Remittances Review
July 2024,
Volume: 9, No: S 3, pp.1204-1231
ISSN: 2059-6588(Print) | ISSN 2059-6596(Online)

Received: 17 June 2024, Accepted: 18 July 2024
DOI: https://doi.org/10.33282/rr.vx9i2.58

Optimization of Cobalt Oxide Nanoparticles via Aqueous Synthesis and Evaluation of Antimicrobial Potential Against *Staphylococcus haemolyticus* and *Staphylococcus aureus*

Muhammad Umair Akhtar[1], Muhammad Shafiq[2], Madeeha Shakeel[1], Muhammad Imran Alvi[3], Misbah Akbar[4], Kaynat Akber[5], Hasnain Zafar[5], Humayoun Islam Aswad[6], and Muhammad Khuram Sajjad[1]

[1]Department of Zoology, University of Okara, Okara Pakistan
[2]Department of Zoology, University of Education, Township Lahore, Pakistan
[3]Department of Physics, University of the Punjab, Lahore Pakistan
[4]Department of Zoology, Ghazi University, DG Khan Pakistan
[5]Institute of Zoology, Bahauddin Zakariya University, Multan Pakistan
[6]Department of Zoology, University of Education Lahore, Multan Campus

Abstract

This study aims to investigate the synthesis and antimicrobial properties of cobalt oxide nanoparticles (Co3O4 NPs). Due to their small size and magnetic characteristics, nanoparticles show promise across applications like biomedical uses. Co3O4 NPs were synthesized using a green chemistry method involving cobalt chloride, trisodium citrate, and leaf extract from Phytolacca dodecandra. X-ray diffraction and Fourier transform infrared spectroscopy were used to characterize the crystalline structure and functional groups. Scanning electron microscopy revealed a cubic spinel structure with porous morphology. Antimicrobial efficacy against clinically isolated Staphylococcus aureus and S. haemolyticus was determined using disc diffusion and well diffusion assays. Minimum inhibitory concentration values indicated S. haemolyticus was more susceptible to Co3O4 NPs than S. aureus. Optimization of nanoparticle synthesis and examination of antibacterial mechanisms against drug-resistant pathogens can contribute to new therapeutic modalities. Understanding interactions between nanomaterials and biological systems also supports customized applications in fields like targeted drug delivery.

Keywords: Cobalt Oxide, Nanoparticle, X-ray, Haemolyticus, Staphylococcus

Remittances Review
July 2024,
Volume: 9, No: S 3, pp.1204-1231
ISSN: 2059-6588(Print) | ISSN 2059-6596(Online)

INTRODUCTION

Nanoparticles may exhibit novel characteristics not seen in bulk materials, according to research, because of their very small size and high surface-to-volume ratio (Jamuna et al., 2023). As a bridge between the bulk and the atomic level, nanoparticles are recognized to have distinct chemical, electrical, and magnetic property sets (Madkour & Madkour, 2019). The utilization of these systems in pigments in paints, magnetic tapes, ferrofluids, magnetic refrigerants, colour imaging, catalysts, drug delivery systems, etc., creates an immediate technological push (Behera & Behera, 2022; Bilal[a,b], 2021).

Cobalt, a transition metal (d-block), finds its most common use in the ceramics and paints sectors. Dehydrogenation, hydrogenation, and hydroformylation reactions are only a few of the many organic processes that make use of its constituents as catalysts. Cobalt has several potential uses as a catalyst, nano-sensor, or component in nanoelectronic systems due to its unique electrical, catalytic, magnetic, and optical characteristics.

In the past, cobalt and its three metastable phases—the epsilon, face-centered cubic, and hexagonal tightly packed—had a major impact as ferromagnetic metals (Iravani & Varma, 2020). Cobalt nanoparticles (NPs) may be synthesized using several well-established physicochemical techniques. Some examples of such approaches are solvothermal processes, hydrothermal treatment decomposition, liquid-phase reduction, and template-based procedures (Ranjbar Bahadori et al., 2020).

These NPs have a wide range of important uses, including as photocatalysts, field effect transistors, energy storage, anode materials for rechargeable Lion batteries, solar cells, and electrochromic sensors. Because of their exceptional biological applications, these NPs also possess antioxidant, antifungal, antibacterial, anticancer, and enzyme-inhibitory characteristics. Although cobalt is an essential element, mammals only have limited amounts of it in their bodies, and they typically receive it from eating grains and green vegetables (Islam et al., 2023).

Its involvement in cobalamin, vitamin B12, and a limited number of other cobalt-containing enzymes that have been discovered so far are important aspects of its biological activity (Balabanova et al., 2021). Owing to cobalt's significance in human metabolism, cobalt-based

materials appropriate for biomedical applications are becoming more and more significant (Vodyashkin et al., 2022).

Cobalt nanoparticles have several applications, including energy storage, microelectronics, pharmaceuticals, contrast agents, and drug delivery systems(Thalji et al., 2021). The ability of cobalt nanoparticles to be magnetic is a crucial component(Oyarzún et al., 2015), This creates new opportunities for the targeted administration of drugs via nanoparticles(Mukherjee et al., 2009).

Furthermore, it should be mentioned that cobalt nanoparticles' small size and physio-chemical characteristics make them useful as sensors for identifying a variety of chemicals(Luyts et al., 2013).

The main techniques for producing cobalt/cobalt oxide nanoparticles and their applications in areas such as biomedicine and catalysis were the subjects of this study. Cobalt nanoparticles are highly sought-after and useful in several fields, such as diagnostics and cancer treatment, and they are also quite easy to obtain. Right now, there's a lot of interest in creating new cobalt nanoparticle production techniques because of how important nanoparticles are in a lot of different areas of human life(Gaur et al., 2021).

Current techniques for producing cobalt nanoparticles must satisfy several critical requirements to guarantee high process efficiency: 1. A high yield of the desired crop (Xinran Li et al., 2020). The potential to produce nanoparticles with a specific structure (size, shape, and crystallinity). The method's safety and viability of acquisition(Shabatina et al., 2020).

Environmentally friendly methods (Rodríguez De Luna et al., 2020). The ability to scale by using the liquid phase reduction approach, Co NPs were successfully synthesized at room temperature, showing the reducing agent's reaction mechanism as well as the particle formation mechanism. Additionally, it was looked into how citric acid additions affected the size and form of the NPs that were produced(Usami et al., 2021).

Ever since Meiklejohn and Bean discovered it in partially oxidized Co particles in 1956 (Belkacem et al., 2023), There have been reports of exchange bias (EB) in several different

nanostructured systems. It happens when two materials are exchange-coupled at their interface, often one being ferromagnetic and the other antiferromagnetic, and they have considerably differing magnetic anisotropies(Gareeva et al., 2023). Exchange coupling of this kind usually shows up as a variety of phenomena, such as coercivity enhancement and changes of the hysteresis loop along the axis of the applied magnetic field (HE) and the magnetization axis (vertical shift, VS) (Jeon et al., 2022).

Spin valves and other spintronic devices heavily utilize EB. (Ryu et al., 2020), It has enormous promise for making new nanostructured permanent magnets, for example (Mohapatra et al., 2020). As a result, the impact has been thoroughly investigated, mainly about thin films and multilayers(F.-H. Wang et al., 2022), including discontinuous multilayers as well as in nanoparticles made using a range of methods (Anasthasiya et al., 2020), where the use of exchange bias has been suggested as a tactic to stabilize the FM core with an eye toward(Vivek et al., 2023) e.g., to postpone magnetic recording's superparamagnetic limit. Specifically, Co/CoO nanoparticles (NPs) have been investigated quite a bit since conventional equipment can easily perform magnetic characterization at the Curie and Néel temperatures. Cobalt NPs can be partially oxidized to generate (i) core-shell (metallic Co cores surrounded by a shell of native oxide(s)) or entirely oxidized to form cobalt oxides (CoO or Co3O4). (Du et al., 2020), Depending on the particle size, annealing temperature, and oxidant quantity, (ii) hollow or (iii) yolk structures (Xiang et al., 2022).

Reactive gas-phase aggregation can also yield NPs with Co and CoO phase mixing and large exchange bias fields (Ghoshani et al., 2021). To maximize the HE value, the diameter of the Co core and the thickness of the CoO shell are both important. It goes without saying that between the pure Co and CoO extremes (both with 0% bias), there must be an ideal core/shell geometry that varies with oxidation.) (Jaffari et al., 2021) , However, its existence at a certain Co/CoO ratio is associated with intricate aspects of the interface spin structure, which are frequently dictated by the CoO shell's size(Ghoshani et al., 2021) who discovered a critical size (based on energy concerns) below which EB vanishes (Bustamante-Torres et al., 2022), Our study indicates that the spin canting caused by lattice strain is a result of the strain dependence of the respective amplitudes of two distinct antiferromagnetic modulations in the shell, which results in a net

(uncompensated) moment of the (100) planes of the oxide at the core-shell interface(Gogoi et al., 2023).

According to those latter writers, the development of a maximum in the EB field is mostly dependent on the fluctuation of that moment with the shell thickness. Furthermore, we note the emergence of a soft FM phase in the field-cooled hysteresis loops, which results in the shape referred to as "hummingbird-like." (Deltell et al., 2021). Before discarding such interpretations in the context of our current findings, we critically examine earlier hypotheses regarding the phase's origin(Bragazzi et al., 2023).

Globally, infectious diseases rank as the second leading cause of death for people(Bray et al., 2021). Fever, elevated heart rate, fast breathing, anxiety, and insanity are common signs of infectious disorders. In certain cases, unchecked infection conditions might result in sudden death. The mainstay of treatment for severely ill patients with infections, antibiotics have prevented millions of deaths globally (Coates et al., 2020). However, patient compliance frequently prevents antibiotics from being delivered as effectively as they should, which invariably leads to less favourable patient outcomes and medication resistance (Bonkat et al., 2022). The overuse of antibiotics in recent years has led to a widespread development of drug resistance to some routinely used antibiotics, which accelerates the formation of extremely deadly multidrug-resistant mutants (Terreni et al., 2021).

Investigating novel therapeutic approaches is therefore crucial for improving the effectiveness of infectious disease control. In nature, cobalt (Co) and its compounds are extensively spread, and many human activities involve it (Ouyang et al., 2022).

Because cobalt is an essential component of vitamin B12, it is regarded as an essential trace element (González-Montaña et al., 2020). Cobalt has an impact on nerve cell protein and amino acid production as well as fatty acid metabolism. Additionally, cobalt compounds show promise in the medicinal field (Sayadi et al., 2022). Cobalt-based alloys, for instance, have exceptional strength, machinability, corrosion, and wear resistance, and can be utilized to create femoral heads, acetabular liners, femoral components, and stems—the frictional and supporting portions of artificial joints.

Nanoparticles (NPs) are ultra-small particles with a diameter of 1 to 1000 nm. They can be made of various materials, including metals, polymers, and lipids, and each has its benefits for delivery (Wang et al., 2020). Metal nanoparticles (MNPs), one of the most significant catalysts for biomedical applications, have drawn more attention in the last few decades (Bilal et al., 2019). Because of these MNPs' developing qualities, cobalt nanoparticles, or Co NPs, are employed in both the medicinal and technical domains(Flores-Rojas et al., 2022). As an illustration, a study revealed that Co-NPs can cause cell death (Bajpai et al., 2020), It suggests that Co NPs are feasible and relevant as new nanomedicines for possible phototherapy, thermotherapy, and chemotherapy (Xiaokun Li et al., 2022).

Cobalt can boost the production of certain enzymes and improve protein metabolism, which can improve a person's immunity (Ma et al., 2022). Cobalt nanoparticles (Co NPs) are utilized as possible therapeutic agents for the treatment of a variety of infectious disorders due to the metal's promising qualities (Ma et al., 2022). Co NPs have been shown to produce reactive oxygen species (ROS), which are in charge of counteracting the inhibitory effects of Co NPs on certain viruses, fungi, and bacteria. The biological activity and biomedical applications of Co NPs were compiled here, with a focus on their potential for anti-infectious treatment, which could aid in the future development of anti-infection tactics (Jiang et al., 2022).

In rocks, soil, plants, and animals, cobalt is a silvery-grey, glossy, brittle, but durable metal that is found in large quantities in the natural world. With an atomic weight of 58.9, cobalt is a transition metal in the fourth row of the periodic table that is adjacent to nickel and iron (Ma et al., 2022). Cobalt can exist in both organic and inorganic forms and is a necessary trace element for human health (Jenkins et al., 2023). Vitamin B12 and its derivatives, which are the main component of cobalamin and act as cofactors for many different enzymes and protein components, are the most well-known organic forms (Lovelock et al., 2022). Vitamin B12, being a water-soluble vitamin, has the mineral cobalt in its center, arranged in a corrin ring with upper and lower ligands (Pereira et al., 2019).

Because of this, substances having vitamin B12 action are consistently referred to as "cobalamins." (Halczuk et al., 2023) To the best of the author's knowledge, no studies have established the antibacterial impact of Co NPs against all three of the selected bacteria in this

investigation, even though several publications on the destruction of bacteria with Co or cobalt complex NPs are available. Additionally, there are currently insufficient studies comparing the antibacterial properties of Co NPs with widely used antibiotics. To ascertain the physical characteristics and gauge the antibacterial activity of the Co NPs against three bacteria that are resistant to several drugs, the current study was carried out (Abass et al., 2021).

The aims and objectives of this study are to optimizing the synthesis and customizing the unique properties of cobalt nanoparticles is the overarching aim of this research. By precisely controlling characteristics like size, shape, and surface features through adjusted synthesis methods, we seek to enhance magnetic properties, biocompatibility, catalytic activity, and antimicrobial efficacy. Achieving improved size and morphology control at the nanoscale would support nanoparticles designed for tailored applications. Potential outcomes include more effective magnetic materials, catalysts, targeted drug delivery systems, and novel antibacterial therapies. Exploring cobalt nanoparticles' performance across industries, from energy storage and biomedicine to environmental remediation and data storage, could solve scientific and technological problems while advancing disciplines such as materials science and nanotechnology. Realizing their full potential will promote innovation, progress and interdisciplinary collaborations across sectors through customized cobalt nanoparticles optimized for diverse functions.

MATERIALS AND METHODS

Experimental Details

Preparation and Optimization of Cobalt oxide Nanoparticles

With minor adjustments, cobalt oxide nanoparticles will be created at ambient temperature using the simple chemical technique described by (Liang & Zhao, 2012). Initially, 10 ml of deionized water will combine with 0.235 g of tri-sodium citrate trihydrate ($C_6H_5Na_3O_7.3H_2O$) while being constantly stirred at 150 rpm. Subsequently, 0.1 g of sodium borohydride ($NaBH_4$) and 0.2 g of cobalt chloride hexahydrate ($CoCl_2.6H_2O$) will be added simultaneously while being constantly stirred (at 150 rpm). When a significant amount of hydrogen is released during the reduction reaction, a boil in the solution can be observed. After the hydrogen emission ceases, a

magnet will be used to gather the greyish-black powder of nanoparticles, which will then be repeatedly cleaned with ethanol and deionized water before being allowed to dry for 24 hours at room temperature in the open.

At ambient temperature, the magnetic characteristics of cobalt oxide nanoparticles will be investigated with the use of a Lakeshore 7407 Vibrating Sample Magnetometer (VSM).

An X-ray powder diffractometer (D-maxIIA, Rigaku, Japan) will be used for structural analysis and crystallite size determination. The cobalt oxide nanoparticles will be annealed at 500 °C in a controlled environment and then pulverized gently using a mortar and pestle at room temperature before XRD analysis. Within the 2θ range of 20° to 80°, this diffractometer will be run at 30 kV using CuKα radiations with a wavelength (WL) of λ= 1.54060 ″.

Optimization technique

X-ray diffraction (XRD, Bruker D8 Advance, and Cuka) will be used to study the crystal structure of cobalt oxide nanopowders (CO3O4) in two ranges of 20 to 80. The CO3O4 NPs' FTIR spectra will be analyzed using FTIR Shimadzu, and utilizing the KBr sample approach, absorption values will be collected in the range of 4000 cm1 to 400 cm1. A Carl Zeiss SUPRA-55 scanning electron microscope will be used to examine the surface morphology. An EDX (Quantax 200 with X-Flash Bruker) analyzer connected to a Carl Zeiss SUPRA-55 apparatus will be used to determine the chemical composition.

The Jasco V-670-UV visible reflectance spectrometer will be used to study the optical bandgap. Using a 250 nm excitation wavelength, the luminescence spectrometer LS45 captured the sample's emission spectra. An Elico SL-150 spectrophotometer, a UV-Vis spectrophotometer with a diffuse reflectance attachment for powder samples, will be used to measure the optical band gap of the produced Co3O4 nanoparticles. FTIR (Perkin Elmer 65, PerkinElmer, Inc., and Waltham, USA) techniques will be used to characterize both green and chemically generated NPs to gather information about the functional groups.

Antibacterial Activity Test

Preparation of inoculum

The first step involved taking 2.5 mg of nutritional broth, dissolving it in 100 mL of distilled water, and then preparing it in two conical flasks before sterilizing them. A clinically isolated strain of Staphylococcus aureus will be injected into a single conical flask. A clinically isolated strain of Staphylococcus haemolyticus will be introduced to the other conical flask. After being injected with nutritional broth, these bacterial cultures will rotate at 100 rpm for about 24 hours (Chen et al., 2007).

Disc Diffusion Method

Using a modified approach, the disc diffusion method will be used to conduct antibacterial testing on nutrient agar with a suspension of bacteria. Submerging the swab in the bacterial broth culture is the next step. To extract the excess fluid, push the swab gently against the interior of the tube. Grass may be grown by streaking an agar or nutrient agar plate with a swab. If you want your stripes to stand out, try striping the plate in three different directions: diagonally, at an angle to the first, and then second. Subsequently, the swab will be utilized to streak the agar's outside diameter. For 24 hours, the infected plates will be incubated at 37 °C (Mohandes et al., 2023).

Dispensers with spaced-out antibiotic discs or flame-sterilized forceps used to place individual discs on top of the agar surface are also viable options for this task. Next, to evaluate the antibacterial activity, the diameter of the zone of inhibition against the tested strains of Staphylococcus aureus and Staphylococcus haemolyticus will be measured using a ruler and calipers. Using the agar disk diffusion technique, the antibacterial activity of the CO nanoparticles will ascertained. C-1 (1 mg/ml), C-2 (50 mg/ml), and C-3 (100 mg/ml) were the doses employed in the disk diffusion method to assess the antibacterial activity of the as-prepared cobalt oxide (Co2O4) nanoparticles against three distinct bacterial strains: B. subtilis, Staphylococcus haemolyticus, and Staphylococcus aureus

First, the bacteria will be cultivated for a whole night in nutritional broth. Distribute 50 µl of the bacterial solution evenly across the nutrient agar plates until they seem dry. After that, 5 mm sterile paper disks will be placed on the dishes. Water served as the negative control (Huh &

Kwon) and the antibiotic ciprofloxacin as the positive control (Ab). After that, the disks will be covered with around 20µl of the antibiotic solution, water, and a suspension of Co2O4 nanoparticles. For twenty-four hours, plates will incubate at 37°C. By determining the diameter of the zone of inhibition (ZOI) surrounding the disks, antibacterial activity will be determined.

Antibacterial assay of synthesized Co3O4 nanoparticle

The good diffusion method will be used to analyze the produced Co3O4 nanoparticles against S. aureus and Staphylococcus haemolyticus germs to investigate their antibacterial properties. The antibacterial test will be conducted using the broth cultures of both strains. On a Muller Hinton Agar medium plate, a well with a diameter of around 5 mm will be created using gel puncture. A sterile cotton swab will be used to clean the colonies on the test medium. Five milligrams of Co3O4 nanoparticles will be added to one millilitre of distilled, sanitized water to create the nanoparticle suspension. A 0.10 µL suspension of the produced Co3O4 nanoparticles will add to the well as an inoculant. The plates will then be incubated for 24 hours at 37 °C in an incubator before the zones of inhibition are assessed.

Statistical Analysis

The antibacterial impact of cobalt nanoparticles (Co NPs) at various doses, the inhibition zone widths of Co NPs in comparison to the antibiotics, and the Co NPs' activity index (AI) in comparison to the antibiotics will all be investigated using a statistical design of experiments.

RESULTS

Characterization of Cobalt oxide NPs

XRD Analysis

Utilizing X-ray diffraction research, the cobalt oxide nanoparticles' crystalline nature was checked. The diffraction peaks in Figure 1's XRD pattern of Co3O4 nanoparticles have 2θ values of 10.84°, 34.31°, and 48.99°, which correspond to the 450, 1000, and 348 crystal planes of the crystalline Co3O4 phase, respectively. To a pure cubic phase structure, these peaks are indexed.

By plugging the nanoparticles' peak locations into the Scherrer equation, we could predict their average crystalline size. Produced Co3O4 nanoparticles had an average size of 36.24 nm.

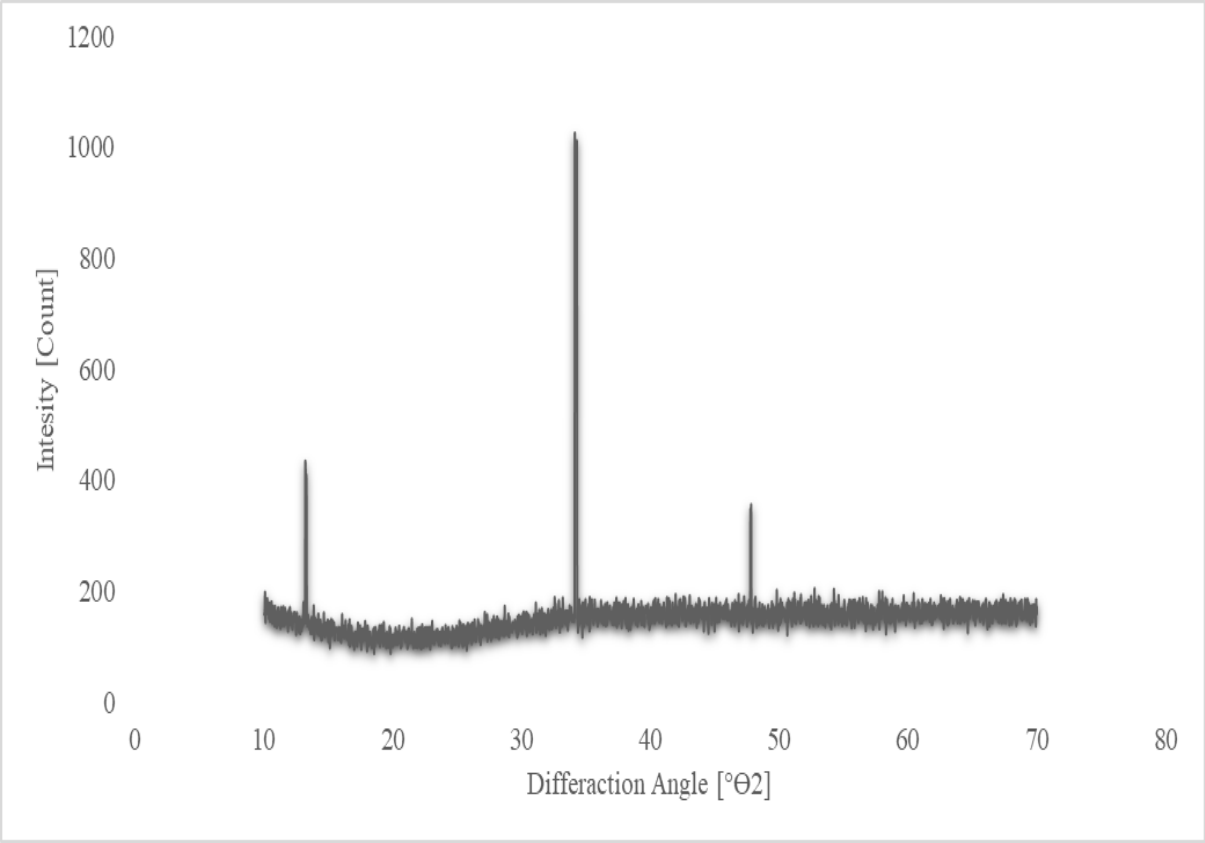


Figure 1: XRD analysis

FTIR analysis

A study using fluorescence time-of-flight (FTIR) may uncover the functional groups and chemical composition of the nanoparticles. Figure 2 shows the FTIR spectrum of the Cobalt oxide NPs that were synthesized. The vibrational characteristics of Cobalt oxide NPs were investigated using the Fourier transform infrared spectra. Figure 2 shows these characteristics are within the 0–4000 cm-1 range. By measuring peaks at 566 cm-1 and 667 cm-1, respectively, the stretching frequencies of CO and OCO3 were discovered, indicating the existence of a Co3O4 structure. So, a peak at 667 cm−1 indicates that there is Co2+ at the tetrahedral site while a signal at 566 cm−1 indicates that there is Co3+ at the octahedral site. Around 3450 cm−1 is the designated broadband frequency for the adsorbed water.

Remittances Review
July 2024,
Volume: 9, No: S 3, pp.1204-1231
ISSN: 2059-6588(Print) | ISSN 2059-6596(Online)

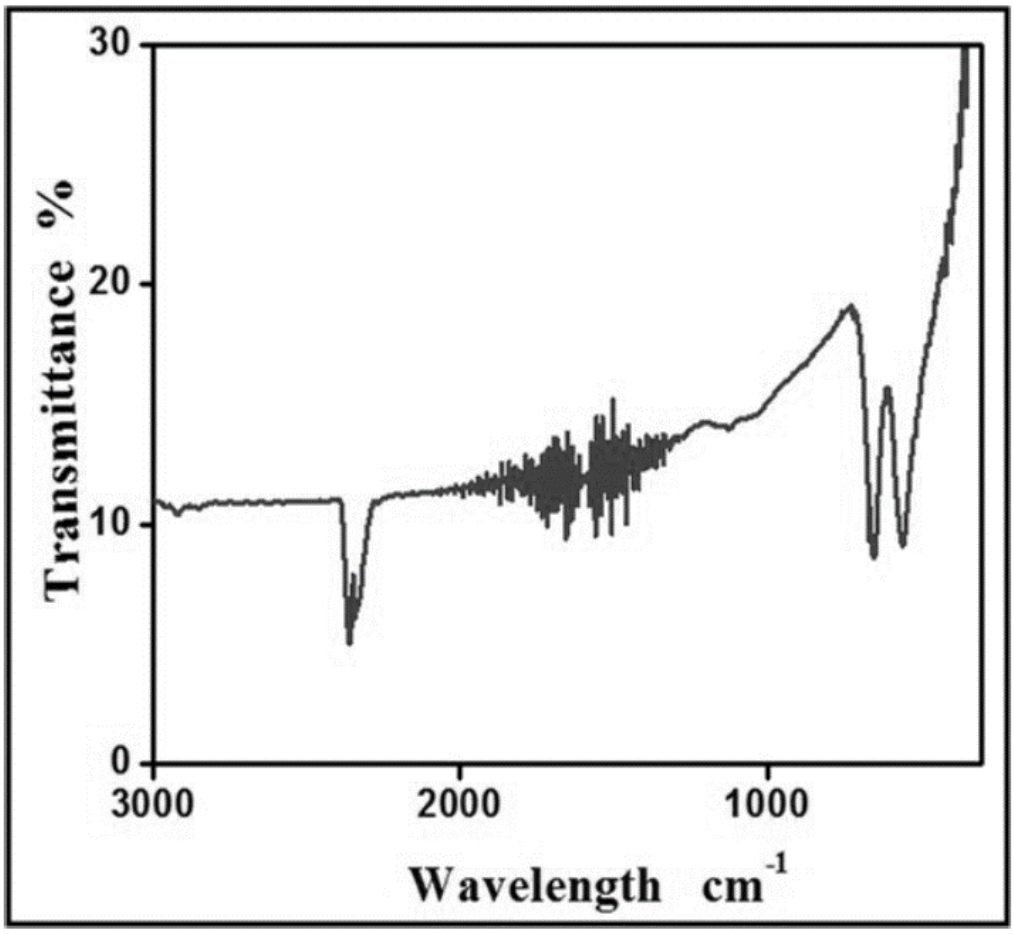


Figure 2: FTIR analysis

SEM analysis

The scanning electron microscopy (SEM) imaging of the cobalt oxide nanoparticles demonstrated their quality. The preparation of Co3O4 NPs is shown in Figure 3 via scanning electron microscopy illustrations. The Co3O4 nanoparticles that were created exhibited a porous network and a cubic spinel layout. In terms of average size, the pure nano-sized crystal particles were 36.24 nm.

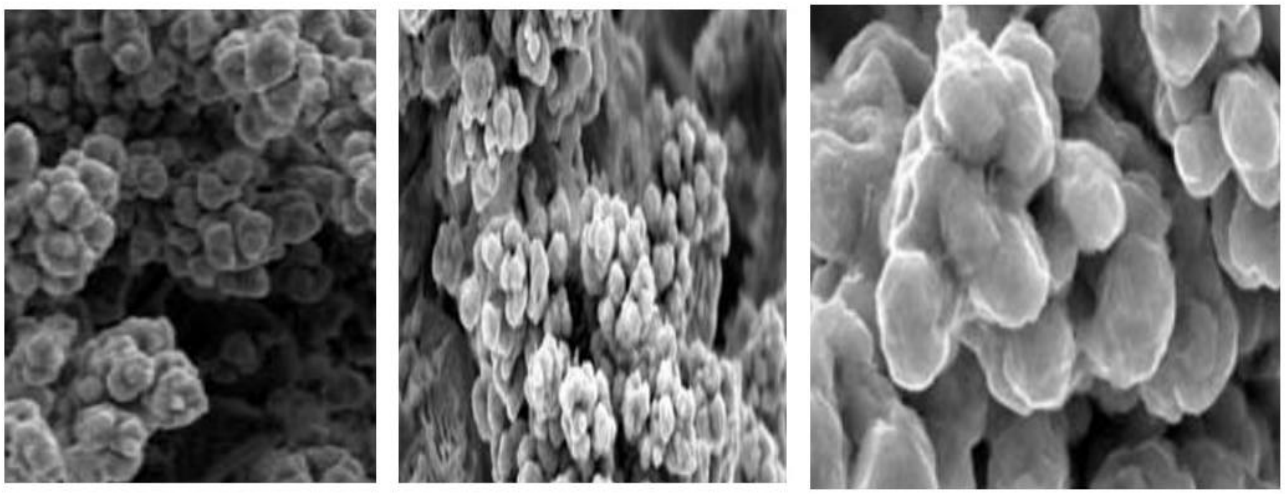


Figure 3: Shape of Cobalt oxide NPs, SEM analysis

Antimicrobial activity

Zone of Inhibition and MIC of Cobalt Oxide Nanoparticles

When compared to the control dosage at baseline, the 5 ug/ml dose significantly inhibits Staphylococcus aureus and Staphylococcus hemolyticus. The result is statistically significant. The 5 ug/ml dose is more effective against Staphylococcus haemolyticus compared to Staphylococcus aureus. A dosage of 10 ug/ml has less antimicrobial action against Staphylococcus haemolyticus than Staphylococcus aureus. A concentration of 15 ug/ml had a greater antimicrobial effect on Staphylococcus haemolyticus than on Staphylococcus aureus. Table 1 shows that even at dosages of 5 ug/ml, 10 ug/ml, or 15 ug/ml, the findings retain statistical significance.

Table 1: Antimicrobial activity of Cobalt oxide (20 mM) against Staphylococcus haemolyticus and Staphylococcus aureus.

Dose	***Staphylococcus haemolyticus***	***Staphylococcus aureus***		**P value**
Control	0 mm	0 mm	-	-
5 ug/ml	7±0.01 mm	5±0.02 mm	154.9	0.001***
10 ug/ml	5±0.02mm	7±0.01mm	154.9	0.001***
15 ug/ml	10±0.01mm	8±0.01mm	5.153	0.0067***

Remittances Review
July 2024,
Volume: 9, No: S 3, pp.1204-1231
ISSN: 2059-6588(Print) | ISSN 2059-6596(Online)

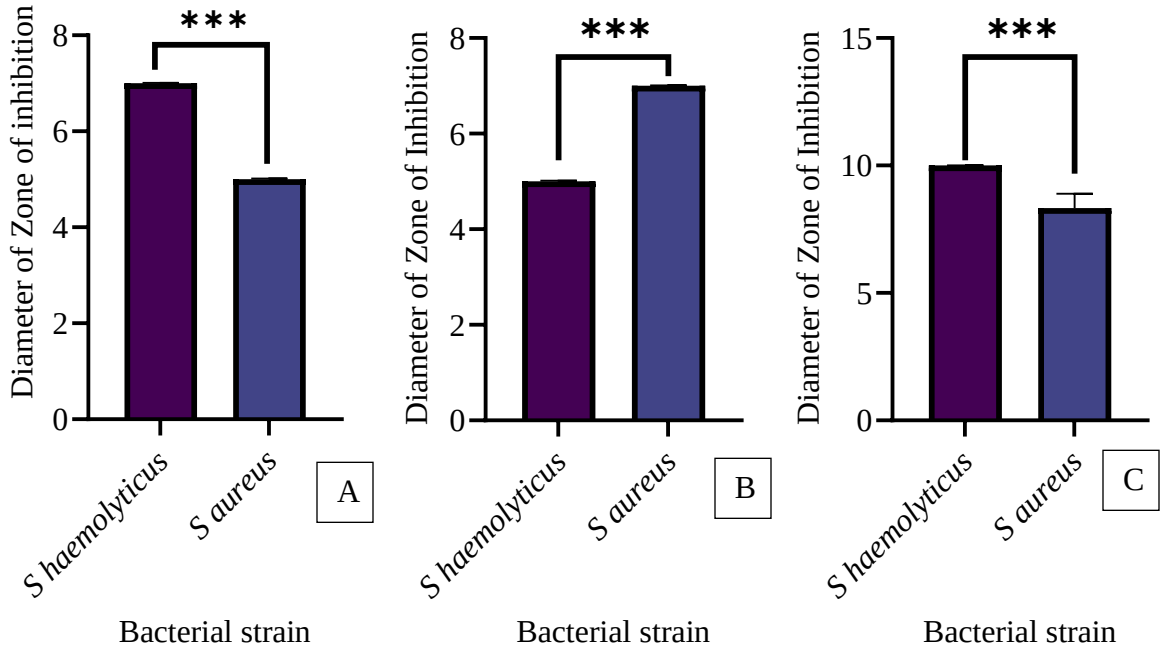


Figure 4: Antimicrobial activity of Cobalt oxide (20 mM) against Staphylococcus haemolyticus and Staphylococcus aureus (A) 5 ug/ml (B) 10 ug/ml (C) 15 ug/ml

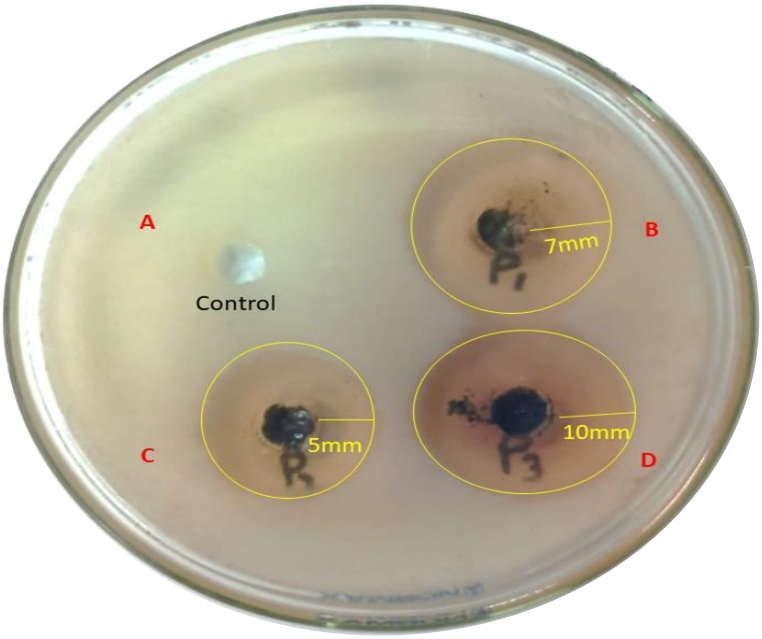


Figure 5: Antimicrobial activity of Cobalt Oxide Nanoparticles against Staphylococcus haemolyticus; (A) Control (B) 5ug/ml (C) 10 ug/ml (D) 15 ug/ml

Remittances Review
July 2024,
Volume: 9, No: S 3, pp.1204-1231
ISSN: 2059-6588(Print) | ISSN 2059-6596(Online)

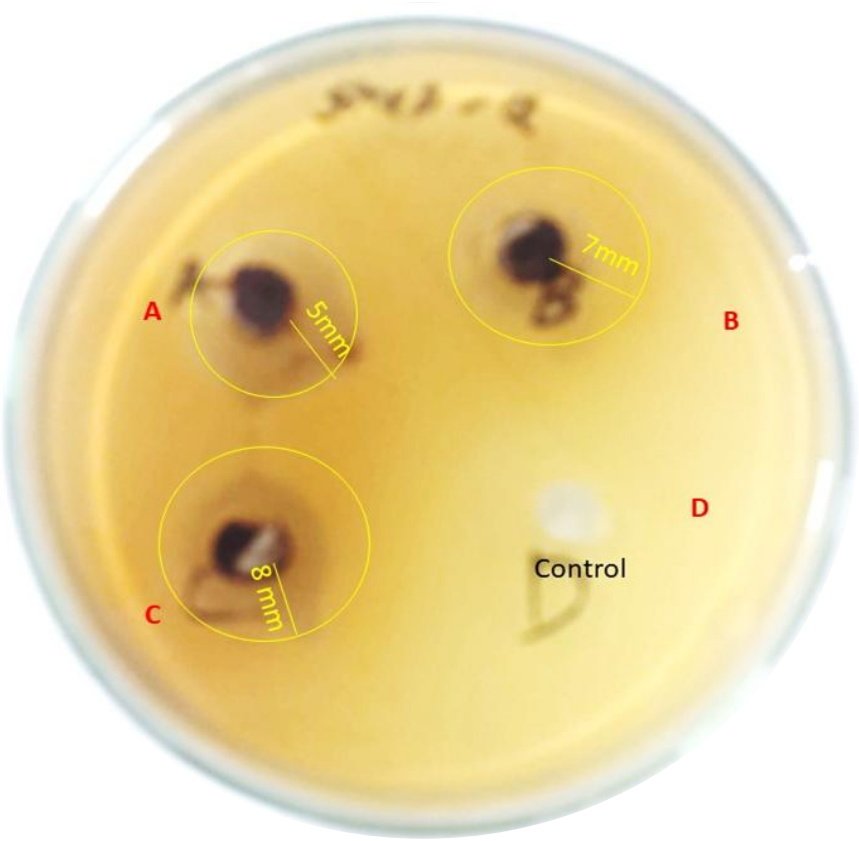


Figure 6: Antimicrobial activity of Cobalt Oxide Nanoparticles against Staphylococcus aureus; (A) 5ug/ml (B) 10 ug/ml (C) 15 ug/ml (D) Control

MIC

The MIC value for Staphylococcus haemolyticus is 1.79. The MIC value for Staphylococcus aureus is 2.68. Generally, the readings are expressed in micrograms per millilitre (µg/mL) or, on occasion, in milligrams per liter (mg/L), contingent upon the particular conditions of the study or medical environment. The efficacy of antibiotics against certain bacterial strains can be ascertained in part by looking at these MIC values. An increased susceptibility to the antibiotic is shown by lower MIC values. The MIC value for Staphylococcus haemolyticus has more potential against microbial activity than Staphylococcus aureus.

Table 2: MIC value of the synthesized cobalt oxide nanoparticles against Staphylococcus haemolyticus and Staphylococcus aureus.

Dose	***MIC value***
Staphylococcus haemolyticus	1.79`
Staphylococcus aureus	2.68

DISCUSSION

The X-ray diffraction examination was used to determine if the cobalt oxide nanoparticles were crystalline or not. Cobalt oxide nanoparticles and other materials may have their crystalline structure studied via X-ray diffraction (XRD) analysis (Mehta et al., 2017).

The XRD pattern of Co3O4 nanoparticles in Figure 1 shows diffraction peaks with 2θ values of 10.84°, 34.31°, and 48.99°, which are accordingly assigned to the 450, 1000, and 348 crystal planes of the crystalline Co3O4 phase. Diffraction patterns are produced by XRD analysis and appear as peaks on a graph. The arrangement of atoms within the material's crystalline lattice is represented by these peaks. Because the crystal structure of cobalt oxide is understood, distinctive peaks can be distinguished in the material (Ingham, 2015; Sajjad et al., 2024).

These peaks are indexed to a pure cubic phase structure. The XRD pattern's peak positions reveal details about the interatomic gap inside the crystal lattice. To verify the phase of the material, these coordinates can be compared to reference data for other crystal structures (e.g., cubic, hexagonal) (Mourdikoudis et al., 2018). The Scherer equation about the peaks was utilized to determine the mean crystalline size of the nanoparticles. The synthesized Co3O4 nanoparticles had an average size of 36.24. The average crystallite size of the nanoparticles can be calculated from the peak broadening using methods like the Scherer equation. The size and crystallinity of the nanoparticles can be inferred from the breadth of the peaks in the XRD pattern. More widely

spaced peaks indicate the presence of structural flaws or smaller crystallite sizes (Zhao et al., 2004).

Fluorescence time-of-flight (FTIR) analysis reveals the nanoparticles' chemical makeup and functional groups. The existence of functional groups on the surface of nanoparticles can also be detected by FTIR. These functional groups might result from surface changes or the application of ligands or stabilizing agents during the manufacture of nanoparticles. FTIR spectra show the presence of common functional groups such as amino (-NH2), carbonyl (C=O), and hydroxyl (-OH) (Movasaghi et al., 2008). Assigning absorption peaks to particular chemical bonds or functional groups based on their distinctive vibrational frequencies is a crucial step in the interpretation of FTIR spectra. Databases and reference spectra can help with this assignment (Madejová, 2003; Sattar et al., 2024).

The prepared Co3O4 nanoparticles showed a cubic spinel structure with a porous network. SEM investigation reveals a cubic spinel structure, indicating a well-defined crystal lattice arrangement in the cobalt oxide nanoparticles. Since the spinel structure is well-known for its stability and advantageous qualities, which make it appropriate for a variety of applications ranging from energy storage to catalysis, this structural knowledge is extremely significant (Carlson & Donavan, 2008). The high surface area-to-volume ratio of the Co3O4 nanoparticles is shown by the detection of a porous network in the SEM images. Because of their porous structure, the nanoparticles may be helpful in gas sensing, catalysis, and energy storage devices due to their increased reactivity and accessibility to active sites (Mohammed & Abdullah, 2018; Bilal et al., 2022).

Staphylococcus haemolyticus is more susceptible to the 5 ug/ml dosage than Staphylococcus aureus. The 5 ug/ml dosage of the antibiotic may be used through methods that are especially effective against Staphylococcus haemolyticus to achieve its antibacterial action. Differential susceptibility between the two species may arise, for instance, if the antibiotic targets a particular metabolic pathway or cell structure that is more essential to Staphylococcus haemolyticus survival than to Staphylococcus aureus (Kernodle & Kaiser, 1994).

Antimicrobial activity against Staphylococcus haemolyticus at a dose of 10 ug/ml is lower than that against Staphylococcus aureus. Because of a dose-response relationship, an antibiotic's effectiveness can change based on the dosage used. Staphylococcus haemolyticus may need a larger concentration of the antibiotic to achieve a similar amount of inhibition or death, even though Staphylococcus aureus may be more sensitive to it at the prescribed dosage (Hosaka et al., 1992; Afzal et al., 2024).

Antimicrobial activity against Staphylococcus haemolyticus is higher at a dose of 15 ug/ml compared to Staphylococcus aureus. Genetic differences between Staphylococcus aureus and Staphylococcus haemolyticus can influence their susceptibility to antibiotics. Staphylococcus aureus strains may possess specific genetic elements that confer decreased susceptibility to the antibiotic at the given dosage compared to Staphylococcus haemolyticus strains (Saeed et al., 2023). The MIC value for Staphylococcus haemolyticus is 1.79. The MIC value for Staphylococcus aureus is 2.68. In this case, Staphylococcus haemolyticus has a lower MIC value (1.79) compared to Staphylococcus aureus (2.68). This suggests that Staphylococcus haemolyticus is more susceptible to the antimicrobial agent being tested than Staphylococcus aureus (Terwee et al., 2021).

CONCLUSIONS AND RECOMMENDATIONS:

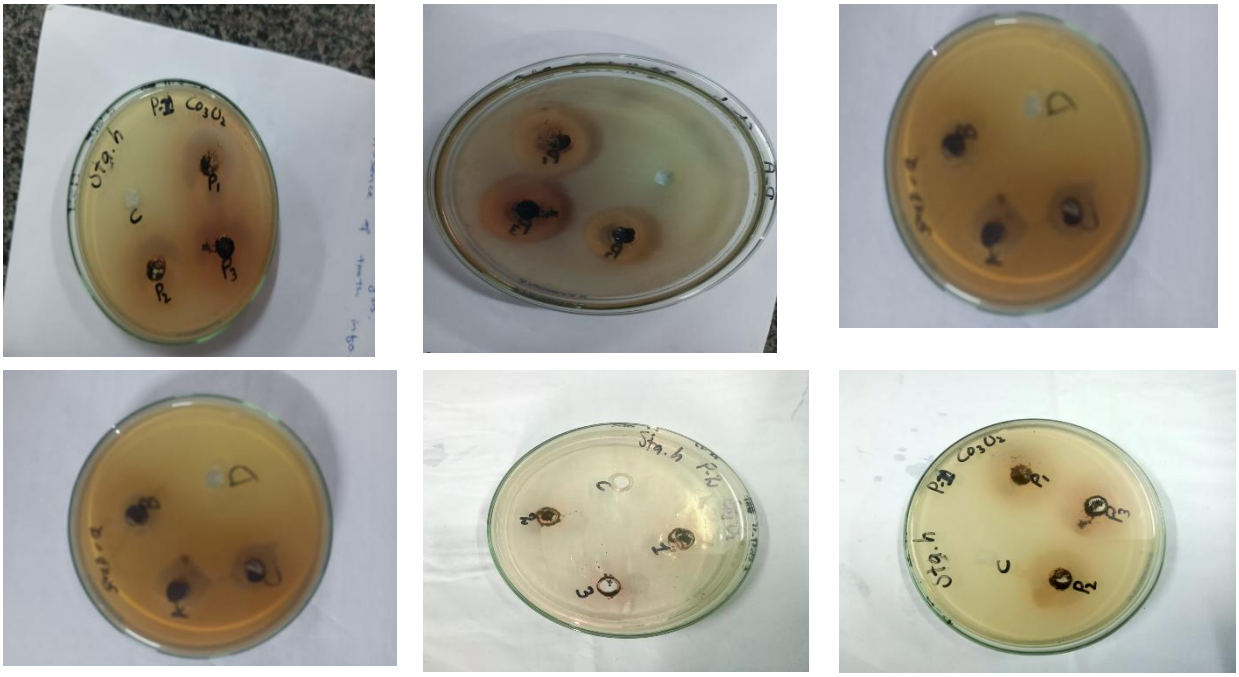
We found that X-ray diffraction (XRD) examination confirmed a cubic spinel structure with unique diffraction peaks, revealing the crystalline structure of cobalt oxide nanoparticles. The average size of the crystallites was found to be 36.24 according to the Scherrer equation. Because of its porous network structure, which was also shown by scanning electron microscopy, this material has potential uses in gas sensing, catalysis, and energy storage, where its enhanced reactivity and accessibility to active sites might be useful. An antibiotic's antimicrobial activity against Staphylococcus haemolyticus and Staphylococcus aureus at several doses was also examined in the research.

The results showed differential susceptibility between the two species, with Staphylococcus haemolyticus being more susceptible at a lower dosage (5 ug/ml), whereas Staphylococcus aureus was more susceptible at higher dosages (10 ug/ml and 15 ug/ml). Genetic differences and

metabolic pathways may contribute to this differential susceptibility. The MIC (Minimum Inhibitory Concentration) values for the two bacterial species were provided, with Staphylococcus haemolyticus having a lower MIC value (1.79) compared to Staphylococcus aureus (2.68). This indicates that Staphylococcus haemolyticus is more susceptible to the tested antimicrobial agent than Staphylococcus aureus.

It is recommended to foster interdisciplinary collaborations between researchers in materials science, microbiology, medicine, and engineering to leverage expertise and resources for comprehensive investigations into nanoparticle synthesis, characterization, and applications, as well as antibiotic resistance mechanisms and treatment strategies.

Supplementary



REFERENCES

Waifalkar, P., Parit, S., Chougale, A., Sahoo, S. C., Patil, P., Patil, P. J. J. o. C., & Science, I. (2016) Immobilization of invertase on chitosan coated γ-Fe2O3 magnetic nanoparticles to facilitate magnetic separation. 482, 159-164.

Abo-Shama, U. H., El-Gendy, H., Mousa, W. S., Hamouda, R. A., Yousuf, W. E., Hetta, H. F., & Abdeen, E. E. (2020) Synergistic and antagonistic effects of metal nanoparticles in combination with antibiotics against some reference strains of pathogenic microorganisms. Infection and drug resistance, 351-362.

Abass, A. A., Alaarage, W. K., Abdulrudha, N. H., & Haider, J. (2021) Evaluating the antibacterial effect of cobalt nanoparticles against multi-drug resistant pathogens. Journal of medicine and life, 14(6), 823.

Bilal, A. (2021). Rabies is a zoonotic disease: a literature review. Occup. Med. Health Aff, 9(2).

Bilal, A. (2021). Impacts of depression on pregnancy: A review. Occup Med Health Aff, 9(2).

Abdelwahab, T. A. M., Mohanty, M. K., Sahoo, P. K., Behera, D., & Fodah, A. E. M. (2021) Cobalt nanoparticles to enhance anaerobic digestion of cow dung: focusing on kinetic models for biogas yield and effluent utilization. Biomass Conversion and Biorefinery, 1-13

Ajarem, J. S., Maodaa, S. N., Allam, A. A., Taher, M. M., & Khalaf, M. (2021) Benign synthesis of cobalt oxide nanoparticles containing red algae extract: antioxidant, antimicrobial, anticancer, and anticoagulant activity. Journal of Cluster Science, 1-12.

Anasthasiya, A. N. A., Prabhu, E., Jayaraman, V., & Gnanasekar, K. (2020) Wide range H2 sensors for process streams, based on multilayered Pd thin films dispersed with Ag nanoparticles and chromium as buffer layer. International Journal of Hydrogen Energy, 45(41), 22195-22203.

Aslan Korkmaz, Ş. (2021) Green synthesis of cobalt-oxide nanoparticles with an endemic species Allium tuncelianum and anticancer activity. Inorganic and Nano-Metal Chemistry, 1-7.

Afzal, M., Ali, U., Riaz, A., Tanvir, F., Bilal, A., & Ahmad, S. (2024). In-silico analysis of deleterious single nucleotide polymorphisms (SNPs) of leukemia inhibitory factor (LIF), and their conformational predictions. Journal of Population Therapeutics and Clinical Pharmacology, 31(1), 2792-2811.

Ávila-Gutiérrez, M., Moisset, A., Ngo, A.-T., Costanzo, S., Simon, G., Colomban, P., Petit, M., Petit, C., & Lisiecki, I. (2023) Influence of the nanocrystallinity on exchange bias in Co/CoO core/shell nanoparticles. Colloids and Surfaces A: Physicochemical and Engineering Aspects, 676, 132281.

Bajpai, V. K., Khan, I., Shukla, S., Kang, S.-M., Aziz, F., Tripathi, K. M., Saini, D., Cho, H.-J., Heo, N. S., & Sonkar, S. K. (2020) Multifunctional NP-doped carbon dots for regulation of apoptosis and autophagy in B16F10 melanoma cancer cells and in vitro imaging applications. Theranostics, 10(17), 7841.

Balabanova, L., Averianova, L., Marchenok, M., Son, O., & Tekutyeva, L. (2021) Microbial and genetic resources for cobalamin (vitamin B12) biosynthesis: From ecosystems to industrial biotechnology. International journal of molecular sciences, 22(9), 4522.

Behera, A., & Behera, A. (2022) Nanomaterials. Advanced Materials: An Introduction to Modern Materials Science, 77-125.

Bilal, A., Ahmad, S., Tanvir, F., Tariq, M., Ramzan, K., Saleem, M., & Saleem, H. G. M. (2022). Predictive modeling of N-acetyl transferase 2 single nucleotide polymorphisms and breast cancer risk using in-silco approaches. The Journal of Microbiology and Molecular Genetics, 3(2), 105-121.

Belkacem, W., Belhi, R., Thabet Mliki, N., & Bouazizi, A. (2023) Exchange Bias and Vertical Magnetic Shift on Co− CoO Thin Films. Crystal Research and Technology, 58(9), 2300059.

Bilal, M., Mehmood, S., Rasheed, T., & Iqbal, H. M. (2019) Bio-catalysis and biomedical perspectives of magnetic nanoparticles as versatile carriers. Magnetochemistry, 5(3), 42.

Bonkat, G., Cai, T., Galeone, C., Koves, B., & Bruyere, F. (2022) Adherence to European Association of Urology Guidelines and State of the Art of Glycosaminoglycan Therapy for the Management of Urinary Tract Infections: A Narrative Review and Expert Meeting Report. European Urology Open Science, 44, 37-45.

Bragazzi, N. L., Kong, J. D., Mahroum, N., Tsigalou, C., Khamisy-Farah, R., Converti, M., & Wu, J. (2023) Epidemiological trends and clinical features of the ongoing monkeypox epidemic: A preliminary pooled data analysis and literature review. Journal of medical virology, 95(1), e27931.

Bray, F., Laversanne, M., Weiderpass, E., & Soerjomataram, I. (2021) The ever-increasing importance of cancer as a leading cause of premature death worldwide. Cancer, 127(16), 3029-3030.

Bustamante-Torres, M., Romero-Fierro, D., Estrella-Nuñez, J., Arcentales-Vera, B., Chichande-Proaño, E., & Bucio, E. (2022) Polymeric composite of magnetite iron oxide nanoparticles and their application in biomedicine: a review. Polymers, 14(4), 752.

Carlson, B. D., & Donavan, D. T. (2008) SEM. Sport Marketing Quarterly, 17, 154-162.

Chen, Y., Zhang, Y., & Fu, S. J. M. L. (2007) Synthesis and characterization of Co3O4 hollow spheres. 61(3), 701-705.

Sattar, R. Z., Bilal, A., Bashir, S., Iftikhar, A., & Yaqoob, I. (2024). Embryotoxicity of fluconazole on developing chick embryos. The Journal of Basic and Applied Zoology, 85(1), 8.

Sajjad, M. K., Bilal, A., Iftikhar, A., Awais, M., Asif, I., Shaheen, F., & Zahoor, G. (2024). Examining the Association Between Pesticide Exposures and Chronic Diseases in Agricultural Workers. Remittances Review, 9(2), 2153-2176.

Coates, A. R., Hu, Y., Holt, J., & Yeh, P. (2020) Antibiotic combination therapy against resistant bacterial infections: synergy, rejuvenation and resistance reduction. Expert review of Anti-infective therapy, 18(1), 5-15.

Deltell, A., Mohamed, A. E.-M. A., Álvarez-Alonso, P., Ipatov, M., Andrés, J., Gonzalez, J. A., Sánchez, T., Zhukov, A., Escoda, M., & Suñol, J. (2021) Martensitic transformation, magnetic and magnetocaloric properties of Ni–Mn–Fe–Sn Heusler ribbons. journal of materials research and technology, 12, 1091-1103.

Ding, D., Shen, K., Chen, X., Chen, H., Chen, J., Fan, T., Wu, R., & Li, Y. J. A. C. (2018) Multi-level architecture optimization of MOF-templated Co-based nanoparticles embedded in hollow N-doped carbon polyhedra for efficient OER and ORR. 8(9), 7879-7888.

Dong, X., Choi, C., & Kim, B. (2002) Chemical synthesis of Co nanoparticles by chemical vapor condensation. Scripta Materialia, 47(12), 857-861.

Du, J., You, S., Li, X., Zhang, Y., Yu, Y., Li, Q., Wang, F., Ren, N., & Zou, J. (2020) In situ immobilization of copper oxide thin-layer on zeolitic imidazolate framework-67-derived cobalt oxide@ nitrogen-doped carbon with multi-level architecture and versatile active sites for enhancing oxygen evolution/reduction reactions. Journal of Power Sources, 478, 228707.

Flores-Rojas, G. G., López-Saucedo, F., Vera-Graziano, R., Mendizabal, E., & Bucio, E. (2022) Magnetic nanoparticles for medical applications: Updated review. Macromol, 2(3), 374-390.

Gareeva, Z., Shulga, N., Doroshenko, R., & Zvezdin, A. (2023) Electric field control of magnetic states in ferromagnetic–multiferroic nanostructures. Physical Chemistry Chemical Physics, 25(33), 22380-22387.

Gaur, M., Misra, C., Yadav, A. B., Swaroop, S., Maolmhuaidh, F. Ó., Bechelany, M., & Barhoum, A. (2021) Biomedical applications of carbon nanomaterials: fullerenes, quantum dots, nanotubes, nanofibers, and graphene. Materials, 14(20), 5978.

Ghaem, E. N., Dorranian, D., Sari, A. H. J. P. E. L.-d. S., & Nanostructures. (2020) Characterization of cobalt oxide nanoparticles produced by laser ablation method: Effects of laser fluence. 115, 113670.

Ghoshani, M., Mozaafari, M., Normile, P. S., De Toro, J. A., & Al-Nabhani, A. (2021) Core size and interface impact on the exchange bias of cobalt/cobalt oxide nanostructures. Magnetochemistry, 7(3), 40.

Gogoi, L., Gao, W., Ajayan, P. M., & Deb, P. (2023) Quantum magnetic phenomena in engineered heterointerface of low-dimensional van der Waals and non-van der Waals materials. Physical Chemistry Chemical Physics, 25(3), 1430-1456.

González-Montaña, J.-R., Escalera-Valente, F., Alonso, A. J., Lomillos, J. M., Robles, R., & Alonso, M. E. (2020) Relationship between vitamin B12 and cobalt metabolism in domestic ruminant: an update. Animals, 10(10), 1855.

Gupta, V., Kant, V., Sharma, A., & Sharma, M. (2020) Comparative assessment of antibacterial efficacy for cobalt nanoparticles, bulk cobalt and standard antibiotics: a concentration dependant study. Наносистемы: физика, химия, математика, 11(1), 78-85.

Halczuk, K., Kaźmierczak-Barańska, J., Karwowski, B. T., Karmańska, A., & Cieślak, M. (2023) Vitamin B12—Multifaceted In Vivo Functions and In Vitro Applications. Nutrients, 15(12), 2734.

Hosaka, M., Yasue, T., Fukuda, H., Tomizawa, H., Aoyama, H., & Hirai, K. (1992) In vitro and in vivo antibacterial activities of AM-1155, a new 6-fluoro-8-methoxy quinolone. Antimicrobial agents and chemotherapy, 36(10), 2108-2117.

Huh, A. J., & Kwon, Y. J. J. J. o. c. r. (2011) "Nanoantibiotics": a new paradigm for treating infectious diseases using nanomaterials in the antibiotics resistant era. 156(2), 128-145.

Ingham, B. (2015) X-ray scattering characterisation of nanoparticles. Crystallography Reviews, 21(4), 229-303.

Iravani, S., & Varma, R. S. (2020) Sustainable synthesis of cobalt and cobalt oxide nanoparticles and their catalytic and biomedical applications. Green Chemistry, 22(9), 2643-2661.

Islam, M. R., Akash, S., Jony, M. H., Alam, M. N., Nowrin, F. T., Rahman, M. M., Rauf, A., & Thiruvengadam, M. (2023) Exploring the potential function of trace elements in human health: a therapeutic perspective. Molecular and Cellular Biochemistry, 1-31.

Jaffari, G. H., Bah, M., & Shah, S. I. (2021) Strong magnetic coupling between antiferromagnetic and ferromagnetic phases in polycrystalline hollow nanoparticles composed of spinel solid solution. Journal of Alloys and Compounds, 857, 157607.

Jamuna, G., Yasodha, S., Thamarai, P., Vickram, A., Swaminaathan, P., Saravanan, A., & Yaashikaa, P. (2023) Design strategies, utilization and applications of nano-engineered biomaterials for the enhancement of bioenergy: A sustainable approach. Biochemical Engineering Journal, 109104.

Jenkins, J. A., Musgrove, M., & White, S. J. O. (2023) Outlining Potential Biomarkers of Exposure and Effect to Critical Minerals: Nutritionally Essential Trace Elements and the Rare Earth Elements. Toxics, 11(2), 188.

Jeon, J. H., Na, H. R., Kim, H., Lee, S., Song, S., Kim, J., Park, S., Kim, J., Noh, H., & Kim, G. (2022) Emergent topological Hall effect from exchange coupling in ferromagnetic Cr2Te3/Noncoplanar antiferromagnetic Cr2Se3 bilayers. ACS nano, 16(6), 8974-8982.

Jiang, T., Li, Q., Qiu, J., Chen, J., Du, S., Xu, X., Wu, Z., Yang, X., Chen, Z., & Chen, T. (2022) Nanobiotechnology: applications in chronic wound healing. International journal of nanomedicine, 3125-3145.

Kernodle, D. S., & Kaiser, A. B. (1994) Comparative prophylactic efficacies of ciprofloxacin, ofloxacin, cefazolin, and vancomycin in experimental model of staphylococcal wound infection. Antimicrobial agents and chemotherapy, 38(6), 1325-1330.

Khizar, S., Ahmad, N. M., Ahmed, N., Manzoor, S., Hamayun, M. A., Naseer, N., Tenório, M. K., Lebaz, N., & Elaissari, A. (2020) Aminodextran coated CoFe2O4 nanoparticles for combined magnetic resonance imaging and hyperthermia. Nanomaterials, 10(11), 2182.

Li, X., Chen, L., Luan, S., Zhou, J., Xiao, X., Yang, Y., Mao, C., Fang, P., Chen, L., & Zeng, X. (2022). The development and progress of nanomedicine for esophageal cancer diagnosis and treatment. Paper presented at the Seminars in cancer biology.

Li, X., Wang, C., Xue, H., Pang, H., & Xu, Q. (2020) Electrocatalysts optimized with nitrogen coordination for high-performance oxygen evolution reaction. Coordination Chemistry Reviews, 422, 213468.

Liang, X., & Zhao, L. J. R. A. (2012) Room-temperature synthesis of air-stable cobalt nanoparticles and their highly efficient adsorption ability for Congo red. 2(13), 5485-5487.

Liaqat, I., Ali, R., Hanif, U., Latif, A., Bibi, A., Saleem, S., Naseem, S., Ulfat, M., Mubin, M., & Rashid, F. (2022) Antimicrobial Efficacy of Biogenic Cobalt and Copper Nanoparticles against Pathogenic Isolates. Journal of Oleo Science, 71(11), 1669-1677.

Lovelock, S. L., Crawshaw, R., Basler, S., Levy, C., Baker, D., Hilvert, D., & Green, A. P. (2022) The road to fully programmable protein catalysis. Nature, 606(7912), 49-58.

Luyts, K., Napierska, D., Nemery, B., & Hoet, P. H. (2013) How physico-chemical characteristics of nanoparticles cause their toxicity: complex and unresolved interrelations. Environmental science: processes & impacts, 15(1), 23-38.

Ma, Y., Lin, W., Ruan, Y., Lu, H., Fan, S., Chen, D., Huang, Y., Zhang, T., Pi, J., & Xu, J.-F. (2022) Advances of Cobalt Nanomaterials as Anti-Infection Agents, Drug Carriers, and Immunomodulators for Potential Infectious Disease Treatment. Pharmaceutics, 14(11), 2351.

Madejová, J. (2003) FTIR techniques in clay mineral studies. Vibrational spectroscopy, 31(1), 1-10.

Madkour, L. H., & Madkour, L. H. (2019) Introduction to nanotechnology (NT) and nanomaterials (NMs). Nanoelectronic Materials: Fundamentals and Applications, 1-47.

Mehta, B., Chhajlani, M., & Shrivastava, B. (2017). Green synthesis of silver nanoparticles and their characterization by XRD. Paper presented at the Journal of physics: conference series.

Mohammed, A., & Abdullah, A. (2018). Scanning electron microscopy (SEM): A review. Paper presented at the Proceedings of the 2018 International Conference on Hydraulics and Pneumatics—HERVEX, Băile Govora, Romania.

Mohandes, A., Aghamaali, M. R., Sabouri, Z., Darroudi, M. J. M. S., & B, E. (2023) Biosynthesis of cobalt oxide nanoparticles (Co3O4-NPs) using Caccinia macranthera extract and evaluation of their cytotoxicity and photocatalytic activity. 297, 116782.

Mohapatra, J., Xing, M., Elkins, J., & Liu, J. P. (2020) Hard and semi-hard magnetic materials based on cobalt and cobalt alloys. Journal of Alloys and Compounds, 824, 153874.

Mohsin, H., Sultan, U., Joya, Y., Ahmed, S., Awan, M., & Arshad, S. (2016). Development and characterization of cobalt based nanostructured super hydrophobic coating. Paper presented at the IOP Conference Series: Materials Science and Engineering.

Moradpoor, H., Safaei, M., Rezaei, F., Golshah, A., Jamshidy, L., Hatam, R., & Abdullah, R. S. (2019) Optimisation of cobalt oxide nanoparticles synthesis as bactericidal agents. Open access Macedonian journal of medical sciences, 7(17), 2757.

Mourdikoudis, S., Pallares, R. M., & Thanh, N. T. (2018) Characterization techniques for nanoparticles: comparison and complementarity upon studying nanoparticle properties. Nanoscale, 10(27), 12871-12934.

Wu, H., Zhang, X., Zhang, H., Liu, C., Huang, F., & Li, S. (2022) Structure-dependent electrochemical properties of cobalt (II) carbonate hydroxide nanocrystals in supercapacitors. Journal of Colloid and Interface Science, 607, 1633-1640.

Yoshikawa, H., Hayashida, K., Kozuka, Y., Horiguchi, A., Awaga, K., Bandow, S., & Iijima, S. J. A. p. l. (2004) Preparation and magnetic properties of hollow nano-spheres of cobalt and cobalt oxide: Drastic cooling-field effects on remnant magnetization of antiferromagnet. 85(22), 5287-5289.

Zhang, J., Dai, Z., Bao, J., Zhang, N., López-Quintela, M. A. J. J. o. c., & science, i. (2007) Self-assembly of Co-based nanosheets into novel nest-shaped nanostructures: Synthesis and characterization. 305(2), 339-344.

Movasaghi, Z., Rehman, S., & ur Rehman, D. I. (2008) Fourier transform infrared (FTIR) spectroscopy of biological tissues. Applied Spectroscopy Reviews, 43(2), 134-179.

Mukherjee, S., Ray, S., & Thakur, R. (2009) Solid lipid nanoparticles: a modern formulation approach in drug delivery system. Indian journal of pharmaceutical sciences, 71(4), 349.

Omran, B., Nassar, H., Younis, S., El-Salamony, R., Fatthallah, N., Hamdy, A., El-Shatoury, E., & El-Gendy, N. S. J. J. o. a. m. (2020) Novel mycosynthesis of cobalt oxide nanoparticles using Aspergillus brasiliensis ATCC 16404—optimization, characterization and antimicrobial activity. 128(2), 438-457.

Ouyang, J., Xie, A., Zhou, J., Liu, R., Wang, L., Liu, H., Kong, N., & Tao, W. (2022) Minimally invasive nanomedicine: nanotechnology in photo-/ultrasound-/radiation-/magnetism-mediated therapy and imaging. Chemical Society Reviews, 51(12), 4996-5041.

Oyarzún, S., Tamion, A., Tournus, F., Dupuis, V., & Hillenkamp, M. (2015) Size effects in the magnetic anisotropy of embedded cobalt nanoparticles: from shape to surface. Scientific reports, 5(1), 14749.

Panda, J., Das, S., Kumar, S., Tudu, B., & Sarkar, R. (2022) Investigation of antibacterial, antioxidant, and anticancer properties of hydrothermally synthesized cobalt ferrite nanoparticles. Applied Physics A, 128(7), 562.

Pereira, J., Simões, M., & Silva, J. L. (2019) Microalgal assimilation of vitamin B12 toward the production of a superfood. Journal of food biochemistry, 43(8), e12911.

Ranjbar Bahadori, S., Hart, R., & Hao, Y.-W. (2020) Synthesis of cobalt, palladium, and rhenium nanoparticles. Tungsten, 2, 261-288.

Raza, M. A., Kanwal, Z., Riaz, S., & Naseem, S. (2016). Synthesis, characterization and antibacterial properties of nano-sized cobalt particles. Paper presented at the Proceedings of the 2016 World Congress on Advances in Civil, Enviromental, and Materials Research (ACEM16), Jeju Island, Korea.

Rodríguez De Luna, S. L., Ramírez-Garza, R., & Serna Saldívar, S. O. (2020) Environmentally friendly methods for flavonoid extraction from plant material: Impact of their operating conditions on yield and antioxidant properties. The Scientific World Journal, 2020.

Ryu, J., Lee, S., Lee, K. J., & Park, B. G. (2020) Current-Induced Spin–Orbit Torques for Spintronic Applications. Advanced Materials, 32(35), 1907148.

Saeed, S. I., Vivian, L., Zalati, C. S. C., Sani, N. I. M., Aklilu, E., Mohamad, M., Noor, A. A. M., Muthoosamy, K., & Kamaruzzaman, N. F. (2023) Antimicrobial activities of graphene oxide against biofilm and intracellular Staphylococcus aureus isolated from bovine mastitis. BMC Veterinary Research, 19(1), 10.

Sajid, M., & Płotka-Wasylka, J. J. M. J. (2020) Nanoparticles: Synthesis, characteristics, and applications in analytical and other sciences. 154, 104623.

Sayadi, M. H., Pavlaki, M. D., Loureiro, S., Martins, R., Tyler, C. R., Mansouri, B., Kharkan, J., & Shekari, H. J. E. (2022) Co-exposure of zinc oxide nanoparticles and multi-layer graphenes in blackfish (Capoeta fusca): evaluation of lethal, behavioural, and histopathological effects. 31(3), 425-439.

Shabatina, T. I., Vernaya, O. I., Shabatin, V. P., & Melnikov, M. Y. (2020) Magnetic nanoparticles for biomedical purposes: Modern trends and prospects. Magnetochemistry, 6(3), 30.

Silva, A. S., Roman, F. F., Dias, A. V., de Tuesta, J. L. D., Narcizo, A., da Silva, A. P. F., Çaha, I., Deepak, F. L., Bañobre-López, M., & Ferrari, A. M. (2023) Hybrid multi-core shell magnetic nanoparticles for wet peroxide oxidation of paracetamol: Application in synthetic and real matrices. Journal of Environmental Chemical Engineering, 11(5), 110806.

Staniuk, M., Hirsch, O., Kränzlin, N., Böhlen, R., van Beek, W., Abdala, P. M., & Koziej, D. J. C. o. M. (2014) Puzzling mechanism behind a simple synthesis of cobalt and cobalt oxide nanoparticles: in situ synchrotron X-ray absorption and diffraction studies. 26(6), 2086-2094.

Terreni, M., Taccani, M., & Pregnolato, M. (2021) New antibiotics for multidrug-resistant bacterial strains: latest research developments and future perspectives. Molecules, 26(9), 2671.

Terwee, C. B., Peipert, J. D., Chapman, R., Lai, J.-S., Terluin, B., Cella, D., Griffith, P., & Mokkink, L. B. (2021) Minimal important change (MIC): a conceptual clarification and systematic review of MIC estimates of PROMIS measures. Quality of life Research, 30, 2729-2754.

Thalji, M. R., Ibrahim, A. A., & Ali, G. A. (2021) Cutting-edge development in dendritic polymeric materials for biomedical and energy applications. European Polymer Journal, 160, 110770.

Usami, T., Salman, S., Kuroda, K., Gouda, M., Mahdy, A., & Okido, M. (2021) Synthesis of Cobalt-Nickel Nanoparticles via a Liquid-Phase Reduction Process. Journal of Nanotechnology, 2021, 1-7.

Viñas, S. L. (2016). Anisotropic cobalt-based nanostructures: synthesis and characterization. Universitätsbibliothek Duisburg-Essen.

Vivek, S., Kumar, A., Lekha, C. C., Kalarikkal, N., Banerjee, A., & Nair, S. (2023) Exchange bias studies of CoFe2O4 coated BiFeO3 nanoparticles. Journal of Alloys and Compounds, 172066.

Vodyashkin, A. A., Kezimana, P., Prokonov, F. Y., Vasilenko, I. A., & Stanishevskiy, Y. M. (2022) Current methods for synthesis and potential applications of cobalt nanoparticles: A review. Crystals, 12(2), 272.

Wang, F.-H., Chen, M.-S., Jiang, Y.-L., Liu, H.-W., & Kang, T.-K. (2022) Fabrication and characterization of sputtered Mg and F co-doped ZnO thin films with different substrate temperature for silicon thin-film solar cell applications. Journal of Alloys and Compounds, 897, 163174.

Wang, L., Hasanzadeh Kafshgari, M., & Meunier, M. (2020) Optical properties and applications of plasmonic-metal nanoparticles. Advanced Functional Materials, 30(51), 2005400.

Wang, S., Wang, C., Zhang, W., Fan, W., Liu, F., & Liu, Y. (2022) Bioactive nano-selenium antagonizes cobalt nanoparticle-mediated oxidative stress via the Keap1-Nrf2-ARE signaling pathway. Journal of Nanoparticle Research, 24, 1-12.

Waris, A., Din, M., Ali, A., Afridi, S., Baset, A., Khan, A. U., & Ali, M. (2021) Green fabrication of Co and Co3O4 nanoparticles and their biomedical applications: A review. Open life sciences, 16(1), 14-30.

Xiang, N., Bai, Y., Li, Q., Han, X., Zheng, J., Zhao, Q., Hou, Y., & Huang, Z. (2022) ZIF-67-derived hierarchical hollow Co3O4@ CoMn2O4 nanocages for efficient catalytic oxidation of formaldehyde at low temperature. Molecular Catalysis, 528, 112519.

Younus, N., & Zuberi, A. (2022) Significance of extrinsic factors for the optimization of dietary cobalt supplementation in Tor putitora fingerlings. Fish Physiology and Biochemistry, 48(4), 883-897.

Zhang, J., Dai, Z., Bao, J., Zhang, N., López-Quintela, M. A. J. J. o. c., & science, i. (2007) Self-assembly of Co-based nanosheets into novel nest-shaped nanostructures: Synthesis and characterization. 305(2), 339-344.

Zhang, S., Wang, Y., Song, H., Lu, J., Yuan, Z., & Guo, J. (2019) Copper nanoparticles and copper ions promote horizontal transfer of plasmid-mediated multi-antibiotic resistance genes across bacterial genera. Environment international, 129, 478-487.

Zhao, Y., Zhang, Z., & Dang, H. (2004) A simple way to prepare bismuth nanoparticles. Materials Letters, 58(5), 790-793.

Zorai, A., Souici, A., Dragoe, D., Rivière, E., Ouhenia, S., Belloni, J., & Mostafavi, M. (2023) Superparamagnetic cobalt ferrite nanoparticles synthesized by gamma irradiation. New Journal of Chemistry, 47(5), 2626