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EFFECT OF TANNERY WASTEWATER ON THE GROWTH AND YIELD OF SUNFLOWER (*HELIANTHUS ANNUUS* L.)

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Abstract

The effects of tanneries wastewater on growth and yield of three cultivars (SF-187, CRN-1435 and DK-3915) of sunflower (*Helianthus annuus* L.) showed that wastewater was not fit for irrigation due to extremely high mineral and heavy metal contents. In addition, effluents were highly alkaline with high load in EC, BOD, COD and SAR values. The amount of iron, manganese, zinc and chromium was also quite high. The growth and yield of plants was adversely affected by effluents treatment, especially at higher doses from seedling stage to maturity leading to reduce biomass and seed production as compared to control. Effluents of tanneries used for irrigation purposes proved to be alarmingly toxic to the growth and yield of the important crop in Kasur region, Pakistan.

Introduction

There are as many as 800 tanneries including small tannin units located in Pakistan. Out of these, 237 units are located in Kasur District (Anonymous 2009). Effluents of tanning industries containing Cr and other metals are discharged into the local water bodies which are used for irrigation purposes. Presence of chromium beyond the tolerance limit (< 2 ppm) makes water unsuitable for crop growth (Sahu *et al*. 2007). The high level of Cr and nutrient contents in the effluent has been reported to inhibit the seed germination and seedling growth, which might be due to the presence of excessive amount of dissolved solids, chlorides, sulphides, chromium, high BOD and COD values of the effluent (Mishra and Bera 1995).

Wastewater laden irrigation water affects the plant growth and yield (Barman and Lal 1994) and the accumulation of toxic heavy metals are biomagnified at different trophic levels through food chain (Rai *et al*. 2002, Saxena *et al*. 2007, Perez and Sarma 2008). The accumulation, however, depends on the plant species, the elements, its bioavailability, redox, pH, cation exchange capacity, dissolved oxygen, temperature and secretion of roots. Marked toxicity of chromium has been reported with respect to photosynthetic pigments in algae and other higher plants (Nath *et al*. 2005). Chromium toxicity produces chlorosis and necrosis in plants (Cervantes *et al*. 2001). The decrease in chlorophyll contents is due to chromium, competing for iron at functional site which might be interfering with the functional metal (Mg^{2+}) in the porphyrin ring (Mengel and Kirkby 2001). Chromium exposure results into complete loss of growth in lateral roots while lesser concentration starts damaging root cap, stomata and cotyledonary hair seem to be collapsed and plasma membrane appears to be detached from the cell wall under cytological studies (Mariappan *et al.* 2001).

The present study was undertaken to investigate the chemical composition of the effluents released by the tanneries and their effects on the growth and yield of three cultivars of sunflower.

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Materials and Methods

The sunflower (*Helianthus annuus* L.) cultivars selected for the present study were SF-187, CRN-1435 and DK-3915. The experiment was performed in a wire house at the Botanical Garden, GC University, Lahore during the growing season 2004. Seeds of the cultivars were sown in 126 earthen pots, allocating 42 pots per genotype, having 36 cm dia. All the pots were filled with 11.5 kg fine field soil. After germination plants were thinned out and single plant per pot was maintained in five pots while five plants per pot were maintained in two pots for each treatment arranged in a completely randomized design with three replicates.

Effluents from the tanneries were collected prior to any treatment from the Water Treatment Plant Kasur and these were kept under shade. The chemical analysis of the wastewater was carried out in the Environment Department, Govt. of the Punjab, Lahore and Department of Botany, GC University, Lahore. Various dilutions *viz.,* To (control), T_1 (20%), T_2 (40%), T_3 (60%), T4 (80%) and T_5 (100%) were prepared by mixing appropriate amount of fresh water with polluted water.

Sunflower plants were maintained throughout the growth period according to local agricultural practices. Plants were irrigated after every 4 - 5 days interval. Plant height (cm), number of leaves per plant, number of senescent leaves per plant, diameter of node (cm) and number of flowers per plant were recorded.

In the mid season (March 10, 2004) destructive harvesting was carried out by randomly selecting two pots per treatments (10 plants per treatment) to determine the shoot and root lengths as well as their fresh and dry biomass. At the completion of vegetative growth (during 2^{nd} week of March, 2004) chlorophyll content of selected leaves of different treatments was determined (Arnon 1949). At maturity capitulum diameter, capitulum fresh and dry weight, number of seeds, weight of seeds, 100- seeds weight, straw weight and harvest index were recorded.

The data collected was analyzed statistically using software package costat version 3.03. Analysis of variance and Duncan's Multiple Range Test as described by Steel and Torrie (1996) was carried out at 0.05% probability.

Results and Discussion

Analytical data of tanneries wastewater of Kasur city, collected on January, 2004 (Table 1) showed that it belonged to irrigation water quality class C_4 - S_4 (Soil Survey Bulletin No.14) and is regarded as poor in quality for irrigation point of view. It had nearly all hazardous metals, minerals and constituents in excess amount compared with National Environment Quality Standards (NEQS). The results of tanneries effluent analysis are in accordance with the findings of Tahira (1999). Irrigation waters which has been used successfully for considerable time has electric conductivity less than 2.25 dS/m and SAR (Sodium Absorption Ratio) values less than 26.

Increasing levels of effluents caused reduction in all the parameters of vegetative growth and biomass in all the cultivars of sunflower i.e., SF-187, CRN-1435 and DK-3915 as compared to control. Moreover, higher effluent concentrations (T_4 and T_5) delayed seedling emergence a few days and also exhibited marked difference in their morphology and appearance as compared to control. The plants from the control and T_1 treatments of all the three sunflower cultivars were healthier and lush green with well developed and expanded leaves as compared to T_2, T_3, T_4 and T_5 treatments. Plants grown in control and T_1 treatments exhibited vigorous growth as compared to rest of the treatments. Wastewater adversely affected the root and shoot development of all the cultivars of sunflower in higher treatment concentration as compared to counterparts grown in controls (T_0). Plant length along with fresh and dry weights of shoots and roots indicated that highest effluent treatments caused severe reduction in the weights of all the cultivars of sunflower as compared to control (Table 2). Difference between various treatment means for the plant length, were highly significant and ranged from 28 - 31, 24 - 26 and 29 - 33% for cultivars SF-187, CRN-1435 and DK-3915 respectively, compared with control. Similarly fresh shoot and root weight was also reduced corresponding to the gradual increase in the effluent concentrations and ranged from 80 - 81% in all the cultivars. Fresh weights of plants from T_0 treatment were highest followed by successive and statistically significant reduction in plants of other treatments. It can be observed from data that root and shoot dry weights were reduced by 75% in cultivar SF-187 than that of 85% in cultivar CRN-1435 and 93% in cultivar DK-3915 in T_5 treated plants when compared with control. A similar trend was also noted in other effluent treatments. The results are in accordance with the findings of Nath *et al*. (2009) who reported that different concentrations of tannery effluents and Cr^{6+} showed significant reduction in germination percentage, seedling growth and pigments with increase in concentrations.

Table 1. Physicochemical variables of tanneries wastewater collected from pretreatment plant of Kasur District.

Variables	Treatments (Effluents concentrations)						
	T_0 (0 %)	T_1 (20 %)	T_2 (40 %)	T_3 (60 %)	T_4 (80 %)	T_5 (100 %)	NEQS*
Color	Colorless	Light grey	Light grey	Black grey	Black grey	Black grey	Grey
Temp. (^{o}C)	20	20	22	22	23	25	24
pH	7.4	7.1	6.9	7.7	8.1	8.42	6-8.5
EC (µS/cm)	0.75	1.8	2.5	3.0	3.6	3.5	1.5
TSS	50	334	757	1845	2063	2150	400
TDS	200	712	1325	4532	4352	85500	2500
BOD	60	257	463	697	868	987	250
COD	92	454	867	1514	1923	2275	400
SAR	9.8	18.2	25.8	32.3	39.8	45.74	10.2
Calcium (meq/l)	50	214	332	450	750	900	200
Magnesium (meq/l)	25	168	285	425	520	625	150
Sulphate (meq/l)	150	512	1050	2280	2562	3525	1000
Sodium (meq/l)	37	57	145	278	364	397	250
Chloride (meq/l)	200	812	1436	2032	2197	2206	1000
Copper µg/g	Nil	Nil	Nil	Nil	Nil	Absent	1.0
Iron µg/g	2.0	2.8	5.5	8.4	10.5	11.5	8.0
Manganese µg/g	0.05	0.04	0.4	0.9	1.5	2.5	1.5
Zinc µg/g	0.08	1.2	1.8	2.1	3.0	4.0	5.0
Cobolt µg/g	Nil	Nil	Nil	Nil	Nil	Negligible	1.0
Lead µg/g	Nil	Nil	Nil	Nil	Nil	Negligible	0.5
Nickle µg/g	Nil	Nil	Nil	Nil	Nil	Negligible	1.0
Chromium** µg/g	Nil	204	750	1598	1220	3550	1.0
Hardness	50	215	550	985	1758	1924	500

*NEQS source: EPA (2007). NEQS: National Environmental Quality Standards for Municipal wastewaters of Pakistan (values in mg/l unless otherwise defined); **Chromium as both trivalent and hexavalent)

The amount of chlorophyll *a* was less than chlorophyll *b* in the plants of all the treatments (Table 2). Percentage reduction in effluents treated plants relative to control was significantly higher and were not much greener and also the senescence started earlier. Table 2 also shows that the amount of chlorophyll (chl. *a* and *b*) was high in plants from T_0 and T_1 treatments followed by successive statistically significant gradual reduction in plants of increasing effluent levels. The reduction in photosynthetic material in plants grown in high effluent concentrations produced comparatively less productivity than the plants grown in less polluted habitat (Camplin 2001).

Table 3. Effects of tanneries effluents on yield of sunflower at maturity during 2004 growth season.

Sunflower Cultivar	Treatments (effluents %)	Capitulum fresh wt. (g)	Capitulum dry wt. (g)	Capitulum diam. (cm)	Seeds per plant	Seed wt/ plant (g)	Straw wt/ plant (g)	Harvest index
SF- 187	T_o (Cont)	49.70a	45.09a	8.00a	313.0a	44.02a	22.65a	1.94a
	T_1 (20)	33.20b	32.72b	6.7b	262.0b	30.04b	18.72b	1.60a
	T_2 (40)	24.83c	19.09c	5.40c	211.0c	17.45c	11.09c	1.57a
	T_3 (60)	22.22d	17.14cd	4.30cd	168.0d	15.89cd	10.14cd	1.56a
	T_4 (80)	22.18d	16.83cd	3.52de	138.0e	13.49d	8.83cd	1.52a
	T_5 (100)	20.06d	15.15d	3.11e	121.0f	12.31d	8.15d	1.51a
	L.S.D.	2.413	2.330	1.137	2.408	3.619	2.697	0.639
CRN- 1435	T_o (Cont)	54.90a	52.69a	9.80a	383.0a	50.85a	27.30a	1.86a
	T_1 (20)	46.20b	42.00b	9.00ab	352.0b	40.25b	21.90b	183a
	T_2 (40)	39.34c	35.27c	8.52b	333.0c	33.73c	18.88c	1.78a
	T_3 (60)	32.35d	30.02d	6.80c	266.0d	28.69d	18.57c	1.54a
	T_4 (80)	22.14e	17.30e	5.40d	211.0e	15.26e	10.37d	1.46a
	T_5 (100)	20.90e	15.40e	3.80e	148.0f	11.19e	8.22d	1.36a
	L.S.D.	3.615	3.785	0.895	2.082	4.600	2.772	0.923
DK- 3915	T_o (Cont)	61.71a	59.75a	10.65a	416.0a	57.38a	33.57a	1.70a
	T_1 (20)	52.09b	49.20b	7.45b	291.0b	47.25b	28.06b	1.68a
	T_2 (40)	36.54c	34.05c	6.50c	254.0c	32.80c	20.81c	1.58a
	T_3 (60)	30.84d	26.49d	5.30d	207.0d	22.42d	14.44d	1.55a
	T_4 (80)	26.44e	23.48d	4.75e	186.0e	20.03d	13.82d	1.44a
	T_5 (100)	10.77f	8.42f	3.75f	147.0f	5.75e	4.65e	1.23a
	L.S.D.	2.982	1.837	05.32	1.167	4.540	2.242	1.193

Treatment means followed by different letters in the same column are significantly different at $p = 0.05$ according to Duncan's Multiple Range Test; L.S.D.: Least Significant Difference at $p = 0.05$.

Early senescence in plants treated with high doses along with reduction in other vegetative parameters than that of counterparts plants treated with lower doses in present study is strongly supported by Wahid *et al.* (2000) and Gilbert and Dubey (2003).

Table 2. Effects of tanneries effluents on yield and biomass of 11week old sunflower cultivars at midseason harvest during 2004 growth season.

Sunflower cultivars	Treatments (Effluents %)	Plant length (cm)		Fresh weight (g)		Dry weight (g)		Chlorophyll (mg/g fresh tissue)	
		Shoot	Root	Shoot	Root	Shoot	Root	a	b
SF- 187	T_o (Cont)	109.42a	9.27a	41.22a	3.27a	19.53a	2.05a	0.027a	0.033a
	T_1 (20)	102.38a	7.38ab	22.47b	2.37a	14.80b	1.42ab	0.025b	0.032a
	T_2 (40)	82.10b	6.81ab	10.47c	1.11b	8.75c	0.73bc	0.021c	0.027b
	T_3 (60)	79.45b	6.60b	9.93cd	0.89b	8.23.c	0.66bc	0.018d	0.026bc
	T_4(80)	77.58b	6.49b	9.80cd	0.85b	4.89d	0.60bc	0.015e	0.024c
	T_5 (100)	77.53b	5.48b	8.12d	0.79b	4.89d	0.42c	0.011f	0.021d
	L.S.D.	12.607	2.475	1.997	0.941	2.662	0.819	0.002	0.002
CRN- 1435	T_o (Cont)	126.26a	12.16a	35.30a	18.19a	19.77a	5.69a	0.034a	0.047a
	T_1 (20)	123.41b	9.20b	23.42b	3.53b	17.95ab	2.20b	0.026b	0.042b
	T_2 (40)	115.42c	9.08b	20.39c	3.37b	15.60b	1.74bc	0.024c	0.039c
	T_3 (60)	107.32d	7.99c	18.50d	2.50bc	15.51b	1.73bc	0.021d	0.038c
	T_4 (80)	96.16e	7.18e	11.46e	1.72c	6.93c	1.43bc	0.017e	0.035d
	T_5 (100)	95.06e	7.16c	10.86e	1.56c	3.40d	0.61e	0.014f	0.030e
	L.S.D.	1.530	1.014	1.560	1.385	2.618	1.277	0.002	0.002
DK- 3915	T_o (Cont)	126.78a	8.81a	50.59a	10.39a	29.36a	1.84a	0.038a	0.050a
	T_1 (20)	109.46ab	8.07a	45.21b	8.35b	25.10b	1.00ab	0.030b	0.046b
	T_2 (40)	96.40b	7.02a	39.99c	7.30c	18.57c	0.99ab	0.024c	0.038c
	T_3 (60)	95.82b	6.96a	30.29d	6.02d	9.56d	0.81b	0.021d	0.035d
	T_4 (80)	89.00b	6.80a	25.08e	5.51e	9.56d	0.81b	0.019e	0.034d
	T_5 (100)	88.06b	6.31a	15.04f	3.38f	1.43e	0.55b	0.017e	0.031e
	L.S.D.	20.619	2.949	0.650	0.750	2.467	0.940	0.002	0.002

Treatment means followed by different letters in the same column are significantly different at $p = 0.05$ according to Duncan's Multiple Range Test; L.S.D.: Least Significant Difference at $p = 0.05$.

At final harvest, the growth was also higher in control and as the pollutant treatment successively increased the growth of plants was gradually reduced (Table 3). All the parameters of reproductive growth decreased significantly from T_2 to higher effluent concentrations i.e., T_4 and T_5 in all the cultivars of sunflower. Capitulum diameter was greater in control plants in all the cultivars and exhibited gradual decrease with corresponding increase in effluent treatments. The magnitude of reduction in capitulum diameter in T_5 for cv. DK-3915 remained higher (86%) as compared to cv. SF-187 (64%) and cv. CRN-1435 (70%), depicting that DK- 3915 cultivar is more sensitive. Seed production was higher in control (To) plants while gradual significant loss in seeds number was observed in all the effluent treated plants. The reduction in seed number of T_5 treated plants in cultivar SF-187 was 61% while it was 62% in CRN-1435 and 65% in DK-3915 depicting its greater sensitivity to seed production. Plants treated with T_4 and T_5 were extremely stunted, so they produced fewer light weight seeds. Percentage differences recorded were highly different and statistically significant in higher pollution levels. Seed weight was also higher in T_0

and T_1 treatments as compared to seeds produced in T_4 and T_5 treatments. The magnitude of reduction in seed weight per plant in T_5 compared to control was 72% in cultivars SF-187, while it was a bit higher in cultivar CRN-1435 (78%) and DK-3915 (90%) exhibiting drastic affect on seed production and seed weight of sunflower cultivars. 100- seed weight was also gradually reduced in higher pollution levels. Harvest index was higher in plants collected from control and T_1 treatments when compared with that of higher effluents treatments. The magnitude of reduction was higher in cv. DK-3915 (27%) as compared to cv. SF-187(23%) and cv. CRN-1435 (22%). The results are in line with Gillbert and Dubey (2003) who reported that biomass reduction by effluents in first step results in severe yield reduction at latter stages of growth.

So, high concentrations of effluents (80 - 100%), proved deleterious to plant growth, both at vegetative and reproductive stages. Effluents treatment especially at higher concentrations along with the increased EC values of treated soil suggested that treatment with higher effluent concentrations induced some kind of stress on plants (salinity stress), which affected various metabolic processes resulting in reduction of vegetative and later on reproductive growth of the plant. Hewitt and Keller (2003) reported injurious effects of wastewater of a chemical manufacturing plant on the growth of maize, soybean and wheat. Similar views were also reported by other investigators (Kilicel and Dag 2006, Yasir 2003, Rusan *et al*. 2007).

However, diluted effluents may be used for the crop. Significant growth and yield losses due to tannery effluents is a matter of serious concern from view point of agriculture as Pakistan is an agrarian country and its soils are being contaminated with toxic loads of industries.

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