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Amelioration of chlor-alkali waste contaminated soil by incorporation of *Sesbania aculeata* Pers.

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ABSTRACT

An attempt was made to decrease the toxic effect of waste soil from a chlor-alkali factory by Sesbania aculeata.Pers. Sesbania cultivated in varying waste/soil combinations for 35 days added about 120.59% more nitrogen and 220.33% more organic carbon to the medium containing 60% of the waste. Alkaline pH of the waste/soil combinations also decreased with time. After 35 days of incorporation of Sesbania, a significant increase in the growth of rice var. IR 36 was reported over that of control set.

Key words: *Chlor-alkali, waste soil, mercury, amelioration Sesbania*

The hazardous effect of mercury was realized after the outbreak of Minamata Bay disease in Japan, since then several research papers have been published pertaining to damaging effect of the heavy metal on living organisms. Nanda *et al* (1993) have established that waste of chlor-alkali factory loaded with various pollutants along with mercury is toxic inducing depletion in biochemical variables of rice plant. Dash (2002) observed that when *Sesbania aculeata* was applied to the waste soil containing mercury the decrease in morphological as well as biochemical variables took place. Mishra (1986) reported contamination of local crop fields with discharges from a chlor-alkali factory resulting depletion in production of cultivated cereals. It has been estimated that 7 million hectares of land was never in use due to its alkaline nature. Alkaline discharges released from chlor-alkali industry further degrade crop fields in the vicinity.

Although reports on biological reclamation of disturbed lands are very few Singh (1950) reclaimed saline “user land” by cyanobacteria. Subashini and Kaushik (1981) reported amelioration of sodic soil by blue-green algae. The mercury contaminated waste of the chlor-alkali factory M/s Jayashre Chemicals, Ganjam, Orissa, is alkaline in nature and also has affected local crop fields. Therefore an attempt was made to decrease toxicity caused by the same waste soil with *Sesbania aculeata*. *Sesbania* has been recognized as green manure with its capacity to fix atmospheric nitrogen and release them through decay along with organic acids (Panda *et al.*, 2005).

MATERIALS AND METHODS

Mercury contaminated brine solid waste of the chlor-alkali factory which was periodically removed and dumped by M/s Jayashre Chemicals, Ganjam, Orissa, was collected in gunny bags, air dried and was subjected to analysis for important constituents.

The dried and powdered waste soil was passed through a fine mesh and was used for the preparation of culture pots. Varying concentrations of the waste soil was prepared with normal garden soil (pH 6.44 ± 0.02) ranging from 10 per cent to 60 per cent with an increment of 10 per cent. Final weight of the soil combinations prepared was 4 kg. They were kept in earthen pots of equal size in triplicate to use as the medium to culture *Sesbania aculeata* seedlings. The pots were watered equally. The waste soil combinations and water were mixed thoroughly and allowed to settle.Four seeds of *Sesbania* were sown in each pot containing varying contaminated waste soil combinations.

Soil variables were measured after growth and decomposition of *Sesbania* in waste soil combinations at 14 and 35 days. Nitrogen content was analyzed by

Micro-Kjeldhal distillation method, soil organic carbon was measured by Walkley and Black (1934) rapid titration method.

After the decomposition of *Sesbania aculeata*, seedlings of rice var. IR 36 were transplanted in the waste soil combinations to measure possible bioremediation of the mercury contaminated waste soil.

RESULTS AND DISCUSSION

The pH of the alkaline gray coloured soil was measured to be 9.30 ±0.03. It contained 0.95 ± 0.05g of mercury per kg of the waste as analyzed by cold vapour atomic absorption technique using a mercury analyzer (MA 5800A). Sodium content of the waste soil, analysed by Flame Photometer, was 6 g per kg of the waste soil and chloride content was 18g kg^{-1}. Potassium and phosphate contents of the waste soil were 55 and 60 mg kg^{-1} of the waste soil, respectively.Mercury accumulation by root of *Sesbania* seedlings was found to be dependent on both waste soil percentage and time. Mercury accumulation was found to increase with increase in waste soil from 10 per cent to 60 per cent and also time period after the growth of *Sesbania aculeata*. In the control set with only garden soil mercury accumulation was not reported. Fourteen days after growth of *Sesbania*, mercury accumulation increased from 7.11 ± 0.11 µg g^{-1} in 10 percent to 18.27 + 0.07 µg g^{-1} fresh weight in 60 per cent waste soil combination. The correlation (r = 0.991) between waste soil combinations and mercury uptake by root was highly significant ($P < 0.01$). After 35 days growth of the *Sesbania* seedlings mercury uptake increased from 12.85 ± 0.05 µg g^{-1} fresh weight in 10 percent to 25.61 ± 0.06 µg g^{-1} fresh weight in 60 per cent waste soil combination. A highly significant correlation was obtained between waste soil and mercury uptake by root (r = 0.971, $p<0.01$).

The accumulation of mercury in shoot was dependent on amount of waste soil and time period. It increased with increased waste soil and time period. It increased with increasing waste soil form 10 per cent to 60 per cent and from 14 days after growth to 35 days. Highly significant correlations were observed between mercury uptake by shoot and varying waste soil combinations. 14 days after the growth of *Sesbania aculeata*, mercury accumulation increased from 3.88 ± 0.07 µg in 10 per cent to 8.22 ± 0.20 µg in 60 per cent waste soil combination per g fresh weight of shoot. A highly significant ($P < 0.01$) correlation (r = 0.986) was obtained between mercury accumulation and waste soil combination. 35 days after the growth of *Sesbania*, mercury accumulation increased from 7.00 ± 0.09 µg in 10 per cent to 11.15 ± 0.05 µg in 60 per cent waste soil combination per g fresh weight of shoot. A highly significant ($P < 0.01$) correlation (r = 0.984) was obtained between mercury accumulation and waste soil combination.

Culture of *Sesbania* in varying waste soil combinations for different time intervals resulted changes in soil pH (Table 1). Before incorpration of *Sesbania*, soil pH increased from 8.45 ± 0.06 in 10% waste soils to 8.65 ± 0.07 in 60% waste soils. After 14 and 35 days decomposition of *Sesbania* it decreased from 8.29 ± 0.05 and 8.21 ± 0.06 in 10% waste soil to 8.57 ±0.05 and 8.48±0.05 in 60% waste soils, respectively.

Nitrogen content of soil measured as g pot^{-1} changed after decomposition of *Sesbania* for varying time periods (Table 2). The changes in nitrogen content also varied with increasing concentration of the waste

Table 1. Changes in soil pH following the decomposition of *Sesbania*

Days of Culture	Percent waste soil combination						
	0	10	20	30	40	50	60
0	6.75 ± 0.05	8.45 ± 0.06	8.58 ± 0.20	8.59 ± 0.15	8.60 ± 0.16	8.64 ± 0.25	8.65 ± 0.07
14	6.30 ± 0.05	8.29 ± 0.05 (-1.89)	8.37 ± 0.04	8.47 ± 0.10	8.50 ± 0.06	8.51 ± 0.05	8.57 ± 0.05 (-0.92)
35	6.50 ± 0.05	8.21 ± 0.06 (-3.56)	8.23 ± 0.50	8.33 ± 0.75	8.43 ± 0.05	8.45 ± 0.03	8.48 ± 0.05 (-1.97)

Level of significance = <0.001
Values are mean of 3 samples ± standard deviation.
Figure in parentheses indicates percent decrease over respective control.

Table 2 Changes in Nitrogen content of 100g of soil following the decomposition of *Sesbania*

Days of Culture	Percent waste soil combination						
	0	10	20	30	40	50	60
0	18.21 ± 3.30	15.60 ± 2.16	13.40 ± 0.10	11.80 ± 1.85	9.00 ± 1.60	7.55 ± 1.11	6.80 ± 0.75
14	35.35 ± 1.25 (94.12)	26.80 ±2.00 (71.79)	21.50 ± 0.05 (60.45)	18.71 ± 0.10 (58.56)	13.55 ± 0.06 (50.56)	10.45 ± 0.05 (38.41)	8.90 ± 0.98 (30.88)
35	50.15 ±2.50 (174.14)	39.48 ± 2.85 (153.08)	33.75 ± 0.50 (151.87)	28.21 ± 0.95 (139.01)	21.11 ± 0.05 (134.56)	17.50 ± 0.07 (131.79)	15.00 ± 1.55 (120.59)

Level of significance = <0.01
Values are mean of 3 samples ± standard deviation.
Figure in parentheses indicates percent decrease over respective control.

soil. Before *Sesbania* inoculation nitrogen content of the waste soil combinations varied from 15.60±2.16 g in 10% to 6.80 ± 0.75 g in 60% waste soil. Following 14 and 35 days culture of *Sesbania* it increased to 26.80 ± 02.00 and 39.48 ±2.85 in 10% to 8.90 ± 0.98 and 15.00±1.55 in 60% waste soil combination. Percent increase over respective controls ranged between 71.79 in 10% to 30.88% in 60% waste soils in 14 days and153.08% to 120.59% in 35 days. The changes in nitrogen content were also statistically significant.

The organic carbon content of the waste soil combinations changed with concentration and time (Table 3). Before *Sesbania* inoculation organic carbon content of the waste soil combinations varied from 0.48±0.18 g in 10% to 0.25 ± 0.13 g in 60% waste soil. Following 14 and 35 days culture of *Sesbania* it increased to 1.05 ±0.22 and 1.89 ±0.26 in 10% to 0.45± 0.15 and 0.80±0.25 in 60% waste soil combination. Percent increase over respective controls ranged between 114.89% in 10% to 55.61% in 60% waste soil in 14 days and 289.98% in 10% waste soil to 220.33% in 60% waste soil.

In the control set (35 days), at maximum tillering stage height of the seedlings decreased from 48.00 ±1.67 cm in garden soil to 20.67 ±1.53 cm in 30% waste soil combinations (Table 4).In 40%, 50%, 60% waste soils rice seedlings failed to survive. Following growth and decomposition of *Sesbania* after 35 days rice seedlings not only survived in 40%, 50%, and 60% waste soil but also there was growth of the seedlings. In the treated set however reduction in survivability of rice seedlings started from 40% waste soil combinations. In 40% waste soil combination 7 out of 12 transplanted survived, in 50% waste soil 5 survived and in 60% only one survived. However heights of the rice plants decreased from the garden soil. Similarly at maximum tillering stage tiller number of the seedlings decreased from in garden soil to 30% waste soil combinations. In 40%, 50%, 60% waste soils rice seedlings failed to survive. Following growth and decomposition of *Sesbania* after 35 days rice seedlings not only survived in 40%, 50%, and 60% waste soil but also the tiller numbers increased.

The presence of various pollutants in fairly high amount indicated toxicity of the waste soil. Mishra and Mishra (1984), Mishra *et al* (1985) and Dash (2002)

Table 3. Changes in organic carbon content of soil following the decomposition of *Sesbania*

Days of Culture	Percent waste soil combination						
	0	10	20	30	40	50	60
0	0.48 ± 0.18	0.47 ± 0.13	0.42 ± 0.12	0.38 ± 0.13	0.32 ± 0.11	0.27 ± 0.11	0.25 ± 0.13
14	01.05 ± 0.22 (118.75)	1.01 ± 0.29 (114.89)	0.84 ± 0.25 (100.81)	0.73 ± 0.27 (92.11)	0.61 ± 0.16 (90.63)	0.51 ± 0.21 (88.89)	0.45 ± 0.15 (55.61)
35	1.89 ±0.26 (293.75)	1.80 ± 0.36 (282.98)	1.55 ± 0.30 (269.05)	1.35 ± 0.35 (255.26)	1.10 ± 0.25 (243.75)	0.90 ± 0.28 (233.33)	0.80 ± 0.25 (220.33)

Level of significance = <0.001
Values are mean of 3 samples ± standard deviation.
Figure in parentheses indicates percent decrease over respective control.

Table 4. Height and tiller number of IR 36 (without and after decomposition of *Sesbania*)

	Percentage of solid waste													
	0		10		20		30		40		50		60	
	WDS	ADS	WDS	ADS	WDS	ADS	WDS	ADS	WDS	ADS	WDS	ADS	WDS	ADS
Height	48.00±1.67	50.48±2.11 (5.17)	33.35±1.94	47.09±3.01 (41.20)	23.44±2.78	34.92±2.11 (48.98)	20.67±1.53	28.39±1.62 (37.35)	0 0	27.88±2.80	0 0	21.33± 4.00	0 0	15.00
Tiller number	10.84±1.01	15.08±1.10 (43.89)	3.29±0.67	11.83±1.79 (259.57)	1.38±0.25	7.50±1.69 (418.12)	1.07±0.58	6.08±1.75 (469.22)	0 0	4.40±1.88	0 0	2.00± 1.00	0 0	1.00

*WDS = Without decomposition of *Sesbania* ADS=After decomposition of *Sesbania*
Figures in the parentheses indicate percent increase over respective control

opined that the waste from chlor-alkali factory cause damaging effect on plant-systems. Shaw and Panigrahi (1986) reported contamination of vegetation in the nearby area of a chlor-alkali factory by the pollutants released with the industrial waste.

A decrease in alkaline pH of the waste soil was reported following culture of *Azolla* for various periods. Kaushik and Krishnamurthy (1981) reported decrease in alkaline pH of soil following BCA culture. Lewin (1962) and Stewart (1967) opined that cultured algae secreted peptides, polysaccharides and lipids into the medium. It is pertinent to suppose that a similar excretion of polysaccharides, peptides and other organic molecules by *Sesbania* could have resulted decrease in the alkaline pH of the waste soil combinations.

One of the consequences of biological activity of nitrogen fixing organisms is the nitrogen acceleration of the soil (Moore, 1969, Rogers and Kaulasorriya, 1980), *Sesabania* cultured in the waste soil mixtures increased nitrogen status with time. Singh (1961) and Mishra (1985) reported a similar increase in saline soil following culture of cyanobacteria. Raychoudhury *et al* (1979) reported enhancement in aggregation status of alkaline soil with fixation of nitrogen by blue-green algae. However, the decrease in nitrogen, fixed with increasing waste soil concentration in the waste soil mixtures indicated the toxic effect of the waste soil on *Sesbania*.

Along with changes in the alkaline pH and nitrogen status of the soil a significant increase in percent organic carbon content of the waste soil combinations was reported after growth and decomposition of *Sesbania*. Singh (1961) reported annual addition of organic acid to the order of 36.5% to 59.7% by blue-green algae. Moors (1969) reviewed organic significance of *Azolla* due to its capacity to fix organic carbon to the growing medium. The possible release of organic acids by *Azolla* to the growing medium resulted in enhancement of organic status of the waste soil combination was already reported (Behera, 1996).Similar reason may be traced out for the increase in organic carbon content of the waste soil after the decomposition of *Sesbania aculeata.*

The increase in height of rice seedlings over respective controls reflect reduction in toxicity of the waste soil induced by *Sesbania aculeata.* Singh (1961) strongly suggested that reclaimed use land supported rice and sugarcane in the third year with addition of high amount of nitrogen and organic carbon to the medium.It is pertinent to believe that increase in nitrogen and organic status of the waste soil combinations along with decrease in pH was induced by *Sesbania.*

This experiment indicated that *Sesbania aculeata* can be cultivated in alkaline soil containing mercury, to decrease the toxicity of the heavy metal and to increase soil fertility.

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