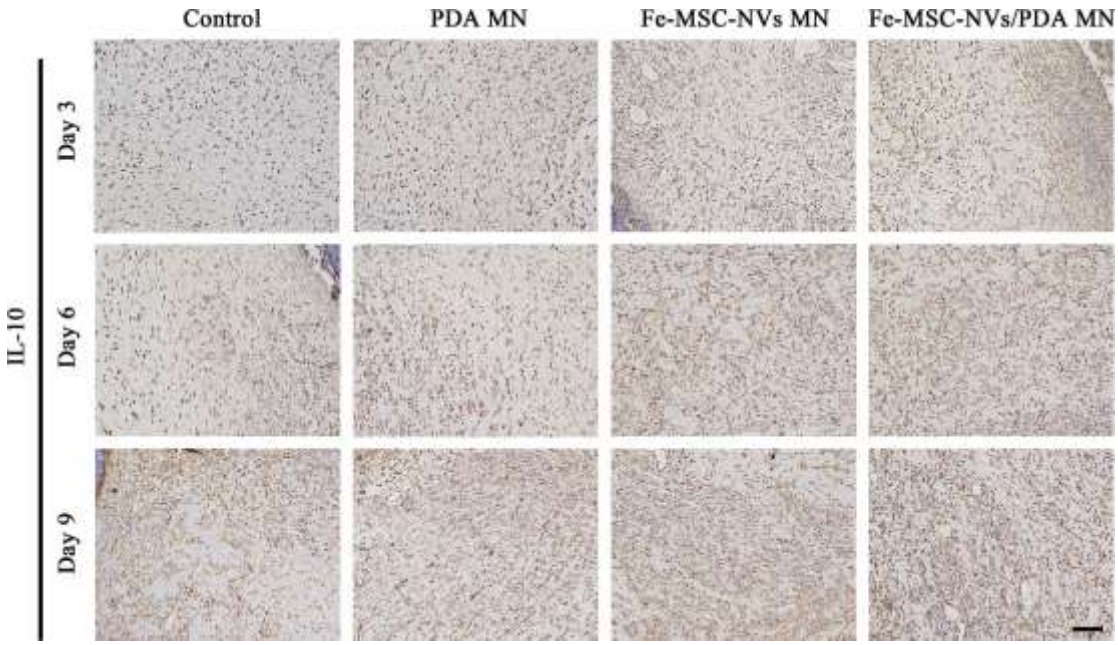

Figure S 16 Immunohistochemical staining of IL-10 of granulation tissues in different treatment groups: control group, PDA MN, Fe-MSC-NVs MN, and Fe-MSC-NVs/PDA MN on days 3, 6, 9. Scale bar: 100 μm.

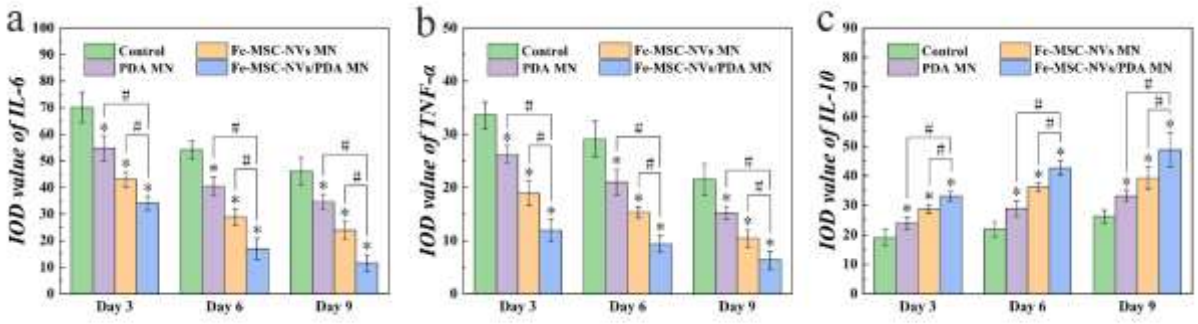

Figure S 17 Corresponding quantitative analysis of the mean value of the integrated optical density (IOD) for immunohistochemical staining of (a) IL-6, (b) TNF-α, and (c) IL-10 in different treatment groups. n = 6, * $p < 0.05$ versus control. # $p < 0.05$.

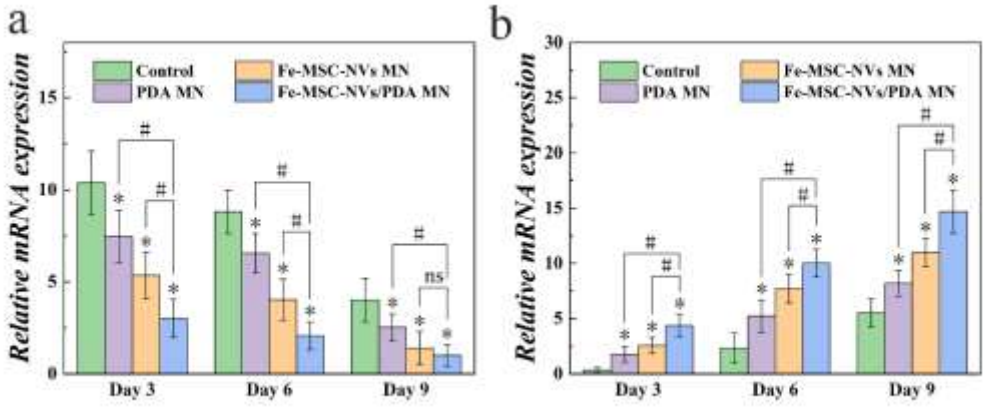

Figure S 18 Relative mRNA expression of (a) iNOS and (b) Arg-1 in granulation tissues of wounds in different treatment groups: control group, PDA MN, Fe-MSC-NVs MN, and Fe-MSC-NVs/PDA MN on days 3, 6, 9. n = 6, * $p < 0.05$ versus each control group. # $p < 0.05$.

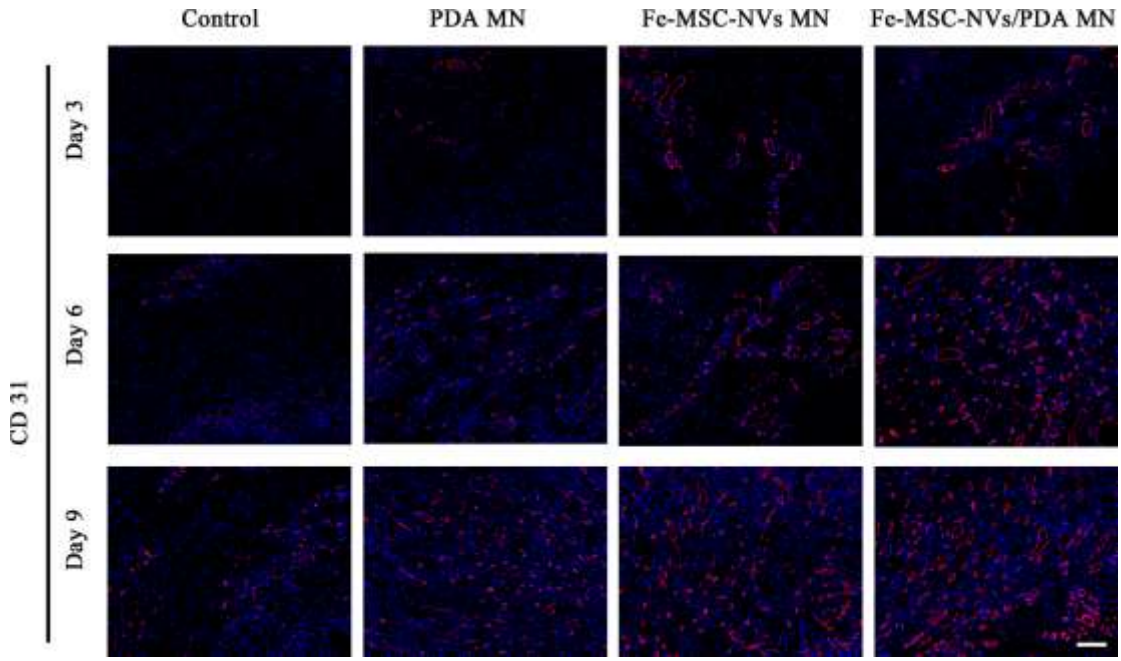

Figure S 19 Immunofluorescence staining of CD31 of granulation tissues in different treatment groups: control group, PDA MN, Fe-MSC-NVs MN, and Fe-MSC-NVs/PDA MN on days 3, 6, 9. Scale bar: 200 μm.

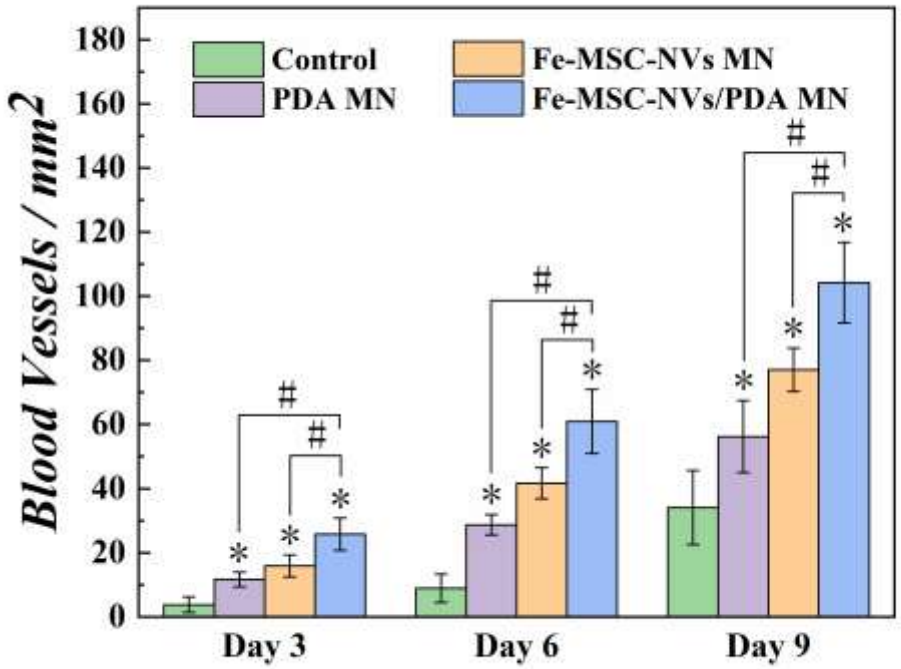

Figure S 20 Corresponding quantitative analysis of microvessel area stained by CD31 in different treatment groups on days 3, 6, 9. n = 6, * $p < 0.05$ versus control. # $p < 0.05$.

Table 1 Target gene primer sequences

Gene	Forward	Reverse
Human VEGF	TTGCCTTGCTGCTCTACCTC	AGCTGCGCTGATAGACATCC
Human HIF-1α	GTGGTGGTTACTCAGCACTTTT	CGTCCCTCAACCTCTCAGTT
Human HGF	GACGCAGCTACAAGGGAACA	AAAAGCTGTGTTCGTGTGGT

Human FGF2	CCACCTATAATTGGTCAAAGTGG	GAAACGAGGGAGAAAGGATGGA
Human Ang-1	GTGCTCACGTGGCTCGACTATA	GCACAGCAAGCTCAGCAGTT
Human TGFβ-3	TACTGCTTCCGGTGAGACTG	CAGGGACCCTCTGTTGAGTG
Human IL-6	GACAGCCACTCACCTCTTCA	CCTCTTTGCTGCTTTCACAC
Human GAPDH	ATGGGGAAGGTGAAGGTCG	GGGGTCATTGATGGCAACAATA
Rat iNOS	CCCTAAGAGTCACAAGCATC	AGGGTGTCGTGAAAAATCTC
Rat Arg-1	TGCCCTCTGTCTTTTAGGGCT	CTCCTCGAGGCTGTCCCTTA
Rat GAPDH	GCATCTTCTTGTGCAGTGCC	TACGGCCAAATCCGTTCACA