Journal of
Plant Pathology & Microbiology

Chung and Chung, J Plant Pathol Microbiol 2016, 7:6
DOI: 10.4172/2157-7471.1000358

Research Article Open Access

Development of the Biochemical Parameters of Selected Plants Exposed with Ozone Gas

Chung-Yi Chung[1]* and Pei-Ling Chung[2]

[1]*Department of Tourism Management, Tajen University, Pingtung, Taiwan, R.O.C*
[2]*Department of Hospitality Management, Tajen University, Pingtung, Taiwan, R.O.C*

Abstract

Air pollution tolerance index (APTI) is used by landscapers to select plant species tolerant to air pollution. To develop the usefulness of plants as bioindicators requires an appropriate selection of plant species, which is of utmost importance in specific situations. In order to evaluate the tolerance level of plants to air pollutants, four parameters, namely ascorbic acid, chlorophyll, relative water content, and leaf-extract pH, were determined and computed together in a formulation signifying the air pollution tolerance index (APTI) of plants. The study examined the APTI of seven selected species which were fumigated with 60 ppb ozone gas (O_3) for 10 days. The results showed that the APTI of *Palaquium formosanum* exhibited the most significant decrease after fumigation with O_3. Based on the nature of their tolerance, the experimental plants could be arranged in tolerance order as *Plaquium formosanum* > *Aglaia formosana* > *Cerbera manghas* > *Nageia nagi* > *Millettia pinnata* > *Terminalia catappa* > *Tournefortia argentea*.

Keywords: Biochemical; Plant; Ozone gas; Air pollution tolerance index

Introduction

Air pollution is the human introduction into the atmosphere of chemicals, particulate matter, or biological materials that cause harm or discomfort to humans or other living organism, or damage the environment. Air pollutants can directly affect plants via leaves or indirectly via soil acidification [1]. Plants provide an enormous amount of leaf area for impingement, absorption and accumulation of air pollutants and thus reduce the pollutant levels in the air [2], with a varied extent of mitigation for different species [3]. Plants also can green the environment, clean the air and adjust the climate. In both city and industrial areas, it is crucial to improve the air quality with long term planting and maintenance efforts, so plants should therefore readily adapt to the surrounding environment, even in a polluted environment [4,5]. Different plant species vary considerably in their tolerance to air pollutants [6]. The identification and categorization of plants into tolerant groups is important because the former can serve as indicators and the latter as sinks for the abatement of air pollution. It has been reported that when exposed to air pollutants, most plant experience physiological changes before exhibiting visible damage to leaves. Previous studies also showed the impact of air pollution on ascorbic acid content [7], chlorophyll content [8], leaf extract pH [9] and relative water content [10]. These separate parameters gave conflicting results for same species. However, the air pollution tolerance index (APTI) based on all parameters has been used for identifying tolerance levels of plants species [11-15].

Ozone gas (O_3) can harm the human respiratory system, and it can also impact plant health. The cells membranes of plant leaves are damaged under conditions of high concentrations O_3 in the environment. Cells die, and then symptoms appear on the leaves. When a plant comes into contact with O_3, the common symptoms are leaf rolling and drooping, as well as dark brown and ashen spots, among other visible symptoms. In order to achieve better air quality in industrial settings and urban environments, planted trees should have high tolerance toward pollutants. In this research, the O_3 concentrations of the experiment were set at 60 ppb. This research assess the tolerance of common green trees to O_3 in order to use this as an indicator of air quality, and fumigation equipment was utilized for this purpose. The results lead to a better understanding of plant tolerance to O_3.

Material and Methods

Experiment setup

Leaves of seven kinds of Taiwan protophyte plants were collected for this experiment, including *Aglaia formosana* (*A. f.*), *Cerbera manghas* (*C. m.*), *Millettia pinnata* (*M. p.*), *Nageia nagi* (*N. n.*), *Palaquium formosanum* (*P. f.*), *Tournefortia argentea* (*T. a.*), and *Terminalia catappa* (*T. c.*). The tested plants were put in a fumigation chamber for this experiment. The chamber was transparent and was placed in natural sunlight. An air conditioner was placed in the chamber to control the temperature between 25 to 30°C during the fumigation experiment. A fan was used on the inside ceiling of the chamber to mix the O_3 with the inner air in order to achieve a uniform O_3 concentration environment in the chamber. An O_3 gas manufacture machine (OW-K1/A-O, Taiwan) supplied the O_3 gas into the chamber using a calibrated flow meter to control the fumigation volume of the O_3. An O_3 gas detector (Photometric O_3 Analyzer-model-400a, USA) measured the O_3 concentration in the chamber, and as the concentration changed, it was adjusted to keep the gas concentration constant in the chamber during the experiment. The O_3 concentration in the sample was calculated as following equation [16],

$$C_{O_3} = -\frac{10^9}{a \times L} \times \frac{T}{T_0} \times \frac{P_0}{P} \times \ell n \frac{I}{I_0}, \tag{1}$$

where

C_{O3} = O_3 concentration

a = Absorption efficiency

***Corresponding author:** Chung-Yi Chung, Associate Professor, Department of Tourism Management, Tajen University, Pingtung, Taiwan, R.O.C, Tel: +88687624002; Fax: +88687628033; E-mail: yi@tajen.edu.tw

Received May 24, 2016; **Accepted** June 14, 2016; **Published** June 16, 2016

Citation: Chung CY, Chung PL (2016) Development of the Biochemical Parameters of Selected Plants Exposed with Ozone Gas. J Plant Pathol Microbiol 7: 358. doi:10.4172/2157-7471.1000358

L = Light wave length

I_0 = Incidence ultraviolet light strength

I = Outlet ultraviolet light strength

T_0 = Temperature of standard gas temperature (273 K)

P_0 = Pressure of standard gas (29.92 in-Hg)

T = Temperature of sample

P = Pressure of sample

4.2 Air pollution tolerance index

During present investigation ascorbic acid content, leaf extract pH, total chlorophyll content and relative water content was taken together in the form of mathematical expression to obtain an empirical value, signifying the air pollution tolerance index (APTI) [17].

$$\mathrm{APTI} = \frac{\mathrm{A} \times (\mathrm{T} + \mathrm{P}) + \mathrm{R}}{10}, \quad (2)$$

where

A = Ascorbic acid content (mg g^{-1} fresh wt.)

P = Leaf extract pH

T = Total chlorophyll (mg g^{-1} dry wt.)

R = Percent relative water content of leaf (%).

Ascorbic acid content was measured using spectrophotemetric method. 1 g of the fresh foliage was put in a test-tube, 4 mL oxalic acid - EDTA extracting solution was added, then 1 mL of orthophosphoric acid and then 1 mL 5% tetraoxosulphate (VI) acid added to this mixture, 2 mL of ammonium molybdate was added and then 3 mL of water. The solution was then allowed to stand for 15 minutes. After which the absorbance at 760 nm was measured with a spectrophotometer. The concentration of ascorbic acid in the sample was then extrapolated from a standard ascorbic acid curve.

Fresh leaf (0.5 g) sample was homogenized in 10 ml deionized water. This was then filtered and the pH of leaf extracted determined after calibrating pH meter with buffer solution of pH 4 and pH 9.

Total chlorophyll content in the leaves, 3 g of fresh leaves were blended and then extracted with 10 mL of 80% acetone and left for 15 minutes for thorough extraction. The liquid portion was decanted into another test-tube and centrifuged at 2,500 rpm for 3 minutes. The supernatant liquid was then collected and the absorbance taken using a spectrophotometer at 645 nm and 665 nm.

Relative leaf water content the fresh leaves were weighed and then immersed in water over night, blotted dry and then weighed to get the turgid weight. The leaves were dried overnight in an oven at 55°C and reweighed to obtain the dry weight.

Results and Discussion

The highest leaf ascorbic acid content of tested trees was recorded in *Palaquium formosanum* (6.67 mg/g FW), and the lowest was recorded in *Nageia nagi* (1.16 mg/g FW) before fumigation (Table 1). After 60 ppb O_3 fumigation for 10 days, most of the tested trees' ascorbic acid was lower than before fumigation except *Aglaia formosana*, *Nageia nagi* and *Terminalia catappa.* The *Palaquium formosanum* showed the greatest drop with the reducing rate 56%. But the ascorbic acid value of *Palaquium formosanum* also was the highest of all tested trees after fumigation. The ascorbic acid of *Nageia nagi* displayed a 62% increase, which was the most increasing rate than other plants.

A range from 38.6 mg/g of *Tournefortia argentea* to 67.4 mg/g of *Nageia nagi* was observed in this experiment for chlorophyll content before fumigation (Table 2). After fumigation with 60 ppb O_3 for 10 days continually, the chlorophyll content did not show significant variance, the *Tournefortia argentea* and *Millettia pinnata* had a decrease of 1 - 2% after O_3 gas fumigation, the other plants showed increase with a range 1 - 9%.

The leaf extract pH was recorded from 5.21 of *Terminalia catappa* to 6.47 of *Aglaia formosana* before fumigation. After 60 ppb O_3 fumigation for 10 days, the pH of the *Tournefortia argentea*, *Millettia pinnata* and *Palaquium formosanum* increased by 3%, and the other trees showed a lower pH than before fumigation ranging from 3% (*Cerbera manghas*) to 8% (*Terminalia catappa*) (Table 3).

The results of the study showed the leaf relative water content to vary from 83% for *Cerbera manghas* to 94% for *Tournefortia argentea* before fumigation. After fumigation, the leaf relative water content of the test trees ranged from 86% to 93%. The variance under the O_3 environment of tested plants was -3% for *Millettia pinnata* to +8% for *Aglaia formosana* (Table 4).

The APTI is an index indicating the tolerance of plants to air

Species	Before (mg/g)	After (mg/g)	Relative (%)[a]
A. f.	1.82	2.28	125
C. m.	2.53	1.74	69
M. p.	1.79	1.45	81
N. n.	1.16	1.87	162
P. f.	6.67	2.89	43
T. a.	1.56	1.29	83
T. c.	1.77	1.79	101

[a]Relative rate(%) = After/Before fumigation

Table 1: Leaf ascorbic acid content of experiment plants before and after fumigation with 60 ppb O_3 gas for 10 days.

Species	Before (mg/g)	After (mg/g)	Relative (%)[a]
A. f.	64.2	64.6	101
C. m.	48.5	51.6	106
M. p.	50.0	49.7	99
N. n.	67.4	67.2	100
P. f.	58.8	61.1	104
T. a.	38.6	38.0	98
T. c.	42.2	46.1	109

[a]Relative rate(%) = After/Before fumigation

Table 2: Leaf total chlorophyll content of experiment plants before and after fumigation with 60 ppb O_3 gas for 10 days.

Species	Before	After	Relative (%)[a]
A. f.	6.47	6.12	95
C. m.	6.11	5.95	97
M. p.	6.47	6.68	103
N. n.	6.17	5.89	95
P. f.	5.62	5.81	103
T. a.	6.44	6.63	103
T. c.	5.21	4.8	92

[a]Relative rate(%) = After/Before fumigation

Table 3: Leaf extract pH of experiment plants before and after fumigation with 60 ppb O_3 gas for 10 days.

Species	Before (%)	After (%)	Relative (%)[a]
A. f.	85	92	108
C. m.	83	87	105
M. p.	94	91	97
N. n.	93	93	100
P. f.	86	91	106
T. a.	94	92	98
T. c.	84	86	102

[a]Relative rate = After/Before fumigation

Table 4: Leaf relative water content of experiment plants before and after fumigation with 60 ppb O_3 gas for 10 days.

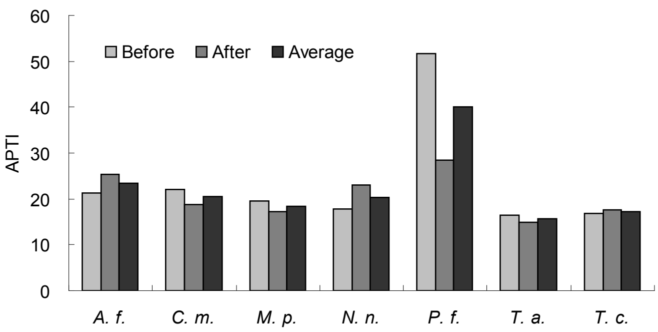

Figure 1: APTI of experiment plants fumigated with 60 ppb O_3 gas for 10 days.

pollution. Higher values indicate higher resistance to air pollutants [18,19]. The average APTI before and after fumigation showed in Figure 1. The results showed the range for *Tournefortia argentea* (16.4) to *Palaquium formosanum* (51.6) before fumigation. After fumigated with 60 ppb O_3 gas for 10 days, the *Tournefortia argentea* and *Palaquium formosanum* also had the lowest and highest APTI which were 15.0 and 28.4, respectively. The *Palaquium formosanum* showed a most decrease rate of 45% as compared to before fumigation and the *Nageia nagi* had the highest increase rate 29%. The next higher raise was *Aglaia formosana* with increase rate 18% and *Terminalia catappa* was 5% higher than before fumigation. The final tolerance order to O_3 could display as *Palaquium formosanum* > *Aglaia formosana* > *Cerbera manghas* > *Nageia nagi* > *Millettia pinnata* > *Terminalia catappa* > *Tournefortia argentea*. The observations in this study suggest that plants have the potential to serve as excellent quantitative and qualitative indices of pollution. Since biomonitoring of plants is an important tool by which to evaluate the impact of air pollution on plants, *Palaquium formosanum* can be used as biomonitors of O_3 gas pollution.

Conclusions

APTI index combined a variety of parameters can yield more reliable results as compared to an APTI only based on a single biochemical parameter. This study provided useful information by which to select tolerant species fit for landscape on sites continuously exposed to O_3 gas. In this research, a fumigation experiment was used to evaluate the air pollution tolerance index characteristics of seven kinds plants with 60 ppb O_3 gas for 10 days. The results showed *Palaquium formosanum* had the most tolerance to O_3 and *Tournefortia argentea* was the weakest among seven tested plants. The species are ranked by ability to tolerate O_3 gas, the experimental plants could be arranged in the following order: *Palaquium formosanum* > *Cerbera manghas* > *Aglaia formosana* > *Millettia pinnata* > *Nageia nagi* > *Terminalia catappa* > *Tournefortia argentea*.

Acknowledgements

This research was financially supported by the National Science Council, Taiwan, R.O.C. (NSC 99-2632-H-127-001-MY3 and NSC 100-2632-E-127-001-MY3).

References

1. Steubing L, Fangmeier A, Both R (1989) Effects of SO_2, NO_2, and O_3 on population development and morphological and physiological parameters of native herb layer species in a beech forest. Environment and Pollution 58: 281-302.
2. Escobedo FJ, Wagner JE, Nowak DJ, De la Maza CL, Rodriguez M, et al. (2008) Analyzing the cost effectiveness of Santiago, Chile's policy of using urban forests to improve air quality. J Environ Manage 86: 148-157.
3. Hove LWA, Bossen ME, Bok FAM (1999) The uptake of O_3 by poplar leaves: The impact of a long-term exposure to low O_3-concentrations. Atmospheric Environment 33: 907-917.
4. Chung CY, Chung PL, Liao SW (2011) Carbon fixation efficiency of plants influenced by sulfur dioxide. Environ Monit Assess 173: 701-707.
5. Chung CY, Chung PL, Gau HS, Liao SW, Lai WL, et al. (2012) Assessment of the Tolerance of Trees to High Concentration Sulfur Dioxide. Advanced Materials Research 518-523: 5423-5428.
6. Dwivedi AK, Tripathi BD (2007) Pollution tolerance and distribution pattern of plants in surrounding area of coal-fired industries. J Environ Biol 28: 257-263.
7. Hoque MA, Banu MN, Okuma E, Amako K, Nakamura Y, et al. (2007) Exogenous proline and glycinebetaine increase NaCl-induced ascorbate-glutathione cycle enzyme activities, and proline improves salt tolerance more than glycinebetaine in tobacco Bright Yellow-2 suspension-cultured cells. J Plant Physiol 164: 1457-1468.
8. Flowers MD, Fiscus EL, Burkey KO (2007) Photosynthesis, chlorophyll fluorescence, and yield of snap bean (*Phaseolus vulgaris* L.) genotypes differing in sensitivity to ozone. Environmental and Experimental Botany 61: 190-198.
9. Klumpp G, Furlan CM, Domingos M, Klumpp A (2000) Response of stress indicators and growth parameters of Tibouchina pulchra Cogn. exposed to air and soil pollution near the industrial complex of Cubatão, Brazil. Sci Total Environ 246: 79-91.
10. Agbraire PO, Esiefarienrhe E (2009) Air Pollution tolerance indices (apti) of some plants around Otorogun Gas Plant in Delta State, Nigeria. Journal of Applied Sciences and Environmental Management 13: 11-14.
11. Singh SK, Rao DN, Agrawal M, Pandey J, Narayan D (1993) Air pollution tolerance index of plants. Journal of Environmental Management 32: 45-55.
12. Karthiyayini R, Ponnammal NR, Joseph R (2005) Air pollution tolerance index of certain plants of Coimbatore-ooty highyways, near I.T.I area, Coimbatore, Tamilnadu. Pollution Research 24: 363-365.
13. Lakshmi PS, Sravanti KL, Srinivas N (2008) Air pollution tolerance index of various plant species growing in industrial areas. The Ecoscan 2: 203-206.
14. Agbaire PO (2009) Air pollution tolerance indices (APTI) of some plants around Erhoike-Kokori oil exploration site of Delta State, Nigeria. International Journal of Physical Science 4: 366-368.
15. Jyothi JS, Jaya DS (2010) Evaluation of air pollution tolerance index of selected plant species along roadsides in Thiruvananthapuram, Kerala. Journal of Environmental Biology 31: 379-386.
16. Ingle JDJ, Crouch SR (1988) Spectrochemical Analysis. New Jersey: Prentice Hall.
17. Singh SK, Rao DN (1983) Evaluation of the plants for their tolerance to air pollution. (Paper presented at the air pollution control, IIT, Delhi) pp: 218-224.
18. Ninave SY, Chaudhari PR, Gajghate DG, Tarar JL (2001) Foliar biochemical features of plants as indicators of air pollution. Bull Environ Contam Toxicol 67: 133-140.
19. Panigrahi T, Das KK, Dey BS, Mishra M, Panda RB (2012) Air Pollution Tolerance Index of various plants species found in F.M. University Campus, Balasore, Odisha, India. Journal of Applicable Chemistry 1: 519-523.