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Cocoon Yield Pattern and Analysis of Water, Soil and Leaves from Mulberry Gardens Irrigated with Polluted Water Around Bangalore, India

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Sericulturists in the vicinity of Bangalore city irrigate their mulberry gardens with Vrishabhavathy stream water, which is polluted with domestic and industrial wastes from the city. To investigate the effect of pollutants on silkworm crops, a detailed field survey was conducted to study the cocoon yield pattern of the crops raised on mulberry irrigated with wastewater as against irrigation by bore/open wells. The villages along the stream were grouped into five test batches at about a distance of 5~8 km from each other. The seasonal yield data with relevant information were collected through questionnaires from 117 rearers using stream water and 35 rearers using bore/open wells, the latter serving as control group. The average yield for 100 layings was 46 to 57 kg in the control group whereas in test groups, it ranged between 34 to 51 kg in the first test group and 22 to 38 kg in the rest. The difference in yield was 9~19 kg depending on the season between control and test batches. In summer, this difference was higher, with high co-efficient of variation in test groups (33~52%). Further, water, soil and leaf samples were collected from selected rearers and were analyzed for zinc, copper, iron, lead and nickel. Results indicated significantly higher contents of these metals in samples from gardens using wastewater when compared control samples. Significantly ($p<0.05$ & $p<0.01$)) higher levels of zinc (24~122 ppm) and iron (208~683 ppm) were noticed in mulberry leaves during summer followed by winter and rainy season. The significance of high content of heavy metals in mulberry leaves and cocoon yield pattern of this area in relation to the quality of irrigation water is discussed.

Key words: Heavy metals, Waste water irrigation, Mulberry, Silkworm, Cocoon yield

Introduction

The problem of pollution of streams and rivers with industrial effluents and domestic waste is common especially around cities in India. This polluted water is used for irrigation of agricultural crops and studies on its effect on agricultural produce are intensified in recent years (Shrivastava, 1993). Vrishabhavathy stream passes near Bangalore City and flows towards south where, at Byramangala village, 35 km from the city, it forms a large tank or reservoir before joining Arkavathy, a tributary of Cauvery River, near Kanakapura town. The stream passes through the industrial area of the city hence it contains more of effluents as well as domestic wastewater. Sunitha and Phene (1995) have reported that Vrishabhavathy stream is highly polluted till about 18 km from Bangalore City and medium pollution is observed for another 11 km till Byramangala tank/reservoir. The farmers practicing sericulture along this valley use this stream water to irrigate their mulberry gardens. The potential hazard of accumulated pollutants with continuous irrigation with wastewater poses a threat to sustainable sericulture activity in this area since mulberry plant is perennial. It is reported that plants and crops grown in this region are affected by trace metal contamination (Chikkaswami and Shivashankar, 1999; Somashekar and Ramaswamy, 1982). N, P, K and micronutrient contents in soil and mulberry leaf samples from sericulturists' gardens at Byramangala area were analyzed by Subbarayappa *et al.* (1996) and at Kengeri area by Bongale and Krishna (2000), but information on heavy metal content of mulberry leaves and soil was not available. Hence, to investigate the effect of pollutants on silkworm cocoon crops, a detailed field study involving sericulturists was conducted along Vrishab-

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Table 1. Yield pattern of cocoon crops in selected points in Vrishabhavathy Valley area collected during the field survey

Sampling point	No. of farmers		Mulberry area covered (acres)	Average yield/100dfls	
	w.w. source[1)]	bw. Source[2)]		w.w. source[1)] (kg)	bw. source[2)] (kg)
I Kengeri (Outskirts of Bangalore City)	29	2	41.1	45.7 (25-75)	45 (30-60)
II. Kengeri (12 km from Bangalore City)	22	4	50	40.5 (15-70)	56.3 (25-75)
III. Byramangala (25 km from Bangalore City)	34	11	34.2	35.0 (6-60)	38.2 (15-60)
IV. Harohalli (35 km from Bangalore City)	20	6	25.5	25.5 (8-55)	30.0 (20-40)
V. Kanakapura (45 km from Bangalore City)	12	12	43.7	38.1 (15-40)	45.6 (20-60)

[1)]Mulberry leaves used from wastewater irrigated gardens for rearing.
[2)]Mulberry leaves used from borewell water irrigated gardens for rearing.

havthy area from the outskirts of Bangalore city till Byramangala tank/reservoir and the observations are presented in this paper.

Material and Methods

Field Survey

Vrishabhavathy stream area from a village called Mylasandra close to Bangalore City near Byramangala was divided into 5 sampling points with 8~10 km distance. The farmers using both wastewater and bore well water were interviewed through questionnaires seeking the information about the age of mulberry garden, cultivation practices, rearing techniques and annual crop yields pattern. The information collected from 117 and 35 farmers were recorded and compiled later to obtain a comprehensive description of their status as sericulturists (Table 1). Cocoon yield data collected from the rearers were compiled and compared between seasons and different points taking both the control (bore well water irrigation) and test (wastewater irrigation) groups. Data were analyzed statistically with ANOVA test and co-efficient of variation and presented in Table 2.

Table 2. Yield pattern (yield/100 Dfls in Kg) of cocoon crops during different seasons along Vrishabhavathy valley area from Kengeri to Byramangala

Area	Summer	Winter	Rainy	Average
I Point Kengeri TSC (12 km from Bangalore city)	33.90[1)] ±11.77	50.7[1)] ±0.88	43.93[3)] ±4.61	42.84[1)] ±11.22
II Point Kengeri TSC (24 km from Bangalore city)	28.75[1)] ±14.93	37.5[2)] ±15.55	32[1)] ±15.66	32.75[2)] ±14.42
III Point Byramangala TSC (36 km from Bangalore city)	21.95[1)] ±7.34	37[2)] ±4.67	32.13[1)] ±3.71	30.36[2)] ±8.22
Control (borewell water irrigated)	46.13 ±8.82	57.43 ±8.89	49.5 ±7.82	51.02 ±9.16
F test	S[1)]	HS[2)]	S[1)]	HS[2)]
CD @ 5%	9.46	7.15	7.12	8.58
CD @ 1%	–	10.02	–	11.51

Values are mean±Standard deviation.
[1)]Significant at 5%; [2)]Significant at 1% and [3)]not significant when compared to control.

Analysis for heavy metal content

Sample Collection: For heavy metal analysis, the area was divided into three points one each from Mylasandra and Byramangala villages and one in the middle region along Vrishabhavathy stream at 8, 20, 30 km distance from Bangalore City, respectively. Samples were drawn from sewage and bore well water irrigated mulberry gardens for water, soil, tender and coarse mulberry leaves from both control and treated groups. In total 16 sets of samples were taken from the test groups and 5 from control group during summer, rainy and winter seasons to compare the seasonal differences in heavy metal concentration.

Analysis using AAS: Water samples were diluted to 50 ml and readings were taken on AAS (Model 939, Unicam, UK). The soil and leaf samples were dried and known amounts were digested with nitric acid-perchloric acid (metal free grade) until the solutions were clear on a hot plate. The readings were taken after filtration by using AAS after calibration with the standard solutions of Copper, Lead, Nickel, Iron and Zinc (purchased from Merck Co.) according to the standard methods (AOAC, 1995). The analyte concentrations of blank and unknown samples were determined and recorded as concentrations in mg/L. The final content was calculated as below:

$$\text{Analyte Content} = \frac{(\text{Digest mg/L} - \text{Blank}) \times (\text{Final Digest Dilution Volume in Liters})}{(\text{Sample Mass (mg)}) \times 1000}$$

The calculated values were tabulated, statistically ana-

Table 3. Heavy metal content (ppm) in mulberry leaf samples collected from farmers' gardens along Vrishabhavathy valley area

Element	Borewell irrigated	S.p. 1[1)]	s.p. 2[2)]	s.p. 3[3)]	F Test	CD@ 5%	CD@ 1%
Summer							
Zn	31.7	46.3[5)]	43.3[5)]	122.3[5)]	HS[5)]	7.3	11.1
Cu	10.4	15.3[5)]	13.9[4)]	18[4)]	S[4)]	3.1	–
Fe	186.7	605.3[4)]	340.9[4)]	639.7[4)]	S[4)]	233.6	–
Pb	7.7	11.3[4)]	11[4)]	11.7[4)]	S[4)]	1.6	–
Ni	3.2	6.5[4)]	5.6[4)]	6.7[4)]	S[4)]	0.92	–
Rainy							
Zn	22	38.3[4)]	31.7[4)]	55[4)]	S[4)]	7.82	–
Cu	4	9[4)]	6.7[4)]	8.3[4)]	S[4)]	0.96	–
Fe	146.7	449[4)]	206.7[6)]	245[6)]	S[4)]	159.4	–
Pb	11.6	15[4)]	15[4)]	18.3[4)]	S[4)]	2.58	–
Ni	0.91	5.8[5)]	3.1[5)]	4.6[5)]	HS[5)]	1.39	2.11
Winter							
Zn	21	29.7[6)]	24[6)]	90.7[5)]	HS[5)]	10.3	15.6
Cu	9.3	11.9[5)]	11.4[5)]	13.7[5)]	HS[5)]	0.76	1.15
Fe	149.3	485.2[5)]	305.3[4)]	683.3[5)]	HS[5)]	113.3	171.6
Pb	8.7	10.3[4)]	9.3[6)]	11.7[4)]	S[4)]	1.1	–
Ni	2.1	4.9[5)]	4.3[5)]	5.3[5)]	HS[5)]	0.29	0.44

[1),2),3)]Samples collected from farmers of Kengeri, Mayaganahalli and Byramangala areas (at 12, 24 & 36 km from Bangalore city), respectively.
[4)]Significant at 5%; [5)]Significant at 1% and [6)]not significant when compared to control(borewell irrigated).

lyzed adopting ANOVA test and while the values for water and soil samples are presented as histograms (Figs. 1~4), the analyzed values for heavy metals observed in mulberry leaves are represented in Table 3.

Results

Field Survey

From the yield data collected from the field survey in the area of wastewater (polluted with industrial and domestic effluents) irrigated gardens, it could be inferred that there is a difference of 5 to 16 kg in cocoon yield between the gardens irrigated with bore well and wastewater at the identified areas along Vrishabhavathy valley (Table 1). There was a wide difference in the yield range in wastewater-irrigated gardens. Cocoon yield from 117 rearers using polluted water ranged from 6 to 70 kg/100 dfls and the average 25 to 40 kg. The rearers using bore well water obtained yield ranging from 15 to 75 kg on an average of 30 to 56 kg. A difference of 6~12 kg between the test and control groups was noticed in villages of Mayaganahalli & Harohalli TSC range, where the crop yield is generally lower when compared to other areas surveyed.

When the seasonal variations between the cocoon yield in test batches were statistically analyzed, significant differences could be observed during all the seasons and overall average yield (Table 2). The yield pattern showed that the yield was higher in all the areas during winter when compared to summer and rainy seasons. There was significant to highly significant differences in yield between bore well-irrigated gardens and wastewater irrigated gardens. When the range of yield is examined it is observed that at the third sampling point, the average yield was lowest (22 kg) while highest was obtained in control batch (57.4 kg) during winter. In general, between the sampling points tested, the yield was higher in first point (33.9 to 50.7 kg) followed by the second point (28.8 to 37.5 kg). The cocoon yield was lower i.e., ranged from 22 to 37 kg even in favourable season i.e., during winter at third point, indicating the problem of water as the wastewater connects to a tank at Byramangala, where the accumulation of pollutants is severe.

Analysis of heavy metals in samples

Water samples used for irrigation: There is considerable difference in the zinc content of irrigation water, though the difference in bore well water between seasons is negligible (0.053 to 0.071 ppm) (Fig. 1). In wastewater collected at different points along Vrishabhavathy stream, it was high during summer (0.199 ppm at third point) followed by winter (0.157 ppm). The copper content showed similar trend, showing low levels during rainy but high during summer (ranging from 0.032 to 0.563 ppm) (Fig. 1). But in sampling points 2 & 3, higher levels observed even during rainy season (0.210 & 0.417 ppm) while least values obtained in winter (0.159 and 0.316 ppm). There was drastic difference in iron content between bore well water and wastewater sources, being low in the former and higher in the latter and in summer the highest levels of 5.3 ppm was observed in sampling point 3 (Fig. 1). The lead content was on the other hand highest (0.302 ppm) in sampling point 1 probably because of the proximity of battery industries located at the outskirts of the city. The values were higher in summer (0.159 to 0.302 ppm) followed by winter (0.125 to 0.183 ppm) and during rainy season show the least values in all the batches (0.060 to 0.098 ppm) (Fig. 2). Interestingly, nickel levels in bore well water were at undetectable level whereas in wastewater it ranged between 0.001 to 0.027 ppm (Fig. 2). It was low in sampling point 2, highest in third point fol-

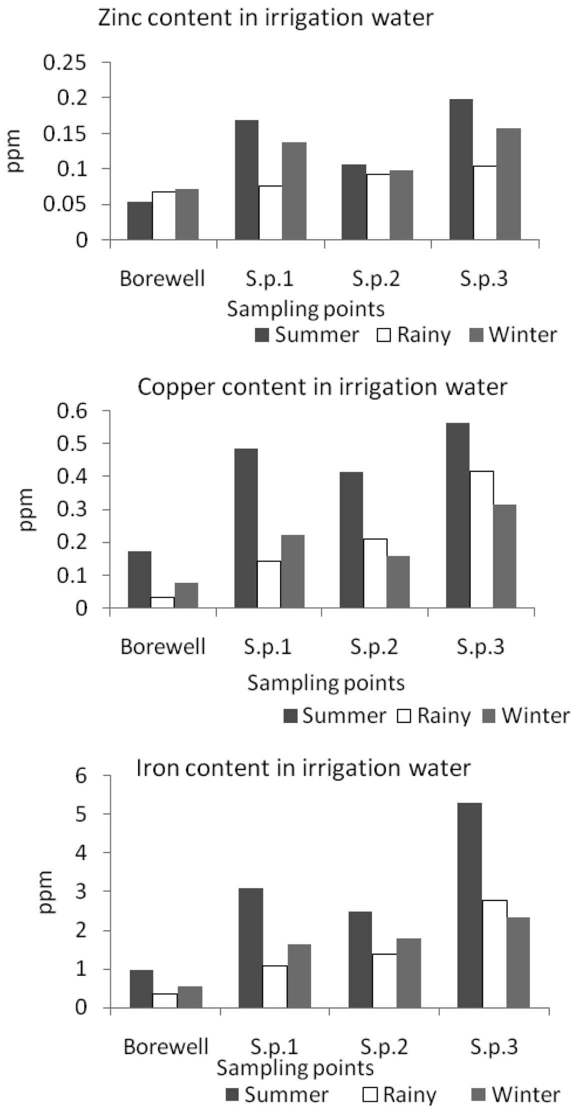

Fig. 1. Levels of Zinc, Copper and Iron (ppm) in irrigation water from bore wells and wastewater sources in areas from Bangalore Urban district in summer, rainy and winter seasons. S.p.1, S.p.2 & S.p.3: Samples collected at three points along the waste water stream from Bangalore City, at 12, 24 & 36 km respectively.

lowed by the second.

Soil samples: In soil samples from all the areas, zinc content was highest in summer, followed by winter and lowest in rainy season (Fig. 3). The bore well water showed lower levels (23 to 82 ppm) than the wastewater source, especially in 1 and 3 sampling points (114 to 210 ppm). Copper content was higher in wastewater source, being more in summer and winter seasons (Fig. 3). Highest levels (210 ppm) were observed in sampling point no.3. Iron contents ranged from 12030 ppm to 18940-ppm levels, lowest in bore well water during rainy season while highest value in wastewater source no. 3 during

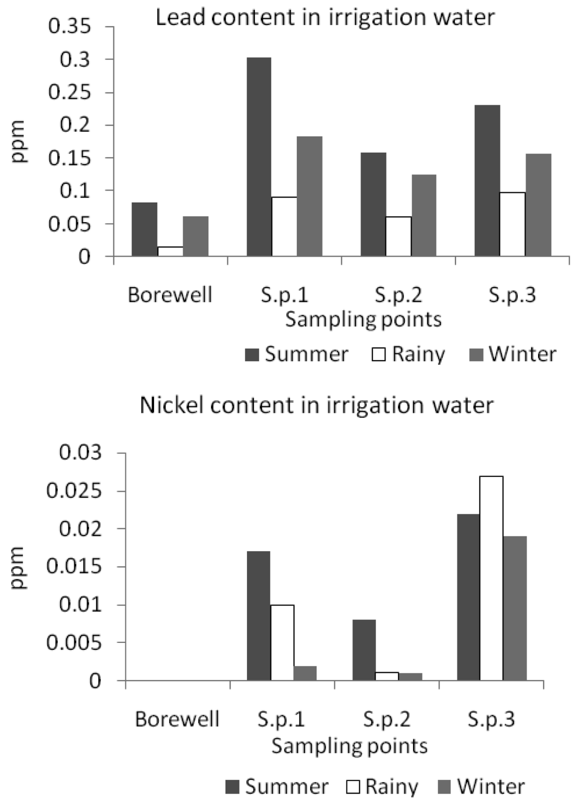

Fig. 2. Levels of Lead and Nickel (ppm) in irrigation water from bore wells and wastewater sources in areas from Bangalore Urban district in summer, rainy and winter seasons. S.p.1, S.p.2 & S.p.3: Samples collected at three points along the waste water stream from Bangalore City, at 12, 24 & 36 km respectively.

summer. During all the seasons, bore well water exhibited lower levels, followed by sampling point 2, 1 and 3 (Fig. 3). The concentration of lead was higher in wastewater source in all the seasons as compared to the soil samples from irrigated-irrigated gardens. Relatively higher contents were noticed in summer (50 to 82 ppm) and winter seasons (51 to 66 ppm) as against rainy season (43 to 7 ppm) (Fig. 4). In contrast to the irrigated water, soils irrigated with irrigated water exhibited 10 to 20 ppm levels of nickel. But, the levels were lower when compared to the soil from wastewater-irrigated gardens as unto 122-ppm levels could be observed at sampling point 3 during summer (Fig. 4).

Mulberry leaf samples: The statistically analyzed data from the values obtained after analysis of mulberry leaves collected from the gardens of rearers at the test area are presented in Table 3. During summer, zinc levels were higher compared to other seasons in all the groups. It was at lowest level in leaves raised with irrigated gardens during winter (21 ppm) and the values were significantly ($p<0.01$) lower when compared to the levels found in

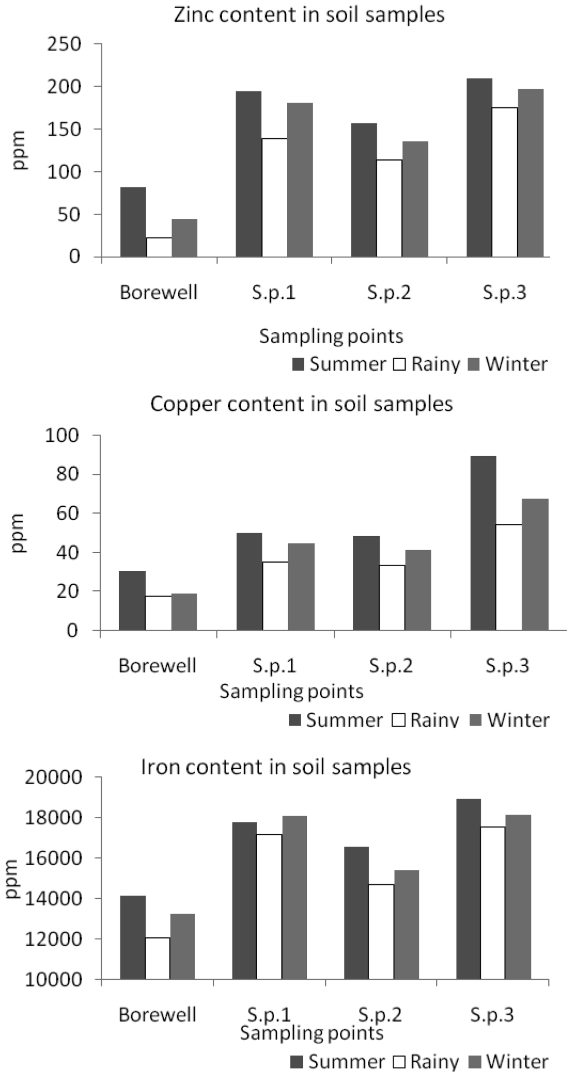

Fig. 3. Levels of Zinc, Copper and Iron (ppm) in soils from bore wellsand wastewater irrigated mulberry gardens in areas from Bangalore Urban district in summer, rainy and winter seasons. S.p.1, S.p.2 & S.p.3: Samples collected at three points along the waste water stream from Bangalore City, at 12, 24 & 36 km respectively.

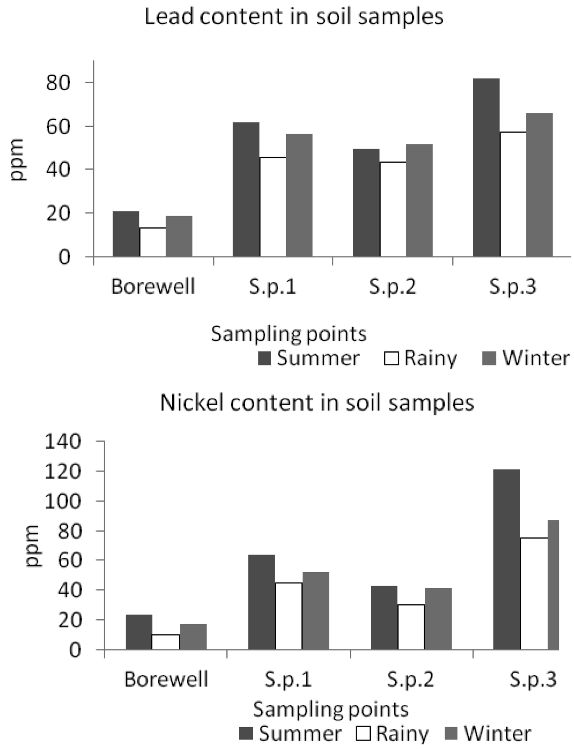

Fig. 4. Levels of Lead and Nickel (ppm) in soils from bore wells and wastewater irrigated mulberry gardens in areas from Bangalore Urban district in summer, rainy and winter seasons. S.p.1, S.p.2 & S.p.3: Samples collected at three points along the waste water stream from Bangalore City, at 12, 24 & 36 km respectively.

leaves from wastewater-irrigated gardens. The highest value was observed at third sampling point during summer (122.3 ppm). The copper content was also high during summer in all the groups though significant ($p<0.05$) difference was observed between control and test batches in all the seasons. The values were lowest during rainy season in all the batches. Significantly higher iron content was found in test batches as against the control group. In the test batches high levels were noticed during summer and winter, reaching unto 683 ppm at the third sampling point (Table 3) and higher values were noticed during rainy season (449 ppm) at first point, which may be due to the accumulative effect. The levels of lead varied from 7.7 ppm to 18.3 ppm in control batch during summer and in point no.3 batch during rainy seasons. There was no wide difference in the content between seasons in contrast to those observed for other elements, which may also be due to the cumulative effect. In case of nickel, during rainy season, the levels were negligible in leaves of the control batch, whereas the test batches showed a range of 3.1~6.7 ppm and variation was not very wide between the sampling points (Table 3).

Discussion

The present study indicates that irrigation with wastewater increases the heavy metal concentration in soil and mulberry leaves, which, though not at toxic levels individually; their combination might affect the growth and development of silkworms. The cocoon yield pattern shows that during summer, there are crop losses and though the adverse environmental conditions are a common factor, the loss is heavy in cases where wastewater is used for irrigation. The low yield is noticed throughout the year around Byramangala village, where the rearers use the polluted water from the reservoir for irrigation. This is the third sampling area where heavy accumulation of heavy metals is noticed probably because they tend to precipitate in the reservoir. The results obtained from the analysis of heavy metals in water, soil and leaves corroborate this suggestion. Though some of the earlier studies indicate the beneficial effect of using wastewater (Subbarayappa *et al.*, 1996), many recent reports have highlighted the dangers of using polluted water for irrigation (Chikkaswamy and Shivashankar, 1999; Das *et al.*, 2003). Vrishabhavathy stream contains metals and solids due to the influx of industrial effluents, which have a propensity to accumulate in soils and hence their levels may attain dangerous concentrations to affect the physiology of silkworms, as it is a sensitive insect. Subbarayappa *et al.* (1996) found that available phosphorus was high in sewage irrigated mulberry garden soil but potassium, calcium and magnesium were more in irrigated water. Besides, sulfur and manganese content were higher in mulberry leaves after wastewater irrigation though no additional inputs were given to the garden. Similarly, Bongale and Krishna (2000) reported significantly higher cocoon yield harvested from sewage water irrigated mulberry gardens. On the contrary, observations recorded by Chikkaswamy and Shivashankar (1999) indicate that soil status is contaminated with heavy metals in this study area. Gowda *et al.* (2000) have noted that irrigation with sewage water leads to soil sickness due to accumulation of soil organic matter and other molecules in capillary pores which adversely affect soil aeration and infiltration resulting in poor root growth and plant growth of mulberry. A few reports have shown that the nutritive component of mulberry leaves is found to be highly influenced by the soil nutrient status (Rupa *et al.*, 1993; Subbaswamy *et al.*, 1990). Kuno (1984) had observed that cadmium, copper and nickel at high concentrations in the soil showed reduced leaf weight and photosynthetic rate in mulberry. Saha *et al.* (2003) observed that growth of mulberry is poor when wastewater is used and lower contents of nitrogen, potassium and chlorophyll were found in these leaves. While examining the mobility of heavy metals in food chain, in case of cadmium and copper contents, higher mobility from soil to root was observed, followed by leaf to larva, larva to feces and finally root to leaf (Prince *et al.*, 2001). They demonstrated that in case of leaf to larval transfer, copper has a tendency of higher mobility showing a concentration dependent accumulation.

Though the plants can accumulate heavy metals at concentrations not toxic to them, they can affect the animals, which feed on them. Earlier studies indicate that though zinc, iron and copper are essential (Ito, 1978), excess content is evidently harmful to silkworms. These studies suggest that the influence of zinc or copper on the silkworms depend on the concentrations either as supplement or in the feed itself. Magadum *et al.* (1992) observed that there was deleterious effect at 50 and 100 ppm levels of oral copper supplementation affecting the survival. This deleterious effect was observed at higher levels of zinc (more than 90 μg) supplemented with food of silkworms (Hugar *et al.*, 1998). When nickel chloride treatment was given, the lipid content increased in *Bombyx mori* at 0.25 and 0.5%, but at higher concentration i.e., 0.75% the reverse effects were noticed (Saha and Khan, 1995), clearly indicating the deleterious effect at higher concentrations. In silkworms, cocoon and shell weights decreased when there was heavy metal concentration in the feed (Miyoshi *et al.*, 1978). These reports along with the present observations identify the safe levels of copper, iron and zinc in the feed for silkworms should be below 10, 50 and 50 ppm, respectively, whereas lead and nickel are not required for their growth (Miyoshi *et al.*, 1978). Thus, importance of monitoring heavy metal levels in the feed cannot be ignored, as otherwise, due to their accumulation effect; higher contents can result in growth inhibition leading to toxicity-induced mortality (Gintenreiter *et al.*, 1993). This might have reflected in the cocoon yield pattern of rearers using wastewater since low yield is apparent at the area (third sampling point), where the precipitation of heavy metal at the reservoir is noticed in the present study. Hence, close monitoring of heavy metal contents in soil and mulberry leaves is essential in the area where irrigation is conducted with industrial wastewater for maintaining the sustainability of silkworm cocoon crops. Further, more in-depth studies are required to be taken up to further emphasize the need for taking appropriate measures to treat the industrial and domestic wastewater sources before using them for irrigation purposes.

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