Electronic Supplementary Material (ESI) for RSC Advances.

Supplementary Information

Zinc oxide nanocrystals as nano-antibiotic and osteogenic/osteoinductive agents

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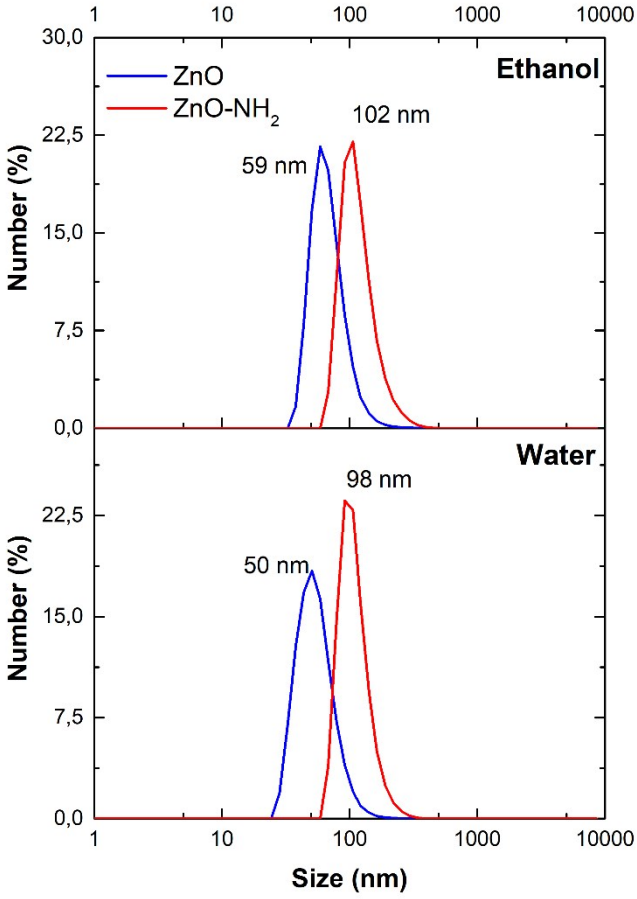


Figure S1. Size distribution curves deriving from Dynamic Light Scattering (DLS) measurements for ZnO and ZnO-NH_2 NCs in Ethanol (top) and water (bottom)

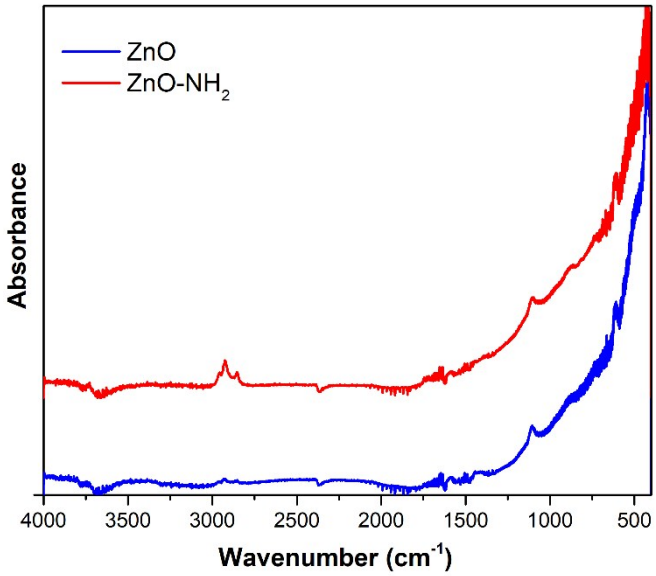


Figure S2. Fourier Transform Infra-Red (FT-IR) spectra of ZnO and ZnO-NH_2 NCs

The FT-IR spectra (Figure S2) of ZnO and ZnO–NH_2 nanocrystals show both an intense mode at 440 cm^{-1} typical of Zn–O vibration . In addition, the two spectra show, with different intensities, the symmetric and antisymmetric stretching of –CH_2 and –CH_3 groups at 2860 and 2925 cm^{-1}, respectively. These vibrations are attributed to residual acetate
groups on the ZnO surface due to the precursors used in the synthetic procedure. However, in the case of ZnO–NH_2 NCs, the 2860 and 2925 cm^{-1} stretching modes are more intense because of the presence of the propyl chain of the amine-functional group, thus confirming the successful functionalization of the ZnO surface.
Moreover, the broad band from 3600 to 3400 cm^{-1}, due to the stretching vibrations of hydroxyl groups on the ZnO surface, are less pronounced in the ZnO–NH_2 sample with respect to the pristine ZnO. Since the APTMS moiety links through hydroxyl groups to the oxide surface (Zn–OH), leading to Zn–O–SiR bonds, the above-mentioned-observation further confirms the successful amine functionalization of our ZnO NCs.

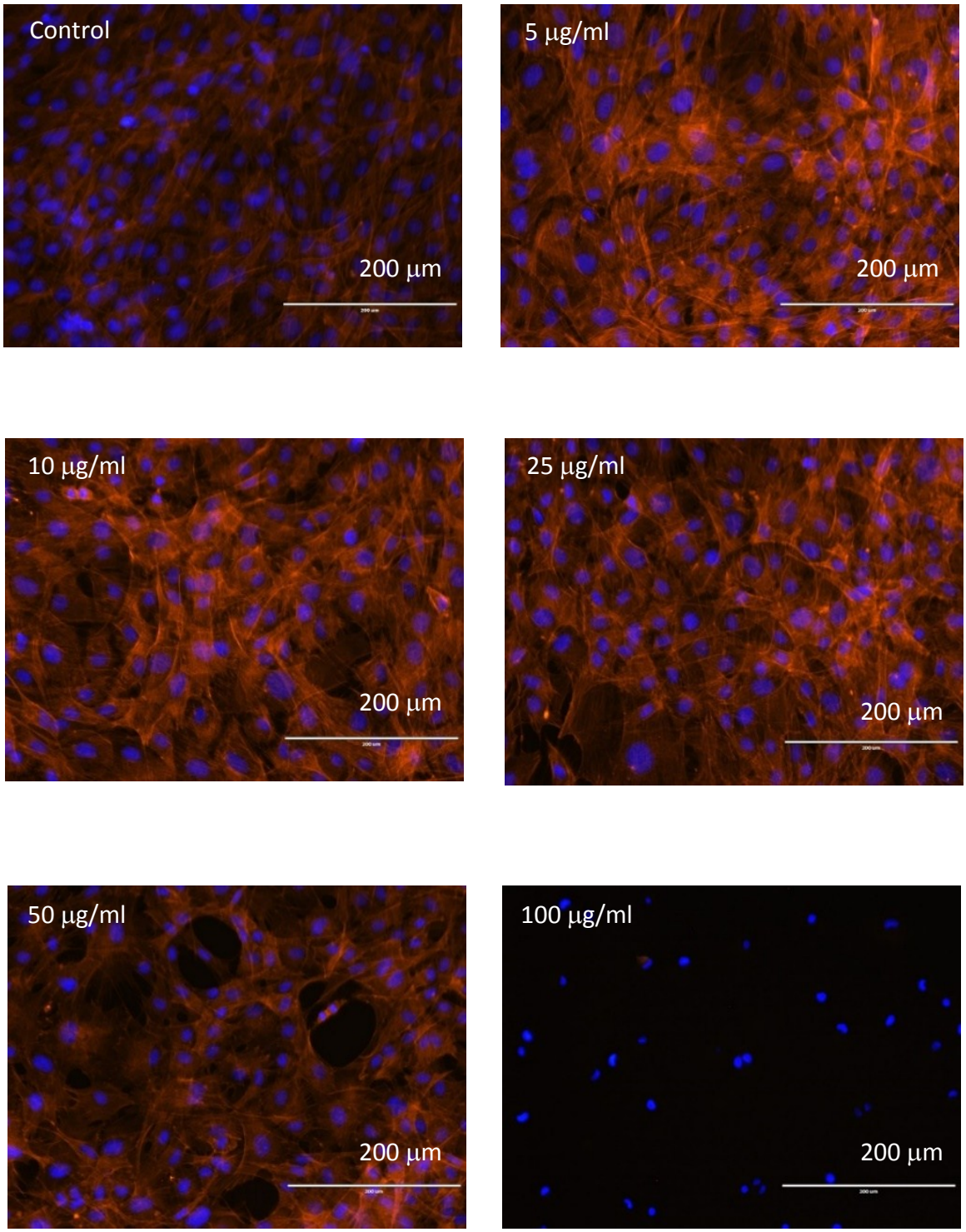


Figure S3. Fluorescent microscope images of pre-osteoblasts cultured up to 70 % of confluence after incubation for 4 days with ZnO NCs at different concentrations.

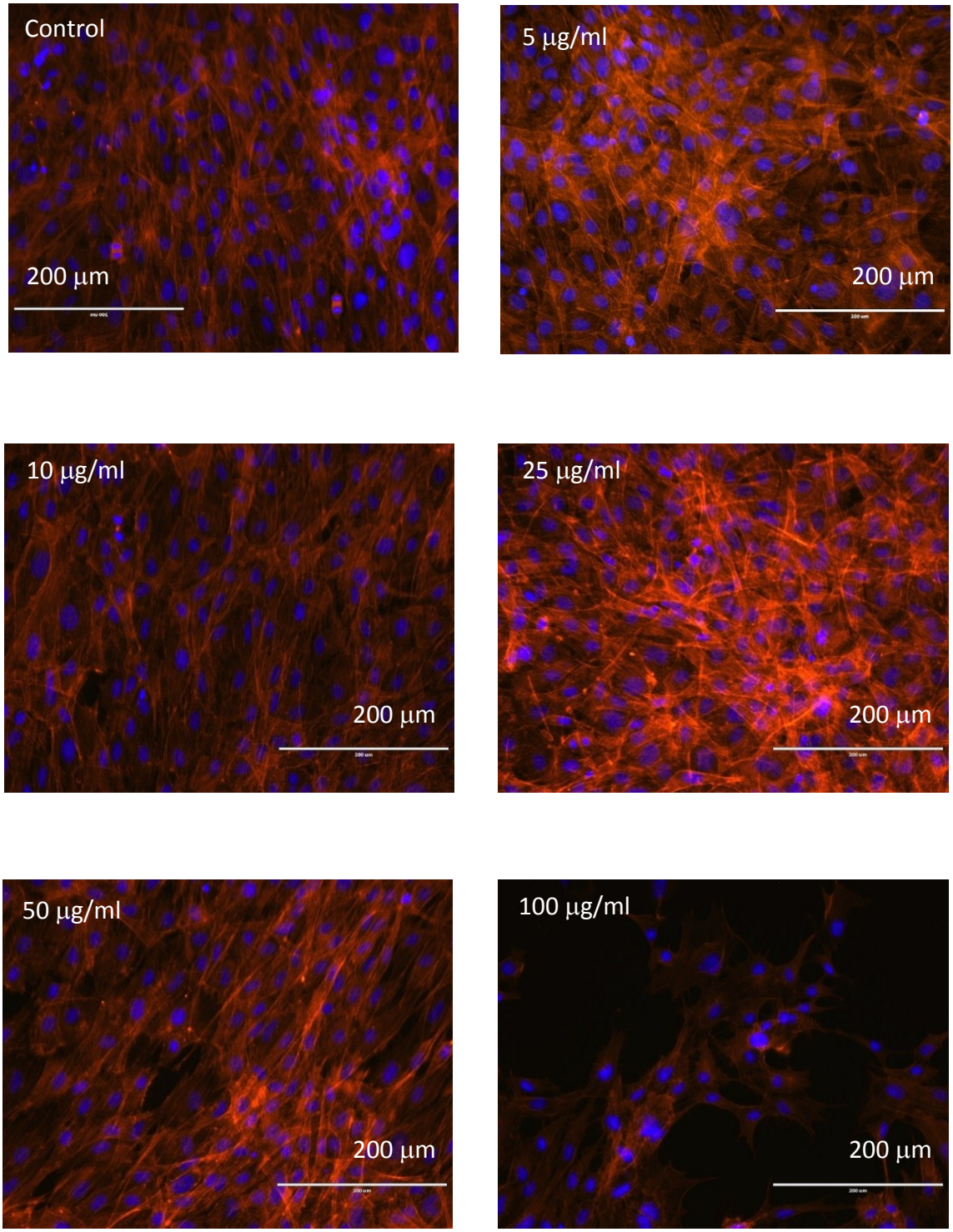
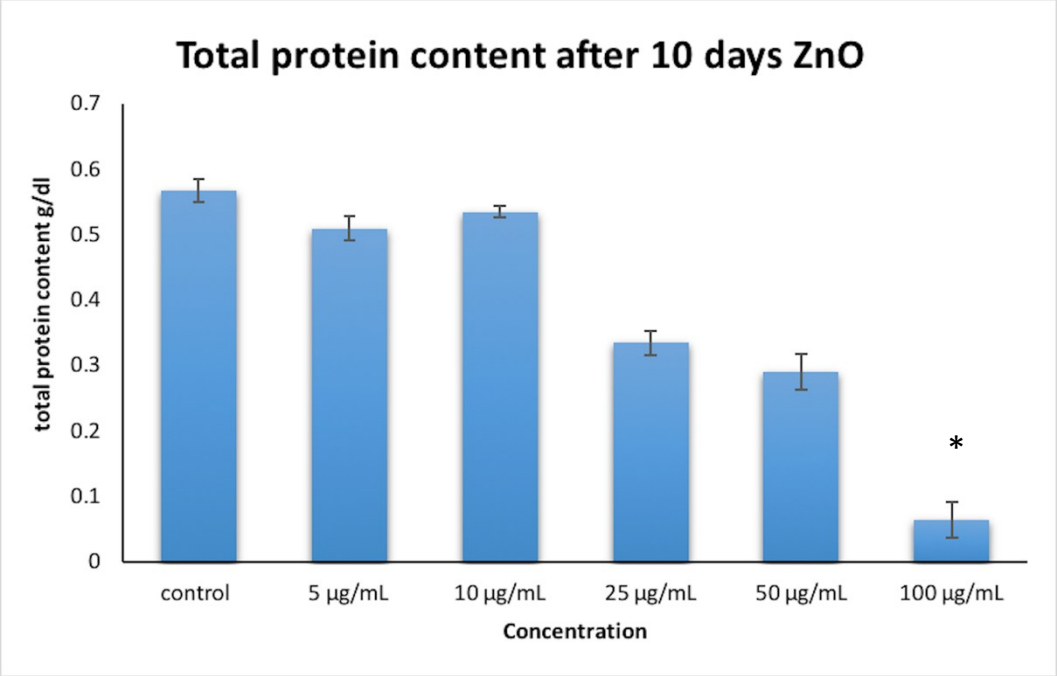


Figure S4. Fluorescent microscope images of pre-osteoblasts cultured up to 70 % of confluence after incubation for 4 days with ZnO-NH_2 NCs at different concentrations.



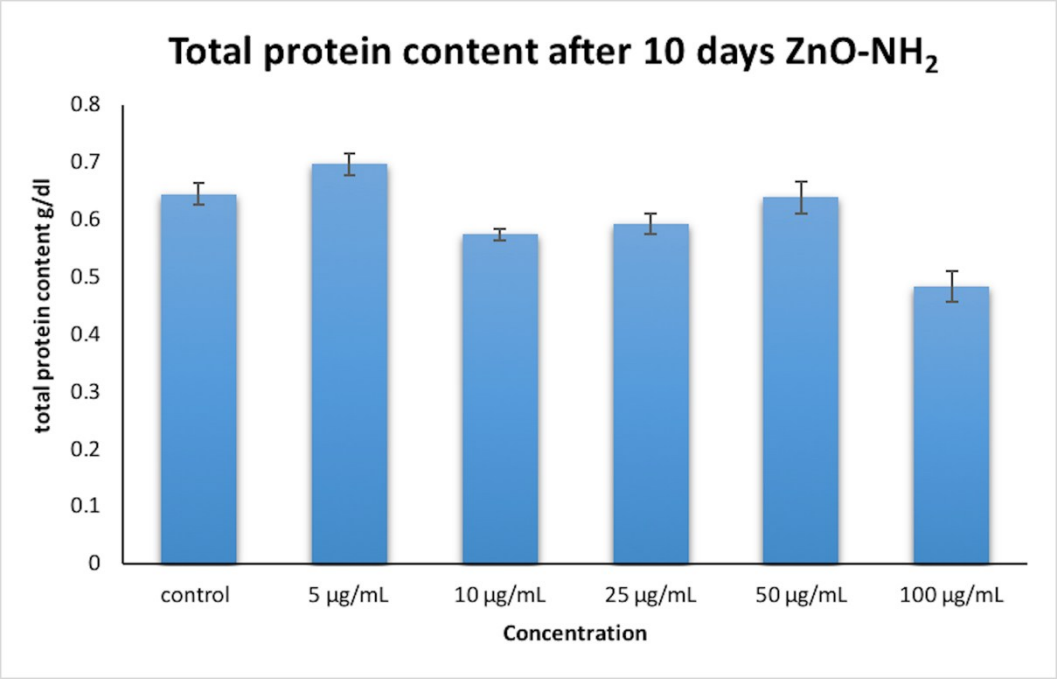


Figure S5. Differentiation assays in term of total protein content after 10 days of incubation with ZnO (top) and ZnO-NH_2 (bottom) NCs at different concentrations. ($p < 0.05$, significant differences compared to control denoted by an asterisk.

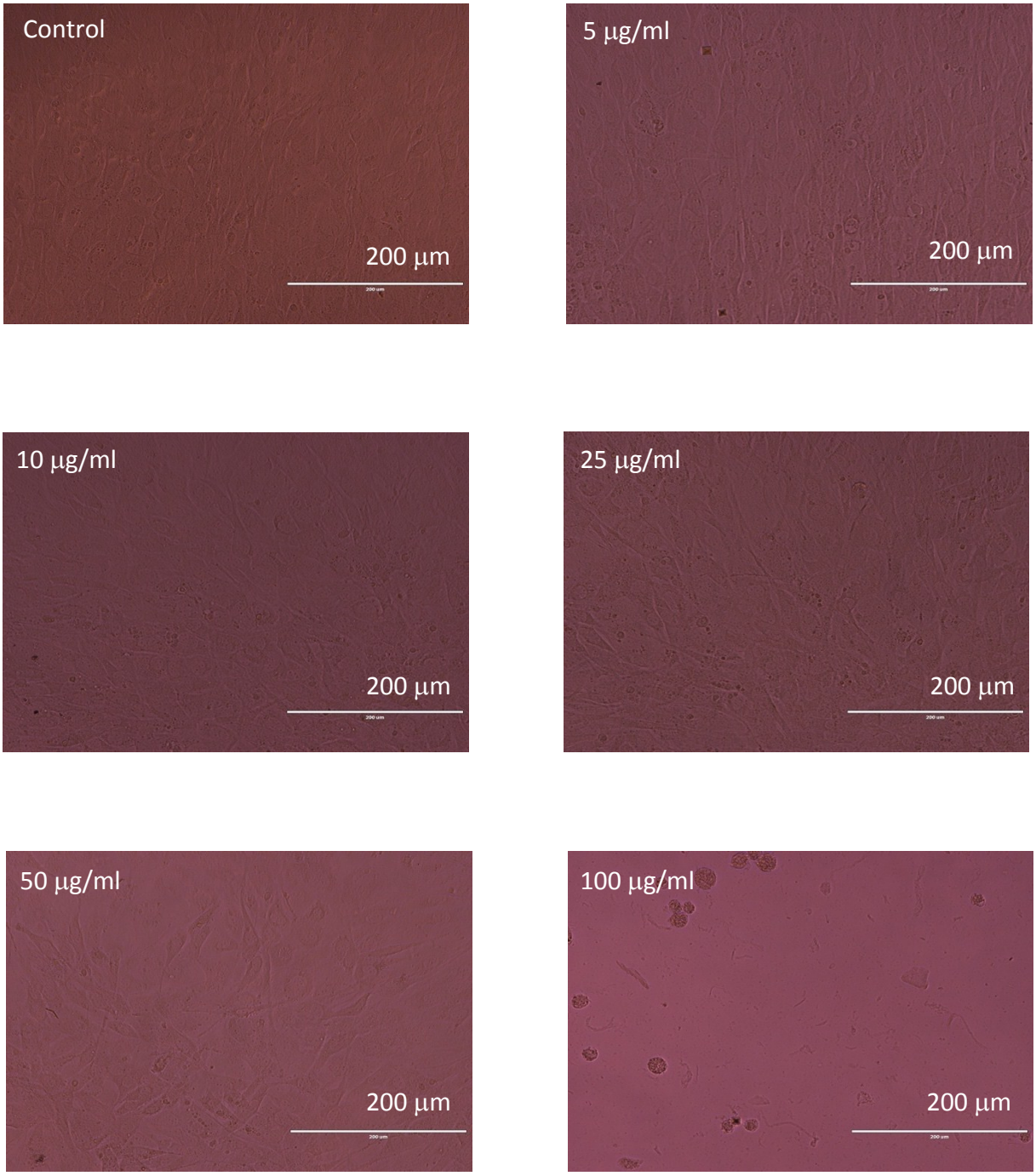


Figure S6. Cell morphology evaluation by optical microscopy of MC3T3-E1 pre-osteoblast cells after 10 days of incubation with ZnO NCs.

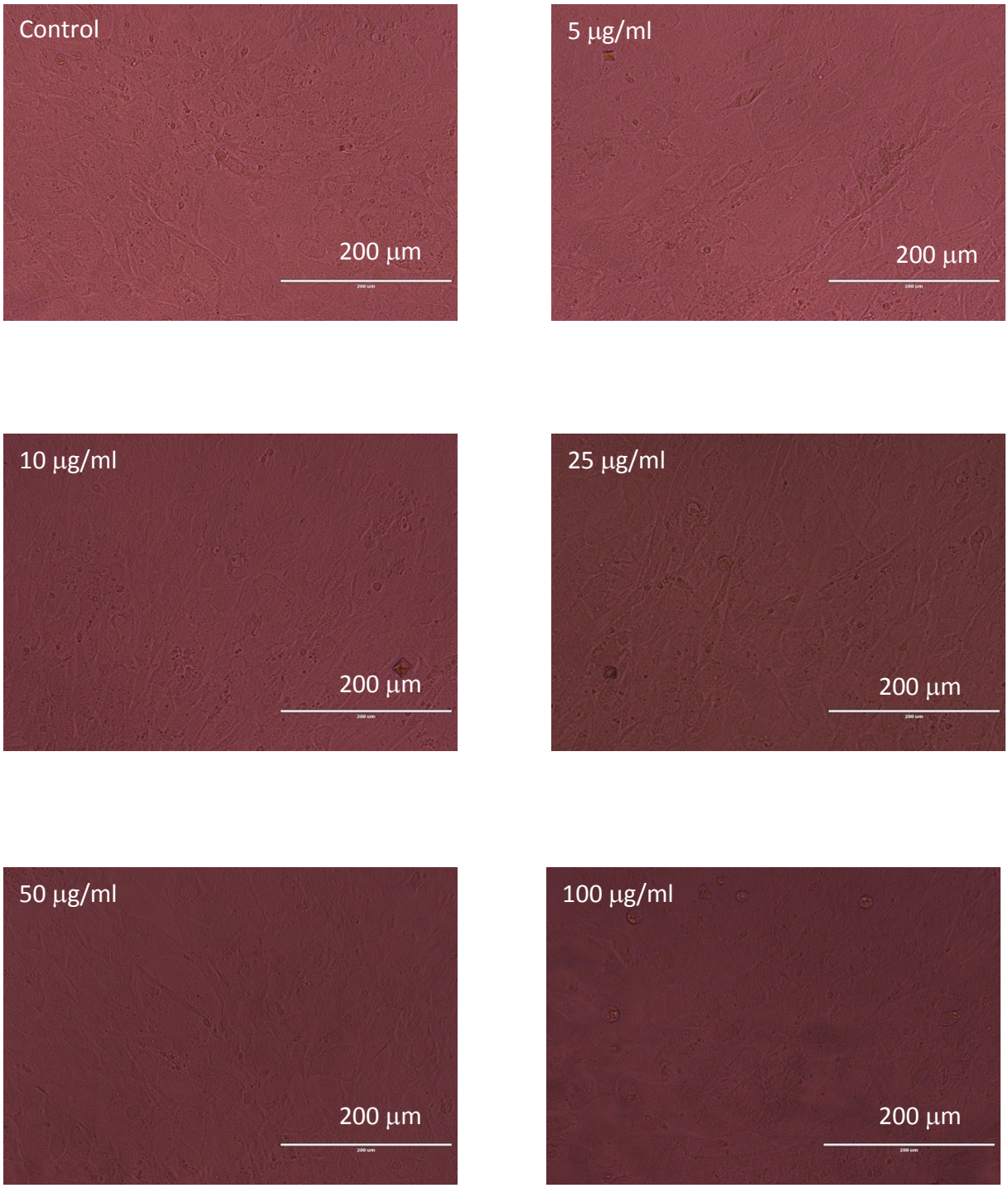


Figure S7. Cell morphology evaluation by optical microscopy of MC3T3-E1 pre-osteoblast cells after 10 days of incubation with ZnO-NH_2 NCs.

Table S1. Reduction in bacteria viability after 24 h of incubation with ZnO and ZnO-NH_2 NCs, respectively. The values shown in bold indicate a CFU concentration less than 10^2 CFU/ml, which is considered as positive effectiveness.

Concentration	***E. coli* vs ZnO**	***E. coli* vs ZnO-NH_2**	***S. aureus* vs ZnO**	***S. aureus* vs ZnO-NH_2**
5 µg/mL	**99.999%**	**100%**	99.923%	99.935%
10 µg/mL	**100%**	**100%**	99.965%	99.963%
25 µg/mL	**100%**	**100%**	**99.985%**	**99.990%**
50 µg/mL	**100%**	**100%**	99.940%	99.853%
100 µg/mL	**100%**	**100%**	99.963%	99.883%