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Green Synthesis of Titanium Nanoparticles prepared from Citrus reticulata peels; its Characterization, Antioxidant and Anti-Inflammatory Activities

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Abstract

This study investigates the green synthesis, characterization, and biological importance of titanium dioxide (TiO_2) nanoparticles formed from the peels of *Citrus reticulata* (mandarin orange). The synthesized nanoparticles were characterized using various analytical techniques, including UV-vis spectrophotometry (UV-vis), Fourier-transform infrared spectroscopy (FTIR), and scanning electron microscopy (SEM). The FTIR of $CrTiO_2NP$ shows some noticeable peaks at 3430.53 (-OH), 1779.04, 1639.76, 1449.41, 1162.46, and 682 cm^{-1} revealed the presence of functional groups from the peel extract, indicating their role in the stabilization of the nanoparticles. SEM analyses provided insights into the morphology, shape, and size composition of the synthesized nanoparticles, SEM shows the spherical shape and particle size of $CrTiO_2NP$, which ranged from 72 to 135 nm. The LCMS revealed Neohesperidin, 3-feruloylquinic acid, Hesperidin, Rhamnoside, Nobiletin, Tangeretin, Dihydroquercetin, 4,5-dihydroxy-6,7-dimethoxyflavone, 7-methylcapillarisin. The antioxidant and anti-inflammatory activities of EEPCr) and $CrTiO_2NP$) were evaluated i.e. Cyclooxygenase % (21.42±0.32 (EEPCr), 23.13±1.62 ($CrTiO_2NP$), 63.252 while Dicloflenac was employed as the positive standard); Lipo-oxygenase %(32.46±1.42 (EEPCr), 53.53±0.13 ($CrTiO_2NP$), 75.141(Diclofenac)); NO % (31.73±0.79 (EEPCr),41.53±0.79 ($CrTiO_2NP$), 60.2321 (Diclofenac)). The findings of this study highlight the successful green synthesis of TiO_2 nanoparticles using *Citrus reticulata* peels, their physicochemical characteristics, and their promising biological applications, particularly in the fields of antiinflammation and antioxidant therapies. This eco-friendly approach to nanoparticle synthesis holds great promise for the development of sustainable and biocompatible nanomaterials with diverse applications in various industries.

Keywords: TiO_2, green synthesis, $CrTiO_2NP$, anti-inflammatory

Introduction

The development of green and sustainable nanotechnology has gained significant attention in recent years due to the growing concerns over the environmental impact and potential health risks associated with conventional chemical synthesis methods. One promising approach is the utilization of natural plant-based resources, such as citrus fruit peels, for the synthesis of nanoparticles. *Citrus reticulata*, commonly known as mandarin orange or tangerine, is a widely cultivated and consumed citrus species. The peels of *C. reticulata* are often considered waste products, despite their rich phytochemical composition, including various bioactive compounds like flavonoids, phenolic acids, and essential oils. These compounds possess potent antioxidant, anti-inflammatory, and antimicrobial properties, making them attractive candidates for the green synthesis of nanoparticles. Titanium nanoparticles (TiNPs) have garnered significant interest in various applications, including biomedical, cosmetic, and environmental remediation, due to their unique physicochemical properties, such as high surface area, enhanced reactivity, and strong antimicrobial activity.

The green synthesis of TiO_2NPs using natural plant-based resources, like *C. reticulata* peels, offers a sustainable and eco-friendly alternative to conventional chemical methods, which often involve the use of toxic reagents and generate hazardous waste. This study aims to explore the green synthesis of TiO_2NPs using *C. reticulata* peel extracts, characterize the synthesized nanoparticles, and evaluate their antioxidant and anti-inflammatory activities. The findings of this research contribute to the development of innovative, sustainable, and bioactive nanomaterials derived from waste citrus peels, with potential applications in various fields, including biomedicine, cosmetics, and environmental remediation.

MATERIAL AND METHODS

Materials

Collection of Plant Sample

Tangerine peels were collected from fruit sellers in Katsina Metropolis. The collected tangerine peels were air-dried in a room in the absence of sunlight. The dried peels were crushed using mortar and pestle to make a fine powder. The powder was stored in a polythene bag and kept for further analysis.

Extraction of Plant Sample

500 g of the powdered tangerine peels were subjected to extraction using n-hexane, ethyl acetate, and ethanol respectively. Each solvent was allowed to extract for four days after which the extract was concentrated using a rotary evaporator.

Preparation of Nanoparticles

Titanium dioxide (TiO_2, analytical grade, purity $\geq$ 99.85%) was used as a precursor directly without further purification for the synthesis of TiO_2 NPs and purchased from Fisher Scientific, U.K. The ethanol extract of the powdered citrus peel was utilized for the green synthesis of TiO_2 NPs. The TiO_2 nanoparticles were prepared at the Department of Chemistry, Faculty of Physical Science, Federal University Dutsin-Ma, Dutsin-Ma.

Preparation of TiO_2 solution

A concentration of 0.01M TiO_2 solution was prepared by dissolving 4.04 g TiO_2 in 1000 ml distilled water and was used for the green synthesis of iron nanoparticles.

Synthesis of peels of *Citrus reticulate* Nanoparticles (Cr-TiO_2NPs)

The nanoparticles, and peels of *Citrus reticulate* TiO_2 Nanoparticles (Cr-TiO_2NPs) were synthesized using the ethanol extract of peels of *Citrus reticulate* with 0.01M TiO_2 in the ratio 1:1 by volume. The ethanolic extracts of the peels of *Citrus reticulate* were added slowly to 0.01M aqueous TiO_2 solution and stirred for 60 minutes using a magnetic stirrer for the reduction (Sravanthi *et al*., 2018). A change of colour from an orange solution to a black precipitate indicates the formation of Nanoparticles.

Characterization of the Green Synthesized Nanoparticles

Ultraviolet-visible (UV-Vis) Spectroscopy

The UV-Vis spectroscopy will be used to validate the formation of NPs centered on their optical properties. The wavelength of green synthesized NPs based on light absorbance in the range of 300 to 600 nm with a resolution of 1 nm will be measured by loading the sample of 2 ml in quartz cuvette in UV-Vis spectroscopy.

Fourier Transform Infrared Spectroscopy (FTIR)

The FTIR is an analytical tool meant for the measurement of infrared intensity, wavelength, or wave number of light of the green synthesized NPs. Therefore, for FTIR analysis a sample of 0.5 g of NPs will be used and the spectra will be scanned in the range of 4000–400 cm^{-1} at a resolution of 4 cm^{-1} for the characterization of compound functional groups present over the surface of green synthesized NPs. The FTIR data measures the interaction between the metallic ion and protein molecules. Therefore, when an infrared light interacts with the sample of NPs, the chemical bonds show the stretching, contracting, and bending nature of nanoparticles as indicated by a peak in FTIR and the result will be matched with the standard library search of Infrared charts.

Scanning Electron Microscope (SEM)

In this study, SEM will be used to characterize the size and morphology of the nanoparticles. SEM with magnification ranging from 20X to approximately 30,000X with a spatial resolution of 50 to 100 nm will be used. For the imaging, a sample of 0.5 mg NPs will be dusted on one side of the double-sided adhesive carbon conducting tape. The tape will be mounted on an 8 mm aluminum stub. The sample will be observed at different magnifications and the images picturized.

Biological Activity of Ethanol Extract of Tangerine Peels and its TiO_2 Nanoparticles

Determination of Total Phenol

The total phenol content of the extract is determined by the method of (Singleton et. al., 1999). 0.2 ml of the extract was mixed with 2.5 ml of 10% Folin-ciocalteau's reagent and 2 ml of 7.5% Sodium carbonate. The reaction mixture was subsequently incubated at 45°C for 40 mins, and the absorbance was measured at 700 nm in the spectrophotometer, garlic acid would be used as standard phenol.

Determination of NO radical scavenging ability

Sodium Nitroprusside in aqueous solution at physiological pH spontaneously generates NO, which interacts with oxygen to produce nitrite ions that can be estimated by use of a Greiss reagent. Scavengers of NO compete with oxygen, leading to reduced production of NO. Briefly, 5mM sodium nitroprusside in phosphate-saline was mixed with the extract, before incubation at 25°C for 150 min. Thereafter the reaction mixture was added to the Greiss reagent. Before measuring the absorbance at 546 nm, relative to the absorbance of a standard solution of potassium nitrate treated in the same way with Greiss reagent (Ebrahimzadeh et al., 2010).

Determination of total carotenoid

Weigh 2.5 g of fine blended sample into a conical flask, add 30 ml of hexane and 20 ml of ethanol, and then add 2 ml of 2% NaCl. Mix very well and transfer content into a separating funnel, allowing it to stand for about 10 minutes to allow for extraction of carotenoid run the lower content off and collect the upper layer (extractant phase). Measure the absorbance @ 436 nm using a UV spectrophotometer (Bao et al., 2005).

Determination of Total Flavonoid

The total flavonoid content of the extract was determined using a colourimeter assay developed by (Bao, 2005). 0.2 ml of the extract was added to 0.3 ml of 5% $NaNO_3$ at zero time. After 5 min, 0.6 ml of 10% $AlCl_3$ was added and after 6 min, 2 ml of 1M NaOH was added to the mixture followed by the addition of 2.1 ml of distilled water. Absorbance was read at 510 nm against the reagent blank and flavonoid content was expressed as mg rutin equivalent

Anti-proteinase determination

The test was performed according to the modified method of Oyedepo et al. (2007) and Sakat et al. (2009) The reaction mixture (2 ml) contained 0.06 mg trypsin, 1 ml 20 mM Tris HCl buffer (pH 7.4) and 1 ml test sample was used. The mixture was incubated at 37°C for 5 min and then 1 ml of 0.8% (w/v) casein was added. The mixture was incubated for an additional 20 min. 2 ml of 70% perchloric acid was added to arrest the reaction. The cloudy suspension was centrifuged and the absorbance of the supernatant was read at 210 nm against the buffer as blank.

Heat-induced hemolysis

The reaction mixture (2 ml) consisted of 1 ml test sample and 1 ml of 10% RBCs suspension, instead of the test sample only saline was added to the control test tube. All the centrifuge tubes containing the reaction mixture were incubated in a water bath at 56 ºC for 30 minutes. At the end of the incubation, the tubes were cooled under running tap water. The reaction mixture was centrifuged at 2500 rpm for 5 min and the absorbance of the supernatants was taken at 560 nm.

Lipoxygenase activity

Anti-lipoxygenase activity was studied using linoleic acid as substrate and lipoxidase as enzyme. Test samples were dissolved in 0.25 ml of 2M borate buffer pH 9.0 and added 0.25 ml of lipoxidase enzyme solution (20,000U/ml) and incubated for 5 min at 250C. After which, 1.0ml of linoleic acid solution (0.6mM) was added, mixed well, and absorbance was measured at 234nm. Indomethacin was used as the reference standard.

LC-MS Analysis

Protocol for LCMS Analysis (Generic Method) using LC Waters e2695 separation module with W2998 PDA and couple to ACQ-QDA MS The samples were analyzed using liquid chromatography (LC) tandem mass spectrophotometer (MS) as described by (Piovesana*et al*., 2018) with some modifications. The extracted samples were reconstituted in methanol and filtered through a polytetrafluoroethylene (PTFE) membrane filter with 0.45 µm size. After filtration, the filtrate (10.0 µl) was injected into the LC system and allowed to separate on Sunfire C18 5.0 µm 4.6 mm x 150 mm column. The run was carried out at a flow rate of 1.0 mL/min, with Sample and Column temperature at 25°C. The mobile phase consists of 0.1% formic acid in water (solvent A) and 0.1% formic acid in acetonitrile (solvent B) with a gradient as below:

Table 1: Solvent Gradients A and B

Time	%A	%B
0	95	5
1	95	5
13	5	95
15	5	95
17	95	5
19	95	5
20	95	5

From the ratio of A/B 95:5 this ratio was maintained for a further 1 min, then A/B 5:95 for 13 min, to 15 min. then A/B 95:5 to 17 min, 19 min, and finally 20 min. the PDA detector was set at 210-400 nm with a resolution of 1.2 nm and a sampling rate of 10 points/sec. The mass spectra were acquired with a scan range from m/z 100 – 1250 after ensuring the following settings: ESI source in positive and negative ion modes; capillary voltage 0.8 kv (positive) and 0.8 kv (negative); probe temperature 600oC; flow rate 10 mL/min; nebulizer gas, 45 psi. MS is set in automatic mode applying a fragmentation voltage of 125 V (Piovesana*et al*., 2018). The data was processed with Empower 3. The compounds were identified based on the following information, elution order, retention time (Rt), fragmentation pattern, and Base m/z.

RESULTS AND DISCUSSION

Results

Extraction

The result for the extraction yield of *Citrus reticulate* of 500 g of powdered plant sample concerning the solvent medium used in the extraction is given in Table 1.

Formation of TiO_2Nanoparticles

Titanium dioxide (TiO_2, analytical grade, purity $\geq$ 99.85%) was used as a precursor directly without further purification for the synthesis of TiO_2 NPs and purchased from Fisher Scientific, U.K. The ethanol extract of the powdered citrus peel was utilized for the green synthesis of Cr-TiO_2NPs.

Table 2: Extraction of *Citrus reticulata*

SOLVENT	Weight of extract (g)	Percentage yield (%)
Ethanol	50	10.00
Ethyl acetate	28	5.60
n-hexane	15	3.00

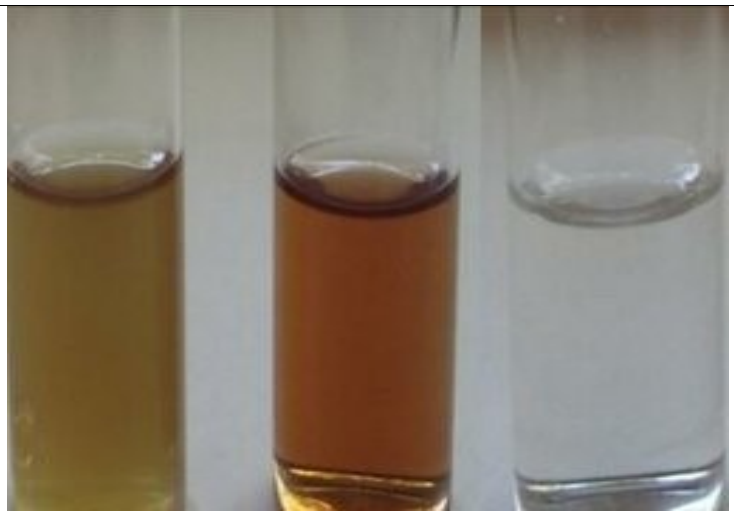

Figure 1: Colour Change of synthesized TiO_2nanoparticles from ethanol extract of *Citrus reticulate* peels

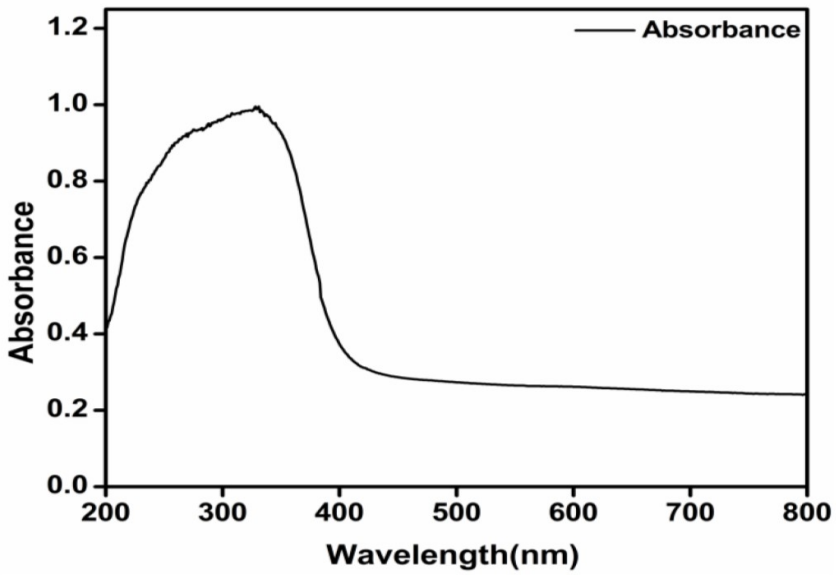

Figure 2: UV-Vis absorption spectra for the synthesized $CrTiO_2$-NP

Characterization

Ultraviolet-visible (UV-Vis) Spectroscopy

This Uv-vis spectroscopy was employed to examine further that $CrTiO_2$-NP has been formed, **Figure 4.1** shows that the ethanol extracts of *C. reticulate* were able to reduce ferric nitrate nonahydrate to iron nanoparticles at 380 nm being the surface plasma resonance. **Figure 4.1** revealed the curve absorbed by each spectrum that gave the iron nanoparticles within the wavelength range of 380 nm for $CrTiO_2$NP formed.

FTIR Spectroscopy

To evaluate ethanol extract and green-produced TiO_2 NPs from the ethanol extract of *C. reticulate*, FTIR spectroscopy was carried out on FTIR (Alpha II, Brucker, Billerica, MA, USA) spectrometer equipment. Additionally, the sample scan was performed using 64 scans at a resolution of 4 cm^{-1}, covering the wavenumber range of 4000–400 cm^{-1}.

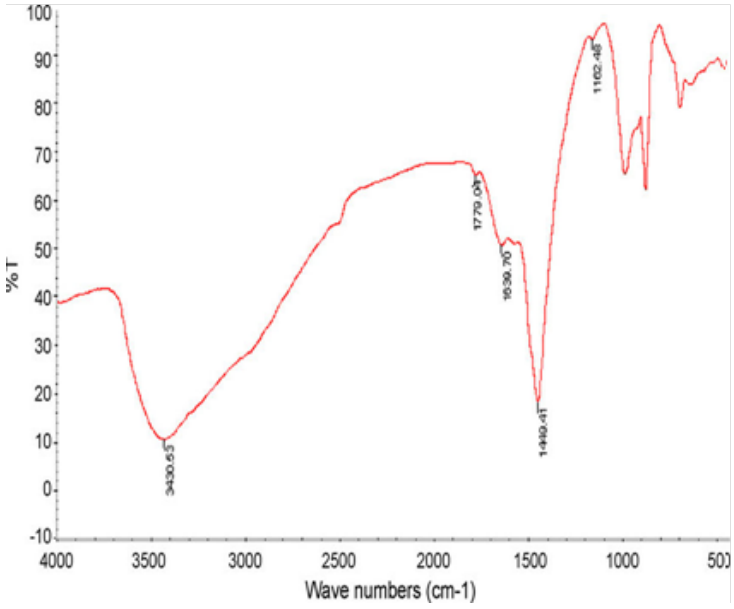

Figure 3: FTIR spectrum of TiO_2 nanoparticles

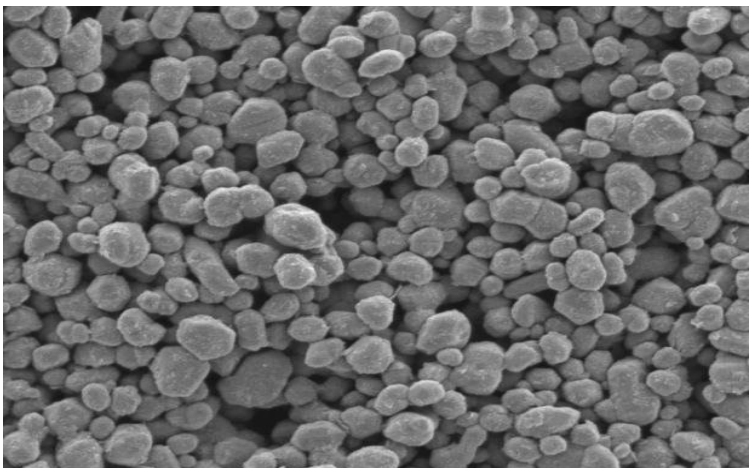

Figure 4: Scanning electron microscopy image showing nanoparticles

Table 3: Antioxidant Activity of EEPCr and $CrTiO_2$-NP

		C. reticulata	$CrTiO_2$-NP	Control
1	Flavonoid mg QE/g	32.15±1.32	34.45±0.37	35.552 µg/ml (Quercetin)
2	Phenol mg GAE/g	47.81±0.22	49.91±1.53	58.492 µg/ml (Gallic acid)
3	Carotenoids µg/mL	4.54±0.21	2.30±0.18	

SEM

The SEM images show that the NP conglomerate caused an increase in the TiO_2 NP size and that the TiO_2 NPs had a uniform spherical morphology. Figure 4.3 shows the SEM image of TiO_2 NPs prepared by green synthesis. Microscopic analysis revealed that the developed NP system is spherical and polygonal. In the current investigation, the surface morphology and size of the green synthesized TiO_2 NPs were studied using SEM analysis.

Biological Activities

Content Phenolic

Numerous studies indicate that because of their antioxidant action, phenolic chemicals are vital for human health. The total phenolic content of the ethanol extract of the peels of *C. reticulate* and its TiO_2NP was compared in Table 4.2. This was determined using the equation y = 0.006x + 0.027 (R2 = 0.990) to yield (47.81±1.71) mg gallic acid equivalent/g of extract and (49.91±1.53) mg gallic acid equivalent/g of its synthesized nanoparticle

Content of Flavonoids

As indicated by Table 4.2, the ethanol extract of the peels of *C. reticulata* and its TiO_2NP had a good quantity of flavonoid content (32.15 ± 1.32) mg quercetin equivalent/g of extract after the calibration curve (y = 0.022x + 0.182; R2 = 0.994) was constructed based on the obtained data while that of $CrTiO_2$-NP is 34.45±0.37 mg quercetin equivalent/g

Content of Carotenoids

Table 4.2 compares the carotenoid content of ethanol extract of the peels of *C. reticulata* and $CrTiO_2$-NP. This indicates that the ethanol extract of the peels of *C. reticulata* and $CrTiO_2$-NP has a moderately low carotenoid concentration of 4.54±0.21 and 2.30±0.18

Table 4: Anti-inflammatory Activity of EEPCr and $CrTiO_2$-NP

		C. reticulata	$CrTiO_2$-NP	Control
1	Cyclooxygenase %	21.42±0.32	23.13±1.62	63.252 (Dicloflenac)
2	Lipo-oxygenase %	32.46±1.42	53.53±0.13	75.141 (Dicloflenac)
3	NO %	31.73±0.79	41.53±0.79	60.2321 (Dicloflenac)
4	Protein denaturation %	47.61±0.71	21.62±1.32	72.628 (Aspirin)
5	Anti-proteinase %	59.04±0.49	58.32±1.05	73.985 (Aspirin)
6	Heat-induced %	57.14±0.13	42.42±1.63	70.431 (Aspirin)

Anti-lipoxygenase Activity

The assay quantifies the $CrTiO_2$-NP and the peel extracts' inhibitory activity against LOX, the primary enzyme in the manufacture of mediators involved in inflammatory lipids, including leukotrienes, hepoxilins, lipoxins, and other hydroxylated fatty acid derivatives. The peel extract of *C. reticulate* showed low activity (32.46±1.42) as compared to $CrTiO_2$-NP (53.53±0.13) while the positive control employed Diclofenac (75.141) as shown in Table 4.3.

Cyclooxygenase Activity

The effects of $CrTiO_2$-NP and ethanol extract from the peels of *C. reticulate* on the production of prostaglandins were determined by the percentage inhibition of Cyclooxygenase activity. The results are tabulated in (Table 4.3). The extract showed moderate activity as compared to the positive control employed Diclofenac (63.252) though the synthesized nanoparticle (23.13±1.62) displayed a better activity than the extract (21.42±0.32)

Inhibition of protein denaturation

The $CrTiO_2$-NP and ethanol extract of the peel of *C. reticulate* were analyzed for their anti-inflammatory activity and compared with the standard Aspirin. The denaturation of proteins is one of the well-documented causes of inflammation. The results of the present study show that the extract (47.61±0.71) is moderately effective in inhibiting heat-induced albumin denaturation compared to the positive control (72.628) though fairly better than the $CrTiO_2$-NP (21.62±1.32)

Nitric Oxide Activity

The $CrTiO_2$-NP and ethanol extract of the peel of *C. reticulate* was assessed for percentage inhibitory activity against nitric oxide produced *in vitro*. Nitric oxide radical, generated by photochemical decomposition of sodium nitroprusside and eventually leading to stable nitrite ions, was found to be averagely inhibited by the extract (31.73±0.79) and good activity for the nanoparticle formed the peels of *C. reticulate* (41.53±0.79) as compared with the positive control as Dicloflenac (60.2321).

Heat-induced Haemolysis

The ethanol extract from the peels of *C. reticulata* and its synthesized nanoparticles were effective in inhibiting heat-induced haemolysis when compared with the standard Aspirin. The results from Table 4.3 displayed that the extract at a concentration of 100 µg/ml shows 57.14±0.13 while $CrTiO_2$-NP shows 42.42±1.63 as compared with the standard (70.431).

Anti-Proteinase Inhibitory Activity

Neutrophils are found in lysosomes and are recognized to be a rich source of serine proteinase. It has been previously documented that leukocyte proteinase contributes significantly to the development of tissue damage during inflammatory reactions, and proteinase inhibitors significantly reduce tissue damage. (Das & Chatterjee, 1995). The ethanol extract from the peels of *C. reticulata* and its synthesized nanoparticles exhibited significant anti-proteinase activity as shown in Table 4.3 (59.04±0.49 and 58.32±1.05). It showed a percentage inhibition of 48.15±0.38 as compared with the standard employed which is Aspirin (73.985).

LCMS

The result from the LCMS is shown in **Table 5**, nine compounds were revealed in the extracts and drawn in Figure 4.5.

Table 5: Compounds Identified from EEPCr

S/N	Identified compounds	Molecular weight
1	Neohesperidin	$C_{28}H_{36}O_{15}$
2	3-feruloylquinic acid	$C_{17}H_{20}O_9$
3	Hesperidin	$C_{28}H_{34}O_{15}$
4	Rhamnoside	$C_{21}H_{20}O_{11}$
5	Nobiletin	$C_{21}H_{22}O_8$
6	Tangeretin	$C_{20}H_{20}O_7$
8	Dihydroquercetin	$C_{15}H_{10}O_7$
9	4,5-dihydroxy-6,7-dimethoxyflavone; 7-methylcapillarisin	$C_{17}H_{14}O_6$

Discussion

UV-vis spectroscopy

To confirm the presence of nanoparticles in the resulting solutions, the UV-vis spectra

were analyzed. UV-vis absorption spectroscopy is an important technique to monitor the formation and stability of metal NPs in aqueous solution. The absorption spectrum of metal NPs is sensitive to several factors, including the shape, size of the nanoparticles formed and the particle-particle interaction with the medium and Sonnichesen et al. (2002) asserted that the absorption maximum (λmax) depends on the shape and size of the plants' nanoparticles. Figure 4.1 shows the UV-vis absorption spectra for $CrTiO_2$NPs nanoparticles between 200 and 400 nm. The absorption for $CrTiO_2$NPs appears at 380 nm in UV–Vis spectroscopy. Similar studies were conducted by Roopan et al. (2012). The optical absorbance of the TiO_2 nanoparticles was evaluated using the UV–visible absorption spectrum. Since all of the samples are extracted from the same solvent (ethanol), as can be seen in the UV-visible spectrum above, they all have the same pattern of absorbance (Zhao et al., 2007). According to Rathod and Waghuley's (2015) findings, which also indicated that % absorption is higher on the lower wavelength side, the optimum absorption in this study is higher on the lower wavelength side. The crystal structure of TiO2 determines its various properties. According to Amano et al. (2022), anatase TiO2 can only absorb UV radiation with wavelengths less than 390 nm due to its 3.2 eV band gap. Conversely, rutile TiO_2 can absorb visible light at wavelengths shorter than 410 nm and has a band gap of 3.0 eV (Alijani et al., 2022). One-dimensional TiO_2 nanotubes (TNTs) have a high active surface area and are a good choice for UV photodetectors. TiO_2 nanowire arrays have remarkable UV light sensitivity as well; at 7 μW/cm2 UV illumination, a high photoresponsivity of 7.7 × 103 A/W is attained. It was discovered that TiO_2 semiconducting thin films were transparent in the UV-visible range and that their refractive index varied with wavelength.

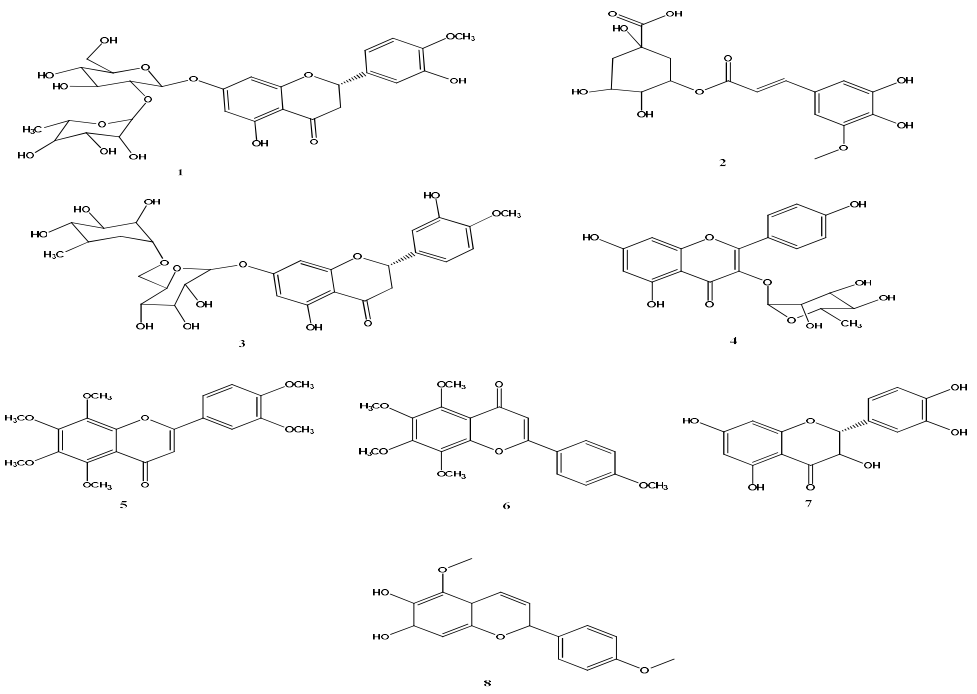

Figure 5: Structures of Compounds identified in the ethanol of *Citrus reticulata*

FTIR Spectroscopy

These functional organic groups' presence in the nanoparticle formed suggests that the organic source was in charge of reducing TiO_2 (Algahtani et al., 2020b; Algahtani et al., 2022; Anbumani et al., 2022; Ansari et al., 2022). 1449.41 cm^{-1} is the absorption peak of the C=C backbone stretching vibration of the

aromatic ring. The Ti–O-O bond is responsible for the peak located at 682 cm^{-1} in the TiO_2 NPs FTIR spectra. This study validated the ethanol extracts of the peels of C. reticulate 's capping action for the generated TiO2 NPs, which stabilized them (Thandapani et al., 2018; Thakur et al., 2019; Algahtani et al., 2020a). The absorption peaks around 3430 cm^{-1} for the characteristic absorption peaks of the joined hydroxyl group. The peaks of some intensity at 3043 cm^{-1} are the absorption peaks of C-H stretching vibrations on the benzene ring. Increasing aromatic ring condensation and higher aromatic ring substitution. FTIR spectrum of TiO_2 nanoparticles, Ti-O bending mode, and deformative vibration of Ti-OH stretching mode may be observed at 483 cm^{-1} and 1623.50 cm^{-1} respectively. There were noticeable peaks at 3430.53, 1779.04, 1639.76, 1449.41, 1162.46, and 682 cm^{-1} (Figure 4.2). The presence of alkyl halide, aliphatic nitro, aromatic amide, carbonyl amide, flavonoids, phenol, and secondary alcohol that may have contributed to the environmentally friendly synthesis of TiO_2 NPs was indicated by the detected peaks. Asymmetrical and symmetrical stretching vibrations of a hydroxyl group (-OH) may be observed at 3434.53 cm^{-1}.

SEM

The SEM image verified the formation of titanium nanoparticles and displayed the high-density titanium nanoparticles produced by the ethanol extracts of the peels of *C. reticulata*. Under a SEM, the majority of the nanoparticles aggregated while very few were dispersed. The produced titanium nanoparticles from the peels of *C. reticulata* were examined using a scanning electron microscope (SEM) to reveal their spherical shape and particle size, which ranged from 72 to 135 nm. The nanoparticles formed from the ethanolic extract of the peels of *C. reticulata* were homogeneously dispersed. This complements the studies of some authors on the green synthesis of TiO_2NPs employing various plant extracts (Narayanan *et al.,* 2021; Selvi *et al.,* 2022; Ravi *et al.,* 2023; Pradeep *et al.,* 2024). The Scanning Electron Microscopy result obtained in this study is in tandem with the obtained by Haidar et al. (2017) where TiO_2 nanoparticles calcined at 400°C have a roughly spherical spongy shape with small size nanoparticles around the range of less than 20 nm. When the temperature increases up to 600°C, the sizes become bigger and the agglomeration becomes significant, as shown in Figure 4.3. Further increase in temperature up to 800°C, and 1000°C, TiO_2 nanoparticles exhibited non-uniform particle shape due to the agglomeration of initial particles with the increase in crystalline size.

LCMS

Flavanones occur in higher amounts in the Citrus genus than flavones. Flavanones are contained more in the albedo and membranes than in the juice sacs. The main flavonoids in citrus fruits include hesperidin, naringin, and neohesperidin. Table 4.3 shows the presence of hesperetin, neohesperidin, tangeretin, and nobiletin in the peels of *Citrus reticulata*. Compounds identified in the ethanol extract were more than the ones identified in the TiO_2-NP

These compounds are renowned for having so many pharmacological activities

Neohesperidin, a flavonoid glycoside found in citrus fruits, exhibits diverse medicinal properties (Akhtar *et al.* 2022). It has been studied for its potential therapeutic effects against various conditions such as hypertension, obesity, and inflammatory diseases (Zhang *et al.,* 2022). Neohesperidin has shown promise in inhibiting angiotensin II-induced hypertension and vascular remodeling, acting as an antioxidant (Kumar *et al.,* 2023). Additionally, it has been explored for its role in treating bronchial asthma and diseases related to Th1/Th2 cell immune imbalance. Furthermore, neohesperidin has demonstrated beneficial effects in attenuating obesity, inflammation, and insulin resistance by restoring gut barrier damage and modulating intestinal microbiota composition. These findings highlight neohesperidin's potential as a therapeutic agent for a wide range of complex disorders, although further research is needed to confirm its safety and efficacy for clinical use (Lu *et al.,* 2020).

Nobiletin, a polymethoxyflavone found in citrus fruits, exhibits promising medicinal properties. Research highlights its role in inhibiting cancer cell proliferation through cell cycle arrest and mitochondrial-dependent mechanisms (Moazamiyanfar*et al.,* 2023). Moreover, nobiletin has shown potential as an effective cancer chemoprevention agent by targeting various cellular and molecular pathways, overcoming issues like toxic effects and multidrug resistance (Chen *et al.,* 2023).

Additionally, nobiletin's antioxidative, anti-inflammatory, and anti-viral properties make it a valuable phytochemical for various health conditions (Lin *et al.,* 2020). Its ability to induce cell death in malignant cells while reducing normal cell toxicity underscores its dual effect, making it a compelling candidate for enhancing tumor suppression with low toxicity (Arshad *et al.,* 2023). Overall, nobiletin's diverse mechanisms and low side effects position it as a significant natural compound with therapeutic potential in cancer treatment and beyond.

Tangeretin, a flavonoid compound found in citrus fruit peels, exhibits diverse pharmacological properties (Agarwal & Murti, 2022). It shows promise as a natural anticancer agent, synergizing with chemotherapeutic drugs and reversing cancer resistance. Tangeretin's antioxidant, anti-inflammatory, and anticancer effects make it a potential candidate for reducing chemotherapy-induced toxicity (Ahamad *et al.,* 2021). Additionally, tangeretin's hepatoprotective, neuroprotective, and antitumor activities highlight its therapeutic potential in various disorders. Studies suggest that combining tangeretin with cytotoxic agents like 5-Fluorouracil enhances chemotherapy effectiveness, particularly in colorectal cancer, by modulating miR-21 expression and PI3K/Akt signaling. Overall, tangeretin's multifaceted pharmacological profile positions it as a promising natural compound for medicinal applications, especially in cancer treatment and reducing chemotherapy-related side effects (Arafa *et al.,* 2021).

Dihydroquercetin (DHQ) is a bioflavonoid with diverse pharmacological activities, including antioxidant, anti-inflammatory, and capillary-protective properties. It has shown therapeutic potential in various conditions such as chronic venous insufficiency, chronic obstructive pulmonary disease, bronchial asthma, atherosclerosis, Alzheimer's disease, and even as a potential regulator of oxidative stress in COVID-19 therapy (Zhang *et al.,* 2022). Different dosage forms of DHQ have been developed, including tablets and gels for topical use, to enhance its bioavailability and efficacy (Orlova *et al.,* 2022). Studies have demonstrated the safety and efficacy of DHQ in mitigating neuroinflammation and related neurodegenerative disorders, making it a promising candidate for managing conditions like Alzheimer's and Parkinson's diseases.

Conclusion

The study investigated the green synthesis, characterization, and biological importance of titanium dioxide (TiO2) nanoparticles derived from the peels of *Citrus reticulata* (mandarin orange). The nanoparticles were synthesized using a simple, eco-friendly, and cost-effective method by exploiting the reducing and capping properties of the Citrus reticulata peel extract. The synthesized nanoparticles were characterized using various analytical techniques: UV-vis spectrophotometry, FTIR, and SEM analysis. FTIR showed the presence of functional groups from the peel extract, indicating their role in stabilizing the nanoparticles. SEM analysis revealed the spherical shape and size range of 72-135 nm for the nanoparticles. LCMS analysis identified several phytochemicals in the nanoparticles, including Neohesperidin, Hesperidin, Nobiletin, Tangeretin, and Dihydroquercetin, which are known to have good biological activities such as antibacterial, antioxidant, anticancer, anti-inflammatory, antidiabetic, and anti-Alzheimer's. The study evaluated the antioxidant and anti-inflammatory activities of the ethanol extract of *Citrus reticulata* peels (EEPCr) and the synthesized nanoparticles ($CrTiO_2NP$). The nanoparticles demonstrated significant anti-inflammatory and antioxidant properties, which were attributed to the phytochemicals present in the peel extract. The findings highlight the successful green synthesis of TiO_2 nanoparticles using *Citrus reticulata* peels, their physicochemical characteristics, and their promising biological applications, particularly in the fields of anti-inflammation and antioxidant therapies. This eco-friendly approach to nanoparticle synthesis holds great promise for the development of sustainable and biocompatible nanomaterials with diverse applications.

Reference

Agrawal, K. K., & Murti, Y. (2022). Tangeretin: a biologically potential citrus flavone. Current Traditional Medicine, 8(4), 31-41.

Ahmed FY. Comparative study of antibacterial effects of Titanium dioxide nanoparticles alone and in combination with antibiotics on MDR

Pseudomonas aeruginosa strains. Int J Nanomed. 2020;15:3393–404.

Akhtar S, Shahzad K, Mushtaq S, Ali I, Rafe MH, Fazal-ul-Karim SM. Antibacterial and antiviral potential of colloidal Titanium dioxide (TiO2) nanoparticles suitable for biological applications. Mater Res Express. 2019;6(10):105409.

Akhtar S. Antibacterial and antiviral potential of colloidal Titanium dioxide (TiO2) nanoparticles suitable for biological applications. Mater Res Exp. 2019;6(10):105409.

Akhter, S., Arman, M. S. I., Tayab, M. A., Islam, M. N., & Xiao, J. (2022). Recent advances in the biosynthesis, bioavailability, toxicology, pharmacology, and controlled release of citrus neohesperidin. Critical Reviews in Food Science and Nutrition, 1-20.

Algahtani, M.S.; Ahmad, M.Z.; Ahmad, J. (2020b) Nanoemulgel for Improved Topical Delivery of Retinyl Palmitate: Formulation Design and Stability Evaluation. *Nanomaterials* , *10*, 848.

Algahtani, M.S.; Ahmad, M.Z.; Ahmad, J. (2022) Investigation of Factors Influencing Formation of Nanoemulsion by Spontaneous Emulsification: Impact on Droplet Size, Polydispersity Index, and Stability. *Bioengineering 9*, 384.

Algahtani, M.S.; Ahmad, M.Z.; Nourein, I.H.; Ahmad, J. (2020a) Co-Delivery of Imiquimod and Curcumin by Nanoemugel for Improved Topical Delivery and Reduced Psoriasis-Like Skin Lesions. *Biomolecules* , *10*, 968

Alhamad, D. W., Elgendy, S. M., Al-Tel, T. H., & Omar, H. A. (2021). Tangeretin as an adjuvant and chemotherapeutic sensitizer against various types of cancers: a comparative overview. Journal of Pharmacy and Pharmacology, 73(5), 601-610.

Alijani, M., Wiltshire, B. D., Zarifi, M. H., & Macak, J. M. (2022, July). TiO2 Nanotube Integrated Microwave Resonator UV Sensor. In Electrochemical Society Meeting Abstracts 241 (No. 52, pp. 2167-2167). The Electrochemical Society, Inc..

Allahverdiyev AM. Investigation of antileishmanial activities of TiO2@Ag nanoparticles on biological properties of L. tropica and L. infantum parasites, in vitro.ExpParasitol. 2013 Sep;135(1):55–63.

Alvi, G.B., Iqbal, M.S., Ghaith, M.M.S. (2021). Biogenic selenium nanoparticles (SeNPs) from citrus fruit have anti-bacterial activities. *Sci Rep* **11**, 4811

Amano, F., Yamamoto, A., & Kumagai, J. (2022).Highly Active Rutile TiO2 for Photocatalysis under Violet Light Irradiation at 405 nm.Catalysts, 12(10), 1079.

Anbumani, D.; Dhandapani, K.v.; Manoharan, J.; Babujanarthanam, R.; Bashir, A.K.H.; Muthusamy, K.; Alfarhan, A.; Kanimozhi, K. (2022) Green synthesis and antimicrobial efficacy of titanium dioxide nanoparticles using Luffa acutangula leaf extract. *J. King Saud Univ. Sci. 34*, 101896. [

Ansari, A.; Siddiqui, V.U.; Rehman, W.U.; Akram, M.K.; Siddiqi, W.A.; Alosaimi, A.M.; Hussein, M.A.; Rafatullah, M. (2022) Green Synthesis of TiO_2 Nanoparticles Using Acorus calamus Leaf Extract and Evaluating Its Photocatalytic and In Vitro Antimicrobial Activity. *Catalysts 12*, 181

Arafa, E. S. A., Shurrab, N. T., & Buabeid, M. A. (2021).Therapeutic implications of a polymethoxylated flavone, tangeretin, in the management of cancer via modulation of different molecular pathways.Advances in Pharmacological and Pharmaceutical Sciences, 2021.

Aravind, M., Amalanathan, M. & Mary, M.S.M. Synthesis of TiO2 nanoparticles by chemical and green synthesis methods and their multifaceted properties. *SN Appl. Sci.* **3**, 409 (2021).

Arshad, R., Masood, S., Sharif, H. R., Goksen, G., Xu, B., & Faisal Manzoor, M. (2023).New Insights into Health-Promoting Effects of Nobiletin from Citrus Fruits. Food Reviews International, 1-20.

Aryal S, Bahadur KCR, Bhattard N, Kim CK, Kim HY (2006) Study of

electrolyte induced aggregation of gold nanoparticles capped by amino acids. *J. Colloid Interface Sci.* . 299:191–7.

Aslam M, Abdullah AZ, Rafatullah M.Recent development in the green synthesis of titanium dioxide nanoparticles using plant-based biomolecules for environmental and antimicrobial applications. J IndEng Chem. 2021;98:1–16.

Avila-Juarez L. Nanoparticles as potential antivirals in agriculture. Agriculture. 2020;10(10):444.

Ayoob, A.K. Gupta, V.T. Bhat, (2008). A conceptual overview on sustainable technologies for the defluoridation of drinking water, Crit. Rev. Environ. Sci. Technol. 38 (2008) 401–470.

BabakKakavandi, Ahmad Jonidi, Jafari, RoshanakRezaei, Kalantary, SiminNasseri, Ahmad Ameri, and Ali Esrafily 2013. Synthesis and properties of Fe3 O4 -activated carbon magnetic nanoparticles for removal of aniline from aqueous solution: equilibrium, kinetic and thermodynamic studies. Iranian Journal of Environmental Health Science & Engineering, 10-19.

Bartram, J. C. Brocklehurst, M.B. Fisher, R. Luyendijk, R. Hossain, T. Wardlaw, B. Gordon, (2014). Global monitoring of water supply and sanitation: history, methods and future challenges, Int. J. Environ. Res. Public Health 11 (8) 8137–8165.

Behbahani, M. M.R. AlaviMoghaddam, M. Arami, (2011). Techno-economical evaluation of fluoride removal by electrocoagulation process: optimization through response surface methodology, Desalination 271 209–218.

Behnam MA, Emami F, Sobhani Z, Dehghanian AR. The application of titanium dioxide (TiO2) nanoparticles in the photo-thermal therapy of melanoma cancer model. Iran J Basic Med Sci. 2018;21(11):1133.

Benov L. Effect of growth media on the MTT colorimetric assay in bacteria.PLoS One. 2019;14(8):e0219713.

Bindhu MR and Umadevi M. Synthesis of monodispersed silver nanoparticles using Hibiscus cannabinus leaf extract and its antimicrobial activity. SpectrochimicaActa Part A: Molecular and Biomolecular Spectroscopy. 2013; 101: 184-190.

BogumilaReidy, Andrea Haase, Andreas Luch, Kenneth, A. Dawson and Iseult Lynch 2013. Mechanisms of silver nanoparticle release, transformation and toxicity: a critical review of current knowledge and recommendations for future studies and applications materials. 6 : 2295-2350.

Bottero J-Y, Rose J and Wiesner MR. Nanotechnologies: tools for sustainability in a new wave of water treatment processes. Integrated Environmental Assessment and Management. 2006; 2: 391–395

Brame, J. Q. Li, P.J.J. Alvarez, (2011) Nanotechnology enabled water treatment and reuse: emerging opportunities and challenges for developing countries, Trends Food Sci. Technol. 22 618–624.

Chen Y, Wan Y, Wang Y, Zhang H, Jiao Z. Anticancer efficacy enhancement and attenuation of side effects of doxorubicin with titanium dioxide nanoparticles. Int J Nanomed. 2011;6:2321–6.

Chen, Y. Y., Liang, J. J., Wang, D. L., Chen, J. B., Cao, J. P., Wang, Y., & Sun, C. D. (2023). Nobiletin as a chemopreventive natural product against cancer, a comprehensive review. Critical Reviews in Food Science and Nutrition, 63(23), 6309-6329.

Choi H, Stathatos E and Dionysiou, DD. Sol-gel preparation of mesoporousphotocatalyticTiO2 films and TiO2/Al2O3 composite membranes for environmental applications. Applied Catalysis B: Environmental. 2006; 63: 60–67.

Das, R. Md.E. Ali, S.B.A. Hamid, S. Ramakrishna, Z.Z. Chowdhury, (2014). Carbon nanotube membranes for water purification: A bright future in water desalination, Desalination 336 97–109.

Devipriya D, Roopan SM (2017) Cissusquadrangularis mediated

ecofriendly synthesis of copper oxide nanoparticles and its antifungal studies against Aspergillus Niger, Aspergillusflavus. Mater SciEng C 80:38–44

Dhermendra K. Tiwari, Behari, J. and Prasenjit Sen. 2008. Application of Nanoparticles in Waste Water Treatment World Applied Sciences Journal. 3 (3) : 417-433.

Doan Thi, T. U., Nguyen, T. T., Thi, Y. D., Ta Thi, K. H., Phan, B. T., & Pham, K. N. (2020). *Green synthesis of ZnO nanoparticles using orange fruit peel extract for antibacterial activities. RSC Advances, 10(40), 23899–23907.*

Droste RL. Theory and practice of water and wastewater treatment. New York: Wiley (Book). 1997.

Elsharkawy MM, Derbalah A. Antiviral activity of titanium dioxide nanostructures as a control strategy for broad bean strain virus in faba bean. Pest Manag Sci. 2019;75(3):828–34.

Farhadian, B., Alizadeh, M., &Sepahi, M. (2022).A comprehensive review on the synthesis methods of TiO2 nanoparticles. Ceramics International, 48(11), 14598-14617.

Fawell, J. M.J. Nieuwhuijsen, (2003). Contaminants in drinking water, Br. Med. Bull.68 199–208.

Fouad GI. A proposed insight into the anti-viral potential of metallic nanoparticles against novel coronavirus disease-19 (COVID-19). Bull Natl Res Cent. 2021;45(1):1–22.

Gholami, S., Ramakrishna, S., &Ganesan, P. (2021). Synthesis of titanium dioxide nanoparticles and their biomedical applications: A review. Colloids and Surfaces B: Biointerfaces, 200, 111551.

Gupta S, Behari J and Kesari K. Low frequency ultrasonic treatment of sludge. Asian Journal of Water Environment and Pollution.2006; 3: 101-105.

Gutman RL, Peacock G, Lu DR. Targeted drug delivery for brain cancer treatment. J Controlled Rel. 2000;65(1–2):31–41.

Haider, A. J., AL– Anbari, R. H., Kadhim, G. R., & Salame, C. T. (2017). Exploring potential Environmental applications of TiO2 Nanoparticles. Energy Procedia, 119, 332–345.

Hamza, I.A. L. Jurzik, A. Stang, K. Sure, K. Uberla, M. Wilhelm, (2009). Detection of human viruses in rivers of a densely-populated area in Germany using a virus adsorption elution method optimized for PCR analyses, Water Res. 43 (10) 2657–2668.

Hasan, S. (2015). A review on nanoparticles: their synthesis and types. Res. J. Recent Sci, 2277, 2502.

HuifengQian, Lori A. Pretzer, Juan C. Velazquez, Zhun Zhao, Michael S. Won 2013. Gold nanoparticles for cleaning contaminated water. Journal of Chemical, 735-741.

Ichinose N, Ozaki Y and Kashu S. Superfine particle technology.Springer, London, (Book). 1992.

Irshad MA, Nawaz R, Rehman MZ, Adrees M, Rizwan M, Ali S, et al. Synthesis, characterization and advanced sustainable applications of titanium dioxide nanoparticles: a review. Ecotoxicol Environ Saf. 2021;212:111978.

Jaberzadeh A, Moaveni P, Moghadam HRT, Zahedi H. Influence of bulk and nanoparticles titanium foliar application on some agronomic traits, seed gluten and starch contents of wheat subjected to water deficit stress. Not BotanicaeHortiAgrobotanici Cluj-Napoca. 2013;41(1):201–7.

Jayaseelan C, Rahuman AA, Roopan SM, Kirthi AV, Venkatesan J, Kim S-K, et al. Biological approach to synthesize TiO2 nanoparticles using Aeromonashydrophila and its antibacterial activity. SpectrochimicaActa A MolBiomolSpectrosc. 2013;107:82–9.

Kim, B., Park, C.S., Murayama, M., Hochella, M.F., Jr. 2010. Discovery and characterization of silver sulfide nanoparticles in final sewage sludge products. Environ. Sci. Technol. 44(19) : 7509–7514

Kumar, D., Dey, Y. N., Pramanik, S. D., Singh, L. P., Selvaraja, M., & Rajagopal, M. (2023). Potential of Hesperidin in the Medicinal Field.

Current Bioactive Compounds, 19(5), 17-25.

Kurutas EB. The importance of antioxidants which play the role in cellular response against oxidative/nitrosative stress: current state. Nutr J. 2016;15:71.

Li, Y. H., Luan, Z. K., Zhao, Y. M., Hu, W. B., Ahmad, I., Zhu,Y. Q. , Peng, X.J. 2007. Carbon nanotubes – the promising adsorbent in wastewater. Treatment Journal of Physics: Conference Series. 61 : 698-702

Lin, C. X., Tu, C. W., Ma, Y. K., Ye, P. C., Shao, X., Yang, Z. A., & Fang, Y. M. (2020). Nobiletin inhibits cell growth through restraining aerobic glycolysis via PKA-CREB pathway in oral squamous cell carcinoma. Food Science & Nutrition, 8(7), 3515-3524.

Liu E, Zhou Y, Liu Z, Li J, Zhang D, Chen J, et al. Cisplatin loaded hyaluronic acid modified TiO2 nanoparticles for neoadjuvant chemotherapy of ovarian cancer. J Nanomaterials. 2015;2015:390358.

Lu, J. F., Zhu, M. Q., Zhang, H., Liu, H., Xia, B., Wang, Y. L., ...& Wu, J. W. (2020). Neohesperidin attenuates obesity by altering the composition of the gut microbiota in high-fat diet-fed mice. The FASEB Journal, 34(9), 12053-12071.

Lunge SS, Singh S and Sinha A. International Conference on Chemical, Civil and Environmental Engineering (CCEE'2014), Magnetic Nanoparticle: Synthesis and Environmental Applications, Singapore. 2014; 18-20.

Maheswari P. Bio-modified TiO2 nanoparticles with Withaniasomnifera, Ecliptaprostrata and Glycyrrhizaglabra for anticancer and antibacterial applications. Mater SciEng C Mater Biol Appl. 2020 Mar;108:110457.

Mahmoodzadeh H, Aghili R. Effect on germination and early growth characteristics in wheat plants (Triticumaestivum L.) seeds exposed to TiO2 nanoparticles. J Chem Health Risks. 2014;4(1):29–36.

Manoj A. Lazar, Shaji Varghese and Santhosh S. Nair 2012.Photocatalytic Water Treatment by Titanium Dioxide: Recent Updates Catalysts, 2 : 572-601.

Mazurkova NA, Spitsyna YE, Shikina NV, Ismagilov ZR, Zagrebel'nyi SN, Ryabchikova EI.Interaction of titanium dioxide nanoparticles with influenza virus.Nanotechnol Russia. 2010;5(5):417–20.

Michael A. Perez 2012. The Effects of Silver Nanoparticles on Wastewater Treatment and Escherichia Coli Growth. Honors These Paper 68

Mishra B., Bhavesh B., Patel B. B., TIwari (2010).,Nanomedicine: Nanotechnology, Biology, and Medicine 2010, 6: 9 -24

Moazamiyanfar, R., Rezaei, S., AliAshrafzadeh, H., Rastegar-Pouyani, N., Jafarzadeh, E., Mouludi, K., ...& Najafi, M. (2023). Nobiletin in cancer therapy; mechanisms and therapy perspectives.Current pharmaceutical design, 29(22), 1713-1728.

Murthy, S.K. (2007). Nanoparticles in modern medicine: state of the art and future challenges, *Int. J. Nanomed.* 2.129–141

Murugan K, Dinesh D, Kavithaa K, Paulpandi M, Ponraj T, Alsalhi MS, et al. Hydrothermal synthesis of titanium dioxide nanoparticles: mosquitocidal potential and anticancer activity on human breast cancer cells (MCF-7). Parasitology Res. 2016;115(3):1085–96.

Nabi, G., Raza, W., Tahir, M., 2019.Green synthesis of TiO2 nanoparticle using cinnamon powder extract and the study of optical properties. J. Inorg. Organomet.Polym.Mater.1–5.

Nakano R, Ishiguro H, Yao Y, Kajioka J, Fujishima A, Sunada K, et al. Photocatalytic inactivation of influenza virus by titanium dioxide thin film. PhotochemPhotobiol Sci. 2012;11(8):1293–8.

Narayanan, M.; Devi, P.G.; Natarajan, D.; Kandasamy, S.; Devarayan, K.; Alsehli, M.; Elfasakhany, A.; Pugazhendhi, (2021) A. Green synthesis and characterization of titanium dioxide nanoparticles using leaf extract of *Pouteria campechiana* and larvicidal and pupicidal activity on Aedes aegypti. *Environ. Res.*, *200*, 111333.

Nikam, A. P., Ratnaparkhiand, M. P., &Chaudhari, S. P. (2014).Nanoparticles–an overview. Int. J. Res. Dev. Pharm. Life Sci, 3, 1121-1127.

Nowack B. Pollution prevention n treatment using Nanotechnology.In Environmental Aspects. 2008

Obare SO and Meyer GJ.Nanostructured materials for environmental remediation of organic contaminants in water. Journal of Environmental Science and Health Part A. 2004; 39: 2549-2582.

Orlova, S. V., Tatarinov, V. V., Nikitina, E. A., Sheremeta, A. V., Ivlev, V. A., Vasil'ev, V. G., ... & Goryainov, S. V. (2022). Bioavailability and safety of dihydroquercetin. Pharmaceutical Chemistry Journal, 55(11), 1133-1137.

OussamaOuerghi, Mohammed H. Geesi, YassineRiadi&Elmutasim O. Ibnouf (2022) Limon-citrus extract as a capping/reducing agent for the synthesis of titanium dioxide nanoparticles: characterization and antibacterial activity, Green Chemistry Letters and Reviews, 15:3, 483-490,

Ovais M, Khalil AT, Ayaz M, Ahmad I, Nethi SK, Mukherjee S. Biosynthesis of metal nanoparticles via microbial enzymes: a mechanistic approach. Int J Mol Sci. 2018 Dec;19(12):4100.

Owolade O, Ogunleti D. Effects of titanium dioxide on the diseases, development and yield of edible cowpea. J Plant Prot Res. 2008;48(3):329–35.

Parthasarathi, V. and Thilagavathi, G. 2009.Synthesis and characterization of titanium dioxide nanoparticle and their applications to textiles for microbe resistance. Journal of Textile, Apparel Technology and Management, 2(2) : 1-8.

Parveen, K., Banse, V., &Ledwani, L. (2016). *Green synthesis of nanoparticles: Their advantages and disadvantages.*

Peng, T., Zhao, D., Dai, K., Shi, W., &Hirao, K. (2005). *Synthesis of Titanium Dioxide Nanoparticles with MesoporousAnatase Wall and High Photocatalytic Activity. The Journal of Physical Chemistry B, 109(11), 4947–4952.*

Petrusevski, B. S. Sharma, W.G. van der Meer, F. Kruis, M. Khan, M. Barua, J.C. Schippers, (2008). Four years of development and field-testing of IHE arsenic removal family filter in rural Bangladesh, Water Sci. Technol. 58 (1) 53–58.

Pinkerton KE. Health effects of inhaled engineered and incidental nanoparticles. Crit Rev Toxicol. 2009;39(8):629–58.

Prachi, PranjaliGautam, DeepaMadathil and A.N. Brijesh Nair 2010.Nanotechnology in waste water treatment. A Review International Journal of Chem Tech Research 5(5) : 2303-2308

Pradeep C. Dathan, Deepak Nallaswamy, S. Rajeshkumar, Suja Joseph, Shahin Ismail,Tharani, Naziya Rasheed (2024). *In vitro* Biological Activity of (Anti-inflammatory, Anti-oxidant) Pomegranate Peel Extract Mediated Titanium Dioxide. Trends Biomater. Artif. Organs, 38(1), 23-28.

Pradeep, T. and Anshup 2009. Noble metal nanoparticles for water purication: A critical review. Thin Solid Films, 517 : 6441-6478.

Qiao, J. Cui, Z. Y. Sun, Q. Hu, X. Guan, (2014). Simultaneous removal of arsenate and fluoride from water by Al-Fe hydr(oxides), Front. Environ. Sci. Eng. 8 (2) 169–179.

Rathod, P. B., & Waghuley, S. A. (2015).Synthesis and UV-Vis spectroscopic study of TiO2 nanoparticles.International Journal of Nanomanufacturing, 11(3/4), 185.

Ravi Saini, Pradeep Kumar (2023). Green synthesis of TiO2 nanoparticles using Tinospora cordifolia plant extract & its potential application for photocatalysis and antibacterial activity, Inorganic Chemistry Communications 156, 111221,

Ren W, Zeng L, Shen Z, Xiang L, Gong A, Zhang J, et al. Enhanced doxorubicin transport to multidrug resistant breast cancer cells via TiO2nanocarriers. RSC Adv. 2013;3(43):20855–61.

Ripolles-Avila C, Martinez-Garcia M, Hascoët AS, Rodríguez-Jerez JJ. Bactericidal efficacy of UV activated TiO2 nanoparticles against Gram-positive

and Gram-negative bacteria on suspension. CyTA J Food. 17(1):408–18.

Roopan S M, Bharathi A, Prabhakaran A, Rahuman AA, Velayutham K, Rajakumar G, Padmaja RD, Leksami M, Madhumitha G. Efficient phyto-synthesis and structural characterization of rutile TiO_2 nanoparticles using Annona squamosa peel extract. *Spectrochim Acta Part A* . 98:86–90

S.V. Jadhav, E. Bringas, G.D. Yadav, V.K. Rathod, I. Ortiz, K.V. Marathe, (2015). Arsenic and fluoride contaminated groundwaters: A review of current technologies for contaminants removal, J. Environ. Manage. 162 306–325.

Saha, S. K., Islam, M. S., &Hossain, M. A. (2021). Synthesis of titanium dioxide nanoparticles: A review. Materials Today: Proceedings, 42, 329-334.

Santhoshkumar, T., Rahuman, A. A., Jayaseelan, C., Rajakumar, G., Marimuthu, S., Kirthi, A. V., … Kim, S.-K. (2014). Green synthesis of titanium dioxide nanoparticles using Psidiumguajava extract and its antibacterial and antioxidant properties. Asian Pacific Journal of Tropical Medicine, 7(12), 968–976.

Santos, C.S., Gabriel, B., Blanchy, M., Menes, O., García, D., Blanco, M., Neto, V., 2015.Industrial applications of nanoparticles–a prospective overview.Mater. Today Proc. 2 (1), 456–465.

Savage N and Diallo MS. Nanomaterials and water purification: opportunities and challenge. Journal of Nanoparticle Research. 2005;7: 331-342.

Selvi, Jayaram Mari; Murugalakshmi, Mariappan; Sami, Ponnusamy; Gnanaprakash, Mariappan; Thanalakshmi, R. (2022). Green synthesis, characterization and applications of TiO_2 nanoparticles using aqueous extract of *Erythrina variegata* leaves. Current Science, 123 (1):59

Shah M, Fawcett D, Sharma S, Tripathy SK, Poinern GE. Green synthesis of metallic nanoparticles via biological entities.Mater (Basel). 2015 Nov;8(11):7278–308.

Singh P, Kim Y-J, Zhang D, Yang D-C. (2016) Biological synthesis of nanoparticles from plants and microorganisms.Trends Biotechnol.;34:588–99.

Singh, S., Maurya, I. C., Tiwari, A., Srivastava, P., &Bahadur, L. (2022). Green synthesis of TiO2 nanoparticles using Citrus limon juice extract as a bio-capping agent for enhanced performance of dye-sensitized solar cells. *Surfaces and Interfaces*, *28*, 101652.

Sönnichsen C, Franzl T, Wilk T, von Plessen G, Feldmann J, Wilson O (2002) Mulvaney Drastic reduction of plasmon damping in gold nanorods. *P. Phys. Rev. Lett.* .88: 077402/1

Stucchi M, Bianchi CL, Argirusis C, Pifferi V, Neppolian B, Cerrato G, Boffito DC (2018) Ultrasound assisted synthesis of Ag-decorated TiO2 active in visible light. UltrasonSonochem 40:282–288

Sujitha, M. V., &Kannan, S. (2013). Green synthesis of gold nanoparticles using Citrus fruits (Citrus limon, Citrus reticulata and Citrus sinensis) aqueous extract and its characterization. SpectrochimicaActa Part A: Molecular and Biomolecular Spectroscopy, 102, 15–23

Syngouna VI, Chrysikopoulos CV. Inactivation of MS2 bacteriophage by titanium dioxide nanoparticles in the presence of quartz sand with and without ambient light. J Colloid Interface Sci. 2017;497:117–25.

Thakur, B.K.; Kumar, A.; Kumar, D. (2019**)** Green synthesis of titanium dioxide nanoparticles using Azadirachta indica leaf extract and evaluating their antibacterial activity. *S. Afr. J. Bot.* , *124*, 223–227.

Thandapani, K.; Kathiravan, M.; Namasivayam, E.; Padiksan, I.A.; Natesan, G.; Tiwari, M.; Giovanni, B.; Perumal, V. (2018**).** Enhanced larvicidal, antibacterial, and photocatalytic efficacy of TiO(2) nanohybrids green synthesized using the aqueous leaf extract of Parthenium

hysterophorus. *Environ. Sci. Pollut. Res. Int.* , *25*, 10328–10339.

Tiwari D.K, Behari J and Sen P. Application of Nanoparticles in Waste Water Treatment.World Applied Sciences Journal.2008; 3: 417-433.

United Nations, Transforming our world (2015): the 2030 Agenda for Sustainable Development. United Nations, A/RES/70/1.

US Bureau of Reclamation and Sandia National Laboratories.Desalination and water purification technology roadmap a report of the executive committee Water Purification. 2003.

US Environmental Protection Agency.Alternative disinfectants and oxidants guidance manual.EPA Office of Water Report.1999; 815: 99-114.

US Environmental Protection Agency.Microbial and disinfection by-product rules.Federal Register.1998; 63: 69389-69476.

Venkatasubbu GD, Ramasamy S, Reddy GP, Kumar J. In vitro and in vivo anticancer activity of surface modified paclitaxel attached hydroxyapatite and titanium dioxide nanoparticles. Biomed Microdevices. 2013;15(4):711–26.

Walmsley GG, McArdle A, Tevlin R et al. Nanotechnology in bone tissue engineering Nanomedicine. 2015; 11: 1253-1263.

Wang L. The antimicrobial activity of nanoparticles: present situation and prospects for the future. Int J Nanomed. 2017;12:1227–49.

Wang, L.K. D.A. Vaccari, Y. Li, N.K. Shammas, (2006). Chemical precipitation (in) handbook of environmental engineering, Physicochemical Treatment Processes, Humana Press, NJ, pp. 141–197.

WHO. Progress on Drinking Water and Sanitation. 2012.

WHO/UNICEF, Progress on Drinking-water and Sanitation 2015 Update.JointMonitoring Programme: World Health Organization and United Nations ChildrenEducation Fund, 2015, 1, 1.

Wiltshire, B. D., Alijani, M., Mohammadi, S., Hosseini, A., Macak, J. M., & Zarifi, M. H. (2022).High-frequency TiO2 nanotube-adapted microwave coplanar waveguide resonator for high-sensitivity ultraviolet detection. ACS Applied Materials & Interfaces, 14(4), 6203-6211.

World Health Organization (WHO) (2011). Guidelines for drinking water quality, fourth ed., IWA Publishing, London, 2011

Yang F, Hong F, You W, Liu C, Gao F, Wu C, et al. Influence of nano-anataseTiO2 on the nitrogen metabolism of growing spinach.Biol Trace Elem Res. 2006;110(2):179–90.

Zahir AA, Chauhan IS, Bagavan A, Kamaraj C, Elango G, Shankar J, et al. Green synthesis of silver and titanium dioxide nanoparticles using Euphorbia prostrata extract shows shift from apoptosis to G0/G1 arrest followed by necrotic cell death in Leishmaniadonovani. Antimicrobial Agents Chemotherapy. 2015;59(8):4782–99.

Zayed, M., Ghazal, H., Othman, H.A. et al. Synthesis of different nanometals using Citrus Sinensis peel (orange peel) waste extraction for valuable functionalization of cotton fabric. Chem. Pap. 76, 639–660

Zhang, J., Chen, K., Ding, C., Sun, S., Zheng, Y., Ding, Q., ...& Liu, W. (2022). Fabrication of chitosan/PVP/dihydroquercetin nanocomposite film for in vitro and in vivo evaluation of wound healing. International Journal of Biological Macromolecules, 206, 591-604.

Zhang, J., Hui, Y., Liu, F., Yang, Q., Lu, Y., Chang, Y., ...& Ding, Y. (2022). Neohesperidin protects angiotensin II-induced hypertension and vascular remodeling. Frontiers in Pharmacology, 13, 890202.

Zhao, Y., Li, C., Liu, X., Gu, F., Jiang, H., Shao, W., He, Y. (2007). Synthesis and optical properties of TiO2 nanoparticles. Materials Letters, 61(1), 79–83