https://doi.org/10.33472/AFJBS.6.6.2024.891-898

African Journal of Biological Sciences

Journal homepage: http://www.afjbs.com

Research Paper Open Access

Green synthesis of SnO_2 nanoparticles and its cytotoxicity and antioxidant properties

D. Madhumalathi[1, 2] and S. Kanimozhi[3]

[1]*Department of Science and Humanities, Bharath Institute of Higher education and Research, Selaiyur, Chennai, India*

[2]*Department of Biochemistry, Auxilium College (Autonomous), Vellore, India*

[3]*Department of Industrial Biotechnology, Bharath Institute of Higher education and Research, Selaiyur, Chennai, India*

Article Info

Volume 6, Issue 6, May 2024

Received: 28 March 2024

Accepted: 30 April 2024

doi: 10.33472/AFJBS.6.6.2024.891-898

Abstract:

This study focused on synthesizing tin dioxide (SnO_2) nanoparticles using extracts from Sugarcane leaves (*Saccharum officinarum)* of Poaceae family. The aim is to use green methods by using plant materials to create the nanoparticles, as opposed to chemical methods that can be hazardous. Tin dioxide nanoparticles (SnO_2 NPs) are considered essential and interesting nanomaterial, and this study aimed to use agricultural waste to produce them. XRD, SEM with EDX, TEM and were used to characterize the nanoparticles. XRD spectrum shows the crystalline nature of SnO_2 nanoparticle. The presences of functional groups in the nanoparticle are confirmed by FT-IR analysis. SEM and TEM images illustrate the spherical shape of nanoparticle. Synthesized SnO_2 nanoparticles were evaluated for cytotoxicity and antioxidant properties. The study revealed that, SnO_2 nanoparticle shows remarkable cytotoxic and antioxidant activity.

Keywords: Tin dioxide nanoparticles, green synthesis, Characterization, cytotoxicity, antioxidant

1. Introduction

Nanotechnology has grown rapidly in recent years and has many potential applications, including medical applications (Salata et al 2004, Gwinn et al 2006). Various techniques have been utilized to create nanoparticles, encompassing physical, chemical, and biological methodologies. Other methods are expensive and often involve the use of harmful chemicals (Geoprincy et al 2013). However, the biological approach is cost-effective, environmentally friendly, efficient, safe, and requires less energy (Santhoshkumar et al 2014). The use of plant-mediated synthesis has proven to be a superior method compared to conventional physicochemical techniques. Not only is it simple and environmentally friendly, but it produces a high-quality product free of impurities (Suresh et al 2019, Sudha et al 2019; Chakraborty et al 2022). This method does not require expensive equipment or the use of high temperatures and pressures. Various extracts from different plant species have been utilized for the eco-friendly synthesis of SnO_2 NPs (Garrafa-Galvez et al 2019). It is a non-toxic substance, making it widely used in the cosmetic industry Tin dioxide (SnO_2) possesses a distinctive property of reflecting infrared radiation, which makes it an excellent choice for a range of applications, including heat-reflecting windows and mirrors, dye-sensitized materials, liquid crystal displays, and humidity sensors (Ahmad et al 2021; Preethi et al 2021). There are various techniques for producing tin dioxide nanoparticles.

Sugarcane, which grows in subtropical regions, is an important source of sugar. Various products derived from sugarcane, such as cane brown sugar, sugarcane juice, and sugarcane molasses, have been studied for their biological effects (Grof et al 2001). During the sugar production process, sugarcane leaves are removed. These leaves, along with other byproducts like seed testa, hulls, and peels, are often wasted or used as feedstuff and fertilizer (Abdel-Rahman and Ahmed 2007). However, the disposal of sugarcane leaves has become a concern, and finding more rational uses for them is necessary. Plant leaves have been found to possess biological activity.

Sugarcane has been used to create various types of nanoparticles including iron, copper, silica, silver, carbon quantum dots, gold, and zinc oxide. Sugarcane is able to act as a precursor, stabilizing agent, and reducing agent in the synthesis process. This method of synthesis is environmentally friendly and cost-effective since it does not produce any toxic by-products and occurs at room temperature. Additionally, it is safe, quick, reproducible, easy to carry out, and bio-compatible (Velu et al 2017). Therefore, further research is encouraged in order to produce tin dioxide nanoparticles from sugarcane leaves that can be used in a variety of biomedical applications.

This study highlights the potential use of sugarcane leaf an abundant and eco-friendly agricultural waste, as a support for synthesis tin dioxide nanoparticles. Based on current research, there have been limited studies conducted on this particular subject. These produced metal nanoparticles will be examined and analyzed for their potential biomedical uses.

2. Materials and Methods

2.1.Preparation of Plant Extract

To prepare the leaf extract, we collected whole sugarcane leaves from local agricultural land in Kanchipuram district, Tamil Nadu, India. The leaves were washed multiple times in

distilled water to remove any dirt. We then cut 50 grams of sugarcane leaves into small pieces. The pieces were heated at the temperature of 60˚C for 30 mins with 250 mL of sterile distilled water in a 500 mL Erlenmeyer flask. We were able to get the extract from the leaves and let it cool down to room temperature undisturbed. Then, we filtered it using Whatman No. 42 and put it in the refrigerator at 4°C for future experiments.

2.2.Biosynthesis of SnO_2 Nanoparticles

The synthesis of SnO_2 NPs through plants is a straightforward process. To produce SnO_2 NPs, 0.529 grams of Tin chloride dihydrate ($SnCl_2 \cdot 2H_2O$) (0.05 M) were mixed with 30 mL of double-distilled water. Then, 30 mL of aqueous extract from Sugarcane leaves was slowly poured into the mixture while stirring at 60°C for 30 mins. The solution changed from a light green to a brown colour as a result of the heat. After cooling to room temperature, the mixture was centrifuged for 30 minutes at 5000 rpm, and the residue was washed three times with absolute ethanol and double-distilled water. Finally, it was dried on a hot plate at 60°C and calcined at 200°C for 3 hours.

2.3.Characterization techniques

The sample that was synthesized is analyzed for its phase and structural characterization using an XRD. To evaluate the surface morphology and composition, a scanning electron microscope and transmission electron microscope is used. The functional groups were analyzed by studying their FTIR spectrum.

2.4.Biomedical Applications

2.4.1. Cytotoxic activity

The effectiveness of SnO2 NPs in fighting cancer was studied using the MTT assay. MCF-7 human breast cancer cell line were tested using a colorimetric-based 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) assay(Ciapetti et al1993). In the experiment, cells were cultured in a 96-well plate containing 10% (v/v) FBS and DMEM and then treated with varying concentrations of SnO2 (25, 50, 100, 150, and 200) prepared in 0.4% DMSO. The cells were treated separately for 24 hours before performing the MTT assay and measuring the optical density with a micro plate reader at 595 nm, with 655 nm as reference.

2.4.2. Free Radical Scavenging activity

The scavenging abilities of both natural and synthetic substances have been studied using a DPPH assays (Brand-Williams et al. 1995). To determine the free-radical scavenging activity of SnO2 NPs from sugarcane leaves various concentrations of 5, 10, 25, 50 and 100µg/ml sample solutions were prepared with 10 ml of methanol. Meanwhile, a 100 ml flask containing 1M 7.89 mg of DPPH was dissolved in 99.5% methanol. The absorbance of DPPH decreases over time, stabilizing after about 1 hour, so the solution was kept in the dark for 4 hours. Next, different concentrations of the analytical sample were taken and mixed with 1 ml of DPPH in a sampling tube. The absorbance was measured at 517 nm using a UV/VIS Spectrophotometer.

3. Results and Discussion

3.1.Characterization of SnO_2 Nanoparticles

3.1.1. XRD

Figure 1 depicts the XRD pattern of synthesized SnO2 nanoparticle. The XRD peaks are observed at angles (2θ) of 26.74°, 33.98°, and 38.06°, which correspond to the (110), (101),

and (200) planes of SnO2 NPs. These peaks indicate the presence of tetragonal rutile-type SnO2 crystalline structure (JCPDS No. 41-1445) as per the standard diffraction peaks. The Scherrer equation revealed that the SnO2's average crystallite size was 38.9 nm.

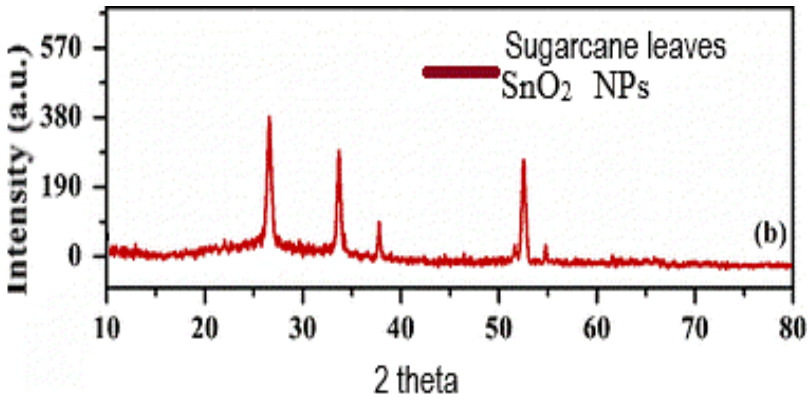

Fig. 1: XRD pattern of synthesized SnO_2

3.1.2. FT-IR Spectroscopy

FT-IR spectrum of SnO_2 NPs at 4500–500 cm-1 was depicted in figure 2. The spectra exhibit a significant absorbance peak for SnO2 at 3368.56 cm^{-1}, which is associated with the polyphenols' -OH stretching vibration. Both the symmetric and asymmetric -CH stretching vibrations of the methyl group are responsible for the absorption bands that are positioned at approximately 2967.32 cm^{-1}. Sugarcane leaves existed strong absorption bands are present for the -C-O-H bending vibration at 500–650 cm^{-1}, and the -C-H bending vibration of the alkane at 594.89 cm^{-1}.

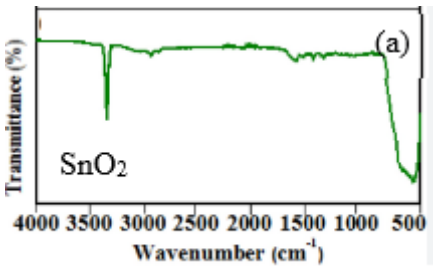

Fig. 2 FT-IR spectrum of SnO_2 nanoparticle

3.1.3. Morphology study of catalyst

The size and surface appearance of SnO2 nanoparticles were investigated using scanning electron microscopy (SEM), as displayed in Fig 3. SEM demonstrates that the shape of aggregated SnO2 NPs was studied using a 500 nm scale bar. The image reveals the presence of numerous ridge and valley structures. Figure 4 depicts the EDX spectrum, which shows peaks indicating the presence of O and N at 1.9 keV and 0.8 keV respectively. Sharp peaks at 3.8 keV and 3.7 keV indicate the presence of Sn in the sample. The EDX spectrum also revealed the presence of C in the sample, which could be due to the plant leaf extract. Morphology study of SnO2 NP was also determined by TEM analysis which is depicted in

figure 5. TEM image (Fig 4) shows the majority of the SnO2 NP is agglomerated and spherical in shape**.**

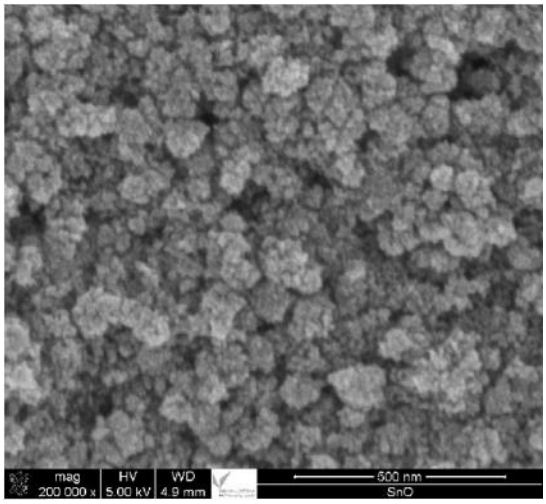

Fig. 3 SEM image of SnO_2 nanoparticles

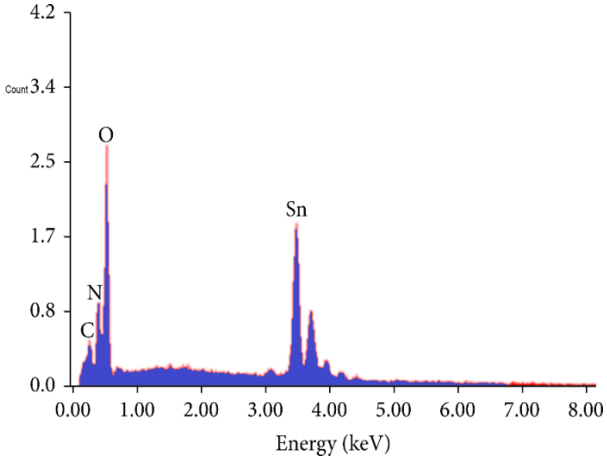

Fig. 4 EDX Spectrum of SnO_2 nanoparticle

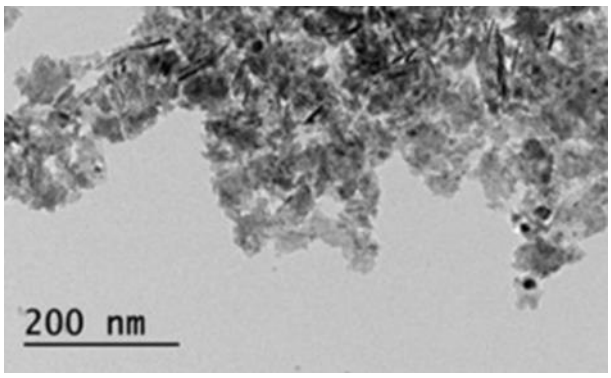

Fig. 5 TEM image of SnO_2

3.2.Biomedical Applications

3.2.1. Cytotoxic activity

Cytotoxicity analysis was performed to evaluate the properties of SnO_2 nanoparticle. MTT assay method was utilized to perform cytotoxicity using MCF 7 human breast cancer cell

line. Figure 6 illustrated the cell viability at different concentrations (25, 50, 75, 100, 125, 150, 175 and 200 µg) of SnO_2 nanoparticle. When SnO_2 nanoparticles were applied to cell lines, a notable proportion of cytotoxicity was noted. Increases in nanoparticle concentration result in a decrease in cell viability %, indicating that the concentration of SnO_2 nanoparticles was a determining factor in the reduction in percentage. A key criterion in the cytotoxicity investigation was the IC50 value. The concentration at which 50% of cell growth was inhibited is known as the IC50 value. Based on the findings, SnO_2 had a maximum IC50 value of 118 µg on the MCF7 cell line.

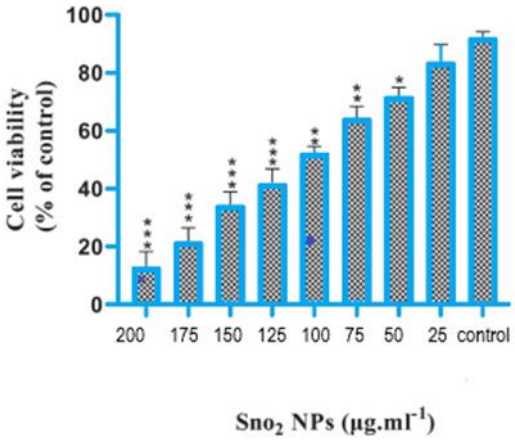

Fig. 6 Cytotoxic activity of SnO_2

3.2.2. Free radical Scavenging activity

The standard drug Ascorbic acid was used as standard for DPPH radical scavenging activity. It was carried out in order to have better comparative study with SnO2. The concentration of nanoparticle 10, 25, 50 and 100 µg/ml for all antioxidant a ssay, where ascorbic acid was taken in 5, 10, 25 and 50 µg/ml. Because it is found that if standard exceeds 50 µg/ml, it reached maximum absorbance value. So, in antioxidant assay the standard concentration are taken at range of 5-50 µg/ml to have better comparative study. The well-known and more stable free radical DPPH is based on the reduction of absorbing hydrogen or electron from donors. A colour change was used to measure the SnO2 ability to reduce DPPH, and the control exhibits no color change at all. The color shift that occurs when SnO2 are added to a DPPH solution is caused by the DPPH being scavenged by the contribution of a hydrogen atom to stabilize the DPPH molecule. SnO2 nanoparticle effectively inhibited the DPPH scavenging property (Figure 7). The DPPH assay showed the maximum activity of 89.25% at 100 µg/mL. The radical scavenging activity increased by increasing the concentrations of nanoparticles. Hence, the radical scavenging activity increased in dose dependent manner.

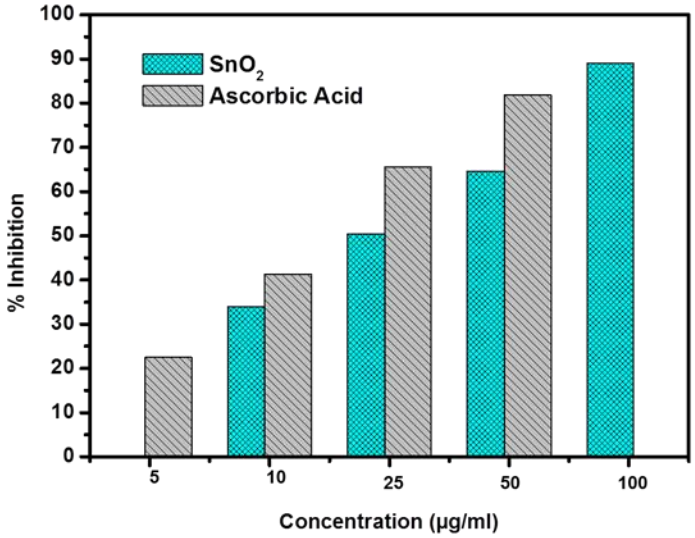

Fig. 7 Antioxidant activity of synthesized of SnO_2 Nanoparticle

4. Conclusion

The green synthesis approach, which is regarded as one of the cost-effective techniques with a high yield at lower concentrations of plant extract, was used to create the SnO_2 nanoparticles from agricultural waste of *Saccharum officinarum* leaves extract. SnO_2 nanoparticle production was confirmed using XRD and FTIR methods. The XRD result suggested that the particles were organized in a crystal structure and were crystalline in nature. The morphology of SEM and TEM images reveals the agglomerated and spherical shape of SnO_2 nanoparticles. The presence of functional groups in SnO_2 nanoparticles was verified by FTIR analysis. *Saccharum officinarum* leaves extract was used to create the SnO_2 nanoparticles, which demonstrated exceptional cytotoxic activity against MCF7 human breast cancer cell lines. Free radical scavenging assay clearly indicate that the medicinal role of SnO_2 nanoparticle could be due to its strong antioxidant potential. Hence the synthesized SnO_2 nanoparticles from the agricultural waste can be used for biomedical applications.

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