

EFFECTS OF ACID RAIN ON EPIPHYTIC ORCHID GROWTH

Sr. John Karen Frei, Chris Orenic, Natalie Smith,
Heather Jeffer
Division of Biological & Biomedical Sciences
Barry University
Miami, Florida 33161, U.S.A.

ABSTRACT

The Big Cypress Swamp in South Florida is known for its subtropical flora and fauna. Among the species of subtropical vegetation occurring in this area are many epiphytic plants which are directly vulnerable to atmospheric inputs.

Some preliminary evidence indicates that this general area may be experiencing elevated concentrations of gaseous and particulate air pollutants, but very little is known about their potential impact on biological resources.

The purpose of this study was to examine the effects of simulated acid rain at several pH levels in the growth and development of the epiphytic orchid plant Encyclia tampensis.

The sixty plants used in this experiment were collected from the Fakhahatchee Strand located within the Big Cypress Swamp. Plants used were mature plants, intermediate sized plants and young seedlings, which were exposed to the following pH ranges of simulated acid rain (pH 2.5, 4.0, 5.6) for a period of six months.

Observations were made concerning: number and conditions of flower spikes and flower buds; number and condition of new and old leaves; the general condition of the plant.

The results indicated that there is a negative effect on the growth rate of these plants exposed to varying levels of simulated acid rain mistings.

1. INTRODUCTION AND RATIONALE

Comprehensive knowledge of epiphytic ecology requires knowledge not only of the interaction between substrate and epiphytic plants, but also an understanding of the effect of air and water borne pollutants present in the habitat of these plants. The objective of this study was to investigate the growth and development of the epiphytic orchid Encyclia tampensis collected from the Fakhahatchee Strand in the Big Cypress Swamp, Florida, and grown under simulated acid rain conditions in a controlled

environmental setting. This orchid was chosen for study because of the reported decrease in its population numbers from within the Strand. Due to the plant's physical location, the decrease was thought not to be completely attributed to collection pressures. It was thought that this geographic region could be experiencing air and water borne pollutants.

Air quality is continuing to improve in the United States, although serious problems exist in many areas. Changes by the federal government lessening the requirements of anti-pollution devices in automobiles could have an adverse effect on air quality. Ozone and carbon monoxide continue to be the pollutants most often in excess. Florida, east and south of Lake Okeechobee is experiencing ozone pollution (6). In a study of air quality in Florida, it was shown that in addition to ozone, NO₂ and carbon monoxide are important pollutants in many areas of Florida, and that total suspended particle (TSP) levels are generally high throughout the state. Also, the fact that Florida is a southern state with strong photo-chemical processes, adds to its air pollution problems (12). Growth and development of plants are directly related to air quality. A number of pollutants not only suppress growth, but they also cause disease and death in plants (17, 19).

In his second environmental message to Congress in 1979, President Carter identified acid rain as one of the two most serious environmental problems associated with the continued use of fossil fuels (22). It is a growing problem in the northeast and is believed to be created when sulfur oxide and nitrogen oxide combine with water in the atmosphere (5). Acid precipitation may fall to the earth many miles from where the pollutants were emitted. Scientists estimate that 10-30 percent of the acid problem may result from dry deposition. This is significant because the farther the polluted air travels, the more sulfur dioxide is transformed into sulfuric acid which then coats other particles. There are documented cases of sulfate particles formed from emissions in the midwest and northeast traveling as far as the Gulf of Mexico (24). The Hubbard Brook watershed, located in northern New Hampshire, containing a much damaged ecosystem, receives about one-third of its sulfur in dry form (16). Acid rain, in its liquid form, has been shown to adversely affect plant growth and cause foliage damage (3, 8, 10, 13, 14, 18). It also may exert subtle effects on the reproductive potential of plants. Evans and Buzzone (7) have shown that reproductive development in ferns is very sensitive to the effects of acid rain in that sperm motility is decreased at pH levels below 5.8. Fertilization, which in all lower plants, i.e. bryophytes, involves movement of the sperm through the liquid environmental medium to the egg, would be subject to the effects of acid rain. It has been suggested that fertilization in the fern Pteridium may be used as a bioindicator of contaminants in rain water (11). Acid rain has been shown to affect the germination rate and seedling growth of many forest woody species. Acidic precipitation has been shown to adversely affect the microbiotic components of the environment as well (1).

The acidity of rain in northern and central Florida has increased markedly in the past twenty-five years with the average sulfate and nitrate concentrations increasing by factors of 1.6 and 4.5 since the early 1950's (20). Since Florida's human population is still expanding rapidly, the demands for electric power are increasing more rapidly in Florida than in most other states. Florida is shifting to coal as the energy source for increasing electric power-generating capacity. In southern Florida, there are six coal-fired plants operating now with fifteen additional plants scheduled to be operating by 1987 (2). Many experts believe that fossil fuel burning power plants are responsible for much of the SO₂ problem (21).

Because of the described factors including the known detrimental effect of pollutants emitted into the atmosphere from fossil fuel burning, it is imperative to monitor the fragile epiphytic ecosystem for the effect of pollution while the ecosystem is still present in south Florida.

Published reports regarding the effect of air pollutants on orchids are mainly concerned with the adult plant, i.e. how buds and flowers are affected by ethylene gas (23). Adult plants are reported to be relatively resistant to the air pollutants sulfur dioxide and ozone. Using untested pesticide materials and formulations has caused serious injury to orchid plants. Pesticide injury varies from stunting growth to leaf-scorch, necrotic spotting, white banding on newly developing leaves and abnormal pigmentation in the blades. Necrosis may cause collapsed blade margins (4). There are no published studies concerning the effect of air or water borne pollutants on the germination of orchid seed or on early protocorm development.

2. MATERIALS AND METHODS

The purpose of this study was to examine the effects of simulated acid rain/mist (hereafter referred to as acid rain) at several pH levels on the growth and development of Encyclia tampensis plants. On May 16, 1983, sixty E. tampensis plants were collected in the Fakahatchee Strand. Some of the orchids when collected were physically removed from their bark substrates, leaving them with bare roots, while others were removed intact on their substrate bark using a machete to shave off the bark from the tree with its attached orchid plant. The plants were divided into three age groups (young seedlings, intermediate sized plants, and large mature plants) based on their stage of development. Of the total number of plants collected, seven were designated as young seedlings, two of which were collected attached to the bark substrate; 45 young plants were designated as intermediate sized plants, 15 of which were collected attached to the bark substrate; and 8 were designated as large mature plants of which one collected was attached to the bark substrate.

The average size of the young seedlings was as follows: the number of leaves was 2.9, the leaf length was 6.3 cm and the number of pseudo-bulbs/plant was 7.6.

The intermediate sized young orchid plants average size was as follows: the number of leaves was 6.3, the leaf length was 13.0 cm and the number of pseudo bulbs/plant was 14.4.

The mature plants average size was as follows: the number of leaves was 19.5, the leaf length was 15.3 cm and the number of pseudo-bulbs/plant was 45.3.

Designation into the three age groups was also based on the number and size of pseudo bulbs as well as the number and size of mature leaves. It was observed that young seedlings had pseudo-bulbs about 0.5 to 1.0 cm in diameter, and their leaves were short and slender. The mature plants had a large number of pseudo-bulbs some of which were large and thick while the intermediate sized plants had pseudo-bulbs of a size in between the other two. In nature, as the older pseudo-bulbs die, they decompose in place and new pseudo-bulb growth occurs on the outside of the cluster of the decayed pseudo-bulbs. Often the center of the pseudo-bulb cluster is totally decomposed.

Among the sixty plants collected, eighteen plants (5 mature and 13 intermediate) were in spike and some of the spikes were in flower at the time of the collection, while others were still in bud. There was a total of twenty-eight spikes among the eighteen plants in spike.

The average spike length was 27.5 cm. Five of the mature plants were in spike accounting for ten of the eighteen spikes. One of the intermediate sized orchid plants had the most spikes - five, which had a total of twenty-eight developing flower buds.

Those orchid plants which had been physically removed from their bark substrates were attached to pieces of tree fern - one plant per piece - with plastic coated wire. With time as the plants' roots grew into the tree fern securing the plant to the tree fern, the wires were removed. Five young seedlings, thirty intermediate sized plants and seven mature plants were secured in this manner. Four days after they were secured, 23 of the plants were placed into a growth chamber and allowed to acclimatize to the chamber for a period of fourteen weeks. The number of plants which could be placed in the chamber was limited by the physical size of the chamber as well as by the physical area misted by the water spray system. The misting during this fourteen week period was non-acidic. The remainder of the plants were placed in a saran greenhouse which had a sprinkling system on a time clock. The pH of the water in the greenhouse was 7.3

The growth chamber used was a Percival Model PT-80. During the experimental period, conditions in the chamber were programmed to simulate those found in the Fakhahatchee Strand and adjusted periodically to reflect seasonal changes. Light conditions were established using eight F 72 T12/cw/1500 fluorescent lamps and ten 60 watt incandescent lamps; these were under the control of two separate and independent 24 hour timers. The incandescent timer was set to turn the incandescent lights on one hour before and one hour after the fluorescent lighting timer turned its light source on to simulate sunrise and sunset. The light range was from 12 hours (10 fluorescent 12 incandescent) to 14 hours

(12 fluorescent 14 incandescent). The light entered the environmental chamber through translucent thermal barriers. Temperature controls were set to values approximating the Fakahatchee Strand temperature as recorded during field trips to the Strand. The temperature range was 24° - 30° C daytime and 18 - 24° C nighttime. The relative humidity settings were kept near 60% and allowed to fluctuate \pm 10%.

The watering system components were constructed of the following materials: spray nozzles - #316 stainless steel, white nylon tubing, #304 stainless steel water holding tank, brass solenoid valve stainless steel strainer. The nozzles had 0.010 inch orifices and were rated at one gallon/hour at forty PSIG. The chamber had six nozzles located at the top of the chamber and spaced around the chamber. The watering system was set at 40 PSIG. The sprinkling system had been specially designed for this experiment to insure that the area below the nozzles was well watered. The plants were watered five times daily with three-minute sprayings per time interval. Spray times were 9:00 a.m., 12:00 noon, 2:30 p.m., 5:30 p.m. and 9:00 p.m. The pH of the spray was non-acidic.

Weekly observations were made on the sixty plants (individually numbered, and tagged for identification purposes), during the fourteen week acclimatization period as well as throughout the experimental period. The following measurements were taken during the acclimatization and experimental periods.

1. Number of live flower spikes.
2. Length of live flower spikes recorded in centimeters.
3. Number of flower buds, flowers and/or seed pods/spikes. Spikes which died were also recorded.
4. Total number of leaves - all leaves included - new and mature. Dead leaves excluded.
5. Average leaf length. Measurements were made from the base of the leaf considered as the leaf-pseudo-bulb junction to the tip of the leaf. At the leaf tip, measurement was made only of the live portion of the leaf. All leaves included - new and mature. Dead leaves excluded.
6. Number of new leaves - immature leaves.
7. Length of new leaves - each new leaf was measured separately.
8. Range of mature leaf lengths.
9. Number of live pseudo-bulbs.
10. Number of new pseudo-bulbs. Those considered new were those formed at the base of an immature leaf. General comments concerning the overall condition of the plants were also recorded on a weekly basis.

On August 21, 1983, after the acclimatization period was concluded, the 60 plants were divided into six groups of ten plants each, with an attempt made to include the three age distributions of plants previously described into each group. Three of the groups were arbitrarily chosen as experimental groups while the remaining three served as the control group.

The experimental groups and the acid rain pH ranges to which they were subjected were as follows:

Plant Group 1A - pH 4.0	Plant Group 1B - control group
Plant Group 2A - pH 5.6	Plant Group 2B - control group
Plant Group 3A - pH 2.5	Plant Group 3B - control group

The first experimental group (1A) was placed into the growth chamber under the same environmental conditions as the plants had previously been subjected to; however, the spray which they received was acid rain.

A plant was exposed to only one of the three pH levels throughout the experiment.

The artificial acid rain formula used was based on that provided in February, 1983, by Virginia Polytechnic Institute.

The following chemical amounts were mixed in deionized, distilled water (maximum conductivity, 2.0 umhos) and diluted to 1 liter.

Na ₂ SO ₄	2.1621g
CaSO ₄	1.2208g
MgSO ₄	0.3465g
K ₂ SO ₄	0.0555g
KCl	1.2478g
(NH ₄) ₂ HPO ₄	0.0124g
NH ₄ NO ₃	0.7440g
HNO ₃ , 1.0N	10.54ml
HCl, 1.0N	4.84ml

The stock solution was diluted 1 to 1000 after being adjusted to the desired pH with NaOH or H₂SO₄. Ionic concentration in the simulated rain solution should have been:

<u>Ion</u>	<u>Mg/l</u>
Na ⁺	0.70
Ca ⁺²	0.38
NH ₄ ⁺	0.70
Mg ⁺²	0.07
K ⁺	0.05
SO ₄ ⁻²	2.68
NO ₃	1.23
Cl ⁻	0.29
PO ₄ ⁻³	0.01

Each batch (10 liters) made was adjusted to the desired pH using NaOH and H_2SO_4 . A corning model 610A expand portable pH meter was used with a set of Coleman Tri-Purpose shielded glass electrodes to determine pH. At the end of each acid rain treatment, the system was thoroughly rinsed with deionized water before the next acid rain treatment began.

The control groups were maintained in the greenhouse throughout the course of the experiment. Each experimental plant group was placed for one week into the growth chamber undergoing the acid rain treatment, followed by a two week period in the greenhouse, and then placed back into the growth chamber for another week of acid rain treatment. This cycle - one week in - two weeks out - one week in - was repeated for twenty weeks so that each experimental group of plants received an acid misting seven days out of every twenty-one days for thirty weeks.

3. RESULTS

3.1. Flower Spike Development

During the experimental period, the plants which had been in flower and spike development when collected underwent normal flower and seed pod development. After the plants were established, no new spikes developed until the first week of October. On the original spikes, all the seed pods which had developed had done so before any of the plants were subjected to the acid rain treatments.

During the first two weeks of October, a new flower spike developed on plant #13 of group 1A (pH 4.0). This spike was initiated after 21 days of exposure to the simulated acid rain (pH 4.0). This plant is an intermediate sized plant mounted on tree fern bark.

By the fourth week of October, this plant had reached its maximum length of 5.7 cm and had produced a single flower. The length of this flower spike was considerably shorter than the 27.5 cm average length of the eighteen original plants collected in spike. It should be noted that it was also shorter than the smallest spike recorded after the initial orchid collection on May 16. The flower produced was of normal size but had brown spot damage along the edge of two of its sepals. This damaged area was a small indentation of tissue which had turned brownish in color. This flower was hand pollinated in late October, but the flower fell off the plant after showing no signs of seed pod development. No more flower buds developed and the spike died back after an additional 21 days of exposure to the acid rain.

As can be seen in Table 1, four flower spikes developed during the acid rain treatment. They were initiated (all within 12 days of one another) after a period of 42 days of acid rain treatments. After an additional period of 21 days of acid rain treatment (for a total period of 63 acid rain treatment days) they reached the recorded spike size as indicated in Table 1. This represents a total of 190 days of growth including a recovery period of 137 days in the greenhouse.

Table 1. Comparison of Flower Spike Development after 63 Days of Acid Rain Treatment

PLANT #	pH	SIZE	LENGTH (cm)	NUMBER OF BUDS
51	Control	Mature	38.2	6
24	5.6	Medium	11.9	0
55	4.0	Mature	4.3	0
17	2.5	Medium	2.9	0

The longest spike development (38.2 cm) was in a mature plant #51 in the control group. The average diameter of its five flower buds was 0.5 cm. The longest spike from a plant receiving the acid rain treatments (pH 5.6) was from plant #24. This is a medium sized plant whose flower spike was 11.9 cm long. No buds have been initiated on this plant. The second longest spike on a plant receiving the acid rain treatment (pH 4.0) was from plant #55. This is a mature sized plant whose flower spike was 4.3 cm long. No buds have been initiated on this plant. The last plant with flower spike development on a plant receiving the acid rain treatment (pH 2.5) was from plant #17. This is a medium sized plant whose flower spike was 2.9 cm long.

3.2. Development of New Leaves

One new leaf was initiated on plant #25, a young seedling plant. This new leaf was initiated during the acclimatization period in the growth chamber and just prior to the acid rain treatment. This plant was in group #3A which received acid rain treatment (pH 2.5). Immediately after receiving one week's exposure to the acid rain, the new leaf died.

A total of ten new leaves were initiated on plants #21, #27 and #43 - medium sized plants - as well on plant #30, a young seedling plant. These new leaves were initiated during the acclimatization period in the growth chamber and just prior to the acid rain treatments. These four plants were in group #1A which received acid rain treatment (pH 4.0). Within three weeks after undergoing a one week treatment of acid rain, seven of the ten leaves died.

Fifteen new leaves were initiated on plants #6, #14, #37 and #60 - all young seedling plants - and on plants #31 and #41 which were medium sized plants. These new leaves were initiated during the acclimatization period in the growth chamber and just prior to the acid rain treatments. These plants were in group #2A which received acid rain treatment (pH 5.6). Immediately after

receiving one week's exposure to the acid rain, three of the new leaves on plant #60 died.

Nine new leaves were initiated on plants #3, #5, and #7 - all young seedling plants - and on plant #51 which was a mature plant. These new leaves were initiated during the acclimatization period in the greenhouse on these control plants, and just prior to the beginning of the acid rain treatments on Group #3A, #1A, and #2A. Control plant #7 lost two leaves six weeks after the end of the acclimatization period.

3.3. Leaf Initiation During Acid Rain Treatment

Table 2 records leaves of plants still considered to be new - 160 days (6 months) into the acid rain experimental period. A new leaf is defined as one which has a pseudo-bulb under development but not yet visible. During the experimental treatment with acid rain, additional new leaves were initiated. Once pseudo-bulbs developed at their bases, they were considered as mature leaves. For that reason they are not recorded in this table.

As can be seen from the table, the rate of growth of leaves, average growth of leaves, and average leaf length is greatest in plants exposed to acid rain of pH 5.6 and slowest in plants which were exposed to acid rain pH 4.0.

3.4. Leaf Tissue Damage

Leaf tissue damage was studied in mature and intermediate sized plants. Those plants reported in Table 3 were selected based on similarity in size among the mature and intermediate sized plants.

An examination of group 3A plants (pH 2.5) on the 180th day after the acid rain treatment began, revealed a leaf tissue damage which was rarely present on leaves of the control plants. This tissue damage (tissue necrosis) first noticed on February 17, 1984, was characterized by a blackening of the tissue and additionally in some cases a slight loss of tissue turgidity. The blackening of the tissue was found mainly along the lateral edges of the leaf, upper leaf epidermis and at leaf tips, but also all along the leaf including the upper and lower epidermis.

An examination of three of the mature leaves on plant #17 (in group 3A), a mature plant, indicated that on the longest leaf there was the most tissue damage. This damage on the upper side of the leaf was localized along the lateral edges of the leaf which in several areas extended several millimeters in from the edge of the leaf. On the under surface of the leaf, the damage was present in a more "spotted" random manner, although there was lateral edge damage corresponding to that present on the leaf's upper surface. There appeared to be damage on older leaves, possibly of a mechanical nature, or from insects, around which the blackening of the tissue also developed. Ninety-three percent of the leaves on plant #17 were damaged.

Table 2. Growth Rate of New Leaves After 180 Days of Acid Rain Treatment

pH	PLANT NO.	PLANT SIZE	AGE OF LEAF	FINAL LENGTH OF LEAF CM	RECORDED GROWTH CM	GROWTH RATE CM
Control	5	Young	5	0.3	0.1	0.0200
	5	Seedling	5	0.3	0.1	0.0200
	7	Young	42	2.3	1.9	0.0452
	7	Seedling	42	2.5	2.1	0.0500
			days		new leaves	
				$\bar{x} = 1.35$	$\bar{x} = 1.05$	$\bar{x} = 0.033$
5.6	41	Medium	40	3.2	2.9	0.0725
	41	Medium	40	3.3	2.8	0.0700
	41	Medium	26	3.1	2.2	0.0846
	41	Medium	12	1.5	0.8	0.0667
	53	Mature	26	3.4	1.9	0.0731
				$\bar{x} = 2.90$	$\bar{x} = 2.12$	$\bar{x} = 0.077$
4.0	21	Medium	20	1.0	0.5	0.0250
	27	Medium	167	2.3	2.0	0.0120
	30	Young	62	1.4	1.1	0.0177
	43	Medium	93	0.9	0.1	0.0011
	33	Mature	83	1.9	1.3	0.0157
				$\bar{x} = 1.50$	$\bar{x} = 1.0$	$\bar{x} = 0.014$
2.5	25	Young	27	2.8	2.0	0.0741
	35	Mature	13	1.3	0.6	0.0462
	59	Medium	27	2.4	1.3	0.0481
				$\bar{x} = 2.16$	$\bar{x} = 1.3$	$\bar{x} = 0.056$

An examination of Group 1A plants (pH 4.0) revealed less leaf damage, only 40% as compared to the 93% observed in Group 3A plants (pH 2.5). The ratio of the number of damaged leaves to healthy leaves was less in Group 1A plants as was the ratio of tissue damage to healthy leaf tissue on those leaves showing damage (see Table 3).

Table 3. Leaf Damage in Representative Plants of Each Experimental/Control Group

GROUP-(pH)	PLANT #	TOTAL # OF LEAVES	\bar{x}	# & % LEAVES DAMAGED	% LEAF TIPS DAMAGED	# LEAVES HIGHLY DAMAGED
MATURE PLANTS						
3A-(2.5)	17	30	14.7	28 - 93%	74%	6
1A-(4.0)	55	5	21.7	2 - 40%	0	0
2A-(5.6)	24	27	13.3	10 - 37%	0	0
CONTROL	51	8	14.5	2 - 25%	0	0
INTER. PLANTS						
3A-(2.5)	59	17	9.1	14 - 82%	86%	2
1A-(4.0)	29	7	17.6	3 - 43%	0	0
2A-(5.6)	41	8	9.1	1 - 12.5%	0	0
CONTROL	39	11	13.1	5 - 45%	0	0

An examination of the five leaves on plant #55, a mature plant in Group 1A indicated that the most tissue damage was located on the two oldest leaves. One of these leaves had black spotting on the upper surface both medially and laterally with the lateral damage apparent from both sides. There was blackening damage around sites of previous damage; in particular there was a hole in the center of the leaf approximately 0.3 cm diameter with tissue necrosis spreading out from the hole. There were numerous small black spots on the under surface of the leaf.

Two of the leaves of fairly recent origin showed no sign of blackening tissue damage.

An examination of group 2A plants (pH 5.6) revealed very little leaf damage. Where there was damage, it was located on the underside of the leaf. The damage was that of a tiny spotting with no blackening of leaf tissue. The degree of damage on the affected leaves was less than on group 3A leaves, but the

difference between group 1A and 2A plants was difficult to discern. There was no damage to leaf tips. An examination of the 27 leaves on plant #24 reveal slight damage to ten of the leaves. This damage was a black spotting (0.1 cm diameter) located on the undersurface of the leaves.

An examination of group 3B control plants, revealed very little damage of the type found on the plants exposed to the acid rain treatments. Of the eight leaves on plant #51, a mature plant, six leaves showed no signs of damage. Concerning the two remaining leaves one had damage which was not consistent with acid rain damage, and the other had a single 0.1 cm indentation along the edge of the leaf which appeared similar to acid rain damage. The leaf tips were whole on the six undamaged leaves, and the damage on the other two leaves appeared to be mechanical in nature. The two damaged leaves had a slight spotting on the underside of the leaf while the other six did not.

4. DISCUSSION AND CONCLUSION

This experiment was performed to determine the effect of acid rain on the growth and development of the epiphytic plant - E. tampensis.

Reduced flower spike and bud development and spike and bud initiation were observed. In the fall (October) the only spike initiated did not reach a normal size and produced only one flower which was visually damaged. When the flower was hand pollinated it aborted. By late winter (February), those plants which had received the most acid of the mistings (pH 2.5) had the shortest flower spikes initiated with no buds formed. As the mistings became less acid, the length of the spikes increased. The spikes initiated at pH 2.5 achieved only 7% growth of that grown under control conditions, those initiated at pH 4.0 achieved 11% growth of those grown under control conditions and the spikes initiated at pH 5.6 achieved 31% growth of those grown under control conditions. The only buds initiated were on the plant grown under control conditions. The lack of bud initiation on flower spikes will ultimately lead to a lessening of a plant's reproductive potential and ability to survive. The effect of the acid rain on plant reproductive capacity has been noted by Evans and Buzzone (7) who observed reduction to Pteridium aquilinum sporophyte production at pH levels of 4.2 and 3.8. Further work by Evans and Conway (11) indicated a reduction in fertilization of P. aquilinum gametophytes at pH levels of 4.5 and 3.6. In studies of bush bean exposure to simulated acid rain, Hindawi, Rea and Griffis (13) reported reduced seed and pod growth at pH 4.0 and below. In the bush bean studies of Johnston et al. (15) statistically significant reductions in growth and pod number of the bean plants occurred in response to acid rain exposures between pH 3.2 and 4.0. The current study appears to support the general plant reproductive response to acid rain already reported. Such a response would appear to indicate that a plant's reproductive potential/response to acid rain is a good measure of its use as a bioindicator to judge the potential effect of this pollutant on plant survival in the environment.

The immediate effect of acid rain mistings on new leaves initiated during the acclimatization period was that at all acid rain levels (2.5, 4.0, and 5.6) newly initiated leaves were adversely affected. Twenty percent of those new leaves treated with acid rain of pH 5.6 dropped off the plant one week after treatment, 70% of those treated with acid rain of pH 4.0 dropped off the plant within three weeks of the misting and the only new leaf exposed to acid rain of pH 2.5 dropped off within one week of the misting. In a study of the effect of simulated acid rain in Phaseolus vulgaris and Helianthus annuus by Evans, Gmur and DaCosta (8), it was found that at pH levels 2.3 and 3.1 injury to Helianthus plants was greatest at the 3-4 leaf stage than at the 5-6 and 7-8 leaf stage. No significant plant age effects to acid rain were noted in the Phaseolus plants.

The leaf initiation results on plants in which new leaves were initiated during the acid rain treatments is difficult to explain. As might be expected, the rate of growth of leaves, average growth of leaves and average leaf length was greatest in plants exposed to simulated acid rain of pH 5.6 but slowest in plants which were exposed to simulated acid rain of pH 4.0. It is not clear why leaf growth on plants which received an acid rain misting of pH 2.5 grew longer, had a larger average growth rate and grew faster than plants at pH of 4.0. The growth rate of all leaves in the acid rain treatment was much less than in the control plants.

Leaf tissue damage was similar to previously reported studies (8, 9, 10). The leaf damage observed on the control plants is most likely due to mechanical injury (effect of the wind, cold) or insect injury. As other researchers have reported, and as was found in this study, the greatest number of leaves damaged and the percent of leaf damage was found on those plants exposed to the most acid (pH 2.5) of the acid rain mistings. Only at pH 2.5 was there any damage to leaf tips.. Further study of the injury to the leaf cells themselves at a microscopic level should be made. The purpose of this study was to investigate whether acid rain had an effect on the growth and development of the epiphytic plant - E. tampensis. It has been shown that there is a negative effect on the growth rate of these plants exposed to varying levels of acid rain mistings. These results should be added to the qualitative evidence accumulating to indicate the negative effect of acid rain on plant growth.

ACKNOWLEDGEMENTS

The research in this paper was sponsored by the National Park Service.

REFERENCES

1. Babich, H., D.L. Davis and G. Stotsky. 1980. Acid precipitation causes and consequences. *Environ.* 22(4):6-13, 40-41.
2. Brezonik, P.L., E.S. Edgerton and C.D. Hendry. 1980. Acid precipitation and sulfate deposition in Florida. *Sci.* 208:1027-1029.
3. Cowling, E.B. and R.A. Linthurst. 1981. The acid precipitation phenomenon and its ecological consequences. *Bioscience.* 31(9):649-654.
4. Davidson, O.W. 1967. Orchid ailments not caused by insects or diseases-II. *Amer. Orchid Soc. Bull.* 36(7):564-574.
5. Environmental Quality. The Eleventh Annual Report of the Council on Environmental Quality. Dec. 1980. Gus Speth, Chairman.
6. EPA:Pilot National Environmental Profile: 1977. Office of the Administrator, U.S. Environmental Protection Agency, Washington, D.C. Oct. 1980.
7. Evans, L.S. and D.M. Buzzone. 1977. Effect of buffered solutions and sulfate on vegetative and sexual development in gametophytes of Pteridium aquilinum. *Amer. Jour. Bot.* 64(7):897-902.
8. Evans, L.S., N.F. Gmur and F. DaCosta. 1977. Leaf surface and histological perturbation of leaves of Phaseolus vulgaris and Helianthus annuus after exposure to simulated acid rain. *Amer. Jour. Bot.* 64(7):903-913.
9. Evans, L.S., N.F. Gmur and J.J. Kelsch. 1977. Perturbations of upper leaf surface structures by simulated acid rain. *Envir. and Exp. Bot.* 17:145-149.
10. Evans, L.S. and T.M. Curry. 1979. Differential responses of plant foliage to simulated acid rain. *Amer. Jour. Bot.* 66(8):953-962.
11. Evans, L.S. and C.A. Conway. 1980. Effects of acidic solutions on sexual reproduction of Pteridium aquilinum. *Amer. Jour. Bot.* 67(6):866-875.
12. Green, A.E.S., D.E. Rio and R.A. Hedinger. 1978. Florida's air quality, present and future. *Florida Sci.* 41(3):182-190.
13. Hindawi, I.J., J.A. Rea and W.L. Griffis. 1980. Response to bush bean exposed to acid mist. *Amer. Jour. Bot.* 67(2):168-172.
14. Jacobsen, J.S. and A.C. Hill (Eds.). 1970. Recognition of Air Pollution Injury to Vegetation: A Pictorial Atlas. Air Pollution Control Association. Pittsburgh, PA.

16. Johnston, J. William et al. 1982. Effect of rain pH on senescence, growth, and yield of bush bean. *Environ. and Exp. Bot.* 22(1):329-337.
16. Kerr, R.A. 1981. There is more to "Acid Rain" than rain. *Sci.* 211:692-693.
17. LeBlanc, F. and J. DeSloover. 1970. Relation between industrialization and the distribution and growth of epiphytic lichens and mosses in Montreal. *Can. Jour. Bot.* 48:1485-1496.
18. Likens, G.E. and F.H. Bormann. 1974. Acid rain: A serious regional environmental problem. *Sci.* 184:1176-1179.
19. Noble, R.D. and K.F. Jensen. 1980. Effects of sulfur dioxide and ozone on growth of hybrid poplar leaves. *Amer. Jour. Bot.* 67(7):1005-1009.
20. Rosencrantz, A. and G. Wetstone. 1980. Acid precipitation national and international responses. *Environ.* 22(5):6-8.
21. Walton, Susan. 1980. Coal conversion will increase acid rain damage. *Bioscience.* 30(5):293-295.
22. Wetstone, G.S. 1980. The need for a new regulatory approach. *Environ.* 22(5):9-14, 40-41.
23. Withner, C.L. 1974. (edited) *The Orchids: Scientific Studies.* John Wiley & Sons. New York, NY.
24. Wolff, G.T., N.A. Kelly and M.A. Ferman. 1981. On the sources of summertime haze in the eastern United States. *Sci.* 21:703-705.