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Chitosan-Stabilized Copper Nanoparticles: Synthesis, Characterization, and Anticancer Potential in Pancreatic Cancer Cells

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Abstract

It is an excellent polymer because of its high capping and stabilizing, biocompatibility, biodegradability, environmental friendliness, poly cationic, and low toxicity that has been widely utilized as a support in the synthesis of metallic nanoparticles (MNPs). The nanoparticles made of metallopolymeric chitosans may be prepared through different chemical methods, such as sol-gel, co-precipitation, and hydrothermal procedures, each possessing its own strengths of regulating particle size, shape and homogeneity. The current research used the solution-casting method to produce copper-chitosan nanoparticles (CS-CuNPs). These particles were then systematically investigated in order to test their physicochemical and biological characteristics. The produced CS-CuNPs were characterized by a stable brick-red color and a clear UV-visible absorption peak at 214 nm, which are evidence of the creation of nanomaterials and the availability of surface plasmon resonance. The X-ray diffraction (XRD) studies revealed a crystalline face-centered cubic (fcc) structure that has big diffraction peaks at 36.78°, 43.38°,

50.56°, and 74.26°. The particle size based on the atomic force microscopy (AFM) was 62.45 nm on average. Fourier transform infrared (FTIR) spectroscopy revealed the existence of strong interactions between chitosan functional groups and copper ions with distinctive absorption bands at 3370-3226 cm^{-1} , 1633 cm^{-1} , and 680 cm^{-1} . Further testing was done by measuring the zeta potential (ZP) and SEM analysis to determine the stability of the colloidal particle and surface integrity of the cell lines studied and revealed that when the biological evaluation was taken into account, CS-CuNPs had much anticancer efficacy. Conversely, the cytotoxic responses of the pancreatic cancer cells were a little less encouraging, which indicated that the efficiency of nanotechnology depends on the cell line. The CS-CuNPs showed high cytotoxic and anticancer activities at a concentration of 25 $\mu\text{L/mL}$, which portrays the potential of the new nanotherapeutic agent in cancer treatment and this is against the breast cancer cells. Graphical abstract Fig 1.

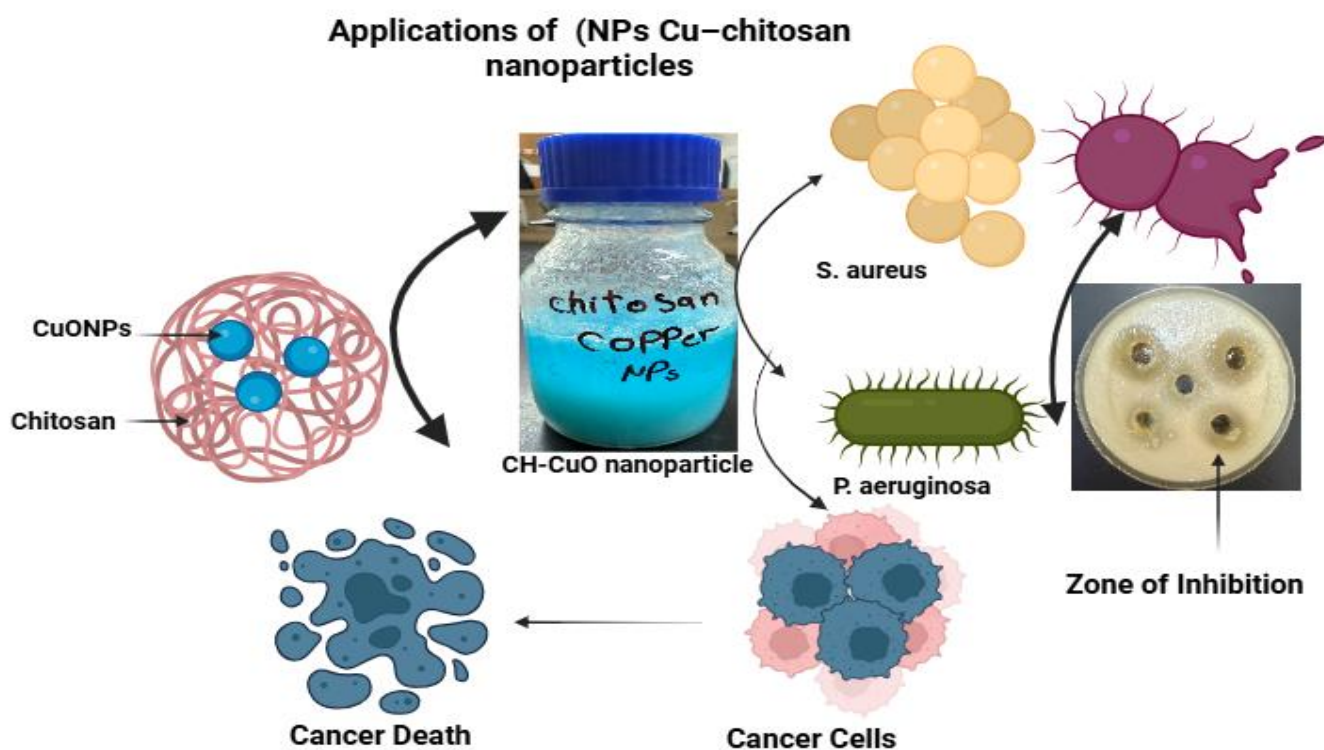


Figure 1 : Graphical abstract Applection of Chitosan-copper nanoparticles

Keywords: Chitosan, X-ray diffraction (XRD), Pancreatic Cancer Cells.

Introduction

Natural polymers such as chitin, cellulose, and starch have attracted considerable research interest over the past decade because of their diverse applications in areas such as signaling, energy storage, transportation, and structural materials [1]. Chitosan, a linear polymer derived from the deacetylation of chitin, is primarily obtained from the shells of shrimp, prawns, and crabs. Its excellent biocompatibility and biodegradability, together with its ability to interact with macromolecules and biological systems, have contributed to its widespread application in biomedical and sensing technologies. These properties, along with its physiological compatibility and versatile functional characteristics, make chitosan an attractive biomaterial for various applications [2].

The use of chitosan and its derivatives has expanded considerably in recent years, with applications in high-energy battery components, biosensors, actuators, artificial muscles, environmentally responsive membranes, wastewater treatment, and electrolytes [3]. In addition, emulsified and cross-linked chitosan nanoparticles have been investigated as potential carriers for anticancer drug delivery. Since their initial development, chitosan-based nanoparticulate systems have been extensively studied for controlled and targeted drug delivery, and various formulations incorporating different matrix-forming materials have been developed [4]. Copper nanoparticles (CuNPs) have also attracted considerable attention because of their favorable physicochemical properties, particularly their high thermal and electrical conductivity. In addition, copper is relatively inexpensive compared with noble metals such as gold and silver, making copper-based nanomaterials attractive for a range of technological and biomedical applications. For example, Esteban-Cubillo et al. reported a bactericidal effect of copper nanoparticles prepared using a sepiolite scaffold [5]. Chitosan is a naturally occurring linear biopolyaminosaccharide that has gained considerable attention as a versatile biopolymer in the food, medical, cosmetic, and pharmaceutical industries. In the present study, shrimp shells, representing an abundant and sustainable source of chitin, were used as the starting material for chitosan production. The resulting chitosan was subsequently employed in the fabrication of copper oxide nanoparticles. The preparation and characterization of chitosan-based nanoparticles are

of considerable interest because of their broad potential applications in biological and non-biological systems [6]. Previous studies have reported that chitosan–copper oxide (CH–CuO) nanoparticles can exhibit antimicrobial and antibiofilm activities. Furthermore, phase-contrast microscopy has demonstrated pronounced morphological alterations in treated cells compared with untreated controls, providing additional evidence of cellular damage following nanoparticle exposure [6]. Chitosan also possesses inherent antioxidant and antimicrobial properties, which may complement the functional characteristics of CuO nanoparticles and potentially enhance their biological activity. Accordingly, chitosan has been employed as a stabilizing, coating, and functionalizing agent in the synthesis of various metal oxide nanomaterials, highlighting its versatility in nanomaterial development [7]. CuO nanoparticles possess distinctive physicochemical and biological properties that have attracted substantial scientific interest. Their potential applications are associated with characteristics such as high stability, photocatalytic activity, thermal conductivity, relatively low cost, and reported antibacterial, anticancer, and anti-inflammatory activities. Incorporation of CuO nanoparticles into chitosan matrices may therefore produce chitosan–CuO nanocomposites with complementary or synergistic properties, potentially enhancing their antimicrobial performance while benefiting from the biocompatibility and functional properties of chitosan [8]. CuO is considered a promising inorganic antimicrobial agent, particularly when combined with biopolymers possessing complementary biological and physicochemical properties. The combination of CuO with chitosan may result in nanocomposite systems with enhanced antimicrobial activity through interactions between the constituent components [9]. In addition to contributing to the antimicrobial properties of the composite, chitosan can improve the dispersion and biocompatibility of CuO nanoparticles and may function as both a stabilizing and capping agent during nanoparticle synthesis. Overall, chitosan–CuO nanocomposites have demonstrated considerable potential for applications in biomedical, pharmaceutical, food, and environmental fields. Their biological activity and physicochemical versatility make them promising candidates for further investigation, particularly in antimicrobial and nanomedicine-related applications. However, their therapeutic potential should be evaluated through appropriate **in vitro and in vivo studies** before any clinical or therapeutic conclusions can be drawn.

Material and methods

Preparation of Chitosan

The chitosan was made according to Mallick et al. (2012) [10]. Concisely, 1.0g chitosan was dissolved into 100mL of 1% (v/v) acetic acid solution by stirring continuously until a clear solution resulted. Fig 2.

CS-Cu Nanoparticles can be synthesized in the following way.

The procedure outlined by 11 was done to produce chitosan-copper nanoparticles (CS-Cu NPs). Simply put, 50 mL of 5% (v/v) acetic acid solution was combined with 3% (w/v) chitosan at room temperature, which was constantly stirred. 25 mL of 1 M ascorbic acid was put into the solution in order to decrease pH to 3.5 and achieve a clear blue solution. 0.1 M copper(II) acetate was dissolved in 20mL of distilled water separately. A continuous 60-min stirring at 60 °C was done when adding copper solution slowly to the chitosanascorbic acid mixture. The abrupt emergence of a brick-brown color was an indication of the creation of CS -Cu NPs.

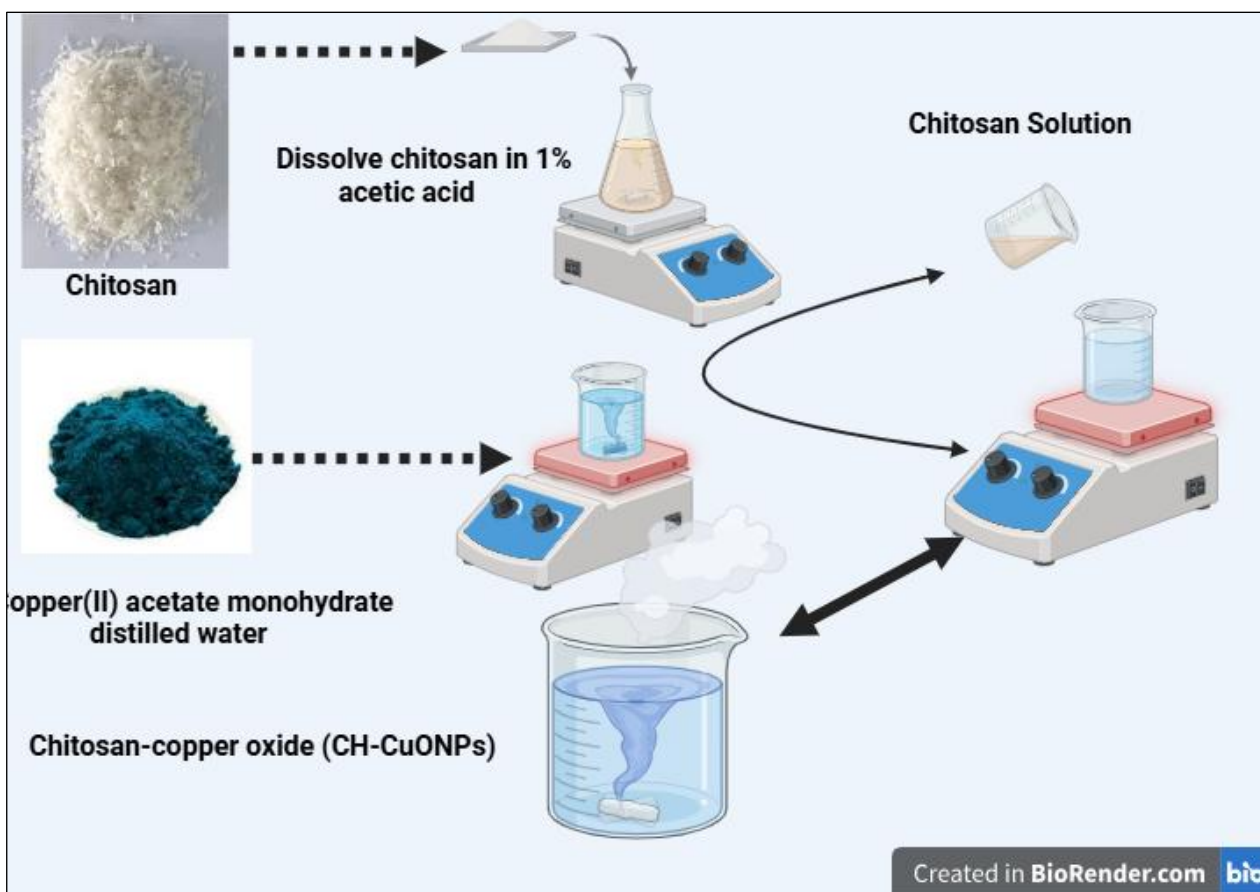


Figure 2 : Synthesis of Chitosan-copper nanoparticles

Characterization of Chitosan-copper nanoparticles

The functional groups included in the generated nanoparticles were detected using Fourier Transform Infrared Spectroscopy (FTIR) (Bruker, Ettlingen, Germany) spanning a spectral range of $400\text{--}4000\text{ cm}^{-1}$. Utilizing UV-visible spectroscopy (UV/Vis-1800, Shimadzu, Kyoto, Japan), the optical properties of the nanoparticles in the $200\text{--}600\text{ nm}$ wavelength range were investigated. The form and surface topography of the nanoparticles were examined using atomic force microscopy (AFM) and scanning electron microscopy (SEM; Hitachi S-3400N) with a resolution of 500 nm . The crystalline structure of the nanoparticles

was investigated using X-ray diffraction (XRD), and their elemental composition was verified using Energy Dispersive X-ray (EDX) spectroscopy [12].

Minimum Inhibitory Concentration

To find out the minimum quantity of AMB required to prevent the visible development of microorganisms, a MIC experiment was conducted. The Clinical and Laboratory Standards Institute (CLSI) microdilution protocol was followed for conducting the test. The prepared nano-antibiotics were serially diluted two times on a 96-well microtiter plate, yielding concentrations ranging from 64 $\mu\text{g/mL}$ in the first column to 0.125 $\mu\text{g/mL}$ in the tenth column. Column 12 was the negative sterility control, which included just the culture media, and Column 11 was the positive growth control, which contained the microbial inoculum but no antimicrobial agent [13].

Cells, cell culture, and CS-Cu NPs preparation

The CAPAN-2 human pancreatic cancer cell line was used in this investigation. As previously reported [9], cells were grown in Roswell Park Memorial Institute-1640 (RPMI-1640) media supplemented with L-glutamine (Capricorn Scientific GmbH). RPMI-1640 was supplemented with 10% (v/v) fetal bovine serum (FBS) and 1% (v/v) penicillin–streptomycin solution (100 $\times$; Euroclone S.p.A.) to create the full culture medium. In a humidified incubator with a 5% CO_2 environment and 95% relative humidity, cells were kept at 37 $^{\circ}\text{C}$. The produced nanoparticles were first dispersed in deionized (DI) water to provide a stock solution for in vitro tests. Complete culture medium was used to dilute the stock solution in order to provide the new working concentrations needed for the next tests [14].

Cytotoxicity assay.

To assess harmful effects, an in vitro 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) assay was employed [10]. Each well of a 96-well plate contained 7,000 cells, which were incubated for the whole night to ensure cell adhesion. Following that, cell lines (MCF7) were exposed to compounds at increasing concentrations of CS-Cu NPs

(6.25, 12.5, 25, 50, and 100 µg/ml); each treatment was carried out in three duplicate wells. After the media was removed from the plate after a 24-hour incubation period, each well received 20 µl of MTT solution (5 mg/ml) from Shanghai Macklin Biochemical Co., Ltd. Following that, the wells were incubated for three hours at 37°C in the dark. The MTT was stirred for 10 minutes after 50 µl of DMSO (Bio Basic Inc.) was added to dissolve it [15]. Absorbance at 490 nm was measured using a microplate reader (BioTek Instruments, Inc). The fraction of live cells was calculated from the raw absorbance data using the algorithm below.

$$\text{Viability (\%)} = (\text{Absorbance of treated cells} / \text{Absorbance of control cells}) \times 100$$

The dose-response curve was made using GraphPad Prism software version 6 (Dotmatics), where the absorbance is represented by "A." The growth inhibitory concentration (GI50), which reduces viability by 50%, was determined using the same curve [16].

Results

Synthesis of Chitosan-copper nanoparticles

The morphology, functionality, and overall performance of the created copper nanoparticles are largely determined by their physicochemical characteristics, particularly are reflected in their optical behavior. The aqueous reaction mixture exhibited pronounced colour changes during the synthesis, which acted as qualitative indicators of nanoparticle formation. As chitosan is most soluble in an acidic environment, the chitosan–acetic acid solution was milky white and colour of this solution did not change when ascorbic acid was introduced. The greenish-yellow of the

reaction mixture turned to a very stable brick-brown upon adding some pale blue colour to copper(II) acetate solution, which suggests formation of copper nanoparticles. For the creation of metallic nanoparticles employing affordable experimental settings, chemical reduction of metal salts has been accepted as a viable and scalable method. Copper salts were used as the precursor material in this procedure, and chitosan coupled with Cu^{2+} ions to act as a stabilizing and capping agent. By acting as a reducing agent, ascorbic acid prevented the unwanted formation of copper hydroxide $[\text{Cu}(\text{OH})_2]$ and copper oxides (Cu_2O , CuO) while additionally assisting in the conversion of Cu^{2+} to metallic Cu^0 . When the copper solution was added to the chitosan–ascorbic acid mixture while continuously stirring it at 60°C for 60 minutes, a homogenous grayish-brown colloidal suspension appeared, which further demonstrated the effective manufacturing of chitosan–copper nanoparticles. A fine nanoparticle powder was produced by drying the ensuing colloidal nanoparticles using microwave irradiation. Fig 3.

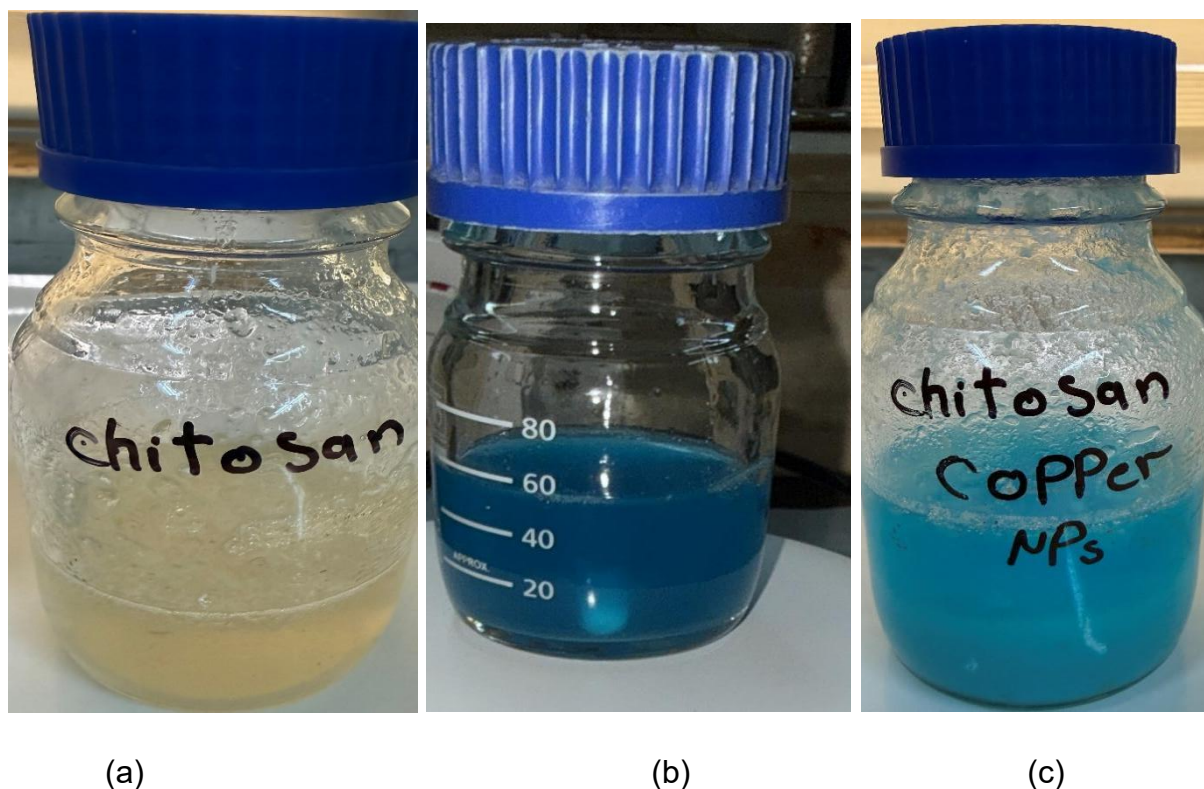
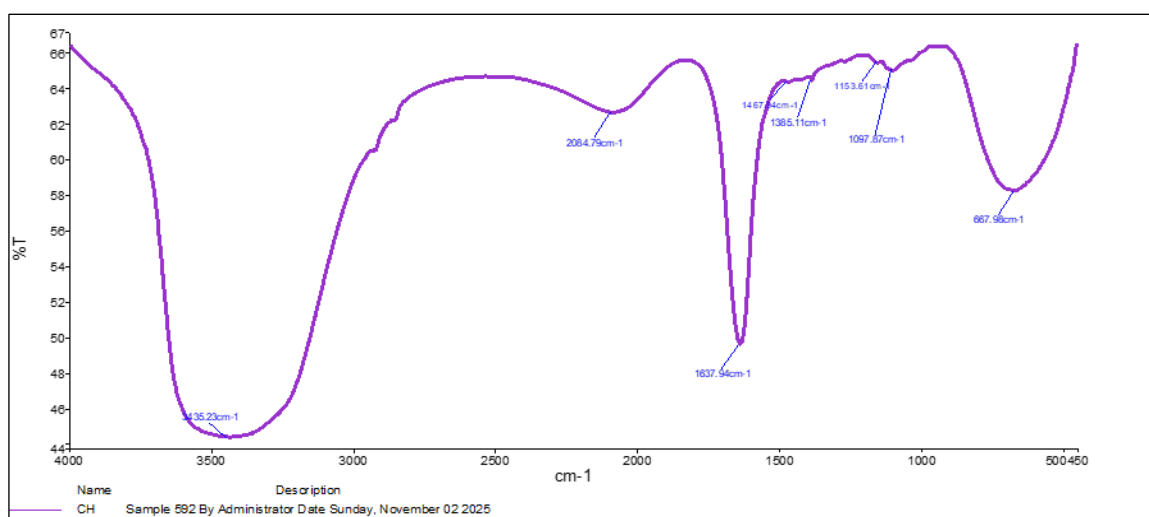


Figure 3 : Synthesis of Chitosan-copper nanoparticles: a) Chitosan, b) Copper(II) acetate, c) Chitosan-copper nanoparticles NPs

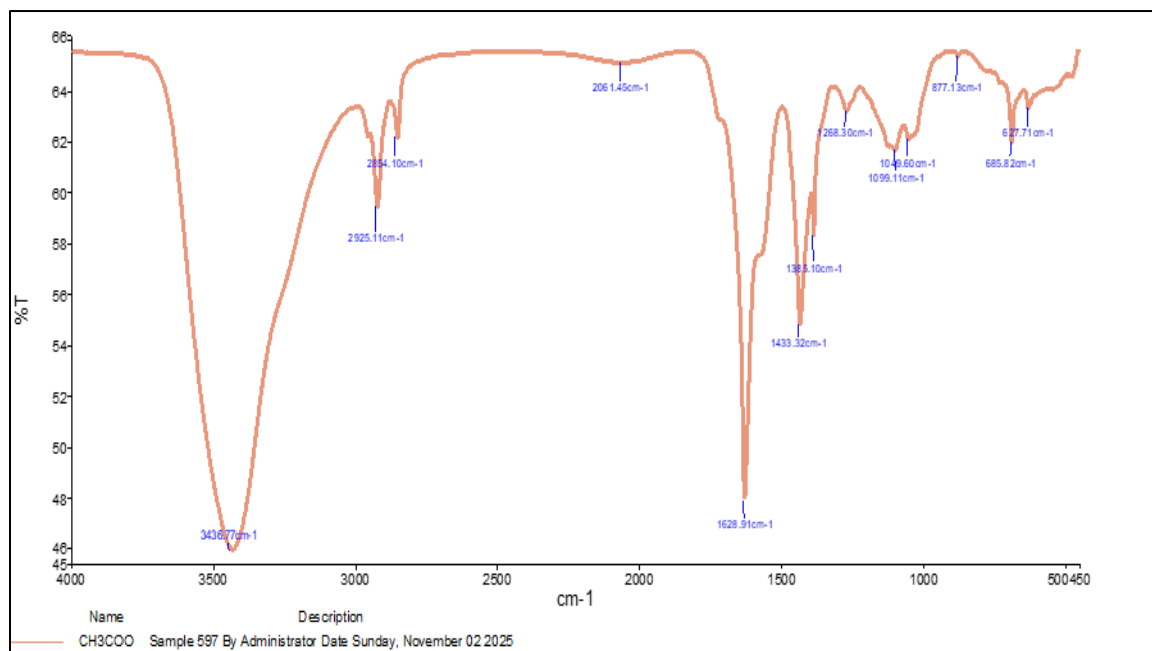
Characterization Chitosan-copper nanoparticles

FTIR analysis

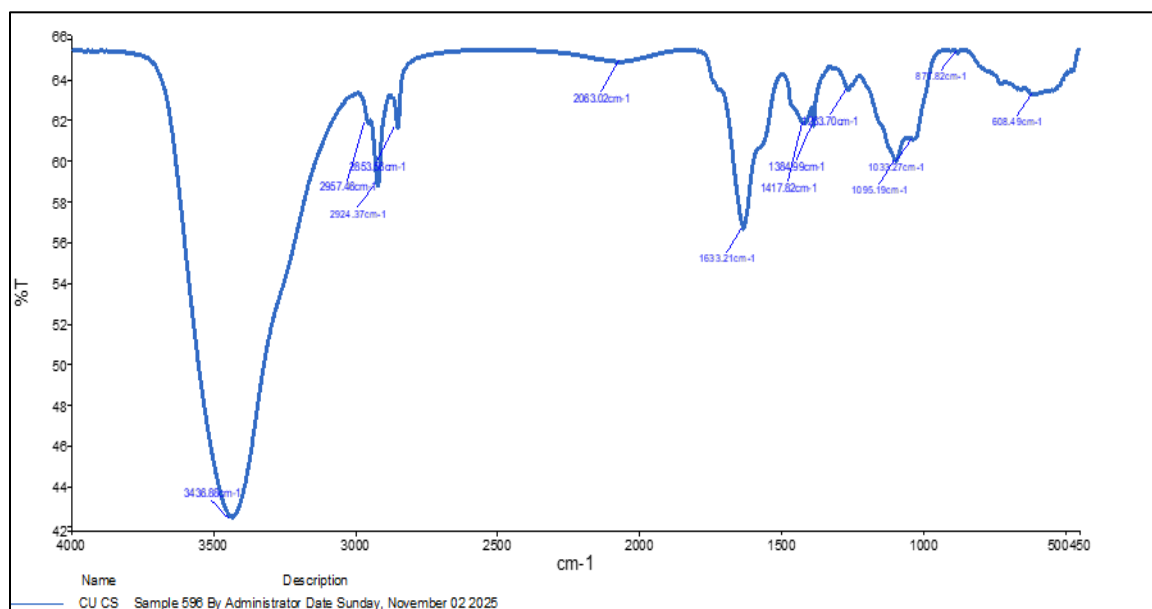
When free electrons in their outer electronic orbitals resonate with incoming electromagnetic radiation, they produce surface plasmon resonance (SPR), which gives copper nanoparticles in aqueous solution their distinctive brick-green color. The relationship between chitosan and copper nanoparticles was investigated using Fourier transform infrared (FTIR) spectroscopy (Fig. 4a). Pure chitosan's FTIR spectra revealed a wide absorption band about 3000 cm^{-1} in addition to discrete bands at 2878 cm^{-1} and 1658 cm^{-1} . This band can be attributed to overlapping N–H and O–H stretching vibrations. In contrast, copper sulfate showed a prominent absorption peak at 2927 cm^{-1} . The additional absorption bands seen at 3196 cm^{-1} and 3361 cm^{-1} , respectively, were recognized as S bending and Cu stretching vibrations (Fig. 4b). Major reductions in band intensities and changes in peak positions were seen after the creation of chitosan–copper interfaces (Fig. 4c), showing the effective production of CS–Cu nanoparticles and the strong interactions between chitosan functional groups and copper ions. The CS–CuNPs showed a rather narrow size range, with an average particle size of 87–89 nm and a polydispersity index of 24.74%, as determined by Zetasizer analysis. According to complementary structural analysis, the CS–CuNPs had an overall nanometric particle size approximately 131 nm and a hybrid core–shell architecture with a copper core wrapped in a chitosan shell.



(a)



(b)



(c)

Figure. 4. FTIR spectra of a) chitosan, b) Copper(II) acetate and c) CS-Cu NPs

UV-Visible (UV-VIS) Spectroscopy

One fundamental method for characterizing metal nanoparticles is UV–visible absorption spectroscopy. The different absorption features of the synthesized materials are shown in Fig. 5. UV-vis spectra. CuSO_4 , CS–CuNPs, and chitosan all showed distinctive absorbance maxima at 207 nm, 330 nm, and 214 nm, respectively, indicating that copper nanoparticles had been formed. The chitosan being used as a stabilizing and a reducing agent in the synthesis of the nanoparticles, by inhibiting the oxidation processes and agglomeration of the nanoparticles and thus limiting the formation of the particles. The synthesis and characterization of chitosan-copper nanoparticles (Ch-CuNPs) was also an important aspect of this investigation. The surface plasmon resonance effect, which was caused by the incident light interacting with the free electrons in the outermost orbitals of the copper nanoparticles, was found to be the cause of the characteristic brick-brown color of the aqueous dispersion.

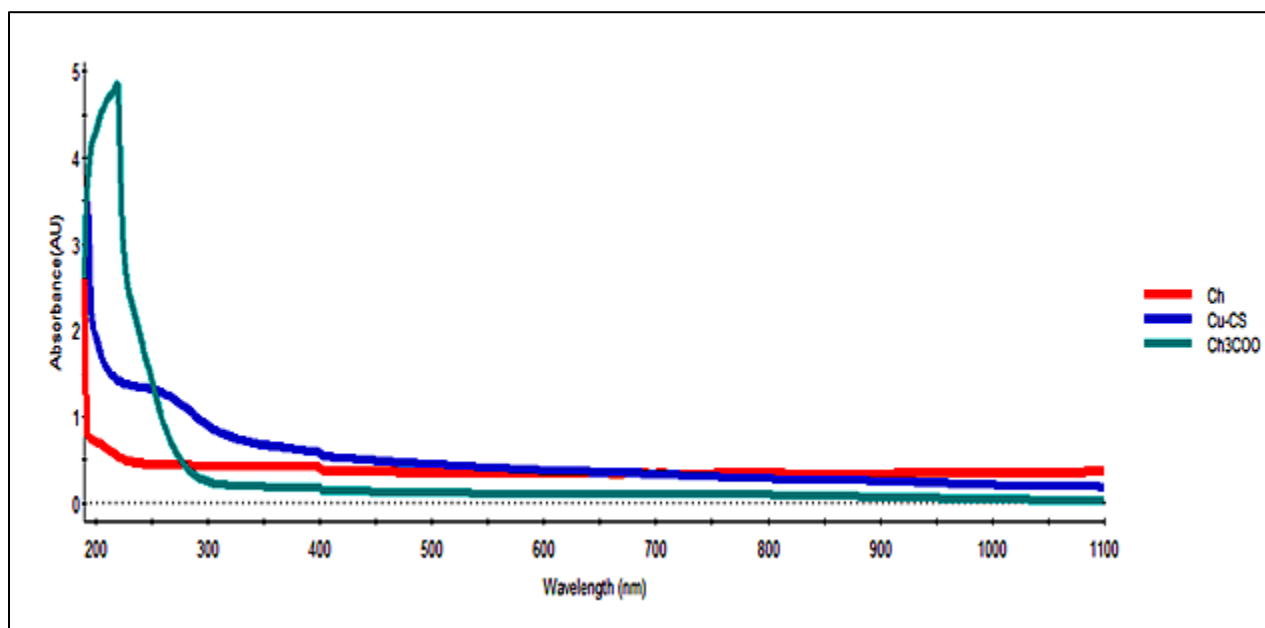


Figure 5 : UV–vis spectroscopy analysis a) Chitosan , b) +Copper(II) acetate and c) CS-Cu NPs

Zeta Potential Analysis

The CS-CuNPs' surface charge properties and colloidal stability were evaluated using zeta potential analysis. The nanoparticles' negative zeta potential was -35.36 ± 0.6 mV, as seen in Figure 6. This amount is presumably due to the presence of ionizable functional groups, including carboxyl and amino moieties adsorbed in the nanoparticle surface. Since the absolute magnitudes of the zeta potential are used as an important measure of colloidal stability, whereby increasing values of the zeta potential are associated with greater repelling forces between particles due to electrostatic interaction, the observed relatively high negative zeta potential indicates that the CS360 CuNPs have good colloidal stability in aqueous suspension. The hydrodynamic diameter and particle size distribution of the chitosan-copper nanoparticles were measured by dynamic light scattering (DLS) technique using a Zetasizer Nano Series instrument (Malvern Instruments). There were three measurements, and the average hydrodynamic particle size was obtained as 495.17 nm. DLS and the zeta potentials were measured at a controlled temperature of 25 °C.

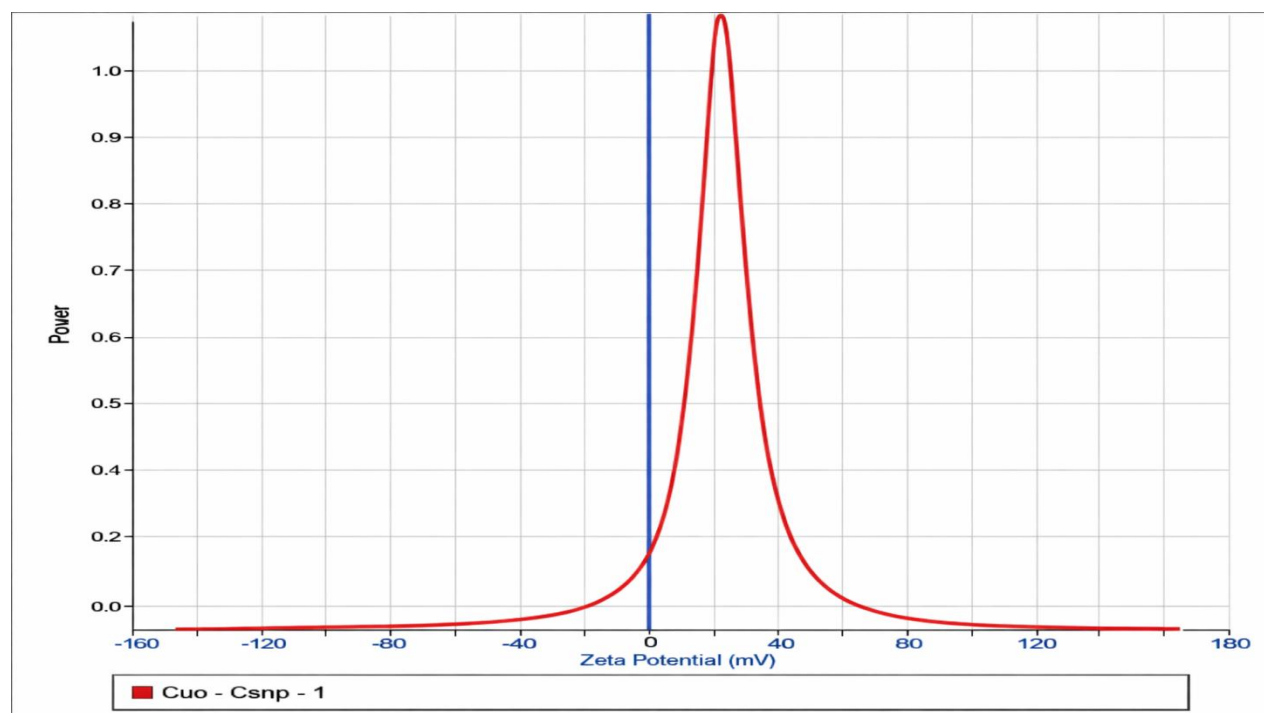


Figure. 6. b) Zeta potential, b) DLS potential of CS-CuNPs

Atomic force microscopy (AFM)

Surface morphology and particles dispersion of the produced CS–CuNPs were also analysed via contact scanning probe microscopy (CSPM) and atomic force microscopy (AFM). Microstructural features of the chitosan–copper nanocomposite were fully studied via AFM. Three-dimensional AFM images showed that the CS-CuNPs displayed irregular and triangular clustered morphologies with noticeable height fluctuations. According to the results of the particle size analysis, the average particle diameter was around 64 nm. The AFM data confirming the effective manufacturing and the nanoscale properties of the CS-CuNPs are shown in Figure 7.

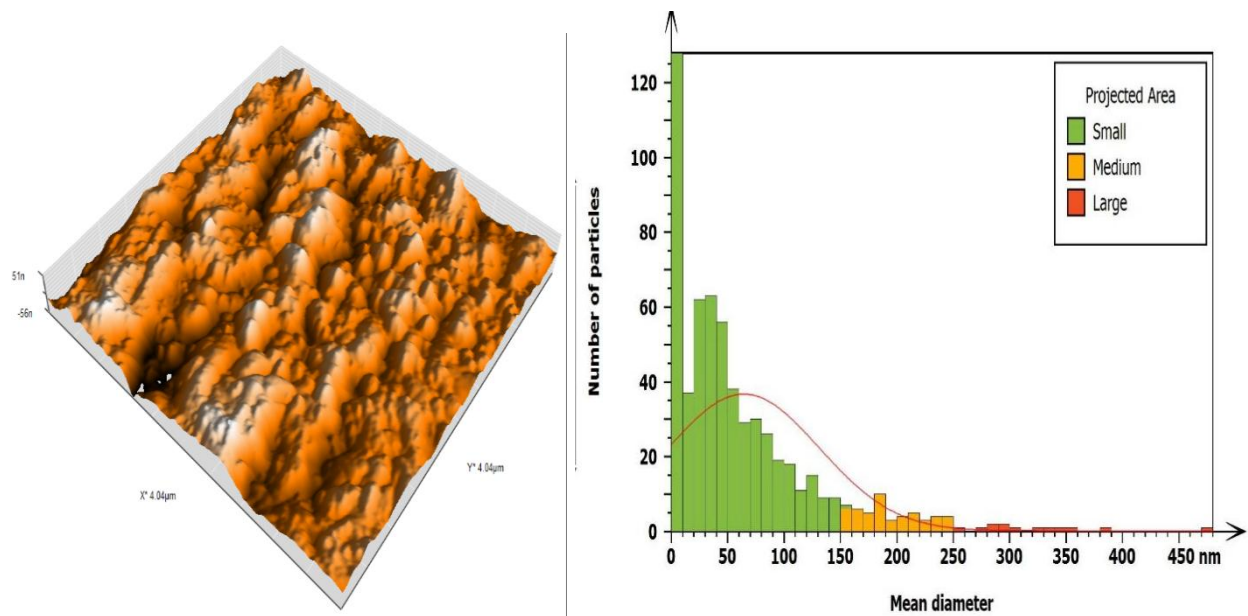


Figure 7 : The Chitosan–CuNPs Sunder AFM 2D and 3D .

Energy dispersive X-ray (EDX)

The elemental composition of CS-CuNP powder was determined from energy-dispersive X-ray analysis (EDX). As seen from the EDX spectrum (Fig. 8) and the quantitative data (Table 1), the main ingredients carbon C, nitrogen N, oxygen O and copper Cu

characteristic of the chitosan–copper nanocomposite system are detected. The copper signal indicates that the copper nanoparticles were successfully integrated into the chitosan structure, while the carbon, nitrogen, and oxygen signals originated from the chitosan polymer matrix. According to quantitative EDX analysis, oxygen was the most common element (79.97 wt%), followed by copper (12.25 wt%), carbon (5.28 wt%), and nitrogen (2.50 wt%). The hydroxyl and amine functional groups of chitosan as well as potential surface oxidation of copper nanoparticles could be the cause of the elevated oxygen content. The high purity of the produced CS–CuNPs is indicated by the existence of copper peaks without any additional impurity signals. Overall, as shown in Table 1 and Fig. 8, the EDX results verify the successful production and elemental composition of the chitosan–copper nanocomposite.

Table 1: Energy dispersive X-ray (EDX) of Chitosan–CuNPs

Elt	Lin	Int	Error	K	Kr	W%	A%	ZAF	For	Ox%	Pk/B	Clas	LCo	HCo	Cat#
e									mula		g	s	nf	nf	
C	Ka	19.3	11.3	0.03	0.02	5.28	7.56	0.56		0.00	6.48	A	4.90	5.66	0.00
			751	91	99			71							
N	Ka	11.3	11.3	0.02	0.01	2.50	3.07	0.70		0.00	11.3	A	2.26	2.73	0.00
			751	30	76			61			0				
O	Ka	416.9	11.3	0.85	0.65	79.9	86.0	0.82		0.00	175.	A	78.7	81.2	0.00
			751	66	65	7	5	08			66		4	1	
Cu	La	38.6	11.3	0.08	0.06	12.2	3.32	0.50		0.00	13.4	A	11.6	12.8	0.00
			751	14	24	5		90			3		3	7	
				1.00	0.76	100.	100.			0.00					0.00
				00	65	00	00								

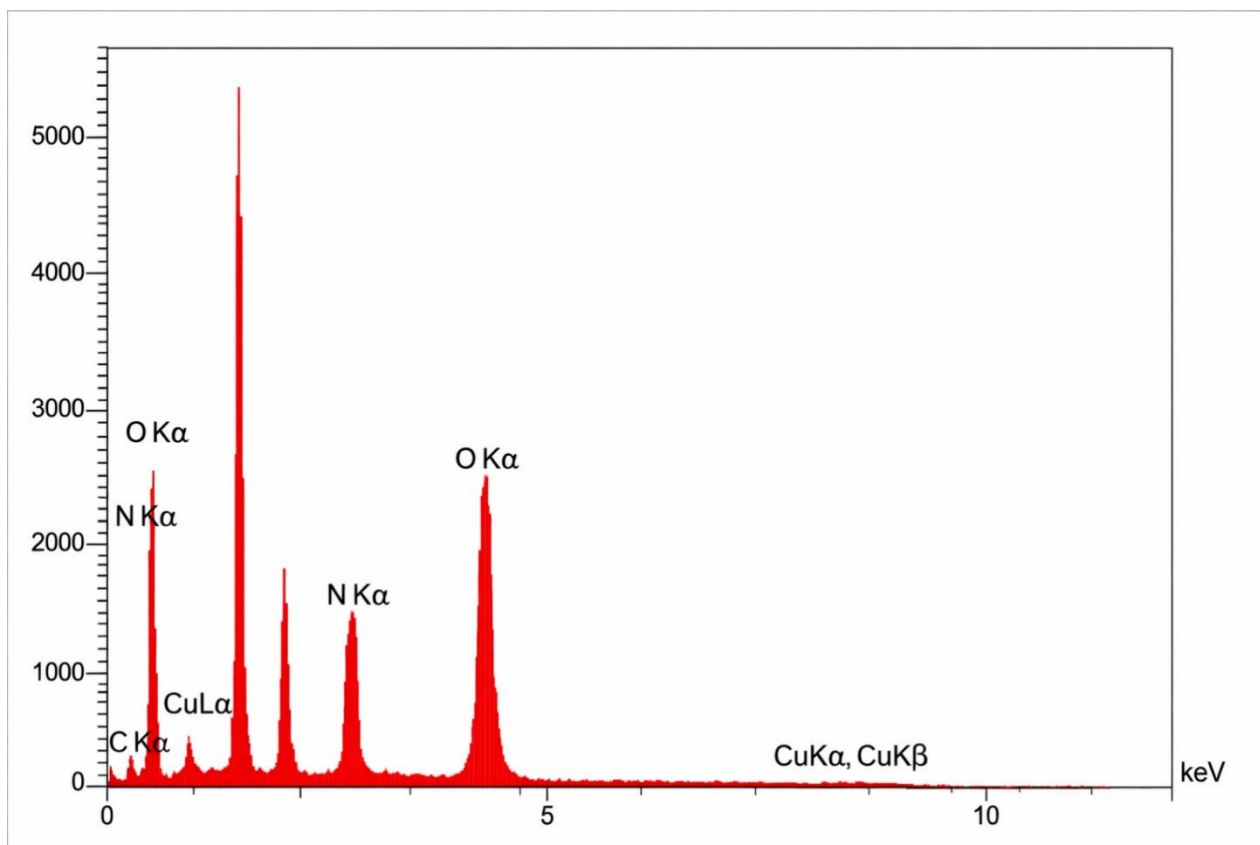


Figure 8: The images of EDX analysis show pure Chitosan–CuNPs

Transmission Electron Microscope and Scanning Electron Microscopy TEM

To determine the morphology and size distribution of the prepared CS-CuNPs, Transmission electron microscopy (TEM) was used as represented in Figure 9. The nanoparticles exhibited abnormal morphologies and a high tendency to aggregate which was probably due to electrostatic interactions and hydrogen bonding between the meropenem molecules and chitosan chains. The existence of the individual nanoparticles in the nanometer regime is supported by the presence of the 100nm scale bar, which further validates the nanoscale dimensions of the individual nanoparticles. The observed heterogeneity of the particle size and shape is considered to be symbolic of the nanocomposites that were produced in biologic or chemical pathways. All the TEM data confirm the successful synthesis of CS–CuNPs and support their nanoscale nature and aggregation tendency.

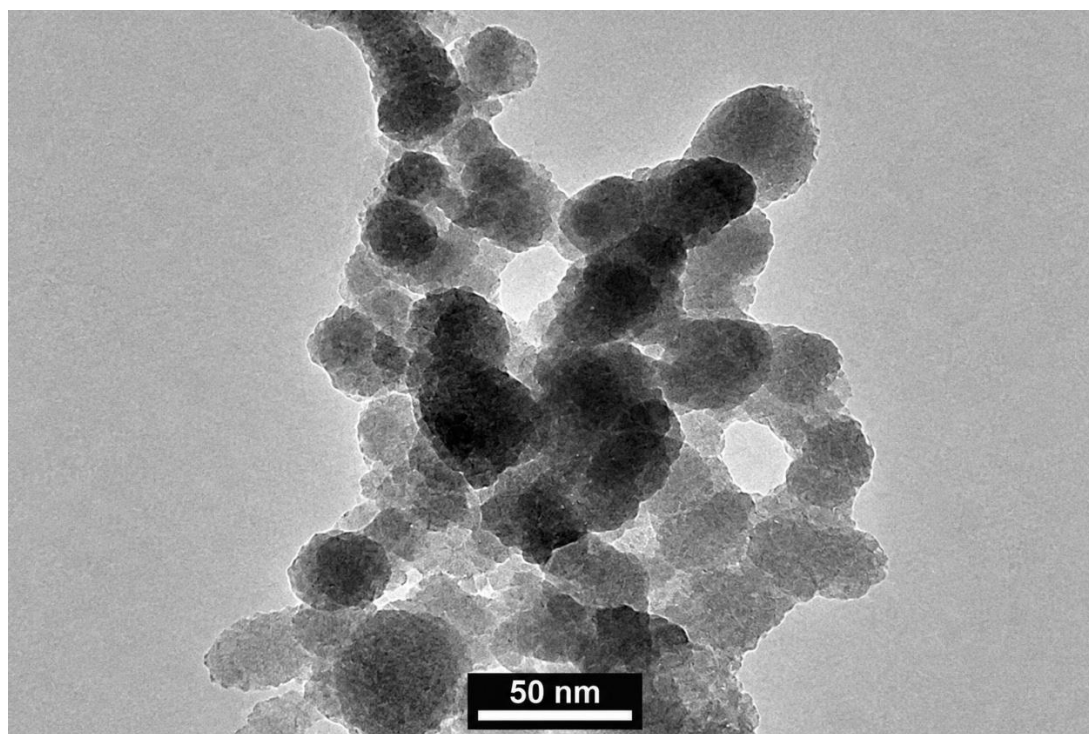


Figure 9: TEM of CS-CuNPs, Scanning Electron Microscopy (SEM)

The cytotoxic effect of CS-CuNPs on CAPAN-2 Pancreatic Cancer Cells lines

The nature of the tumour vascular structures that has a wide and high-porosity structure makes the accumulation of nanoparticles in the tumour microenvironment through vascular apertures easy, which promotes the nutrient and oxygen delivery. Cell viability reduction in the study under current investigation was a selective concentration-dependent effect that was induced by CS-CuNPs treatment. Cytotoxic potential of the CS-CuNPs was significant as the GI^{21} value was nearly 11 $\mu\text{g/mL}$. The cytotoxic properties of chitosan-coated copper nanoparticles (CS-CuNPs) to CAPAN-2 pancreatic cancer cells were tested through various concentrations (Figure 10). The results showed that cell viability was near 100% at low doses (0–1 $\mu\text{g/mL}$) and therefore the cytotoxicity in these doses was minimal. The viability of the cell declined with the increase in concentration of CS-CuNPs up to about 95% at 5 $\mu\text{g/mL}$ showed in Figure 10. The more significant decrease in viability was at 10 $\mu\text{g/mL}$ when viability dropped to about 87 percent marking when cytotoxic effects were starting to be significant. Cell viability was significantly decreased to about 60% at the highest tested concentration (50 $\mu\text{g/mL}$), indicating a potent dose-dependent inhibitory

effect of CS-CuNPs on CAPAN-2 cell survival. These results demonstrate the potential of CS-CuNPs as anticancer nanomaterials and show that they have considerable cytotoxic effects at increasing concentrations.

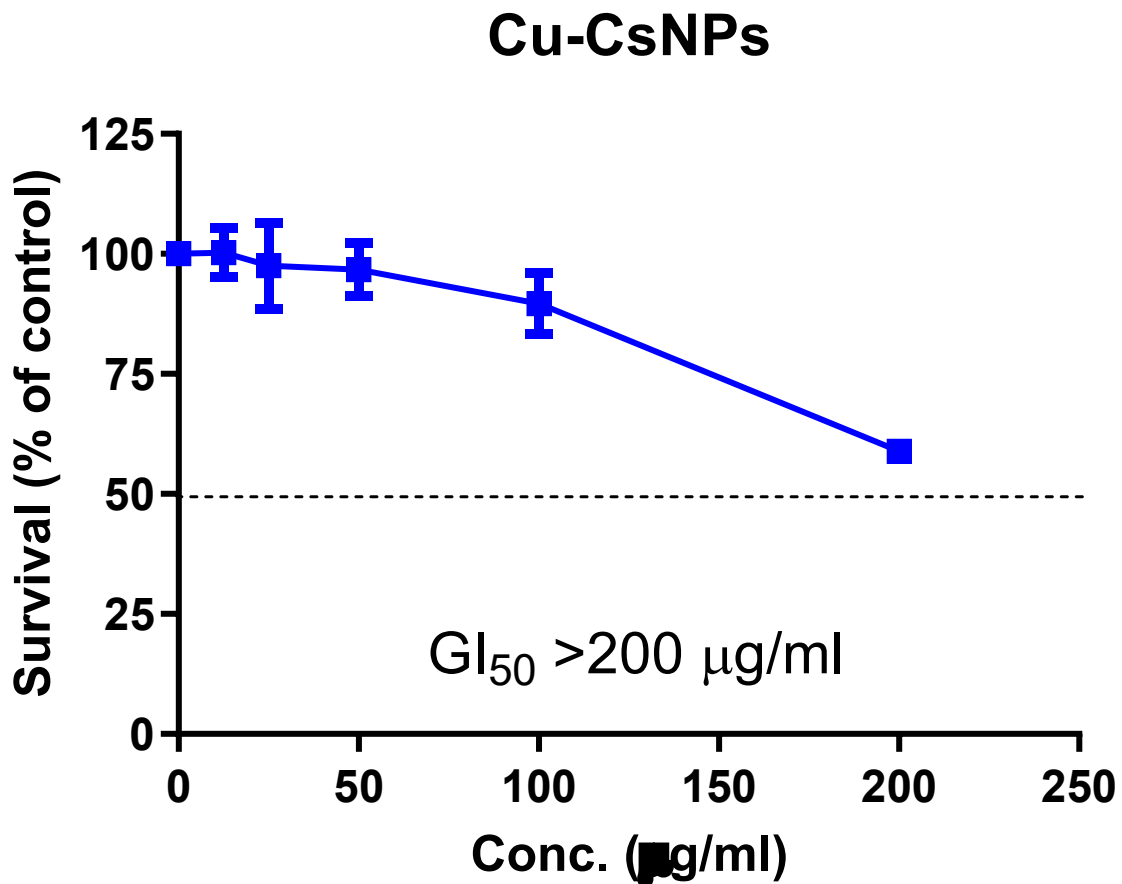


Figure.10. The cytotoxic effect of CS-CuNPs on CAPAN-2 Cells

Discussion

Chitosan, a biopolymer well known for its biocompatibility, biodegradability, eco-friendliness, and strong stabilizing properties, has been widely used for the preparation of copper-based nanoparticles (Chi-CuNPs). Nanoparticles prepared using a solution-casting approach have been characterized by Fourier-transform infrared spectroscopy (FTIR), ultraviolet–visible (UV–Vis) spectroscopy, scanning electron microscopy (SEM), energy-dispersive X-ray spectroscopy (EDX), and X-ray diffraction (XRD). These techniques confirmed the successful formation of nanomaterials with particle sizes ranging from 1 to 100 nm [17]. Copper oxide nanoparticles (CuO NPs) possess excellent optical, catalytic, electrical, antibacterial, and anticancer properties and have therefore attracted considerable scientific interest. Although several synthetic approaches have been developed, biologically mediated synthesis offers important advantages over conventional chemical methods, including improved safety, greater environmental compatibility, and operational simplicity. Various biological agents, including algae, plants, bacteria, fungi, and other biomolecules, have been employed for the synthesis of nanoparticles [18]. The biodistribution of nanoparticles and their interactions with biological membranes are strongly influenced by their physicochemical characteristics, including particle size, morphology, and surface properties. Copper-based nanoparticles can disrupt membrane integrity, increase membrane permeability, and ultimately induce cellular damage and death [19]. Owing to their high surface-area-to-volume ratio, metal and metal oxide nanoparticles (MONPs) can exhibit considerable biological activity even at relatively low concentrations, making them promising candidates for antibacterial applications. In particular, copper nanoparticles have attracted considerable attention because they are more economically accessible than noble-metal nanoparticles while exhibiting several functional properties comparable to those of silver and gold nanoparticles [20]. Nanoparticles have also been extensively investigated as drug-delivery platforms for the treatment of various diseases, including malignancies, cardiovascular disorders, and neurological diseases such as Alzheimer's disease. Their small size and tunable physicochemical properties allow certain nanocarriers to cross or facilitate drug transport across the blood–brain barrier, thereby improving the delivery of therapeutic agents to affected regions of the brain [21].

Pancreatic cancer remains a major health challenge because of its high mortality rate and poor prognosis. In recent years, cannabinoids derived from *Cannabis sativa* have attracted increasing attention as potential complementary or alternative agents in cancer research because of their reported antiproliferative and pro-apoptotic effects. Despite these promising findings, relatively few studies have investigated the potential therapeutic effects of cannabis-derived compounds in pancreatic cancer. Accordingly, one study evaluated the anticancer effects of cannabis in a pancreatic cancer xenograft mouse model [22]. The differential uptake of nanoparticles by cancer cells, potentially mediated by the mucin glycosylation mesh, may play an important role in regulating intracellular delivery and therapeutic efficacy. This phenomenon provides opportunities for the rational design and optimization of nanomedicine-based cancer therapies [23]. In addition, copper complexes have emerged as promising chemotypes for the treatment of human cancers. Several studies have demonstrated that copper-based complexes can exert cytotoxic effects through mechanisms that differ from those of conventional chemotherapeutic agents and may exhibit preferential activity toward tumor cells compared with normal cells. Furthermore, some copper-based compounds have demonstrated favorable safety profiles compared with conventional platinum-based chemotherapeutic agents [24]. Nanomaterials have emerged as versatile platforms for drug delivery across a broad range of diseases because of their biocompatibility, tunable physicochemical properties, surface-functionalization capabilities, and potential for scalable production. A wide variety of nanocarriers have been investigated for the delivery of anticancer therapeutics, including nucleic acid therapeutics (NATs) and small-molecule drugs. These systems can facilitate tumor accumulation and targeted delivery while protecting therapeutic cargo from degradation by chemical and biological factors during circulation. In the present study, copper oxide–chitosan nanoparticles (CuO–Ch NPs) were synthesized using a chemical approach, and their anticancer activity was evaluated against CAPAN-2 pancreatic cancer cells. The physicochemical properties of chitosan, including its stabilizing capacity, biocompatibility, and ability to facilitate cellular internalization, may contribute to the effective delivery of copper oxide nanostructures to malignant cells. The CuO–Ch NPs exhibited a concentration-dependent cytotoxic effect against CAPAN-2 cells, indicating their potential antiproliferative activity under the experimental conditions investigated.

Conclusion

The current paper shows that CuO chitosan nanoparticles are successfully produced and have high anticancer properties against CAPAN-2 pancreatic cancer cells. The latter is promoted by improved cellular uptake, oxidative stress caused by copper, and the pathways associated with apoptosis that are involved in the development of the desired cytotoxicity of the resulting nanocomposite, which varies with concentration. Chitosan is actively involved in enhancing the stability, biocompatibility as well as interaction of the nanoparticles with the malignant cells. All these findings suggest that CuO–chitosan nanoparticles are a promising nanotherapeutic approach to the pancreatic cancer, which should be investigated further in vivo to determine their safety profile and therapeutic effectiveness.

Declarations

Author Contributions: The author contributed to the study conception and design. Material preparation, data collection and analysis were performed by [Maryam Ibrahim, Nada H. A. L. Al-Mudallal]. The first draft of the manuscript was written by [Mais Emad Ahmed], who commented on previous versions of the manuscript and read and approved the final manuscript.”

Ethical Approval : This case report was conducted in according with the declaration of Helsinki. The collection and evaluation of data was confidential according to the health policy of the Iraqi Ministry of Health

Competing Interests : The authors declare no competing interests.

Funding Declaration :

The authors funded this study, and no other funding was obtained.

Research Involving Humans and Animals Statement:

All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the Helsinki Declaration and its later amendments or comparable ethical standards.

Informed Consent: None.

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