

## ORIGINAL RESEARCH ARTICLE

# Synthesis of carbon nanoparticles from glucose, evaluation of their phenol adsorption capacity, and study their potential as drug delivery systems for treating human pancreatic cancer cells

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## ABSTRACT

This study reports the creation of carbon nanomaterials via the wet chemical method using glucose as the precursor. The particles' sizes were characterized via XRD, and the particle's phenol adsorption capability characterized using UV-VIS and FTIR. The degree of loading efficiency (DLE) showed a very high amount for loading of phenol onto carbon nanomaterials. The particle size and shape were characterized after phenol loading using Transmission Electron Microscopy (TEM), showing the produced particles had multiple small particles in the nanometer-sized range. The cytotoxicity of the phenol-loaded nanosized carbon particles was tested against human pancreatic cancer cells with various dose amounts using the MTT assay. This study indicates that the phenol-loaded nanosized carbon particles show potential to kill cancer cells.

**Keywords:** carbon nanoparticles; phenol; drug delivery; MTT assay; pancreatic cancer; cytotoxicity

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## 1. Introduction

The pancreas, situated behind the stomach, is essential for digestion and regulating glucose levels by means of insulin secretion. Pancreatic cancer is among the most deadly cancers in the world today, with risk factors being smokings, chronic pancreatitis, genetic predisposition, obesity, and advanced age, with the majority of cases diagnosed after the age of 65<sup>[1,2]</sup>. Further epidemiology studies show that there is a higher occurrence of men than there are of females<sup>[3-5]</sup>. The most common treatment options for pancreatic cancer involve surgical resection, chemotherapy, and radiotherapy, although in general these types of treatments have several downsides such as poor treatment targeting and a high rate of side effects<sup>[6-8]</sup>.

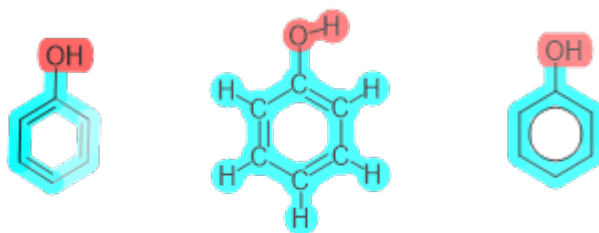
Nanodelivery systems provide a new opportunity to improve the drug efficaciousness and to decrease the negative aspects associated with drug treatments, as well as increasing the selectivity of the cancer cells that receive treatment as a result of their unique characteristics, including nanoscale sizes, large surface areas, and capacity for controlled release of chemicals<sup>[12-14]</sup>. One example of

promising nanomaterials are carbon nanoparticles, which possess unique physical and chemical characteristics that will provide improved drug loading and delivery properties in oncology applications as a result of the physical and chemical characteristics of carbon nanoparticles<sup>[15-17]</sup>.

In this study, carbon nanoparticles were synthesized via a wet chemical method, which involves chemical reactions in liquid media, using glucose as a carbon source and nickel chloride as a catalyst under acidic conditions<sup>[18-21]</sup>. These nanoparticles exhibit a large surface area conducive to efficient adsorption<sup>[22]</sup>. Phenol, a hydroxyl-containing organic molecule with antioxidant properties, was selected for loading onto the carbon nanoparticle surface (**Figure 1**). Phenol's capacity to mitigate oxidative stress caused by free radicals has been linked to protective effects against diseases such as cardiovascular disorders, diabetes, and cancer<sup>[23,24]</sup>. The adsorption process, involving the accumulation of phenol molecules onto the nanoparticle surface, was utilized to achieve drug loading<sup>[25,26]</sup>.

Cytotoxicity of the phenol-loaded nanoparticles was assessed using the MTT assay, a widely employed, sensitive, and cost-effective method that evaluates cell viability through mitochondrial metabolic activity<sup>[27-35]</sup>. This study aims to deepen our understanding of nanoparticle-based drug delivery mechanisms in pancreatic cancer treatment and to provide foundational data supporting the development of novel, safer therapeutic strategies.

Recent advances in nanomaterial applications and phenol-based therapeutics highlight the potential of such nanoformulations in oncology, emphasizing the importance of controlled release, targeting efficiency, and biocompatibility<sup>[36-41]</sup>.



**Figure 1.** shows the chemical structure of phenol.

## 2. Experimental section

### 2.1. Materials and methods

#### Synthesis of Carbon NPs

To synthesize nanocarbon molecules, 50 grams of glucose were dissolved in 100 millilitres of distilled water in a 500-milliliter flask. After that, one gram of nickel chloride was added as a catalyst, and the flask was placed on a magnetic stirrer. After that, a 70 ml acidic mixture was created using 35 ml of nitric acid and 35 ml of sulfuric acid. As shown in **Figure 2A**, the acid was gradually added to the sugar solution using a pipette while the mixture was continuously stirred in a customized ventilation chamber. After that, we heated the substance to 120 degrees Celsius, and yellow gas emission was observed, indicating the release of nitrogen oxides. After four hours, we saw a color change to black, which suggests that sugar has broken down and carbon has formed, as shown in **Figure 2B**. As seen in **Figure 3**.

To eliminate impurities and acid residues, we then performed the filtering technique shown in **Figure 2C** and washed the precipitate ten times with distilled water. Subsequently, we desiccated it at 100 degrees Celsius in a convection oven.

To ascertain the dimensions and purity of the synthesized particles, we gathered the resultant powder as seen in **Figure 2D** and used the XRD method for analysis.

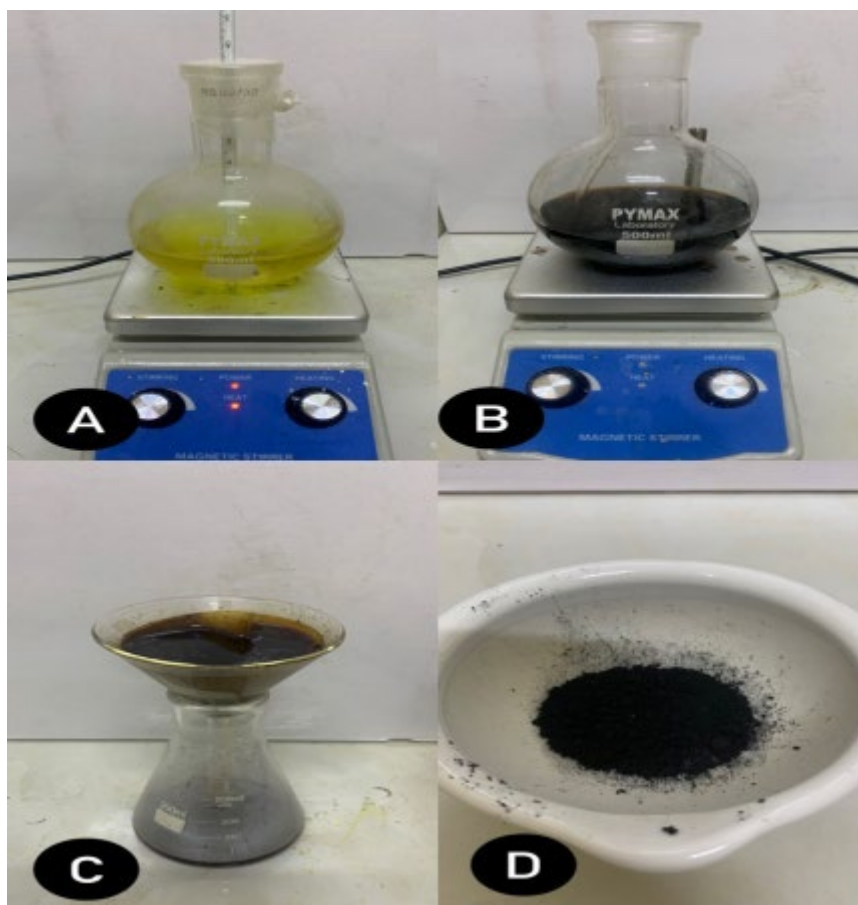


Figure 2. shows the steps of preparing nano carbon.

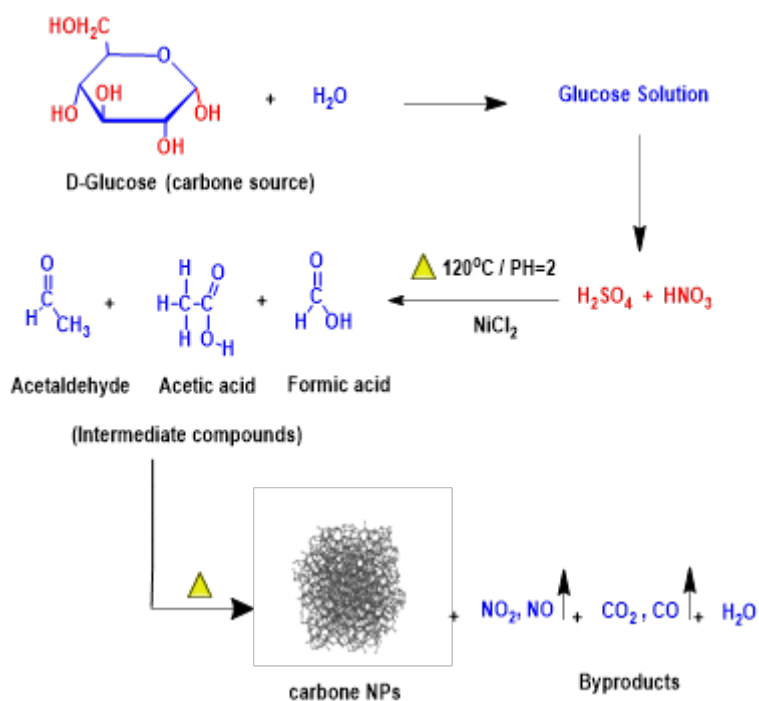


Figure 3. shows the schematic of the process of preparing nanocarbon from glucose.

### Drug loading

The drug loading process involved the adsorption of prepared carbon nanoparticles onto the phenol compound. This was achieved by mixing 1.3 grams of phenol in 60 ml of deionized distilled water within a

150 ml beaker, followed by continuous stirring for 5 minutes using a magnetic stirrer. Subsequently, 5 grams of carbon nanoparticles were incrementally introduced while continuously swirling for 15 minutes. Subsequently, the mixture was allowed to rest in a dark bottle for 24 hours to attain equilibrium. Refer to **Figure 4**.



**Figure 4.** Shows the components of the mixture: water, nanocarbon, and phenol.

Once the stipulated time had passed, we filtered the precipitate using filter paper, dried it in an air oven set to 80 °C, and prepared it for testing and diagnosis. The filtrate was collected in order to carry out the loading efficiency test in the procedures that follow.

### 3. Characterization

The following approach was used to confirm that phenol was adsorbed onto the nanocarbon surface after the precipitates had been collected and dried with care:

- 1- Using a UV-VIS spectrophotometer to read the absorbance.
- 2- Performing an FTIR study to verify the existence of phenol.

After that, we used an atomic force microscope (TEM) to analyze the form and size of the nano carbon particles after they had been adsorbed.

#### **Drug Loading Efficiency (DLE)**

Five different concentrations of phenol solution were prepared: 1%, 3%, 6%, 9%, and 12%. In order to carry out the calibration technique and create the calibration curve, the absorbance of each solution was measured at a wavelength of 285 nm, which is the maximum absorption wavelength of the phenol component. In order to evaluate how well phenol is loaded onto the nanocarbon surface, the residual concentration must be calculated by measuring the absorbance of the solution (the solution that remains after adsorption).

#### **Cell culture and cytotoxicity**

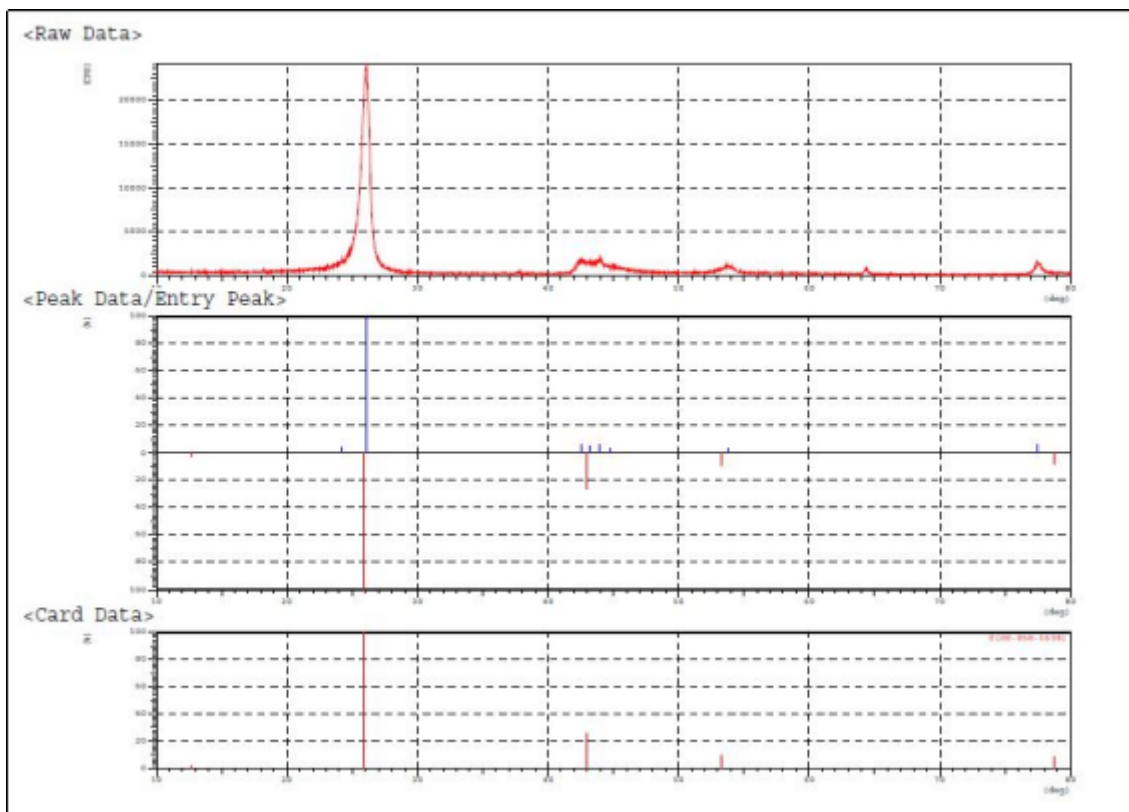
The cytotoxic potential of nanoparticles was assessed using the MTT assay. Pancreatic cancer cells were chosen to evaluate cell viability using the MTT assay following treatment with phenol-loaded carbon nanoparticles. The cells were inoculated in a 96-well plate at a density of  $1 \times 10^4$  cells per well and incubated at 37 °C with 5% CO<sub>2</sub> for 24 hours to promote optimal stability and adherence. The culture medium was discarded, and the cells were subsequently cultured with fresh medium containing different concentrations of phenol-loaded carbon nanoparticles (10, 20, 30, 40, 50, and 60 µg/ml) for a duration of 48 hours. After 48 hours of cell culture, 10 µl of MTT (5 mg/ml) was added to each well. The incubation lasted

for 4 hours, followed by the addition of 100  $\mu\text{L}$  of DMSO to each well to solubilize the MTT crystals. The responses were measured with an ELISA reader at a wavelength of 570 nm.

## 4. Results and discussion

### XRD analysis

The produced carbon nanoparticles were subjected to XRD examination in order to ascertain the atoms' particle size. The outcomes are displayed in **Figure 5**.



**Figure 5.** shows the analysis result of the prepared carbon npt using XDR technology.

The crystallite size of the synthesized nanoparticles was estimated based on the X-ray diffraction (XRD) pattern using the Scherrer equation, which is widely applied for determining average crystallite dimensions in the nanometer scale. The equation is expressed as:

$$\tau = \frac{K\lambda}{\beta \cos \theta}$$

where:

$\tau$ : average crystallite size (nm)

K: shape factor, typically = 0.9

$\lambda$ : X-ray wavelength (typically 0.15406 nm for Cu-K $\alpha$ )

$\beta$ : full width at half maximum (FWHM) of the diffraction peak = 0.00285 radians

$\theta$ : Bragg angle (degrees) = 14°

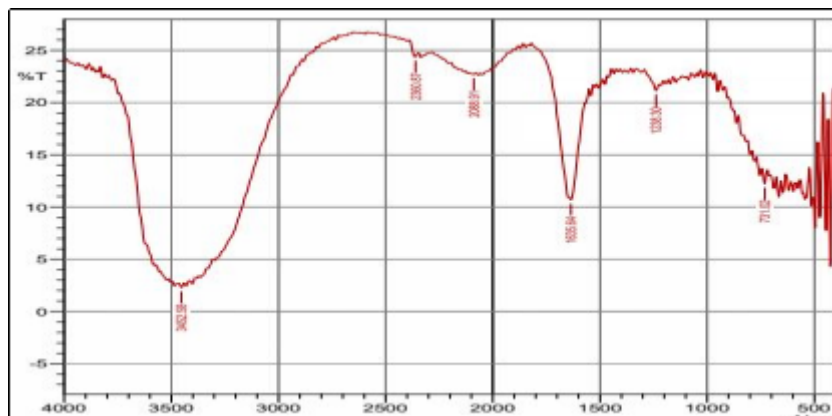
Using the diffraction data, we calculated the crystal size to be approximately 50.12 nm, confirming that the particles are within the nanoscale range. It should be noted that this method is valid for estimating sizes less than 100–200 nm, due to the effects of peak broadening in X-ray diffraction.

## UV-VIS Spectrometer

The presence of phenol on the surface of the carbon nanoparticles was confirmed using UV–Vis spectroscopy. A characteristic absorbance peak was observed at 285 nm, which corresponds to the maximum absorbance wavelength ( $\lambda_{\text{max}}$ ) of phenol. The sample was prepared by dispersing 0.1 g of phenol-loaded carbon nanoparticles in 5 mL of distilled deionized water, and the measured absorbance at 285 nm was 1.536. This result confirms the successful loading of phenol onto the nanoparticle surface.

## FTIR Spectrum

FTIR analysis was carried out to identify the functional groups present on the surface of the nanocarbon particles after phenol adsorption, as shown in **Figure 6**.



**Figure 6.** shows the FTIR spectrum of the carbon nanoparticles after adsorption and loading of phenol molecules onto their surface.

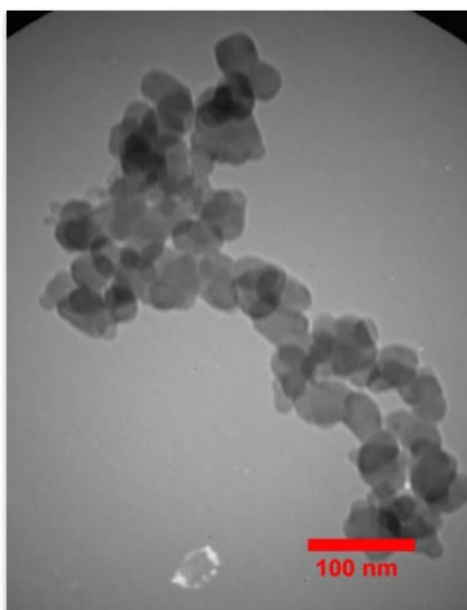
A broad absorption band at  $3452.58\text{ cm}^{-1}$  corresponds to the O–H stretching of hydroxyl groups attached to the aromatic ring. The peak at  $1645.64\text{ cm}^{-1}$  is assigned to C=C stretching vibrations in the aromatic structure. A band at  $1238.30\text{ cm}^{-1}$  is attributed to C–O stretching, while the peak at  $731.02\text{ cm}^{-1}$  corresponds to O–H bending vibrations.

These characteristic peaks are consistent with the structure of phenol and indicate successful adsorption onto the carbon nanoparticle surface. Although only qualitative FTIR analysis was conducted, the appearance and positions of the functional group bands strongly support the presence of phenol.

## TEM Analysis

In order to analyze the nanostructure and determine the size of the carbon nanoparticles, as well as to investigate the geometric form after phenol has been successfully loaded onto the surface and adsorbed. We performed an atomic force microscope study, which provided us with a clear image by demonstrating that all of the particles were smaller than 100 nanometers and inside the nano scale.

It also demonstrated the geometric form of the particles, which were minuscule and had a variety of shapes that were not defined. As seen in **Figure 7**.



**Figure 7.** shows the shape and size of the carbon nanoparticles after adsorption as analyzed by TEM.

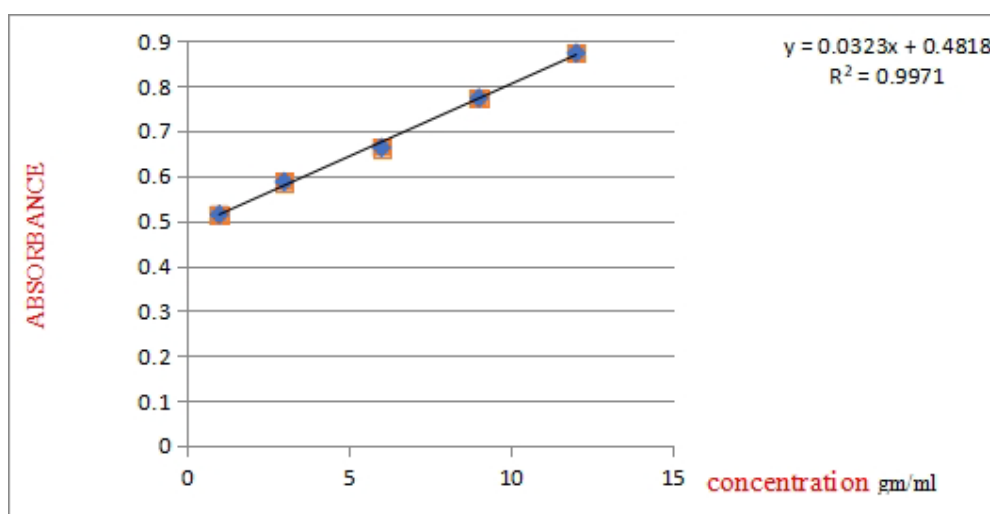
### Drug Loading Efficiency (DLE)

The absorbance was measured for all prepared phenol concentrations and the results were as shown in **Table 1**.

**Table 1.** shows the concentration and absorbance values.

Concentration	Absorption
1	0.514
3	0.587
6	0.663
9	0.773
12	0.873
1	0.514

Based on the concentration versus absorbance data in **Table 1**, the calibration curve and the equation of the straight line were plotted as shown in **Figure 8**.



**Figure 8.** shows the standard curve plot, straight line equation calculation, and R value for the prepared phenol concentrations.

Following the measurement of the absorbance of the residual filtrate from the adsorption process at a wavelength of 285 nm, the concentration value was obtained by referencing the standard calibration curve, yielding a concentration of 0.002 g/mL. To calculate the phenol loading efficiency on the surface of the carbon material, the following equation was applied:

$$\text{Loading Efficiency (\%)} = (C_0 - C_e / C_0) \times 100$$

$C_0$ =Phenol concentration before adsorption and it was equal to 0.026 g/ml

$C_e$ =Phenol concentration after adsorption and it was equal to 0.002 g/ml

The drug loading efficiency (DLE) was extracted and it was equal to 92%

### **Cytotoxicity of phenol-loaded carbon nanoparticles on pancreatic cancer.**

Following the successful cultivation of pancreatic cancer cells and the MTT assay to evaluate the cytotoxicity of the prepared concentrations, the cell viability percentage and cytotoxicity percentage were determined using the following formulas

$$\% \text{ Cell viability} = \text{Sample absorbance} / \text{absorbance} * 100$$

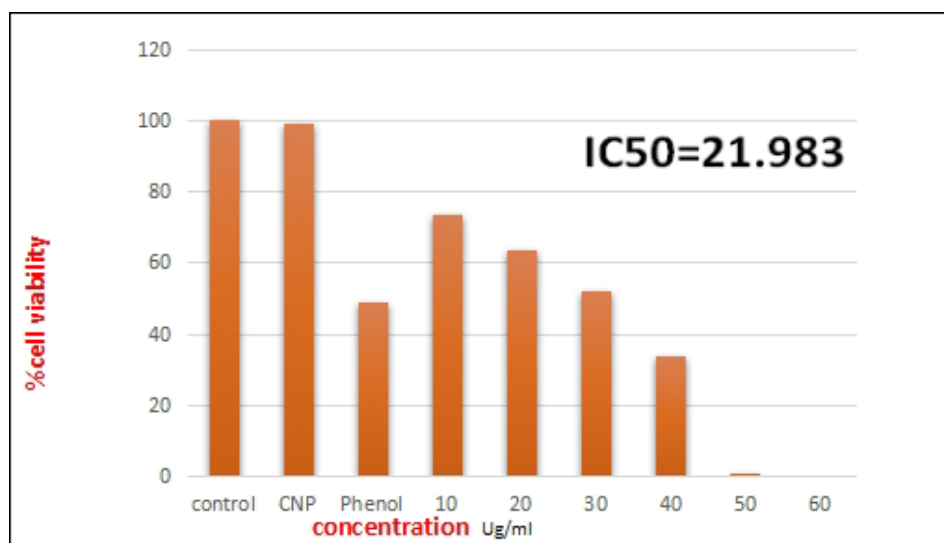
$$\% \text{ Cell viability for control sample} = \text{Control absorbance} / \text{Control absorbance} * 100$$

$$\% \text{ Cytotoxicity} = 100 - \text{Cell viability}$$

Control experiments were conducted to isolate the contribution of each component to the observed cytotoxicity. Pancreatic cancer cells treated with unloaded carbon nanoparticles exhibited minimal cytotoxicity, with a cell viability of approximately 98.99%, indicating negligible toxicity. In contrast, treatment with free phenol at a concentration of 50  $\mu\text{g/mL}$  reduced cell viability to 48.86%, suggesting moderate cytotoxic potential.

The phenol-loaded carbon nanoparticles exhibited the highest cytotoxic effect at concentrations of 50–60  $\mu\text{g/mL}$ , where cell viability dropped to 0%. The lowest observed toxicity was at 10  $\mu\text{g/mL}$ . The average inhibitory concentration was calculated to be 21.983  $\mu\text{g/mL}$ , at which approximately 50% of the cancer cells were no longer viable.

These results suggest that the combined system has enhanced cytotoxic activity compared to either component alone, indicating a potential synergistic effect between phenol and the nanocarrier. as seen in **Figure 9.**



**Figure 9.** presents the viability of pancreatic cancer cells treated with phenol-loaded carbon nanoparticles at various concentrations, compared to cells treated with free phenol and unloaded nanoparticles.

The enhanced cytotoxicity of the phenol-loaded nanoparticles may be attributed to biochemical mechanisms involving oxidative stress and intracellular damage. Phenol is known to generate reactive oxygen species (ROS), which can compromise mitochondrial integrity, cause DNA fragmentation, and trigger apoptosis pathways. These cellular disruptions collectively reduce viability and contribute to the observed effect. However, further mechanistic investigations are necessary to confirm the precise cellular pathways affected in pancreatic cancer cells.

## 5. Conclusion

This study demonstrates successful preparation (or synthesis) of carbon based material (i.e., carbon nanoparticles) using environmentally friendly and renewable carbon sources, specifically glucose, via an acid wet chemistry method. The resultant carbon was shown to possess a very high surface area, leading to a very successful (i.e., loading of phenol) absorption of phenol onto the surface of the carbon nanoparticles. The carbon nanoparticles thus produced exhibited very high levels of toxic effects towards pancreatic cancer cells when compared to untreated controls; furthermore, there was found to be a positive correlation between increasing size of the carbon nanoparticles and an increase in their toxic effect. This work demonstrated that the prepared carbon nanoparticles will have potential for future use as a drug to treat cancer through larger studies and clinical trials.

## Ethics Information

None.

## Fundings

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## Author Contribution Statement

Ghufran Hammood: Conceptualization, methodology, investigation, writing- original draft; Mustafa Allawi: Visualization, project administration, writing- reviewing and editing; Mina Faris: Supervision, visualization, project administration, writing- reviewing and editing; Shahad Hammad: Investigation and resource.

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## Conflict of interest

The authors declare no conflict of interest.

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