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Biogenic Production and Physicochemical Characterization of ZnO Nano Fertilizer from *Punica Granatum* Peel Extract and their Impact on *Sorghum Bicolor* Growth

Amarvani P Kanjikar
Department of Biotechnology, Karnatak Art's Science and Commerce College, Bidar, Karnataka, India

Abstract: ***The present study focused on the green synthesis, characterization, and agricultural application of zinc oxide (ZnO) nanoparticles using* Punica granatum *(pomegranate) peel extract as a natural reducing and stabilizing agent. The synthesized ZnO nanoparticles were characterized using UV–Visible spectroscopy, Fourier Transform Infrared Spectroscopy (FTIR), X-ray Diffraction (XRD), and Scanning Electron Microscopy (SEM). UV–Visible spectroscopic analysis showed a characteristic absorption peak is 350-360 in the UV region, confirming the formation of ZnO nanoparticles due to surface plasmon resonance. FTIR analysis revealed the presence of various phytochemicals such as phenols, flavonoids, and other functional groups responsible for the reduction and stabilization of nanoparticles. XRD analysis exhibited distinct diffraction peaks corresponding to the hexagonal wurtzite crystalline structure of ZnO nanoparticles, confirming their crystalline and pure nature. SEM analysis demonstrated predominantly quasi-spherical to irregular morphology with slight agglomeration and nanoscale particle size distribution. The biosynthesized ZnO nanoparticles were further evaluated for their nanofertilizer effect on sorghum (*Sorghum bicolor*) seed germination and plant growth. The ZnO nanofertilizer treatment significantly improved seed germination, root and shoot, and overall plant growth compared to untreated control plants. The enhanced growth performance may be attributed to improved nutrient uptake, higher surface area, and efficient zinc availability provided by the nanoparticles. The study concludes that green synthesized ZnO nanoparticles derived from* Punica granatum *peel extract possess promising structural and morphological properties and can be utilized as an eco-friendly and sustainable nanofertilizer for enhancing sorghum growth and agricultural productivity.***

Keywords: **Punica granatum, Sorghum bicolor, *UV–Visible spectroscopy, Fourier Transform Infrared Spectroscopy (FTIR), X-ray Diffraction (XRD), and Scanning Electron Microscopy (SEM).***

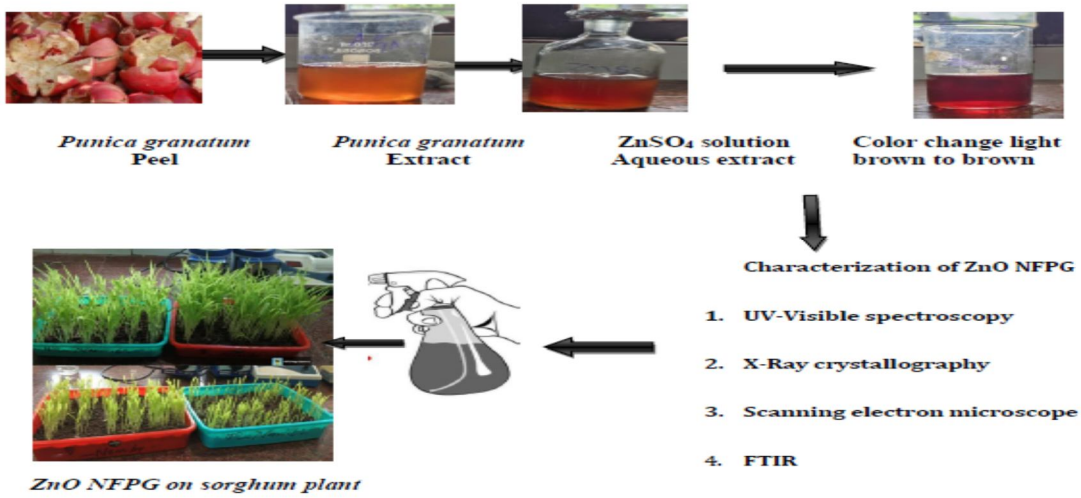


Figure-1: Graphical abstract of biosynthesis and characterization of zno nanofertilizer from *Punica granatum* peel extract and its effect on Sorgham growth.

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I. INTRODUCTION

Agricultural sustainability is a critical global priority in the face of increasing population pressure, climate change, and declining soil fertility [1]. Conventional agricultural practices rely heavily on chemical fertilizers to enhance crop productivity; however, these inputs often exhibit poor nutrient use efficiency due to leaching, volatilization, and chemical fixation in soil matrices. Such inefficiencies not only reduce crop yield but also contribute to environmental pollution, including groundwater contamination and soil degradation [2]. Therefore, there is a pressing need for innovative and sustainable nutrient management strategies.

Micronutrients play a vital role in plant growth and development, among which zinc is particularly significant. Zinc functions as a structural, catalytic, and regulatory component of numerous enzymes and proteins. It is involved in essential physiological processes such as chlorophyll biosynthesis, photosynthesis, carbohydrate metabolism, auxin synthesis, and membrane integrity. Zinc deficiency is one of the most widespread micronutrient deficiencies in agricultural soils worldwide, especially in tropical and subtropical regions. Deficiency of zinc in plants leads to stunted growth, chlorosis, reduced grain yield, and poor nutritional quality [3].

Nanotechnology has emerged as a promising tool in modern agriculture, offering novel solutions to improve nutrient delivery systems. Nanofertilizers, particularly zinc oxide nanoparticles (ZnO NPs), have gained considerable attention due to their unique physicochemical properties, including high surface area-to-volume ratio, enhanced reactivity, and controlled release behavior. These properties facilitate improved nutrient absorption, translocation, and utilization within plant systems compared to conventional fertilizers [4].

Despite their advantages, traditional methods for synthesizing nanoparticles—such as chemical reduction and physical methods are often associated with high energy consumption, toxic reagents, and environmental hazards [5]. In response, green synthesis approaches using biological materials have gained prominence. Plant-mediated synthesis of nanoparticles is particularly advantageous due to its simplicity, cost-effectiveness, scalability, and eco-friendliness. Plant extracts contain a diverse array of bioactive compounds such as polyphenols, flavonoids, alkaloids, and tannins, which act as reducing, capping, and stabilizing agents during nanoparticle formation [6].

Among various plant-derived materials, the peel of *Punica granatum* is an abundant agro-industrial waste rich in phenolic compounds, ellagitannins, and flavonoids. These photochemical not only facilitate nanoparticle synthesis but also enhance stability and functionality. Utilizing such waste materials aligns with sustainable development goals and circular economy principles by converting waste into value-added products [7, 8].

Sorghum bicolor is a globally important cereal crop known for its adaptability to semi-arid environments and its role as a staple food, fodder, and biofuel source [9,10]. However, sorghum productivity is often constrained by micronutrient deficiencies, particularly zinc, which affects both yield and grain nutritional quality [11] . Improving zinc availability through nanofertilizer application can significantly enhance sorghum growth, stress tolerance, and nutrient content.

In this context, the present study aims to (i) synthesize ZnO nanoparticles using pomegranate peel extract via a green route, (ii) characterize the synthesized nanoparticles using advanced analytical techniques, and (iii) evaluate their efficacy as a nanofertilizer on sorghum growth, physiological, and biochemical parameters. This work contributes to the integration of nanotechnology and sustainable agriculture, providing an eco-friendly alternative to conventional fertilization practices.

II. MATERIALS AND METHODS

A. Materials

Fresh peels of *Punica granatum* were collected from local markets. Zinc nitrate hexahydrate ($Zn(NO_3)_2 \cdot 6H_2O$) was used as the precursor for ZnO nanoparticle synthesis. Sodium hydroxide (NaOH) was used for pH adjustment. Ethanol and distilled water were used for washing and preparation. Certified seeds of Sorghum bicolor were obtained from an agricultural research supplier. All chemicals used were of analytical grade.

B. Preparation of Pomegranate Peel Extract

1) Collection, Cleaning, and Drying

Fresh pomegranate peels were thoroughly washed with running tap water followed by rinsing with distilled water to remove surface contaminants. The cleaned peels were shade-dried at ambient temperature (25–30°C) for 5–7 days to preserve thermo labile photochemical. The dried material was cut into small pieces and ground into a coarse powder using a mechanical grinder.

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2) Extraction Procedure

Approximately 20 g of powdered peel was mixed with 200 mL of distilled water and heated at 80°C for 40 minutes under continuous stirring. The mixture was allowed to cool to room temperature and filtered through Whatman No.1 filter paper. The filtrate containing bioactive compounds was stored at 4°C and used within 48 hours [12].

C. Green Synthesis of ZnO Nanoparticles

For the biosynthesis of zinc oxide nanoparticles, 50 mL of pomegranate peel extract was added dropwise to 100 mL of zinc sulphate solution under continuous magnetic stirring. The reaction mixture was maintained at 70 °C for 2 h to facilitate the reduction of zinc ions and nucleation of nanoparticles. Subsequently, the pH of the mixture was gradually adjusted to approximately 10 using 0.1 M sodium hydroxide (NaOH), which promoted the formation of ZnO nanoparticles. The appearance of a white precipitate confirmed the successful biosynthesis of zinc oxide nanoparticles [13].

The reaction mixture was centrifuged at 10,000 rpm for 15 minutes. The obtained precipitate was washed multiple times with distilled water and ethanol to remove residual impurities and unreacted components. The purified sample was dried at 60°C overnight and calcined at 400°C for 3 hours in a muffle furnace to obtain crystalline ZnO nanoparticles [14].

1) Fourier Transform Infrared (FTIR) Spectroscopy

FTIR analysis was performed to identify the functional groups present in the pomegranate peel extract and their role in nanoparticle synthesis and stabilization. The spectra were recorded in the range of 400–4000 cm^{-1} [15].

2) Scanning Electron Microscopy (SEM) Analysis

SEM analysis was performed to examine the surface morphology, particle size, and aggregation behavior of ZnO nanoparticles. The samples were coated with a thin layer of gold prior to imaging to enhance conductivity [16].

3) X-ray Diffraction (XRD) Analysis

The crystalline structure of the synthesized ZnO nanoparticles was analyzed using X-ray diffraction (XRD). Prior to analysis, the nanoparticles obtained through green synthesis using *Punica granatum* peel extract were purified by repeated centrifugation and washing with distilled water and ethanol to remove residual impurities and photochemical. The purified sample was then dried in a hot air oven at 60–80°C to obtain a fine powder, which was gently ground to ensure uniform particle size distribution. The XRD analysis was carried out using a Bruker D8 Advance X-ray Diffractometer (or equivalent instrument) with Cu Kα radiation (λ = 1.5406 Å) operated at 40 kV and 30 mA. The diffraction pattern was recorded over a 2θ range of 20° to 80° with a scanning rate of 2° per minute and a step size of 0.02°. The powdered sample was evenly spread on a sample holder to obtain a smooth surface for accurate measurement. The obtained diffraction peaks were analyzed by comparing them with standard data from the Joint Committee on Powder Diffraction Standards (JCPDS) database to confirm the phase and crystalline structure of ZnO nanoparticles [17].

4) Evaluation of ZnO Nanofertilizer on Sorghum Growth

The effect of biosynthesized ZnO nanoparticles on seed germination and plant growth of sorghum (*Sorghum bicolor*) was evaluated under laboratory conditions. Healthy and uniform sorghum seeds were surface sterilized using 0.1% mercuric chloride solution for 2–3 minutes and thoroughly washed several times with sterile distilled water to remove any contaminants. The synthesized ZnO nanoparticles were dispersed in distilled water using sonication to obtain a uniform nanofertilizer suspension at the desired concentration. The sterilized seeds were divided into control and treatment groups. For the treatment group, sorghum seeds were soaked in ZnO nanoparticle suspension for 6–8 hours, while control seeds were soaked in distilled water. After treatment, the seeds were placed in sterile Petri dishes lined with moist filter paper and incubated under controlled environmental conditions at room temperature. Distilled water was added regularly to maintain adequate moisture throughout the experimental period [18].

Seed germination was recorded after 5–7 days, and the germination percentage was calculated. The root length and shoot length of germinated seedlings were measured using a ruler. The seed vigor index was calculated using the formula [19].

Seed Vigor Index=Germination Percentage× (Root Length+Shoot Length) Fresh weight of seedlings was measured immediately after harvesting, while dry weight was determined after drying the seedlings in a hot air oven at 60°C until constant weight was obtained. Chlorophyll content was estimated using standard acetone extraction methods. All experiments were conducted in triplicates, and the obtained results were expressed as mean ± standard deviation. The growth parameters of ZnO nanoparticle-treated plants were compared with untreated control plants to determine the nanofertilizer efficiency of the synthesized ZnO nanoparticles [20].

III. RESULT AND DISCUSSION

The complete study of green synthesis and characterization of ZnO nanoparticles using *Punica granatum* fruit peel extract as carried out and is reported in this paper. An UV-Vis Spectroscopy is one of the major techniques carried out to study the formation of metal nanoparticles that shows the absorbance peak at 355nm. Fig.1 provided Surface Plasmon resonance exits for the metal. Fig.2. shows the sample tubes containing UV-Vis Spectrophotometer Analysis of (sample-3) *Punica granatum* peel Extract, (sample-1) ZnSO4, (sample-2) ZnNps.. It is observed that the colour of the solution has changed from light colour to brown with increasing incubation time. The colour yellowish brown colour arises due to surface Plasmon vibration of metal nanoparticles.

A. FTIR Spectrum Revealed several Characteristic Peaks

The FTIR spectrum of the synthesized ZnO nanoparticles showed a broad peak around 3200–3400 cm^{-1} corresponding to O–H stretching vibrations of phenols and alcohols present in the pomegranate peel extract. Peaks near 1600–1700 cm^{-1} indicated C=O stretching of carbonyl groups, suggesting the involvement of phytochemicals in nanoparticle synthesis and stabilization. A peak around 1400 cm^{-1} was attributed to C–H bending vibrations, while peaks below 600 cm^{-1} confirmed Zn–O stretching vibrations, indicating successful formation of ZnO nanoparticles (Figure-3). Similar FTIR findings were reported by Sangeetha et al. (2011)**,** Vijayakumar et al. (2018)**,** and Ramesh et al. (2015) in plant-mediated synthesis of ZnO nanoparticles, confirming the role of bioactive compounds as reducing and capping agents.

B. SEM analysis of ZnO NF of Punica granatum

The SEM analysis of the biosynthesized ZnO nanoparticles revealed predominantly quasi-spherical to irregularly shaped particles with moderate agglomeration (Figure 4). The nanoparticles were distributed in the nanometer range and exhibited relatively uniform morphology. However, slight clustering of particles was observed, which may be attributed to the high surface energy and polarity of ZnO nanoparticles. The aggregation behavior could also be influenced by the presence of phytochemicals such as polyphenols, flavonoids, and tannins from *Punica granatum* peel extract, which act as reducing and capping agents during nanoparticle synthesis. The obtained SEM results are consistent with previous reports on green synthesized ZnO nanoparticles. Gunalan et al. (2012) reported spherical and aggregated ZnO nanoparticles synthesized using *Aloe vera* extract, while Bhuyan et al. (2015) observed irregular and clustered nanoparticles produced using *Azadirachta indica* leaf extract. Similarly, Diallo et al. (2015) reported quasi-spherical ZnO nanoparticles with slight agglomeration due to intermolecular interactions and stabilization by plant biomolecules. The similarity between the present findings and earlier studies confirms the successful biosynthesis of ZnO nanoparticles using pomegranate peel extract.

C. XRD analysis

The XRD analysis confirmed the crystalline nature and phase purity of the synthesized ZnO nanoparticles. The diffraction pattern showed characteristic peaks at 2θ values around 31.7°, 34.4°, 36.2°, 47.5°, 56.6°, 62.8°, 66.3°, 67.9°, and 69.1°, corresponding to the planes (100), (002), (101), (102), (110), (103), (200), (112), and (201), respectively as shown in Figure-5. These peaks are characteristic of the hexagonal wurtzite structure of ZnO nanoparticles and are in agreement with standard JCPDS data.

Similar diffraction patterns were reported by Fakhari et al. (2019)**,** who synthesized ZnO nanoparticles using plant extracts and observed intense peaks corresponding to the hexagonal crystalline structure of ZnO. Likewise, Elumalai and Velmurugan (2015) reported characteristic ZnO peaks at similar 2θ positions, confirming high crystallinity and purity of biosynthesized nanoparticles. The absence of additional impurity peaks in the present study suggests that the synthesized ZnO nanoparticles were highly pure and crystalline. The sharp diffraction peaks further indicate good crystallization of nanoparticles, which may enhance their effectiveness as nanofertilizers.

D. Evaluation of ZnO Nanofertilizer on Sorghum Growth

The application of biosynthesized ZnO nanofertilizer significantly enhanced seed germination and growth parameters of *Sorghum bicolor* compared to the untreated control. Seed germination increased from 72% in the control to 91% in the ZnO nanoparticle-treated group, indicating improved seed metabolic activity and nutrient availability (Figure-6). Root length and shoot length also showed substantial increases of 65.38% and 72.97%, respectively, suggesting enhanced cell division and elongation due to zinc-mediated enzymatic activity.

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The seed vigor index exhibited a remarkable increase of 114.66%, demonstrating the positive influence of ZnO nanoparticles on early seedling establishment (Table-1). Similarly, fresh and dry biomass accumulation increased significantly in treated plants, indicating improved nutrient uptake and photosynthetic efficiency. Chlorophyll content also increased by 60.15%, which may be attributed to the role of zinc in chlorophyll biosynthesis and activation of photosynthetic enzymes (Table-1).

These findings are consistent with the reports of Tarafdar et al. (2014)**,** who observed enhanced germination and biomass production in pearl millet treated with ZnO nanoparticles. Similarly, Raliya and Tarafdar (2013) reported improved shoot growth, chlorophyll content, and nutrient uptake in cluster bean following ZnO nanoparticle application. Subbaiah et al. (2016) also demonstrated that ZnO nanoparticles significantly promoted seedling vigor and plant growth due to better zinc availability and enhanced physiological activities. The present study therefore confirms the potential of green synthesized ZnO nanoparticles as an efficient nanofertilizer for improving sorghum growth and productivity.

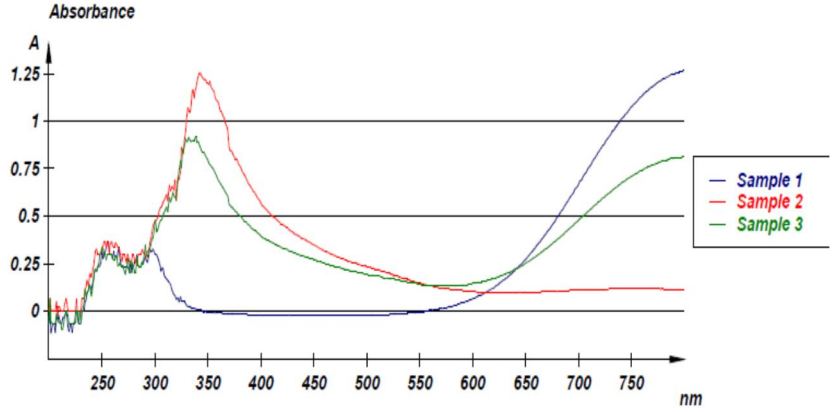


Figure-2: UV-Vis Spectrophotometer Analysis of (sample-3) *Punica granatum* peel Extract, (sample-1) ZnSO4, (sample-2) ZnNps.

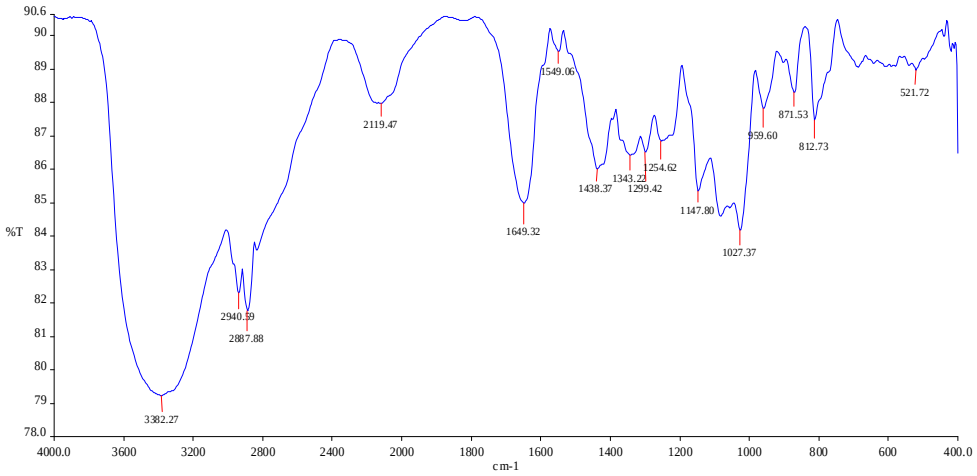


Figure-3: FTIR Analysis of ZnO nanoparticles of *Punica granatum* peels extract.

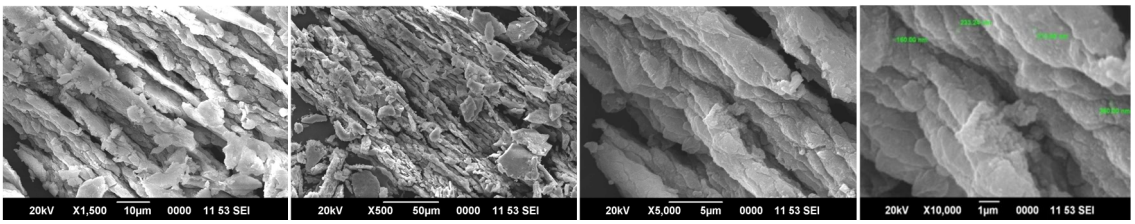


Figure-4: SEM evaluation of ZnO nanoparticles of *Punica granatum* peel extract.

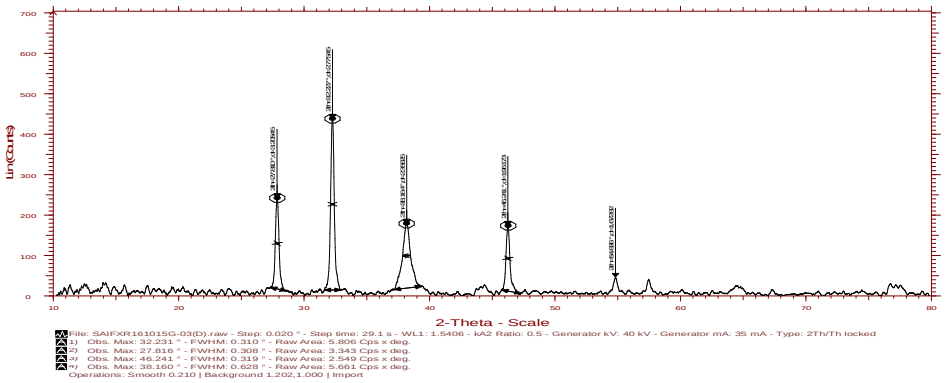


Figure-5: XRD results of of Zno-Nanofertilizer synthesized from Pomegranate.



Figure-6: Plant growth effect of Zno-Nanofertilizer synthesized from Pomegranate and control.

Table-1: Effect of ZnO Nanofertilizer on Sorghum Seed Germination and Plant Growth.

Parameters	Control	ZnO Nanofertilizer Treatment	Percentage Increase (%)
Seed Germination (%)	72 ± 1.5	91 ± 1.2	26.38
Root Length (cm)	5.2 ± 0.3	8.6 ± 0.4	65.38
Shoot Length (cm)	7.4 ± 0.5	12.8 ± 0.6	72.97
Seed Vigor Index	907 ± 12	1947 ± 18	114.66
Fresh Weight (g/plant)	1.84 ± 0.08	3.12 ± 0.11	69.56
Dry Weight (g/plant)	0.62 ± 0.03	1.15 ± 0.05	85.48
Chlorophyll Content (mg/g)	1.28 ± 0.04	2.05 ± 0.06	60.15

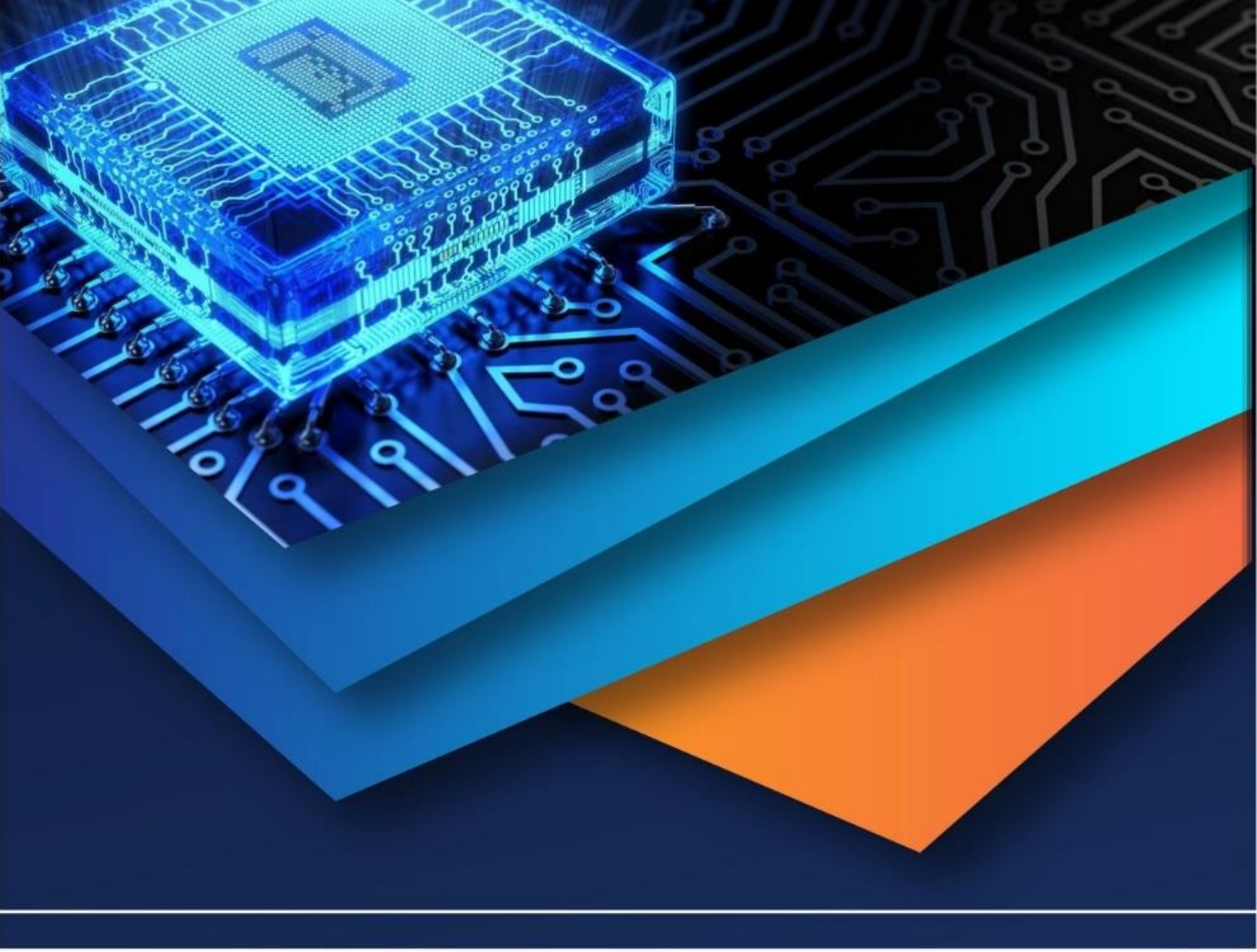
IV. CONCLUSION

The present study successfully demonstrated the green synthesis of zinc oxide (ZnO) nanoparticles using *Punica granatum* peel extract as an eco-friendly and sustainable reducing and stabilizing agent. Characterization studies confirmed the successful formation of ZnO nanoparticles. UV–Visible spectroscopy revealed the characteristic absorption peak of ZnO nanoparticles, while FTIR analysis confirmed the involvement of phytochemicals such as phenols and flavonoids in nanoparticle synthesis and stabilization. XRD analysis verified the highly crystalline hexagonal wurtzite structure of ZnO nanoparticles, and SEM analysis showed predominantly quasi-spherical to irregular morphology with slight agglomeration in the nanoscale range.

Furthermore, the synthesized ZnO nanoparticles exhibited significant nanofertilizer potential by enhancing seed germination percentage, root length, shoot length, seed vigor index, biomass accumulation, and overall growth performance of sorghum (*Sorghum bicolor*) compared to untreated control plants. The improved plant growth may be attributed to enhanced zinc availability, increased surface area, and efficient nutrient uptake provided by the nanoparticles. The findings suggest that biosynthesized ZnO nanoparticles derived from agricultural waste such as pomegranate peel can serve as an effective, low-cost, and environmentally friendly nanofertilizer for sustainable agricultural applications. This study highlights the potential of green nanotechnology in improving crop productivity while reducing dependence on conventional chemical fertilizers.

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