A Comparative Analysis of Physiological and Biochemical Responses to Low Doses of Cadmium in Two Important Varieties of *Oryza sativa* L. of Odisha, India

Pallavi Jali¹, Anath Bandhu Das², Chimay Pradhan^{1*}

¹P.G. Department of Botany, Utkal University, Vani Vihar, Bhubaneswar - 751004, Odisha, India

²Department of Agricultural Biotechnology, College of Agriculture, Orissa University of Agriculture and Technology , Bhubaneswar -751003 , Odisha , India

^{1*}P.G. Department of Botany, Utkal University, Vani Vihar, Bhubaneswar - 751004, Odisha, India

Coresponding Author

Dr. Chinmay Pradhan Lecturer in Botany, P.G.Department of Botany, Utkal University, Vani Vihar,, Bhubaneswar-751004, Odisha, India Email- chinmay_pr@yahoo.com Ph: 91- 9438676755

Abstract: Cadmium (Cd) is a non-essential heavy metal as well as an important environmental pollutant, which have a significant impact on plant metabolic system. The current study aims at investigating the effect of cadmium on physiological and biochemical processes of rice plants (variety - Khandagiri, Tejaswini). The germination rate of Khandagiri was 87.78 % in 20 μ M, 24.44 % in 400 μ M and that of Tejaswini was 81.11 % in 20 μ M, 18.88 % in 400 μ M. The total chlorophyll content of Khandagiri and Tejaswini at 400 μ M was 201.01 μ g/g , 177.47 μ g/g respectively. Carotenoid content of Khandagiri and Tejaswini at 400 μ M was 113.44 μ g/g , 92.7 μ g/g respectively. Starch content for both Khandagiri and Tejaswini was found to be 13.28 μ g/g , 6.23 μ g/g respectively at 400 μ M. The total sugar content at 400 μ M was 2.99 μ g/g and 1.882 μ g/g for Khandagiri and Tejaswini respectively. The proline content increased upon exposure to 20 μ M to 400 μ M cadmium . The above result revealed that with increased Cd concentration in the medium showed decreased germination rate, photosynthetic activity, total sugar, starch, but there was an increase in the proline content at different time intervals (7-21 days).

Keyword: Cadmium, heavy metal, chlorophyll, carotenoid, proline

1. Introduction

Heavy metals compete with minerals that serve as essential nutrients for uptake by plants and therefore are known to disturb the nutritional value of plants [1] and after that they get absorbed by the plant by roots, hence they get accumulated in various tissues and cell compartments of the plant and disturb the metabolic activities of that plant [2] [3] [4]. Cadmium is a trace element which is not considered to have any biological functions, but is regarded as highly toxic for both plants and animals [5]. Among other heavy metals it is considered as one of the most harmful pollutants, and large amount is discharged into the environment mostly by anthropogenic activities [6]. It enters the environment and disturbs the biogeochemical cycle and generally gets accumulated in soil and sediments . Cd gets absorbed through the roots of the plants and then tends to get transported into the stem and leaf tissues [7]. In plants, Cd gets readily absorbed and translocated at a faster rate, which exerts a strong toxic effect even at relatively low concentrations [8]. Cadmium salts are also used against fungal infections [9] [10]. Cd is regarded as phytotoxic, at high doses it causes plant growth retardation and creates obstacle for plant development [11]. Cadmium efficiently inhibits photosynthesis [12] [13] [14]. It affects stomatal opening in higher plants [15] and an overall retardation of photosynthetic pigments and other processes [16] [17] [13].

Rice is considered as an important crop throughout the world and is a model plant among monocots for biological research due to its small genome size [18] [19] [20]. Cd contaminated rice grains causes major health risk among half of the world's population, as rice is a basic staple food in almost all continents. It induces toxicity in rice plant and causes inhibition of growth and retardation of chlorophyll biosynthesis [21]. Seed germination is generally regarded as a complex physiological process , which is highly sensitive to heavy metal contamination . Cd reduces germination rate and affects growth of seedlings [22] [23]. The present paper discusses the toxic symptoms induced by Cd on germination process, photosynthetic activity, total sugar, starch and proline content of two rice varieties Khandagiri and Tejaswini .

2. Materials and Methods

2.1. Plant Material Collection

Rice seeds (*Oryza sativa* L.) were collected from the Plant Genetics and Breeding department, Orissa University of

Agriculture and Technology (OUAT), Bhubaneswar, Odisha and used as primary explants. Two rice cultivars (Khandagiri and Tejaswini) were used for all experiments in this study.

2.2. Seed Treatment and Germination Assay

Seeds of rice were taken and surface sterilized in 70% ethanol for 5 min followed by treatment with 0.01% HgCl₂ for 10 min and then washed with sterile distilled water thrice [10]. 30 sterilised seeds were kept on moist filter paper and 10 ml of Cadmium chloride monohydrate (CdCl₂.H₂O) solution of different concentrations (20 µM, $50 \,\mu\text{M}$, $100 \,\mu\text{M}$, $200 \,\mu\text{M}$, $400 \mu\text{M}$) were given. Controls were kept on filter paper soaked with 10 ml distilled water. The seeds were kept for four days under dark in a controlled culture room. In the present study, germination was considered when the radicals were longer than 2 mm. Three replicates were carried out for each treatment, each one consisting of 30 bulked seeds. The radicle length was measured after 4 days of germination of seeds . The seedling vigour index [24], Metal tolerance index [25] and Phytotoxicity percent were calculated. The experiments were repeated five times with three replicates in each Cd concentration.

2.3. Extraction and Estimation of Photosynthetic Pigments

Fresh 0.5 gm leaves were taken and thoroughly homogenized in chilled 80% acetone with the help of a morter and pestle. The homogenates were taken and centrifuged at 10,000 rpm for 10 min in the dark as described previously [26]. The supernatant were collected and absorbance at 470nm, 646.8nm and 663.2nm were recorded using UV –Visible double beam spectrophotometer (Perklin Elmer, India). The experiments were repeated three times with three replicates in each concentration of Cd.

2.4. Extraction and Estimation of Total sugar

0.1gm leaves were taken, chopped and were kept in a test tube to which 5ml of 2.5N HCl was added to hydrolyze it by keeping in a boiling water bath for 1 to 1.30 hr. The sample was then cooled to room temperature, neutralized by adding solid sodium carbonate until the effervescence ceases. The volume was made up to 10 ml and centrifuged. The supernatant was collected and 0.5ml of the aliquots were taken for analysis. The volume was made up to 1ml by adding distilled water. 4ml of anthrone reagent was added, heated for 8 min in boiling water bath. Cooled rapidly and absorbance was taken at 630nm as described earlier [27].The experiments were repeated three times with three replicates in each concentration of Cd.

2.5. Extraction and Estimation of Starch

0.1gm leaves were homogenized in hot 80% ethanol in a mortar and pestle. The homogenate was centrifuged to retain the residue. The residue was repeatedly washed with hot 80% acetone . The supernatant was discarded and the residue was dried well over a water bath. 5ml of distilled

water and 6.5 ml of 52% perchloric acid was added and centrifuged for 20 min and the supernatant was collected. 4ml of anthrone reagent was added and the tubes heated for 8 min in a boiling water bath. Cooled rapidly and the absorbance was taken at 630 nm [27] [28]. The experiments were repeated three times with three replicates in each Cd concentration.

2.6 Extraction and Estimation of Proline

Proline content was estimated by [29] . 0.5gm leaf sample were taken and homogenized with sulfosalicylic acid and filtered through Whattsman No-2 filter paper. 2ml of the filterate was taken, 2ml ninhydrin and 2ml glacialacetic acid was added. The mixture was then incubated at 100° C for 1 hr, the reaction was stopped by keeping the test tubes in ice containing chamber. 4ml toluene was added and the mixture was shaken vigorously for 15 - 20 sec. The aqueous toluene layer was separated and warmed up to room temperature. The red colored aliquote was measured at absorbance 520nm. The experiments were repeated three times with three replicates in each concentration of Cd.

3. Results and Discussion

3.1. Toxicity of Cadmium on germination rate and radical length

Germination rate of rice seeds responds differently to increasing concentration of cadmium and extremely reduced when the concentration reaches about 400µM as compared to 20µM (Table.1 and fig.1). The germination percentage was 87.78% (20 μ M) , 24.44% (400 μ M) for Khandagiri and 81.11% (20µM), 18.88% (400 µM) for Tejaswini . The radicle length of Khandagiri decreased with increased cadmium doses 5.17 cm (20µM), 2.1 cm (400 µM). Similar result was seen in case of Tejaswini 4.5cm (20µM), 0.73cm (400µM). Seedling vigour index of Khandagiri was 453.82 and 51.324 for 20µM and 400µM respectively. Metal tolerance index was recorded as 78.69 and 31.963 for 20µM and 400µM respectively in case of Khandagiri and 71.05 and 11.58 for 20µM and 400µM respectively in Tejaswini. Phytotoxicity percent for Khandagiri was 21.31% and 68.037 % at 20µM and 400µM respectively and for Tejaswini was 28.91% and 88.47% at 20µM and 400µM respectively.

Table 1: Effect of Cadmium on seed germination of Oryza sativa L. (Variety- Khandagiri, Tejaswin	ni)
---	-----

Variety	Treatment	Germination Percentage	Radical length (cm)	Seedling Vigour Index	Metal Tolerance Index	%Phytotoxicity
	Control	97.78 ± 0.471	6.57 ± 0.492	642.414	100	0
	20µM	87.78 ± 0.471	5.17 ± 0.249	453.82	78.69	21.31
	50µM	87.77 ± 0.942	5.03 ± 0.124	441.48	76.56	23.44
khandagiri	100µM	72.22 ± 1.247	3.77 ± 0.339	272.269	57.382	42.618
Kilailuagiii	200µM	52.22 ± 0.942	2.53 ± 0.262	132.116	38.508	61.492
	400µM	24.44 ± 0.471	2.1 ± 1.42	51.324	31.963	68.037
	Control	91.11 ± 1.247	6.33 ±0.236	577.03	100	0
	20µM	81.11 ± 0.942	4.5 ± 0.432	364.092	71.05	28.91
	50µM	74.44 ± 1.885	4.3 ± 0.566	320.092	67.89	32.07
Tojocwini	100µM	51.11 ± 1.247	2.77 ± 0.34	141.40	43.68	56.24
rejaswiii	200µM	42.22 ± 1.699	1.87 ± 0.56	78.811	29.47	70.46
	400µM	18.88 ± 0.942	0.73 ± 0.4	13.845	11.58	88.47

*Values in the table are mean \pm SD of 3 replicates.



Figure 1: Effect of Cadmium on seed Germination of Oryza sativa (variety- Khandagiri, Tejaswini)

The seedling vigour index and metal tolerance index decreased with increasing Cd concentration whereas the phytotoxicity percentage increased with increasing Cd concentration. Hence from above all these results we can clearly observe that cadmium has an adverse effect on the germination and growth of rice seedlings upon exposure to increasing Cd toxicity.

3.2. Effect of Cadmium on Chlorophyll and Carotenoid content

The changes in photosynthetic pigments of Khandagiri and Tejaswini is shown in the Table-2a,2b. A very significant decrease of total chlorophyll was noticed with increasing Cd concentrations . The carotenoids also decreased with increasing days of exposure to different levels of Cd toxicity. The total chlorophyll content of Khandagiri at 21 days was 850.23 μ g/g and 201.01 μ g/g at 20 μ M and 400µM respectively. Similarly total chorophylll content of Tejaswini exposed at 21 days was 578.89 µg/g and 177.47 μ g/g at 20 μ M and 400 μ M respectively. Similar trend was seen in case of carotenoids. The carotenoid content of Khandagiri at 21 days interval was 217.59 µg/g and 113.44 $\mu g/g$ for 20 μM and 400 μM respectively. Carotenoids of Tejaswini was seen to follow the similar trend 194.48 μ g/g and 92.7 μ g/g at 20 μ M and 400 μ M respectively. The increasing cadmium concentrations might have disrupted the chloroplasts as a result of which there was a reduction in the photosynthetic pigments [30] of the two rice varieties when exposed to increasing Cd concentrations in different days of treatment.

or or yzu sativa E. (Variety- Khahdagiri, Tejaswini)				
	Treatme	Total chlorophyll (µg/g) f.wt.		
Variety		Days of treatment		
	nı	7	15	21
	Control	1056.75±13.	1125.87±4.	1151.14±14.
	Colluor	52	37	51
	20 µM	953.13±10.9 8	906.3±1.5	850.23±2.89
Khandag	50 µM	851.72±3.99	722.52±4.1	472.68±3.91
iri	100 M	500 47 2 11	493.24±16.	376.63±29.1
	100 µM	399.47±2.11	78	1
	200 µM	493.67±13.7 9	336.3±6.35	229.12±2.29
	400 µM	386.14±0.81	320.46±1.6	201.01±5.1
	Control	1046.35±1.9	1062.73±8.	1113.13±4.9
		3	47	8
	20 µM	768 8+8 83	654.33±8.8	578.89 ± 21.4
		700.0±0.05	2	4
	50 µM	654.79±10.6	527.86±18.	370 1+6 01
Tejaswin i		9	23	579.1±0.01
	100 µM	431.06±2.83	268.05±6.7 3	202.91±2.96
	200 µM	328.9±0.66	292.32±5.8 9	274.08±0.67
	400 µM	216.68±1.22	191.46±6.9 1	177.47±0.77

 Table 2a: Effect of Cadmium on total Chlorophyll content of Orvza sativa L. (Variety- Khandagiri, Tejaswini)

*Values in the table are mean \pm SD of 3 replicates.

Table 2b: Effect of Cadmium on Carotenoid content of Oryza sativa L. (Variety- Khandagiri, Tejaswini)

Variety	Treatment	Carotenoid $(\mu g/g)$ f.wt.			
		Days of treatment			
		7	15	21	
	Control	252.85 ± 4.49	275.12±1.19	277.33±1.94	
	20 µM	232.4±0.28	221.27±24.84	217.59±4.61	
Whan do airi	50 µM	228.95 ± 4.44	201.34±0.98	195.74±3.25	
Knandagiri	100 µM	204.78±3.85	187.26±3.81	179.55±0.85	
	200 µM	189.88±3.63	149.19±0.41	127.18±0.46	
	400 µM	159.49±0.91	147.22±1.53	113.44±0.91	
Tejaswini	Control	259.17±2.73	278.77±1.03	300.56±3.29	
	20 µM	216.4±0.49	212.7±4.93	194.48±3.64	
	50 µM	212.94±1.29	192.16±0.93	175.69±0.74	
	100 µM	171.77±4.57	139.47±0.47	136.11±0.55	
	200 µM	142.51±1.13	127.99±0.07	98.74±5.35	
	400 µM	123 22+1 53	11699+07	92 7+4 95	

*Values in the table are mean \pm SD of 3 replicates

3.2. Effect of Cadmium on Sugar content

The total sugar content of Khandagiri at 21 days interval was 6.85 μ g/g and 2.99 μ g/g for 20 μ M and 400 μ M respectively. Similarly for Tejaswini at 21 days the total sugar content was 5.338 μ g/g and 1.882 μ g/g respectively.The total sugar content of the two rice varieties decreased with increasing concentrations of cadmium stress whereas the control rice varieties showed increased total sugar content (Table.3 and fig.2a, 2b). The total sugar content of the plants solely depends on its photosynthetic activity. As the photosynthetic mechanism was not efficient so we can conclude that the total sugar also reduced with increasing Cd toxicity when exposed to different days interval.

Table 3 : Effect of Cadmium on total sugar con	ntent of
Oryza sativa L. (Variety- Khandagiri, Tejas	wini)

		Total sugar in µg/g f.wt.			
Variety	Treatment	Days of treatment			
		7	15	21	
	Control	10.19 ±	13.02 ±	13.87 ±	
	Control	0.199	0.087	0.021	
	20M	9.29 ±	7.69 ±	$6.85 \pm$	
	20 µM	0.004	0.002	0.014	
	50. uM	7.68 ±	7.42 ±	$6.92 \pm$	
	50 µM	0.003	0.001	0.005	
Khandagiri	100 uM	7.119 ±	6.44 ±	4.61 ±	
	100 μΜ	0.002	0.003	0.005	
	200M	5.27 ±	4.21 ±	3.24 ±	
	200 µM	0.012	0.003	0.015	
	400 µM	$5.24 \pm$	4.6 ± 0.047	$2.99 \pm$	
		0.0145	4.0 ± 0.047	0.017	
	Control	$10.004 \pm$	11.195 ±	13.41 ±	
		0.003	0.027	0.006	
	20 µM	7.77 ±	$7.619 \pm$	$5.338 \pm$	
		0.002	0.0025	0.0015	
	50 µM	6.705 ±	5.016 ±	4.243 ±	
		0.005	0.002	0.112	
Tojogujuj	100 .uM	5.31 ±	3.899 ±	3.285 ±	
Tejaswiiii	100 μΜ	0.014	0.008	0.018	
	200 µM	4 ± 0.0125	3.381 ±	2.311 ±	
		4 ± 0.0133	0.008	0.002	
	400 µM	3.416 ±	2.666 ±	1.882 ±	
		0.01	0.003	0.0205	

*Values in the table are mean \pm SD of 3 replicates



Figure 2a: Effect of Cadmium on Total sugar of Oryza sativa (Variety- Khandagiri)





3.3. Effect of Cadmium on Starch content

The starch content of Khandagiri at 21 days interval was 45.74 μ g/g and 13.28 μ g/g for 20 μ M and 400 μ M respectively. Similarly for Tejaswini at 21 days the total sugar content was 49.75 μ g/g and 6.23 μ g/g for 20 μ M and 400 μ M respectively.The starch content of the two rice varieties decreased with increasing concentrations of cadmium stress whereas the control rice varieties showed increase in starch content (Table.4 and fig. 3a, 3b).

		Starch content in $\mu g/g$ f.wt.		
Variety	Treatment	Days of treatment		
		7	15	21
	Control	65.16 ± 0.01	68.05 ± 0.049	74.59 ± 0.008
	20 µM	58.58 ± 0.05	51.21 ± 0.002	45.74 ± 0.006
Khandagiri	50 µM	51.67 ± 0.003	51.49 ± 0.185	44 ± 0.003
-	100 µM	47.76 ± 0.119	40.36 ± 0.013	36.41 ± 0.0025
	200 µM	36.14 ± 0.003	31.67 ± 0.042	24.86 ± 0.005
	400 µM	20.29 ± 0.049	16.92±0.001	13.28 ± 0.0015
Tejaswini	Control	59.68 ± 0.045	64.23 ± 0.014	72.53 ± 0.002
	20 µM	53.58 ± 0.007	52.49 ± 0.035	49.75 ± 0.003
	50 µM	49.87 ± 0.003	43.53 ± 0.042	38.33 ± 0.006
	100 µM	39.17 ± 0.059	19.05 ± 0.002	15.8 ± 0.013
	200 µM	19.1 ± 0.003	15.65 ± 0.001	11.51 ± 0.001
	400 µM	20.24 ± 0.061	15.73 ± 0.093	6.23 ± 0.006

Table 4: Effect of Cadmium on starch content of Oryza sativa L. (Variety- Khandagiri, Tejaswini)

*values in the table are mean \pm SD of 3 replicates



Figure 3a: Effect of Cadmium on Starch content of Oryza sativa (Variety- Khandagiri)



Figure 3b: Effect of Cadmium on Starch content of Oryza sativa (Variety- Tejaswini)

3.4. Effect of Cadmium on Proline content

The Proline content of Khandagiri at 21 days interval was 9.62 μ g/g and 85.365 μ g/g for 20 μ M and 400 μ M respectively. Similarly for Tejaswini at 21 days the Proline content was 38.62 μ g/g and 101.38 μ g/g respectively. The Proline content of the two rice varieties increased with increasing concentrations of cadmium stress whereas the control rice varieties showed slight increase in proline content but remained less that the treated plants. Proline content of the two varieties is shown in the Table. 5 and

fig. 4a, 4b. Proline accumulation is a general phenomenon in all the stressed plants. As the rice plants were subjected to cadmium stress, estimation of proline was very important to know whether increase of proline content can be a protection mechanism by increasing osmotic compounds in the cell sap.

		<i>Proline content</i> $\mu g/g f.wt$.			
Variety	Treatment	Days of treatment			
		7	15	21	
	Control	2.6 ± 0.0033	3.83 ± 0.0024	4.022 ± 0.0009	
	20 µM	2.8 ± 0.0024	4.16 ± 0.0012	9.62 ± 0.0106	
Khandagiri	50 µM	14.7 ± 0.0179	23.67 ± 0.0165	25.166 ± 0.329	
	100 µM	23.402 ± 0.0156	36.92 ± 0.0115	45.321 ± 0.0033	
	200 µM	37.29 ± 0.0084	56.71 ± 0.0434	66.12 ± 0.0437	
	400 µM	50.858 ± 0.0009	70.987 ± 0.0045	85.365 ± 0.0447	
Tejaswini	Control	11.15 ± 0.0082	20.29 ± 0.0085	30.3 ± 0.0103	
	20 µM	11.18 ± 0.0024	26.72 ± 0.013	38.62 ± 0.031	
	50 µM	18.37 ± 0.003	37.1 ± 0.032	57 ± 0.0009	
	100 µM	30.57 ± 0.024	44.87 ± 0.004	64.67 ± 0.003	
	200 µM	43.91 ± 0.042	66.07 ± 0.083	76.8 ± 0.023	
	400 uM	44.95 ± 0.0046	75.57 ± 0.018	101.38 ± 0.09	

Table 5 : Effect of Cadmium on proline content of Oryza sativa L. (Variety- Khandagiri , Tejaswini)

*values in the table are mean \pm SD of 3 replicates



Figure 4a: Effect of Cadmium on Proline content of Oryza sativa (Variety- Khandagiri)



Figure 4b: Effect of Cadmium on Proline content of Oryza sativa (Variety-Tejaswini)

4. Conclusion

Cadmium at higher concentrations might inhibit the growth of the plant directly by inhibiting the root growth which in return inhibits the uptake of water and other essential mineral elements through roots, resulting in the uptake of cadmium itself and causing several mineral deficiencies. The water content therefore decreases at higher cadmium toxicity. At higher concentrations it is highly toxic to plants. Similar results were reported on the effect of cadmium [31][32]. Results from the germination studies indicated that Khandagiri showed higher resistance to cadmium as compared to Tejaswini . This study also revealed that at higher cadmium concentrations there was a significant decrease in photosynthetic activity, total sugar content, starch content whereas proline accumulation increased with increase in cadmium toxicity. As compared to Tejaswini , Khandagiri showed good resistance effect . This study may help for selection of resistant variety for carrying out further research on the purpose of finding out which concentration of cadmium salts might be useful to control fungal infections.

Acknowledgement

The authors acknowledge the financial support provided by DST- INSPIRE fellowship, UGC-DRS-SAP scheme and PURSE Grant to P.G Department of Botany, Utkal University Bhubaneswar, India.

References

- D.T. Clarkson, U. Luttge, "Mineral nutrition: Divalent cations, transport and compartmentalization", Prog. Bot., (51), pp. 93-112, 1989.
- [2] M.A. Turner, "Effect of cadmium treatment on cadmium and zinc uptake by selected vegetable species", J. Environ. Qual., (2), pp. 118-119, 1997.
- [3] D.A. Thurman, J.C.L. Collins, "Metal tolerance mechanism in higher plants review", In Proceedings of International Conference on Heavy Metals in the Environmental", Heidelberg, CEP Consultan's Edimburg. pp. 298-300, 1983.
- [4] K. Taylor, L.G. Albrigo, C.D. Chase, "Zinc complexation in the phloem of blight affected citrus", J. Am. Soc. Hortic. Sci., (113), pp. 407-411, 1988.
- [5] B. Alloway, "Cadmium Heavy Metals in Soil", John Wiley & Sons, New Yersey, pp. 100-124, 1990.
- [6] J.O. Nriagu, J.M. Pacyna, "Quantitative assessment of worldwide contamination of air, water and soils with trace metals", Nature 333, pp. 134–139, 1988.
- [7] D.E. Salt, R.C. Prince, I.J. Pickering, I. Raskin, " Mechanisms of cadmium mobility and accumulation in indian mustard", Plant Physiol. , (109), pp. 1427-1433, 1995.
- [8] I. V. Seregin, V.B. Ivanov, "The transport of cadmium and lead ions through root tissues", Russ. J. Plant Physiol., (45), pp. 780–785, 1998.
- [9] A.E. Osbourn, "Performed antimicrobial compounds and plants defense against fungal attack", Plant Cell, (8), pp. 1821-1831, 1996.
- [10] B. Mittra, P. Ghosh, S.L. Henry, J. Mishra, T.K. Das, S. Ghosh, C.R. Babu, P. Mohanty, "Novel mode of resistance to *Fusarium* infection by mild dose preexposure of cadmium to wheat", Plant Physiol. Biochem., (42), pp. 781-787, 2004.
- [11] P. Carrier, A. Baryla, M. Havanz, "Cadmium distribution and micro localization in oil seed rape (*Brasica napus*) after long-term growth on cadmium-contaminated soil", Planta , (216), pp. 939-950, 2003.
- [12] M. Greger, M. Johansson, D. Stihi , K. Humza, "Foliar uptake of Cd by pea (*Pisum sativum*) and sugar beet (*Beta vulgaris*)", Physiol. Plant, (88), pp. 563-570, 1994.
- [13] L.K. Chugh, S.K. Sawhney, "Photosynthetic activities of *Pisum sativum* seedlings grown in presence of cadmium", Plant Physiol. Biochem., (37), pp. 297-303, 1999.
- [14] A. Vassilev, A. Perez-Sanz, B. Semane, R. Carteer, J. Vangronsveld, "Cadmium accumulation and tolerance

of two salix genotypes hydrophonically grown in presence of cadmium", J. Plant Nutr., (28), pp.159-2177, 2005.

- [15] C. Poschenrieder, B. Gunse, J. Barcelo, "Influence of cadmium on water relation, stomatal resistance, and absicisic acid content in expanding bean leaves", Plant Physiology, (90), pp. 1365-1371, 1989.
- [16] I.S. Sheoran, H.R. Signal, R. Singh, "Effect of cadmium and nickel on photosynthesis and the enzymes of photosynthetic carbon reduction cycle in pigeon pea (*Cajanus cajan L.*)", Photosynth. Res., (230), pp. 345-351, 1990.
- [17]Z. Krupa, G. Quist, N.P.A. Hurner, "The effect of cadmium on photosynthesis of *Phaseolus vulgaris* – A fluorescence analysis", Physiol. Plant., (88), pp. 626-630, 1993.
- [18] S.A. Goff, D. Ricke, T.H. Lan, G. Presting, R. Wang, M. Dunn, "A draft sequence of the rice genome (*Oryza sativa* L. ssp. japonica)", Science, (296), pp. 92–100, 2002.
- [19] J. Yu, S. Hu, J. Wang, G.K.Wong, S. Li, B. Liu, "A draft sequence of the rice genome (*Oryza sativa* L. ssp. indica)", Science ,(296), pp. 79–92, 2002.
- [20] G.K. Agrawal, R. Rakwal, "Rice proteomics: a cornerstone for cereal food crop proteomes", Mass Spectrom Rev ,(250), PP. 1–53, 2006.
- [21] A.K Stobart, W.T. Griffihs, I. Ameen-Bukhari ,R.P Sherwood, "The effect of Cd²⁺ on the biosynthesis of chlorophyll in leaves of barley", Physiol. Plant.,(63), pp. 293-298, 1985.
- [22] M. Stiborova, M. Ditrichova, A. Brezinova, "Effect of heavy metal ions on growth and biochemical characteristics of photosynthesis of barley and maize seedlings", Biol. Plant., (29), pp. 453–467, 1987.
- [23] N. Rascio, F. Dallavecchia, M. Ferretti, L. Merlo, R. Ghisi, "Some effects of cadmium on maize plants", Arch. Environ. Con.Tox., (25), pp. 244–249, 1993.
- [24] A.A. Abdul Baki, J.D. Anderson, "Vigour determination in soybean seed by multiple criteria", Crop. Sci., (3), pp. 630-633, 1973.
- [25] R.C. Turner, C.Marshal, "Acculmulation of zinc by sub cellular fraction of root *Agrostis tenuis* sibth in relation to zinc tolerance", New Phytol.,(71) ,pp. 671-676, 1972.
- [26] D.I. Arnon, "Copper enzymes in isolated chloroplasts polyphenol oxidase in *Beta vulgaris*", Plant Physiol., (24), pp. 1-15, 1949.
- [27] J.E. Hedge, B.T. Hofreiter , In: Carbohydrate Chemistry, (Eds. Whistler R.L. and Be Miller, J.N.), Academic Press, New York, (17), 1962.
- [28] B.Thayumanavan, S. Sadasivam, Qual Plant Foods Hum Nutr , (34) , pp.253 , 1984.
- [29] I.s. Bates, R.P.Walden, I.D Teare, "Rapid determination of free proline for water-stress studies", Plant and soil, (39), pp. 205-207, 1973.
- [30] J.D. Dodge, G.B. Lawes, "Plastic ultra-structure in some parasitic and semi-parasitic plants" ,Cytobiologie, (9), pp. 1-9, 1974.
- [31] M.C. Kalita, P. Devi, I. Bhattacharya, "Effect of cadmium on seed germination, early seedling growth and chlorophyll content of *Triticum aestivum*", Indian J. Plant Physiol., 36(3), pp. 189-190, 1993.

[32] Sunil kumar, Urmil J.Mehta, Sulekha Hazra, "Accumulation of cadmium in growing peanut (*Arachis hypogea* L.) seedlings- its effect on lipid peroxidation and on the antioxidative enzymes catalase and guaiacol peroxidase", Plant nutr.soil.sci, (171), pp. 440-447, 2008.

Author Profile

Dr. Anath Bandhu Das is working as Associate Professor in Department of Agriculture Biotechnology, College of Agtriculture, Orissa University of Agriculture and Technology, Bhubaneswar, Odisha.

Dr. Chinmay Pradhan is working as Lecturer in Post Graduate Department of Botany, Utkal University, Bhubaneswar, Odisha.

Pallavi Jali is doing Ph.D in Post Graduate Department of Botany, Utkal University, Bhubaneswar, Odisha.