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Effects of simulated acid rain on the injury and physiological responses of crops

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Abstract— Experiment was performed to determine the injury and physiological responses of plant foliage to simulated acid rain. Cross section of injured leaves, pH value of foliar homogenate, electrolyte leakage, malondialdehyde content, content of peroxidase and some other enzymes were examined after exposure to simulated acid rain. Plants exhibited visible leaf injury when exposed to simulated acid rain at pH values of 2.5 and 3.0. Peroxidase content and isozyme bands increased significantly after the treatment at pH 3.0. Injured plants had higher electrolyte leakage and higher levels of malondialdehyde. This showed that simulated acid rain induced lipid peroxidation and membrane damage.

Keywords: acid rain; crops; plant foliage; biological effects.

1 Introduction

Effects of acid deposition on plant have been widely studied. The results showed that the biological effects are numerous and complex. Experimental results about the impact of acid rain on crop yields have been especially controversial among investigators. To solve these problems, further work on responses of plants to the stress of acid deposition and other air pollutants is necessary. Since early year, physiological responses of plants have been used to reflect the stresses which have been exposed, and much work was reported on the effects of acid rain on physiological activities such as foliar leaching, photosynthesis and respiration. However, few results have been given about the effects on enzyme and membrane systems, which are important for evaluating the possible mechanisms of the impacts of acid deposition on plants.

The present study examines the changes in electrolyte leakage, peroxidase and catalase contents and malondialdehyde levels on which acid rain brings about, exploring the sensitivity of the physiological index and some possible mechanism of the effects.

2 Material and methods

Seedlings were grown in a green chamber at a mean temperature of 25°C with a 14-hour-photoperiod daily in each treatment with 5 repetitions. After 10 days, the plants were sprayed with simulated acid rain for 1 hour daily for 10 days. The acid rain solution was prepared in accordance with the composition of statistical result of Chongqing rainwater during 1982－1985, which contains mixtures of sulfuric and nitric acid with background concentrations of chloride, potassium sodium, calcium, ammonium and magnesium ions. These amounts were based on measurements taken of concentrations in rain water at Chongqing City in southwestern China.

Electrolyte leakage was measured according to the methods described by Robert (Robert, 1980). Peroxidase (POD) and catalase (CAT) activities were measured as described by Jin (Jin, 1981). Levels of MAD (malondialdehyde) were measured as described by Chia (Chia, 1981).

3 Results and discussions

3.1 Leaf injury

Substantial foliar injury was observed with the treatment of simulated acid rain at pH 2.5 and 3.0, but not at pH 3.5 and above. The injured plants' sizes were inhibited, and the young leaves wrinkled and abscised prematurely. Upon microscopic examination, within cross sections of injured leaves outer epidermal cells in some areas collapsed, and the palisade and spongy mesophyll layer were disordered and undistinguished.

3.2 pH value of foliar homogenate

No significant change of the pH values of foliar homogenate (1g of fresh leaf was homogenized with 6 ml deionized H_2O) was observed after exposure to simulated acid rain at different pH values (Table 1), neither after steeping and immersing the leaved in acidic solution for 5 hours. This shows that there is no relationship between the pH value of foliar homogenate and the effect of simulated acid rain on the acidity of treatment tissue.

The epidermal cells and bundle sheath parenchyma of leaves treated at pH 2.5 were acidified by staining fresh tissue sections with Congo red indicator. Using a new technique to measure the acidity of intercellular environment will be helpful to the determination of the effect of acid rain on the intercellular environment of foliage.

Table 1 Effect of simulated acid rain on pH values of foliar homogenate

Treatment pH	2.5	3.0	3.5	4.0	Control
Exposure	6.58	6.52	6.42	6.67	6.53
Soaking for 5 h	6.58	6.66	6.63	6.53	6.53

3.3 Electrolyte leakage

Cowpea (Vigna sinensis Endl) exposed to simulated acid rain below pH 3.0 increased foliar electrolyte leakage significantly (Fig. 1). The extent of imbibitional leakage is one of the indexes of differential permeability of plasma membranes. The foliar ion leakage has been evaluated to increase during the exposure to some s' -sses such as SO_2, O_3, coldness, drought and high temperature (Irving, 1981).

The result shows that simulated acid rain also induced imbibitional leakage and destruction of differential permeability of the plasma membrane.

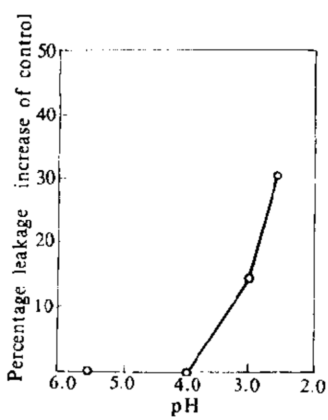

Fig. 1 Electrolyte leakage-exposure to simulated acid rain

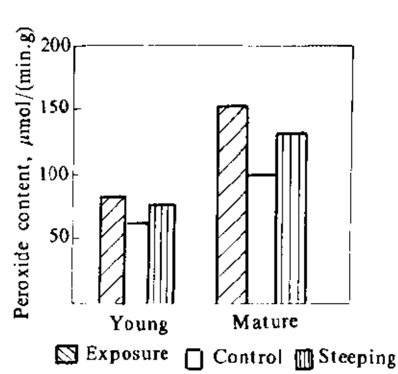

Fig. 2 Peroxide content response to simulated acid rain at pH 3.0

1. Exposure; 2. Control; 3. Soaking

3.4 Peroxidase content and isozyme patterns

Peroxidase is one of the enzyme systems with sprayed in aerobic cells protecting against deleterious substances. Some reports suggested that plant exposed to sulfur dioxide enhance peroxidase activity (Wellbum, 1976). In this experiment, the result shows that the peroxidase activity increased by 45% (mature leaves) and 51% (young leaves, Fig. 2), peroxidase isozyme bands also increased, which is determined by polyacrylamide gel electrophoresis (Fig. 3) . This indicates that peroxidase is a sensitive physiological index to stress of acid rain.

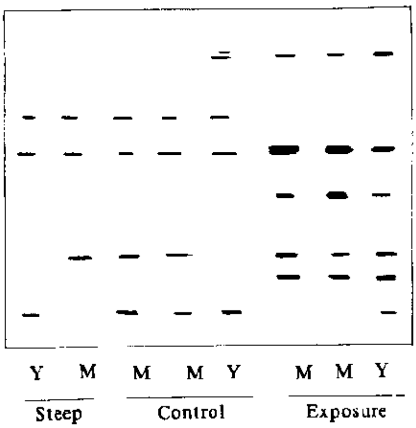

Fig. 3 Responses of peroxidase isozyme patterns to simulated acid rain at pH 3.0
Y— Young leaf; M— Mature leaf

After steeping the foliage of the plants in acid solution for 5 hours, peroxidase activity increased, but there was no change in peroxidase isozyme patterns. This shows that the chronic effect of acid rain induces the biosynthesis of some new peroxidase isozymes.

In this experiment, the treated plants are inhibited. This effect may be related to the enhancement of peroxidase activity, as peroxidase plays an important role in adjusting the biosynthesis of auxin.

3.5 Catalase content

Catalase is another important enzyme of profecting aerobic cells. It is concentrates in peroxisome and degrades H_2O_2 to prevent the occurrence of deleterious H_2O_2-depended reactions. The treated plant shows slight decreases of catalase activity (Table 2). Compared with peroxidase, it is less sensitive to the treatment of acid rain.

Table 2 Catalase content responses to simulated acid rain at pH 3.0

Unit: μm/(min. g FW)

Leaf age	Treatment		
	Control	Exposure	Steeping
Young	929.6	909.6	840.0
Mature	928.0	909.2	901.6

3.6 Malondialdehyde content

Malondialdehyde (MDA) is the product of lipid peroxidation. The results show that the content of MDA increased after treatment of simulated acid rain at pH 2.5 and 3.0 (Fig. 4) . The same results were found on another plant, green pepper (Capsicum frutescens Uar. Loungun Bailey). MDA was suggested as an index of lipid peroxidation, and some other symptoms of lipid peroxidation were observed in this experiment such as electrolyte leakage and premature foliar abscission. It indicates that simulated acid rain induces lipid peroxidation and membrane damage of foliage.

The lipid peroxidation can be induced by some stresses such as chilling, heavy metal ions, SO_2 and O_3 (Omran, 1980; Lizada, 1981). Lipid peroxidation was induced directly by free radical arising from the metabolism process or from the environment, and hydrogen ion concentration is the phytotoxic composition of simulated acid rain to plant, thus, free radical that induced lipid peroxidation after treatment of simulated acid rain would arise from metabolism rather that from environment. Further study is needed to determine the levels of free radicals such as the superoxide radical produced probably from acid-rain-treated tissue using electron spin renonance (ESR).

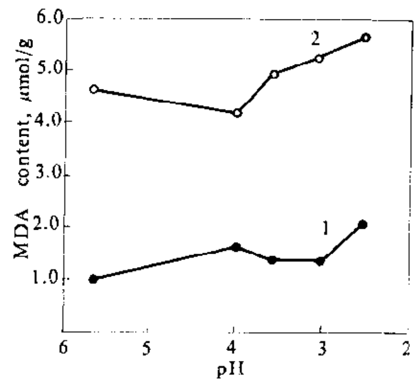

Fig. 4 Level of malondialdehyde (MDA) in foliage after exposure to simulated acid rain
1. Cowpen 2. Green pepper

It has been suggested that peroxidation of membrane lipid plays an important role in SO_2 and O_3 injuries to plants, and lipid peroxidation is considered to be the toxicological mechanism of the interaction of SO_2 and O_3 on plants. Other results show that acid precipitation may have an interaction effect with sulfur dioxide and ozone on plants. Thus lipid peroxidation would be one of the possible toxicological mechanisms of this interaction.

3.7 Superoxide dismutase isozyme patterns

Superoxide dismutases (SOD), present in almost all parts of higher plants, are specific enzymes that play a role in prevention of cell organelles from oxygen toxicity such as lipid peroxidation. The result from polyacrylamide gel electrophoresis shows that there are two more isozyme bands in young leaves than mature leaves, but there is no difference between control and treated plants (Fig. 5).

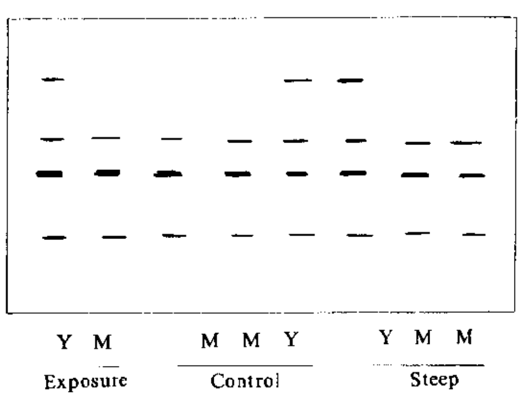

Fig. 5 Response of superoxide dismutase isozymes to simulated acid rain at pH 3.0
Y— Yong leaf; M— Mature leaf

Assay on superoxide dismutases activity may determine the toxicological effect of simulated acid rain on plants.

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