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EFFECTS OF SIMULATED ACID RAIN ON THE GERMINATION AND GROWTH OF *AMARANTHUSDUBIUS*

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The experiments were conducted to investigate the effect of simulated acid rain on the germination and the growth of Amaranthusdubius Mart. ex Thell. The effect of simulated acid rain using sulphuric acid with different pH concentrations (2, 3 and 4) were determined by exposing the seeds and seedlings to them for ten consecutive days. A control (pH 6.8) was placed along with the treatments. The seed germination was found reduced at pH 2. The effect on seedlings showed remarkable reductions on the growth parameters and the chlorophyll content. A large number of reductions were noted at pH 2. The leaves were with marked irregular lesions, necrotic spots and chlorosis at pH 3 and pH 2 levels. The total phenol content showed an increasing trend as pH level reduced from 4 to 2. Thus this study points out that Amaranthusdubius Mart. ex Thell. has less tolerance to acid rain which demands new resistant varieties through breeding and other plant propagation methods.

Keywords: *Amaranthusdubius,* simulated acid rain, seed germination, growth

Air pollution becomes a worldwide critical problem for the healthy existence of living organisms. The term acid rain is used to mean the deposition of acidic components in rain, snow, fog, dew or dry particles. Unpolluted rain is slightly acidic, because carbon dioxide and water in the air react together to form carbonic acid, a weak acid. Rain acquires additional acidity through the reaction of air pollutants with water in the air, to form strong acids (such as sulphuric acid and nitric acid). The main sources of these pollutants are emissions from vehicles, industrial plants and power generating plants (Kita et al., 2004).The pH of water in equilibrium with atmospheric CO2 is approximately 5.6 and hence only rain events with a pH lower than this value are classified as acid rain(Elsom, 1987).

The studies have shown that acid rain has serious negative effects on the vegetative organs of plants and it induces modification to the physiology and cellular biochemistry of plants (Babu and Madhavan, 2011; Lal and Singh, 2012;Odiyi and Bamidele, 2014). Biological effects of acid rain on plants are vast and complex and include visible symptoms such as chlorosis and necrosis and non-visible effects such as diminished photosynthesis and leaf nutrient loss. The effect of simulated acid rain (pH 2) on fir seedlings, investigated by Izutaet al. (1993) reported severe damage such as visible foliar injury and depression of growth. The present study focuses analyzing the effect of different pH of simulated acid rain (SAR) on Amaranthusdubius Mart. ex Thell and it gives awareness regarding the effect of SAR on different growth parameters of Amaranthusdubius Mart. ex Thell.

Materials and Methods

The present investigation is concerned with theeffects of simulated acid rain (SAR) on the germination and growth of Amaranthusdubius Mart. ex Thell. and this plant is used as leafy

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vegetable in Kerala.

i) Preparation of simulated acid rain: 1 N Sulphuric acid was prepared by diluting 4.9 ml of conc. H2SO4in 1 litre of distilled water. It was then calibrated using 1% NaOH and distilled water to the desired pH (2, 3 and 4) using pH meter.

ii) Test on seed germination: The seeds of Amaranthusdubius Mart. ex Thell.were collected from Agricultural university, Mannuthy, Kerala. About 25 of them were allowed to germinate to check the viability. Then 50 of the seeds were allowed to germinate in cotton soaked with different pH solutions in a seed tray. Theseeds were placed in a way of 10 seeds in one cotton piece. 250 ml of pH (2, 3 and 4) solutions prepared were sprayed to them. A control set was placed having cotton soaked with distilled water along with each treatment. The data was collected on the 11th day of the treatment and each treatment had 50 replicates along with control set and repeated three times. The percentage of germination was analyzed.

iii) Test on seedling growth: One month old seedlings of Amaranthusdubius Mart. ex Thell. were grown underex vitro conditions. Of these healthy ones were selected and transferred to the polythene bags having 15 cm diameter, which were filled with autoclaved soil and manure in the ratio 2:1. 200 ml of (pH 2, pH 3 and pH 4) was showered separately for ten minutes at a time for the ten consecutive days for each treatment. The control plants were sprayed with distilled water. Each treatment was repeated three times with five replicates each along with control.

On 11th day the plants were uprooted and growth parameters such as (i) height of the plant,(ii) number of leaves, (iii) fresh weights of root and stem,(iv) circumference of the stem,(v) single leaf area and (vi)root lengthwere studied. Biochemical parameters like chlorophyll content (Arnon, 1949) and total phenol content (Bray and Thorpe, 1954) were also estimated.

Results and Discussion

i)Effect on seed germination

The simulated acid rain effects the germination of the seeds by reducing the average germination percentage at all different pH levels. Highest reduction was seen at pH 2 (Fig. 1).

ii)Effect on physical growth parameters

The SAR application on seedlings also results in reduction in growth parameters at all the three pH levels studied compared to the control(Table 1). The reduction was more significantly seen at pH 2.

The chlorotic and necrotic spots appeared in the leaves from the 7th day of the treatment at all the levels of pH studied. The plants exposed to pH 3 were more subjected to chlorosis. Necrotic symptoms were observed at pH 2 and pH 3. Leaf damages including primary abscission and foliar injuries were observed from the 3rd day of the treatment and then leaves get completely dried off within the onset of 8th day. Similar kind of observations was noted by Sant'Anna-Santos et al. (2006) on the leaf blades of Genipaamericanaat pH3. The change of pigments involved in photosynthesis is linked to the variation of the photosynthetic process, with drastic consequences on the metabolism of plants.

The reduction of germination of the seeds was found in rice seeds at lower pH as reported by Zabawiet al. (2008). Babu and Madhavan (2011) also reported similar observations in all the growth parameters on Phaseolusradiatus L. and Phaseolus vulgaris L. The impact of simulated acid rain with pH levels of 5, 4 and 3 was studied by Vermaet al. (2010) in Capsicum annuum, Lycopersiconesculentum and Solanummelongena and the growth parameters was severely curtailed with increasing acidity in all these three species. Odiyi and Bamidele (2014) reported reduction in growth parameters in ManihotesculentaCrantz. Simulated acid rain studies in Helianthus annuusL. revealed that biomass and length (root and shoot) of the plant decreased with decreasing pH (Lal and Singh, 2012).

iii)Effect on biochemical parameters

The chlorophyll content, which is the photosynthetic pigment, was also inhibited with respect to all acidity levels (Fig. 2). Varshney et al. (2005) also noticed similar results in radish. Higher amounts of phenol accumulation were seen in the leaves of Amaranthusdubius Mart. ex Thell. subjected to pH 2 and pH 3 compared to the control plants (Fig. 3). Zobel (1996) reported that atmospheric pollutants can induce an increase in

the amount of phenol content in plants.

Summary and Conclusion

The effect of simulated acid rain on the germination and growth parameters reveals that simulated acid rain at lower pH levels had great impacts germination percentage and all other growth parameters of Amaranthusdubius Mart. ex Thell. The highest amount of reduction was seen at pH 2. The chlorophyll content was also seen decreased with the lowering of pH. This indicates the inhibition of photosynthetic pigments at high acidity levels. The phenol accumulation in the leaves shows the necessity of reducing atmospheric pollution. The primary abscission of leaves, chlorosis and necrosis were also the symptoms of toxicity to the leaves and thereby the whole plant due to air pollutants.

Thus this study points out that AmaranthusdubiusMart. ex Thell. has less tolerance to acid rain which demands new resistant varieties through breeding and other plant propagation methods. The foliar applications of some growth tonics comprising micronutrients also help in developing the resistance to withstand simulated acid rain in plants. Controlling the industrialization and air pollution canreduce the impact of acid rain.The present study is very useful in getting the information about simulated acid rain treatment and its effects.

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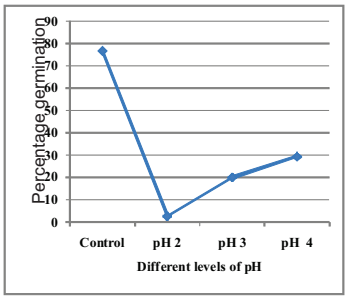

Fig. 1. Average germination percentage of seedsafter application of SAR.

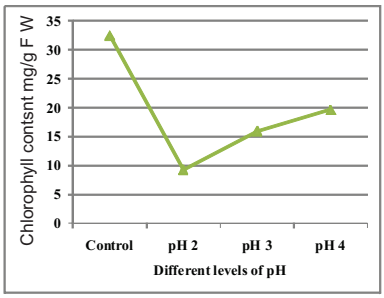

Fig. 2. Chlorophyll content after application of SAR.

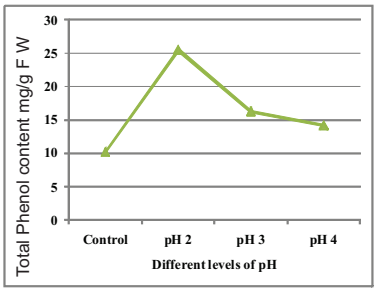

Fig. 3. Total phenol content on leaves after application of SAR.

Table 1. Effect of SAR on growth parameters

Growth parameters	pH levels			
	pH 2.0	pH 3.0	pH 4.0	Control
Height (cm)	8.17±0.3	11.32±0.2	14.63±0.2	25.81±0.7
Number of leaves	1.39±0.3	2.43±0.7	2.69±0.3	5.20±0.6
Circumference (cm)	0.44±0.07	0.59±0.04	0.86±0.03	1.42±0.08
Single leaf area (cm^2)	5.89±0.5	8.93±0.7	10.89±0.3	15.13±0.6
Leaf weight (g)	0.08±0.02	0.12±0.02	0.19±0.02	0.23±0.02
Total leaf weight (g)	0.1±0.02	0.29±0.11	0.5±0.07	1.23±0.2
Root weight (g)	0.04±0.00	0.06±0.03	0.09±0.01	0.15±0.01
Stem weight (g)	0.09±0.75	0.12±0.02	0.16±0.04	0.25±0.08
Root length (cm)	0.75±0.07	1.5±0.09	2.0±0.17	2.56±0.3

NOTES

SAR- Simulated Acid Rain

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