



Chemical and Spectroscopic Characterization of Novel Dexamethasone/Zn Complex as a Potent Antioxidant and Antibacterial Agent

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ABSTRACT

Background: Dexamethasone (DX) is a glucocorticoid drug, its side effects, which include oxidative stress, despite being used to treat inflammatory diseases. Therefore, the primary goal of the study is to evaluate the new DX/Zn drug complex's potent capacities with an increase in its antioxidant and antibacterial activities.

Methods: The DX/Zn novel complex was synthesized and characterized by means of magnetic susceptibility, SEM, TEM, elemental analysis and spectroscopic techniques (IR, ¹HNMR, UV and X-ray diffraction). Antioxidant activities were performed *via* a lot of *invitro* antioxidant assays (ORAC, ABTS and Metal chelation), antibacterial actions against *B. subtilis* and *E. coli*.

Result: DX is coordinated toward Zn ion as a monodentate ligand through the oxygen of (C=O). DX/Zn induced antioxidant activities via different antioxidant tests, it exhibits strong antibacterial properties against the bacterial strains *B. subtilis* and *E. coli*. Novel DX/Zn novel complex is an effective antioxidant and antibacterial agent.

Key words: Antibacterial activity, Antioxidant agent, Dexamethasone/Zn complex.

INTRODUCTION

Under the control of the hypothalamic-pituitary-adrenal axis, the adrenal cortex produces and releases steroid hormones known as glucocorticoids (Batista *et al.*, 2024). They influence various physiological processes through their interaction with the glucocorticoid receptor (Gans and Coffman, 2021). However, elevated body fat mass, osteoporosis, depression, infection risk, muscle atrophy and hyperglycemia can result from high GC levels (Vandewalle *et al.*, 2018).

Clinical use of glucocorticoid is essentially linked to lipid, protein and glucose disruptions that are repeatable in humans and rodents (Rafacho *et al.*, 2014; Burén *et al.*, 2008; Kaikaew *et al.*, 2019 and Schneider and Tappy *et al.*, 1998). The non-selective synthetic glucocorticoid, Dexamethasone (DX) is frequently for its anti-inflammatory, anti-allergic and immunosuppressive qualities. It has a potency that is between 50 and 100 times greater than cortisol. DX, however, causes adverse effects *in vitro* and *in vivo*, including muscle catabolism, increased adiposity, dyslipidemia and hyper-triacylglycerolemia, when used excessively (Kaikaew *et al.*, 2019; Ahrén, 2008; Cea *et al.*, 2016; Yoshikawa *et al.*, 2021; Asensio *et al.*, 2004; Ferreira *et al.*, 2017; Zuo *et al.*, 2008 and Barbera *et al.*, 2001). Furthermore, long-term DX administration produces free radicals like hydrogen peroxide that exacerbate oxidative stress (Bjelakovic *et al.*, 2007).

Oxidative stress, an imbalance between antioxidant and oxidant systems, leads to the production of free radicals during metabolic processes, which are neutralized

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by antioxidants. It is a significant factor in asthma pathogenesis. Novel antioxidant agents, like glucocorticoids, may be beneficial for asthma treatment due to their ability to inhibit smooth muscle hyper-contraction (El-Megharbel *et al.*, 2022). According to Li *et al.* (2018), oxidants can control the expression of multiple genes in eukaryotic cells, including those that control transcriptional, transduction and mRNA stability.

Oxygen radicals produced by inflammatory cells in the respiratory tract or exposure to outside air can contribute to

chronic obstructive airway diseases (Nounou *et al.*, 2010). These include apoptosis, mucous hypersecretion, membrane lipid peroxidation and damage to alveolar epithelium. Oxygen radicals can also damage proteins, lipids and DNA, leading to asthma pathology and affecting the normal physiological antioxidant system (Fujisawa, 2005).

Oxidative stress, an imbalance between oxidant and antioxidant systems, is crucial in asthma pathophysiology and severity (Rahman, 2005). Antioxidants could help treat asthma. Studies show that increased dietary vitamin C and vitamin E intake can lower asthma risk (Kirkham and Rahman, 2006). Combining antioxidant supplements with vitamin E can effectively reduce ozone-induced broncho constriction, thereby reducing the incidence of asthma (Fogarty *et al.*, 2003).

Corticosteroids affect airway vasculature through genomic and nongenomic mechanisms, with their beneficial effects in asthma treatment partly due to their ability to inhibit smooth muscle hypercontraction (Pearson *et al.*, 2005). This study suggests that combining antioxidant vitamin supplements with lower dosages of dexamethasone may reduce exposure to side effects of corticosteroids in patients with bronchial asthma (Nounou *et al.*, 2010).

Halliwel and Gutteridge (2007) define antioxidants as materials that prevent substrate oxidation even at low concentrations. Both humans and animals have an antioxidant defense system, including enzymes like catalase, glutathione reductase, GPx, SOD and non-enzymatic antioxidants like vitamins and glutathione. Oxidative stress occurs when these defense systems fail (El-Megharbel *et al.*, 2022).

Dexamethasone (DX) (Fig 1), a glucocorticoid hormone, is used to treat autoimmune and inflammatory diseases but has serious side effects like oxidative stress by (Feng and Tang, 2014). Long-term use decreases renal tissue's antioxidant capacity, increasing reactive oxygen

species formation. Dex inhibits endothelium-dependent vasodilatation of resistance arterioles by blocking endothelium nitric oxide synthetase (Bera *et al.*, 2005), leading to increased blood pressure.

Hence, the present study was conducted to evaluate the new capacities of a novel complex of dexamethasone and Zn metal as an antioxidant and antibacterial agent, aiming to demonstrate its potential physiological benefits in reducing free radical production and chelation.

MATERIALS AND METHODS

Dexamethasone and ZnCl_2 used in this manuscript were provided from sigma aldrich chemical company and all the analyses were performed in both labs of taif university (synthesis) and zagazig university (analysis) between march and July 2024; both are analytical reagent grade and didn't require any additional purification. Vario EL Fab. Elemental analysis instrument CHNS has been used for the C and H elements. The TGA technique was used to estimate the percentage of Zn (II). Using a Bruker infrared spectrophotometer, the zinc mixed ligand complex's FT IR spectrum was obtained in the $400\text{--}4000\text{ cm}^{-1}$ range. At room temperature with freshly prepared solutions, molar conductance's of 10^{-3} M solutions in DMSO solvent was measured using a HACH conductivity meter model. The electronic spectra was scanned in *situ* DMSO within $200\text{--}800\text{ nm}$ range by Unicam (Ultraviolet/visible spectrophotometer) UV/Vis spectrometer. Effective magnetic moment (μ_{eff}) of the zinc complex was measured using Gouy's method by the help of a magnetic susceptibility balance from Johnson Metthey and Sherwood model. ^1H NMR spectra measured using Bruker AM-500 NMR spectrometer.

Thermogravimetric analysis (TGA) experiment were measured using Shimadzu TGA-50H thermal analyzers. All experiments were performed using a single loose top loading platinum sample pan under N_2 atmosphere at a flow rate of 30 mL/min and a 10°C/min heating rate for the

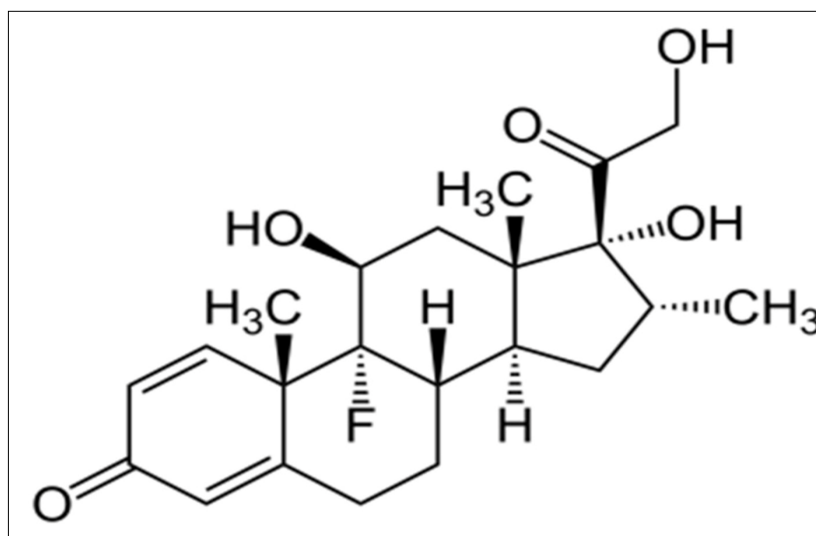


Fig 1: Dexamethasone structure.

temperature range 25-1000°C. SEM images were obtained using a Jeol Jem-1200 EX II Electron microscope at an acceleration voltage of 25 kV. The transmission electron microscopy images (TEM) were performed using JEOL 100s microscopy. The X-ray diffraction patterns were recorded on X 'Pert PRO PAN analytical X-ray powder diffraction, target copper with secondary monochromate.

Synthesis of zinc mixed ligand complex

The $[\text{Zn}(\text{DX})_2(\text{Cl}_2)] \cdot \text{H}_2\text{O}$ complex was prepared from ZnCl_2 and dexamethasone (DX) as explained below (Fig 2). A 30 ml methanol solution of DX (2 mmol) was added to an aqueous solution of ZnCl_2 (1 mmol). Then make stirring and heating at 60°C for one hour. Then total solution mixture was heated at temperature 80°C. The pH of the solution mixture is adjusted to 9 by using 1M ammonium hydroxide and immediately a novel complex was precipitated. Next, cool the mixture, filter the solid precipitate and wash with methanol. The prepared complexity was dried under vacuum. The solid brownish yellow product equal to a yield of 80% with (1:2) (Zn:DX) molar proportion.

Antioxidant activity

Assay of ORAC

DX it's metal complex antioxidant capacities with "Zn" metal, was carried out based on the method of Liang *et al.*, 2014 briefly; Dx and it's metal complex sample was incubated with 30 μL fluoresceine at 37°C for ~ 10 minutes. The fluorescence measurement was carried out for 3 cycles. Then, addition of 70 μL of (2-amidinopropane) dihydrochloride to each well immediately.

Metal chelation assay

The assay was conducted using the Santos *et al.*, 2017 method, which involved mixing 50 μL of DX/Zn with 20 μL of

FeSO_4 (0.3 mM). 30 μL of ferrozine was then poured into each well. After that, the plate was incubated for about ten minutes at 37°C. Next, at 562 nm, the color intensity started to decrease.

ABTS assay

The antioxidant assay was carried out based on Arnao *et al.*, 2001, 192 mg of ABTS were dissolved in dist. H_2O . 1 mL of the previous solution was added to 17 μL of $\text{K}_2\text{S}_2\text{O}_8$ and the mixture was left for 24 hrs in dark. Then, 1 mL of the mixture was completed with methanol to obtain the final ABTS dilution. 190 μL of the ABTS reagent was mixed with the sample, the reaction was incubated at 37°C. The notable decrease in ABTS intensity, measured at 734 nm, at the conclusion of the dark incubation period.

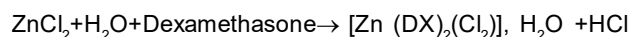
Antibacterial activity against *Escherichia coli* and *Bacillus subtilis*

Bacillus subtilis (ATCC 6633) and *Escherichia coli* (ATCC 8739) discs were injected into the broth medium and incubated for a day at 30°C. An agar medium was lightly streaked with a scoop of broth using a culture agar plate and the mixture was then incubated at 30°C. The control well was added to the plate without any DX or its Zn complex. A control well designated as (-ve) that holds the broth medium (El-Megharbel *et al.*, 2024).

RESULTS AND DISCUSSION

Microanalytical and molar conductance values

The preparation of the Zn (II) dexamethasone complex occurred as the following equation:



Zn(II) chloride and dexamethasone ligand chelate to form a 1:2 metal:dexamethasone complex known as

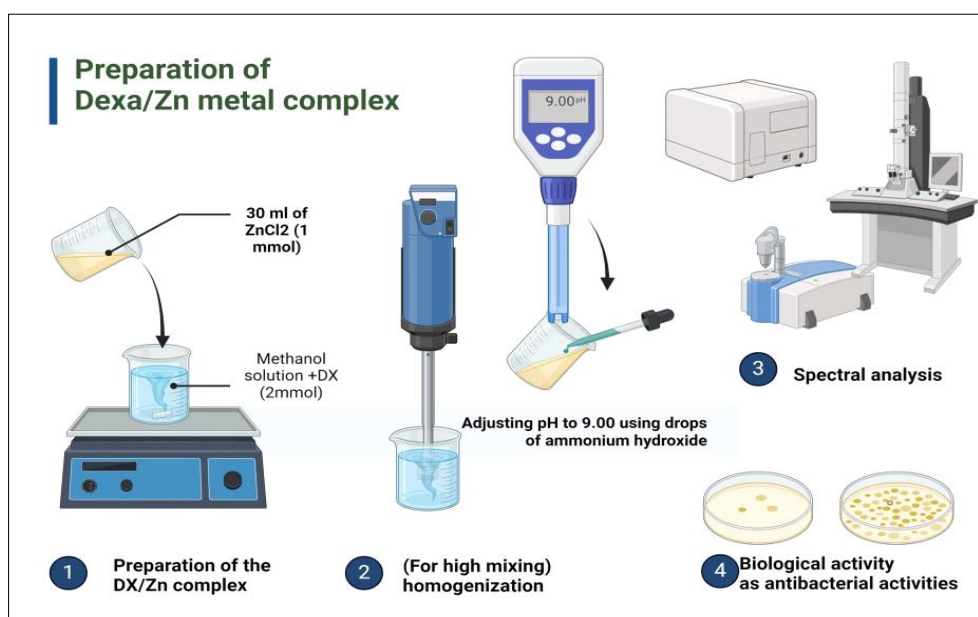


Fig 2: Experimental procedures.

$[\text{Zn}(\text{DX})_2(\text{Cl})_2] \cdot \text{H}_2\text{O}$. The zinc complex that is synthesized is a stable solid that is soluble in water but varies in solubility in common organic solvents. The complex is painted a pale-yellow hue. The thermal stability of the solid zinc complex suggests a robust metal-ligand bond. The zinc complex physical microanalytical data agrees with the general formulation $\text{Zn} \cdot \text{DX}_2 \cdot \text{Cl}_2 \cdot \text{H}_2\text{O}$. At a concentration of 10^{-3} M in dimethyl sulfoxide (DMSO), the complex's molar conductance value is $\mu\text{m} = 24$ ($\Omega^{-1} \text{mol}^{-1} \text{cm}^{-1}$) suggesting that it is not electrolytic in nature (Nakamoto., 1970, Al-Salmi *et al.*, 2023; Hamza *et al.*, 2022). This can be attributed to the presence of chloride ions inside the chelation sphere. The molar conductance data and the elemental analyses of C, H and Zn agree well with the proposed structure of $[\text{Zn}(\text{DX})_2(\text{Cl})_2] \cdot \text{H}_2\text{O}$ is the formula for the complex $\text{Zn} \cdot \text{DX}_2 \cdot \text{Cl}_2 \cdot \text{H}_2\text{O}$. Dexamethasone ligand's coordination modes with Zinc (II) metal ions were examined in relation to molar conductance, electronic spectra, magnetic moment and infrared spectra.

Infrared spectra

The Fourier transform infrared spectroscopy (FT-IR) spectrum for the representative complex, $[\text{Zn}(\text{DX})_2(\text{Cl})_2] \cdot \text{H}_2\text{O}$ is shown in Fig 3. For dexamethasone free ligand (Fig 3A)

the ring carbonyl absorption frequency appeared at 1690 cm^{-1} , this band is shifted to lower wave number at 1674 cm^{-1} . In the free dexamethasone the $\nu(\text{OH})$ vibrational stretching bands appeared at 3266 cm^{-1} , where this band appeared without shifting in the IR spectrum of mixed ligand complex, confirming that OH group not participated in chelation with the Zinc ion.

The $[\text{Zn}(\text{DX})_2(\text{Cl})_2] \cdot \text{H}_2\text{O}$ complex (Fig 3B) has new bands within wavenumber $660\text{--}533 \text{ cm}^{-1}$, these bands are assigned to the $\nu(\text{M-O})$ stretching vibration motions (Bellamy, 1975 and Sachan *et al.*, 2012), therefore the DX ligand is coordinated toward zinc ion as monodentate ligand through oxygen of the ring carbonyl group.

UV-Vis spectra and magnetic data

The UV-Vis spectrum of the free dexamethasone showed an absorbance band in the UV region at 245 nm , which can be assigned to the $\pi \rightarrow \pi^*$ transition. The electronic absorption spectra for $[\text{Zn}(\text{DX})_2(\text{Cl})_2] \cdot \text{H}_2\text{O}$ complex has three types of absorption bands at 255 nm , 340 nm and

Table 1: UV-Vis data of dexa and dexa/zn.

Compound	$\pi \rightarrow \pi^*$	$n \rightarrow \pi^*$
Dexamethasone	245	335
$[\text{Zn}(\text{DX})_2(\text{Cl})_2] \cdot \text{H}_2\text{O}$	255	340, 370 nm

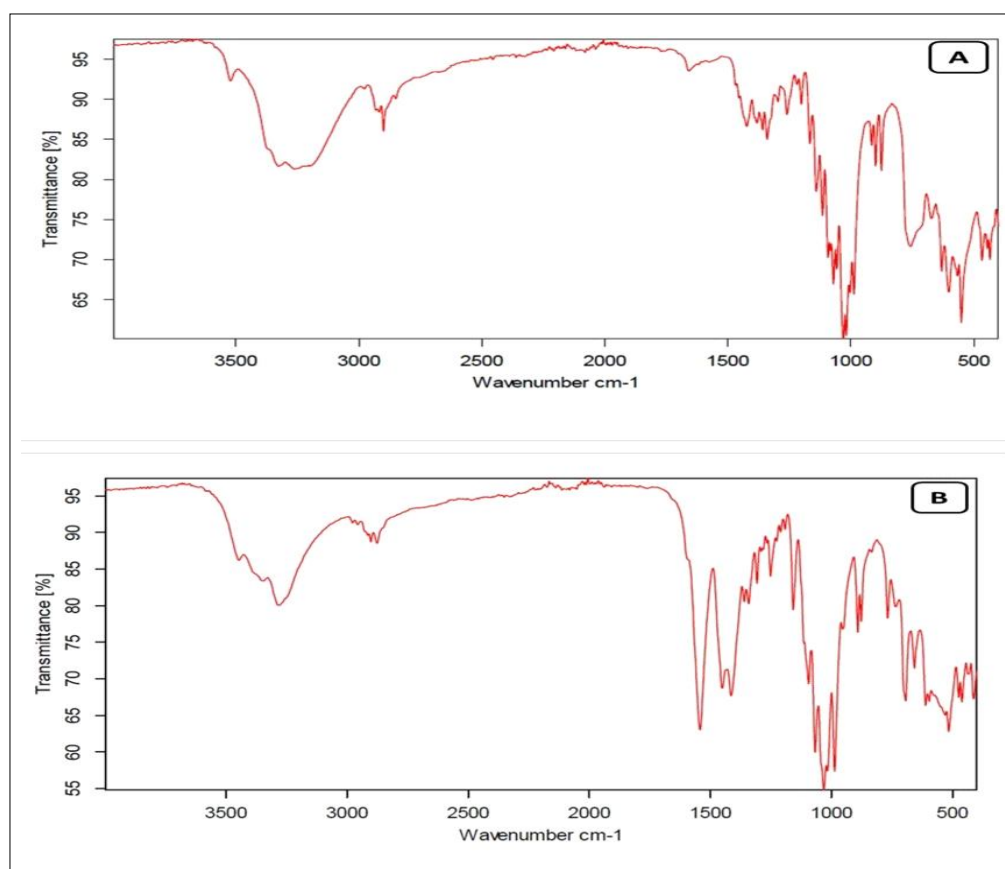


Fig 3: FT-IR of (A) DX, (B) DX/Zn.

370 nm due to $\pi \rightarrow \pi^*$ and $n \rightarrow \pi^*$ electronic transitions respectively (Table 1) and considered as diamagnetic.

X-ray powder diffraction

X-ray powder diffraction patterns that provide a crystallinity explanation. Utilizing X-ray powder diffraction patterns, the nano-structural form of $[\text{Zn}(\text{DX})_2(\text{Cl})_2] \cdot \text{H}_2\text{O}$ complex was examined within the diffraction angle range of (2θ) 0-100°. The X-ray powder diffraction (XRD) patterns revealed that the Zinc complex is amorphous as shown in Fig 4.

(SEM) Scanning electron microscopy and (TEM) Transmission electron microscopy studies

The $[\text{Zn}(\text{DX})_2(\text{Cl})_2] \cdot \text{H}_2\text{O}$ SEM pictures. Fig 5 displays the complex. The images under investigation for surface morphology included some with regular grains and many with irregular shapes. The $[\text{Zn}(\text{DX})_2(\text{Cl})_2] \cdot \text{H}_2\text{O}$ TEM pictures. Fig 6 displays the nanoparticles that were produced when one mole of DX and zinc chloride salt reacted. Following

the full formation of new $[\text{Zn}(\text{DX})_2(\text{Cl})_2] \cdot \text{H}_2\text{O}$, the particles have spherical black spots and range in size from 20 to 45 nm.

^1H -NMR spectra

The ^1H -NMR spectra of the Zn(II) complex and free DX were found in Fig 6A,B and DMSO- d_6 . A distinct signal was observed in the free DX spectrum at $\delta(\text{ppm}) = (\text{CH}, 1.45 \text{ and } 2.70)$, $(\text{OH alcohol}, 5.07)$ and $(\text{OH}-\text{CH}_2, 6.63)$. After chelating with DX, OH groups did not change, as evidenced by the similarity between the H of the OH in the ^1H NMR spectra of DX and its Zn(II) complex. As a result, the coordination did not involve the OH group (Fig 7).

Analyses using thermogravimetric test

The DX/Zn complex was subjected to thermogravimetric and differential thermogravimetric analysis (TGA-DTG) at 1000°C, as illustrated in Fig 5. The Zn(II) complexity has been observed to exhibit weight loss up to 140°C, indicating the loss of crystalline water and confirming the presence

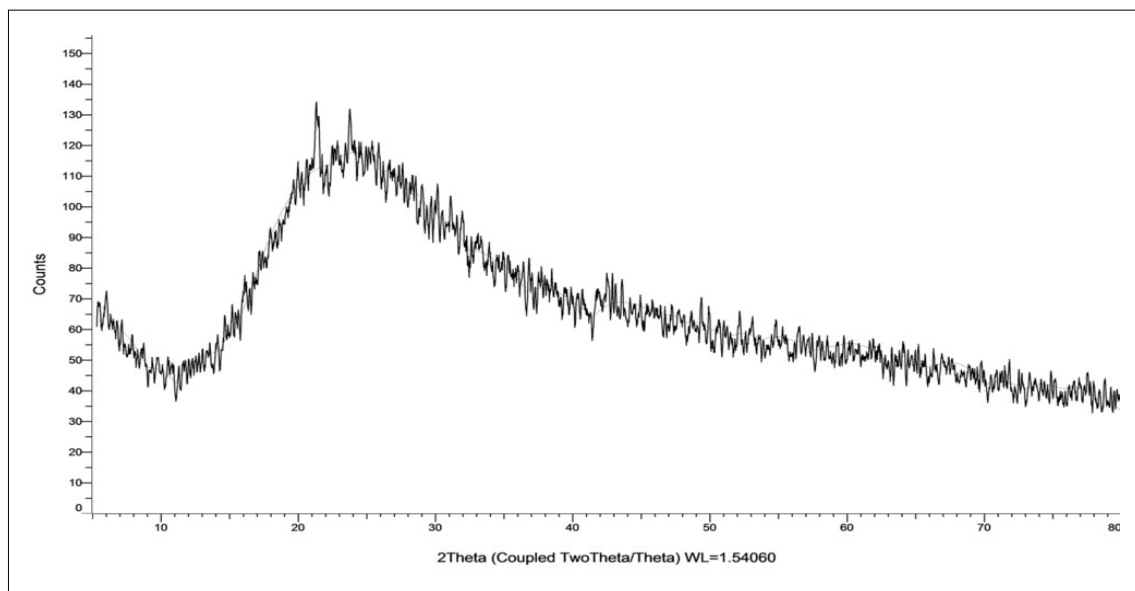


Fig 4: XRD of DX/Zn novel complex.

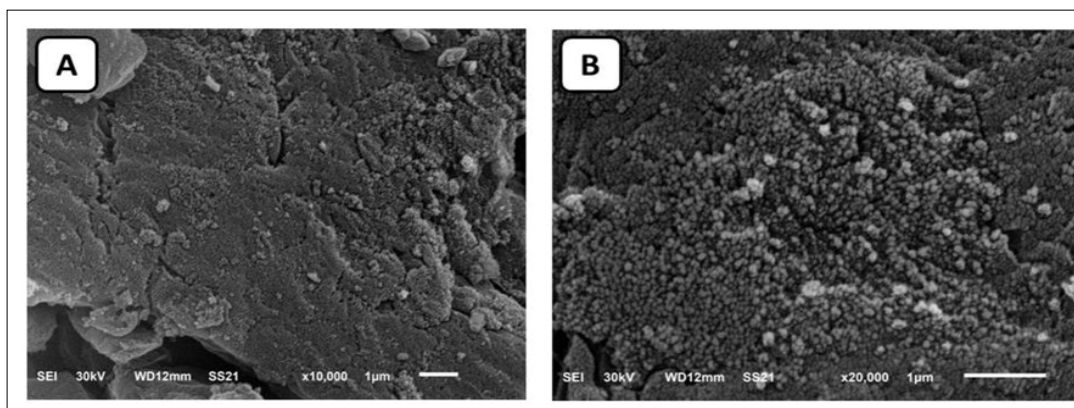


Fig 5: SEM sections of (A): DX, (B) DX/Zn.

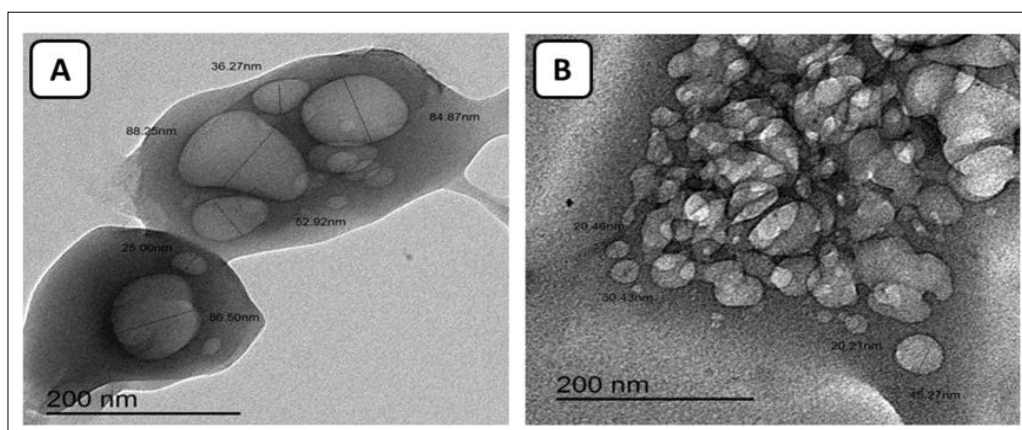


Fig 6: TEM sections of (A): DX, (B) DX/Zn.

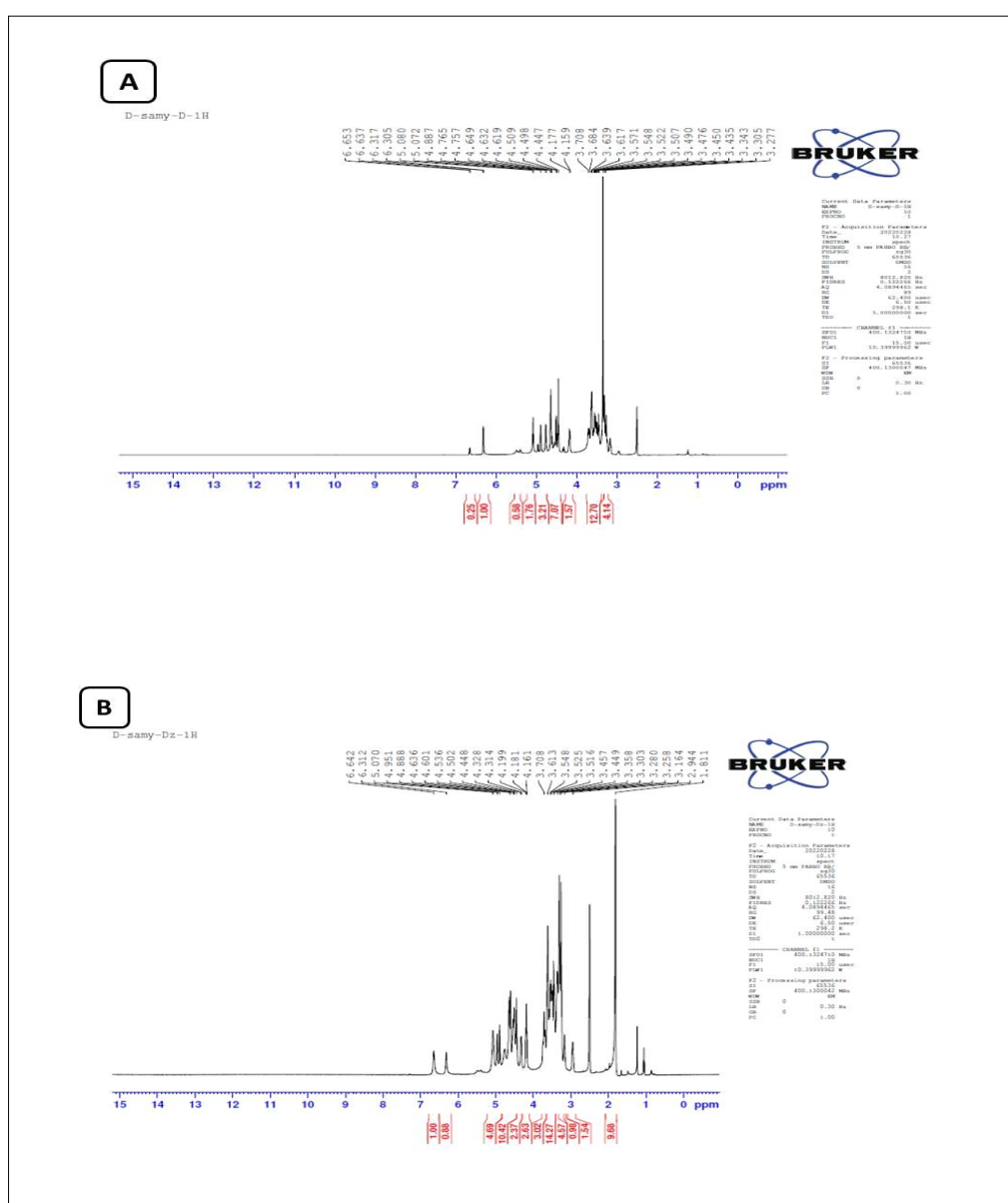


Fig 7: ^1H -NMR of (A) DX, (B) DX/Zn.

of external water molecules. The Zn(II) complex thermal analysis curve indicates a weight loss up to 496°C, indicating that the weight loss for Zn(II) complex is related to the breakdown of the DX ligand. There are five steps in the thermal breakdown process for zinc complexity. As can be seen in (Fig 8), the thermogram of Zinc complexity revealed that ZnO was the most stable final product tainted with some carbon atoms.

Antioxidant capacities of Dexamethasone/Zn (DX/Zn) metal complex

Fig 9 displays the percentages of chelating activity determined by three distinct techniques. ABTS, metal chelation assay and oxygen Radical Absorbance Capacity (ORAC) as a measure of the hydrophilic antioxidant capacity of the DX metal complex with zinc were employed as free radical scavenging techniques. As seen in (Fig 9), the DX/Zn complex had a greater ability than DX alone to scavenge the ABTS, ORAC and chelating activity radical. The current results show that DX/Zn has a higher free radical absorbance activity than DX alone, which is extremely encouraging. This observation may be explained by DX/Zn's high lipophilic activity.

Antibacterial activity evaluation

Biological evaluations of the target complex (DX/Zn) was performed on Gram-positive (*Bacillus subtilis*) and Gram-negative (*Escherichia coli*) bacteria. The results of the antimicrobial activities of the DX and/or its metal complex is presented in Fig 10. The inhibition concentrations of the DX metal complex (samples with the same concentration) against both Gram-positive and Gram-negative bacteria (*B. subtilis* and *E. coli*) were found to be high at low concentrations. Based on the standard conditions, the DX complex was found to be sufficient, with high antimicrobial activity as compared to DX treatment only (Fig 10).

Glucocorticoids, steroid hormones or synthetic compounds used as anti-inflammatory drugs, are linked to

inflammation and increased production of oxygen reactive species (ROS) (Liu *et al.*, 2018 and Kenanidis *et al.*, 2015). These ROS, derived from biological aerobic metabolism, destroy proteins, lipids and DNA, affecting cell function. Prolonged or excessive use of glucocorticoids can lead to necrosis and oxidative stress, affecting cell function. Recent research confirms the production of novel glucocorticoid metal complexes, such as the DX/Zn novel complex, with potent antioxidant activities, which could potentially mitigate the negative effects of oxidative stress. Bronchitis is a common symptom of this condition.

Oxidative stress, an imbalance between antioxidant and oxidant systems, leads to the production of free radicals during metabolic processes, which are neutralized by antioxidants. Overproduction can interfere with cell regeneration and repair, leading to accelerated aging and certain diseases. Oxidative stress is also crucial in the development of chronic diseases like cardiovascular disease, diabetes, neurodegenerative diseases and cancer. Novel antioxidant agents may be beneficial for treating asthma, as they can inhibit smooth muscle hyper-contraction when administered systemically with fewer side effects when complexed with novel transition metals (Nounou *et al.*, 2010; Fujisawa, 2005). Accordingly, it was concluded that the treatment of mother with betamethasone prior to fertilization, adversely affect the heart of newborn rabbits by biochemical, histological and ultrastructural changes (Al Nwaiser *et al.*, 2023). So, according to the above mentioned, it is recommended to evolve new metal drug complexes categories to stop the serious alterations in the heart of the off spring.

Inflammatory cells in the airways or direct inhalation of outside air can produce oxygen radicals (Fujisawa, 2005). Important insights into the pathophysiology of chronic obstructive airway diseases can be gained from oxidative stress. These include apoptosis, mucous hypersecretion, membrane lipid peroxidation, oxidative inactivation of antiproteases and surfactants, mitochondrial respiration,

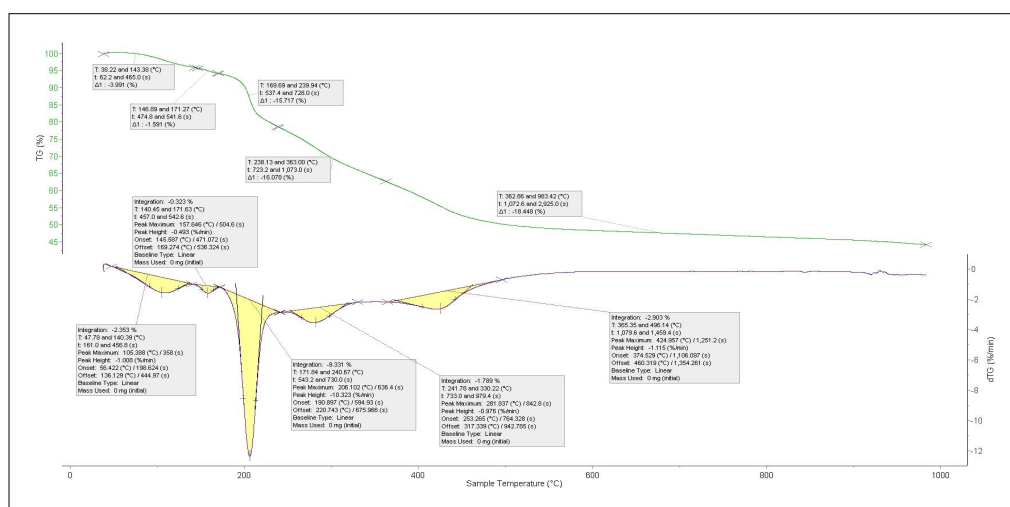


Fig 8: TGA/DTG of DX/Zn.

damage to the alveolar epithelium, extracellular matrix remodeling and mitochondrial respiration (Barnes, 2020; Horvath, 2006 and Chiba *et al.*, 2008).

Zinc is essential for maintaining normal immunophysiological performance in animals, as it is not stored in

the body. Zinc is an antioxidant that lowers reactive oxygen species generation, maintains membrane stability and prevents lipid peroxidation. Cu-Zn superoxide dismutase, a vital enzyme, requires zinc as a cofactor. A study aimed to create a new DX/Zn complex to validate its strong

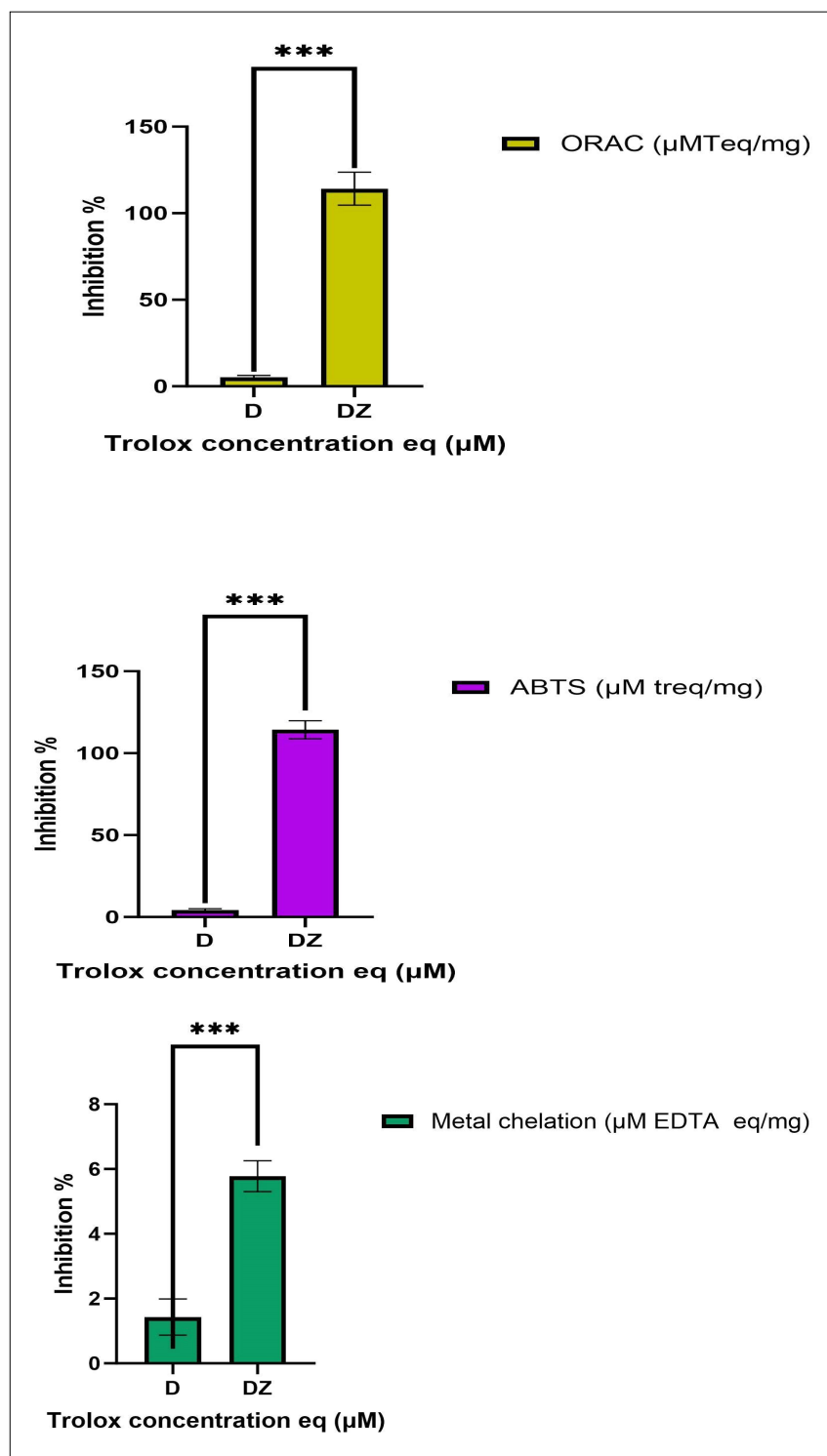


Fig 9: Antioxidant capacities of DX/Zn novel complex.

antioxidant and antibacterial properties *in vitro* and mitigate potential adverse effects of DX (Manimaran *et al.*, 2024., Deori *et al.*, 2024 and Jain *et al.*, 2024).

Zn, like other metals like Fe and Cu, is crucial for prokaryotic and eukaryotic cellular functions and protein activity. Controlling the amount of Zn attached to various molecules may impact the activity of proteins that require Zn (Sudhakar *et al.*, 2025). Zn regulation may be influenced by a number of membrane-associated Zn transporters that have been discovered (DeMoor *et al.*, 2001). Thus, the idea of chelation of zinc to DX drugs was evolved to increase DX's cellular capacities and decrease potential adverse effects. Sudhakar *et al.* (2025) previously reported that supplementation of zinc from different sources affects blood chemistry and pathological condition in RIR birds. This is because the current research has shown that zinc effectively increases DX's capacities and produces new antioxidant activities, confirming the critical role of zinc in activating DX's capacities (DeMoor *et al.*, 2001).

Singh and Singh (2002) study found that daily injections of glucocorticoids like DX increased zinc uptake in neonatal buffalo calves. The subcellular fractions of liver and muscle showed an increase in zinc concentration, while the

cytosolic fraction showed an 80% increase. This suggests a redistribution of zinc in the body, confirming the importance of complexation of zinc with DX drug for novel antioxidant capacities and antibacterial activities and as Girdhar *et al.*, 2021 reported previously that zinc (Zn) is one of the most essential minerals.

Previous study indicated alterations in Zn metabolism and homeostasis in neonatal buffalo calves, who were under constant stress due to continuous administration of glucocorticoid (dexamethasone) injection. Singh and Singh (2002) confirmed that in the DX treated group, the weekly plasma Zn concentrations were significantly lower than the control group averages. This underscored the significance of complexing ZN with DX, which will be a magic solution for alleviating any glucocorticoid side effects and compensating for the decline in its cellular level and thus greatly elevate antioxidant capacities.

The current study supports evidence from previous study Toledano *et al.* (2023) who confirmed that the presence of DX in the NPs improved the mechanical performance when compared with the undoped-NPs such as the nano-range of DX/Zn complex in the current study as the particles appeared with spherical black spots and range in nano-size from 20 to 45 nm based on TEM analysis.

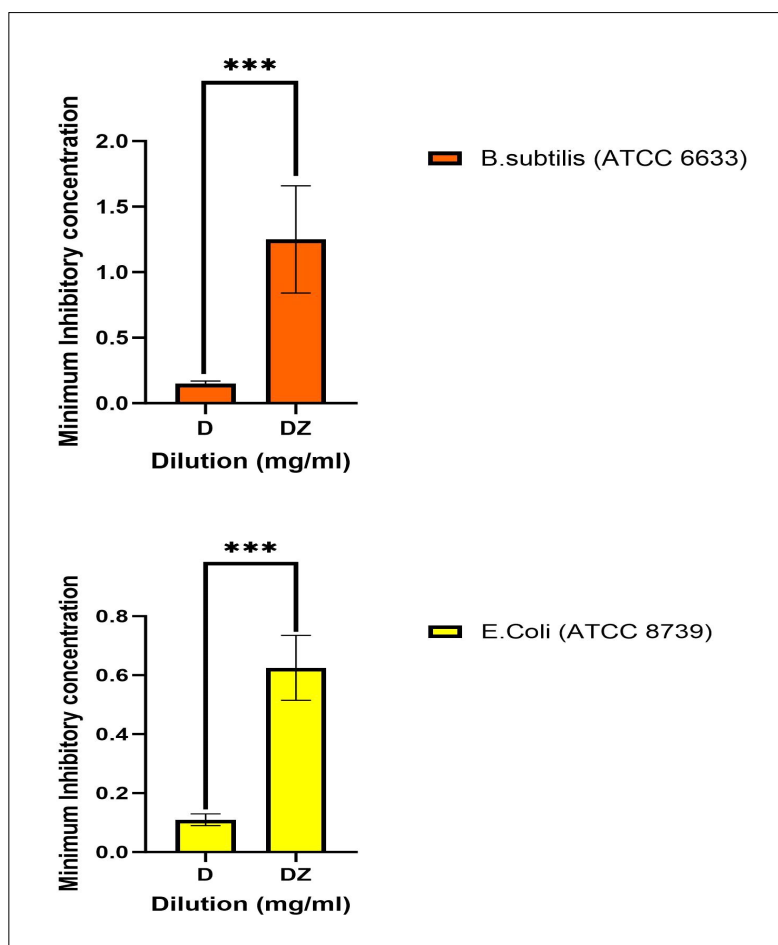


Fig 10: Antibacterial activities of both DX and DX/Zn.

Additionally, the need for DX treatment for diabetics in COVID-19 pandemics is another key idea for this study. As previously confirmed in (Eucilene *et al.*, 2024), diabetics are the most susceptible patients due to lowered immune functions and the expression of specific receptors in diabetics that are sensitive to SARS-CoV-2. Thus, recent studies have confirmed the same idea by confirming the role of oxivanadium as a treatment for declining insulin resistance induced by DX in mice, which revealed better results in treatment in case of combination of DX with other transition metals like the current study and confirmed the oxidative stress induced by DX alone (Eucilene *et al.*, 2024). The results offer promising data for enhancing DX's efficacy when chelated with other metals, reducing stress and may be tested for potential new capacities.

The current study showed potent antibacterial activity of the DX/Zn novel complex, which is in accordance with the previous finding of Neher *et al.* (2008), which confirmed that DX killed *S. milleri* and *A. flavus*, but when DX was combined with N-chlorotaurine in low concentration, it led to a 90% reduction of *S. aureus* and *P. aeruginosa* within 30 minutes and a 99.9% reduction within 50 minutes. Which confirmed the same concept of the current study, as DX chelated with Zn showed high antibacterial activity against the bacterial strains *B. subtilis* and *E. coli* at low concentrations, as the combination of DX and Zn showed significantly stronger antimicrobial effects, which might be a promising therapeutic option, producing high efficacy with low side effects.

Thus, the current study confirms the novel complex of DX with Zn as a potent antioxidant agent, scavenging and capturing excessive free radical production and showing high antibacterial activities against both gram positive and gram-negative bacteria.

CONCLUSION

The study highlighted synthesis of novel DX/Zn complex and its characterization using spectroscopic techniques such as IR, ¹HNMR, UV, X-ray diffraction, elemental analysis, magnetic susceptibility, SEM and TEM. The study focused on the synthesis of a novel DX/Zn complex. Through metal chelation, ORAC and ABTS tests, this novel complex reduced the oxidative stress that was induced *in vitro*. It also demonstrated strong antibacterial activity against *B. subtilis* and *E. coli*.

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Conflict of interest

All authors confirm that there is no conflict of interest.

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