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AMELIORATION OF ADVERSE EFFECTS OF SIMULATED ACID RAIN (SAR) ON GROWTH AND YIELD ATTRIBUTES OF SUNFLOWER (*HELIANTHUS ANNUUS* L.) BY GROWTH TONICS

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Abstract

A pot experiment was conducted to mimic the adverse effects of air pollution in the form of simulated acid rain (SAR) on morphological and yield attributes of two sunflower (*Helianthus annuus* L.) hybrids viz. FH-385 and FH-37 and their amelioration with the application of two growth tonics (Fashion and Micron-T i.e., macro & micronutrients). The experiment was performed in the net house of Old Botanical Garden, University of Agriculture, Faisalabad, in a Completely Randomized design with four replicates. Simulated acid rain (containing HNO_3 at pH 3) alone and in combination with foliar spray of Fashion or Micron-T were applied 20 days after seed germination. Acid rain treatment remarkably reduced growth and yield in both hybrids while the application of Fashion excelled Micron-T in ameliorating the toxic effects of acid rain. A comparison between sunflower hybrids indicated FH-37 to be more tolerant to SAR as compared to FH-385. Based on these findings it may be recommended that growth tonics may ameliorate the toxic effects of SAR. There is need for further investigation to improve agricultural produce using such growth tonics.

Introduction

Air pollution has become a worldwide critical problem for the healthy existence of biota (Ling *et al.*, 2010). The important air pollutants i.e., oxides of sulphur (SO_2), nitrogen (NOx) and carbon (CO_2) get emitted into atmosphere through industries and fossil fuels burning. After reacting with atmospheric moisture they reach living surface or bare soil as acid deposition which considerably affects soil chemistry by leaching some essential micronutrients and detrimentally influences agricultural produce especially at its pH < 3 (Bouwman *et al*., 2002; Singh & Agrawal 2008).

Micro-nutrients (Zn, Fe, B, Cu) play an essential role during plant growth and metabolism. As in many other Asian countries, Pakistani soils being calcareous possess high pH and owing to prolonged drought experience salt stress. The high bicarbonate contents in irrigation water and imbalanced application of fertilizers remain other constraints for the cultivated crops (Imtiaz *et al*., 2010) which get exacerbated by acid rain. The adverse effects of acid rain on soil attributes and agricultural crops can be minimized with the addition of micronutrients to the soil. In this regard foliar application of growth tonics comprising some micronutrients has also shown promising results (Akhtar *et al.*, 2009; Alam *et al*., 2010). In present study two growth tonics were used as foliar spray. These growth tonics have been introduced by Green Crop Pvt. Limited., Pakistan with trade names i.e., "Fashion" comprised of macronutrients (N = 8%, P_2O_5 = 8% and K_2O = 6% w/v) and mixture of micronutrients named "Micron –T" (Zn = 6%, Fe = 2% and Mn = 2%), both manufactured by Apna Zarai Markaz Lahore with the claim to improve plant growth.

Some studies relating to effects of SAR at pH 3 on different plant species indicated development of visible injury symptoms on leaves (Saubhagy, 2003) and metabolic changes (Anna-Santos *et al.*, 2005). Nevertheless, the use of micronutrients has been reported to substantially increase seed yield in mungbean (Panpruik *et al*., 2002) and onion (Alam *et al*., 2010) and oil contents of *Mentha piperita* (Akhtar *et al.*, 2009). In a resembling study the application of nutrient solutions increased fresh biomass of sunflower and safflower plants under stressful conditions (Jabeen & Ahmed 2012).

Sunflower (*Helianthus annuus* L.) is an important oil yielding crop. It can potentially fit in the agricultural cropping system in Pakistan. Its annual production in Pakistan is 619.4 thousand tons (Anon., 2012; Mukhtar *et al*., 2009). Its vegetable oil is a healthy supplement of calcium, nicotinic acid and vitamin E (Skoric *et al*., 2008). However its production in the country remains low as compared to some developed countries of the world owing to a variety of biotic and abiotic environmental stresses. Faisalabad is an industrial city where many toxic gases are emitted by various industries. Therefore, the purpose of this study was to examine the consequences of foliar application of acid rain separately and in combination with growth tonics on different attributes of sunflower (*Helianthus annuus* L.).

Materials and Methods

The seeds of 2 sunflower hybrids *i.e*., FH-385 and FH-37 obtained from Ayub Agricultural Research Institute (AARI) Faisalabad were sown in plastic pots lined with polyethylene bags each having 10 kg of soil and plant manure at a ratio 3:1. After the germination, the plants were thinned to six plants in each pot. Simulated acid rain (SAR) of HNO_3 at pH 3 alone and in mixture with growth tonics i.e., Fashion or Micron-T (each @ 1250 mL ha^{-1}) comprising macro and micro-nutrients were sprinkled once 20 days after germination of the crop.

The data for relative increase per week for four consecutive intervals were recorded for various growth attributes and converted to per day growth rate. The root and shoot lengths of each hybrid were measured with the help of a measuring tape after the harvest. The fresh weight of plant of each hybrid was measured on analytical balance (AND-GF-300), thereafter samples were oven-dried at 75°C to get a constant dry weight. The yield parameters were taken after the crop matured. The data

were analyzed through the statistical software COSTAT v 6.303 (Cohort software, Monterey, California) (Steel & Torrie 1984).

Results and Discussion

The growth of both sunflower hybrids was negatively affected by the application of acid rain. This deteriorating effect was observed in all vegetative parameters. Shoot length of FH-385 plants was reduced up to 24.85% by simulated acid rain (T_1) during 1st harvest as compared to control (Table 1). Macro- & micro-nutrient application of Fashion (T_2) and Micron-T (T_3) increased growth rate of shoot up to 10.05% and 9.46% as compared with control (T_0) for FH-385. In the combined application of SAR and growth promoters, the adverse effects of acid rain got reduced to 18.93% (T_4) and 20.11% (T_5) as compared to control (T_0). A similar tendency was observed in shoot length during 2nd and 3rd harvest interval for both hybrids.

During the 1st and 2nd harvest effect of SAR was more adverse on root growth in FH-37 as compared to FH-385 (Table 1). The joint application of SAR & growth promoters had remarkable positive effect on growth rate. It seems SAR reduced shoot and root lengths in both hybrids. The combination of SAR and growth tonics reduced the adverse effects of HNO_3 to some extent and showed positive impact on both sunflower hybrids.

The more pronounced effect of foliar application of SAR (T_1) was observed during the 2nd harvest of crop after the spray of SAR. The plant dry weight was reduced by 39.72% and 34.88% due to SAR in FH-385 and FH-37 respectively during the 2nd harvest. In T_2 and T_3 this reduction seems to be sustaining in Fashion and Micron-T treated hybrids when compared to their control plants (T_0) (Table 2). The deteriorating effect of acid rain that was 39.72% in FH-385 got reduced to 20.54% and 23.28% in T_4 and T_5 due to the effect of macro & micronutrients. The negative effect of acid remained less prominent during 3rd harvest. The reduction in plant dry weights of FH-385 and FH-37 was due to induced acid stress but their adverse effects were reduced by the growth tonics. FH-37 proved comparatively more tolerant to SAR than FH-385. A similar trend was observed for plant fresh weights in both sunflower hybrids (Table 2).

Capitulum diameter was very much affected by the application of SAR and growth tonics. Statistically highly significant differences were noted among various treatments regarding this parameter (Table 3). Spray of HNO_3 at pH 3 (T_1) showed a strong negative effect on size of capitulum in both hybrids (Fig. 1). Treatment of plants with growth tonics enhanced the capitulum diameter and slightly better results were indicated by Fashion (T_2) as compared to its counterpart Micron-T (T_3). The deteriorative effect of SAR was reduced to some extent in treatments when growth tonics were applied along with acid spray. Fashion excelled Micron-T for counteracting the negative effect of SAR. The two hybrids differed significantly in their response to various treatments. FH-385 performed better than FH-37.

Capitulum dry weight was also very much affected by the SAR. The application of acid (T_1) strongly reduced the capitulum dry weight in both hybrids. Growth tonics reduced the adverse effect of acid rain on capitulum dry weight in T_4 and T_5. SAR effect was suppressed by the growth tonics in their combined application either with Fashion (T_4) or Micron-T (T_5). FH-385 gave better results as compared to FH-37 as shown in the Fig. 2.

Hundred achene weight was significantly reduced by the SAR treatment (T_1). Applications of Fashion (T_2) and Micron-T (T_3) enhanced the 100 achene weight as compared to control (T_0). They also imposed a positive impact when used in combination with acid. They ameliorated the adverse effects of acid rain to some extent in T_4 and T_5. The Fashion imparted more positive effect on 100 achene weight as compared to Micron-T. Among hybrids the FH-385 performed better than FH-37 under the effect of all the treatments (Fig. 3).

Achene weight per plant was very much affected by the SAR. The worst effect of SAR was examined in T_1 as it gave the minimum achene weight per plant in both hybrids. This negative effect of SAR was overcome by the effect of growth tonics when they were applied in combination with SAR (T_4, T_5). The Fashion (T_2) and Micron-T (T_3) treated plants gave more achene weight as compared to control (T_0). Both hybrids gave better response with the growth tonic Fashion as compared to Micron-T. However FH-385 showed slightly better achene weight than FH-37 as shown in Fig. 4.

Simulated acid rain application caused negative effect on understudy sunflower hybrids. It decreased the growth in both sunflower hybrids. Previous investigations have also shown similar toxic effects of acid rain on kidney bean and tomato plants (Liao *et al*., 2005; Shaukat & Khan, 2008). Zhou *et al*., (2010) also recorded reduction in growth of soybean with the application of SAR. Acid rain stress reduced yield in both sunflower hybrids in present study. These findings are in close conformity with Fan & Wang (2000). A decrease in yield of green pepper (*Capsicum annuum* L.) was also noticed by Shri & Naresh (2000). This might be due to rapid transfer of chlorophyll to pheophytin 'a' which leads to low photosynthetic rate and ultimately low yield. Same results have been obtained for wheat (Kausar & Khan, 2009).

The application of macro- and micronutrient during this study reduced the adverse effects of acid rain. Phosphorus is present in growth tonic Fashion. Phosphorus being essential element of the ribonucleic acids (RNA) is required in many physiological and biochemical processes including protein metabolism and energy transfer (Prabhu *et al*., 2007). Hence the application of macro & micronutrients may have increased the dry matter production of both sunflower hybrids. Similar results were obtained by Lawlor (2002), Rehman *et al*., (2013) and Hossain & Hamid (2007). This increase in dry matter production may be attributed to the application of high N_2 content in the "Fashion" which might have increased the leaf area and thus enhanced light interception and photosynthesis (Yin *et al*., 2003; Lemaire *et al*., 2005; Masinde *et al*., 2009). Moosavi (2012) also reported increase in grain yield of maize under the effect of different levels of nitrogen fertilizer.

Table 1. Growth in shoot length (cm) and root length (cm) of two sunflower hybrids (FH-385 and FH-37) in response to SAR and growth tonics application.

	FH-385						FH-37					
	T_0	T_1	T_2	T_3	T_4	T_5	T_0	T_1	T_2	T_3	T_4	T_5
Growth in shoot length												
Growth per day in 1st harvest	1.69±0.02	1.27± 0.12	1.86± 0.18	1.85± 0.25	1.37± 0.28	1.35±0.21	1.60± 0.08	1.30± 0.23	1.75± 0.12	1.72± 0.11	1.41± 0.43	1.34± 0.38
% difference		-24.85	10.05	9.46	-18.93	-20.11		-18.75	9.37	7.50	-11.87	-16.25
Growth per day in 2nd harvest	1.50±0.26	0.73±0.14	1.27±.0.26	1.26±0.26	0.80±0.17	0.60±0.21	1.61±0.31	0.88±0.42	1.04±0.34	1.02±0.39	1.30±0.44	1.30±0.44
% Difference		-51.3	15.3	16.0	-46.6	-47.3		-45.34	5.6	4.34	-19.25	-19.25
Growth per day in 3rd harvest	1.60±0.31	0.70±0.21	2.30±0.64	2.04±0.49	0.90±0.32	0.90±0.32	1.65±0.57	1.05±0.41	1.81±0.57	1.70±0.41	1.3±0.23	1.1±0.21
% Difference		-56.2	43.7	27.5	-43.75	-43.75		-36.36	9.70	3.03	-21.21	-33.33
Growth in root length												
Growth per day in 1st harvest	0.12±0.05	0.06±0.02	0.16±0.03	0.14±0.04	0.10±0.03	0.09±0.02	0.12±0.02	0.05±0.02	0.17±0.01	0.13±0.03	0.09±0.02	0.08±0.03
% Difference		-50	33.33	16.66	-16.66	-25.0		-58.33	41.66	8.33	-25	-33.33
Growth per day in 2nd harvest	0.14±0.05	0.06±0.02	0.19±0.08	0.18±0.06	0.13±0.04	0.12±0.04	0.10±0.04	0.04±0.01	0.13±0.04	0.12±0.04	0.09±0.03	0.08±0.02
% Difference		-57.14	35.71	28.57	-7.14	-14.28		-60	30.00	20.0	-10.0	-20.0
Growth per day in 3rd harvest	0.14±0.04	0.06±0.02	0.16±.038	0.13±0.05	0.11±0.04	0.10±0.04	0.11±0.04	0.06±0.02	0.13±.05	0.12±0.02	0.09±0.01	0.08±0.01
% Difference		-57.14	14.29	7.14	-21.42	-28.57		-45.4	18.18	9.09	-18.18	-27.27

T_0= Control, T_1= HNO_3 (pH 3), T_2= Fashion (1250 mL/ ha), T_3= Micron-T (1250 mL/ ha), T_4= Fashion + HNO_3 (pH 3), T_5= Micron-T + HNO_3 (pH 3)

Table 2. Growth in plant fresh weight (g) and plant dry weight (g) of two sunflower hybrids (FH-385 and FH-37) in response to SAR and growth tonics application.

	FH-385						FH-37					
	T_0	T_1	T_2	T_3	T_4	T_5	T_0	T_1	T_2	T_3	T_4	T_5
Plant fresh weight												
Growth per day in 1st harvest	3.68±0.34	3.05±1.20	4.03±0.42	3.82±0.58	3.52±0.28	3.48±0.32	2.52±0.47	2.13±0.42	3.23±0.31	2.48±0.49	2.5±0.67	2.42±0.9
% difference		-17.1	9.51	3.80	-4.3	-5.43		-15.4	28.17	1.58	-0.79	-3.96
Growth per day in 2nd harvest	6.04±1.18	4.80±0.73	7.08±1.28	6.88±0.35	6.03±0.92	5.65±0.49	5.31±0.09	4.29±0.97	5.87±0.79	5.64±0.66	4.5±1.72	4.80±0.8
% Difference		-20.72	17.21	13.90	-0.16	-6.45		-19.20	10.54	6.21	-7.72	-9.60
Growth per day in 3rd harvest	9.57±0.38	8.62±0.39	9.96±0.91	9.8±0.60	9.1±1.16	9.05±0.65	8.4±0.50	7.13±0.70	8.93±0.65	8.72±0.67	7.91±0.87	7.45±0.7
% Difference		-9.92	4.07	2.40	-4.91	-5.43		-15.12	6.31	3.81	-5.83	-11.30
Plant dry weight												
Growth per day in 1st harvest	0.39±0.18	0.25±0.19	0.42±0.22	0.40±0.11	0.28±0.02	0.26±0.04	0.19±0.09	0.17±0.15	0.25±0.23	0.21±0.14	0.18±0.03	0.18±0.3
% Difference		-35.89	7.69	2.56	-28.21	-33.33		-10.52	31.57	10.52	-5.26	-5.26
Growth per day in 2nd harvest	0.73±0.15	0.44±0.28	1.0±0.06	0.99±0.22	0.58±0.21	0.56±0.12	0.43±0.27	0.28±0.25	0.57±0.24	0.53±0.17	0.42±0.24	0.40±0.2
% Difference		-39.72	36.98	35.61	-20.54	-23.28		-34.88	32.55	23.25	-2.32	-6.97
Growth per day in 3rd harvest	1.3±0.15	1.11±0.34	1.44±0.18	1.32±0.22	1.25±0.12	1.22±0.15	1.11±0.28	0.99±0.09	1.22±0.27	1.19±0.18	1.09±0.14	1.07±0.0
% Difference		-14.61	10.76	1.53	-3.85	-6.15		-10.81	9.91	7.21	-1.80	-3.60

Table 3. Analysis of variance (ANOVA) of yield of two sunflower hybrids under simulated acid rain (SAR) and growth tonics application.

Source of variation (SOV)	df	Capitulum diameter (Cm)	Capitulum dry weight (g)	100 achene weight (g)	Achene weight / plant (g)
Treatment (Trt)	5	14.1786 ***	9.8529***	13.8261***	9.6530***
Hybrid (Hyb)	1	1.7519***	4.3495***	2.0295*	0.6327*
Trt x Hyb	5	0.1069^{ns}	0.3141^{ns}	0.1604^{ns}	0.0205^{ns}
Error	36	0.1398	0.2433	0.3949	0.1193

ns = Non-significant; *, ** and *** Significant at 0.05, 0.01 and 0.001 levels respectively

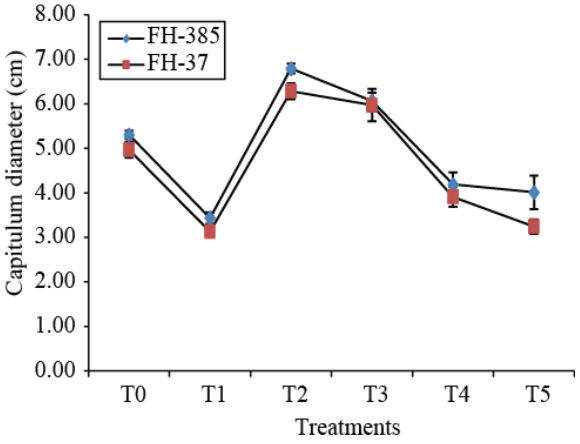

Fig. 1. Capitulum diameter under the effect of all the treatments.

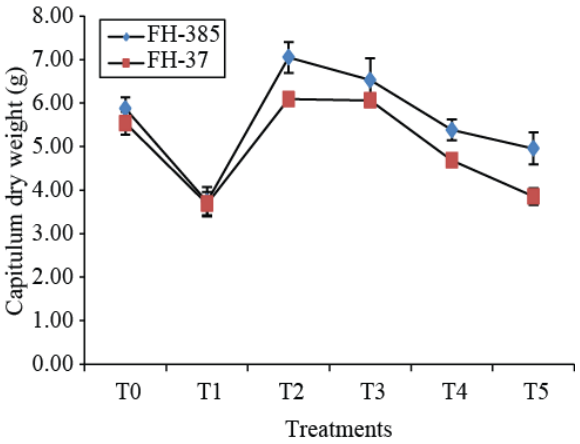

Fig. 2. Capitulum dry weight under the effect of all the treatments.

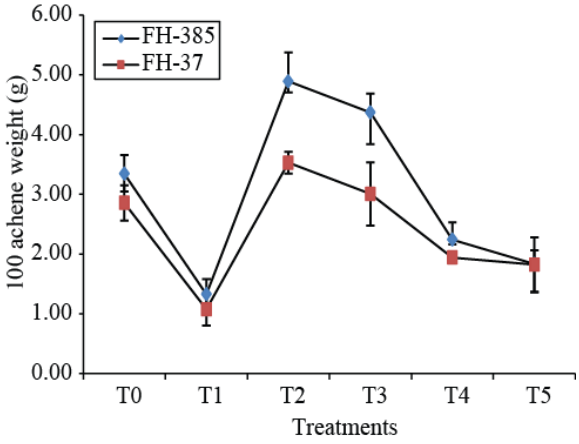

Fig. 3. 100 achene weight under the effect of all the treatment.

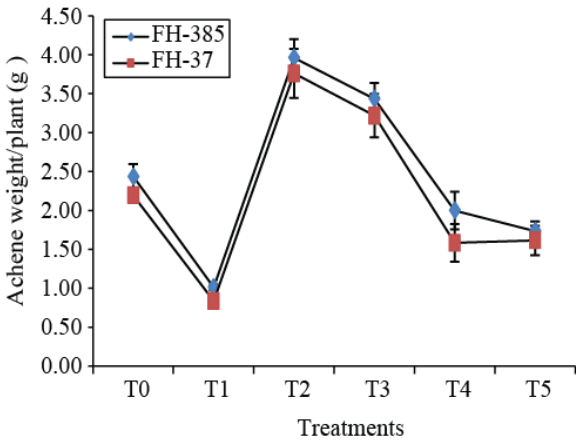

Fig. 4. Achene weight per plant under the effect of all the treatments.

It is possible that negative effect of SAR can be reduced with the addition of micronutrients either in soil or foliar spray. Macro & micronutrients are required in minute quantity and considered essential component of biomolecules (DNA and RNA synthesis proteins, carbohydrates, alcohols and fats). These increase quality of the crop and enhance production efficiency (Adejumo, 2010). The inadequate absorption of micronutrients from soil has been attributed to substantial yield losses in different edible crops (Malakouti, 2008; Habib, 2009; Imtiaz *et al*., 2010).

During this study application of SAR considerably affected the availability of nutrients but the application of macro & micronutrients (Fashion & Micron-T) caused positive effects in both sunflower hybrids by ameliorating the adverse effects of acid rain. More pronounced effect of SAR was seen on growth of FH-385 which proved less tolerant against acid stress.

Conclusions

During this study SAR drastically reduced the growth of both sunflower hybrids, while the combined application of growth tonics (macro- & micro-nutrients) along with SAR reduced the adverse effects of acid rain in both hybrids and gave better growth as compared to application of acid rain alone. Yield was also severely affected by the acid rain, however, growth tonics

counteracted the adverse effects of acid rain. The results showed that FH-37 proved to be more tolerant hybrid towards acid rain as compared to FH-385. On the basis of the present study it is recommended that growth tonics should be used in agricultural practices to overcome the hazardous effects of air pollution.

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