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IRRIGATIONAL IMPACT OF RUBBER FACTORY EFFLUENT ON ELEMENTAL BIOACCUMULATION AND METABOLITE CONCENTRATION IN COMPONENT PARTS OF *HORDEUM VULGARE* VAR. BG - 39

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Rubber factory effluent exhibited high magnitude of pollution. pH (4.2-9.4) showed fluctuation. Other parameters (mg/litre), viz., D.O (1.7-5.6), B.O.D. (31.34-1291.67), C.O.D. (61.0-1610.50), Chloride (295-6060), Free CO_2 (70-250), Oil and Grease (17.0-67.6) violated tolerance limits. Brownish-black colour, foul odour and poor transparency (2-3.5 cm) indicated poor water quality. T.S.S., T.D.S. and heavy metals like Cr, Pb, Zn, Fe and minerals like Na, K, Ca, Mg, SO_4, PO_4 and Total Nitrogen indicated organic and inorganic load. Percentage of Ca, K, PO_4 and Total Nitrogen, Crude Protein and Ether Extract was significantly lower in the seeds of effluent treated cultivar *Hordeum vulgare* var. BG -39. On the contrary, concentration of Na, Fe, SO_4, Total Carbohydrates, Total Ash and Chloride increased significantly.

Key Words : Industrial effluent, Pollution, Chemical composition, Bio-accumulation, Rubber Factory, Heavy Metals.

Indiscriminate disposal of wastes by large number of industrial units has led to rapid deterioration in the quality of aquatic environment at Bareilly. The effluent from Rubber factory located at Fatehganj, Bareilly is disposed off through several drains which carry their pollution load into river Shankha that ultimately causes pollution in river "Ram Ganga" flowing at 8 Km West from Bareilly. Cultivators of adjacent village irrigate their crops with the polluted water of the factory. This has resulted colossal damage to their crops. Several workers have studied the effect of industrial effluent on growth yield and chemical composition of various crops (Tripathi, 1978; Bahadur and Sharma, 1990; Someshekar *et al.,* 1992; Juwarkar *et al.,* 1993; Saini *et al.,* 1993; Sharma and Habib, 1994). No efforts seem to have been made to study the water pollution caused by Rubber Factory, Bareilly and its irrigation impact on mineral bioaccumulation and metabolite concentration in different parts of crop plants. An attempt has, therefore, been made to fulfil the above objective.

MATERIALS AND METHODS

The effluent disposed of from Rubber factory was collected from its disposal points at weekly intervals and was physico-chemically analysed according to A.P.H.A. (1980). Quantitative estimation of heavy metals in the effluent was made by using Atomic Absorption Spectrophotometer (AAS 300 Parkin and Elmer). The data are presented in table-1. Chemical analysis of root, stem, leaf and seeds was done according to Piper (1966). Seeds of *Hordeum vulgare* var. BG -39 were separately sown in unglazed earthen pots (22 cm dia.) containing uniform bulk density of garden loam soil mixed with farm-yard manure. Thinning was done after one week of seedling emergence to permit only one seedling to grow in each pot. The control and treatment sets were maintained separately in triplicate and were irrigated at weekly intervals with tap water and effluent, respectively. Plants were harvested at the time of seed ripening. Different Parts, viz., root, stem, leaf and seed were collected from the plants of control and treatment sets and analysed for total ash, Na, K, Ca, PO_4, Total-N, Fe, SO_4, CI, Crude protein, ether extract and total carbohydrates. Values are expressed in term of dry weight percentage. mean values ± S.E. were recorded for each parameter. Variance ratios, Critical Differences and SE m± were worked out according to Snedecor and Cochran (1967). The data are incorporated in Tables 2 to 5. The soil used in the control and treatment sets was the same and was found to have pH (7.3), Nitrogen (1.42 Kg ha^{-1}), Potassium (290 Kg ha^{-1}), ECe (0.8 mSC m^{-1}), Organic Carbon (1.65%), Organic matter (4.2%), WHC (42.13%), moisture (9.6%), Silt (14%) and Clay (30%).

RESULTS AND DISCUSSION

The data indicate (Table 1) that the brownish-black-colour, foul odour and poor transparency (2-3.5

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Table 1. Physico-chemical parameters of effluent and tap water used in seed germination and irrigational treatments (Nov. 1990 - April 1991)

	Parameters	Effluent	Tapwater
1.	Colour	Brown-Brownish-black	Colourless
2.	Odour	Foul smelling	Odourless
3.	Temp. (°C)	18.5 - 30.00	20.2 - 32.6
4.	pH	4.2 - 9.40	7.0 - 7.3
5.	Transparency (cm)	2.0 - 3.50	100
6.	T.D.S. ((mg/l)	323 -11200	96 - 152
7.	T.S.S. (mg/l)	169 - 1870	17 - 29
8.	D.O. (mg/l)	1.7 - 5.60	10.0 - 13.3
9.	B.O.D. (mg/l) (5 day 20°C)	31.34 - 1291.670	2.5 - 3.0
10.	O.O.D. (mg/l)	61.0 - 1610.50	40 - 45
11.	Total Alkalinity (as $CaCO_3$) (mg/l)	48.3 - 330.10	13 - 62
12.	Total-N (mg/l)	8.8 - 63.0	-
13.	Ca (mg/l)	66.5 - 89.2	1.3 - 4.6
14.	Mg (mg/l)	41.3 - 57.9	9.6 - 14.2
15.	Na (mg/l)	15.90 - 21.5	2.0 - 2.4
16.	K (mg/l)	17.0 - 17.8	1.7 - 2.1
17.	ECe. (mS cm^{-1})	3.9 - 4.8	1.3 - 1.7
18.	Total Hardness (as $CaCO_3$) (mg/l)	41.3 - 529.3	98 -1200
19.	Dissolved silica (mg/l)	0.4 - 1.5	-
20.	Free CO_2 (mg/l)	70 - 2500	1.2 - 2.5
21.	'Cl (mg/l)	295 - 6060	19.4 - 23.2
22.	SO_4 (mg/l)	110 - 144	1.6 - 4.8
23.	PO_4 (mg/l)	1.2 - 4.70	0.2 - 0.4
24.	Oil & grease (mg/l)	17.0 - 67.6	-
25.	Heavy metals		
	Cd (mg/l)	0.02 - ·0.3	-
	Cr (mg/l)	0.3 - 0.5	-
	Cu (mg/l)	0.5 - 0.8	-
	Pb (mg/l)	0.2 - 0.3	0.090
	Ni (mg/l)	0.3 - 0.5	-
	Fe (mg/l)	2.5 - 2.8	0.30
	Zn (mg/l)	3.0 - 5.5	1.20
	Co (mg/l)	0.3 - 0.5	-
	Mn (mg/l)	0.7 - 0.8	0.50

cm) of the effluent are evident in revealing the magnitude of pollution. pH (4.2 to 9.4) is not conducive to inhabitation and survival of aquatic life. Concentration of TDS, TSS, DO, BOD, COD, Total alkalinity, Total-N, Ca, Mg, Na, K, ECe (mScm^{-1}), Total Hardness, Dissolved Silica, Free CO_2, CI, SO_4, PO_4, oil and Grease, Cd, Cr, Cu, Pb, Ni, Fe, Zn, Co, Mn Violated the recommended permissible limits.

Calcium : Data indicate that there was overall decrease in the concentration of calcium in all the four component parts of *Hordeum vulgare* var. BG-39. The percentage decrease in the root, stem, seeds and leaf being 36.36%, 73.48%, 76.00% and 78.80% respectively (Tables 2-5). Calcium is an important constituent of middle lamella which accumulates it in the form of calcium pectate. Poor concentration of calcium in vegetative parts in the treated plants could be due to its poor intake as Ca+ ions through plasma membrane. Precipitation of calcium as calcium hydroxide and calcium carbonate seems to be important causative factor responsible for its restricted availability. its deficiency resulted in thin and weak stems. Poor development of leaves and inhibition in calcium content of seeds as reported by Clarkson and Hanson (1980).

Sodium : Sodium concentration increased significantly in all the four parts, maximum increase (828.57%) being obtained in the case of seed followed by leaf (272.88%), stem (375.00%) and root (163.16%) (Tables 2-5). Higher Na content in the stem of treated crops may be attributed to rapid intake of Na+ ions. There is ample evidence to indicate the Na+ ions always inhibit the entry of K+ ions. However, sodium ion in association with Cl- ions cause particles of plasma membrane to separate and enhance permeability (Bains and Fireman 1968). Accumulation of Na+ ions exerts inhibitory effect on metabolism and growth. Na+ ions play an important role in modifying the edaphic conditions of soil. In association with weak and strong anions it alters the pH of the soil solution and affects crop growth (Tripathi, 1978).

Potassium : Availability of potassium was poor in all the component parts. Decrease was maximum (91.76%) in root of the treated crop followed by seed (77.50%), leaf (29.1%) and stem (12.00%) (Table 2-5). Potassium occurs in the plant cells only in the ionic form as macronutrient. Its poor availability was observed in the form of deficiency symptoms. As an activator of enzyme ALA dehydraseit has been reported to play an important role in the biosynthesis of chlorophyll (Tribe and Whittaker, 1972). Potassium has a marked effect on the weight of the seeds hence its maximum reduction in the seeds may be attributed to its deficiency. Low pH has been reported to cause potassium deficiency and adverse effect on nitrogen metabolism (Sinha and Mehta, 1992).

Phosphate : Decrease in phosphate content was in the order of leaf > seed > stem > root. Maximum decrease (39.34%) was obtained in the leaf of treated crop. Poor intake of phosphate through plasma membrane is closely associated with pH. From pH 4 to 6.5 and 7.5 to 8.5 phosphate remain poorly available. This has been found to be associated with higher intake of sodium ions which exerts toxic effect in the higher concentration (Daubeanmire, 1970). Nutrient imbalance is main causative factor inhibiting growth (Brvok, 1983). Poor intake of phosphate has been found to affect synthesis of protein, carbohydrates and fat (Tables2-5).

Iron : Percentage increase in iron content in root, stem, leaf and seed was 597.67%, 300.00%, 195.77% and 29.41%, respectively. Iron content in the effluent was also much higher than recommended tolerance limit. Due to low pH of the effluent for major span of crop growth iron became excessively soluble and was absorbed mostly as ferric ions through plasma membrane. It appears that beyond its judicious micronutrient limit it exerted toxic effect on ferredoxin which plays key role in nitrogen metabolism. Inhibition in growth has also been found to be associated with suppression of the activity of several enzymes like peroxidase, catalase and of cytochrome. Iron porphyrin which acts as primary catalyst in respiration is possibly inactivated. Bio-accumulation of Fe in plants at low pH is amply documented (Tables 2-5).

Sulphate : Concentration of sulphate has been found to be significantly higher in all the parts in the effluent treated crop over the control, maximum being in the root (173.10%) followed by stem (153.80%), seed (119.74%), leaf (53.42%). Sulphates content was also higher in the effluent as compared to tap water. Sulphates are mostly soluble in water and impart hardness. Sulphates along with other inorganic solutes are absorbed through plasma membrane and become important constituent of proteins and vitamins like biotin, thiamine and coenzyme A. Since the concentration of sulphates inthe effluent was within the permissible limits, their bio accumulation int he seeds does not seem to be involved in growth inhibition. In association with cations like Ca++ and Mg++ they bring about lowering of pH in soil solution. Under reducing anaerobic conditions they are reduced to sulphates and may also lower down the pH and exert adverse effect on plant metabolism (Tables 2-5).

Chloride : Chloride percentage increased significantly in the all component parts of the treated plants. Percentage increase was maximum in seed while minimum (177.24%) values are obtained in case of root. Chloride content was higher in effluent treated crops over their respective controls. Along with other essential anions like SO_4__, NO_3-, Chloride ions were also taken up through plasma membrane and got accumulated in cytoplasm. Under judicious limits chlorides are required in catalytic amount to carry on various enzymatic reactions in the cells. However, when all the negative charges on the particles of protoplasm are neutralised and substituted by negative Cl- units the permeability of plasma membrane is at the maximum. Chloride ions are among strong anions that increase toxicity both in water and the cell sap (Table 2-5).

Total Nitrogen : Total Nitrogen exhibited significant decrease in the treated plants over of the respective control. The decrease was in the following sequence - 26.19% > 8.45% > 5.51% > 4.13% in seed, leaf, stem and root respectively. Though the concentration of nitrates and nitrites was higher in effluent yet the reduction in the total nitrogen in the seeds suggests impairement of nitrification caused by inactivation of microbes at low pH (6.5-4) of the effluent (Tables 2-5). Effect of nitrogen starvation was manifested by yellowing of leaves brought about by reduction in Chlorophyll biosynthesis and depressive effect on nitrogenous bases like, purines and pyrimidines (Sinha *et al.*, 1988).

Total Carbohydrates : Data reveals that their was overall increase in the concentration of total carbohydraes in the root, stem, leaf and the seeds of the cultivars studied. Maximum increase (3.53%) was obtained in the case of seed followed by leaf (0.68%), stem (0.49%) and root (0.24%). Greater concentrations of carbohydrates was found to associated with decrease fat content. Regeneration of organic matter in the form of carbohydrates thus become cyclic process in which CO_2, H_2, O_2 and N_2 which are liberated by the organic matter become available again for the synthesis (Caputto *et al.*, 1967).

Crude Protein : Crude protein contents exhibited over all decrease in seed, leaf, stem and root, being 26.19%, 8.39%, 5.54% and 4.10% respectively. Protein content has been found to be positively correlated with total-N. Inorganic nitrogen taken up as NO_3- ions is converted into NH_2 groups before being elaborated into amino acids (Boulter, 1970). Synthesis of proteins as also of fats is intimately linked with carbohydrate metabolism. Protein breakdown into amino acids is also adversely affected due to effluent

Table 2. Chemical constituents of Root in *H. vulgare* (var. BG-39) as affected by industrial effluent.

Parameters %	Contol Mean±SE	Treatment Mean±SE	% Decrease/ Increase	F	CD	SEm±
Ca	0.11 ± 0.06	0.07 ± 0.05	- 36.36	38.40***	0.02	0.003
Na	0.003 ± 0.002	0.01 ± 0.006	+163.16	10.00**	0.005	0.0004
KO.	18 ± 0.04	0.007 ± 0.004	- 91.76	920.00***	0.12	0.09
PO_4-P	0.08 ± 0.05	0.08 ± 0.04	- 4.76	7.51**	0.003	0.0004
Fe	0.004 ± 0.003	0.03 ± 0.02	+597.67	19.64**	0.01	0.003
SO_4	0.108 ± 0.06	0.29 ± 0.17	+173.10	1178.73***	0.02	0.003
Cl	0.29 ± 0.12	0.80 ± 0.25	+177.24	63.00**	0.10	0.01
Total-N	1.21 ± 0.10	0.16 ± 0.04	- 4.13	7.71**	0.23	0.04
Crude Protein	7.56 ± 0.40	7.25 ± 0.20	- 4.10	8.10**	0.21	0.003
Total Carbohydrates	81.20 ± 35.60	81.18 ± 28.25	+ 0.024	16.77**	0.04	0.005
EDther extract	0.14 ± 0.82	0.06 ± 0.04	- 55.71	44.68***	0.04	0.006
Total ash	11.09 ± 6.40	11.51 ± 6.64	+ 3.78	29.34***	0.26	0.04

** P <0.01, *** P < 0.001.

Table 3. Chemical constituents of Stem in *H. vulgare* (var. BG-39) as affected by industrial effluent.

Parameters %	Contol Mean±SE	Treatment Mean±SE	% Decrease/ Increase	F	CD	SEm±
Ca	0.21 ± 0.12	0.05 ± 0.03	- 73.48	1012.05***	0.02	0.003
Na	0.004 ± 0.003	0.01 ± 0.01	+375.00	30.00***	0.003	0.0004
K	0.10 ± 0.05	0.08 ± 0.05	- 12.00	13.34**	0.014	0.002
PO_4-P	0.16 ± 0.30	0.11 ± 1.00	- 30.86	48.63***	0.33	0.04
Fe	0.007 ± 0.004	0.02 ± 0.02	+300.00	311.29***	0.004	0.0006
SO_4	0.13 ± 0.23	0.33 ± 0.18	+153.80	28.15***	0.17	0.02
Cl	0.16 ± 0.10	0.68 ± 0.04	+315.24	8.00**	0.40	0.04
Total-N	1.27 ± 0.60	1.20 ± 0.02	- 5.51	48.00***	0.03	0.002
Crude Protein	7.94 ± 4.60	7.50 ± 4.33	- 5.54	18.47**	0.34	0.05
Total Carbohydrates	82.50 ± 47.61	82.91 ± 50.26	+ 0.49	8.70**	0.23	0.03
EDther extract	1.32 ± 2.20	1.11 ± 1.00	- 15.90	24.61***	0.20	0.03
Total ash	8.23 ± 0.50	8.98 ± 1.10	+ 8.90	114.61***	0.49	0.06

**P < 0.001, ***P < 0.001

Table 4. Chemical constituents of Leaf in *H. vulgare* (var. BG-39) as affected by industrial effluent.

Parameters %	Contol Mean±SE	Treatment Mean±SE	% Decrease/ Increase	F	CD	SEm±
Ca	0.170 ± 0.09	0.03 ± 0.02	- 78.80	1496.33***	0.01	0.002
Na	0.005 ± 0.003	0.02 ± 0.01	+272.88	40.00***	0.004	0.0004
K	0.134 ± 0.07	0.09 ± 0.05	- 29.1	104.35***	0.01	0.001
PO_4-P	0.244 ± 0.14	0.14 ± 0.08	- 39.34	345.60***	0.02	0.003
Fe	0.007 ± 0.004	0.02 ± 0.01	+195.77	573.89***	0.002	0.0003
SO_4	0.219 ± 0.13	0.33 ± 0.19	+ 53.42	1081.08***	0.012	0.002
Cl	0.205 ± 0.02	0.62 ± 0.03	+204.88	45.00***	0.18	0.02
Total-N	1.42 ± 0.82	1.30 ± 0.75	- 8.45	14.89**	0.10	0.015
Crude Protein	8.87 ± 5.12	8.12 ± 4.69	- 8.39	14.05**	0.64	0.10
Total Carbohydrates	79.46 ± 45.88	80.00 ± 46.18	+ 0.68	8.97**	0.58	0.09
EDther extract	1.36 ± 0.78	1.13 ± 0.65	- 16.90	52.90***	0.10	0.02
Total ash	10.32 ± 5.96	10.75 ± 6.21	+ 4.10	191.27***	0.02	0.01

**P < 0.01, ***P < 0.001

Table 5. Chemical constituents of Seed in *H. vulgare* (var. BG-39) as affected by industrial effluent.

Parameters %	Contol Mean±SE	Treatment Mean±SE	% Decrease/ Increase	F	CD	SEm±
Ca	0.10 ± 0.06	0.02 ± 0.01	- 76.00	1083.00***	0.008	0.001
Na	0.009 ± 0.005	0.09 ± 0.05	+828.57	990.00***	0.01	0.0002
K	0.44 ± 0.25	0.09 ± 0.05	- 77.50	8725.00***	0.009	0.001
PO_4-P	0.30 ± 0.17	0.20 ± 0.12	- 32.89	1034.48***	0.01	0.002
Fe	0.008 ± 0.005	0.01 ± 0.006	+ 29.41	24.95***	0.001	0.0002
SO_4	0.31 ± 0.18	0.69 ± 0.39	+119.74	8.45**	0.49	0.07
Cl	0.10 ± 0.02	0.59 ± 0.02	+490.00	61.00***	0.12	0.009
Total-N	1.68 ± 0.97	1.24 ± 0.72	- 26.19	72.60***	0.17	0.02
Crude Protein	10.50 ± 6.07	7.7 ± 4.48	- 26.19	72.58***	1.06	0.16
Total Carbohydrates	86.05 ± 49.68	89.09 ± 51.44	+ 3..53	68.59***	1.21	0.18
Ether extract	1.84 ± 1.06	1.03 ± 0.59	- 44.02	4845.95***	0.09	0.01
Total ash	1.61 ± 0.93	2.13 ± 1.23	+ 32.29	1014.00***	0.05	0.008

**P < 0.01, ***P < 0.001

toxicity. Hence poor availability of nitrogen may be a causative factor for reduction in crude protein content in different parts. Decrease in protein content has been found to be associated with increase in total carbohydrates which is nutritionally unsound since with decline of protein the subtle balance of amino acids is distrubed (Tables 2-5).

Ether Extract : Ether extract content showed over all decrease in all the component parts of the effluent treated crop. Maximum decrease was observed in the case of root followed by seed, leaf and stem over their respective controls which may be attributed to decline in carbohydrate reserves leading to break down of fats that are first hydrolysed in the presence of lipases to yield fatty acids and glycerol (Tables 2-5). Suppression of fat fetabolism may be accounted to inhibitiory action of pollutants such as heavy metals, sodium and chlorides on fatty acid synthesizing enzymes (Webb, 1966).

Total Ash : Percentage of ash content was higher in the seed of effluent treated cultivar as compared to control. Ash content of the cultivar is the direct manifestation of bioaccumulation of minerals observed as macro-nutrient (C, H, O, N, P, K, Ca, S, Mg, Fe) and micro-nutrients (Mn, Zn, Bo, Cu, Mo) which take up different functions, viz., synthesis of proteins, carbohydrates, lipids and some become important constituents of protoplasm and cell wall while others enter into the composition of large number of enzymes and also that of chlorophyll. Excessive solubility of Zn, Mn, Pb, Cr, Fe and Cu at low pH levels (4 to 6.5) is the chief factor creating toxicity (Singh and Mukhiya, 1980). Intake of toxic metallic ions results in their bioaccumulation in plant tissues. Metabolic attributes such as lipids, crude proteins and total carbohydrates being dependent on the efficiency of enzymes and intake of minerals from soil thus exhibited variation in their concentration. Soil pH plays a decisive role governing their intake. It is, therefore, desirable to ameliorate the pH of effluent without much alteration in ECe levels and treatment designs be modified according to the nature of prevailing pollution (Table 2-%).

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