Degradation Of Methylene Blue using Silver Nanoparticles Synthesized from Cynodon dactylon (L.) Pers. Leaf aqueous extract

R.Anjana, N.Geetha

Abstract: The present study reports a green chemistry approach for the phytosynthesis of silver nanoparticles (AgNPs) by using the leaf extract of cynodon dactylon (L.) Pers and dye degradation property of its AgNPs. Initially, synthesis of AgNPs was confirmed by colour change from pale yellowish reaction mixture to dark brown after 20 minutes of boiling. In order to confirm the formation of silver nanoparticles, UV-Vis, XRD and SEM charaterizations were made. The nanoparticles showed an absorption peak at 350 nm in UV-Visible spectrum corresponding to the Plasmon Resonance Band of the synthesized AgNPs. XRD analysis showed that size of the silver nanoparticles was around 13 nm. Various concentration of nanoparticles (2.0-10.0 mg)were mixed separately with water containing methylene blue dye (10 mgl1000 ml). A control was maintained without the addition of the AgNPs. At specific day intervals, aliquots of 2-3 ml suspension were filtered and used to take absorbance at 660 nm. Using the absorbance values, percentage of dye degradation was calculated. The percentage of dye degradation was increased with increasing the day. Of various concentration of AgNPs used, dye solution containing 10 mg AgNPs showed 75% dye degradation after 5 days of incubation at room temperature.

Index Terms: Cynodon dactylon, silver nanoparticles, dye degradation, methylene blue

1 INTRODUCTION

Nanotechnology is the science of producing and utilizing nanosized particles [1]. Silver nanoparticles (AgNPs) is useful in various fields such as catalysis, optics, biomedical, pharmaceutical and sensor technology [2]. Although many methods are available to synthesize AgNPs, most of them utilized toxic chemicals and also include the use of enormous energy. This scenario is not economic and cause serious pollution to environment. Synthesis of nanomaterials using biological entities is gaining attention because biological methods are less expensive, nontoxic and involving environmentally acceptable procedures [3]. The use of plant and plant extract in nanoparticles synthesis is advantageous compared to microbial based system because it eliminates the intricate process of maintaining cell cultures [4]. Dyes are a major class of synthetic organic compounds used in many industries especially textiles [5] which consume about 60% of total dye production for coloration of different fabrics. Moreover, after the completion of their use nearly 15% of dyes are washed out. These dye compounds dissolve in water bodies with a concentration in between 10 and 200 milligram per liter results in major water pollution worldwide [6,7]. Therefore, treatment of dye effluents from textile industries is a compulsory part of waste water treatment. The release of dye effluents in aquatic systems is chief environmental concern because coloration not only decreases sunlight penetration and dissolved oxygen in water bodies, but also releases toxic compounds during chemical or biological reaction pathway that affects aquatic flora and fauna [8]. Nowadays biosynthesised nanocatalysts are widely used for the efficient removal of dye contaminants. Plant contains a complex network of metabolites and enzyme that can be used to synthesize naoparticles. The presence of different chemical compounds in plant such as polyphenols, flavonoids, sterols, triterpenes, reducing sugar like glucose and fructose, and protein could help produce metallic nanoparticles [9]. Since nanotechnology field has provided a new platform for waste water treatment [10], researchers have explored this technique for degradation of dye [8].

The focus of the present investigation is to apply the accurate principles of green chemistry for the synthesis of AgNPs using leaf extract of Cyanodon dactylon (L.) Pers. (Bermuda grass) as reducing and capping agent. Bermuda grass belongs to the family, Poaceae. it is native to East Asia, Africa, Southern Europe and Australia. Cyanodon is generally considered as a weed and has been found to possess various potential medicinal properties [12]. Many researches showed that Bermuda grass as a wide spread creeping grass could be useful for stabilizing spill affected soil [13,14]. Various studies show that Pb, Ni [15], Mn, Cu, Zn, Pb, Co [16], Cr, Zn, Cd [17] and Cr, U [18] bioaccumulation potential of this plant. Recently, biosorption of thymol blue from industrial wastewater using activated biocarbon from this plant leaves was shown [19]. Methylene blue is a model cationic dye employed by industries such textile industry for a variety of purposes. It is a heterocyclic aromatic chemical compound with a molecular formula $C_{16}H_{18}ClN_3S$. It causes eye burn which may be responsible for permanent injury to the eyes of human as well as aquatic animals. It can also cause irritation to the gastrointestinal tract with symptoms of nausea, vomiting and diarrhea. Methylene blue also causes irritation to the skin when in contact with it [20]. So far, there is no report on exploration of phytosynthesized silver nanoparticles of C. dactylon for degradation of textile dye methylene blue. Therefore, the study was undertaken with the objectives: to synthesis and characterization of silver nanoparticles using leaf extract of C.dactylon and to assess the exploitation feasibility of these nanoparticles for phytoremediation purpose.

2 MATERIALS AND METHODS

Preparation of plant extract

C. dactylon leaves were collected from Bharathiar university campus, Coimbatore, Tamil Nadu, India. Washed leaves were cut into bits and 10 mg of tissue ground well with adding adequate distilled water. The filtrate was made up to 100 ml with distilled water and it was boiled for 20 minutes. After cooling, it was centrifuged at 8000 rpm for 10 minutes. Then the supernatant was taken for the synthesis of silver nanoparticles.

Preparation of silver nitrate solution
Silver nitrate solution was prepared using $AgNO_3$ in different concentrations such as 1 mM, 2 mM, 3 mM, 4 mM and 5 mM and prepared solutions were stored under dark condition.

Synthesis of silver nanoparticles
10 ml of supernatant was taken in a conical flask and 90 ml of different concentrations of silver nitrate solution was added to it separately. Then the solution was boiled in a waterbath for 20 minutes. The formation of silver nanoparticle was indicated by the appearance of dark brown colour in the reaction mixture after boiling.

UV-Visible spectra analysis
UV-visible spectral analysis was performed for samples containing different concentrations of $AgNO_3$ and the absorption maxima were recorded at a wavelength of 200-800 nm using spectrophotometer.

XRD analysis of silver nanoparticles
After confirmation of phytosynthesis of AgNPs by UV-Vis, the solution was centrifuged at 8000 rpm for 10 minutes. The pellet was washed with water two to three times and allowed to dry to get pure AgNPs. It was then subjected to X-ray diffraction (XRD) to examine the crystalline nature of AgNPs in the given sample.

Degradation of methylene blue dye
10 ppm methylene blue dye solution was prepared by adding 10 mg of dye in 1 L distilled water. To study the catalytic degradation of methylene blue by silver nanoparticles, different concentrations of AgNPs such as 2.0, 4.0, 6.0, 8.0, 10.0 mg were weighed and mixed with prepared dye solution separately. Then the flasks were placed in an orbital shaker. A control was also maintained without addition of nanoparticles. The degradation of the dye was observed by the reduction in the colour of the dye. The absorbance values were taken at 660 nm for 5 days with an interval of 24 hours to calculate the reduction of methylene blue dye in solution at room temperature.

Percentage of dye degradation was estimated by the following formula:

$$\%\text{Decolourization} = (C_0 - C)/C_0 \times 100$$

where C_0 is the initial concentration of dye solution and C is the concentration of dye solution after catalytic degradation.

Statistical analysis
Means and standard deviations were derived from measurements of three replicates for each treatment and their related controls.

3 RESULTS AND DISCUSSION

Among various concentrations containing $AgNO_3$ tried, 3 mM showed efficient production of AgNPs during 20 minutes boiling. Reduction of Ag into AgNPs is usually marked by colour change. The synthesized NPs exhibited dark brown colour transition from their original pale yellow colour (Figure 1) when monitored by UV-Vis spectroscope in a range of 200-800 nm. The colour change was due to the excitation of Surface Plasmon Resonance of the synthesized AgNPs [21]. A strong SPR band was appeared at 350 nm (Figure 2). In order to confirm the stability of synthesized AgNPs, OD was taken using small volume of sample for three days before using AgNPs for degradation of dye study and found that appearance of SPR band only at 350 nm. Generally green synthesized AgNPs were found to be stable for six months without relocating the SPR band [22, 23]. This stability may be due to the presence of phytochemicals present in the leaf extract of C.dactylon which act as stabilizing agents.

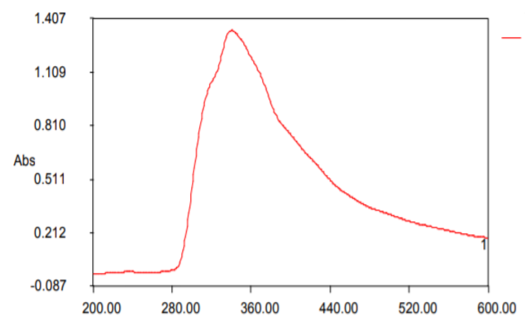

***Figure 1:** Phytosynthesis of AgNPs*

SPR band Wave length

Wave length

***Figure 2.**:UV-VIS Spectrum of phytosynthesized AgNPs*

XRD pattern of synthesized AgNPs was recorded using XRD Shimadzu 6000. The diffractogram obtained was compared with JCPDS library to confirm the crystalline structure of synthesized AgNP (Figure 3). The average size of the AgNPs was calculated using values of XRD (Table 1). Diffractogram shows three distinct diffraction peaks of 32.2, 27.7, 46.2° at 2θ values which can be indexed to the plane of (1 0 1), (3 1 0) and (1 5 1), correspondingly. The inter planar spacing (dA) values were found to be 2.775, 3.208 and 1.963 attributing to the above said planes, respectively. The average size of the AgNPs synthesized is approximately to be ~13 nm which is obtained by applying Debyr Scherrer's formula. Generally, strong peaks are associated with the face centered cubic lattice [24]. The remaining inconspicuous peaks obtained at 2θ values can be ascribed to some of the phytocompounds moieties present on the surface of synthesized AgNPs [25, 26]. SEM analysis indicates the presence of AgNPs in a denser condition with spherical and uniform nature (Figure 4). Mostly, they were found as mono dispersed form. In common, nanoparticles are not mingled with each other even though they are in aggregated form. This is due to the presence of capping agents or phytocompounds which usually stabilize the NPs [27].

.

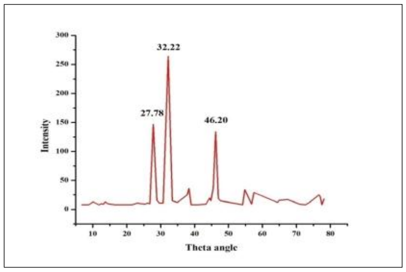

***Figure 3** XRD Diffractogram of Phytosynthesized AgNPs*

***Table 1:** XRD results of Phytosynthesized AgNPs*

Peak No .	2 θ	d(A)	I/I1	FWHM	Intensity counts	Integrated intensity counts
20	32.22	2.78	100	0.64	264	1998
16	27.79	3.21	56	0.62	147	979
31	46.21	1.96	51	0.74	134	942

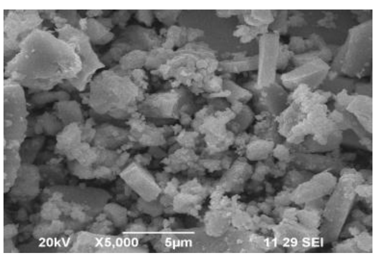

***Figure 4.** SEM image of Phytosynthesized AgNPs*

Catalytic activity of phytosynthesized silver nanoparticles on degradation of dye was demonstrated by using methylene blue at room temperature. The degradation of methylene blue was noted by colour change from the initial first day deep blue colour to light blue at the end of 5th day. For every 24 hours for 5 days, a small volume of filtered aliquot was used to take absorbance value at 660 nm. The absorption spectrum peaks were decreased gradually with the increase of the day exposure which indicates the catalytic degradation reaction of methylene blue dye by phytosynthesized silver nanoparticles (Figure 5). The percentage of dye degradation efficiency of silver nanoparticles was calculated for every 24 h for 5 days (Table 2). Of various concentrations of AgNPs, 10 mg was found effectively degrading the dye at the end of 5th day (75%). Dye
degradation of potential of phytosynthesized AgNPs were reported for various plants [28-30].

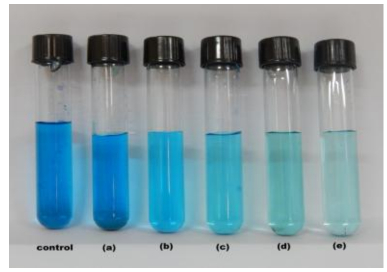

***Figure 5.** Visual observation of methylene dye degradation at different day exposure: control-without dye (a)dyesolution containing 2mg AgNPs (b) 4mg AgNPs (c) 6mg AgNPs (d) 8mg AgNPs and (e) 10mg AgNPs*

***Table 2.** Degradation of Methylene Blue by Phytosynthesized AgNPs*

AgNPs (mg/L)	Methylene Blue dye Degration (%) Day 1	Day 2	Day 3	Day 4	Day 5
2 mg	24± 0.07	30± 0.19	36± 0.17	42± 0.31	48± 0.14
4 mg	31± 0.17	38± 0.06	43± 0.06	49± 0.17	55± 0.24
6 mg	38± 0.17	43± 0.06	49± 0.17	56± 0.13	62± 0.06
8 mg	42± 0.21	49± 0.17	54± 0.24	61± 0.03	69± 0.10
10 mg	48± 0.12	56± 0.13	61± 0.27	68± 0.13	75± 0.13

Means ± SD of three experiments

4 CONCLUSION

Green nanotechnology is gaining importance due to its wide application and elimination of harmful reagent and provides effective synthesis of silver nanoparticles in one step. Here, the silver nanoparticles were synthesised using aqueous leaf extract of C. dactylon. The synthesised nanoparticles proved its efficiency for the degradation of one of the important textile dyes, i.e methylene blue in an eco-friendly manner. In future, an attempt will be made to enhance its efficiency for complete degradation of dye within short exposure by applying various parameters such as sunlight and controlled light exposure. Thus, this study exhibits the dye degradation property of the Bermuda grass especially their eco-friendly synthesised AgNPs.

ACKNOWLEDGEMENT

The authors acknowledge the financial support provided by UGC-SAP and DST-FIST, INDIA for carrying out this research.

REFERENCES

[1]. P. Abrahamsson, R. Moser, W. Pedrycz, A. Sillitti, G. Succi, "Effort Prediction in Iterative Software Development Processes -- Incremental Versus Global Prediction Models", Proceedings of the First International Symposium on Empirical Software Engineering and Measurement (ESEM), 2007

[2]. N. Khatoon, J.A. Mazumder, and M. Sardar, “Biotechnological Applications of Green Synthesized Silver Nanoparticles,” J. Nanosci. Curr. Res, vol.2, no. 1, pp.107. 2017.

[3]. P. Logeswari, S. Silambarasan, and J. Abraham, “Ecofriendly Synthesis of Silver Nanoparticles from Commercially available Plant powders and their Antibacterial properties,” Sci. Iran, vol. 20, no. 3,1049–1054, 2013.

[4]. S. Monda, N. Roy, R.A. Laskar, I, Sk, S. Basu, D. Mandal, and N.A.Begum, “Biogenic Synthesis of Ag, Au and Bimetallic Au/Ag alloy Nanoparticles using Aqueous extract of Mahogany (Swietenia mahogani JACQ.) leaves,” Colloids Surf. B, vol. 82, no. 2, pp. 497–504, Feb. 2011.

[5]. A. Annamalai, S.T. Babu, N.A. Jose, D. Sudha, and C.V. Lyza, "Biosynthesis and Characterization of Silver and Gold nanoparticles using Aqueous Leaf extraction of Phyllanthus amarus Schum & Thonn,” World Appl. Sci. J, vol.13, no. 8, pp.1833-1840, 2011.

[6]. M.H. Habib, and E. Askari, “Photocatalytic Degradation of an Azo textile dye with Manganese-doped ZnO Nanoparticles Coated on Glass.” Iran. J. Catal, vol,1, no. 1,pp. 41–44, 2011.

[7]. K.H. Gonawala, and M.J. Mehta, “Removal of Color from Different Dye Wastewater by using Ferric oxide as an Adsorbent,” Int. J. Eng. Res. Appl, vol. 4, no.5, pp.102-109, May 2014.

[8]. A.K. Dutta, S.K. Maji, and B. Adhikary, “γ-Fe_2O_3 Nanoparticles: an easily Recoverable Effective Photo-catalyst for the Degradation of Rose bengal and Methylene blue dyes in the Waste-water treatment plant,” Mater. Res. Bull, vol. 49, pp. 28–34, Jan. 2014. S.Khan, and A. Malik, "Environmental and Health Effects of Textile Industry Wastewater", In Environmental deterioration and human health, pp. 55-71. Springer, Dordrecht, 2014.

[9]. N.N, Bonnia, M.S. Kamaruddin, M.H. Nawawi, S. Ratim, H.N. Azlina, and E.S. Ali, “Green Biosynthesis of Silver nanoparticles using ‘Polygonum Hydropiper’and study its Catalytic Degradation of Methylene blue,” Procedia Chem, vol.1, no. 19, pp. 594-602, Jan. 2016.

[10]. G. Moussavi, and M. Mahmoudi, “Removal of Azo and Anthraquinone Reactive dyes from Industrial Wastewaters using MgO nanoparticles,” J. Hazard. Mater, vol. 168, no. 2-3, pp. 806-812, Sep. 2009.

[11]. L. Ai, and J. Jiang J, “Catalytic Reduction of 4-nitrophenol by Silver nanoparticles stabilized on Environmentally benign macroscopic Biopolymer Hydrogel,” Bio. Resour. Technol, vol. 132, pp. 374-377, Mar.2013.

[12]. S.K. Singh, P.K. Rai, S. Mehta, R.K. Singh, and Watal G, “Curative Effect of Cynodon dactylon against STZ induced Hepatic injury in Diabetic rats,” Ind. J. Clin. Biochem, vol. 24, no. 4, pp.410-413, Oct. 2009.

[13]. E. R. Smith, R. Naidu, and A.M. Alston, “Arsenic in the Soil Environment: A Review,” Doctoral dissertation, Academic press, 1998.

[14]. P.Madejon, J.M. Murillo, T. Maranon, F. Cabera, and R. Lopez, “Bioaccumulation of As, Cd, Cu, Fe and Pb in Wild Grasses Affected by Aznalcollar Mine Spill (SW Spain),” Sci. Total. Environ, vol. 290, no. 1-3, pp. 105-120, May 2002.

[15]. M. Soleimani, M.A. Hajabbasi, M. Afyuni, A.H. Charkhabi, and H. Shariatmadari, “Bioaccumulation of Nickel and Lead by Bermuda Grass (Cynodon dactylon) and Tall Fescue (Festuca arundinacea) from Two Contaminated Soils,” Caspian J. Env. Sci, vol. 7, no.2, pp. 59-70, 2009.

[16]. G. Mulugisi, J.R. Gumbo, F.A. Dacosta, and C. Muzerengi,”The Use of Indigenous Grass species as part of Rehabilitation of Mine tailings: A Case study of New Union Gold Mine,” Proc. Int. Mine Water Conf. Pretoria, South Africa, 2009.

[17]. S. Saraswat, and J.P. Rai, “Phytoextraction Potential of Six Plant Species in Multimetal Contaminated Soil,” Chemist. Ecol, vol. 25, no.1, pp. 1-11, Feb. 2009.

[18]. K. Nelushi, J. R. Gumbo, and F. A. Dacosta, “An Investigation of the Bioaccumulation of Chromium and Uranium Metals by Cynodon dactylon: A Case Study of Abandoned New Union Gold Mine Tailings, Limpopo, South Africa,” Afr. J. Biotechnol, vol. 12, no. 46, pp. 6517-6525, Nov. 2013.

[19]. G. Mathubala, R. Kalpana Devi, and P. Ramar, “Biosorption of Thymol Blue from Industrial Wastewater Using Activated Biocarbon from Cynodon dactylon Plant Leaves,” Int. J. Chem Tech. Res, vol.7, pp. 2894-2901, 2015.

[20]. L.S, Oliveira, A.S. Franca, T.M. Alves, and S.D.Rocha, “Evaluation of Untreated Coffee Husks as Potential Biosorbents for Treatment of Dye Contaminated Waters,” J. Hazard. Mater, vol.155, no.3, pp. 507-512, Jul. 2008.

[21]. S. Supraja, S. M. Ali, N. Chakravarthy, A. Jaya Prakash Priya, E. Sagadevan, M.K. Kasinathan, S. Sindhu, and P.Arumugam, “Green Synthesis of Silver Nanoparticles from Cynodon dactylon Leaf Extract,”Int. J. Chem. Tech, vol 5, no.1, pp. 271-277, Jan. 2013.

[22]. K. Govindaraju, S. Tamilselvan, V. Kiruthiga, and G. Singaravelu, “Biogenic Silver Nanoparticles by Solanum torvum and their Promising Antimicrobial Activity,” J. Biopesticides, vol. 3, no. 1, pp. 394–399, 2010.

[23]. G. Thirumurugan, and M.D. Dhanaraju, “Novel Biogenic Metal Nanoparticles for Pharmaceutical Applications,” Adv. Sci. Lett, vol. 4, no. 2, pp. 339–348, 2011.

[24]. A. de Jesús Ruíz-Baltazar, S.Y. Reyes-López, D. Larrañaga, M. Estévez, and R. Pérez, “Green Synthesis of Silver Nanoparticles using a Melissa officinalis Leaf Extract with Antibacterial Properties’, Results. Phys, vol 7, pp. 2639–2643, Jan 2017.

[25]. R. Shanmuganathan, D. MubarakAli, D. Prabakar, H. Muthukumar, N. Thajuddin, S.S. Kumar, and A. Pugazhendhi, “An Enhancement of Antimicrobial Efficacy of Biogenic and Ceftriaxone-Conjugated Silver Nanoparticles: Green Approach,” Environ. Sci. Pollut. Res, vol. 25 no.11, pp. 10362-10370, Apr. 2018.

[26]. S. Pirtarighat, M. Ghannadnia, and S. Baghshahi, “Green Synthesis of Silver Nanoparticles using the Plant Extract of Salvia spinosa grown In vitro and their Antibacterial activity assessment,” J. Nanostructure. Chem, vol.9, no.1, pp. 1-9, Mar. 2019.

[27]. A. M. Priya, R. K. Selvan, B. Senthilkumar, M. K. Satheeshkumar, and C. Sanjeeviraja, “Synthesis and Characterization of $CdWO_4$ nanocrystals,” Ceram. Int, vol. 37, no. 7, pp. 2485–2488,Sep. 2011.

[28]. M. Vanaja, K. Paulkumar, M. Baburaja, S. Rajeshkumar, G. Gnanajobitha, C. Malarkodi, M. Sivakavinesan, and G. Annadurai, “Degradation of Methylene Blue using Biologically Synthesized Silver Nanoparticles,” Bioinorg. Chem. Appl, 2014, http://dx.doi.org/10.1155/2014/742346.

[29]. S. Bhakya, S. Muthukrishnan, M. Sukumaran, M. Muthukumar, S.T. Kumar, and M.V. Rao, “Catalytic Degradation of Organic Dyes using Synthesized Silver Nanoparticles: A Green Approach,” J. Bioremediat. Biodegrad, vol.6, no.5, pp.1, Jul. 2015.

[30]. T. Singh, K. Jyoti, A. Patnaik, R. Chauhan, and N. Kumar, “Application of Silver Nanoparticles Synthesized from Raphanus sativus for Catalytic Degradation of Organic Dyes,” In MATEC Web of Conferences, EDP Sciences, vol. 57, p. 05003, 2016.