Comparative assessment of antibacterial efficacy for cobalt nanoparticles, bulk cobalt and standard antibiotics: A concentration dependant study

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Synthesis of compounds that can prevent bacterial resistance is of huge interest and gaining immense popularity. Cobalt (Co) is one of the cheaper transition metals and its nano form has not been studied in details for antibacterial actions. Comparative analysis of Co nanoparticles with bulk Co and standard antibacterials are also lacking. In our study, concentration dependent action of Co nanoparticles was observed from 0.125 to 128.0 µg/ml against *S. aureus* and *E. coli*. Zone of inhibition of Co nanoparticles was better against *E. coli* than *S. aureus*. Co nanoparticles were markedly better than bulk Co, oxytetracycline and gentamicin. Activity index and fold increase of Co nanoparticles were higher at most of the concentrations. In conclusion, Co nanoparticles showed better antibacterial action than other tested compounds against *S. aureus* and *E. coli* particularly at lower concentrations, and their use may be extended in different biomedical fields in future.

**Keywords:** Cobalt nanoparticles, *S. aureus*, *E. coli*, antibacterial activity, activity index, fold increase.

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1. Introduction

There is a continuous battle throughout history between humans and multiple microorganisms. Major discoveries and developments for antibacterial drugs and other ways of infection control during the middle of the 20th century have occurred, which helped significantly for human being and animals. However, rapid emergence of bacterial resistance become as one of the major concerns of the 21st century. Antimicrobial resistance has become a global problem with the fast increase in multidrug-resistant bacteria. *Staphylococcus aureus* (*S. aureus*) and *Escherichia coli* (*E. coli*) are two common bacteria to which almost every human being and animal has been exposed. Some earlier treatable microorganisms are currently becoming untreatable such as methicillin-resistant *Staphylococcus aureus* (MRSA) and vancomycin-resistant enterococcus (VRE) [1]. Different strategies to address this challenge include the designing of improved version of existing antibacterial classes, combinations therapy etc. have been attempted. These strategies can be quite effective, but a high risk of rapid development of bacterial resistance still remains. Bacteria have the genetic capability to transmit and acquire resistance to antimicrobial drugs. In the last three decades, different pharmaceutical industries have produced a lot of new antibiotics, but the bacterial resistance to these drugs has also increased [2]. In view of the increase of bacterial resistance, synthesis of compounds that may prevent bacterial growth with least cytotoxicity are of huge interest and gaining immense popularity in the area of drug development.

During past decades, applications of nanotechnology have brought many breakthroughs. In recent years, much attention has been given to the applications of nanotechnology in various metals and metal oxides for their different uses. Metal nanoparticles are exceptionally appealing better applications in contrast to mass materials due to their high surface-to-volume proportion. There are many nano metals (such as silver, gold, copper, zinc etc.), which exhibit their own structural properties and important biological activities [3, 4]. Metals have been used in the therapy of several diseases of humans and animals since ancient times. Copper and cobalt (Co) ions have been used for centuries by people to inhibit the growth of harmful microbes. Further, transition metals and their complexes are of current interest from numerous point of view, like their use as antioxidant, antibacterial, antifungal, anticancer, anti-inflammatory agent etc. [5, 6].

Co is one of the cheaper transition metals which have wide applications and also possesses antimicrobial properties. Co nanoparticles are of significant interests due to their exceptional magnetic and catalytic properties, and can be used in super alloys, magnetic fluids, computer hard disks, magnetic sensor, catalyst etc [7]. Moreover, Co nanoparticles are also more focused in healthcare system now days due to its antisepctic action. The concentration dependent studies demonstrating antibacterial potentials of Co nanoparticles are very limited, and the studies on concentration dependent comparative efficacy for antibacterial actions of Co nanoparticles with respect to its bulk form as well as standard antibiotics are completely lacking to the best of our knowledge. In this context, we focused the attention on the synthesis of Co nanoparticles to determine their antibacterial efficacy against *S. aureus* and *E. coli* at different

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concentrations, and their comparative analysis of antibacterial potentials with respect to bulk Co, oxytetracycline and gentamicin at different concentrations.

2. Materials and methods

2.1. Materials used

The different chemicals of analytical grade were used for this investigation. Cobalt chloride hexahydrate (CoCl$_2$·6H$_2$O), hydrazine hydrate, sodium hydroxide and ethylene glycol were used for the synthesis of Co nanoparticles/ nanofluids and purchased from Sigma Aldrich, USA. Dimethyl sulfoxide (DMSO) was purchased from SRL. Bulk Co powder was purchased from Central Drug House (CDH), New Delhi, India. Oxytetracycline and gentamicin were procured from Hi-media, Mumbai. The Muller-Hinton Agar (MHA), Muller-Hinton Broth (MHB) and nutrient broth (NB) were purchased from Hi-media, Mumbai, India and used for antibacterial studies. The *S. aureus* (MTCC 1430) and *E. coli* (MTCC 2127) were two bacterial strains in used in this study. These were gifted from the Department of Biotechnology, University of Jammu and revived by using the nutrient broth (NB).

2.2. Synthesis and characterization of Co nanoparticles

The synthesis of cobalt nanoparticles was carried out by reducing a cobalt(II) salt using hydrazine hydrate as reducing agent according to the method of Zhu et al. [8] with some modifications. In a typical procedure, cobalt chloride (10 g) was dissolved into 150 ml ethylene glycol and water mixture (2:1). The solution was stirred vigorously until it completely dissolves. The pH of the solution was adjusted to 12 using aqueous NaOH solution. The solution of CoCl$_2$ was then treated with the mixture of hydrazine hydrate (50 %). After about 30 min, black solid particles appeared. After the completion of reaction, the reaction mixture was centrifuged. The black particles collected were washed several times with triply distilled water and absolute ethanol to remove hydrazine, sodium, and chlorine ions. The final product was then dried in an oven at 60°C.

The reduction reaction can be expressed as:

$$2\text{Co}^{2+} + \text{N}_2\text{H}_4 + 4\text{OH}^- \rightarrow 2\text{Co} + \text{N}_2 + 4\text{H}_2\text{O}.$$  

The characterization studies of synthesized nanoparticles were conducted by using various standard techniques like particle size analyzer (PSA), transmission electron microscopy (TEM) and scanning electron microscopy (SEM). The particle size distribution (PSD) study of the synthesized nanoparticles was conducted by using Malvern Instruments Zetasizer Nano-ZS instrument. The evaluation of the size and morphology of Co nanoparticles was done by using transmission electron microscope (Hitachi H-7500). The scanning electron microscope (SEM-EDS) using SEM (JEOL JSM – 6390LV) was used for the analysis of surface morphology of the Co nanoparticles.

2.3. Assessment of antibacterial efficacy

All the antimicrobial activity assays were performed in triplicate and confirmed by three independent experiments. The antimicrobial activity of synthesized Co nanoparticles and its comparison with oxytetracycline, gentamicin and bulk Co was investigated against the gram-positive i.e. *S. aureus* and gram-negative i.e. *E. coli* bacterial strains. The in *vitro* antibacterial activity was conducted as per agar well diffusion method by using Muller-Hinton agar. The overnight bacterial cultures (108 cfu/ml) were used for seeding of the autoclaved (at 121 °C and 15 lbs) Mueller Hinton agar medium. Seeded agar was later on poured in glass petri plates(30 ml) and allowed for its solidification. Six wells of 6 mm diameter were created into the agar plates and sealed with one drop of 0.8 % melted agar to prevent the leakage of tested compounds. Oxytetracycline, gentamicin, bulk Co and Co nanoparticles were poured (30 µl of each dilution) in the different wells and incubated at 37 °C for 24 hrs. The sensitivities of the bacterial strains to the different concentrations of tested compounds were indicated by clear zone around the wells. Diameters of zone of inhibition produced by of different concentrations of tested compounds were measured to quantify the antibacterial potentials. In present study, DMSO was used as negative control (NC), and oxytetracycline and gentamicin were used as positive control to compare the efficacy of the nanoparticles. Activity index (AI) and fold increase for the comparison of antibacterial potentials of Co nanoparticles with bulk Co, oxytetracycline and gentamicin was also calculated by using the values of zone of inhibition.

$$\text{Activity Index (AI)} = \frac{\text{Inhibition zone of the Co nanoparticles}}{\text{Inhibition zone of the bulk Co or oxytetracycline or gentamicin}},$$

$$\text{Fold increase (}) = \left( \frac{b - a}{a} \right) \times 100,$$

where, $a$ and $b$ refer to the inhibition zones of oxytetracycline/gentamicin/bulk Co and Co nanoparticles, respectively.
3. Results and discussion

Figure 1A is representing the PSD of Co nanoparticles. Sample of Co nanoparticles gives an overall z-average size of 145.6 nm with polydispersity index of 0.372, which revealed that the distribution consists of a single size mode without aggregates. TEM (Fig. 1B) and SEM (Fig. 1C) micrographs of Co nanoparticles showed that synthesized Co nanoparticles have spherical morphology and average size of 13 – 25 nm. SEM image also indicated that the prepared nanoparticles are of spherical shape. SEM picture specifies that individual nanoparticles are aggregated and give large sphere like cumulative structure.

In this study the synthesis of cobalt nanoparticles was followed as per the Zhu et al. method [8] with some modifications. But, the size of Co nanoparticles in our study was very smaller than the size observed in the study of Zhu et al. [8] i.e. 1 to 2 µm. This variation in size in our and study of Zhu et al. [8] was might be due to the variations in the reaction media. The pure ethylene glycol was used as solvent in the study of Zhu et al. [8], whereas ethylene glycol and water mixture (2:1) was used in our study. Previous studies have also reported that solvents usually influence the growth and the morphologies of crystals, and even the formation and the structure of crystals [9–11], because the solvents can selectively control the surface energy of different crystallographic faces [12, 13].

Figure 2 is showing the representative images of bacterial culture plates with zone of inhibition produced by different concentrations of oxytetracycline, gentamicin, bulk Co and Co nanoparticles against S. aureus and E. coli. Table 1 shows the mean value of diameters of zone of inhibition for oxytetracycline, gentamicin, bulk Co and Co nanoparticles at different concentrations against both bacteria. In many of the earlier studies, both S. aureus and E. coli are routinely used to evaluate the antimicrobial activities of diverse nanoparticles [14, 15]. We also selected these two bacterial strains and found that synthesized Co nanoparticles produced larger zone of inhibition against E. coli than S. aureus at the same concentration. This indicated that action of Co nanoparticles is stronger against E. coli than S. aureus.
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*FIG. 2.* Zone of inhibitions of oxytetracycline (A & E), gentamicin (B & F), bulk Co (C & G) and Co nanoparticles (D & H) against S. aureus (A–D) and E. coli (E–H) at different concentrations i.e. (1) 128.0 µg/ml, (2) 32.0 µg/ml, (3) 8.0 µg/ml, (4) 2.0 µg/ml, and (5) 0.5 µg/ml. NC represents negative control i.e. DMSO

**TABLE 1.** Zone of inhibition for oxytetracycline, gentamicin, bulk Co and Co nanoparticles (NPs) against *S. aureus* and *E. coli* at different concentrations

| Conc. (µg/ml) | Zone of inhibition (mm) against *S. aureus* | Zone of inhibition (mm) against *E. coli* |
|---------------|--------------------------------------------|------------------------------------------|
|               | Oxytetracycline | Gentamicin | Bulk Co | Co NPs | Oxytetracycline | Gentamicin | Bulk Co | Co NPs |
| 128.0         | 20.92           | 17.33      | 9.50    | 20.17   | 19.33           | 16.83      | 11.33   | 21.50  |
| 64.0          | 19.17           | 14.17      | 9.33    | 18.00   | 18.50           | 13.83      | 10.67   | 19.33  |
| 32.0          | 17.17           | 13.17      | 9.25    | 17.00   | 16.67           | 12.67      | 9.83    | 18.17  |
| 16.0          | 16.17           | 11.83      | 9.00    | 15.83   | 15.33           | 11.33      | 9.50    | 17.00  |
| 8.0           | 15.42           | 10.92      | 8.92    | 15.33   | 14.42           | 10.33      | 9.33    | 15.75  |
| 4.0           | 14.17           | 10.58      | 8.67    | 13.67   | 12.83           | 9.17       | 9.25    | 15.00  |
| 2.0           | 13.00           | 10.08      | 8.50    | 12.83   | 12.17           | 8.17       | 9.00    | 14.17  |
| 1.0           | 8.50            | 8.17       | 8.33    | 11.75   | 8.17            | 7.17       | 8.92    | 13.33  |
| 0.5           | 0.00            | 0.00       | 8.08    | 10.67   | 0.00            | 0.00       | 8.67    | 11.92  |
| 0.25          | 0.00            | 0.00       | 7.92    | 9.33    | 0.00            | 0.00       | 8.50    | 11.17  |
| 0.125         | 0.00            | 0.00       | 7.50    | 8.50    | 0.00            | 0.00       | 8.25    | 9.67   |

Growth and survival of bacteria is also affected by size, morphology and concentration of nanoparticles. Previous studies have suggested that metal and metal oxide nanoparticles having 1 - 100 nm size and different shapes might have promising antimicrobial actions [16]. Thus, size of 13 – 25 nm and spherical shape of our synthesized Co nanoparticles revealed that our Co nanoparticles might be considered a promising antimicrobial candidate. The smaller size of the metal nanoparticles than the pores of bacterial cell membrane helps the easy transmission of nanoparticles across the cell wall to the cytoplasm, which further causes cell death [17]. Thus, the potent antibacterial potentials of Co nanoparticles might be attributed to their relatively smaller sizes and high amount of surface-area-to-volume ratio which facilitated close interaction with the bacterial membranes [18]. Earlier, it has been also reported that inhibition of bacterial growth increases with the increase in concentration of nanoparticles [19]. In our study, concentration dependent increase in diameter of zone of inhibition for Co nanoparticles was observed only up to 128.0 µg/ml against both bacteria, and beyond this concentration, the size of the zone started to decrease. This might be due to the agglomeration of nanoparticles beyond these concentrations. Previously, it has been reported that optimum concentration exist for nanoparticles at which higher effects are noticed in comparison to more or less concentration [20]. Thus, in
this study, 128.0 μg/ml was the optimum concentrations for Co nanoparticles against S. aureus and E. coli. Oxytetracycline, gentamicin and bulk Co showed increases in diameters of zones of inhibition in concentration dependent manners against both bacteria at all the tested concentrations. Oxytetracycline and gentamicin did not show zone of inhibitions at lower concentrations, i.e., below 1.0 μg/ml against S. aureus and E. coli (Table 1). Thus, oxytetracycline and gentamicin were considered ineffective at lower concentrations. However, synthesized Co nanoparticles of present study were effective below these concentrations and produced marked zone of inhibition up to 0.125 μg/ml. The bulk Co also showed some antibacterial activity at all the tested concentrations, but the sizes of the zones of inhibition produced by it were of smaller size in comparison to it nano form. These results suggested that the range of inhibitory concentration for nano Co was broader with respect to oxytetracycline and gentamicin. Nano form of Co possesses much better antibacterial efficacy in comparison to its bulk form. The markedly better antibacterial effects of nano form of Co particularly at lower concentrations than other tested compounds of present study also revealed that Co nanoparticles may provide antibacterial action for longer duration with less frequency of applications. In the coming years, Co nanoparticles may be considered as a substitute for oxytetracycline, gentamicin and bulk Co in different fields in relation to reduce the occurrence of antibacterial resistance.

Earlier studies have also shown the antibacterial actions of Co nanoparticles prepared by different methods. Environmentally-benign synthesis of Co nanoparticles by using secondary metabolites from Celosia argentea plant extract have showed antibacterial activity against E. coli and Bacillus subtilis (B. subtilis) [21]. Studies have revealed that Co nanoparticles possesses potential for the development of antimicrobial nanomedicine. In a recent study, different varieties of Co nanoparticles synthesized by hydrothermal and ultrasonication methods have been found effective against amoeba species like Acanthamoeba castellani (causative agent for threatening Acanthamoeba keratitis and granulomatous amoebic encephalitis) [22]. The results of this study revealed that cobalt phosphate Co₃(PO₄)₂ hexagonal microflakes, and 100 nm large cobalt hydroxide (Co(OH)₂) nanoflakes showed potent amoebicidal activity as compared to granular cobalt oxide (Co₃O₄). Further, other types of Co based nonmaterials have also shown the antimicrobial actions. Cobalt oxide (Co₂O₃) nanoparticles have revealed the concentration (1, 50 and 100 mg/ml) dependent antibacterial activity against different bacterial strains like E.coli, Pseudomonas aeruginosa (P. aeruginosa) and B. subtilis [23]. Cobalt ferrite (CoFe₂O₄) nanoparticles have shown antibacterial activity against E. coli and its minimum inhibitory concentration was found at 50 mg/L [24]. A comparative study of cobalt ferrite (CoFe₂O₄) and silver-cobalt ferrite (Ag–CoFe₂O₄) nanoparticles, synthesized by wet ferritization and self-combustion methods using Hibiscus rosa-sinensis flower/leaf extracts, showed inhibitory actions against the different strains of bacteria and fungus, and the Ag–CoFe₂O₄ nanoparticles revealed better inhibitory potentials than CoFe₂O₄ nanoparticles obtained by either of the two methods [25].

The surface of the nanoparticles has a significant effect in their bioactivity and materials made up of nanoparticles have a much greater surface area per unit volume as compared to the materials made up of bigger particles (bulk materials). This makes nanoparticles more reactive and interactive to cell wall/membrane, which leads to their impairment and results in leakage of cellular contents [26]. Previous study has revealed that the antibacterial activity of the nanomaterials increased with an increase in the surface-to-volume ratio due to the decrease in size of nanoparticles, and the size of the nanoparticles might play a role in the antibacterial activity [17]. Although, the exact mechanisms by which the metallic nanoparticles produce antibacterial actions are not clear and still under debate. Some possible mechanisms are supposed by which they produce actions include, interaction of nanoparticles with the cell wall/membrane, formation of reactive oxygen species (ROS), release of metal ions etc. Electrostatic attraction, vander waals forces, receptor – ligand interactions and hydrophobic interactions are considered different ways which promote nanoparticles and bacterial contact [27–30]. Nanoparticles can cross microbe membranes after making contact and interfere with different bacterial metabolic pathways, and also induce changes in membrane shape and permeability. Nanoparticles inside cells interact with the different microbial components and cause inhibition of enzymes, deactivation of proteins, production of oxidative stress, modulate the gene expression and electrolyte imbalance [31]. Previous studies have revealed that different metal nanoparticles produce various types of ROS, which resulted in their antibacterial actions [32]. Different types of ROS are observed to inhibit transcription, translation, enzymatic activity, and the electron transport chain in bacteria [33, 34]. Metal ions also have tendency to attach and destroy the DNA molecule after entry into the bacterial cell [26]. It has been observed that metallic nanoparticles have enhanced potential to combat bacterial infections as compared to their salts, and the size of nanoparticles mostly influences the antibacterial mechanism [33, 35–37]. The fact that small sized nanoparticles tend to be more toxic to bacteria than large nanoparticles and their bulk form is due to the larger surface area to volume ratio of small nanoparticles [38]. So, the antibacterial actions by Co metal nanoparticles in this study might be due through different ways, which could be the reason for their better effects as compared to standard antimicrobials.

The calculated values of activity index (AI) as well as fold increase (%) are used to analyze the comparative antimicrobial efficacy of tested compound with respect to other compound [39]. The tested drug/compound can be
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considered better against a particular bacterial strain, if its AI value is more than one. In our study, the AI for Co nanoparticles with respect to (w.r.t.) oxytetracycline, gentamicin and bulk Co against *S. aureus* and *E. coli* were calculated at various concentrations, and the values are presented in Table 2. The AI of Co nanoparticles was higher than 1 w.r.t. oxytetracycline up to 1.0 µg/ml against *S. aureus* and at all the tested concentrations against *E. coli*. The AI of Co nanoparticles w.r.t. gentamicin and bulk Co was higher than 1 at all the tested concentrations against both bacterial strains. The calculation of fold increase (%) is another approach to determine the relative effect of the tested compound in contrast to other compounds. The positive value of fold increase reveals the better potentials of tested compound. However, negative value of fold increase reveals lesser efficacy of the tested compound than other compound against a particular bacterial strain at the same concentration. The Tables 3 present the values of fold increase for Co nanoparticles w.r.t. oxytetracycline, gentamicin and bulk Co against *S. aureus* and *E. coli* at different concentrations. We observed in this study that the positive values of fold increase of Co nanoparticles w.r.t. other tested compounds against both bacterial strains supported the observations of AI. These findings are suggesting that Co nanoparticles may have better antibacterial potentials, particularly at lower concentrations, than other tested compounds of present study. So, Co nanoparticles may be used for applications/coating of different larger surface area, cellulose bandages, uniforms, bed linen, medical equipment, different utensils of medical fields etc., which may be useful as preventive approach to control microbial infections. This will make a substantial impact to reduce the hospitalized bacterial contamination, mortality, costs of treatment etc.

**TABLE 2.** Activity index of Co nanoparticles with respect to (w.r.t.) oxytetracycline, gentamicin and bulk Co against *S. aureus* and *E. coli* at different concentrations

| Conc. (µg/ml) | Activity Index w.r.t. Oxytetracycline | Activity Index w.r.t. Gentamicin | Activity Index w.r.t. bulk Co |
|---------------|--------------------------------------|---------------------------------|-------------------------------|
|               | *S. aureus* | *E. coli* | *S. aureus* | *E. coli* | *S. aureus* | *E. coli* |
| 128.0         | 0.96       | 1.11      | 1.16       | 1.28      | 2.12       | 1.90      |
| 64.0          | 0.94       | 1.05      | 1.27       | 1.40      | 1.93       | 1.81      |
| 32.0          | 0.99       | 1.09      | 1.29       | 1.43      | 1.84       | 1.85      |
| 16.0          | 0.98       | 1.11      | 1.34       | 1.50      | 1.76       | 1.79      |
| 8.0           | 0.99       | 1.09      | 1.40       | 1.52      | 1.72       | 1.69      |
| 4.0           | 0.96       | 1.17      | 1.29       | 1.64      | 1.58       | 1.62      |
| 2.0           | 0.99       | 1.16      | 1.27       | 1.73      | 1.51       | 1.57      |
| 1.0           | 1.38       | 1.63      | 1.44       | 1.86      | 1.41       | 1.50      |
| 0.5           | –          | –         | –          | –         | 1.32       | 1.38      |
| 0.25          | –          | –         | –          | –         | 1.18       | 1.31      |
| 0.125         | –          | –         | –          | –         | 1.13       | 1.17      |

Studies of last few years have showed that the applications of nanoforms of various metals like Ag, Cu, Ti or Zn etc has been augmented due to their bactericidal action against different bacterial strains. Nanoparticles of Ag, Cu, Zn etc. are commonly and widely employed for different purposes like bactericides in catheters, burn wound care, dental practices, antibacterial soaps etc. [40]. However, use of Ag nanoparticles has started to decline due to biological safety of Ag nanoparticles, pigmentation effect of Ag nanoparticles on teeth etc [41]. The Co, metal used in this study, is considered cheaper than silver. In the future, Co nanoparticles may replace the silver nanoparticles for various applications. Additionally, metal nanoparticles are considered better antimicrobial pharmaceuticals in comparison to organic nanoparticles due to their durability, high stability, lower mammalian cell toxicity etc. [42]. The promising antimicrobial efficacy revealed by Co nanoparticles might be very useful for several therapeutic applications in biomedical sciences. Applications of Co nanoparticles may also be extended to food industry, textile industry, nursing homes, water purification, air purifications, paint industry, sewage treatment, etc. There may also be new construction of numerous materials such as antibacterial plastics, antibacterial ceramics, antibacterial clothes, antibacterial stainless steels etc. by using Co nanoparticles. Moreover, applications of Co nanoparticles coated materials in hospitals
TABLE 3. Fold increase (%) of Co nanoparticles with respect to (w.r.t.) oxytetracycline, gentamicin and bulk Co against S. aureus and E. coli at different concentrations

| Conc. (µg/ml) | Fold increase (%) w.r.t. Oxytetracycline | Fold increase (%) w.r.t. Gentamicin | Fold increase (%) w.r.t. bulk Co |
|--------------|------------------------------------------|-----------------------------------|---------------------------------|
|              | S. aureus (E. coli)                       | S. aureus (E. coli)                | S. aureus (E. coli)              |
| 128.0        | -3.59 (11.21)                             | 16.35 (27.72)                      | 112.28 (89.71)                  |
| 64.0         | -6.09 (4.50)                              | 27.06 (39.76)                      | 92.86 (81.25)                   |
| 32.0         | -0.97 (9.00)                              | 29.11 (43.42)                      | 83.78 (84.75)                   |
| 16.0         | -2.06 (10.87)                             | 33.80 (50.00)                      | 75.93 (78.95)                   |
| 8.0          | -0.54 (9.25)                              | 40.46 (52.42)                      | 71.96 (68.75)                   |
| 4.0          | -3.53 (16.88)                             | 29.13 (63.64)                      | 57.69 (62.16)                   |
| 2.0          | -1.28 (16.44)                             | 27.27 (73.47)                      | 50.98 (57.41)                   |
| 1.0          | 38.24 (63.27)                             | 43.88 (86.05)                      | 41.00 (49.53)                   |
| 0.5          | – (–)                                    | – (–)                              | 31.96 (37.50)                   |
| 0.25         | – (–)                                    | – (–)                              | 17.89 (31.37)                   |
| 0.125        | – (–)                                    | – (–)                              | 13.33 (17.17)                   |

may help reduce the prevalence of nosocomial infections. The utilization of Co nanoparticles coated materials in many industries may also be beneficial to check bacterial deterioration of material. Combination therapy of metal nanoparticles with antibiotics has showed some synergistic effects [43, 44]. Therefore, use of combination therapy of Co nanoparticles with standard antibiotics against simple or mixed type microbial infections may be extended for the formulations of more efficacious preparations.

4. Conclusion

In conclusion, the Co nanoparticles possess much better antibacterial efficacy than its bulk form and are also better than standard antibacterials, particularly at lower concentrations, against S. aureus and E. coli. In future, the strong antibacterial potentials of Co nanoparticles may extend its applications in different fields and could create a great revolution in the field of health care.

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