A Synergistic Polydiolcitrate-Copper Metal-Organic Framework Nanocomposite Improves Wound Healing

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Statement of Purpose: Chronic wounds such as non-healing diabetic foot ulcers continue to be a great challenge for the physician and contribute to increasing healthcare costs.[1] Copper ions have been reported to accelerate wound healing due to antibacterial properties and their ability to up-regulate the expression of VEGF and TGF-β. However, elevated non-physiological concentrations of copper ions can be toxic because the ion can interfere with the homeostasis of other metal ions, damage DNA, and generate reactive oxygen species that can adversely modify proteins, lipids and nucleic acids.[2] Entrapping the copper ions within hydrogels can be inefficient and often destabilizes the hydrogel.[3] Therefore, new methods to optimally deliver copper ions to the wound environment are needed. We propose to slowly deliver copper ions through the use of metal-organic frameworks. Metal–organic frameworks (MOFs) are highly porous metal coordination networks that form with organic ligands. However, MOFs are unstable in physiological solutions rendering them unsuitable for use in the wound bed. To address this problem, we investigated whether the use of copper-based metal organic framework (CMOF) nanoparticles dispersed within a thermoresponsive polydiolcitrate would lead to a synergistically stabilized CMOF-hydrogel composite for potential use as a wound dressing. Therefore, the objective of this research was to synthesize a stable CMOF-polydiolcitrate hydrogel nanocomposite and investigate whether it would be suitable as a novel thermoresponsive dressing to accelerate wound healing.

Methods: Poly(polyethyleneglycol citrate-co-N-isopropylacrylamide) (PPCN) was prepared as previously described by us .[4] CMOFs were added to a PPCN solution and mixed thoroughly by vortexing to obtain PPCN/CMOF (PCMOF). The PCMOF was treated with 10% FBS and photographed. Rheological measurements were obtained to confirm hydrogel formation. The morphology of CMOF and PCMOF after treatment with 10% FBS was observed using TEM. The release of copper from PCMOF was measured in PBS with or without 10% FBS. The cytotoxicity of PCMOF was evaluated by MTT assay, and the effects of PCMOF on wound closure over time were measured *in vitro* via a scratch assay on human keratinocytes (HeKa) and human dermal fibroblasts (HDF) cells and *in vivo* in healthy mice.

Results: PCMOF showed higher stability than PPCN/$CuSO_4$ ($PCuSO_4$), as PCMOF hydrogel structure remained intact in PBS containing 10% FBS, while $PCuSO_4$ hydrogel broke into small pieces (Figure 1A). Also, PCMOF formed a stable 3D hydrogel, with a gelation temperature (T_{gel}) of 26.1°C, which was similar to that of PPCN (28.4°C). $PCuSO_4$ could not form a stable 3D hydrogel structure, as no crossover between G' and G" was observed (Figure 1B). PPCN prevented the destabilization of the CMOF due to exposure to FBS (Figure 1C, D). PCMOF exhibited a much slower copper ion release rate compared to $PCuSO_4$. PCMOF exhibited no obvious toxicity against both HeKa and HDF cells (Figure 1E, F) and had the best effect on wound closure rates *in vitro* and *in vivo*.

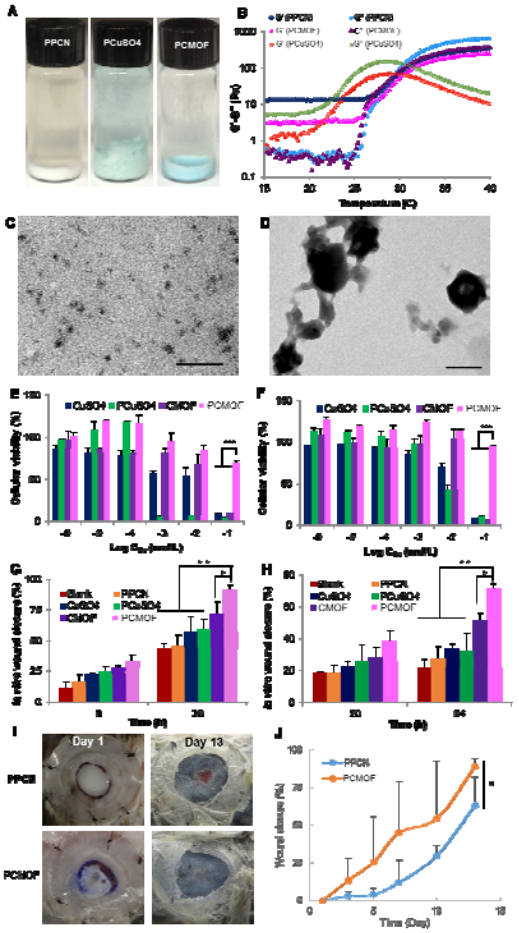

Figure 1. (A) Photograph of hydrogel samples in PBS containing 10% FBS at 37°C. (B) Rheology measurements of PPCN, $PCuSO_4$ and PCMOF. TEM imaging of (C) CMOF and (D) PCMOF after treatment with 10% FBS at 37°C (Scale bars=200 nm). Cytotoxicity of $CuSO_4$, $PCuSO_4$, CMOF and PCMOF when added to (E) HeKa cells and (F) HDF cells. Effects of PPCN, $CuSO_4$, $PCuSO_4$, CMOF and PCMOF on the migration of (G) HeKa cells, and (H) HDF cells cultured on tissue culture plastic. Effects of PPCN and PCMOF on skin wound healing in mice (I, J). (*$P<0.05$, **$P<0.01$)

Conclusions: CMOFs can be stabilized within PPCN hydrogels, enabling the fabrication of a promising novel thermoresponsive wound dressing.

References:

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