KIMIKA 14: (2) 75-82 (1998)
Printed in the Philippines

Assessment of the Ambient Air Quality in Iligan City Using Standard and Phytomonitoring Methods

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Standard chemical and spectroscopic methods for the determination of sulfur dioxide (SO_2), nitrogen dioxide (NO_2), suspended particulate matter (SPM), and lead (Pb) were employed to analyze air samples collected from three study sites in Iligan City, during the period June 1997 to January 1998. At the same time, leaf samples of five trees were collected and analyzed for leaf extract pH, ascorbic acid, chlorophyll, and relative water content. Correlation analysis between air pollutant variables and plant pollution indicators showed that increased levels of pollution resulted in elevated leaf extract pH, depression of chlorophyll, and relative water content. The amount of ascorbic acid was not uniformly affected by the relative dose of air pollutants.

The measured leaf parameters indicated that the responses of the five trees (*Artocarpus heterophyllus, Chrysophyllum caimito, Mangifera indica, Swietenia mahogani,* and *Tsidum guajava*) to air pollution are varied. Among them, *A. heterophyllus* and *M. indica* showed the highest sensitivity towards pollution loading. The least sensitive is *S. mahogani*, which suggests that the latter is a tolerant tree and is suitable for the belt zone around pollution sites.

Key words: phytomonitoring, ambient air pollutants, leaf extract pH, chlorophyll, relative water content, ascorbic acid

INTRODUCTION

In Iligan City, respiratory infection ranks first in causing infant mortality and general morbidity [1]. The annual morbidity rate caused by acute respiratory infection is over 500/100000 (similar to the sum of all the other nine leading causes), higher than the average rate during 1979 to 1983 (which is less than 300/100000) despite the discovery of better and more effective drugs and improved health and medical facilities. There has been no baseline study or an air pollution-monitoring program to evaluate the deterioration of air quality in Iligan City. Hence, the objectives of this study were to assess the ambient air quality in Iligan City using standard methods and to find an alternative method of monitoring ambient air quality (phytomonitoring) so that the monitoring may continue without the use of expensive air samplers.

It has been known that passenger cars and trucks, power plants, steel plants, chemical industries, and refineries generate most of the NO_x, Pb, and SO_2 emissions, respectively [2]. Essentially all industrial operations (furnaces, kilns, boilers, etc.) involving high temperatures, quarries and even domestic biomass burning release a wide range of pollutants. Iligan City, an industrial hub in Northern Mindanao, is home to about twenty industries that includes a steel plant, power plant, chemical industries, and cement factories.

For public health protection, the DENR has set standards for criteria pollutants: SO_2, NO_2, SPM, and Pb concentrations (Table 1). The principal effect of SO_2 is the alteration of the mechanical function of the upper airway, while NO_2 can penetrate the lungs more deeply and cause pulmonary edema. Epidemiological studies have demonstrated an apparent causal relationship between total SPM and adverse health effects, including excess mortality at total SPM levels of 1000 $\mu g/m^3$, aggravation of bronchitis and reversible changes in pulmonary function of children at total SPM levels of 200 to 500 $\mu g/m^3$ [3]. Pb, on the other hand, affects circulatory, reproductive, nervous, and kidney systems, and adversely affects the mental development of children. However, the syn-

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Table 1. A Comparison of the 24-Hour Average Levels of Air Pollutants in the Three Study Sites with the DENR Standards

Sampling Station	Concentration in μg/Ncm			
	SO_2	NO_2	*SPM*	*Pb*
DENR Standard	180	150	150	1.5
Site 1	11.27	9.26	563.74	0.19
Site 2	18.94	17.58	79.91	0.28
Site 3	5.13	8.34	56.32	0.12

ergistic effects of these pollutants are considered even more damaging than the single effect. Moreover, the elderly, children and persons with lung and heart disorders who have lower threshold levels than normal adults are at high-risk. Hence, the no-effect dose of pollutants can not be accurately set.

In plants and crops, low concentrations of airborne SO_2 and NO_2 can be beneficial in satisfying their sulfur and nitrogen requirements. However, at higher concentrations, damage and yield reductions can result. These pollutants penetrate the plants through the stomata of the leaves. NO_2 is transformed into HNO_2 and HNO_3, which when in excess have injurious effects on enzymatic systems, chlorophyll content and biosynthesis of amino acids. SO_2 is converted to toxic bisulfite and sulfite ions and later to nontoxic sulfate ions. In each case, leaf necrosis can result. In the presence of a combination of pollutants, field and experimental studies showed that additive, synergistic or less than additive effects can result depending on the genus, species or cultivar [4]. Atmospheric SO_2 and NO_2 can also deposit in the soil and lower its pH and reduce growth. Pb reduces the growth of plant roots and absorption of nutrients, but can also accumulate in plant parts without causing visible plant injury.

The response of plants to pollutants is in the form of changes or reactions within the plant. These changes (*i.e.*, chlorophyll content, etc.) can be measured and can indicate levels of pollutants in the environment. A number of studies showed that plant parameters such as, visible foliar injury [5] membrane permeability [6], relative water content [7], carotenoid content [8], chlorophyll content [9], peroxidase, dismutase and glutathione activities [10], leaf extract pH [11] can be used as bioindicators inasmuch as they change dynamically as pollutant exposures change. Singh and coworkers [12] have shown that a combination of plant parameters: ascorbic acid, RWC, total chlorophyll and leaf pH can be used as reliable indicator of air pollutant combination in an urban industrial setting.

In this study, three study sites were chosen: two of which are pollution sites located close to the industries and the one farthest was used as the reference site. Air samples were collected three times during the study period and were analyzed for SO_2, NO_2, SPM, and Pb using standard methods. In each of these study sites, five trees were also studied: *A. heterophyllus* (jackfruit), *C. caimito* (starapple), *M. indica* (mango), *S. mahogani* (mahogany), and *T. guajava* (guava); and four leaf parameters (ascorbic acid, leaf pH, relative water content, and chlorophyll level) were analyzed.

EXPERIMENTAL

Standard Air Pollution Monitoring. Air samples were collected from each of the study sites for 24 h in June, November, 1997 and January 1998. Determination of the criteria pollutant levels were carried out using the following methods: Pararosaniline method for SO_2 [18]; Griess-Saltzman method for NO_2 [18]; and, anodic stripping voltametry for Pb.

Pararosaniline Method for SO_2. SO_2 from the air sample (collected at 0.2, L/min, for 24 h) was absorbed in a 50 mL solution of potassium tetrachloromercurate (TCM) forming a dichlorosulfitomercurate complex. A 5 mL aliquot of the complex was diluted with TCM to 10 mL and was allowed to stand for 20 min to decompose any ozone present. Nitrites were eliminated by reaction with 1 mL of 0.6% sulfamic acid for 10 min. The resulting mixture was reacted with 5 mL pararosaniline and 2 mL of 0.2% formaldehyde to form the intensely colored pararosaniline methyl sulfonic acid. The intensity of the color produced was measured by means of a spectrophotometer at 548 nm and was related directly to the amount of sulfur dioxide present in the corresponding air sample by using a calibration curve. Simultaneous blank and control runs were performed.

Calibration was done by using the following volumes of working sulfite-TCM solution in a series of 25 mL volumetric flasks: 0, 0.5, 1, 2, 3, and 4 mL. The remaining reagents were added as described above. The absorbance was plotted against the total concentration (g SO_2 for the corresponding solution).

Griess-Saltzman Method for NO_2. The nitrogen dioxide present in the air sample was absorbed in an azo dye-forming reagent made by mixing anhydrous sulfanilic acid in glacial acetic acid and N-(1-naphthyl)-ethylenediamine dihydrochloride solution. A stable pink color was produced within 15 min and was read in a spectrophotometer at 550 nm using unexposed reagent as reference. A calibration curve (prepared using graduated amounts of the sodium nitrite solution) was used to determine the NO_2 concentration.

***Suspended Particulate Matter* (SPM).** The atmospheric SPM was collected for 24 h on a preweighed glass-fiber filter by sucking air with a high volume air sampler at a flow rate of 500 to 2000 LPM. The exposed filter, after equilibration for 24 h in the dessicator, was accurately weighed. The SPM was calculated by dividing the weight (g) of the suspended particulate by the volume of air sampled (m^3). A control was simultaneously run to eliminate the effects of humidity of the filter and the collected particulate matter.

Pb Level Determination by Anodic Stripping Voltammetry. The fiber glass filter containing the collected sample was transferred to a 250 mL beaker and covered with 15 mL concentrated redistilled nitric acid. The sample was digested over a hot plate, cooled and treated with another 3 mL of concentrated redistilled nitric acid. Digestion was resumed and completed. generally indicated by a light colored residue. The residue was dissolved by adding 10 mL distilled 1:1 hydrochloric acid. The watchglass and beaker were washed with deionized distilled water and the resulting solution was filtered into a volumetric flask and diluted to 100 mL. A 5 mL aliquot of the sample solution was transferred to the voltammetric analyzer (VA Model Metrohm 693) sample vessel and mixed with 10 mL deionized distilled water and 1 mL KCl. The potential reading was noted and Pb concentration determined from the calibration curve. The latter was prepared using standard solutions of Pb (50, 100, 200, 300, 400, 500, and 600 ppb).

Phytomonitoring Method. The following five trees found in all three study sites were selected for the phytomonitoring method: *A. heterophyllus*, *C. caimito*, *M. indica*, *S. mahogani*, and *T. guajava*. Almost simultaneous with the air sample collection (Table 2), mature leaves from these trees were collected three times during the study period using methods adapted from Kovacs [5]. Mature and fully expanded leaves were collected between 8:00 A.M. to 10:00 A.M. The leaves were sampled by the point center method. From the middle of each long shoot, two leaves of opposite poles were cut-off at 5 m above the ground. About 24 leaves were collected per tree and placed in a black plastic bag.

Table 2. Weather Conditions During the Three Sampling Dates

Sampling Period	Date of Collection	Weather Condition
First Sampling		
Air	June 28 to July 3, 1997	Clear (early morning; sunny (daytime; clear (nighttime) for the three sites, except for Site 2 which was rainy nighttime
Phyto	July 12, 1997	Clear (morning)
Second Sampling		
Air	November 11-13, 1997	Clear (early morning); sunny (daytime); clear (nighttime) for the three sites
Phyto	November 20,1997	Clear (morning)
Third Sampling		
Air	January 14-17, 1998	Clear (early morning); sunny (daytime); clear (nighttime) for the three sites
Phyto	January 21, 1998	Clear (morning)

The pH, ascorbic acid, total chlorophyll and relative water content of leaf extracts were determined. Triplicate runs for each measurement were carried out for each sample.

Ascorbic Acid Determination. The ascorbic acid content (mg/g dry weight) was measured by a modified method of Keller & Schwager [11]. Five gram of finely cut fresh leaves were mashed with 50 mL deionized water using a mortar and pestle. The mixture was suction filtered using Whatman number 4 filter paper and diluted to 100 mL. An 8 mL aliquot of the solution was taken, and 200 mL of 0.2% oxalic acid was added to the solution and titrated with standard fresh 0.04 % aqueous sodium dichlorophenolindophenol solution (microburette) with rapid stirring to a light pink endpoint. Control runs were carried out with commercially available ascorbic acid.

Total Chlorophyll Determination. Total chlorophyll determination (mg/g dry weight) was estimated following a modified method of Helrich [19]. 0.6 g of fresh leaves previously cut with scissors were mixed with 85% acetone, mashed, filtered through cotton gauze and diluted to 50 mL. The absorbance at 660 nm was measured using a colorimeter at 660 nm. Its corresponding concentration was determined from the standard calibration curve previously prepared.

Relative Water Content Determination. This determination was adapted from Noggle and Fritz (20). A fresh leaf was accurately weighed and brought to full turgor in a moist chamber (24°C, 5 h) with its end standing in water. The turgid leaf was weighed accurately and dried to constant weight in an oven at 105°C. The relative water content of a leaf is expressed as a percentage based on the water content at full turgor.

Leaf-Extract pH Determination. The leaf extract pH was measured using methods adapted from Singh, *et al.* [21]. A 2 g fresh leaf sample (finely cut) was homogenized with 20 mL deionized water in a mortar and pestle and filtered. The pH of the combined extract was measured with previously calibrated pH meter with glass electrode.

Statistical Methodology. The analysis of variance (ANOVA) was performed to test for the significance of differences between sites and the significance of differences between species. If the hypothesis of no significant difference is rejected, Duncan Multiple Range Test (DMRT) was used to determine which site differed from each other or which species differed from each other. A level of significance of 0.05 was used in performing the tests.

RESULTS AND DISCUSSION

The three study sites selected represented areas of high, moderate and low air pollution loading. Site 1 (Kiwalan area) is in the immediate vicinity of industrial factories located along the Cagayan de Oro-Iligan Corridor, where the majority of industries (*i.e.*, cement, flour, refractories, power plant, and oil industries-Figure 1) are located. It has the highest air pollution loading. Site 2 has medium air pollution loading, located about 0.7 km from a steel plant and about 2 km from two chemical factories. At the furthermost end or the southwestern boundary of the city, away from the industrial plants, is where Site 3 was chosen. All three sites are either a housing area or a public school.

Standard Air Pollution Monitoring. The average air pollution levels in the three different sites were compared in Table 1. In terms of SPM level, pollution Site 1 had the highest level. It ranged from 490 to 656 μg/Ncm in all three trials, exceeding the DENR standard (150 μg/Ncm) by 276% on the average. The 24 h average SPM concentration of 564 μg/Ncm is comparable to that reported in top urban areas in the world reported in the Global Environmental Monitoring System, 1979 to 1980 [3].

In terms of SO_2, NO_2 and Pb levels, pollution Site 2 contained the highest load but these concentrations do not exceed the DENR standards. It is worthwhile to note however, that in extreme situations when the wind velocity is low, at the same time there is full industrial capacity operation, temporary peak pollution can be up to twice the calculated average concentration [13].

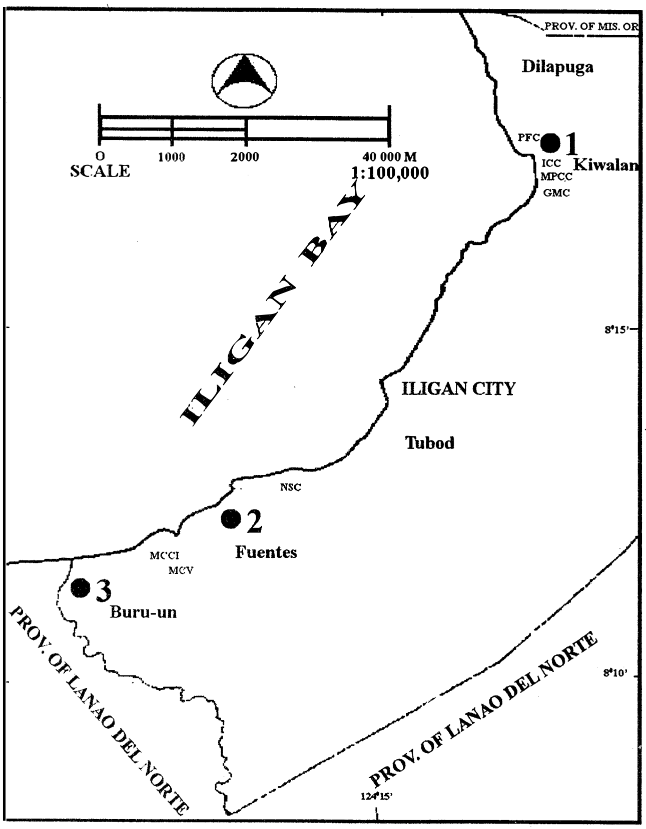

Fig. 1. Map of Iligan City showing location of the three study sites

Analysis of variance for the pollution concentrations (three trials) showed that the control Site 3 is statistically different at the 5% level from the two pollution sites. In effect there are two polluted zones being compared to the reference site, with each polluted zone having a different type of pollution profile. Site 1 is characterized by very high SPM level while Site 2 contain more of the acidic pollutants (SO_2 and NO_2).

Industrial emissions are the main sources of the mentioned air criteria pollutants but the transport sector and domestic burning are also contributory sources. The data in Table 1 concur that Sites 1 and 2 are more polluted than reference Site 3 due to their proximities to the industries and vehicular activities (Fig. 1). It has been known however, that industrial emission through high stacks is easily evenly distributed over a general area, hence it is possible that even the reference site, which is less than 5 km from emission sources and Site 2, may have a pollution profile similar to the latter. Although the reference site had lower pollutant levels than Site 2, the ANOVA test result showed that pollution in Site 2 and reference were not significantly different. Site 1 was, however, significantly different from the other study sites and this is obviously due to its exceedingly high SPM. Hence, for the following discussion, the phytomonitoring results will be discussed in terms of Site 1 versus reference site on one hand, and Site 2 versus reference, on the other.

Leaf pH. The mean leaf pH data (Table 3) at Site 1 were consistently highest in all trees. Between Site 2 and the reference (Site 3), *C. caimito*, *M. indica,* and *T. guajava* measured lower pH in Site 2.

Significant elevation in pH were found in all trees leaf extracts in Site 1 compared to the reference but very small pH changes were observed in Site 2. These results agree with the ANOVA result based on air pollutant levels. In Site 1, where the SPM level is 1000 times higher than reference, all trees studied showed 20% to 30% increase in leaf pH. Studies have shown that cement dust increases soil pH, possibly caused by the hydroxides of calcium and aluminum formed during hydration [14]. Very high pH levels in the leaves can induce coagulation of proteins and hydrolysis of lipids, which ultimately affect crop yield. Among the five trees, the highest pH changes (37% and 30% increase) were registered in *C. caimito* and *M. indica*. The smallest increase in pH was observed in *A. heterophyllus* (19% increase).

Table 3. Mean Leaf pH of FiveTrees Located at the Three Study Sites

Tree Type	Leaf pH*			% Difference	
	Site 1	*Site 2*	*Ref.*	*1 to R*	*2 to R*
A. heterophyllus	7.24 a (0.16)	5.83 b (0.23)	6.07 c (0.16)	+19	-4
C. caimito	6.65 a (0.33)	5.19 b (0.14)	5.11 c (0.20)	+30	+2
M. indica	7.15 a (0.21)	5.32 b (0.30)	5.22 b (0.16)	+37	-2
S. mahogani	6.84 a (0.11)	5.56 b (0.18)	5.66 b (0.07)	+21	-2
T. guajava	6.86 a (0.19)	5.75 b (0.08)	5.61 c (0.13)	+22	-2

* These values represent the means of nine measurements (three trials for each of the three sampling dates). Means followed by a different letter within a row are significantly different at the 5% level. Values in parenthesis represent the standard deviation.

Table 4. Mean Leaf Total Chlorophyll Levels of Five Trees Located at the Three Study Sites

TreeType	Mean Leaf Changes			% difference	
	Site 1	*Site 2*	*Ref.*	*1 to R*	*2 to R*
A. heterophyllus	4.70 c (1.38)	6.84 b (0.35)	9.60 a (1.81)	-51	-29
C. caimito	3.06 c (0.75)	4.58 b (1.41)	6.53 a (1.13)	-53	-30
M. indica	4.61 a (0.86)	3.89 b (0.78)	4.88 a (0.35)	-6	-20
S. mahogani	2.57 b (0.70)	9.52 a (3.88)	7.57 a (2.13)	-66	+26
T. guajava	5.21 b (0.89)	5.03 b (0.72)	6.40 a (0.40)	-19	-21

* These values represent the means of nine measurements (three trials for each of the three sampling dates). Means followed by a different letter within a row are significantly different at the 5% level. Values in parenthesis represent the standard deviation.

A comparison between Site 2 and the reference site showed that the former had higher pollution loading than the latter in all parameters determined. Specifically, Site 2 had 370% SO_2, 210% NO_2, 140% SPM, and 230% Pb content of reference Site 3. Due to the much higher concentrations of the acidic pollutants (SO_2 and NO_2) in Site 2, it was expected that a lower leaf extract pH will be measured in this site. However, at these pollution levels, no significant differences were observed. Except for *A. heterophyllus* leaves (4% decrease), the pH values in the other species vary only by about 0.1 unit. It is possible that the acidic effects of the pollutants in this area may have been partially neutralized by other pollutants, or the acidic pollutant levels were not sufficient to cause drastic reduction in leaf pH. It should be noted that study Site 2 is located between study Sites 1 and 3.

Statistical analysis of leaf pH data showed there is significant difference between Site 1 and the two other sites but none between Site 2 and reference Site 3. This is consistent with the ANOVA result using air criteria pollutant concentration data (standard monitoring). The following species grouping was found significantly different: *A. heterophyllus* (Group I); *S. mahogani* and *T. guajava* (Group II); and, *C. caimito* and *M. indica* (Group III). Among the five trees, *C. caimito* and *M. indica* registered the biggest changes in pH and can be considered as the best indicators studied for pollution in terms of leaf pH.

Total Chlorophyll **(TC).** Except for mahogany, the leaf total chlorophyll content of all leaf species sampled was highest in the reference (Site 3). This result is expected since increased pollution loading reduces chlorophyll level as part of the plants' response to tolerate stress [12]. Comparison between the two pollution sites showed that three out of the five trees (*A. heterophyllus, C. caimito,* and *S. mahogani*) contained higher chlorophyll levels in Site 2 than in Site 1. The mean leaf total chlorophyll levels and their percent difference from the reference site data are given in Table 4.1

The total chlorophyll levels of all trees studied in Site 1 were lower compared to their counterparts in the reference site. Not only SO_2 and NO_2 are known causative agents for lowering of chlorophyll content but cement dust as well [14,15]. Cement dust not only coats leaf surfaces and reduce the amount of light for photosynthesis but is also absorbed in the form of the alkaline solution that can partially denature chloroplasts and decrease the chlorophyll level [16]. Among the five trees, the largest decrease in total chlorophyll content was observed in *S. mahogani* (66%). The tree which registered the smallest depression in total chlorophyll content is *M. indica* (6%).

In Site 2, the trees showing the largest leaf TC reduction (29% and 30%) were *A. heterophyllus* and *C. caimito*. It is worth noting that *S. mahogani* had exhibited a reverse trend in that higher (26% more) TC level was measured in Site 2 than in the reference site. However, *A. heterophyllus* and *C. caimito* remained to be the most sensitive among them in terms of total chlorophyll content since both of them showed the largest TC reductions in both pollution sites.

Using DMRT on the chlorophyll data, it was found that the three sites are significantly different from each other. It is to be noted that the leaf pH and the standard air pollutant data did not show significant variation between Site 2 and reference site. Hence, the leaf TC data is a more sensitive indicator of air pollution loading. Furthermore, the following grouping of tree species was found to be significantly different: *A. heterophyllus* and *S. mahogani* (Group I); *S. mahogani* and *T. guajava* (Group II); and, *C. caimito, M. indica, T. guajava* and (Group III).

Table 5. Mean Leaf Relative Water Content of Five Trees Located at the Three Study Sites

Tree Type	Leaf Relative Water Content*			% Difference	
	Site 1	*Site 2*	*Ref.*	*1 to R*	*2 to R*
A. heterophyllus	80.43 b (7.53)	87.67 a (1.05)	89.23 a (4.13)	-10	-2
C. caimito	82.81 b (6.09)	84.34 b (7.71)	92.11 a (6.65)	-10	-8
M. indica	90.34 b (4.59)	92.55 b (1.87)	97.40 a (1.85)	-7	-5
S. mahogani	84.83 a (4.54)	78.29 b (4.37)	84.61 a (7.25)	<+1	-7
T. guajava	78.75 b (5.84)	84.66 a, b (7.12)	86.41 a (6.37)	-9	-2

* These values represent the means of nine measurements (three trials for each of the three sampling dates). Means followed by a different letter within a row are significantly different at the 5% level. Values in parenthesis represent the standard deviation.

Table 6. Mean Leaf Ascorbic Acid Levels of Five Trees Located at the Three Study Sites

Tree Type	Ascorbic Acid mg/g dry weight*			% Difference	
	Site 1	*Site 2*	*Ref.*	*1 to R*	*2 to R*
A. heterophyllus	1.83 a (0.36)	1.64 a (0.64)	1.51 a (0.10)	+21	+ 9
C. caimito	1.37 b (0.29)	2.30 a (1.25)	1.68 a, b (0.61)	-18	+37
M. indica	3.62 c (0.89)	4.85 b (1.46)	6.54 a (1.19)	-45	-26
S. mahogani	2.87 a (0.56)	2.16 b (0.24)	1.59 c (0.45)	+81	+36
T. guajava	1.92 a (0.65)	1.71 a (0.37)	1.85 a (0.75)	+4	-8

* These values represent the means of nine measurements (three trials for each of the three sampling dates). Means followed by a different letter within a row are significantly different at the 5% level. Values in parenthesis represent the standard deviation).

Relative Water Content **(RWC).** The relative water content data (Table 5) showed an almost uniform relationship among the study sites. Except for mahogany, all the other four trees growing in the reference (Site 3) contained the highest RWC followed by those in Site 2. This implies that pollutants induce increased permeability in cells causing loss of water [11].

On the whole, the DMRT result on the RWC data showed that the reference (Site 3) is significantly different from the two pollution sites but that there is no significant difference between Sites 1 and 2. The following grouping of trees were found to be significantly different: *M. indica* (Group I); *A. heterophyllus, C. caimito*, and *T. guajava* (Group II), *A. heterophyllus, S. mahogani,* and *T. guajava* (Group III). Among the different trees studied, *S. mahogani* is the only one that did not show a marked difference in RWC levels. It is possible that the response of *S. mahogani* toward pollution stress is different from other species, as shown also in total chlorophyll level. Among the different trees studied, the largest change in RWC was observed in *C. caimito* and *A. heterophyllus* (10% lower in Site 1 versus reference). Comparing Site 2 to control, all trees showed lower RWC in the former. The widest differences were found in *C. caimito* and *S. mahogani*, where the Site 2 level measured 8% and 7% lower than the reference, respectively. *A. heterophyllus* and *T. guajava* showed very slight depression (2%) of RWC levels.

With the exception of *S. mahogani* the observed trend in RWC for each species is the following: Site 1< Site 2< reference. ANOVA showed that there is significant difference between Site 1 and the other two sites, which is consistent with the result obtained based on the criteria pollutant (or standard monitoring) and the leaf pH data, but no significant variation was seen between Site 2 and reference site. In terms of RWC, *C. caimito* is the best bioindicator among the five trees studied, in that it showed the biggest reductions of RWC levels in the two pollution sites.

Ascorbic Acid **(AA).** The amount of ascorbic acid in the leaf extract, averaged across the three sampling dates, for *A. heterophyllus, C. caimito, M. indica, S. mahogani*, and *T. guajava* varied with increasing pollution load of the environment. The percent change of mean leaf ascorbic acid content compared levels of those exposed to more pollution loading to those in the reference site (Table 6).

Using Duncan's Multiple Range Test (DMRT), the *M. indica* tree was found to be significantly different from the other four trees. Its leaf ascorbic acid content showed a marked reduction (-45% and -26%) when comparing polluted sites (Sites 1 and 2) to the reference (Site 3). The reducing capability of AA protects the chloroplasts in plant leaves from being oxidized by SO_2, O_3 and other oxidants. The accumulation of oxidation products, like peroxides, is prevented, thus protecting the enzymes of the CO_2 fixation cycle and chlorophyll from inactivation. It is expected to play a significant role in determining the SO_2 sensitivity of plants.

Comparison of AA levels between Sites 1 and 3 indicate that only two trees (*M. indica* and *C. caimito*) show the expected trend. *M. indica* registered the largest AA level depression (45%) in Site 1. In the other two trees (*A. heterophyllus* and *T. guajava*), the AA level in Site 1 is slightly higher while in *S. mahogani*, the AA level had increased by 81% in Site 1.

In comparing Site 2 to the reference, large differences in AA levels were expected as the former registered higher SO_2 and NO_2 levels. However, only *M. indica* and *T. guajava* showed 26% and 8% reduction from reference levels. The other three trees, showed higher AA levels in Site 2 than in Site 3.

Overall, the AA data showed no general trend in variation with pollution loading. This is contrary to previous results [12] showing AA as one of the best bioindicators of urban pollution loading. It is possible that the combination of air pollutants in Iligan City trigger opposing effects on AA levels. Another possibility is that the trees studied here have very different stress coping mechanisms involving AA. Among the five trees, it is only *M. indica* that showed the expected response in the three study sites. Still other possible causes are the fast degradation of ascorbic acid in the fresh leaf samples and very low AA levels that require a more sensitive method of determination.

Standard monitoring of air pollution using SO_2, NO_2, SPM, and Pb levels (Table 1) on one hand, and phytomonitoring on the other, showed similar results: that Site 1 is grossly polluted compared to Site 2 and reference. This is reflected in the increase of leaf pH and reduction of relative water content and total chlorophyll of the leaf extracts in the pollution sites. These trends were observed in all trees tested except for *S. mahogani*. The slightly higher pollution loading in Site 2 versus reference is more sensitively measured by the total chlorophyll level than RWC, leaf pH and standard air monitoring method.

It is to be noted from the foregoing discussion that a single tree (*e.g., M. indica)* could manifest maximum sensitivity to pollution in terms of leaf pH but not in TC. One reason is that different plant species differ in their responses to stress [17]. The sensitivity of a plant is dependent on the rate of pollutant absorption, and this will vary at the genus, species, or even cultivar level. It can also be affected by the soil type, which was not considered in this study. For example, plants growing on sulfur-deficient soils may be stimulated by atmospheric SO_2 instead and those growing on sulfur-rich soils may exhibit very strong sensitivity [3]. Also, the responses of plants to one stress (*i.e.,* cement dust) can alter their capacity to respond to other stresses (*i.e.,* SO_2, etc). *S. mahogani*, for example, showed variable responses to pollution Sites 1 and 2. Apparently, this is the result of multiple interacting stresses, considering that the pollution profile of the pollution sites are dissimilar.

It is clear however, that in the majority of plant parameters tested, *C. caimito* leaves were consistently one of the best tree, especially in total chlorophyll level. Hence, among the five trees tested, it is the best bioindicator, possibly due to its leaf structure. *C. caimito* leaves have hair-like structures on its surfaces, that may enhance the absorption of air pollutants.

CONCLUSION

Determination of air pollution levels by standard methods showed that the area near the cement factories (Site 1, Kiwalan) has SPM levels dramatically exceeding the DENR standard of good quality ambient air. However, the levels of other pollutants, SO_2, NO_x, and Pb are lower compared to those in Site 2. Among the three study sites, ANOVA results showed that Site 1 is statistically different from Site 2 and reference site at the 5% level, but no significant variation exists between the latter two sites.

The results using phytomonitoring showed good correlation with the data gathered by standard methods of air pollution monitoring. The tree leaves collected from Site 1 has significantly higher leaf pH than those from the reference site. The chlorophyl level and relative water content of leaves in Sites 1 and 2 were lower than those from the reference site. The ascorbic acid level however, failed to show a consistent response to different pollution levels.

Among the four phytomonitoring parameters, it is the leaf total chlorophyll level that was reduced significantly in the two pollution sites compared to the reference site. It is to be noted that the standard air pollution data did not show a significant variation between Site 2 and reference site, although it was evident that the pollution loading in Site 2 was relatively higher than in the reference site. The plant parameters, leaf pH data and RWC data, registered consistent results but also failed to show significant difference in the two areas. It is only the leaf total chlorophyll data that had successfully shown this difference.

Hence leaf total chlorophyll level can be used as a bioindicator for air pollution in Iligan City. Statistical analysis of the TC levels of five trees revealed this result despite known variation in sensitivity by plant species. Among the five trees, *C. caimito* was found the most sensitive, with *S. mahogani* as the least sensitive. In between the two extremes are moderately sensitive trees, namely: *A. heterophyllus, M. indica,* and *T. guajava.*

There are a number of plant species that are already used to biomonitor air pollutants. Scots Pine for SO_2 [22] and Bell-W3 variety of tobacco for ozone [23] are just two examples. The present study indicates that starapple leaves are sufficiently sensitive to manifest the relative pollution (mixture of air pollutants) levels in Iligan City. However, before this can be used as a substitute for standard monitoring, standardization of the method has to be achieved. The possible effect of soil and seasonal variations to the leaf parameter levels are now being studied.

ACKNOWLEDGEMENT

Funding for this project from the Philippine Center for Health Research and Development, Department of Science and Technology (PCHRD-DOST) is gratefully acknowledged.

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