

Effects of simulated acid rain on saplings of *Pinus massoniana* and *Cunninghamia lanceolata*

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Abstract—The effects of simulated acid rain with pH values of 6.63 (control, 4.5, 3.0, and 2.0 on saplings of *Pinus massoniana* and *Cunninghamia lanceolata* were studied. The results showed that the pH of *C. lanceolata* leaf sap and soil decreased as the acidity of rainfall increased. The acid rain with very low pH had significant effects on the photosynthetic rates per plant, but not on that of the per unit weight of dry leaves. The respiration rates of the two species were stimulated. Root and leaf biomass, but not stem biomass, were also reduced tremendously during a seven months period.

Keywords: acid rain; net photosynthetic rate; respiration rate; soil pH; biomass.

INTRODUCTION

Acid rain has been known in some areas of southern China since starting acid rain monitoring in 1979. The results (Liu, 1988) of rain water monitoring acid rain for four months (from July to Oct., 1985) indicated that the frequency of acid rain was 100% and the average pH of the rain water ranged from 4.2 to 4.4 with minimum observed values around 3.6 to 4.0. It also indicated that the average pH values of the stemflow ranged from 2.63 to 2.98 with minimum observed values around 2.34 to 2.65. It has been reported that acid rain had seriously damaged the environment (Institute of Chongqing Environmental Sciences, 1985).

There is considerable interest in effects of acid rain on forest. Feng Zongwei *et al.* (1986) reported effects of acid rain on *Pinus massoniana* forest productivity. This paper described effects of simulated acid rain on two dominant species of subtropical forest plants observed in 1986 in an effort to determine the histological, physiological, and developmental effects of acid rain on the plants.

METHODS AND MATERIALS

Materials

Year-old seedlings of *Pinus massoniana* and *Cunninghamia lanceolata* were planted in pots containing forest topsoil for 12 pots per treatment.

Methods of acid rain simulation

Simulated rain solutions with pH 6.63 (control), 4.5, 3.0 and 2.0 were prepared by diluting a mixture of reagent grade sulfuric acid and nitric acid in a 8:1 ratio (by mol concentration) with well water. A Model 51 Digital pH Meter made in Japan was used to prepare the solution with ± 0.01 pH unit of the desired values. Except on rainy days, sprinklers were used to spray simulated rain upon pots and plots with planted seedling for 5 minutes per day from May 22 to July 22, which is equal in total rainfall of 125.4 mm. Atomizers applied simulated acid rain to pots with seeds for 1 minute per day from March 15 to July 22 with a rainfall equivalent of 408.8 mm. Except rainy days, from August 8 to November 26, simulated rain was applied to all the experimental plants by spraying upward through a spraying system manufactured from acid proof material for 30 minutes per day with rainfall of 591.7 mm. Therefore, two

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year-old saplings from seeding, which received ambient rain, would receive 717.1 mm rainfall of simulated acid rain.

Photosynthetic rate determination

Photosynthetic rate was measured by QGD-07 Model Portable Infra-red CO₂ Analyser made by Beijing Analytical Instrument Manufacturer. Open air pipe system was applied. Artificial light density was 20,000 lux. Air temperature in leaf chambers was controlled by a water bath.

Respiration rate determination

Respiration rate was measured by a QGD-07 Model Portable Infra-red CO₂ Analyser. Cycle air pipe system was used. Black paper and black plastic film were used to shade light. Air temperature in leaf chambers was controlled by water bath.

Determination of pH values of leaves sap

Fresh leaf samples were collected, washed, dried with qualitative filter paper and split. 3 grams of samples were taken, ground, and mixed in 30 ml distilled water, then still for 30 minutes. A model 51 Digital pH Meter made in Japan was used to measure the pH values of leaf sap.

Determination of pH values of soil

Surface-soil samples (1-10cm depth) were collected from each pot and mixed up. The pH values of samples were measured using a 1:5 soil / 1 mol/L KCl solution mixture. A Model 51 Digital pH Meter made in Japan was used.

Determination of biomass

After plant growth stopped, biomass of controlled and treated plant was measured to determine if acid rain treatment had had any effect on plant productivity. The roots, stems and foliage of plants were collected, weighed, oven-dried at 70°C, and weighed again. The ratio of water weight for each plant part was observed, then the dry weight of plant parts could be calculated.

RESULTS

Injury symptoms

Visible injury, in the form of leaves necroses, was observed only at treatment of pH 2.0 acid rain. Necroses began at the tip on needle leaves of *Pinus massoniana* and *Cunninghamia lanceolata*. Necroses expanded with increased acid rain. There was a yellow-green band between necroses at a pH treatment of 3.0 or above.

Acidification by acid rain in leaf sap and soil

From November 6 to November 20, simulated acid rain was terminated and was exposure to ambient rain at a pH of 5.64 lasted for 5 days. The pH values of leaf sap were measured for first time on November 20, 1986 (Table 2). From November 21 to November 25, 1986, plants were exposed to simulated acid rain for 30 minutes per day. The second time measurement of leaf sap pH values were taken on November 25, 1986 (Table 1). Results showed that leaf sap was acidified by simulated acid rain. The pH values at which acid rain had caused leaf sap acidification was from 3.0 to 4.5. An acidified leaf sap could restore back to normality when simulated acid rain had terminated for two weeks.

Table 1 Effects of simulated acid rain on pH values of leaf sap

Plant species	Acid rain pH			
	6.6	4.5	3.0	2.0
Sapling of <i>Cunninghamia lanceolata</i>	5.69	5.74	5.50	5.33
Sapling of <i>Pinus massoniana</i>	3.81	3.74	3.73	3.72

Table 2 Leaf sap pH values, after termination of simulated acid rain for two weeks

Sapling species	Acid rain pH			
	6.6	4.5	3.0	2.0
<i>Cunninghamia lanceolata</i>	5.59	5.73	5.39	5.55
<i>Pinus massoniana</i>	3.73	3.75	3.74	3.74

Determination of soil pH values was taken on November 25, 1986. It showed that soil pH values reduced with reduction of acid rain pH values (Table 3), and did not restore back to normality in two weeks when simulated acid rain was terminated. Therefore, soil was more seriously acidified, and more difficult to restore than leaf sap.

Table 3 Effect of simulated acid rain on soil pH values

Soil type	Acid rain pH			
	6.6	4.5	3.0	2.0
Topsoil around roots of <i>C. lanceolata</i>	5.39	4.99	4.44	3.97
Topsoil around roots of <i>P. massoniana</i>	5.00	4.81	4.46	3.53

Effects of simulated acid rain on photosynthetic tissues

Green leaf biomass of *Pinus massoniana* and *Cunninghamia lanceolata* was reduced by 38.2% and 42.5% with acid treatment at pH 2.0 (Table 5). Analysis of variance has indicated significant effects of acid rain treatment on photosynthetic tissues. The decrease of photosynthetic tissues from acid rain should reduce photosynthetic function and therefore was probably an important factor for plant growth reduction.

Table 4 Effect of simulated acid rain on photosynthetic tissues biomass of *P. massoniana* (g) and *C. lanceolata* (g)

Plant species	Acid rain treatment				Significant <i>F</i> value	level <i>p</i>
	pH 6.6	pH 4.5	pH 3.0	pH 2.0		
<i>P. massoniana</i>	29.047	24.384	25.057	17.943	<i>F</i> =2.82	0.05
<i>C. lanceolata</i>	15.977	15.998	17.685	9.188	<i>F</i> =7.31	0.01

Table 5 Effect of simulated acid rain on photosynthetic rate per unit leaf dry weight (CO₂ mg/g leaf dry weight/h)

Plant species	Acid rain treatment				Significant <i>F</i> value	level <i>p</i>
	pH 6.6	pH 4.5	pH 3.0	pH 2.0		
<i>P. massoniana</i> sapling	8.577	8.123	7.930	5.796	2.19	—
<i>C. lanceolata</i> sapling	10.230	10.387	11.609	10.316	0.05	—

Effect of simulated acid rain on photosynthesis

Net photosynthetic rate per unit dry weight of *P. massoniana* sapling leaves decreased with acidity increase, but analysis of variance indicated no significant effects of acid rain. Net photosynthetic rate per unit dry weight of sapling leaves of *C. lanceolata* did not decrease (Table 5).

However, photosynthetic rate that was expressed on a per plant basis decreased at a pH of 2.0. Analysis of variance indicated a significant effect of acid rain (Table 6).

Table 6 Effects of acid rain on net photosynthetic rate per plant unit (CO₂ mg/g per plant/h)

Plant species	Acid rain treatment				Significant <i>F</i> value	level <i>p</i>
	pH 6.6	pH 4.5	pH 3.0	pH 2.0		
<i>P. massoniana</i>	249.145	198.071	198.690	104.005	7.71	0.05
<i>C. lanceolata</i>	163.440	166.173	205.302	94.784	7.44	0.05

Ferenbaugh (1976) reported that acid rain treatment dramatically increased the apparent of photosynthesis of *Phaseolus vulgaris* L. as determined by oxygen evolution. Irving (1985) reported that analysis of variance of experimental results indicated on significant acid rain treatment effects on the photosynthetic rate per unit leaf area of *Raphanus sativas* L., but photosynthesis that was expressed on a per plant basis decreases with increasing acidity. Therefore, response of plant photosynthetic rate to acid rain varies with plant species.

Effects of simulated acid rain on respiration

Dark respiration rate of *P. massoniana* and *C. lanceolata* were all increased significantly at pH 2.0 (Table 7). Ferenbaugh (1976) reported simulated acid rain increased the respiration rate of *Phaseolus vulgaris* L. Therefore, respiration rate increases by acid rain is an significant pattern. Respiration rate increases resulting in increasing material consumption was probably an important factor in plant growth reduction from acid rain.

Effects biomass

Main root biomass of *P. massoniana* and *C. lanceolata* saplings decreased at pH 2.0 treatment by analysis of variance indicated no significant effect of acid rain. Lateral root biomass of *P. massoniana* and *C. lanceolata* saplings decreased at pH 2.0 analysis of variance indicated significant effects of simulated acid rain (Table 8).

Table 7 Effects of simulated acid rain on respiration
(CO₂ mg/g leaf dry weight/h)

Plant species	Acid rain treatment				Significant <i>F</i> value	level <i>p</i>
	pH 6.6	pH 4.5	pH 3.0	pH 2.0		
<i>P. massoniana</i>	1.094	1.143	0.910	1.697	4.09	0.05
<i>C. lanceolata</i>	1.518	1.370	1.728	2.047	3.21	0.05

Table 8 Effects of simulated acid rain on root biomass (g)

Plant species	Acid rain treatment				Significant <i>F</i> value	level <i>p</i>
	pH 6.6	pH 4.5	pH 3.0	pH 2.0		
Sapling main root						
<i>P. massoniana</i>	5.124	5.167	5.406	3.272	1.24	—
<i>C. lanceolata</i>	4.769	4.226	4.150	3.535	1.58	—
Sapling lateral root						
<i>P. massoniana</i>	7.648	8.145	6.990	4.635	3.36	0.05
<i>C. lanceolata</i>	14.075	15.541	17.407	10.504	4.86	0.01

Stem biomass

Stem biomass of *C. lanceolata* saplings slightly increased with increasing acidity, but analysis of variance indicated no significant effects of acid rain. Stem biomass of *P. massoniana* saplings decreased at pH 2.0 treatment. Analysis of variance indicated no significant effects of acid rain (Table 9).

Table 9 Effects of simulated acid rain on stem biomass (g)

Plant species	Acid rain treatment				Significant <i>F</i> value	level <i>p</i>
	pH 6.6	pH 4.5	pH 3.0	pH 2.0		
<i>P. massoniana</i>	21.135	22.492	19.734	20.038	0.16	—
<i>C. lanceolata</i>	11.882	13.889	14.360	13.544	0.79	—

Leaves biomass

Leaves biomass decreased at pH 2.0 treatment. Analysis of variance indicated no significant effect on saplings leaves biomasses of *P. massoniana* and *C. lanceolata* (Table 10).

Table 10 Effects of simulated acid rain on leaf biomass (g)

Plant species	Acid rain treatment				Significant <i>F</i> value	level <i>p</i>
	pH 6.6	pH 4.5	pH 3.0	pH 2.0		
<i>P. massoniana</i>	29.047	24.384	25.057	21.035	1.38	—
<i>C. lanceolata</i>	15.977	15.998	17.685	15.547	0.40	—

DISCUSSION

These experimental results showed that, among three parts of plant, stems were the most resistant one and roots were the most sensitive one to acid rain exposure. Lateral roots were more sensitive than main roots to acid rain exposure. Raynal *et al.* (1982) reported that simulated acid rain at pH 2.0 decreased total plant weight of *Acer saccharum* resulting from weight of root reduction but no change in leaf weight or leaf width. Lee *et al.* (1979) reported that root growth of *Rhus typhina* was restrained, but the growth of above ground parts was not affected. The results of our experiment are consistent with the Lee and Raynal *et al.* A major reason may be that roots have no cuticle and always stay in soil being acidified gradually, but it is also more likely that the reduction of whole plant photosynthesis resulted in less carbon being used for root growth, therefore an indirect effect on roots should also be a possibility. It will be studied in future.

Simulated acid rain at pH 2.0 resulted in restrained plant growth through photosynthetic tissue reduction (Wood and Bormann, 1974). Plants treated with the highest acidity level (pH 2.6) had reduced leaf area which resulted in lower photosynthesis per plant and may have been responsible for the reduced yields of plants (Irving, 1985). These results showed that simulated acid rain caused decreased biomass resulting from increasing respiration which increased dry material consumption and decreased photosynthetic tissue which, in turn, resulted in a dry material production decreasing.

In most cases, growth inhibition occurred only with simulated acid rain of pH < or = 3 (Amthor, 1984). These results showed that the threshold of simulated acid rain pH which resulted in visible injury or significant deleterious effects was at a pH of 3.0 or lower. However, we believe the threshold of atmospheric acid rain pH which resulted in visible injury or significant deleterious effects was 3.0 or lower. However, we believe the threshold of atmospheric acid rain pH which resulted in visible injury or significant deleterious effects was higher than that of simulated acid rain. Five possible factors are discussed as follows: (1) there are also organic acids and other weak acids that result in higher acidity in atmospheric acid rain than in simulated acid rain at same pH level. An additional experiment showed that acetic-simulated acid rain resulted in more severe deleterious effects on leaves of *Metasequoia glyptostroboides* than sulfuric or chloric acid simulated rain at a pH 2.0; (2) atmospheric acid rain contains sulphurous acid which is 30 times as phytotoxic as sulfuric acid to plants; (3) cumulative injurious effects of atmospheric acid rain over several years resulted in more severe deleterious effects on woody perennials than short periods of simulated acid rain over several weeks or months; (4) the combined effects of pollutants (SO₂, O₃, NO_x etc.) and acid rain on plant resulted in more seriously deleterious effects than simulated acid rain alone; (5) indirect effects which resulted from soil acidification result in plants being more seriously damaged and more sensitive to acid rain.

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