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Synthesis and charaterization of TiO_2 nanoparticles and its cytotoxcity in CHO cell line

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ABSTRACT

TiO_2 nanoparticles as photocatalyst were synthesized by sol-gel route involving, and it is efficiently characterized by techniques, such as XRD, TEM, FTIR and DLS. The cytotoxicity of Titanium dioxide was evaluated on CHO cell line. The cytotoxicity of TiO_2 is dependent on the dose and it shows gradual increase in the percentage of dead cell after 24 h of TiO_2 exposure. The cytotoxicity due to TiO_2 increases with the increase in the concentration of TiO_2 nanoparticles from 1 to 75 µg/ml.

Keywords: TiO_2, Nanoparticles, Cytotoxicity, Sol-Gel

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INTRODUCTION

Titanium oxide is considered as the one of the most important semiconductor materials in last few decades due to its wide range of applications in various fields such as cosmetic and pharmaceutical industry [1]. Researchers were focussed on synthesizing TiO_2 nanoparticles because of their unique optical and electrical properties[2]. Number of photocatalytic materials like zinc oxide (ZnO), Ferrous Oxide (Fe_2O_3) and Zinc Tungstate ($ZnWO_4$) were used for the waste water treatment, but titanium dioxide has gained much more focus in removal of pollutant from waste water, thanks to its free electrons and the lesser number generated holes in valence band as compared to electron in valence band[3]. Additionally, TiO_2 is most widely used in the field of electrochemical sensors because of it excellent chemical stable nature, non-toxicity and enhanced transfer rate of electron [2].

Titanium dioxide nanomaterials have also attracted significant interest in the biomedical field because of their non-toxicity, excellent optical and catalytic properties, bio conductivity, and its anticancer applications. Due to their environmentally friendly nature, TiO_2 exhibits excellent chemical and biological stability, which make it suitable for various applications, such as photocatalyst, food packaging industry, rubber industry, solar cell industry, biosensor industry, optoelectronics devices [3]. Compared to other semiconductor nanomaterials, titanium dioxide stands out due to its large bandgap (3.30 eV) and high exciton binding energy, which is almost double than the thermal energy at room temperature (25 meV).

This makes it an attractive material for research in the materials science field [4].

TiO_2 NPs are also widely used for their antimicrobial properties, appearing in products such as antifungal shampoos, antibiotic ointments, antiseptic creams, surgical tapes, and calamine lotions [5]. Furthermore, zinc oxide nanoparticles are incorporated into sunscreen products for their broad-spectrum UV protection [6]. Recently researchers has been working on the applications of titanium dioxide as an antioxidant, biosensors, environmental remediation, & electrochemical sensing due to their low cost, excellent surface area, and stability. In nature titanium dioxide has been found in the two crystalline structure forms, i.e., Anatase, and rutile from [7]. The hexagonal rutile form has been found to be a most stable form of titanium dioxide in various conditions [8]. This stability makes titanium dioxide a preferred semiconductor material for potential applications in the fields such as the rubber industry, lifeguard equipment, and tyre manufacturing [9]. In this work, we synthesize the titanium dioxide nanoparticles by a sol-gel method, and check its cytotoxicity on CHO cell line.

MATERIALS AND METHODS

Synthesis of TiO_2 by sol-gel method

The sol was prepared using titanium isopropoxide, from (Sigma–Aldrich), 2-Propanol from Ranbaxy, Citric acid (Rankem lab). In this synthesis we have taken 10 mL of Ti $(OCH\ (CH_3)_2)_4$ and dissolved in 40 ml of 2-Propanol in a beaker in a moisture free environment. After this, the whole mixture was added dropwise into another mixture containing (10 mL H_2O+10 mL 2-Propanol). The pH of the above mixture was adjusted to 3 by the addition of HCl or ammonium hydroxide. A yellow gel was obtained after 1 hr stirring. The above formed gel was dried at temperature of 105 ^{0}C until a yellow black color crystal are formed. Finally, the above formed product was calcined at about 500 ^{0}C for 7 hr in a furnace.

RESULTS AND DISCUSSIONS

The XRD pattern of titanium dioxide nanoparticles shown in Figure 2. The average grain size was calculated by the highest intense peak (110) using Debye Scherer formula. The value of grain size was found to be 18.8 nm. The highest intensity peak (110) at angle 26.82° corresponds to rutile phase. This measured XRD pattern has been compared with the standard data base JCPDS-card no. 841284.

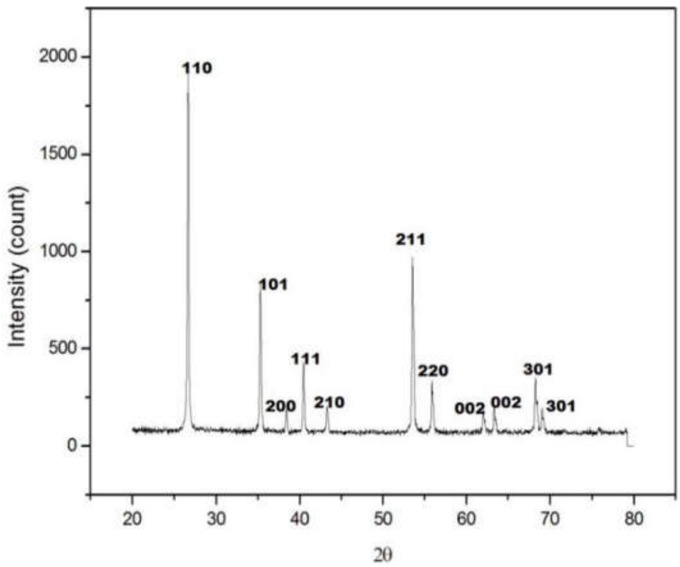

Figure 2: XRD TiO_2 nanoparticles

TEM analysis of TiO_2

The magnified images of TiO_2 nanoparticles are shown in figure 3. TEM images displayed the agglomerated nature of these particles. As shown, the average particle sizes of TiO_2 NPs were smaller or equal to 20 nm.

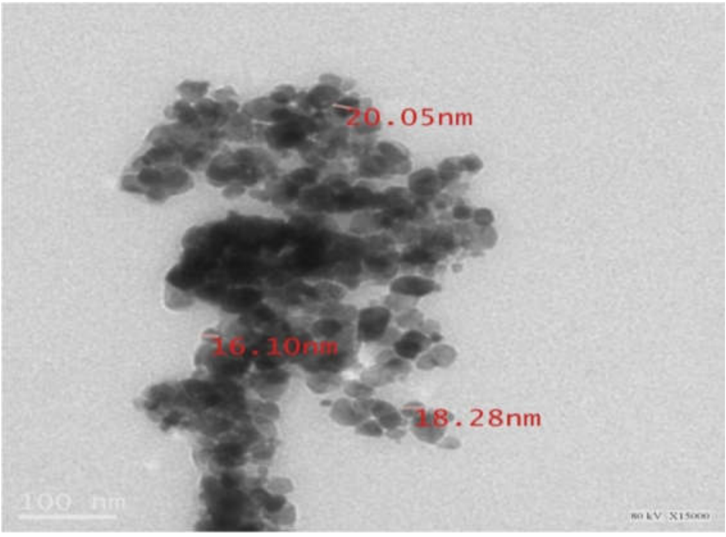

Figure 3: TEM image of TiO_2 nanoparticles

FTIR Analysis: FTIR spectrum of titanium dioxide is displayed in figure 4. The broad intense band below 1000 cm^{-1} is due to Ti-O-Ti vibrations. The metal Oxide peak was found in the range of 400-600cm^{-1} as shown in the Figures. For TiO_2 it appears at 457.42 cm^{-1} as shown in figures 4. The asymmetrical stretch of CO_2 gives a strong band at 2350 cm^{-1} as it is observed in TiO_2 and plots. This band occurs due the presence of CO_2 in the atmosphere.

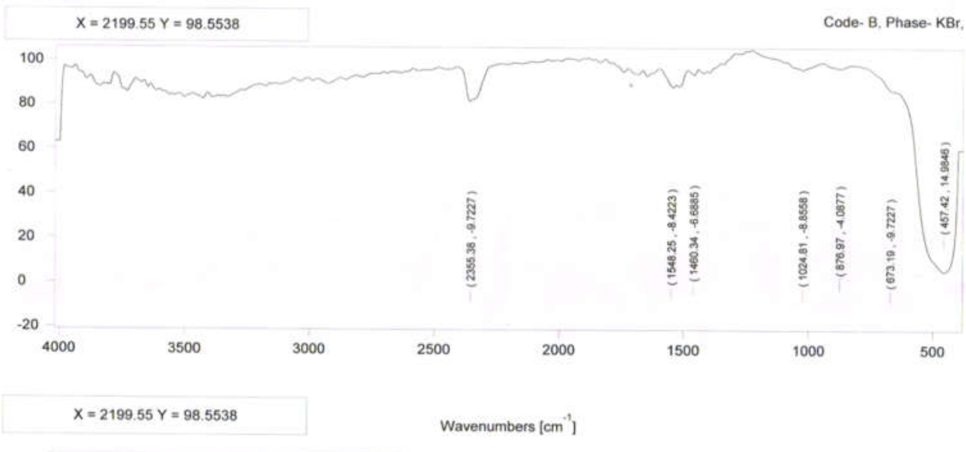

Figure 4: FTIR of TiO2

Dynamic Light Scattering: Nanoparticle size, poly dispersity index and zeta potential were characterized with dynamic light scattering (DLS). DLS data shows the TiO_2 nanoparticles have a hydrodynamic diameter of 225.4 nm and 247.7 nm in water and complete medium, along with the zeta potential values of -27.9 mV and -9.52 mV respectively.

Sample	Average size (nm)	Zeta potential (mV)	PdI
TiO_2 (water)	225.4	-27.9	0.225
TiO_2 (media)	247.7	-9.52	0.243

Particle Size Analyser :- The particle size of TiO_2 is found to be 18.88 nm.

x_{10} = 15.46 nm x_{50} = 18.88 nm x_{90} = 23.89 nm SMD= 18.86 nm VMD = 19.37 nm

x_{16} = 16.06 nm x_{84} = 22.68 nm x_{99} = 28.91 nm S_V = 318.11 m^2/cm^3

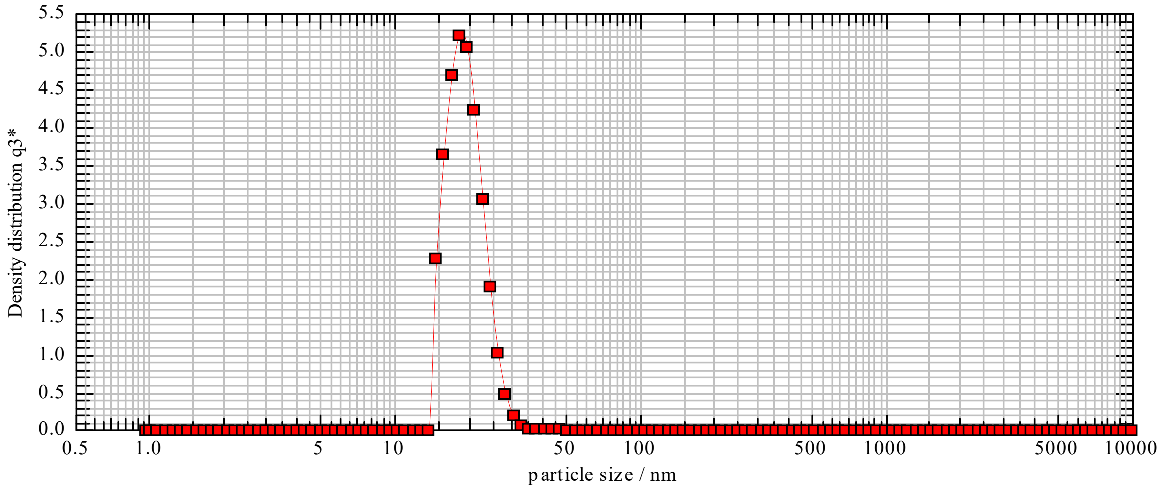

Figure 5: Particle size analysis of TiO_2 nanoparticles

PI uptake:

PI uptake TiO_2 NPs was done to assess the cytotoxicity. The dead cells percentage was calculated in this study. Titanium dioxide has displayed the cytotoxicity which was dependent on dose. The results shows that there is increase in the cytotoxicity from Titanium dioxide as its concentration increase from 1-75 µg/ml (Figure 6).

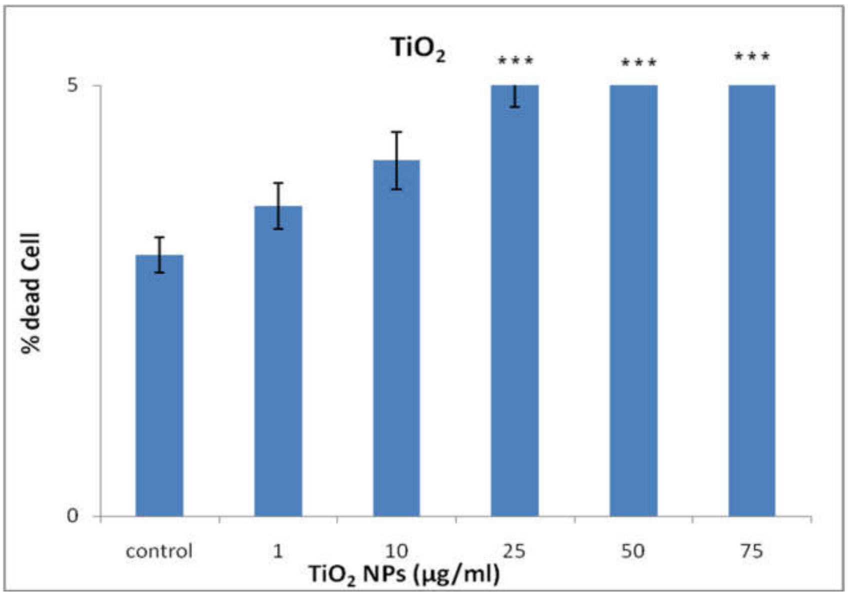

Figure 6: PI uptake analysis by flow cytometry

CONCLUSIONS

In this work, the TiO_2 nanoparticles were synthesized by sol-gel method and their PI uptake in CHO cells was evaluated. Titanium dioxide nanoparticles shows cytotoxicity on CHO cell line, that increase with the increase in concentration of Titanium dioxide 1-75 ug/ml.

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