

Supporting data- An *in vitro* and *in vivo* bio-interaction responses and biosafety evaluation of novel Au-ZnTe core-shell nanoparticles

Nanoparticles display unique optical properties that have the potential to interfere with classical photometric cytotoxicity assays such as WST-1, alamar blue, LDH and MTT by either scattering or absorbing light thereby resulting in distorted cytotoxicity results that could reflect false positive or negative data.¹ Additionally the nanoparticles may display a catalytic ability to convert the cytotoxicity assay dye or reagent into its reduced form thereby indicating a false result. In a thorough cytotoxicity evaluation it becomes imperative to investigate the interference effects of the nanoparticles in order to rule out the possibilities of exaggerated data. In this work two tests were performed using WST-1 and Alamar blue assays.² The first test aimed at understanding the interference and interaction effects of Au-ZnTe nanoparticles with the assay dyes or reagents in the absence of cells. The absorbance was recorded and cell culture media and the test dyes were used as controls (Figure 1 and 3). The second test was performed to establish the interference effects of Au-ZnTe nanoparticles with the reduced test dyes from both WST-1 and alamar blue assays i.e formazan and resorufin respectively. The live cells catalysed the formation of the endpoint indicator products formazan and resorufin (Figure 2 and 4). The absorbance of the products was measured after the supernatant containing the reduced dyes were transferred into a new 96 well plate. This was followed by the addition of Au-ZnTe nanoparticles into each well containing the reduced dye and the subsequent absorbance was evaluated to determine nanoparticle interference with the formazan and resorufin products

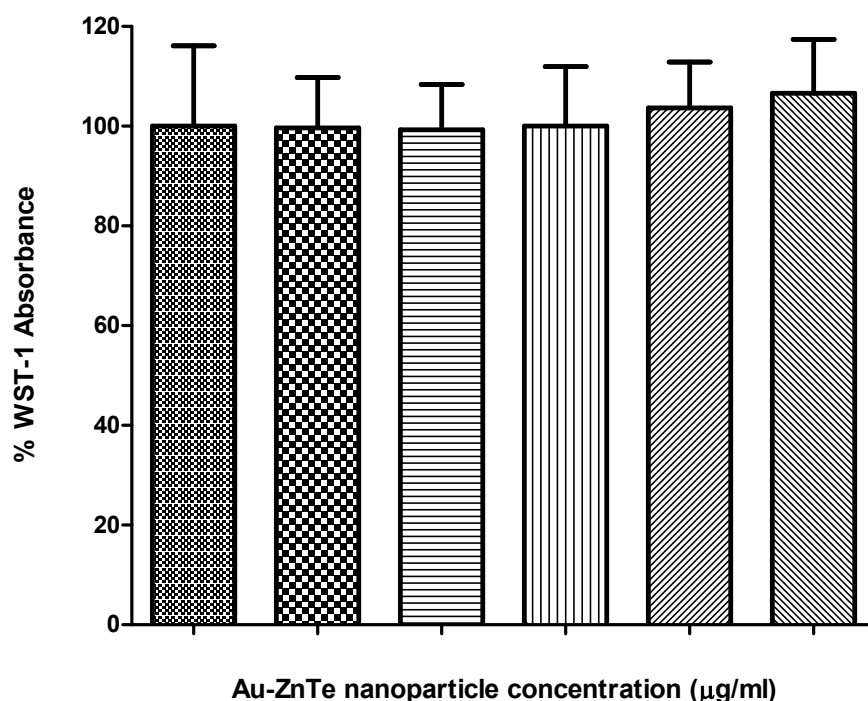


Figure 1. Nanoparticle interference test using WST-1 assay (without cells), n=4

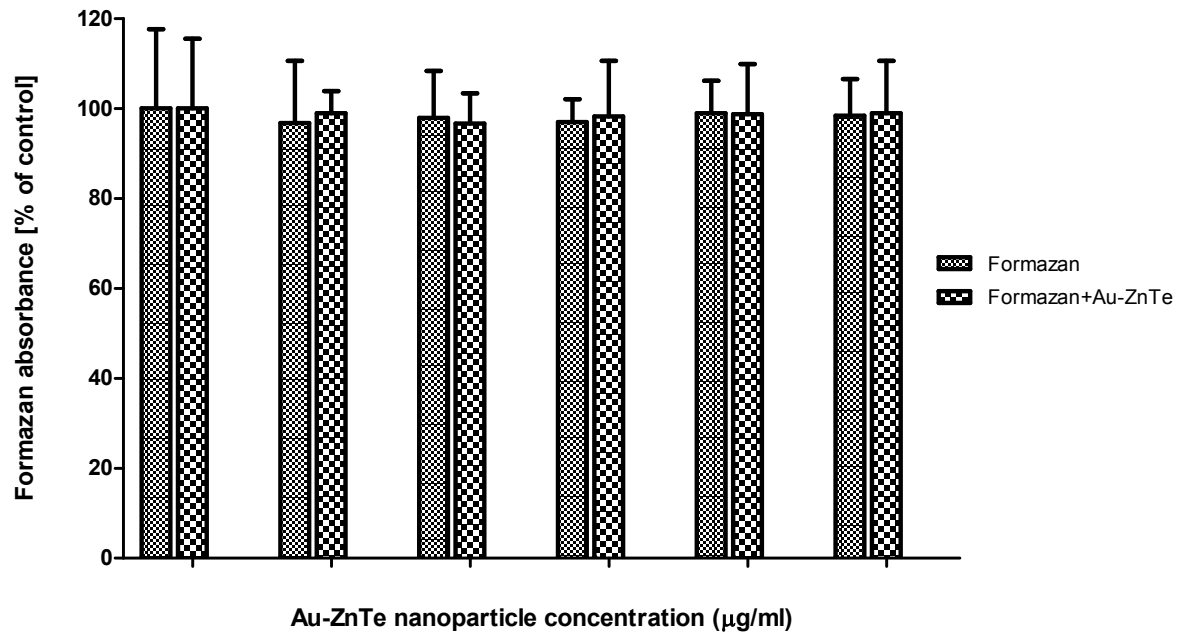


Figure 2. Nanoparticle interference test using WST-1 assay (with MCF7 cells), n=4

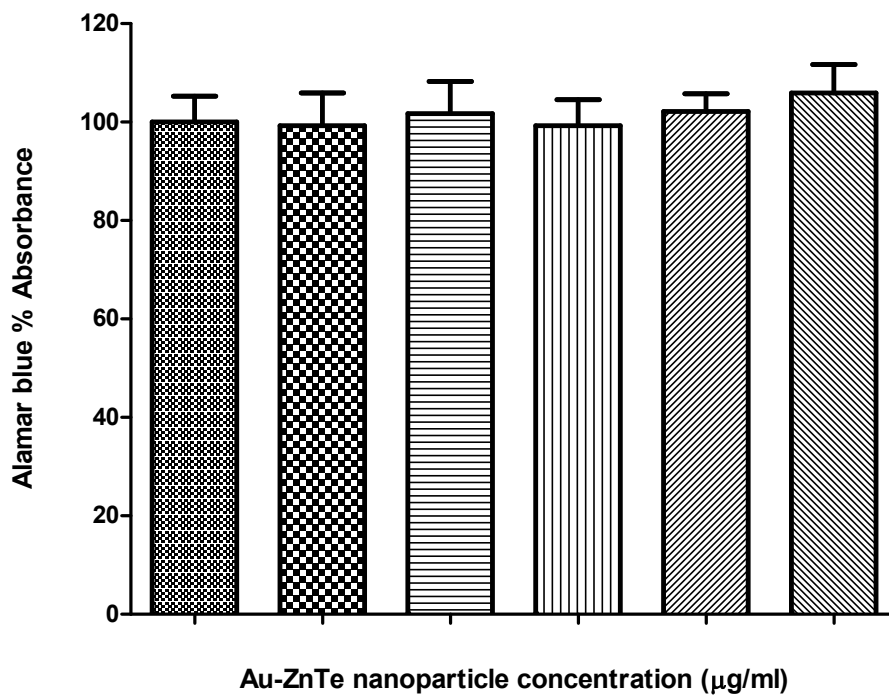


Figure 3. Nanoparticle interference test using alamar blue assay (without cells), n=4

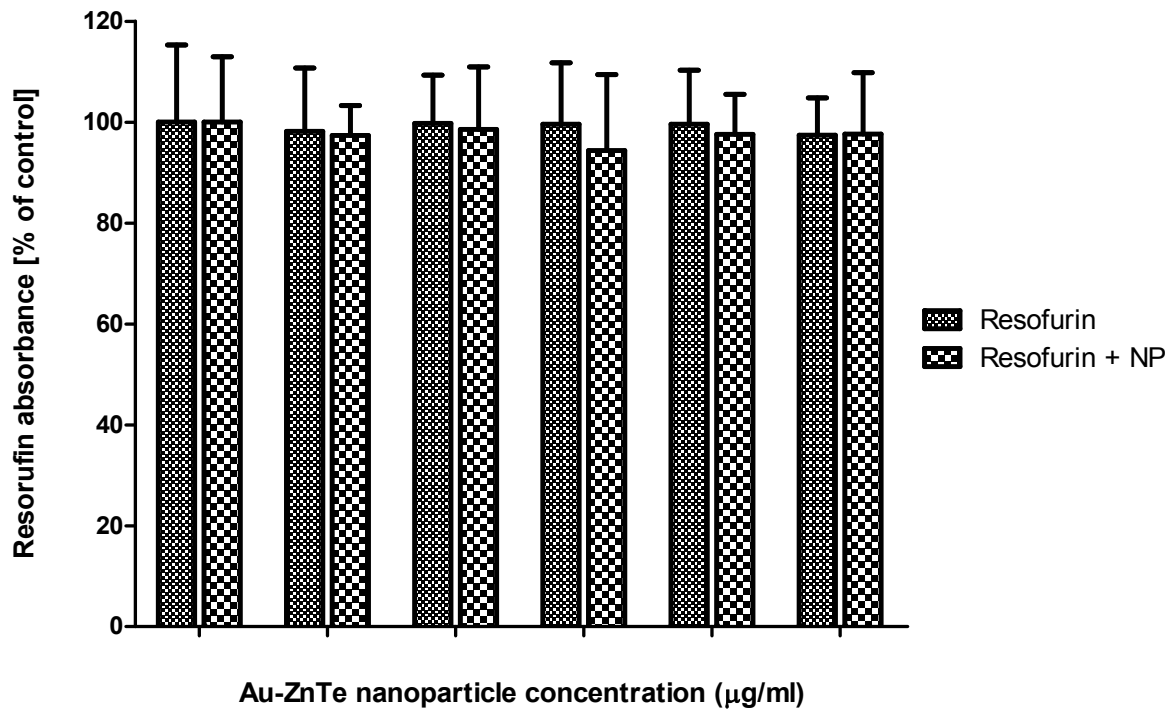


Figure 4. Nanoparticle interference test using alamar blue assay (with PBMC cells), n=4

References

1. Ong KJ, MacCormack TJ, Clark RJ, Ede JD, Ortega VA, Felix LC, Dang MK, Ma G, Fenniri H, Veinot JG, Goss GG. Widespread nanoparticle-assay interference: implications for nanotoxicity testing. *PLoS One*. 2014 Mar 11;9(3):e90650.
2. Park MV, Annema W, Salvati A, Lesniak A, Elsaesser A, Barnes C, McKerr G, Howard CV, Lynch I, Dawson KA, Piersma AH. In vitro developmental toxicity test detects inhibition of stem cell differentiation by silica nanoparticles. *Toxicology and applied pharmacology*. 2009 Oct 1;240(1):108-16.