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Potato peel mediated improvement in organic substances of vigna mungo growing under copper stress

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Abstract

Present study was carried out to investigate the potentials of potato peel powder in the improvement of organic substances of *Vigna mungo* growing under copper stress. Copper is a toxic metal which cause adverse effects in plants, when supplied in high concentrations. Increasing concentration of copper in agricultural soil due to industrialization is an alarming situation for all living organisms. Removal of heavy metals by low cost adsorbents is a recent technique which is more efficient with no side effects. Potato peels were collected free of cost from French fries shops, shade dry and ground. Healthy seeds were surface sterilized and sown in pots containing 1kg soil, each pot was supplied with different concentrations of copper i.e., 0,25,50,75,100 ppm. Treated plants were supplied with 6 gms potato peel powder/kg soil along with different concentrations of copper (0,25,50,75, ppm). Experiment was designed in a randomized block fashion. Photosynthetic pigments regarding chlorophyll a, chlorophyll b, total chlorophyll and carotenoids were examined in experimental as well as treated plants and were compared with control plants. Results were also confirmed with the analysis of carbohydrate, protein and DNA contents. Biophysical parameters like % germination, growth with respect to root, shoot length, leaf area, biomass and relative water contents were also pragmatic to corroborate consequences.

Keywords: Potato peel, copper, chlorophyll, carbohydrates, proteins, carotenoids, DNA, *Vigna mungo*

Introduction

Species of *Solanum* like *S. tuberosum* and *S. nigrum* are best hyperaccumulator and detoxifier for various heavy metal such as cadmium (Zhou, Q.X. and Song, Y.F., 2004) [45]. Potato Peel Waste is a good source of dietary fibre: primarily insoluble carbohydrates – cellulose, hemicellulose, lignin, pectin, gums, etc. (Al-Weshahy and Rao, 2013) [2]. Dietary fiber is well known as a bulking agent, increasing the intestinal mobility and hydration of the feces (Forsythe WA, Chenoweth WL, Bennink MR 1976) [13]. Potato Peel contains high quantities of polyphenols which have a role in the defence mechanism against phytopathogens. Therefore almost 50% of phenolics are located in the peel and adjoining tissues and decrease toward the center of the tuber (Friedman, 1997) [14]. It is believed that binding of bile acids is one of the mechanisms whereby certain sources of dietary fibers lower plasma cholesterol (Lazarov K, Werman MJ 1996) [22]. Potato peel also was able to support the economic growth and production of several extracellular hydrolytic enzymes (Mahmood AU, Greenman J, Scragg AH, 1998) [1]. Heavy metal toxicity is one of the major current environmental health problems and potentially dangerous due to bioaccumulation through the food chain and in plant products for human consumption. Michael, (Wu, *et al.* 2012) [42]. Copper in small concentration is an essential micronutrient for all form of life such as growth, physiological process (Sharma, R. and A. Madhulika, 2005) [36]. However, as a result of the formation of organo copper complexes, excess copper can be consider as a toxic elements leading to reduces shoot and root growth by inhibits cell elongation and cell cycle (Ouzounidou, G., 1993) [30], decrease of chlorophyll content, leaf expansion, disturbance of DNA conformation damage chromatin and the plasma causing ion efflux (Singh, D., K. Nath and Y. Sharma, 2007) [38]. Present study was based on the question that whether free of cost and abundantly available potato peel could improve organic substances of *vigna mungo* growing under copper stress?

Materials and Methods

Three sets each with 3 replicates of soil bags were prepared, each with 1 kg of garden soil. First set named control plants was provided with distilled water only, second set named experimental plants was provided with different concentrations of copper (0,25,50,75, & 100ppm Cu) and third set of soil bags was provided with different concentrations of copper along with 6gm of potato peel/kg soil. Seeds of *vigna mungo* were surface sterilized and carefully sown. The plants were harvested after 17 days of germination for biophysical parameters like root/shoot length, leaf area, biomass and relative water content by Jones and

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Turner (1978) [18], Biochemical parameters like chlorophyll content by Arnon (1949) [15] and McKinney (1941) [27] method, Carotenoid by Kirk and Allen method (1965) [20], carbohydrate content by Yemm and Willits (1945) [43] method, protein content by Lowry's method (1951) [23] and DNA concentration by Jayaraman (1992) [17] were determined.

Results and Discussion

Biophysical Parameters

Percent Germination: The percentage germination of *Vigna mungo* seeds was found to be 46.6 ± 2 , 44.4 ± 2.00 , 35.5 ± 1.57 and 33.3 ± 1.6 at all applied levels of copper whereas it was 68.3 ± 3.88 in control. Decrease in percentage germination of *Vigna mungo* may attribute to the increasing concentrations of copper which cause osmotic stress in seed thus abscisic acid lowers the ability of seed germination (Shun-ying, 2008) [37]. Application of potato peel significantly improved the percentage germination of *Vigna mungo* seeds up to 53.3 ± 2.66 , 47.7 ± 2.1 , 53.3 ± 2.00 and 38.8 ± 2.00 at all supplied concentrations of copper. Potato peels are the best substitute and have been recommended to neutralize the copper toxicity from contaminated soil, because it is the best hyperaccumulator of heavy metals (Haider S *et al.* 2014) [15].

Shoot length: The shoot length of *vigna mungo* was found to be 17.86 ± 2.30 , 18.18 ± 2.17 , 13.09 ± 4.33 and 15.82 ± 2.08 at all applied levels of copper whereas it was 18.43 ± 3.21 in control. Copper at 25 and 50 ppm concentration exhibit beneficial effects on shoot length of *Vigna mungo* so these concentrations may consider as essential for optimal growth, Cu was shown to inhibit growth and to interfere with important cellular processes such as photosynthesis and respiration (Prasad and Strzalka, 1999) [31]. Application of potato peel significantly improved the shoot length of *Vigna mungo* up to 18.88 ± 2.50 , 19.89 ± 4.54 , 13.88 ± 2.79 and 16.86 ± 1.49 at all supplied concentrations of copper. Potato peels contain large amount of carbohydrate constituent, phenolic compound, phosphorus, potassium and other important nutrients (Azadeh M.S. *et al.* 2012) [7].

Root length: The root length of *Vigna mungo* was found to be 15.96 ± 2.48 , 11.94 ± 2.16 , 11.7 ± 2.81 , and 8.12 ± 0.68 cm at all applied levels of copper whereas it was 18.16 ± 0.73 in control. Decrease in root growth may attribute to the reduction in cell division. (Souguir *et al.* 2008) [40]. Application of potato peel significantly improved the root length of *Vigna mungo* up to 16.14 ± 1.88 , 14.51 ± 2.28 , 12.74 ± 1.70 and 10.10 ± 2.97 cm at all supplied concentrations of copper. Being an essential micronutrient copper promoted the growth of plants at lower concentration but reduced growth if present in high level interfering metabolic processes. Root is the main important and first contact part from soil contamination (Azmat, *et al.* 2006c) [46]. Potato peel contains polysaccharides, vitamins and minerals, as well as macro and micro nutrients which may overcome the toxicity of copper.

Leaf area: The leaf area of *Vigna mungo* was found to be 5.82 ± 1.07 , 4.87 ± 0.64 , 5.58 ± 2.04 and 4.34 ± 1.35 at all applied levels of copper whereas it was 7 ± 1.53 in control. The leaf area of *Vigna mungo* showed a gradual decline with an increase in copper level in the soil. Similar observations were reported by McBride (2001) [26]. Application of potato peel significantly improved the leaf area of *Vigna mungo* up to 7.22 ± 1.63 , 7.22 ± 0.83 , 6 ± 0.44 and 6.05 ± 1.13 at all supplied concentrations of copper. Potato peels not only provide organic substances, macro and micro nutrients to the plant but also grab toxic metal copper from the soil forming complexes, leaving the plant in a relaxed state (Haider S *et al.* 2014) [15].

Biomass: The biomass of *Vigna mungo* seeds was found to be 1.11 ± 0.40 , 1.16 ± 0.11 , 1.08 ± 0.05 and 0.8 ± 0.09 at all applied levels of copper whereas it was 1.21 ± 0.50 gm in control. The copper has expressed

an improvement in fresh weight and dry weight of *Vigna mungo* to some extent (50 ppm) at lower concentrations as it is included in micronutrients, but the biomass was significantly reduced with increasing concentrations of copper (Radha solanki and Rajesh dhankhar 2011) [32]. Application of potato peel appreciably improved the biomass of *Vigna mungo* up to 1.16 ± 0.19 , 1.28 ± 0.21 , 1.16 ± 0.08 and 0.88 ± 0.16 gm at all supplied concentrations of copper. Increment in biomass of *Vigna mungo* in the presence of potato peels under Cu stress may attribute to the ligand formation between polysaccharides of potato peels and Cu.

Relative water content (R.W.C): The relative water content of *Vigna mungo* was drastically decreased upto be 78.5 ± 0.70 , 63 ± 5.65 , 73 ± 4.24 and 62 ± 2.82 at all applied levels of copper whereas it was 81 ± 1.41 in control (Table 2). Data showed that relative water content of *Vigna mungo* decreased slightly with low moisture in soil and was reduced even more under stressed conditions (Rodríguez, L 2001) [47]. A huge reduction in relative water content may attribute to the adverse effects of copper on the roots of *Vigna mungo* (Table 2). Application of potato peel significantly improved the relative water content of *Vigna mungo* up to 82.5 ± 2.12 , 66 ± 2.82 , 77.5 ± 3.53 , and 66.5 ± 6.36 at all supplied concentrations of copper. The high relative water content is a resistant mechanism to stress (Morales *et al.* 1991 and Dell'Amico *et al.* 1991) [29, 9]. Organic and inorganic substances of potato peels not only supplies nutrients to the young seedlings but also efficiently adsorb copper ions (Haider S *et al.* 2014) [15].

Biochemical Parameters

Chlorophyll a: According to the data obtained in the present study chlorophyll a of *Vigna mungo* was found to be inhibited upto 0.25 ± 0.04 , 0.20 ± 0.19 , 0.22 ± 0.11 and 0.20 ± 0.09 at all applied levels of copper whereas it was 0.37 ± 0.07 mg/gm f.wt in control. Chlorophyll a contents declined progressively with increasing concentrations of copper in comparison with control. Heavy metals like copper are famous as the direct inhibitor of an enzymatic step (Van Assche and Clijsters, 1990; Sharma *et al.*, 2005) [12, 36]. Heavy metals like copper replaces magnesium of porphyrin ring of chlorophyll molecule and hence make it non-functional. Application of potato peel significantly improved the chlorophyll a contents of *Vigna mungo* up to 0.32 ± 0.02 , 0.28 ± 0.10 , 0.34 ± 0.01 and 0.21 ± 0.09 mg/gm f.wt at all supplied concentrations of copper. Potatoes peels contain large amount of carbohydrate constituents, and phenolic compounds which may adsorb copper hence may stop entry of salt to the plant (Azadeh, *et al.* 2012) [7].

Chlorophyll b: Data showed in Table 3 revealed that chlorophyll b of *Vigna mungo* was found to be decreased as 0.30 ± 0.05 , 0.22 ± 0.18 , 0.20 ± 0.21 and 0.21 ± 0.20 at all applied levels of copper whereas it was 0.30 ± 0.17 mg/gm/f.wt in control. Excessive Cu induces leaf chlorosis which is due to peroxidative breakdown of pigments and membrane lipids and in reduction of pigment content (Maksymiec, 1997; Shainberg *et al.*, 2001) [25, 35]. Application of potato peel significantly improved the chlorophyll b of *Vigna mungo* up to 0.18 ± 0.10 , 0.33 ± 0.02 , 0.37 ± 0.01 and 0.21 ± 0.20 mg/gm/f.wt at all supplied concentrations of copper. Potato peels contain a variety of valuable compounds, including phenols, dietary fibres, unsaturated fatty acids, amides, etc. (Schieber and

Saldaña, 2009; Wu *et al.*, 2012) [34, 42].

Carotenoid: The data of the present study revealed a drastic decrease in carotenoid of *Vigna mungo* upto 0.22±0.08, 0.24±0.06, 0.1±0.07 and 0.07±0.08at all applied levels of copper whereas it was0.35±0.13mg/gmf.wt in control. High concentrations of copper have important effects on plant growth and inhibit the development of various aspects of physiological, biochemical and even cell functions of the crop (Hegedus *et al.*, 2001) [16]. Application of potato peel significantly improved the carotenoid of *Vigna mungo* up to 0.35±0.03, 0.28±0.03, 0.24±0.02, and 0.21±0.007mg/gm f.wt at all supplied concentrations of copper. The potato peel contains vitamins and minerals, as well as macro and micro nutrients, and natural phenols that overcome the toxicity of copper.

Total chlorophyll: The total chlorophyll of *Vigna mungo* found to be inversely proportional to the increasing concentrations of copper upto 0.63±0.08, 0.50±0.29, 0.42±0.33 and 0.40±0.29 at all applied levels of copper whereas it was 0.67±0.10 mg/gm f.wt in control. Decrease in total chlorophyll content may attribute to the inactivation of various proteins which are involved in maintenance of chloroplast bilayer membranes it may also due to replacement of magnesium by copper in porphyrin ring of chlorophyll, inhibition of pigment accumulation and chlorophyll assimilation into photosystems (Kupper *et al.*; 2003) [21]. Application of potato peel significantly improved the carotenoid of *Vigna mungo* up to0.71±0.02, 0.53±0.22, 0.44±0.14 and 0.47±0.06mg/gm f.wt at all supplied concentrations of copper. Potato peels are a potential source of dietary fiber. Potato peel is a nutrient rich waste and it contains adequate amount of nutrients such as carbohydrate and proteins. It also contains alcohol-insoluble solids (pectin, cellulose, and starch), soluble sugars and minerals (mainly Ca, K, P, and Si) which can efficiently adsorb copper ions (Al-Weshahy and Rao, 2012) [3]. Data present in Table 3 exhibited toxicity of copper at higher concentration (75 & 100 ppm) where Potato peel failed to overcome its drastic effects efficiently.

Carbohydrate content: Data presented in (Table 4) revealed that carbohydrate of *Vigna mungo* was inversely proportional to the concentrations of copper and reduced upto 37.63±16.76, 29.46±2.99, 20.5±4.40, and 16.03±12.16 at all applied levels of copper whereas it was62.4±18.40 mg/gm f.wt in control. Inhibition in carbohydrate contents may attribute to the affinity of copper to replace magnesium from the porphyrin ring of chlorophyll, it may also cause damage in the membranes of chloroplast and inhibiting photolysis of water and obstruct both photosynthesis and carbohydrate metabolism (Yruela *et al.*, 1996, Azmat&Askari2013) [44, 6]. Data presented in (Table 4) showed that application of potato

peel significantly improved the carbohydrate contents of *Vigna mungo* at lower concentrations (25 & 50 ppm) up to42.63±22.7 and 35.8±10.36mg/gm f.wt whereas it did not showed any significant improvement at higher concentrations as (75 & 100 ppm) as 18.43±6.36 and 16.73±9.01mg/gm f.wt. Potato peels have high moisture, organic and inorganic contents which makes it a good basis for fertilizer as well as an efficient adsorbent (Arapoglou *et al.*, 2009) [4].

Protein content: Data of current study revealed that protein content of *Vigna mungo* drastically reduced under the stress of copper upto 19.4±4.44, 18.3±5.46, 18.2±5.94 and 23.5±6.07at all applied levels of copper whereas it was40.1±29.8mg/gm f.wt in control. Reduction in protein may attribute to the affinity of heavy metal copper for protein and enzymes, which contain several mercapto ligands to form chelate structure with the metals and hence losing their functional property (Prasad and Freitas, 1999) [31]. Decrease in protein may also attribute to the breakdown of protein under stress. (Dutta *et al.* 2012) [11]. Proteins are most important organic substances of living organisms, exhibit reversible and irreversible changes in physiological functions, are known to respond to a variety of stressors such as natural and xenobiotic (Singh and Tewari, 2003) [39]. Application of potato peel significantly improved the Protein of *Vigna mungo* up to21.4±2.22, 21.1±5.99, 24.2±8.59 and 30.6±4.52mg/gm f.wt at all supplied concentrations of copper. Since proteins were newly synthesized under Copper-stress, it appears to have a role in the mechanism of Copper tolerance which allows making biochemical and structural adjustments that enable the plant to cope with stress conditions as appeared at 100ppm in (table 4) (Jyoti *et al.* 2013) [19].

DNA concentration: Deoxyribonucleic acid is the most significant molecule that hold the genetic information used in the growth and development of all known living organisms and viruses. In plants DNA is enclosed within the nucleus, mitochondria, and chloroplasts. DNA has several properties that are unique among chemical molecules. Data presented in Table 4revealed that the DNA of *Vigna mungo* significantly reduced under the copper stress upto9.8±0.28, 7.24±0.11, 6.7±0.56, and 4.25±0.91at all applied levels of copper whereas it was 10.44±0.16 mg/ml in control. High concentration of copper badly effect DNA synthesis or may even block the cells in the G2 phase of the cell cycle preventing the cells from entering mitosis (Sudhakar *et al.*, 2001) [48]. Application of potato peel significantly improved the DNA concentration of *Vigna mungo* by adsorbing toxic metal copper up to10.8±0.42, 8.07±0.42, 10.15±0.91and 8.45±1.48mg/mlat all supplied concentrations of copper. Potato peel not only efficiently adsorbs toxic metals on their surfaces but also provides a variety of valuable compounds.

Table 1: Effect of copper and its adsorption by potato peel powder on Shoot length, Root length, leaf area in cms of *Vigna mungo*.

S.NO	Cu[ppm]	Shoot Length		Root Length		Leaf Area	
		Experimental	Treated	Experimental	Treated	Experimental	Treated
1	0	18.43±3.21	18.43±3.21	18.16±0.73	18.16±0.73	7±1.53	7±1.53
2	25	17.86±2.30	18.88±2.50	15.96±2.48	16.14±1.88	5.82±1.07	7.22±1.63
3	50	18.18±2.17	19.89±4.54	11.94±2.16	14.51±2.28	4.87±0.64	7.22±0.83
4	75	13.09±4.33	13.88±2.79	11.7±2.81	12.74±1.70	5.58±2.04	6±0.44
5	100	15.82±2.08	16.86±1.49	8.12±0.68	10.10±2.97	4.34±1.35	6.05±1.13

Results expressed as Mean ± SEM of triplicates.

Table 2: Effect of copper and its adsorption by potato peel powder on Biomass and Relative water content of *Vigna mungo*.

S. No	Cu [ppm]	BIOMASS (gm)		R.W.C (%)	
		Experimental	Treated	Experimental	Treated
1	0	1.21±0.50	1.21±0.50	81±1.41	81±1.41
2	25	1.11±0.40	1.16±0.19	78.5±0.70	82.5±2.12
3	50	1.16±0.11	1.28±0.21	63±5.65	66±2.82
4	75	1.08±0.05	1.16±0.08	73±4.24	77.5±3.53
5	100	0.8±0.09	0.88±0.16	62±2.82	66.5±6.36

Results expressed as Mean ± SEM of triplicates.

Table 3: Effect of copper and its adsorption by potato peel powder on photosynthetic pigments of *Vigna mungo* in mg/gmf.wt.

S. No	Cu(ppm)	Chlorophyll a		Chlorophyll b		Total chlorophyll		Carotenoid	
		Experimental	Treated	Experimental	Treated	Experimental	Treated	Experimental	Treated
1	0	0.37±0.07	0.37±0.07	0.30±0.17	0.30±0.17	0.67±0.10	0.67±0.10	0.35±0.13	0.35±0.13
2	25	0.25±0.04	0.32±0.02	0.30±0.05	0.18±0.10	0.63±0.08	0.71±0.02	0.22±0.08	0.35±0.03
3	50	0.20±0.19	0.28±0.10	0.22±0.18	0.33±0.02	0.50±0.29	0.53±0.22	0.24±0.06	0.28±0.03
4	75	0.22±0.11	0.34±0.01	0.20±0.21	0.37±0.01	0.42±0.33	0.44±0.14	0.1±0.07	0.24±0.02
5	100	0.20±0.09	0.21±0.09	0.21±0.20	0.21±0.20	0.40±0.29	0.47±0.06	0.07±0.08	0.21±0.007

Results expressed as Mean ± SEM of triplicates.

Table 4: Effect of copper and its adsorption by potato peel powder CHO, Protein and DNA of *Vigna mungo*.

S. No	Cu[ppm]	CHO (mg/gmf.wt.)		PROTEIN(mg/gmf.wt)		DNA(mg/ml)	
		Experimental	Treated	Experimental	Treated	Experimental	Treated
1	0	62.4±18.40	62.4±18.40	40.1±29.8	40.1±29.8	10.44±0.16	10.44±0.16
2	25	37.63±16.76	42.63±22.7	19.4±4.44	21.4±2.22	9.8±0.28	10.8±0.42
3	50	29.46±2.99	35.8±10.36	18.3±5.46	21.1±5.99	7.24±0.11	8.07±0.24
4	75	20.5±4.40	18.43±6.36	18.2±5.94	24.2±8.59	6.7±0.56	10.15±0.91
5	100	16.03±12.16	16.73±9.01	23.5±6.07	30.6±4.52	4.25±0.91	8.45±1.48

Results expressed as Mean ± SEM of triplicates.

Conclusion

Contamination of agricultural soil by toxic metals due to industrial effluents and other anthropogenic sources has become a universal problem, which not only reducing agricultural productivity but also causing severe human diseases. Detoxification of agricultural soil by green technology is a recent and efficient strategy that involves the use of waste agronomic by-products to adsorb heavy metal contamination from the soil.

Present study revealed that potato peel powder is an efficient and cost effective adsorbent, as it contains large amount of carbohydrates contents and phenolic compounds which have attribute to form metal complex. Beside these, glycoalkaloids, phosphorus, potassium, sulfur and other important nutrients which may act as functional group for adsorption of toxic metal copper are also present (Azadeh *et al.*, 2012; Chaney *et al.*, 2000; Dunbar, 2003)^[7-10]. Potato peels are already been using as a low cost and abundantly available organic fertilizer in various countries, as potatoes are daily diet of poor man of Indo-Pak due to its high nutrition values and low cost. It can be collected free of cost from hotels, restaurants, French fries shops and from residential areas. Daily use of potatoes makes tons of garbage which has to collect by Municipal Corporation daily, so use of potato peels as an adsorbent and as organic fertilizer may also reduce pollution and help directly or indirectly Municipal Corporation in collection garbage or pollutants. It is also concluded that, efficiency of potato peel powder should also be investigated for the adsorption of other heavy metals in major crops and cereals.

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