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Article

Antibacterial Efficacy of ZnO/Bentonite (Clay) Nanocomposites against Multidrug-Resistant *Escherichia coli*

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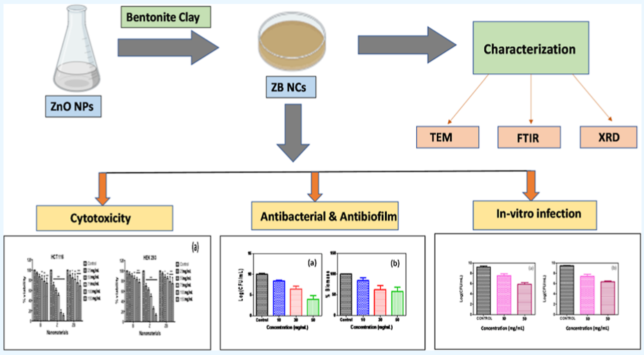
ABSTRACT: The emergence of multidrug-resistant (MDR) bacteria has spurred the exploration of therapeutic nanomaterials such as ZnO nanoparticles. However, the inherent nonspecific toxicity of ZnO has posed a significant obstacle to their clinical utilization. In this research, we propose a novel approach to improve the selectivity of the toxicity of ZnO nanoparticles by impregnating them onto a less toxic clay mineral, Bentonite, resulting in ZB nanocomposites (ZB NCs). We hypothesize that these ZB NCs not only reduce toxicity toward both normal and carcinogenic cell lines but also retain the antibacterial properties of pure ZnO nanoparticles. To test this hypothesis, we synthesized ZB NCs by using a precipitation technique and confirmed their structural characteristics through X-ray diffraction and Raman spectroscopy. Electron microscopy revealed composite particles in the size range of 20−50 nm. The BET surface area of ZB NCs, within a relative pressure (P/P_0) range of 0.407−0.985, was estimated to be 31.182 m^2/g. Notably, 50 mg/mL ZB NCs demonstrated biocompatibility with HCT 116 and HEK 293 cell lines, supported by flow cytometry and fluorescence microscopy analysis. In vitro experiments further confirmed a remarkable five-log reduction in the population of MDR *Escherichia coli* in the presence of 50 mg/mL of ZB NCs. Antibacterial activity of the nanocomposites was also validated in the HEK293 and HCT 116 cell lines. These findings substantiate our hypothesis and underscore the effectiveness of ZB NCs against MDR *E. coli* while minimizing nonspecific toxicity toward healthy cells.



1. INTRODUCTION

The designing, processing, and application of materials in the nanoscale regime has remarkably advanced our gambit against scientific, technological, and social challenges and continues to endure an ever increasing interest.[1,2] Among various nanomaterials, zinc oxide nanoparticles (ZnO NPs), owing to their unique surface chemistry, quantum size dependent properties, and tailorable morphology, have been one of the most widely, scientifically explored, and industrially exploited material.[3,4] ZnO NPs are reported to have unique and remarkable applications in and/or as photocatalysts, optoelectronics, electrical devices, renewable energy devices, etc.[5−7] Moreover, a recent surge of reports and patents regarding the antibacterial potential of the ZnO materials has received ambivalent response from various stake holders.[8−10] The increase in bacterial infection and increasing resistance toward conventional antibiotics can affect public health and cause serious problem. So far, several mechanisms of antibiotic resistance have been discovered which includes the acquisition of transposons or plasmids coding for resistance genes, the upregulation of multidrug efflux pumps that pump out antibiotics, the inactivation of antibiotics by particular enzymes, the sequestration of antibiotics, the thickening of the cell wall preventing antibiotic entry, and the mutation of genes coding for antibiotic target proteins.[11,12] Finding innovative treatments to combat multidrug-resistant (MDR) microorganisms, particularly bacteria, is a crucial concern.[13] Such concerns have further intensified investigation of nanosized ZnO for real life applications.[14] Among various bacteria, *Escherichia coli* although a common flora of human gut, is infamous for causing many common bacterial infections, including cholecystitis, bacteremia, cholangitis, urinary tract infection, traveler's diarrhea, and other clinical complications

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