

Self-assembly of histidine-containing peptides and applications

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Abstract

Self-assembling peptides consist of short chains of amino acids capable of spontaneously forming higher-order structures such as fibers, tubes, and hydrogels [1]. Among these, histidine peptides are widely studied since histidine is an essential amino acid with a unique property. Its imidazole side chain exhibits pH sensitivity, allowing histidine peptides to undergo conformational changes and self-assemble into supramolecular structures in response to environmental factors like pH or salinity. Such versatility holds promise for pharmaceutical applications. Expanding on prior research [2,3], recent studies have focused on evaluating the effectiveness of coordinating histidine-containing peptides with copper and zinc to produce Zn and Cu nanoparticles. Through studying the morphology of these His-peptide NPs (Cyclo-(His-Phe), cHF NPs) using techniques such as FESEM, TEM, and FTIR, our ultimate goal is to evaluate the **antimicrobial properties** of the synthesized materials against selected Gram-positive and Gram-negative bacterial strains, as well as their potential as **anticancer metal-based drugs and antibiotics**. Preliminary experiments have shown promising results in both directions, underscoring the versatility of these nanostructures in biomedical applications.

Structural Characterization of the peptide

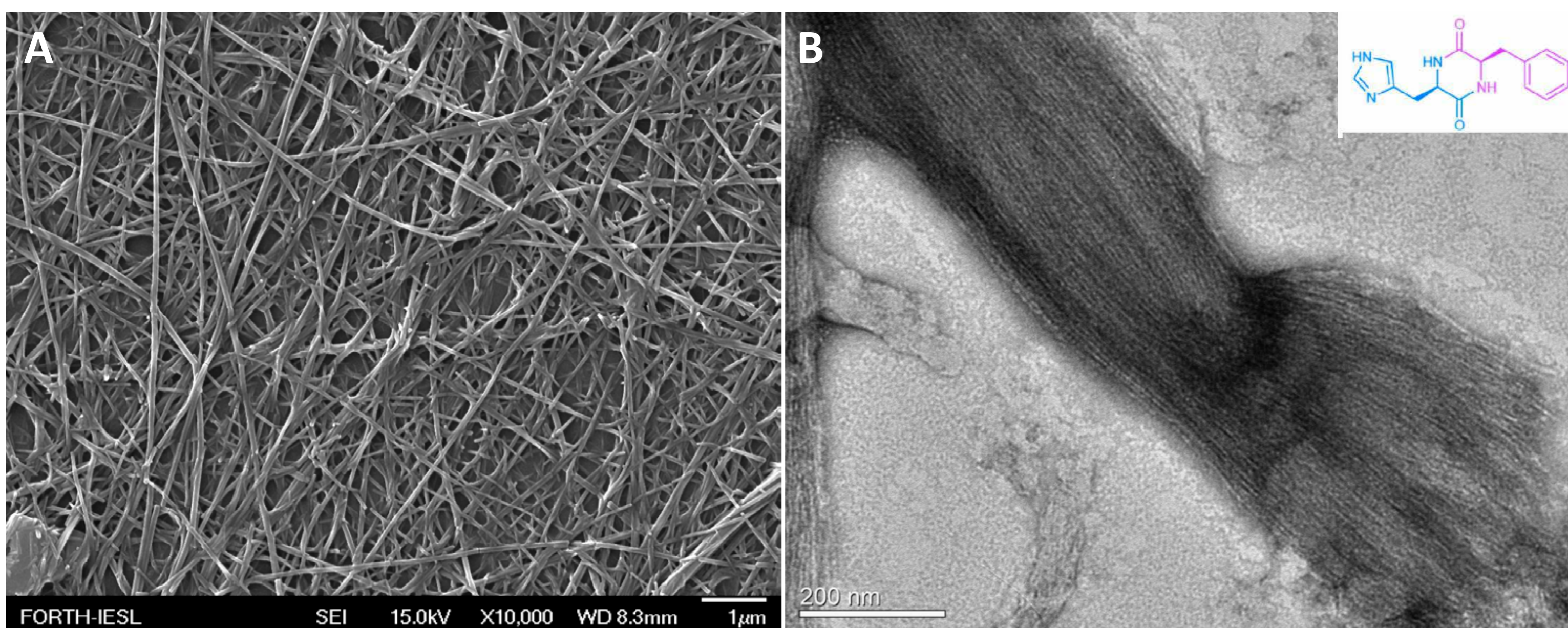


Figure 1: Cyclo-(His-Phe) (CHF) at the concentration of 1 mg/mL in PBS 1X under A) Field-emission SEM B) TEM microscope.

Preparation of cHF-CuNPs and cHF-ZnNPs

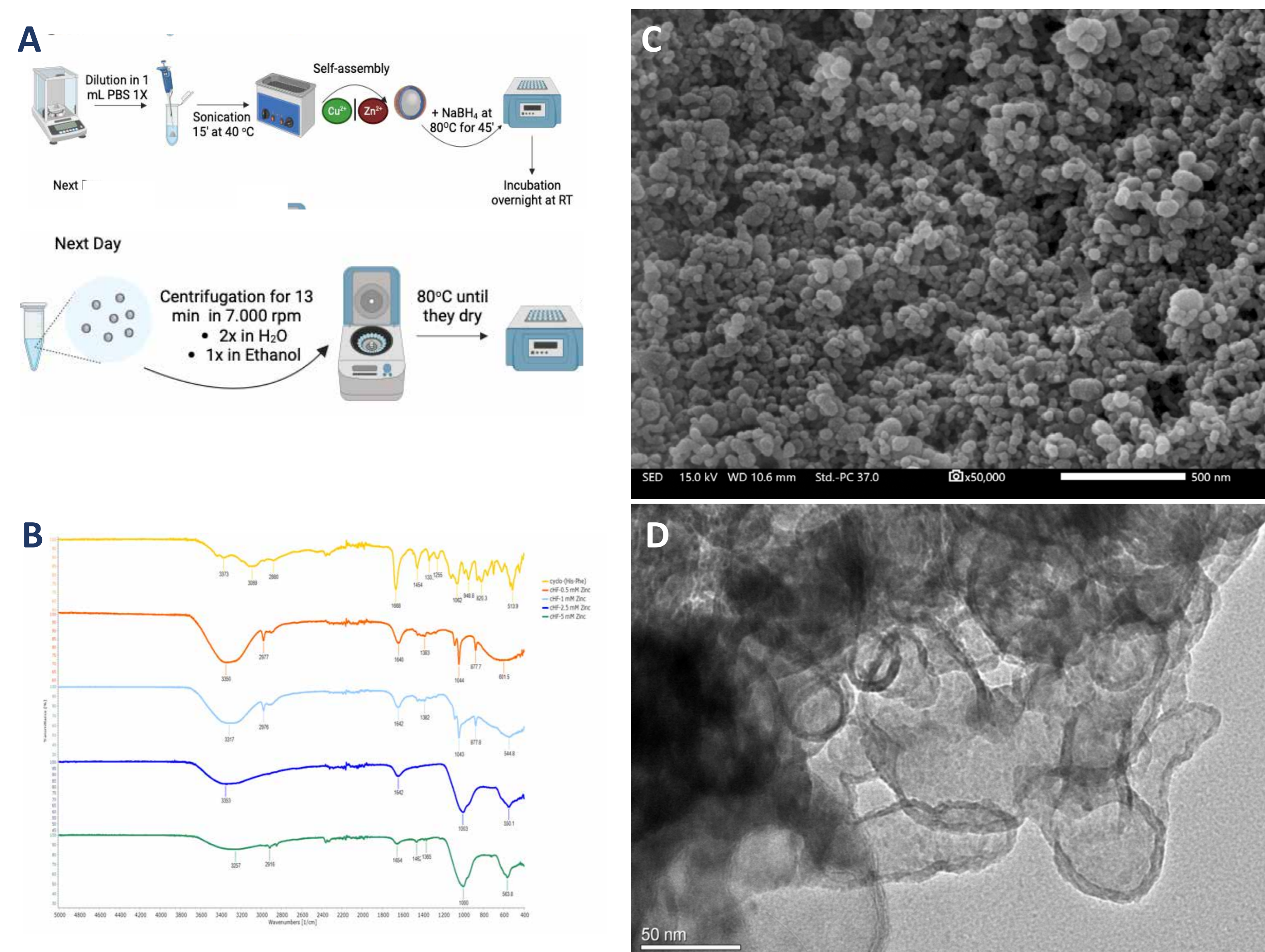


Figure 2: A) Schematic representation of the preparation of cHF-NPs B) FTIR analysis of cHF-ZnNPs at different metal concentrations C, D) Field-emission SEM images of cHF-ZnNPs (initial metal concentration was 1mM).

Conclusions

- Cyclic-(His-Phe) can spontaneously self-assemble into **amyloid fibrils**
- Cyclic-(His-Phe) can coordinate successfully with metal ions, **zinc**, and **copper**, due to the presence of **histidine** residue (FESEM and FTIR), and form peptide-metal NPs.
- Antimicrobial potency: The cHF-CuNPs (1 mM) showed limited cytotoxicity on L929 cells and strong antimicrobial effects against *E.coli* and *St. aureus*.
- Anticancer potency: The cHF-ZnNPs showed strong cytotoxicity against the MG-63 osteosarcoma cell line.

References

- [1] G. Fichman, E. Gazit, Acta Biomater, 2014, 10, (4), 1671–1682.
- [2] E. Glymenaki, et. al., ACS Omega, 2022, 7, (2), 1803–1818.
- [3] C.P. Apostolidou, et. al., Biomolecules, 2024, 14, (2), 226.

Evaluation of the antimicrobial potency of cHF-CuNPs and cHF-ZnNPs

The cHF-CuNPs and cHF-ZnNPs were prepared with different metal concentrations. Then, all the prepared samples were tested for their antimicrobial efficiency against *E. coli* and *S. aureus*.

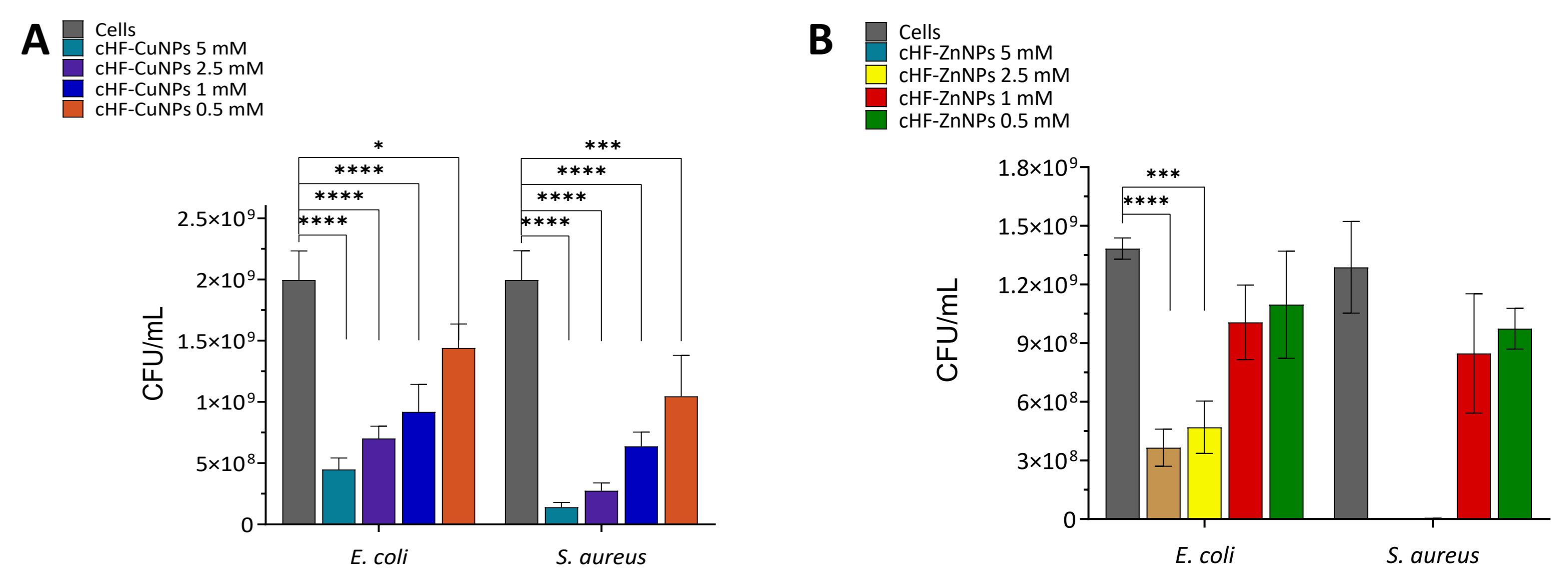
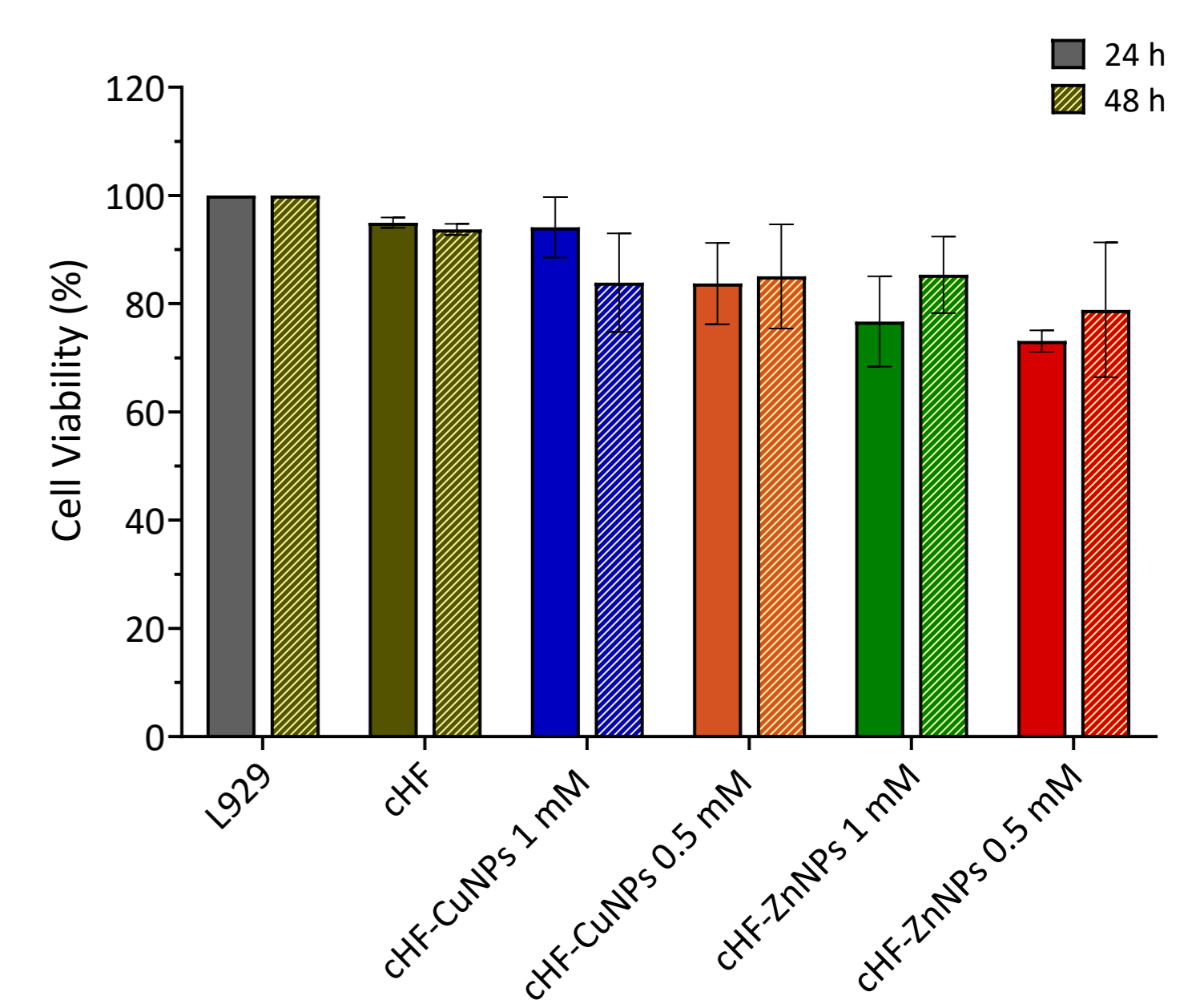


Figure 3: The graph depicts bacterial growth expressed as colony forming unit per mL (CFU/mL) of *E. coli* and *S. aureus* after 24 hours A) cHF-CuNPs and B) cHF-ZnNPs.

Figure 4: Cytotoxicity assessment at 24 and 48 hours on L929 fibroblast cell line. cHF NPs exhibit high cytocompatibility at lower metal concentrations.



Evaluation of the cytotoxicity of cHF-CuNPs and cHF-ZnNPs under different pH on MG-63 osteosarcoma cell line

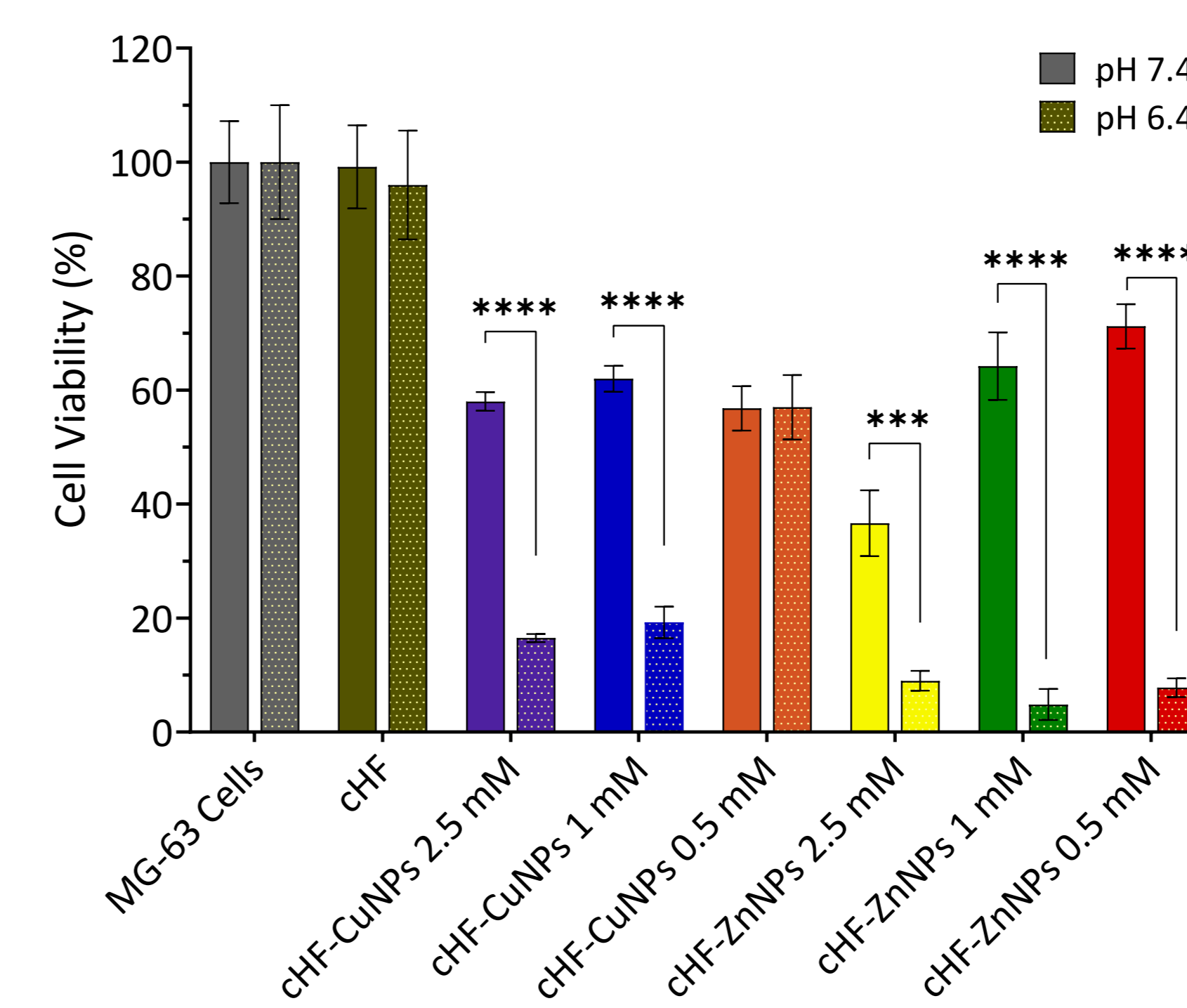


Figure 5: Cell viability in the presence of cHF-NPs after 24 h at pH 6.4 and pH 7.4 was studied by MTT test.