

Green Synthesis and Characterization of Titanium Containing Particles Using Leaf Extract of *Indigofera argentea*

vusqua qamar

vusqaqamar@gmail.com

Islamia University of Bahawalpur <https://orcid.org/0009-0009-1787-3151>

Research Article

Keywords: green synthesis, titanium-containing particles, *Indigofera argentea*, XRD, FTIR, SEM, EDX, UV-Vis

Posted Date: October 6th, 2026

DOI: <https://doi.org/10.21203/rs.3.rs-11258999/v1>

License: © ⓘ This work is licensed under a Creative Commons Attribution 4.0 International License.

[Read Full License](#)

Additional Declarations: The authors declare no competing interests.

Abstract

A nominal 0.5 M titanium tetrachloride (TiCl_4) precursor was reacted with an aqueous leaf extract of *Indigofera argentea* and the resulting powder was characterized by scanning electron microscopy (SEM), energy-dispersive X-ray spectroscopy (EDX), X-ray diffraction (XRD), UV–Vis spectroscopy and Fourier-transform infrared spectroscopy (FTIR). SEM showed round-to-oval particle-like objects occurring in branched clusters, with an image-based mean diameter of 0.247 μm ($n = 454$). EDX gave 75.01 wt% Ti, 19.61 wt% O and 5.38 wt% C, corresponding to an O/Ti atomic ratio of approximately 1.07. XRD showed four principal reflections at 28.94°, 35.55°, 41.74° and 61.79°; apparent Scherrer crystallite sizes were 41–70 nm without instrumental-broadening correction. The observed diffraction pattern did not match conventional anatase or rutile TiO_2 . UV–Vis absorbance increased sharply toward the lower measurement limit, while absorbance above 350 nm remained low; no band gap was assigned because the absorption edge was not adequately captured. FTIR showed hydroxyl and organic functional-group bands and a band near 669 cm^{-1} in the Ti–O region. Taken together, the characterization confirms a titanium-containing, submicron particulate product with nanocrystalline domains, but the available data are insufficient to identify its crystalline phase as TiO_2 or another specific titanium compound.

1. Introduction

Titanium-containing nanomaterials, particularly TiO_2 , have attracted interest in environmental, agricultural and materials applications because of their optical, surface and catalytic properties. Plant-mediated synthesis has been investigated as an alternative to conventional physical and chemical routes, with plant extracts serving as reaction media that can provide organic compounds capable of interacting with metal precursors and the resulting particle surfaces (Odiachi et al., 2023). Several studies have reported plant-mediated preparation of TiO_2 using leaf extracts, including *Erythrina variegata* and *Impatiens rothii* (Selvi et al., 2021; Yitagesu et al., 2023).

An important limitation of green-synthesis studies is that the use of a plant extract does not by itself establish the formation of a particular oxide phase. Phase assignment requires agreement among complementary characterization methods. In particular, morphology, elemental composition, diffraction, optical absorption and surface functional groups can provide different and sometimes non-equivalent information. This is especially important for titanium because EDX establishes elemental composition but does not determine titanium oxidation state or crystalline phase. Therefore, the product obtained from a plant-mediated titanium precursor reaction should not be labelled TiO_2 solely from its morphology or the presence of Ti and O.

Indigofera argentea is a plant species for which, to the best of our knowledge, a published report specifically describing plant-mediated preparation of titanium-containing particles using its leaf extract was not identified during preparation of this manuscript. The present study therefore investigated the product obtained by reacting a nominal 0.5 M TiCl_4 precursor with an aqueous *I. argentea* leaf extract

using SEM, EDX, XRD, UV–Vis and FTIR. The primary objective was to determine whether the resulting material could be assigned to TiO_2 on the basis of the available characterization data.

2. Materials and Methods

2.1 Leaf extract

Indigofera argentea leaves were collected locally, rinsed with distilled water and air-dried for 2–3 days. The dried leaves were ground to a fine powder in a mortar. Fifty grams of the powder were boiled in 200 mL of distilled water to prepare the aqueous extract. The available experimental record does not specify the boiling duration, extraction temperature, filtration procedure, or final extract volume; these details should therefore not be inferred.

2.2 Titanium precursor solution

A nominal 0.5 M TiCl_4 precursor was prepared using the reported 30 mL precursor volume. The calculation was: $0.5 \text{ mol L}^{-1} \times 0.030 \text{ L} = 0.015 \text{ mol TiCl}_4$. Using a molar mass of $189.68 \text{ g mol}^{-1}$ gives approximately 2.85 g TiCl_4 . Because TiCl_4 undergoes rapid hydrolysis in water, the stated concentration should be understood as the nominal precursor loading rather than evidence of an indefinitely stable aqueous TiCl_4 solution. The available record does not specify the addition sequence or rate or the final pH.

2.3 Synthesis

The aqueous leaf extract was mixed with the nominal 0.5 M TiCl_4 precursor and held at 60°C for 2 h under constant stirring. The mixture changed from light yellow to white. The exact volume of leaf extract used in the reaction and therefore the extract-to-precursor ratio were not recorded in the available manuscript. The resulting solid was collected by centrifugation at 10,000 rpm for 15 min and dried at 80°C for 24 h. Washing steps, if any, were not documented in the available record.

2.4 Characterization

- SEM and EDX: SEM imaging was performed using a ZEISS microscope with an SE2 detector at 21 kV, with working distances of 9.2–9.3 mm and magnifications of 2000 $\times$ and 5000 $\times$ according to the data bars. Particle-like objects were measured by automated image analysis using OpenCV/scikit-image with watershed segmentation and calibration against the embedded 1 μm scale bars. EDX was acquired on the same instrument using Zeiss SmartEDX on 6 May 2025. Sample mounting/coating details and the number of EDX acquisition areas were not recorded.
- XRD: diffraction data were collected from $2\theta = 20\text{--}80^\circ$ with a step size of 0.02° using Cu K α radiation; $\lambda = 1.5406 \text{ \AA}$ was used for the Scherrer calculation. Peak positions were fitted with Gaussian functions. Instrumental broadening was not corrected. The instrument model, scan rate and slit configuration were not recorded.
- UV–Vis: the product was scanned from 200 to 800 nm.

- FTIR: the spectrum was recorded from 4000 to 650 cm^{-1} .

SEM and EDX analyses were performed at the University of Engineering and Technology (UET), Lahore. For all characterization methods, details not available in the experimental record are reported as limitations rather than inferred.

3. Results and Discussion

3.1 SEM and particle size

The particles were round to oval and occurred in branched clusters (Fig. 1). A total of 454 particle-like objects from two SEM fields were measured. Calibration against the embedded 1 μm scale bars gave an image-based mean diameter of 0.247 μm (SD 0.069 μm ; median 0.234 μm), with 80% of measured objects between 0.17 and 0.34 μm (Table 1; Fig. 2). None of the measured objects was below 0.1 μm , so the SEM-observed objects are more appropriately described as submicron particles or aggregates rather than nanoparticles in the strict morphological sense. Their smaller XRD-derived crystallite dimensions are not equivalent to the SEM particle size. Because the analysis was based on only two SEM fields and the objects occurred in aggregates, the distribution should be regarded as an image-based estimate rather than a batch-level particle-size measurement.

Table 1
Particle diameter from image analysis.

Image	n	Mean (μm)	SD (μm)
SEM 1 (2000 $\times$)	212	0.257	0.076
SEM 2 (5000 $\times$)	242	0.237	0.061
Both	454	0.247	0.069

3.2 EDX

Table 2 shows the EDX quantification reported by the instrument. Titanium was 75.01 wt% (40.67 at%), oxygen was 19.61 wt% (43.45 at%), and carbon was 5.38 wt% (15.88 at%). The resulting O/Ti atomic ratio is approximately 1.07, substantially below the stoichiometric ratio of 2 expected for TiO_2 . The elemental ratio is therefore inconsistent with assigning the bulk of the detected Ti and O to stoichiometric TiO_2 alone. However, EDX is semi-quantitative and is particularly limited for light elements; moreover, the measurement does not determine titanium oxidation state or crystalline phase. The carbon signal may include plant-derived residue and/or contributions from sample mounting or coating. The carbon map showed faint, relatively even signal without a clear correspondence to particle outlines (Figs. 3 and 4). Consequently, the EDX data should be treated as evidence for a titanium-containing product rather than as proof of TiO , TiO_2 , TiC or another specific phase.

Table 2
EDX quantification (Zeiss SmartEDX report, 6 May 2025).

Element	Weight %	Atomic %	Net intensity	K ratio
C	5.38	15.88	115.59	0.0115
O	19.61	43.45	1423.32	0.0930
Ti	75.01	40.67	755.44	0.6778

3.3 XRD

The XRD pattern contained four clear reflections at 28.94°, 35.55°, 41.74° and 61.79° (Table 3; Fig. 5). Gaussian fitting gave FWHM values of 0.117–0.203°, and apparent Scherrer crystallite sizes of approximately 41–70 nm. Because instrumental broadening was not corrected and the phase identity is unresolved, these values should be interpreted only as approximate apparent crystallite dimensions. The observed reflections do not correspond to the principal diffraction pattern expected for anatase TiO₂, whose strongest reflection is near 25.3°, nor do they provide a satisfactory match to rutile TiO₂. Several of the higher-angle reflections show partial similarity to reflections reported for cubic titanium-containing phases such as TiO- or TiC-type structures. However, these similarities are not sufficient for phase identification. In particular, TiC formation would be chemically unexpected under the reported low-temperature aqueous synthesis conditions, and the carbon detected by EDX may have originated partly from residual plant material or sample mounting/coating. The XRD and EDX results therefore support an unresolved titanium-containing phase rather than a specific assignment to TiO, TiC or TiO₂. Phase-specific techniques such as Raman spectroscopy and X-ray photoelectron spectroscopy (XPS), together with improved reference-pattern matching and, where possible, transmission electron microscopy selected-area electron diffraction, would be needed for definitive identification.

Table 3
XRD peak positions and crystallite size (Gaussian fits, no instrument correction).

2θ (°)	FWHM (°)	d (Å)	Crystallite size (nm)
28.94	0.117	3.08	70
35.55	0.203	2.52	41
41.74	0.186	2.16	46
61.79	0.132	1.50	70

3.4 UV-Vis

Absorbance increased sharply toward the lower end of the measured range, reaching 2.41 at 200 nm, where the scan ended (Fig. 6). A small shoulder was visible around 270–280 nm, whereas absorbance

above 350 nm remained below 0.07. Because the absorption edge was not adequately captured within the measurement range, a Tauc-derived value of approximately 4.1–4.2 eV was not treated as a reliable band-gap estimate. No band gap is therefore reported. The weak absorbance above 350 nm also does not provide support for assigning the product to conventional TiO₂ on its own.

3.5 FTIR

The FTIR spectrum showed a broad band near 3405 cm⁻¹ attributable to O–H stretching from adsorbed water and/or hydroxyl groups. Other prominent bands occurred near 1560, 1390, 1333, 1110, 1020 and 929 cm⁻¹ (Table 4; Fig. 7). These bands are consistent with organic functional groups that may arise from residual extract-derived compounds associated with the product surface. A band near 669 cm⁻¹ lies in the Ti–O vibrational region, but because the scan began at 650 cm⁻¹ only the upper part of this feature was captured. The FTIR spectrum therefore provides evidence of surface hydroxyl/organic groups and a low-frequency metal–oxygen-region band, but it does not establish a specific titanium oxide phase.

Table 4
FTIR bands and likely assignments.

Band (cm ⁻¹)	Likely assignment
3405	O–H stretching (water, hydroxyl)
1560	C = O / carboxylate (asymmetric)
1390, 1333	C–H bending / carboxylate (symmetric)
1110, 1020	C–O stretching
929	C–H or C–O bending
669	Ti–O region (edge of scan)

4. Conclusion

Reaction of a nominal 0.5 M TiCl₄ precursor with *Indigofera argentea* leaf extract produced a titanium-containing powder composed of submicron particle-like objects occurring in clusters. SEM gave an image-based mean diameter of 0.247 μm, while XRD yielded apparent Scherrer crystallite sizes of approximately 41–70 nm. EDX showed an O/Ti atomic ratio of approximately 1.07, which is inconsistent with stoichiometric TiO₂, and the observed XRD reflections did not match conventional anatase or rutile TiO₂. FTIR indicated residual organic functional groups and a band in the Ti–O region, while UV–Vis measurements did not provide a reliable band-gap estimate. Collectively, these data support the formation of a titanium-containing particulate product but do not justify identifying it as TiO₂ nanoparticles or assigning it to another specific crystalline phase. Further phase-specific characterization, particularly Raman spectroscopy and XPS, is required for definitive identification. The incomplete experimental record concerning the exact extract volume, extraction conditions, washing

procedure and some instrument parameters is also a limitation for full reproducibility and should be resolved from the laboratory record before journal submission.

Declarations

Acknowledgments

The author thanks Dr. Ghulam Sarwar, Department of Botany, The Islamia University of Bahawalpur, for supervision and guidance; the University of Engineering and Technology (UET), Lahore, for SEM and EDX analyses; and the Department of Botany, The Islamia University of Bahawalpur, for institutional support.

References

1. Abdel Latef AAH, Srivastava AK, El-sadek MSA, Kordrostami M, Tran L-SP (2018) Titanium dioxide nanoparticles improve growth and enhance tolerance of broad bean plants under saline soil conditions. *Land Degrad Dev* 29(4):1065–1073. <https://doi.org/10.1002/ldr.2780>
2. Alabdallah NM, Alluqmani SM, Almarri HM, Al-Zahrani AA (2024) Physical, chemical, and biological routes of synthetic titanium dioxide nanoparticles and their crucial role in temperature stress tolerance in plants. *Heliyon* 10(4):e26537. <https://doi.org/10.1016/j.heliyon.2024.e26537>
3. Azameti MK, Imoro A-WM (2023) Nanotechnology: A promising field in enhancing abiotic stress tolerance in plants. *Crop Des* 2(2):100037. <https://doi.org/10.1016/j.cropl.2023.100037>
4. Badshah I, Mustafa N, Khan R, Mashwani Z-R, Raja NI, Almutairi MH, Aleya L, Sayed AA, Zaman S, Sawati L, Sohail S (2023) Biogenic titanium dioxide nanoparticles ameliorate the effect of salinity stress in wheat crop. *Agronomy* 13(2):352. <https://doi.org/10.3390/agronomy13020352>
5. Coates J (2000) Interpretation of infrared spectra, a practical approach. In: Meyers RA (ed) *Encyclopedia of Analytical Chemistry*. Wiley. <https://doi.org/10.1002/9780470027318.a5606>
6. Dağhan H (2018) Effects of TiO₂ nanoparticles on maize (*Zea mays* L.) growth, chlorophyll content and nutrient uptake. *Appl Ecol Environ Res* 16(5):6873–6883. https://doi.org/10.15666/aeer/1605_68736883
7. Odiachi IJ, Ize-Iyamu OC, Ize-Iyamu OK, Ikechukwu CD, Ifijen IH (2023) Systematic review of the synthesis of titanium oxide nanoparticles via plant mediation green approach. In TMS 2023 152nd Annual Meeting & Exhibition Supplemental Proceedings (pp. 548–558). Springer. https://doi.org/10.1007/978-3-031-22524-6_49
8. Patterson AL (1939) The Scherrer formula for X-ray particle size determination. *Phys Rev* 56(10):978–982. <https://doi.org/10.1103/PhysRev.56.978>
9. Selvi JM, Murugalakshmi M, Sami P (2021) Synthesis and characterization of nano-TiO₂ using aqueous extract of *Erythrina variegata* leaves. *Asian J Chem* 33(12):3139–3144. <https://doi.org/10.14233/ajchem.2021.23535>

10. Yitagesu GB, Leku DT, Workneh GA (2023) Green synthesis of TiO_2 using *Impatiens rothii* Hook.f. leaf extract for efficient removal of methylene blue dye. ACS Omega 8(46):43999–44012.
<https://doi.org/10.1021/acsomega.3c06142>

Figures

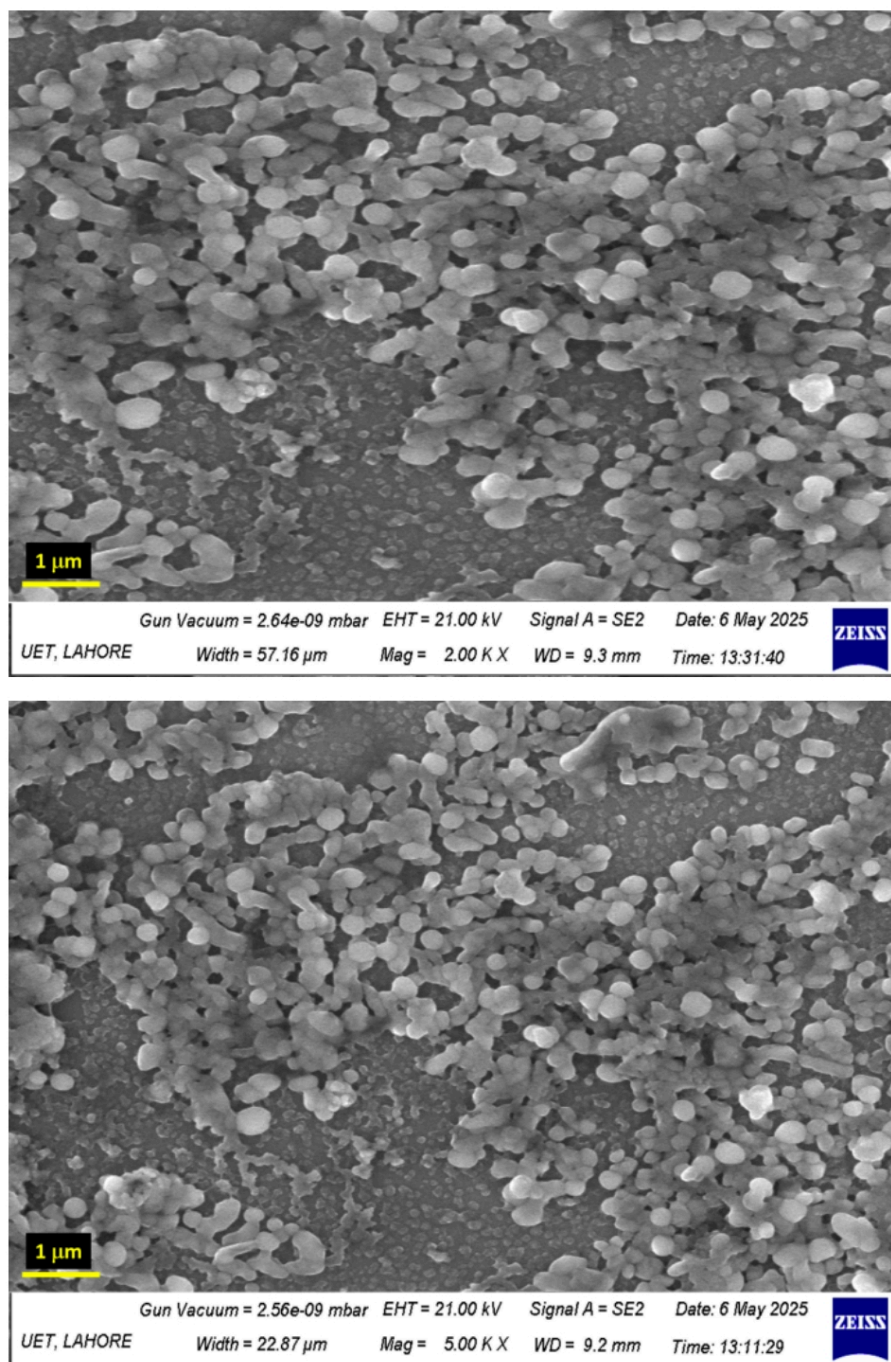


Figure 1

SEM images of the product, 21 kV (top: 2000×, bottom: 5000× by the data bar).

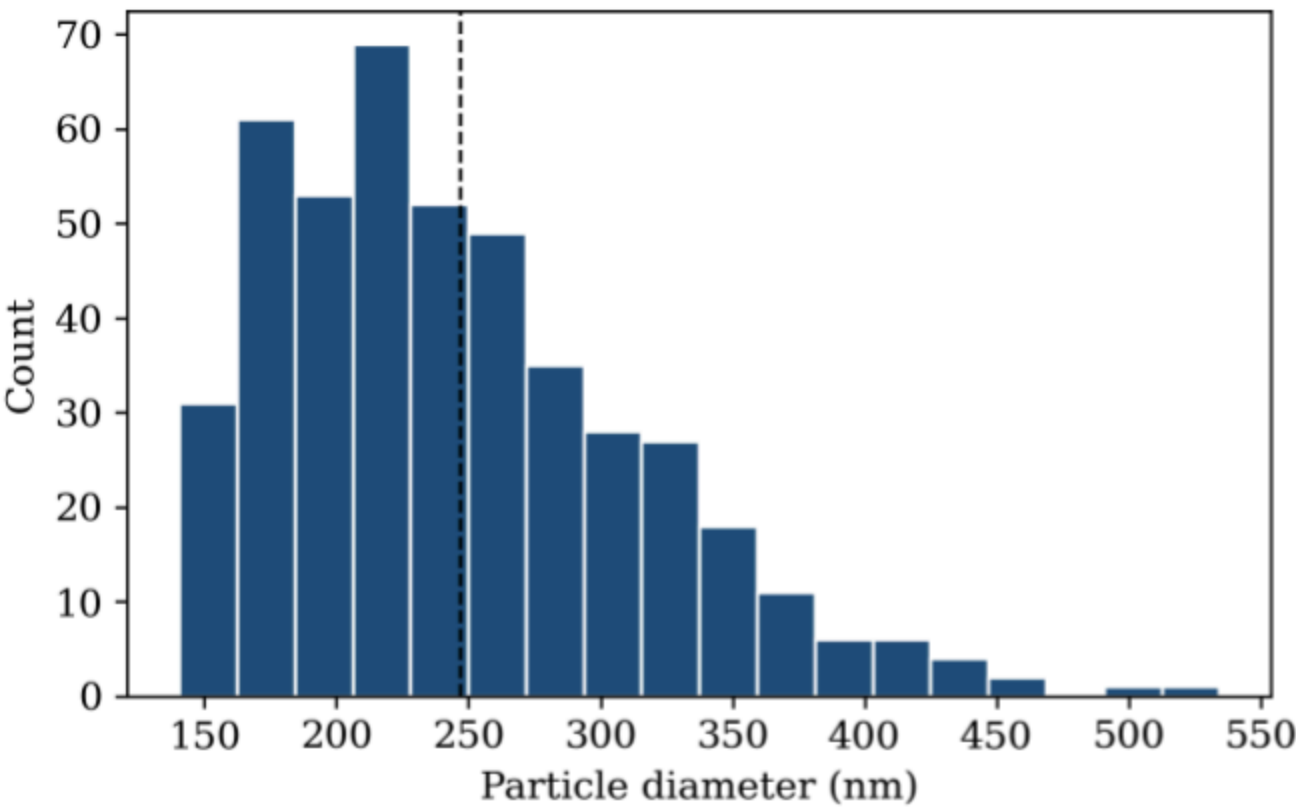


Figure 2

Particle size distribution (n = 454).

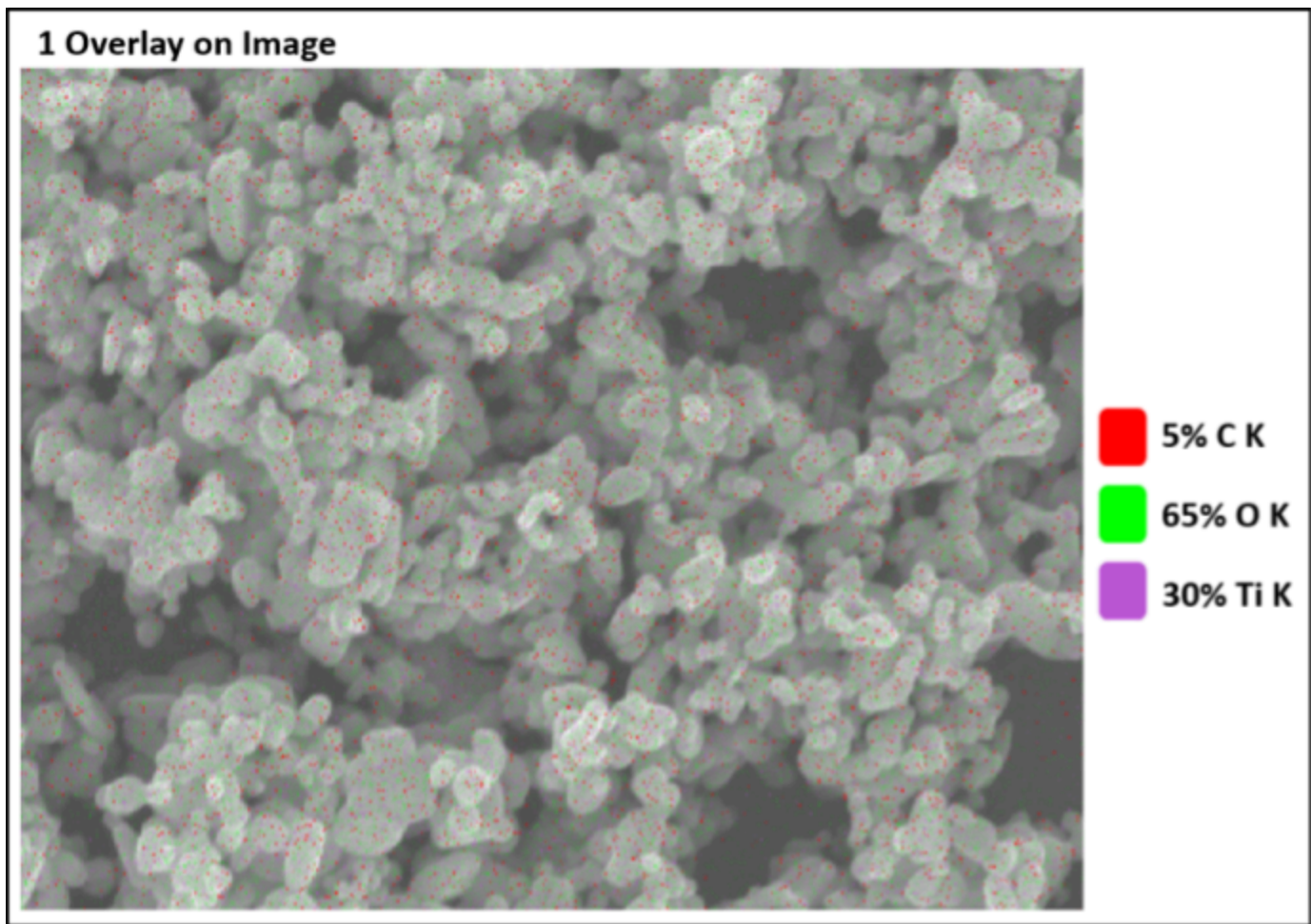


Figure 3

EDX map overlay.

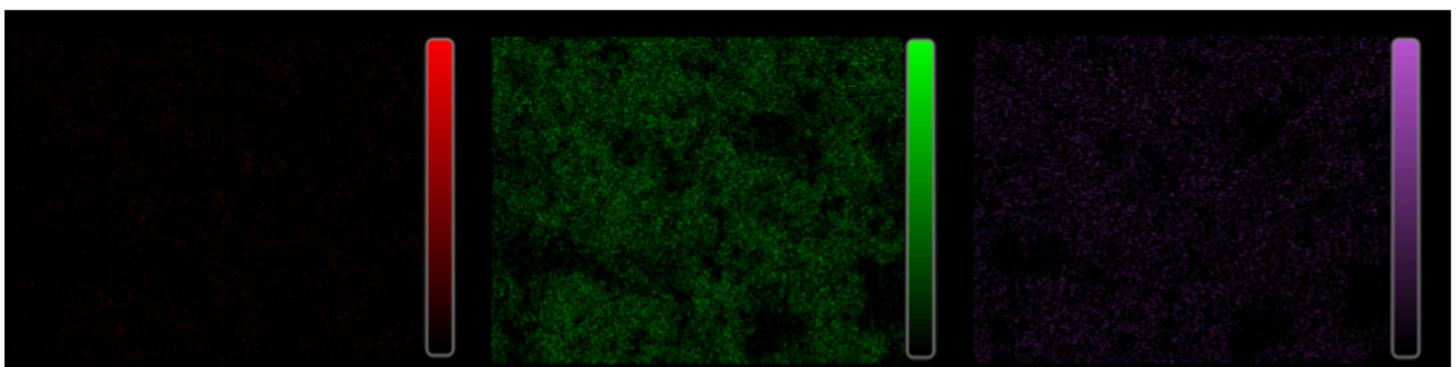


Figure 4

Element maps: C (left), O (middle), Ti (right).

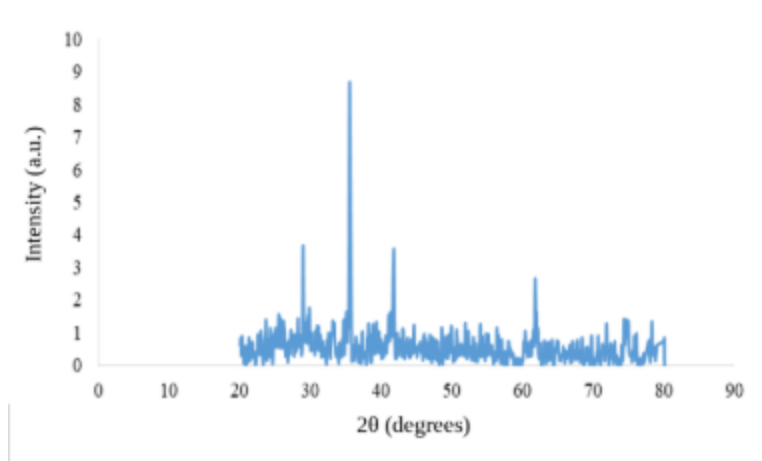


Figure 5

XRD pattern of the product.

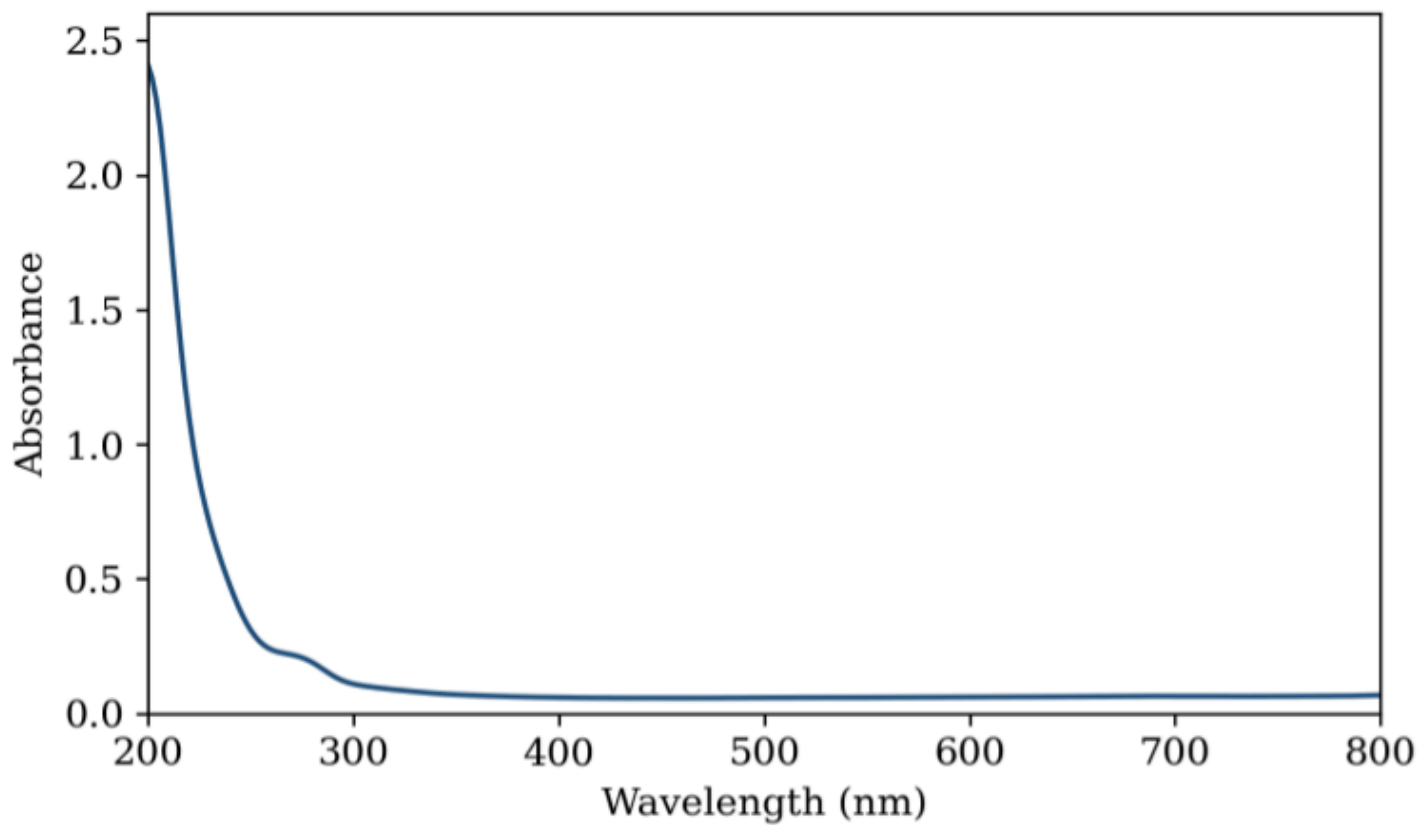


Figure 6

UV-Vis spectrum of the product.

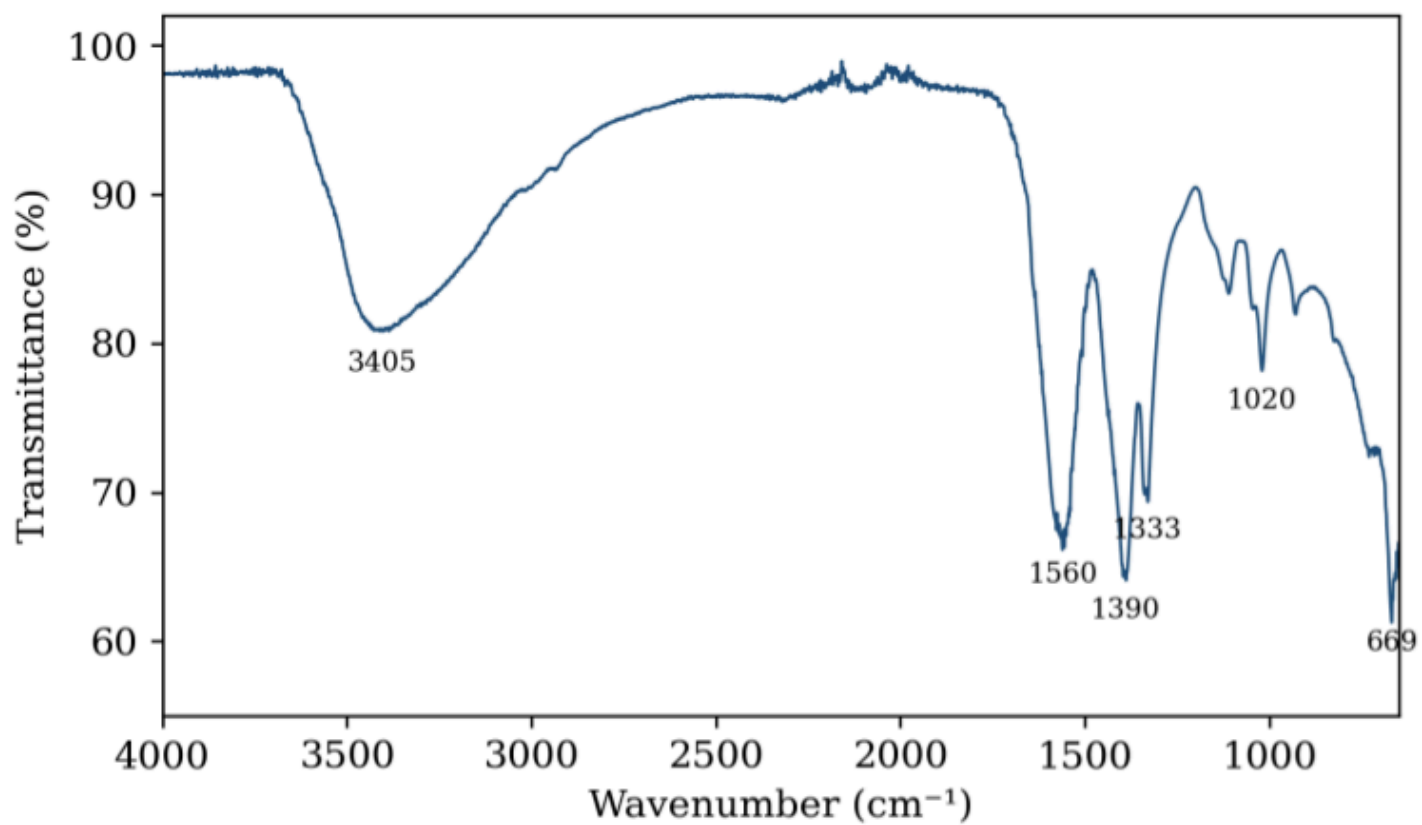


Figure 7

FTIR spectrum of the product.