

Green fabrication of Piper betle leaf extract assisted magnesium oxide nanoparticles with antioxidant potential

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Short Report

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Abstract

The combination of Biotechnology and Nanotechnology is called as Bio-nanotechnology. Bio-nanotechnology is generally used to create nanomaterial utilizing biological method. In this paper, MgO nanoparticles were produced by utilizing extract of *Piper betle* leaf as bio-reducing agent. By employing the standard techniques namely UV-Visible spectroscopy, X-ray diffraction, Fourier transform infrared spectroscopy, scanning electron microscopy with energy dispersive analysis and thermogravimetric analysis to find out the physio chemical properties of green synthesized MgO NPs. All the outcome shows that the produced particles were spherical shape in the range of 18–20 nm, crystalline nature. The antioxidant activity of *P. betle* assisted MgO NPs were performed using DPPH assay. The DPPH assay of *P. betle* assisted MgO NPs could reach 88% at a concentration of 500 µg/ml, respectively. From the outcomes it was determined that the *P. betle* assisted MgO NPs have good antioxidant properties.

Introduction

Magnesium oxide (MgO) is metal oxide and attractive metal, which used as catalyst to manufacture the electronic devices and having the low heat capacity and high melting point (Hulkoti and Taranath, 2014). MgO NPs are produced by the standard methods such as hydrothermal, sol gel, chemical gas phase deposition and wet precipitation methods. MgO NPs produced by conventional method is not used for medical applications. But green synthesis mediated nanoparticles is used for clinical analysis (Palanisamy and Pazhanivel, 2017). Green synthesis using plants is importance due to cost-effective, versatility, efficiency and environmental friendly. Phyto-mediated nanoparticles are produced using leaf extract because phytochemicals act as a reducing and capping agent for formation of nanomaterials (Jeevanandam et al., 2017). *Piper betle* Linn. (Family: Piperaceae) is a perennial dioecious, semi woody climber. Stems are strongly swollen at the nodes, and papillose when young. Leaves were alternate, simple and yellowish green to bright green in color. It is mostly cultivated in India, Sri Lanka, Indonesia, Malaysia, East Africa and Philippine Islands. *Piper betle* show antibacterial (Lubis and Marlisa, 2020), antifungal (Ali et al., 2010), hepatoprotective (Panda et al., 2018), antioxidant (Choudhary and Kale, 2002), wound healing (Santhanam and Nagarajan, 1990) and gastro protective activities (Arawwawala et al., 2014).

Zinc oxide nanoparticles were synthesized using aqueous *P. betle* leaf extract and evaluate the application in surgical sutures by Thi Tran et al. (2021). Few of the researchers have synthesized the silver nanoparticles using *Piper betle* leaf broth as stabilizing and reducing agent (Khan et al.,2020; Ramachandran et al., 2016; Mallikaarjuna et al., 2012). Punuri et al., (2012) reported the fabrication of gold nanoparticles using the aqueous extract of *P. betle* leaf. Palladium (Pd) and platinum (Pt) nanoparticles were biosynthesized and of using *Piper betle* L. by photo reduction method (Rajasekharreddy and Rani, 2014). *Piper betle* leaves mediated synthesis of biogenic SnO₂ nanoparticles were produced by Singh et al., (2018) and determined its photocatalytic degradation of reactive yellow 186 dyes. Kaur et al., (2021) were performed the one-pot biogenic synthesis of TiO₂ nanoparticles using *P. betle* for the degradation of industrial reactive yellow 86 dye. Biogenic synthesis of copper oxide

nanoparticles using leaf extracts of *P. betle* were carried out by Kasi et al., (2021) and they assessed its antibacterial activities.

Thus, the aim of present study is to synthesis of MgO nanoparticles with good antioxidant potential using aqueous extract of *Piper betle* leaf. The extract of betel leaf has been used as a reducing agent for MgO formation.

Materials And Methodology

Collection of P. betle and preparation of P. betle leaf extract

Piper betle leaves were got from the local market of Ukkadam, Coimbatore (District) Tamilnadu, India in the month of September 2021. The obtaind freshly *P. betle* leaves were washed thoroughly via running tap water and distilled water. The extract of *piper betle* was prepared using 100 ml de-ionized water and allowed it for boiling at 100°C for 30 min. The obtained extract was filtered using Whatman No1 filter paper and it was stored in sterile bottles for future analysis (Sivaraj et al., 2014).

Synthesis and characterization of P. betle assisted MgO NPs

10 g of Magnesium nitrate was dissolved in 100 ml distilled water and it was allowed to boil for 20 min at 60°C. Then the 50 ml of *P. betle* leaf extract was added above solution and it kept at 60°C for 30 min. The pH was maintained above 8. Next, the mixture was incubated at room temperature for 12 h. Finally, the dark greenish precipitate was obtained. Then, the precipitate was annealed at 400°C-500°C for 3 h. After annealing process, fine powder was acquired and it stored at 4°C for further studies (Vanathi et al., 2014).

The physicochemical properties of *P. betle* assisted MgO NPs such as optical properties, chemical bonding, crystalline nature, elemental composition, purity, morphology, size and thermal stability were observed using stranded techniques and instruments namely UV–Visible spectrophotometer, XRD, FTIR spectrometer, Scanning electron microscopy with EDX, and thermo gravimetric analyzer (Imani and Safaei., 2019; Al-Ajmi et al., 2018).

Antioxidant Activity of P. betle assisted MgO NPs

The antioxidant activity of P: betle assisted MgO NPs was determined by 2,2-diphenyl-1-picrylhydrazyl (DPPH) assay. Different concentrations of MgO NPs were prepared. 700 μ l of 0.1 mM of DPPH reagents was mixed with 100 μ l of tested solution and it was protected from light. H₂O and Methanol were employed as references. After 30 min of shaking the solution mixtures, the absorbance was recorded at wavelength of 515 nm. The ability to reduce free DPPH radicals was estimated by according to Nguyen et al., (2021).

Result And Discussion

Physicochemical characterization of P. betle assisted MgO NPs

Figure 1 presented the UV-vis spectra of *P. betle* assisted MgO NPs. The UV-vis spectra of MgO NPs showed a single peak at 384 nm in the wavelength range of 200–800 nm. It demonstrated that MgO NPs with a surface plasmon resonance acquired in the reacted mixtures and MgO NPs were formed. The optical properties of MgO NPs were confirmed using UV- Visible spectrophotometer and peak attained at 384 nm. It was a similarity to pervious research reports (Ammulu et al., 2021; Sharma et al., 2017). And they confirmed the formation of MgO NPs by color changes and obtained a peak at 310 nm in UV-Spectrophotometry analysis.

The XRD analysis is broadly used to conclude the nature, structure and size of nanoparticles. Figure 2 shows the XRD pattern of MgO NPs synthesized by using the *P. betle* extract. Debye–Scherrer's formula was employed to calculate the size of MgO NPs and which was found to be 18 nm. The obtained planes in XRD spectra were clearly reveals that bio-fabricated MgO NPs were crystalline nature. Umaralikhan and Jamal Mohamed Jaffar, (2018) were synthesized of MgO nanoparticles using aqueous leaf extracts of *Pisidium guvajava* and *Aloe vera*. They determined the nature and size of the nanoparticles by XRD analysis.

FT-IR analysis was used to find out the potential functional molecules which answerable for the capping and reduction of MgO NPs. Figure 3 refers the FT-IR spectra of MgO NPs synthesized using the P betle extract. FT-IR shows sharp absorption peaks of 486, 699, 754, 1209, 1453, 1789, 1909, 3622 and 3919 cm $^{-1}$. The stretching vibration mode for the Mg $^{-0}$ -Mg moiety was observed at the peaks of 486, 699 and 754 cm $^{-1}$ (Taghavi Fardood et al., 2018). Others peaks were corresponding to $^{-1}$ H stretching vibration and $^{-1}$ C = 0. FT-IR spectra confirms that; MgO NPs have functional groups of phytochemicals.

The surface morphology of *P. betle* assisted MgO NPs was determined using the SEM analysis. It revealed the regular spherical shaped morphology of MgO NPs (Fig. 4). This result agreed with pervious investigation (Nguyen et al., 2021). The elemental composition and purity of *P. betle* assisted MgO NPs were observed by using the EDX technique (Fig. 5). EDX spectra shows the occurrence of Mg, O and C, which confirms the formation of MgO NPs. Dobrucka, (2018) was reported the synthesis of MgO NPs using *Artemisia abrotanum* herba extract and synthesized MgO NPs shows presence of magnesium, other element such as carbon, calcium etc.

The thermal analysis of *P. betle* assisted MgO NPs was performed to assess it weight loss when increasing temperature range from 50 to 1000°C. Figure 6 and b illustrates the TGA curve of MgO NPs. Overall, 33.6% of weight loss was occurred on MgO NPs in the range of 50 to 1000°C. It refers the losses of phyto-molecules in surface of MgO NPs. This study was compare with previous report (Wong et al., 2020).

Antioxidant Assay

P. betle assisted MgO NPs was used to determine its antioxidant activity by DPPH assay (Table 1). The antioxidant activity was shows dependent on concentration of MgO NPs. The low concentration (50 μ g/ml) of MgO NPs showed minimum percentage (25.8%) of free radical scavenging activity, whereas

the highest concentration (500 μ g/ml) of MgO NPs shows the maximum percentage (88.9%) of free radical inhibition. The highest percentage of free radical inhibition was observed in *P. betle* assisted MgO NPs due to presence of bioactive compounds in surface of nanoparticles. Ammulu et al., (2021) reported the synthesis of MgO NPs using *Pterocarpus marsupium* rox. b heartwood extract and evaluate its antioxidant activities. They concluded that, Phyto-assisted MgO NPs have good antioxidant activity.

Table 1
Antioxidant activity of *P. betle* assisted MgO NPs

S.No.	Concentration of <i>P. betle</i> assisted MgO NPs (µg/ml)	Free radical scavenging activity (%)
1	50	25.8 ± 0.20
2	100	48.9 ± 0.78
3	200	68.89 ± 0.99
4	500	92.89 ± 0.98

Conclusion

In the present investigation by using leaf extract of *P. betle*, MgO·NPs were produced and the physicochemical properties were analyzed by the strand techniques. All the analyses were reveals that asbiosynthesized MgO NPs are crystalline nature, spherical with average size of 18 nm. Functional group of phytochemicals were present in the surface of MgO NPs, which was confirmed by FT-IR analysis. Biosynthesized MgO NPs shows good antioxidant properties. Due to presence of phytochemicals from *P. betle* in MgO NPs may be responsible for antioxidant activity. In future, the further research work will be performed at in vivo condition.

Declarations

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Consent to Participate – Not applicable

Consent to Publish - Not applicable

Authors Contributions

PR: Supervision, Funding acquisition and Project administration

MJS: Investigation, Methodology, Writing- Original draft preparation

JD: Data Curation and Writing

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Competing Interests

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Availability of data and materials – Not applicable

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* Authors' information (optional). - Not applicable

Please include the sub-sections below of Compliance with Ethical Standards section. - Not applicable

- * Disclosure of potential conflicts of interest Not applicable
- * Research involving Human Participants and/or Animals Not applicable
- * Informed consent Not applicable

References

- 1. Al-Ajmi, M. F., Hussain, A., Alsharaeh, E., Ahmed, F., Amir, S., Anwar, M. S. ... Koo, B. H. (2018). Green synthesis of zinc oxide nanoparticles using Alstonia macrophylla leaf extract and their in-vitro antiThi Tran, Q.M., Thi Nguyen, H.A., Doan, V.D., Tran, Q.H. and Nguyen, V.C., 2021.,10(3), pp.349–355
- 2. Ali, I., Khan, F. G., Suri, K. A., Gupta, B. D., Satti, N. K., Dutt, P. ... Khan, I. A. (2010). In vitro antifungal activity of hydroxychavicol isolated from Piper betle L. *Annals of clinical microbiology and antimicrobials*, 9(1), 1–9
- 3. Ammulu, M. A., Vinay Viswanath, K., Giduturi, A. K., Vemuri, P. K., Mangamuri, U., & Poda, S. (2021). Phytoassisted synthesis of magnesium oxide nanoparticles from Pterocarpus marsupium rox. b

- heartwood extract and its biomedical applications. *Journal of Genetic Engineering and Biotechnology*, 19(1), 1–18
- 4. Arawwawala, L. D. A. M., Arambewela, L. S. R., & Ratnasooriya, W. D. (2014). Gastroprotective effect of Piper betle Linn. leaves grown in Sri Lanka. *Journal of Ayurveda and integrative medicine, 5*(1), p.38
- 5. Choudhary, D., & Kale, R. K. (2002). Antioxidant and non-toxic properties of Piper betle leaf extract: in vitro and in vivo studies. *Phytotherapy Research: An International Journal Devoted to Pharmacological and Toxicological Evaluation of Natural Product Derivatives*, 16(5), 461–466
- 6. Dobrucka, R. (2018). Synthesis of MgO nanoparticles using Artemisia abrotanum herba extract and their antioxidant and photocatalytic properties. *Iranian Journal of Science and Technology, Transactions A: Science*, 42(2), 547–555
- 7. Hulkoti, N. I., & Taranath, T. C. (2014). Biosynthesis of nanoparticles using microbes—a review. *Colloids and Surfaces B: Biointerfaces*, 121, 474–483
- 8. Imani, M. M., & Safaei, M. (2019). Optimized synthesis of magnesium oxide nanoparticles as bactericidal agents. Journal of Nanotechnology, 2019
- 9. Jeevanandam, J., Chan, Y. S., & Danquah, M. K. (2017). Biosynthesis and characterization of MgO nanoparticles from plant extracts via induced molecular nucleation. *New Journal of Chemistry*, 41(7), 2800–2814
- 10. Kasi, S. D., Ramasamy, J. M., Nagaraj, D., Santiyagu, V., & Ponraj, J. S. (2021). Biogenic synthesis of copper oxide nanoparticles using leaf extracts of Cissus quadrangularis and Piper betle and its antibacterial effects. *Micro & Nano Letters*, 16(8), 419–424
- 11. Kaur, G., Kaur, H., Kumar, S., Verma, V., Jhinjer, H. S., Singh, J. ... Al-Rashed, S. (2021). Blooming approach: one-pot biogenic synthesis of TiO2 nanoparticles using piper betle for the degradation of industrial reactive yellow 86 Dye. *Journal of Inorganic and Organometallic Polymers and Materials*, 31(3), 1111–1119
- 12. Khan, S., Singh, S., Gaikwad, S., Nawani, N., Junnarkar, M., & Pawar, S. V. (2020). Optimization of process parameters for the synthesis of silver nanoparticles from Piper betle leaf aqueous extract, and evaluation of their antiphytofungal activity. *Environmental Science and Pollution Research*, 27(22), 27221–27233
- 13. Lubis, R. R., & Marlisa, D. D. W. (2020). Antibacterial activity of betle leaf (Piper betle I.) extract on inhibiting Staphylococcus aureus in conjunctivitis patient. American journal of clinical and experimental immunology, 9(1), p.1
- 14. Mallikaarjuna, K., Dillip, G. R., Narasimha, G., Sushma, N. J., & Prasad Raju, N. D.,B (2012). Phytofabrication and characterization of silver nanoparticles from Piper betle broth. *Research Journal of Nanoscience and nanotechnology*, 2(1), 17–23
- 15. Nguyen, D. T. C., Dang, H. H., Vo, D. V. N., Bach, L. G., Nguyen, T. D., & Van Tran, T. (2021). Biogenic synthesis of MgO nanoparticles from different extracts (flower, bark, leaf) of Tecoma stans (L.) and their utilization in selected organic dyes treatment. *Journal of Hazardous Materials*, 404, p.124146

- 16. Nguyen, N. H., Nhi, T. T. Y., Van Nhi, N. T., Cuc, T. T. T., Tuan, P. M., & Nguyen, D. H. (2021). Comparative Study of the Silver Nanoparticle Synthesis Ability and Antibacterial Activity of the Piper Betle L. and Piper Sarmentosum Roxb. Extracts. *Journal of Nanomaterials*, 2021
- 17. Palanisamy, G., & Pazhanivel, T. (2017). Green synthesis of MgO nanoparticles for antibacterial activity. *International research journal of engineering and technology*, 4(9), 137–141
- 18. Panda, S., Sharma, R., & Kar, A. (2018). Antithyroidic and hepatoprotective properties of high-resolution liquid chromatography–Mass spectroscopy-standardized Piper betle leaf extract in rats and analysis of its main bioactive constituents. *Pharmacognosy Magazine*, *14*(59), p.658
- 19. Punuri, J. B., Sharma, P., Sibyala, S., Tamuli, R., & Bora, U. (2012). Piper betle-mediated green synthesis of biocompatible gold nanoparticles. *International Nano Letters*, 2(1), 1–9
- 20. Rajasekharreddy, P., & Rani, P. U. (2014). Biosynthesis and characterization of Pd and Pt nanoparticles using Piper betle L. plant in a photoreduction method. *Journal of Cluster Science*, 25(5), 1377–1388
- 21. Ramachandran, K., Kalpana, D., Sathishkumar, Y., Lee, Y. S., & Ravichandran, K. (2016). A facile green synthesis of silver nanoparticles using Piper betle biomass and its catalytic activity toward sensitive and selective nitrite detection. *Journal of industrial and engineering chemistry*, 35, 29–35
- 22. Santhanam, G., & Nagarajan, S. (1990). Wound healing activity of Curcuma aromatica and Piper betle. *Fitoterapia*, 61(5), 458–459
- 23. Sharma, G., Soni, R., & Jasuja, N. D. (2017). Phytoassisted synthesis of magnesium oxide nanoparticles with Swertia chirayaita. *Journal of Taibah University for Science*, 11(3), 471–477
- 24. Singh, J., Kaur, N., Kaur, P., Kaur, S., Kaur, J., Kukkar, P. ... Rawat, M. (2018). Piper betle leaves mediated synthesis of biogenic SnO2 nanoparticles for photocatalytic degradation of reactive yellow 186 dye under direct sunlight. *Environmental nanotechnology, monitoring & management*, 10, 331–338
- 25. Sivaraj, R., Rahman, P. K., Rajiv, P., Narendhran, S., & Venckatesh, R. (2014). Biosynthesis and characterization of Acalypha indica mediated copper oxide nanoparticles and evaluation of its antimicrobial and anticancer activity. *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy*, 129, 255–258
- 26. Taghavi Fardood, S., Ramazani, A., & Woo Joo, S. (2018). Eco-friendly synthesis of magnesium oxide nanoparticles using arabic Gum. *Journal of Applied Chemical Research*, 12(1), 8–15
- 27. Thi Tran, Q. M., Nguyen, T., Doan, H. A., Tran, V. D., Q.H. and, & Nguyen, V. C. (2021). Biosynthesis of zinc oxide nanoparticles using aqueous piper betle leaf extract and its application in surgical sutures. *Journal of Nanomaterials*, 2021
- 28. Umaralikhan, L., & Jamal Mohamed Jaffar, M. (2018). Green synthesis of MgO nanoparticles and it antibacterial activity. *Iranian Journal of Science and Technology, Transactions A: Science*, 42(2), 477–485
- 29. Vanathi, P., Rajiv, P., Narendhran, S., Rajeshwari, S., Rahman, P. K., & Venckatesh, R. (2014). Biosynthesis and characterization of phyto mediated zinc oxide nanoparticles: a green chemistry

- approach. Materials Letters, 134, 13-15
- 30. Wong, C. W., Chan, Y. S., Jeevanandam, J., Pal, K., Bechelany, M., Abd Elkodous, M., & El-Sayyad, G. S. (2020). Response surface methodology optimization of mono-dispersed MgO nanoparticles fabricated by ultrasonic-assisted sol-gel method for outstanding antimicrobial and antibiofilm activities. *Journal of Cluster Science*, 31(2), 367–389

Figures

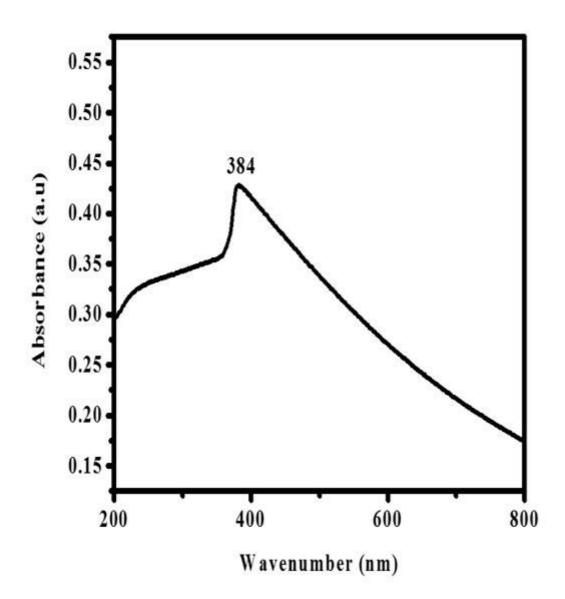


Figure 1

XRD analysis for *P. betle* assisted MgO NPs.

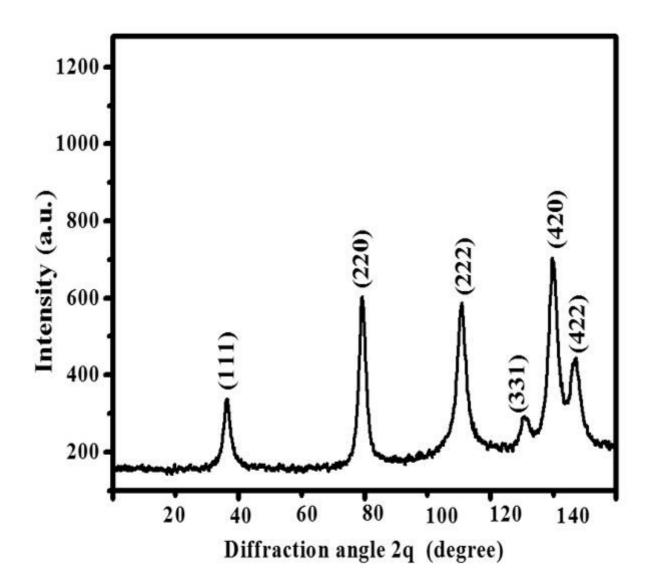


Figure 2XRD analysis for *P. betle* assisted MgO NPs.

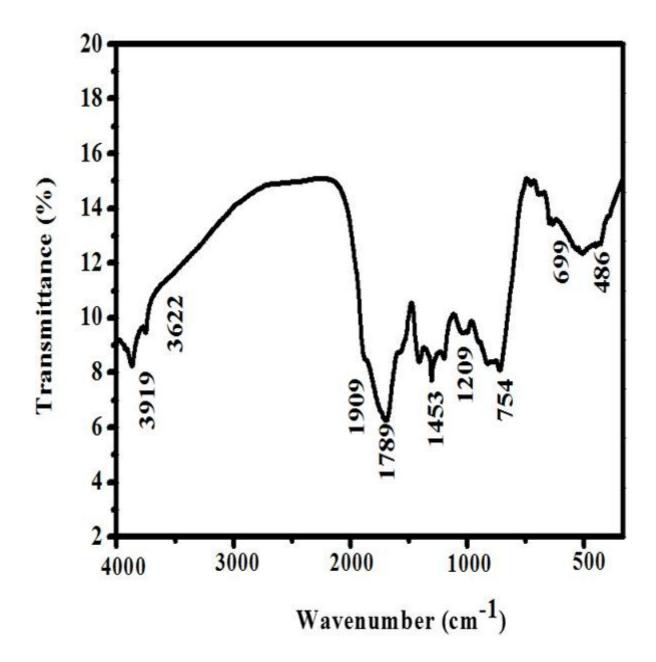


Figure 3
FT-IR analysis for *P. betle* assisted MgO NPs.

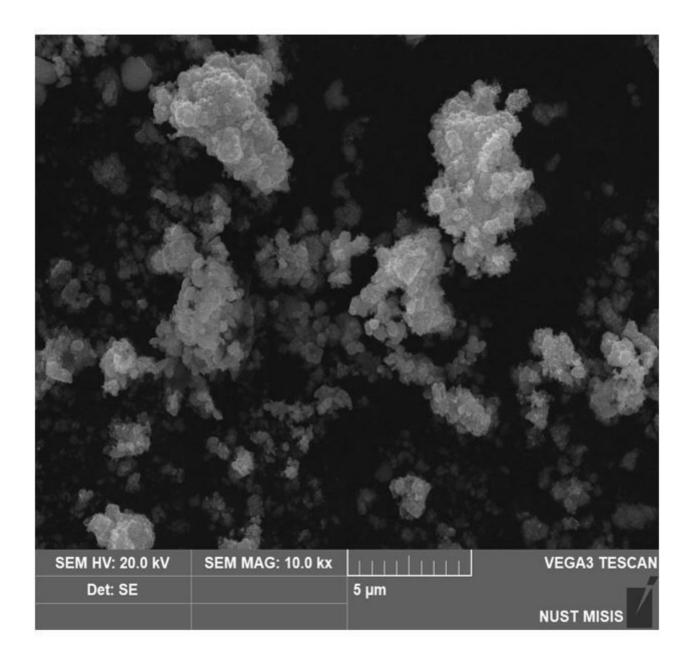


Figure 4SEM analysis for *P. betle* assisted MgO NPs.

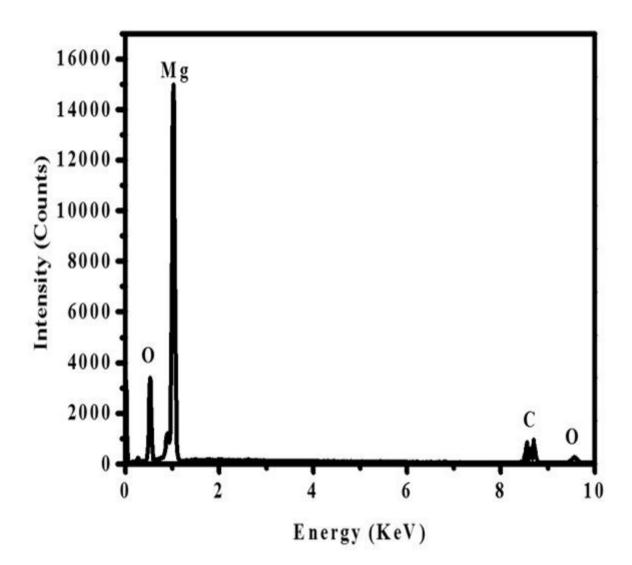
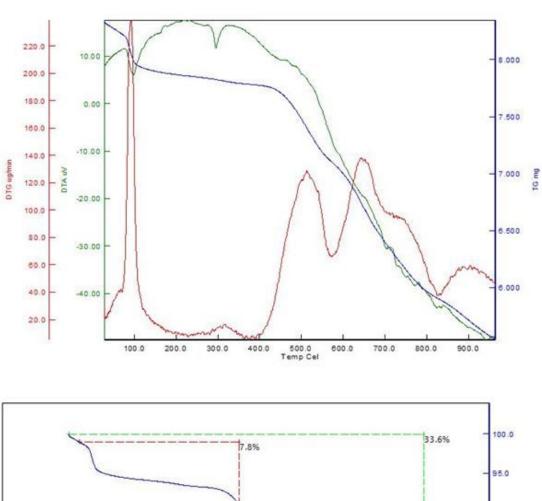


Figure 5EDX analysis for *P. betle* assisted MgO NPs.



77.8% - 100.0 - 95.0 - 95.0 - 90.0 - 90.0 - 75.0 -

Figure 6

6a. TGA analysis for *P. betle* assisted MgO NPs (Red- DTG; Blue-TG; Green-DTA)

6b. Determination of weight loss on *P. betle* assisted MgO NPs by TGA analysis.