JJBS

Jordan Journal of Biological Sciences

Volume 8, Number 1, March .2015
ISSN 1995-6673
Pages 51 - 54

The Effect of Simulated Acid Rain on Plant Growth Component of Cowpea (*Vigna unguiculata*) L. Walps

Bridget O. Odiyi* and Adekunle O. Eniola

Department of Biology, School of Science,Federal University of Technology, PMB 704, Akure, Ondo state, Nigeria.

Received: October 25, 2014 Revised: December 18, 2014 Accepted: December 27, 2014

Abstract

The rain forest belt in Southern Nigeria is potentially susceptible to problems related to acid rain because of the increase in the consumption of petroleum oil products, such as diesel, gasoline and coal, used to produce energy for different sectors of the economy. The effect of simulated acid rain on the plant growth component of cowpea, *Vigna unguiculata* was studied. A greenhouse experiment was conducted at the Federal University of Technology, Akure, Ondo state, Nigeria. Simulated acid rain was prepared with a mixed concentrated sulphuric acid (H_2SO_4) and concentrated nitric acid (HNO_3) in a ratio 2:1 to get the desired pH using a Deluxe pH meter. The plant was exposed to simulated acid rain of pH 2.0, 3.0, 4.0, 5.0, 6.0 and 7.0 which was the control. Simulated acid rain induced morphological changes including chlorosis, early leaf senescence, necrosis, leaf abscission, leaf folding and death. Plant height, leaf area, fresh weight, relative growth rate, chlorophyll content of the leaf and harvest index were highest at 7.0 (control) but significantly ($p<0.05$) decreased with increasing the acidity levels. *V. unguiculata* was seriously affected by the simulated acid rain.

Keywords: Harvest index, Plant growth, Relative growth rate, Simulated acid rain, *V. unguiculata*

1. Introduction

Acid rain is a wet deposition that has been acidified when pollutants, such as oxides of sulphur and nitrogen, contained in power plant emission, factory smoke and car exhaust, react with the moisture in the atmosphere (Kita *et al*., 2004). In natural conditions atmospheric precipitation is slightly acidic due to the dissolution of atmospheric carbon dioxide (Nduka *et al*., 2008). Rain that presents a concentration of H^+ ions greater than 2.5μeq^{-1} and pH values lower than 5.6 is considered acid. Acid deposition may cause a decline in the health and growth of plants (Wyrwicha and Sklodowska, 2006; Liu *et al*., 2010). Several experiments have been carried out in the field and in greenhouses to investigate the effect of acid rain on plants. Acid rain exposure of plants result in a characteristic of foliar injury symptoms, modified leaf anatomy (Stoynora and Velikova, 1998; Park and Yanai, 2009), structural changes in the photosynthetic pigment apparatus and a decrease in the chlorophyll concentrations (Sant' Anna-Santos *et al*., 2006; Wang, 2010). Also, a reduction in plant growth and a yield of field corn (Banwart, *et al*.,1988), green pepper (Shripal *et al*., 2000), tomato (Dursun *et al*., 2002) were reported.

Cowpea, *Vigna unguiculata* (L.)Walp, is a leguminous plant belonging to the family Fabaceae. It is of immense benefit to mankind because it is useful as a rotational cover crop. It is also used to control erosion and to improve soil properties. Cowpea also suppresses weeds and can encourage populations of beneficial insects to defend cash crops from insect pests. It can also be used for the production of high quality hay or silage, when mixed with crops, such as corn or sorghum. Cowpea, being a stable food in Nigeria for millions of Nigerians, can potentially be affected by acid rain, and so, there is a need to examine the potential effects of acidic precipitation on cowpea. Acid rain is an issue in the study site and this experiment has been done on test species. With the increasing rate of population in Nigeria which is leading to the high demand for automobiles, it is evident that acidic rain is a reality in Nigeria. In view of the importance of this plant in human diet and the adverse effect of acid rain, the present study is carried out to assess the effect of simulated acid rain on this plant.

2. Materials and Methods

2.1. Planting Procedure

A greenhouse experiment was conducted in the Department of Biology of the Federal University of Technology, Akure, Ondo State, Nigeria. Disease free seeds were collected from the International Institute of Tropical Agriculture (IITA) Ibadan in Oyo State of Nigeria. The plants were not provided with nutrient solution.

The seeds were tested for viability before planting. Six seeds were planted in each experimental plastic pots. Each pH treatment had four replicates and was arranged in a

* Corresponding author. e-mail: bnyamali@yahoo.com.

Completely Randomized Design (CRD). The plants were watered every other day and grown for a week before the application of the simulated acid rain treatment. The plants were sprayed with 10ml of acidic solution according to their pH values of 2.0, 3.0, 4.0, 5.0, 6.0 and the control (7.0). The solutions were applied every three days using a medium size pressurized sprayer on the plants. The plants were grown for fifteen weeks before the termination of the experiment.

2.2. Preparation of Simulated Acid Rain

The simulated acid rain was formed from a mixture of concentrated sulphuric acid (H_2SO_4) and concentrated nitric acid (HNO_3) in a ratio 2:1. The acidic solution was then calibrated using distilled water with a Deluxe pH meter to get the desired pH (2.0, 3.0. 4.0, 5.0 and 6.0) and cross checked with pH pen. The control pH (7.0) had distilled water.

Several parameters were used in assessing the growth of the plant. The height of the shoots was measured using a tape rule in (cm) from the soil level to the terminal bud. The measurements were taken in an interval of 2 weeks from the day the acid rain treatment commenced to the day of harvest. Leaf area was determined by the proportional method of weighing a cut-out of traced area of the leaves on graph paper with standard paper of known weight to area ratio. The fresh weights and the Relative Growth Rate (RGR) were calculated following the methods of Hunt (1990) and the fresh weight of the whole plant was used to determine the relative growth rate:

$$RGR = (\log W_2 - \log W_1)/(T_2 - T_1)$$

Where: W_2 = final weight

W_1 =initial weight

T_2 =final time

T_1 = initial time.

The chlorophyll content of the leaves was determined by the method of Arnon (1949) and it is expressed on the fresh weight:

Chlorophyll a =$12.7_{D663}-2.69_{D645}$ X Vmg/1000W (mgg^{-1}).

Chlorophyll b=$22.9_{D645}-4.68_{D663}$ X Vmg/1000W (mgg^{-1}).

Total chlorophyll (chlorophyll a & b) = $20.2_{D645}-8.02_{D663}$ X V/1000W.

Where D_x = absorbance of the extract at the wavelength X nm.

V = total volume of the chlorophyll solution (ml).

W = weight of the tissue extracted (g).

The harvest index was determined by the method of Ekanayake (1994).

2.3. Statistical Analysis

Data obtained were subjected to analysis using the Statistical Package for Social Sciences, Version 15.0 (SPSS, 2003). Treatment means were separated using the Duncan Multiple Range Test (Zar, 1984).

3. Results and Discussion

Morphological changes were observed through the 15 weeks after simulated acid rain treatment. The effects of simulated acid rain on the morphology shows that the leaves turned brownish, with red with 70% leaf abscission. The dropping and eventual collapse of leaves stretched over a period of 12-15 weeks at pH 4.0 to pH 2.0. Leaf abscission started with the leaves at the base of the shoot droppings with long petiole at pH 4.0 treatment. Leaves were chlorotic and necrotic. At 2.0 pH treatment, plants died from the base of the shoot. Leaves had 60% leaf abscission.

Table 1. Effect of simulated acid rain (SAR) on the plant height (cm), leaf area (cm), fresh weight (g) of *V. unguiculata*

pH of SAR	Plant height (cm)	Leaf area (cm)	Fresh weight (g)
7.0 (Control)	54.74 ± 2.34 a	38.14 ± 2.27 a	192.11 ± 0.56 a
6.0	46.12 ± 2.06 b	26.08 ± 1.17 b	130.34 ± 0.44 b
5.0	40.10 ± 1.60 b	20.54 ± 1.08 b	122.12 ± 0.20 c
4.0	30.33 ± 1.48 b	15.20 ± 0.20b	94.21 ± 0.12 d
3.0	22.34 ± 0.31 c	11.26 ± 0.18c	64 .10 ± 0.07e
2.0	10.01 ± 0.10 d	06.32 ± 0.04c	42.23 ± 0.04 f

Each value is a mean of ± standard error of four replicates. Means within the same column followed by the same letter are not significantly different at (p>0.05) from each other using New Duncan Multiple Range Test.

The plant had the highest plant height, leaf area and fresh weight significantly higher (p< 0.05) at the control (pH 7.0) compared to the other acidity treatments.

Table 2. Effect of simulated acid rain (SAR) on the relative growth rate ($gg^{-1}d^{-1}$), chlorophyll content (mg/g) and harvest index of *V. unguiculata*

pH of SAR	Relative growth rate	Chlorophyll content	Harvest index
7.0 (Control)	2.6 ± 0.22 a	3.8 ± 0.20 a	0.5
6.0	2.0 ± 0.16 a	3.0 ± 0.16 a	0.4
5.0	1.6 ± 0.12 b	2.2 ± 0.12 b	0.3
4.0	1.2 ± 0.08 b	1.7 ± 0.10 b	0.2
3.0	0.8 ± 0.05 b	0.6 ± 0.05 c	0.1
2.0	0.4 ± 0.02 c	0.2 ± 0.01 c	0.1

Each value is a mean of ± standard error of four replicates. Means within the same column followed by the same letter are not significantly different at (p>0.05) from each other using New Duncan Multiple Range Test.

The effect of simulated acid rain on the Relative Growth Rate (RGR), the chlorophyll content and the harvest index of *V. unguiculata* are presented in Table 2. The plant had a relative growth rate, a chlorophyll content and a harvest index significantly higher (p< 0.05) at pH 7.0 compared to other acidity treatments. There was a significant reduction in relative growth rate, chlorophyll content and harvest index with decreasing pH level.

Symptoms of plants polluted with simulated acid rain include chlorosis, necrosis, stunted growth, lesion, suppression of leaf production, leaf curling, withering of leaves, leaf abscission and even death of plants. Silva *et al*. (2006) and Wang (2010) found that plants exposed to low pH rain (pH 3.0) are generally retarded with leaf chlorosis, necrotic spot coupled with dehydration of the plants. Simulated acid rain exposure caused chlorosis, necrotic lesions and leaf tip injuries at the different pH levels of *V. unguiculata*. Marked chlorotic and marginal necrotic symptoms were observed at pH 4.0 and 5.0. However, this was less pronounced in comparison to pH 3.0 and pH 2.0. Similar symptoms were also observed by Johnston and Shriner (1985) on wheat at pH 4.3 and 2.3.

All the plant growth parameters that studied the plant height, leaf area and fresh weight of *V. unguiculata* were decreased significantly at all acidity levels with respect to the control set; the highest reductions were observed at pH 2.0 level (Table 1). The adverse effects of simulated acid rain on plant growth parameters on several crops were also observed by Evans *et al*. (1997), Banwart *et al.* (1990), Chevone *et al*. (1984) and Liu *et al*. (2010).

Photosynthetic pigments were also inhibited with respect to acidity levels. Chlorophyll content was significantly reduced by simulated acid rain treatment compared to the control at pH 2.0 and pH 3.0 (Table 2). The reduction was due to the removal of Mg^+ from the tetrapyrol ring of the chlorophyll molecules by H^+ (Foster, 1990) or due to the increase of transpiration by acid rain (Evans *et al*., 1997). Recently similar results have also been observed on many crops like mustard, radish, potato (Agrawal *et al*., 2005; Kausar *et al*., 2005; Khan and Devpura 2005; Varshney *et al*., 2005; Park and Yanai , 2009).

Relative growth rate and harvest index were lowest at pH 2.0 and pH 3.0 of *V. unguiculata* compared to the control plants (Table 2), and this have been reported by Seinfield *et al*. (1998). According to Iglesias *et al*. (1994), harvest index of 0.5- 0.6 is the optimum level for crops because at higher values of harvest index, root production decreases due to a reduced leaf area, light interception and photosynthesis. The present paper shows that all the levels of simulated acid rain have a negative effect on the growth and the yield component of cowpea *V. unguiculata* due to the reduction of photosynthesis as a result of chlorosis, necrosis and leaf abscission.

References

AgrawalS, Raghav D and Khan A A. 2005. An evaluation of the impact of simulated acid rain on the growth of mustard in pots. *Test of Agrochem Cultivars*, (Supplement), **21**: 25- 26.

Arnon DI. 1949. Copper enzymes in isolated chloroplast and polyperoxidase in *Phaseolus vulgaris. Plant Physiol.*, 24: 1-15.

Banwart WL, Porter PM, Ziegler EL and Hassett JJ. 1988. Growth parameters and yield component response of field corn to simulated acid rain. *Environ Experimental Bot.*, **28**: 43-51.

Banwart W L, Finke R L, Porter P M and Hassett J J. 1990. Sensitivity of twenty soybean cultivars to simulated acid rain. *J Environ Quality*,**19**:339-346.

Chevone BI, Herzfeld K and Chappelka AH.1984. Effects of acid aerosol and ozone on yellowpoplar seedlings. *Can J Res.,* **14**: 820-891.

Dursun A,Kumlay AM,Yilderin E and Guvenc I. 2002. Effects of simulated acid rain on plant growth and yield of tomato. *Acta Hort*., **579**: 245-248.

Ekanayake I J. 1994.Terminology for growth analysis of cassava.*Tropical Root and Tuber Crops Bulletin,* **8(1):**2.

Evans LS, Gmor NF and Dacosta F 1997. Leaf surface and histological perturbations of leaves of *Phaseolus vulgaris* and *Helianthus annuus* after exposure to simulated acid rain. *Am J Bot*., **4**: 304-313.

Foster JR. 1990. Influence of pH and plant nutrients status onion fluxes between tomato plants and simulated acid mists. *New Phytologist.*,**116** : 475-485.

Hunt R. 1990. **Basic Growth Analysis: Plant Growth Analysis for Beginners**. AcademicPress. London, UK. 41p.

Iglesias C A, Calle F, Hershey C, Jaramillo G and Mesa E. 1994. Sensitivity of cassava (*Manihot esculetna Crantz)* clones to environmental changes. *Field Crops Res.,* **36**: 213-220.

Johnston J W Jr. and Shriner D S.1985. Response of three wheat cultivars to simulated acid rain. *Environ Experimental Bot.,* **25**: 349-353.

Kausar S, Khan A A and Rahman F. 2005. Impact of simulated acid rain on potato.In :**Herbal Drug and Environmental Pollution**. A satellite Session of Third International Conference on Plants and Environmental Pollution, Jamia Hamdard, New Delhi, pp. 56.

Kita I, Sato T, Kase Y and Mitropoulos P. 2004. Neutral rains at Athens, Greece: a natural safeguard against acidification of rains. *Sci Total Environ.,* **327**:285-294.

Khan T I and Deopura S. 2005. Physiological and biochemical effects of simulated acid rain on *Phaseolus vulgaris* Var HUR 15. *The Environ.*, **24**:224-226.

Liu K H, Mansell R S and Rhue R D. 2010. Cation removal during application of acid solution into air dry soil columns. *Soil Sci Society J.*, **4**:1747 – 1753.

Nduka JKC, Oriskwe OE, Ezenweke LO, Ezenwa TE, Chendo MN and Ezeabasili NG.2008. Acid rain phenomenon in Niger Delta region of Nigeria: economic, biodiversity and public health concern. *The SciWorld J* **8**: 811-818.

Park B B and Yanai R D. 2009. Nutrient concentrations in roots, leaves and wood of seedling and mature sugar maple and American beech at two contrasting sites. *Forest Ecol Management,* **258:**1153 – 1160.

Sant' Anna-Santos B F, Silva LC, Azevedo A and Deque-Brasil R. 2006. Effects of simulated acid rain on photosynthesis, chlorophyll fluorescence and antioxidative enzymes in *Cucumis sativus* L. *Photosynthetica,* **40**: 331-335.

Shripal N, Pal KS and Kumar N. 2000. Effects of simulated acid rain on yield and carbohydrate contents of green pepper. *Advanced Plant Sci*., **13**: 85-88.

SPSS .2003. **Statistical Package for Social Sciences** (SPSS 15.0 Inc., Chicago, U.S.A).

Silva L C, Oliva M A, Azevedo A A and Araujo J M. 2006. Responses of resting plant species to pollution from an iron pelletization factory.*Water Air and Soil Pollution,* **175** (1-4): 241-256.

Seinfield J H, Pandis S N and Spyries N. 1998. **Atmospheric Chemistry and Physics from Air Pollution to Climate Damage.** AVI publishing Co, West Print Connecticut, pp 224.

Stoynora D and Velilova V. 1998. Effects of simulated acid rain on chloroplast ultrastructure of primary levels of *Phaseolus vulgaris. Biol Plant*, **40**:589-598.

Varshney P , Kausar S and Khan AA. 2005. An evaluation of the susceptibility of radish inpots to simulated acid rain. *Test of Agrochem Cultivar*, **27**: 29-30.

Wang CQ. 2010. Exogenous calcium alters activities of antioxidant enzymes in *Trifolium repens* L. leaves under peg-induced water deficit. *J Plant Nutr.,* **33**: 1874-1885.

Wyrwicha A and Sklodowska M. 2006. Influence of repeated acid rain treatment on antiocidative enzyme activities and on lipid peroxidation in cucumber leaves. *Environ Experimental Bot.,* **56**: 198-204.

Zar J H. 1984. **Biostatatical Analysis**, 2nd edition, Prentice- Hall International, Englewood Cliffs, N.J.