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Research article

Synthesis, characterization and impact of cadmium sulfide nanoparticles on the growth, pigment content and anti-oxidative defence system of *Pistia stratiotes*

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ABSTRACT

Cadmium Sulfide nanoparticles (CdS NPs) were synthesised and their characteristics were evaluated using XRD (X-ray diffraction), FTIR (Fourier transform infrared), FESEM (Field emission scanning electron microscopy), EDAX (Energy Dispersive X-ray Analysis), and UV-Vis (Ultraviolet-visible). Further, the impacts of CdS NPs on the growth, pigment content and antioxidative defence system of *Pistia stratiotes* were studied. The exposure of CdS NPs to *Pistia stratiotes* caused the negative repercussions, which were tested using a variety of biological tests. The findings demonstrated that CdS NPs were hazardous to the *Pistia* plant at high concentrations, while they were easy to grow at lower concentrations up to 30 ml/l. Both growth and colour characteristics declined at 40 mg/l concentration of CdS NPs. Antioxidant enzymes' activity like superoxide dismutase (SOD)**,** ascorbate peroxidase (APX) and catalase (CAT) also decreased at 40 ml/l on 60 days. The Protein and photosynthetic pigments were declined on 60 days at 40ml/l concentration however, the level of both the parameters was found increased in comparison with control. The results demonstrated that the higher concentrations of CdS NPs are toxic to *Pistia*, resulting in decreased growth and significant alterations in antioxidant enzyme activity. These findings suggest that *Pistia* can be used as a bio-indicator for CdS NPs contaminated water.

Keywords: Cadmium sulfide, Antioxidant Enzymes, Protein, Growth, Water contamination, Photosynthetic pigments.

INTRODUCTION

With the adverse effects on human and environmental health, heavy metal pollution has emerged as a significant environmental issue due to global industrialization. Heavy metal pollution is most widespread in most countries [1, 2]. Global heavy metal emissions have reached 22,000 metric tonnes for cadmium (Cd), 939 Metric Tonnes each year in the last few decades [3]. Metal pollution in soil, especially Cd, has been linked to causing adverse effects on plant growth and yields and nutrient uptake in the plants[4]. Cd enters the food chain through crops and harms and ultimately adversely affect to the human health [4]. Due to the mutagenic nature of Cd, it disrupts DNA and may be carcinogenic human beings[5]. Uncontrolled shaking, muscle atrophy, partial blindness, and abnormalities in children are some of the neurological problems that can arise from chronic exposure to Cd [6] . Around the world, many Phytoremediation procedures have been used to clean up polluted soils and water [7,8]. The national toxicology program (NTP) classifies Cd as a hazardous, non-essential transition metal and human carcinogen [9-13]. Cd exposure has been shown to have harmful effects on biochemical and neurobehavioral parameters and glucose metabolism. Since they have a wide bandgap and are effectively

Group II VI semiconductors, CdS NPs are employed to increase the efficiency of solar cells and other devices [14]. High photosensitivity is also provided by this feature. The efficiency of LED solar cells is increased by their capacity to detect visible light, and they perform effectively as photoconductors [15-17].

In the present study, we employed *Pistia stratiotes L*, aquatic floating macrophytes, as a model to examine how different doses of CdS NPs affect its growth and biochemical parameters, including antioxidant compounds.

MATERIAL AND METHOD

Synthesis of CdS NPs

To synthesize CdS NPs, 0.1 moles of $CdCl_2$ and 0.1 moles of Na_2S were each dissolved separately in 25 ml of deionized water. Total dissolution was then completed after continuous stirring. Later, the reaction was carried out over a range of temperatures (20 to 80°C) by adding Na_2S dropwise to the $CdCl_2$ solution. The light yellow precursor solution gradually changed to a dark yellow colour. The resulting chemical reaction is as follows.

$$CdCl_2 + Na_2S \longrightarrow CdS + 2NaCl \quad \ldots\ldots \quad (i)$$

The finished product was centrifuged five times to remove an additional contaminant before being washed with ethanol five times to produce high purity nanoparticles. The Cd nanoparticles were collected and dried for 30 minutes at 50 degrees Celsius before being used for characterization.

Characterization of CdS NPs

The purity of phase and crystalline size of the prepared CdS NPs was determined using an X-ray diffractometer (XRD, Bruker D8 advance) with monochromatized Cu–Kα radiation (λ=1.5418 Å) at operating accelerating voltage and the applied current of 40 kV and 40 mA, respectively.Using the Debye-Scherer formula, an average crystallite size (D) was calculated (Scherer 1918).

$$D = \frac{K\lambda}{\beta \cos\theta} \ldots\ldots (1)$$

Whereas the lattice parameter for a cubic unit cell was calculated by.

$$\frac{1}{d^2} = \frac{h^2+k^2+l^2}{a^2} \ldots\ldots \quad (2)$$

The density of X-ray (d_x) is evaluated using the relation

$$\text{X-ray density} = \frac{ZM}{Na^3} \ldots\ldots \quad (3)$$

Where, *D* is the average crystalline, K is a dimensionless shape factor (0.94), λ is the X-ray wavelength, β is the FWHM, θ is the Bragg's angle, *d* is the crystal's lattice parameter, atomic lattice spacing, miller indices h, k, and l, andZ is the number of unit cell M is the molecular weight of the sample, N is the Avogadro number.

Further the existence of the functional group in a sample was identified by the Fourier transforms infrared spectroscopy (FTIR, PerkinElmer, UK) techniques, whereas the Field emission scanning electron microscopy (FESEM, Cal Zeiss supra 55) was used for the estimation of the grain size and the surface morphology of the derived sample. Compositional Elemental analysis was done using EDAX. The optical characteristics in terms of photo-energy absorption behavior and bandgap measurement were determined with a UV visible spectrometer (Perkin Elmer) in the range of 200–800 nm using $BaSO_4$ as a reference.

Pot experiment setup

The saplings of fresh *Pistia* were collected from the Ganga River Patna, Bihar. The plants were washed gently with distilled water to remove sediments. The saplings were kept in clean acid-washed plastic tubs filled with tap water for five days. The tubs were filled with 5 litres distilled water having different concentrations of CdS nanoparticles. The concentrations of CdS used were 0, 10, 20, 30 and 40 ml/l. The experiment was performed with three replicates and the tubs kept in a naturally illuminated net-house.

Evaluation of chlorophyll content and carotenoids

The 0.5 gram of fresh plant leaves were crushed with 80% acetone at room temperature using a mortar and pestle. Then it was centrifuged at 4500 RPM for 10 minutes, and the final reading was taken using a UV spectrophotometer. The different pigment absorbance was measured with a spectrophotometer, and chlorophyll a and b were calculated. The absorbance was measured at 663, 645 and470 according to (Lichtenthaler, 1987 and 2001) [18,19].

$$\text{Chlorophyll a} = (19.3\times A663 - 0.86\times A645)\ V/100W$$

$$\text{Chlorophyll b} = (19.3\times A645 - 3.6\times A663)\ V/100W$$

$$\text{Carotenoides} = 100\ (A470) - 3.27\ (\text{mg chl. a}) - 104\ (\text{mg chl. b})/227$$

$$\text{Total chlorophyll} = \text{Chlorophyll a} + \text{Chlorophyll b}$$

Where:

V	=	Volume of filtrate (upper solution of centrifuges)
A	=	Absorption of light at wavelengths of 663, 645 and 470 nm
W	=	Sample fresh weight in grams

Assessing the activity of antioxidant enzymes

Estimation of Ascorbate Peroxidase (APX)

Ascorbate peroxidase activity was determined according to the method of (Chen and Asada,1989). [20]. The reaction mixture included the enzyme fraction, 50 mM potassium phosphate (pH 7.0), 1 mM NaN_3, 0.5 m Mascorbate, and 1.54 mM H_2O_2. Ascorbate began to oxidise when exposed to H_2O_2, and a decrease in absorbance at 290 nm was observed. Utilizing the reduced ascorbate's extinction coefficient, the enzyme activity of APX was estimated (2.8 $mM^{-1}cm^{-1}$). Using a spectrophotometer (Shimadzu UV/VIS 1800) track the reduction in absorbance at 290 nm.

Estimation of Super-Oxide Dismutase (SOD)

The activity of SOD was determined as described by (Chen and Pan, 1996) [21]. In a 3mL reaction mixture, 50 mol sodium phosphate buffer (pH 7.0), 10 mol methionine, 1.17 mol riboflavin, 2 mM EDTA, 50 mM Na_2CO_3, 56 mol NBT, and 100 l test extract were combined. A spectrophotometric graph range rise in absorbance at 560 nm was used to determine the blue formazan formed by NBT photo-reduction. The total enzyme required to prevent 50% of NBT photo-reduction was defined as one SOD unit Shimadzu UV/VIS 1800, 560 nm absorbance measurement.

Estimation of Catalase (CAT)

The absence of H_2O_2 was used to test the Catalase. The 3 mL reaction mixture contained was composed of 70 µlof diluted 30 % H_2O_2 in 2.910 mL of 0.1M phosphate buffer (pH 7.4) with or without 25 µl of light test extract. Absorbance at 240 nm (Shimadzu UV/VIS 1800) is measured. The drop in absorbance at 240nm was measured for 3 minutes, and the amount of H_2O_2 decomposed was used to calculate enzyme activity [22-23].

Protein Activity

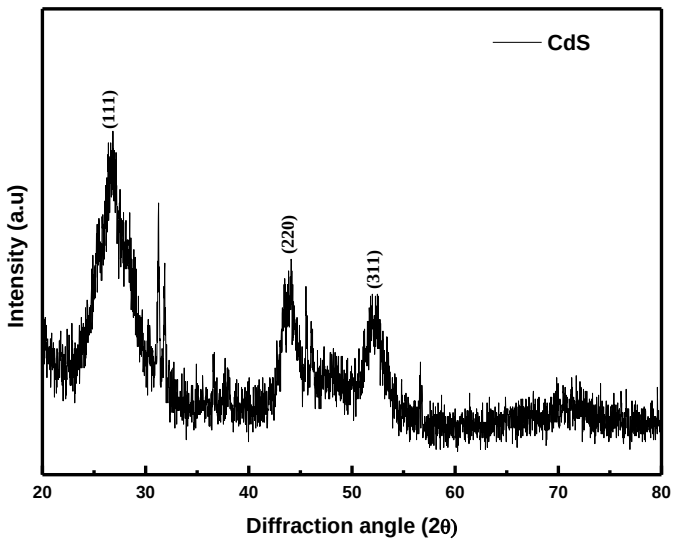
Bovine serum albumin was used as a protein standard by (Lowry et al., 1951) [24] to evaluate the enzyme extracts total soluble protein concentration. The suspension was pipette into spectrophotometer cuvettes, and the absorbance was measured at 595 nm.

RESULT AND DISCUSSION

Structural Analysis

Figure 1 shows the X-ray diffraction patterns of pure CdS nanoparticles. The diffraction patterns of pure CdS show well defined crystalline peaks at 2θ diffraction values ranging from 20-80°. The observed reflections correspond to (111), (220), and (311) planes of cubic structure with space group 'F43m'.

Figure 1: XRD pattern of CdS nanoparticles



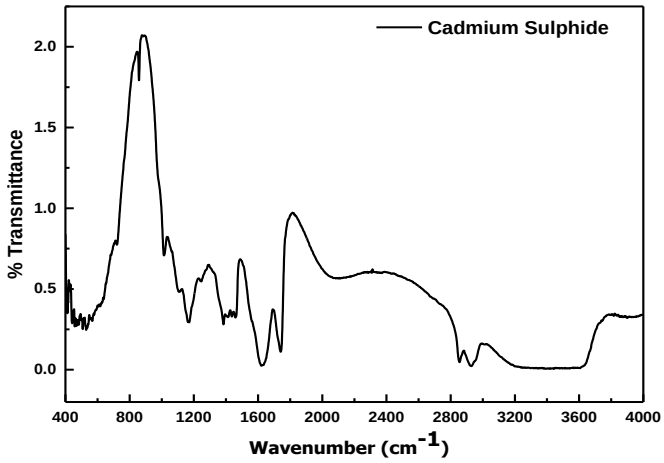
The observed peaks are in good agreement with the reported data and JCPDS card No: 80-0019. The Debye Scherer formula (Equation 1) was used to calculate the average crystallite size (D) of pure CdS. (Scherer 1918)whereas the lattice parameter was calculated by using the equation 2.An estimated average crystallite size was approximately 18 nm, i.e. close to the particle size estimated by FE-SEM, and average lattice parameter a = 0.5799 Å reproduces the hkl values within a standard deviation ± 0.0003 nm, density (ρ) = 5.047 g/cm^3 (Volume = 195.01 Å^3) i.e, close to bulk CdS, a = 0.5812 nm, ρ = 4.87 g/cm^3 (Volume = 196.93Å^3).

It clearly indicates that the CdS particles are agglomerated and in turn form clusters with compact crystalline nature. The CdS nanoparticles are spherical in shape and the distribution of the particles sizes are estimated and presented through histogram in Figure. 2 (b). It is estimated to be in the range of 15-20 nm. Further, the EDS mapping was done on the selected region of CdS sample (as shown in Figure. 2c) for the elemental analysis and the mapping was presented in Figure. 2 (d). The EDS spectrum shows the presence of Cd and S elements with some impurity elements like Cl and Na. The Cl and Na were detected as it was available in the raw materials and did not wash out. The source of C element in the spectrum is because of the CdS sample was placed on the carbon tape for the EDS mapping and oxygen due to surface adsorption. The Cd and S peaks in the EDS spectra confirms the formation of the CdS compound.

Analysis of Functional Group

The FTIR spectrum of CdS nanoparticles produced is shown in Figure 3.

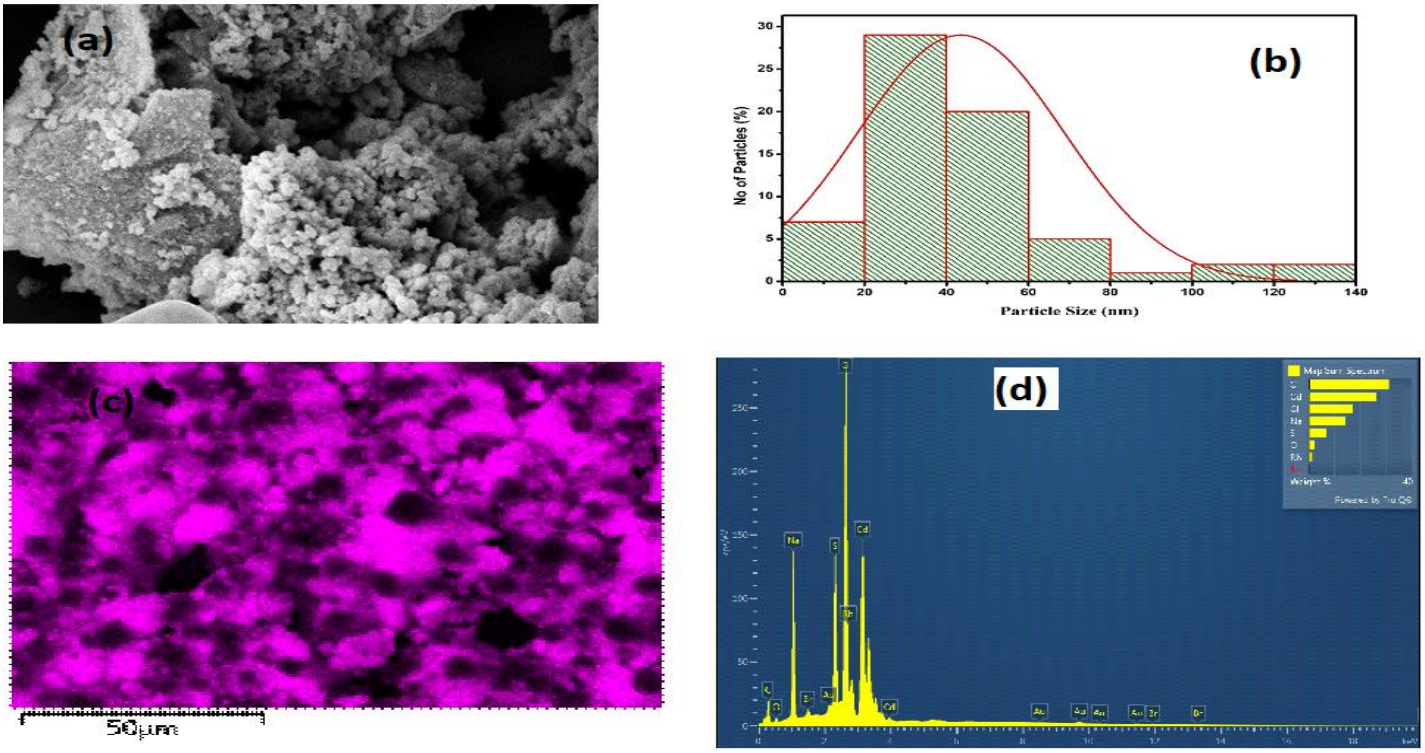
Figure 3: FTIR Spectra for CdS nanoparticles



It is a technique which is used to detect the different functional groups exist in the synthesized sample and their molecular structure. In present studies, the developed sample was scanned from 450 to 400 cm^{-1}. The characteristic vibrational frequencies of the functional groups have been presented in Figure 3. The large absorption peak at 3408 cm^{-1} was attributed to the O-H stretching vibration of water molecules caused by surface moisture adsorption. The asymmetric and symmetric bands around 2931 cm^{-1} gets associated with C-H stretching whereas the absorption peak at 1751 cm^{-1} was attributed to C=C stretching vibration. The weak peaks observed at 1381 and 1623 cm^{-1}, were possibly due to stretching

vibrations of sulphate group and also absorption at 864 and 1011 cm^{-1} was assigned to SO^{4-}. In addition, a strong absorption band at 613 cm^{-1} was assigned to stretching vibration of Cd-S [25], growth study of CdS thin films. Which supports the formation of CdS with less impurities.

Figure 2: (a) FE-SEM image of CdS nanoparticles, (b) Size distribution of histogram of CdS nanoparticles (c) region for EDS mapping and (d) the corresponding EDS spectrum of CdS



Optical Properties and Energy Band Gap Evaluation

For the purpose of determining the bandgap of the generated sample, the UV-Visible spectra of freshly synthesised CdS nanoparticles were measured in the 350–600 nm region. The recorded spectrum of CdS nanoparticles is presented in Figure4. Optical absorption spectrum of CDs nanoparticles exhibits an absorption peak at 425 nm in comparison to 515 nm for bulk CdS material. The absorption peak shows the blue shift behaviour relative to the bulk CdS due to increased binding energy of the exciting. A tauc relation was used to determine the bandgap of the CdS nanoparticles [26].

$$\alpha h\nu = (h\nu - \mathrm{Eg})^{n}$$

Where A is a constant, Eg is a measure of a material's band gap, and the exponent n is based on the nature of the transition. The $(\alpha h\nu)^2$vs h plot is used to calculate the band gap, and the band gap was determined by extrapolating the straight line on the graph to the hν axis. The $(\alpha h\nu)^2$ vs. plot in Figure. 4 was used to determine the band gap values for the sample. It is discovered that the sample's band gap value is 2.25, this value is lower than the bulk value of 2.36 eV and may be due to a size quantization effect in the sample. The sample's optical band gap shifts as a result of the reduction in particle size [27].

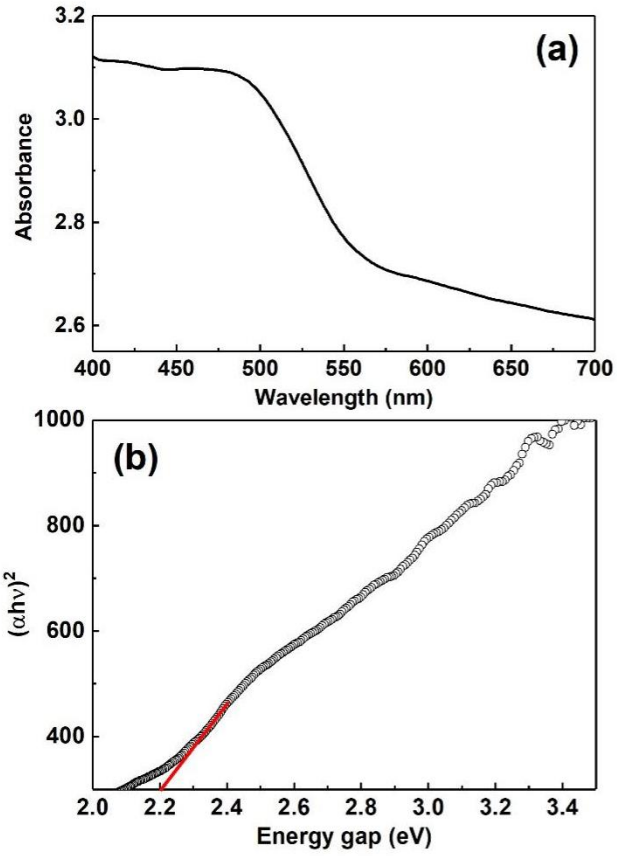


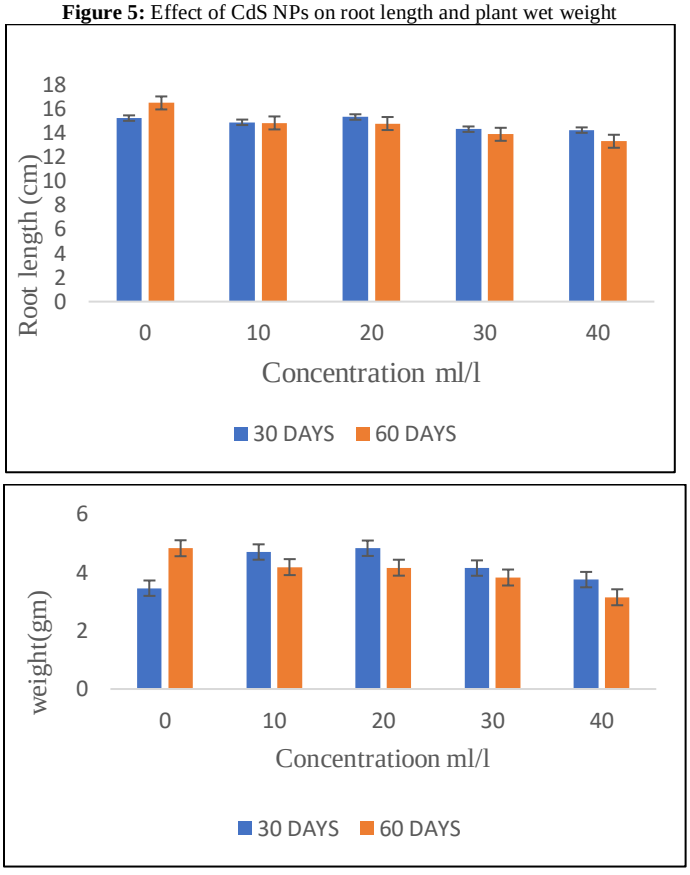
Figure 4: (a). UV-Visible spectra and (b) Energy band calculation of CdS

Plants growth parameters

Effect on Growth Parameters

In some studies, the phytotoxicity of cadmium, which alters morphological, physiological, and biochemical characteristics, is well documented[28]. According to the current study, *Pistia stratiotes* root and shoot biomass decreased when compared to control, with a maximum loss occurring at 40 mg L^{-1} Cd. The effect of CdS NPs on *Pistia stratiotes* were analysed measuring plant fresh weight (gm). The accumulation of Cds caused a major decline in the rates of plant weight and an increased fall in the roots with increasing concentration. The fresh weight declining at 40ml/l on 60 days and root length decreases at 30ml/ l in 0 and 60 days. It was observed that the roots were separated from *Pistia* and leaves started turning brown at 40 ml/l on 60th day completely.

Figure 5: Effect of CdS NPs on root length and plant wet weight



Effect on Chlorophyll and Carotenoids of *Pistia*

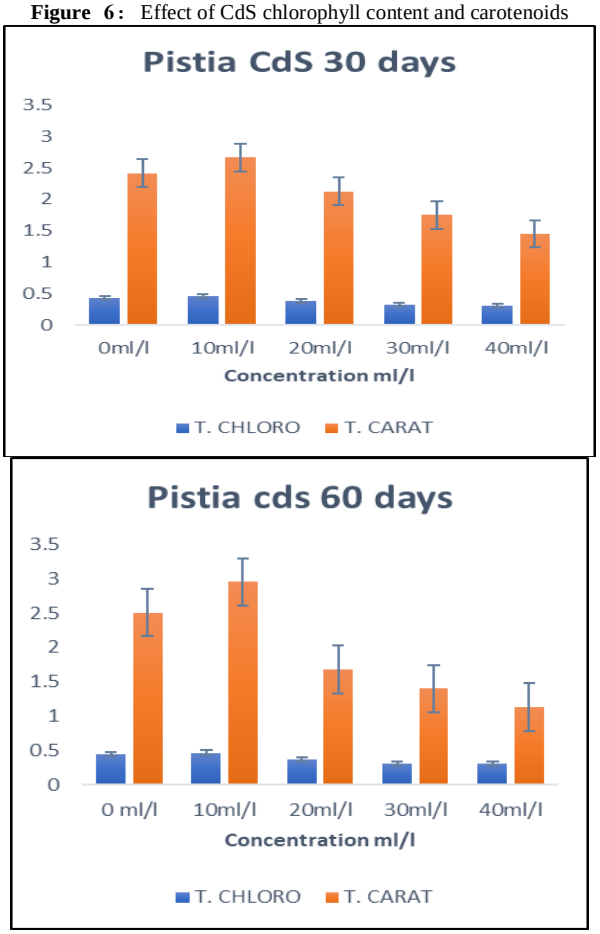
CdS concentration had a negative impact on the photosynthetic pigments in terms of total chlorophyll, and total carotenoids levels. At 40 ml/l on 60 days concentration of CdS NPs decreased the total chlorophyll and total carotenoids content. Due this the wilting and dropping of leaves were observed at this level. It also shows yellow pigmentation at this level.

Effect of Protein and Antioxidants on *Pistia*

The first enzyme in the detoxifying process, superoxide dismutase, transforms O_2 radicals to H_2O_2 at a very quick rate [29]. According to research by [30], cadmium can cause oxidative stress by either increasing the generation of oxygen free radicals or by lowering antioxidant concentrations, both enzymatic and non-enzymatic [31]. The outcomes revealed that the Leaf SOD activity following 100 M Cd exposure whole treatment as well as the root canals on the 15th and 20th days was observed to be considerably higher. But during the CAT observation its shows peroxide liberated in peroxisome following the glycolate conversion during Photorespiration [32]. The analysis of antioxidants was important because it made the slow damage of cells which were caused by free radicles [33]. It was important to do protein analysis because Plant proteins serve a variety of enzymatic, structural, and functional functions (biosynthesis, immunity, photosynthesis, transport etc.) They also serve as storage mediums to meet the nutritional and growth requirements of developing seedlings.

Total protein content in *Pistia* dropped when exposed to CdS NPs, especially at higher concentrations. SOD and APX activity both showed some favourable effects, although they were not dose-dependent on *Pistia*. The *Pistia* aquatic plant's growth (physical qualities) and the degree of stress that leads to increased antioxidant activity must be taken into consideration. An increasing effect was observed in catalase activity with increasing concentration this activity also increasing. There was a noticeable increase in SOD and APX activity in almost after 60 days, after repetitive experiments with same process.

Figure 6: Effect of CdS chlorophyll content and carotenoids



However, there is a decline in protein content in all the groups after 60 days, which indicate stress-induced inhibition of protein synthesis. Necessary co-relation has to be made considering the *Pistia* plant growth (physical characteristics) with the amount of stress resulting in enhanced antioxidant activity [34,35].

Figure 7: Superoxide dismutase (SOD), Ascorbate Peroxidase (APX) and Catalase (CAT) activities on *Pistia* influenced by increasing concentrations of CdS NPs for 30 days.

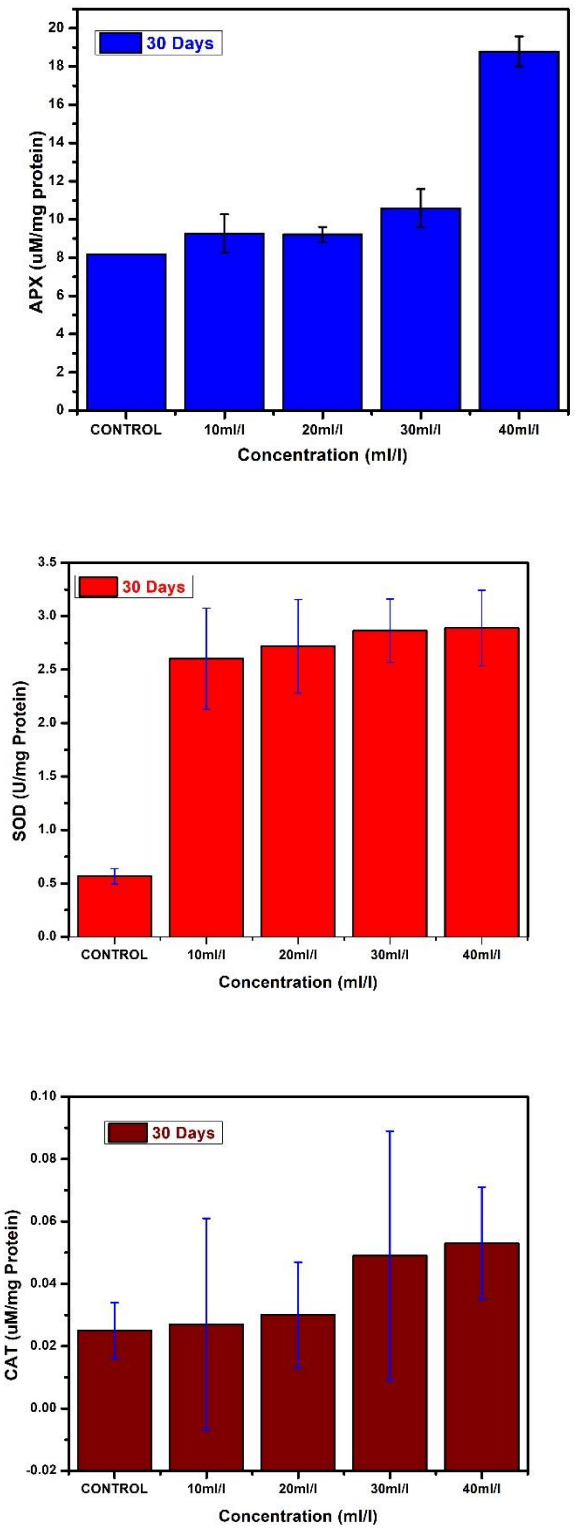


Figure 8 : Superoxide dismutase (SOD), Ascorbate Peroxidase (APX) and Catalase (CAT) activities on Pistia influenced by increasing concentrations of CdS NPs for 60 days

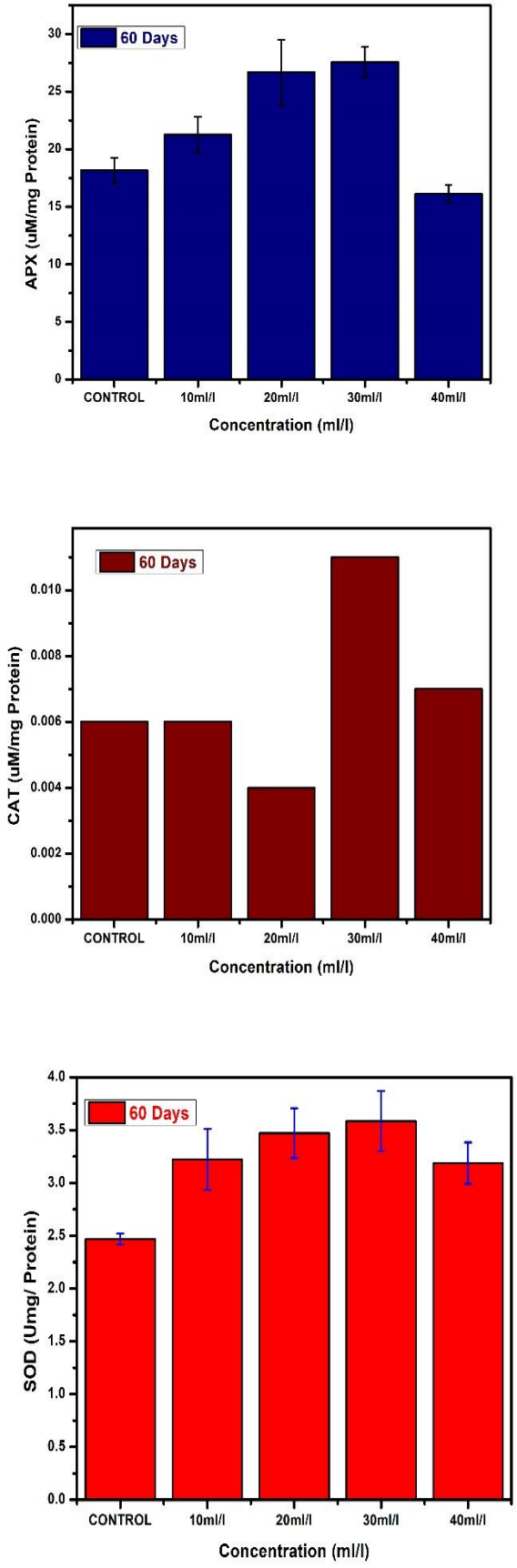
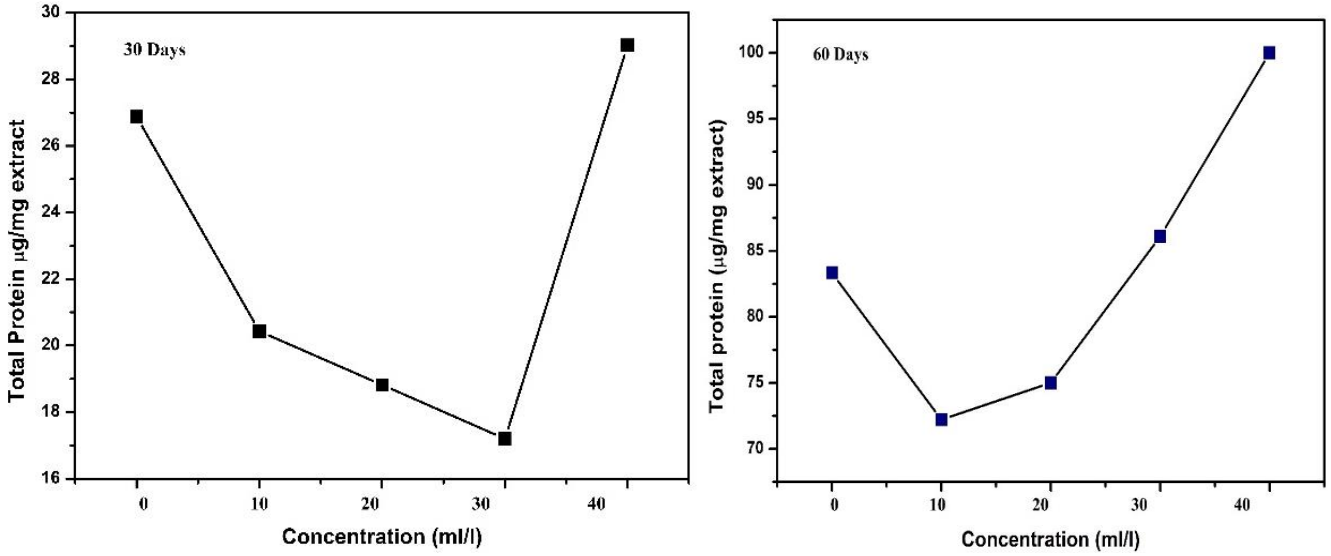


Figure : 9 Effect on protein after 30 and 60 days



In the present work it was observed that the declination in plants root weight and number of leaves during 60 days higher at 40ml/l. Loss of Chlorophyll loss was observed from 60 days at 20ml/l. On 30 days treatment of CdS of 40 ml/l, the highest SOD as 2.889 (uM /mg) and on 60 days 12.650 (uM /mg) highest range was observed at 30 ml/l of concentration. Highest APX was observed 18.788 (uM /mg) at 10ml/l on 30 days and 27.565 (uM /mg) was observed at 30 ml/l on 30 days. No Catalase activities were observed during 30 and 60 days.

As a result, this plant can be suggested for use in phytoremediation due to its abundance presence, ability to grow well without, rapid growth, and short harvest period.

CONCLUSION

It is revealed that application of higher levels of CdS results in decrease in growth, chlorophyll and protein content in the plants. Photosynthetic loss was observed on 60 days at a concentration of 40 ml/l. The application of CdS enhanced the levels of antioxidative defence system which suggests *P. stratiotes* suitability in phytoremediation of CdS from the contaminated water. Its cultivation is easier than other submerged water plant species due to its excellent nutrient removal and uptake capabilities, eco-friendly nature, and rapid growth rate with enormous biomass. Due to its smaller size, it is easy to manage and use in the Phytoremediation process.

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Conflicts of interest

The authors declare no conflict of interest

REFERENCES

1. Batley, Graeme; McLaughlin, Michael. Fate of manufactured nanomaterials in the Australian environment. CSIRO; 2010-04. procite:e4ed7385-6782-402d-ae50-e168cc7f21dd. Doi: 10.4225 /08/58767386e142d.

2. Purkayastha, S.P., Choudhury, M., Deb, D., Paul, C., 2015. Arsenic contamination in ground water is a serious threat in the North Karimganj block of Karimganj district, Southern part of Assam, India. Journal of Chemical and Pharmaceutical Research, 7(8), Pages - 371-378.

3. Shi, J.P., Evans, D.E., Khan, A.A., Harrison, R.M., 2001. Sources and concentration of nanoparticles (< 10 nm diameter) in the urban atmosphere. Atmospheric environment, 35(7), Pages - 1193-1202. Doi: 10.1016/S1352-2310(00)00418-0.

4. Fischer, H.C., Chan, W.C., 2007. Nanotoxicity: the growing need for in vivo study. Current opinion in biotechnology, 18(6), Pages - 565-571. Doi: 10.1016/j.copbio.2007.11.008.

5. Zhao, C.Q., Young, M.R., Diwan, B.A., et al, 1997. Association of arsenic-induced malignant transformation with DNA hypomethylation and aberrant gene expression. Proceedings of the National Academy of Sciences, 94(20), Pages - 10907-10912. Doi: 10.1073/pnas.94.20.10907.

6. Waalkes, M.P., 2000. Cadmium carcinogenesis in review. Journal of inorganic biochemistry, 79(1-4), Pages - 241-244. Doi: 10.1016/S0162-0134(00)00009-X.

7. Khataee, A., Movafeghi, A., Nazari, F., et al, 2014. The toxic effects of L-cysteine-capped cadmium sulfide nanoparticles on the aquatic plant Spirodelapolyrrhiza. Journal of nanoparticle res., 16(12), Pages - 1-10. Doi: 10.1007/s11051-014-2774-7.

8. Dubey, A., Goswami, M., Yadav, K. et al, 2015. Oxidative stress and nano-toxicity induced by TiO2 and ZnO on WAG cell line. PloS one, 10(5), p.e0127493. Doi: 10.1371/journal.pone. 0127493.

9. Gao, Y., Zhang, X., Yin, J., et al, 2019. Castanopsislamontii water extract shows potential in suppressing pathogens, lipopolysaccharide-induced inflammation and oxidative stress-induced cell injury. Molecules, 24(2), Pages – 273-280. Doi: 10.3390/molecules24020273.

10. Siddiqi, K.S., Husen, A., 2017. Plant response to engineered metal oxide nanoparticles. Nanoscale research letters, 12(1), Pages - 1-18. Doi: 10.1186/s11671-017-1861-y.

11. Wilbur, S.B., Hansen, H., Pohl, H., et al, 2004. Using the ATSDR guidance manual for the assessment of joint toxic action of chemical mixtures. Environmental Toxicology and

Pharmacology, 18(3), Pages - 223-230. Doi: 10.1016/j.etap.2003 .03.001.

12. Favero, P.P., Souza-Parise, M.D., Fernandez, J.L.R., et al, 2006. Surface properties of CdS nanoparticles. Brazilian Journal of Physics, 36, Pages - 1032-1034. Doi: 10.1590/S0103-9733200 6000600062.

13. Kouadria, M., djemli, S., Tahraoui, A., 2019. The protective effect of Zinc and Magnesium against subchronic Cadmium toxicity in Wistar rats (Biochemical and neurobehavioral effects). mercury, 3, Pages – 4 - 8.

14. Gawande, M.B., Goswami, A., Felpin, F.X., et al, 2016. Cu and Cu-based nanoparticles: synthesis and applications in catalysis. Chemical reviews, 116(6), Pages - 3722-3811. Doi: 10.1021/acs. chemrev.5b00482.

15. M. Kowshik, N. Deshmukh, W. Vogel, et al, 2002. Microbial synthesis of semiconductor CdS nanoparticles, their characterization, and their use in the fabrication of an ideal diode Biotechnol. Bioeng, 78 5, Pages - 583-588. Doi: 10.1002/bit.10 233.

16. M.F. Kotkata, A.E. Masoud, M.B. Mohamed, E.A, 2009. Mahmoud Synthesis and structural characterization of CdS nanoparticles Phys. E: Low-Dimension. Syst. Nanostruct, 41 (8), Pages - 1457-1465.

17. Lichtenthaler, H.K., 1987. Chlorophylls and carotenoids: pigments of photosynthetic biomembranes. In Methods in enzymology Vol. 148, Pages - 350-382. Academic Press. Doi: Doi: 10.1016/0076-6879(87)48036-1.

18. Lichtenthaler, H.K., Buschmann, C., 2001. Chlorophylls and carotenoids: Measurement and characterization by UV-VIS spectroscopy. Current protocols in food analytical chemistry, 1(1), Pages - F4-3. Doi: 10.1002/0471142913.faf0403s01.

19. Gong-Xiang Chen, KoziAsada, 1989, Ascorbate Peroxidase in Tea Leaves: Occurrence of Two Isozymes and the Differences in Their Enzymatic and Molecular Properties, Plant and Cell Physiology, Volume 30(7), Pages 987–998. Doi: 10.1093/oxford journals.pcp.a077844.

20. Chen, C.N., Pan, S.M., 1996. Assay of superoxide dismutase activity by combining electrophoresis and densitometry. Botanical Bulletin of Academia Sinica, 37.

21. Aebi, H., 1984. Catalase in vitro. In Methods in enzymology Vol. 105, Pages - 121-126. Academic press. Doi: 10.1016/S00 76-6879(84)05016-3.

22. Matsumura, T., Tabayashi, N., Kamagata, Y., et al, 2002. Wheat catalase expressed in transgenic rice can improve tolerance against low temperature stress. PhysiologiaPlantarum, 116(3), Pages - 317-327. Doi: 10.1034/j.1399-3054.2002.1160306.x.

23. Lowry, O. H., Rosebrough, N. J., Farr, A. L., et al, 1951. Proteinmeasurement with the Folin phenol reagent. Journal of Biological Chemistry, 193(1), 265-275. Doi: 10.1016/S0021-9258 (19)52451-6.

24. Demirevska Kepova K, Simova Stoilova L, Stoyanova Z P, et al, 2006. Cadmium stress in barley: growth, leaf pigment, and protein composition and detoxification of reactive oxygen species. Journal of plant nutrition, 29(3), Pages - 451- 468. Doi: 10.1080/01904160500524951.

25. Sabah, A., Siddiqi, S.A., Ali, S., 2010. Fabrication and characterization of CdS nanoparticles annealed by using different radiations. World Academy of Science, Engineering and Technology, 70, Pages - 82-89.

26. Beggas, A., Benhaoua, B., Attaf, A., et al, 2016. Growth study of CdS thin films deposited by chemical bath. Optik, 127(20), Pages - 8423-8430. Doi: 10.1016/j.ijleo.2016.06.030.

27. Devi, R.A., Latha, M., Velumani, S., et al, 2015. Synthesis and characterization of cadmium sulfide nanoparticles by chemical precipitation method. Journal of nanoscience and nanotechnology, 15(11), Pages - 8434-8439. Doi: 10.1166/jnn .2015.11472

28. Hussner, A., Heidbuechel, P., Heiligtag, S., 2014. Vegetative overwintering and viable seed production explain the establishment of invasive Pistiastratiotes in the thermally abnormal Erft River (North Rhine-Westphalia, Germany). Aquatic botany, 119, Pages - 28-32. Doi: 10.1016/j.aquabot.20 14.06.011.

29. Qureshi, M.I., Abdin, M.Z., Qadir, S., Iqbal, M., 2007. Lead-induced oxidative stress and metabolic alterations in Cassia angustifoliaVahl. BiologiaPlantarum, 51(1), Pages - 121-128. Doi: 10.1007/s10535-007-0024-x.

30. Hasan SA, Fariduddin Q, Ali B, Hayat S, Ahmad A, 2009. Cadmium: toxicity and tolerance in plants. J Environ Biol 30: Pages - 165–174.

31. Mohan, B.S., Hosetti, B.B., 2006. Phytotoxicity of cadmium on the physiological dynamics of Salvinianatans L. grown in macrophyte ponds. Journal of Environmental Biology, 27(4), Pages - 701-704.

32. Odjegba, V.J., Fasidi, I.O., 2007. Changes in antioxidant enzyme activities in Eichhorniacrassipes (Pontederiaceae) and Pistiastratiotes (Araceae) under heavy metal stress. Revista de biología tropical, 55(3-4), Pages - 815-823.

33. Qureshi, M.I., Abdin, M.Z., Qadir, S., et al, 2007. Lead-induced oxidative stress and metabolic alterations in Cassia angustifoliaVahl. BiologiaPlantarum, 51(1), Pages - 121-128 .Doi: 10.1007/s10535-007-0024-x.

34. Gupta Amit, Kamboj Vaishali, KambojAbhilasha, 2022. Exploring the antioxidant and anti-inflammatory potential of Cucumissativus and Citrus sinensis, 11 (3), Pages - 4947 – 4950. Doi: 10.55522/jmpas.V11I3.2453

35. Hanane Elazzouzi, Nadia Zekri, Touriya Zair,. Total phenolic and flavonoid contents of Artemisia ifranensis J. Didier plant extracts and their antioxidant activity, 11 (1), Pages - 4419 – 4123. Doi: 10.55522/jmpas.V11I1.264.