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Efficacy of Tree Leaves as Bioindicator to Assess Air Pollution Based on Using Composite Proxy Measure

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ABSTRACT

Air pollution has become a major issue in cities due to urbanization, population growth, industrial development, and increasing number of vehicles. The study used *Gmelina arborea* tree leaves as a bioindicator to determine the Air Pollution Tolerance Index (APTI) as a simple and effective compositional index of environmental health in three cities in the Caraga Region, Philippines. To calculate the APTI, four biochemical parameters of tree leaves were calculated: relative water content, total chlorophyll content, leaf-extract pH, and ascorbic acid content. In terms of the APTI category, results showed that all *G. arborea* species collected in all sample sites are classified as sensitive to air pollution, with the sample collected in Bayugan City being the most sensitive, with an APTI value of 7.66, and the samples collected in Butuan and Cabadbaran City being the least sensitive, with APTI values of 9.54 and 9.11, respectively. A Kruskal-Wallis test revealed a significant difference between the APTI values of *G. arborea* trees in the three sampling areas in the Caraga region. Based on the APTI computed values of the tree leaves determined in all sites, it is concluded that *G. arborea* species can be used as a bioindicator of air pollution, classified as sensitive.

INTRODUCTION

Urbanization, industrialization, population development, and an increase in the number of cars are all contributing to an ever-increasing problem of air pollution in cities. Plants have the potential to significantly reduce air pollution in metropolitan settings (Irshad et al. 2020, Leghari et al. 2019). Air pollution is one of the most serious issues affecting people's health. The problem is especially significant in the Philippines' urban areas. Air pollution represents a major concern that has far-reaching implications for the health and environment of a region (Alpy & Sanjay 2016). Various physical and chemical technologies have been developed and deployed in the past to help improve air quality. Still, the high costs of establishing and maintaining these systems remain a worry (Xie et al. 2017). Biological solutions such as cultivating green plants in and around Metro Manila's urban areas are a viable alternative to employing physical and chemical ways to mitigate air pollution (Su et al. 2018).

Plants have been a tried-and-true solution to various air-related issues (Swami & Chouhan 2015, Achakzai et al. 2017). The content, concentration, and interaction of primary and secondary pollutants in the atmosphere change as the population, urbanization, motorization, and industrialization increase (Sahu et al. 2020). The leaves of plants are the major receptors of air pollution. Because the leaves have a vast surface area for absorption and accumulation, they serve as a sink for pollutants (Liu et al. 2008, Kim et al. 2015). Pollution's impacts are most visible on leaves, which have a direct negative impact on them (Lohe et al. 2015). Because roadside plants' leaves come into close contact with pollutants, they may act as pollutants' stressors (Ogboru et al. 2021). Conveniently, plant leaves have been recommended for testing to determine their ability to absorb and/or adsorb pollutants (Escobedo et al. 2008). Tolerant plants are so effective at absorbing toxins that they can create pockets of clean air (Brilli et al. 2018). Thus, such tolerant trees can help to improve air quality by exchanging gasses and acting as a sink for air pollutants, decreasing pollutant concentrations in the air and contributing to air pollution mitigation. Monitoring for injurious levels of air pollutants by plants is a standard technique to diagnose air pollution injury in plants (Ram et al. 2015). Recently, there has been a greater emphasis on using plants to detect air quality.

Biomonitoring is increasingly used as a cost-effective alternative to instrumental methods for studying local air pollution in the terrestrial environment (Yousaf et al., 2020, Nakazato et al. 2018). Lichens and mosses have already been used as biomonitors. However, dissimilar bioaccumulation dynamics were observed due to differences in morphology, ecophysiology, and habitat (Drava et al. 2019, Al-Khashman et al. 2011, Simon et al. 2014, Gonze & Sy 2016, Margitai et al. 2017). Due to their continual exposure and low-cost sampling, trees provide an alternative means of monitoring urban air quality (Lei et al. 2018, Selmi et al. 2016).

The Air Pollution Tolerance Index can be used to determine how plants react to pollution. APTI is the bio-indicator species' biological monitoring and assessment index. It's a method for determining how plants respond biochemically and physically (Gulliermo & Mallapre 2016.). Plants that are sensitive to pollution help to detect it, whereas those that are tolerant help to reduce pollution by acting as sinks in polluted areas (Lakshmi et al. 2009). The air pollution tolerance index (APTI) is a plant's innate ability to withstand air pollution stress, a major concern in industrial and non-industrial locations (Rai et al. 2013). The suitability of tree species as bioindicators/biomonitors are decided by their tolerance and sensitivity to air pollution, which is frequently tested using the air pollution tolerance index (Ogunkunle et al. 2014). Plants in urban locations are continually exposed to pollutants, resulting in pollutant buildup and integration into their systems, affecting the leaf's character, tolerance, and sensitivity. This sensitivity is assessed using a variety of biochemical measures, followed by an APTI (Tak & Kakde 2017). Among all the trees investigated along Metro Manila's key roadside corridors, *Gmelina arborea* Roxb. had the highest APTI value (Glenn et al. 2018). The study was conducted to determine the Air Pollution Tolerance Indicator (APTI) as a simple and effective compositional index of environmental health by using *G. arborea* tree leaves as a bioindicator in three cities in Caraga Region in the Philippines.

The study aimed to assess the efficacy of biomonitoring methods using *G. arborea* tree species as a bioindicator in investigating the level of tolerance to air pollutants, especially in high-traffic areas of the three cities in Caraga Region on the susceptibility level of *G. arborea* tree to air pollutants in terms of the following biochemical parameters for air pollution tolerance index, namely: relative leaf water content, total chlorophyll content, leaf-extract pH, and ascorbic acid content.

MATERIALS AND METHODS

This chapter discussed the methods and measures undertaken to conduct the study, such as choosing the research design, selecting the locale of the study, sampling technique, data gathering procedure, instrumentation, and the statistical treatment used in data analysis.

Research Design

This study used a quantitative experimental research design with a test configuration performed in a laboratory. Before, during, and after the test experiments, comparative assessments, study guides, and related articles were considered to ensure that there is clear and cut evidence to provide adequate results that could be based on by future researchers who would follow suit in the same or related research. The collected data were analyzed using correlation measures to determine trees' air pollution tolerance index in various sampling areas.

Research Locale

The study sites were conducted in three cities in the Caraga region, particularly on the roadsides of Butuan City, Cabadbaran City, and Bayugan City. *G. arborea* tree species were found and selected on purpose by the researchers in each city, respectively, at Capitol Drive, Butuan City, Ilban Street, Cabadbaran City, and Narra Avenue, Bayugan City. These sampling areas were considered polluted due to the large volume of vehicles on each city's roadsides. Fig. 1 below presents the map of the Caraga Region, Philippines, where three sampling areas were located.

Sampling Technique Used

During the dry season in the morning, fresh leaves of *G. arborea* trees were accumulated from the conveyance congested areas of the three sampling sites, along the road of Capitol Drive, Butuan City, Ilba Street, Cabadbaran City, and Narra Avenue, Bayugan City. Plenarily grown leaves of *G. arborea* were taken from above 3 m height in each of the three sampling areas and were brought to the laboratory. The loose dust particles amassed on the leaf surface were removed with a fine brush before the fresh weight of the leaves was recorded. For the turgid weight, the leaves were marinated for 24 h in distilled water before acquiring the weight. The leaves' dry weight was taken in an oven at 105°C for 2 h. The pH of the leaf extract was measured with an EZDO PH-5011 pH meter. To determine the remaining city samples' total chlorophyll and ascorbic acid content, 1-gram *G. arborea* leaf samples were brought to the Mindanao State University-IIT Chemistry Department Laboratory.

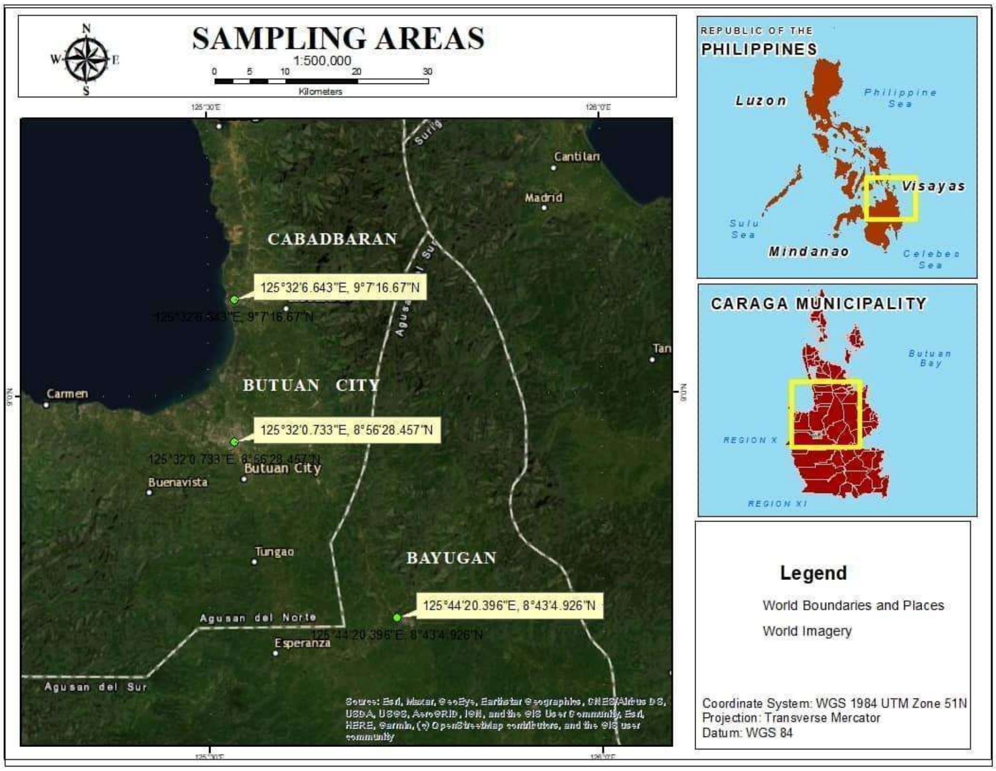

Fig. 1: Map of the study area in Caraga Region, Philippines.

Data Gathering Procedure

The researchers requested permission to conduct an Air Pollution Tolerance Index of *G. arborea* tree species in each city via email to the City Environment and Natural Resources office. Experiments for relative leaf water content and leaf-extract pH were carried out in the laboratory of Ampayon National High School. Due to a lack of equipment in Ampayon National High School and Caraga State University laboratories, the sample leaves' total chlorophyll and ascorbic acid content in three sampling areas were determined at Mindanao State University-IIT. The results of the four biochemical parameters were then used to obtain the Air Pollution Tolerance Index of *G. arborea* leaves in each sampling area. Comparison between the APTI values between each sampling site in Caraga, Philippines, were obtained using the Kruskal-Wallis statistical test.

Samples were handled as follows to obtain four parameters in the APTI (Air Pollution Tolerance Index formula:

Relative Leaf Water Content (RWC): Turner's (1981) formula for calculating RWC was used. Fresh leaves were collected, weighed using a triple beam balance, and recorded. After that, the leaves were soaked in distilled water overnight and weighed to determine the turgid weight (Dash & Dash 2017). The leaves were then dried for 2 h at 105°C in an oven before being reweighed to get the dry weight. The relative water content of leaves was calculated using the formula:

Relative water content (%) = $\{(F-D)/(T-D)\} \times 100$

F = Fresh weight of leaves (g)
D = Dry weight of leaves (g)
T = Turgid weight of leaves (g)

Total Chlorophyll Content (TCC): The sample leaves of each sampling area were brought to the laboratory of the Chemistry Department at Mindanao State University-IIT to determine the total chlorophyll content of *G. arborea* leaves. The chemist used the DMSO method (Ter et al. 2020). The extraction process was carried out using the amphiphilic DMSO solvent. Pepper tissue was sliced into smaller pieces and placed in test tubes with 10 mL of solvent. For an hour, test tubes were incubated in a water bath at 60-65°C. Based on preliminary research (Caabay 2020, Banerjee et al. 2018), this period was deemed adequate for complete tissue decolorization. Cooling at room temperature for 30 minutes was followed by filtration and absorbance measurements at 665 and 648 nm. DMSO was used for the blank determination. The absorption was measured using a HITACHI Spectrophotometer U-2000.

Chlorophyll concentrations were estimated using the methods below and represented as mg/g fresh weight (Barnes et al. 1992).

Chlorophyll a ($mg.g^{-1}$ F.W) = ($14.85\ A_{665} - 5.14\ A_{648}$)

Chlorophyll b ($mg.g^{-1}$ F.W) = ($25.48\ A_{665} - 7.36\ A_{648}$)

Total chlorophyll ($mg.g^{-1}$ F.W) = ($7.49\ A_{665} + 20.34\ A_{648}$)

where: A_{665} = absorption value at 665 nm, A_{648}= absorption value at 648 nm.

Leaf Extract pH: The pH of the leaf extract was calculated using the Datta and Sinha (2018) method. A 5 g of fresh leaves were rinsed in distilled water before being crushed and homogenized in a mortar and pestle with 25 mL of distilled water. A pH meter was used to determine the pH of the leaf extract filtrate (Pandey et al. 2016).

Ascorbic Acid Content (AAC): The concentration of ascorbic acid or Vitamin C content ($mg.100\ g^{-1}$) in the leaf of each sample leaf was determined through a laboratory test at the Chemistry Department at Mindanao State University-IIT. The chemist Singh & Chauhan (2014) spectrophotometric approach to determine ascorbic acid concentration. Fresh leaf samples (1g) were placed in a test tube, followed by 4 mL oxalic acid-EDTA, 1 mL orthophosphoric acid, 1 mL sulfuric acid, 2 mL ammonium molybdate solution, and 2 mL distilled water. After allowing the solution to rest for about 15 minutes, the clear solution from the tube was tested using a Spectrophotometer for absorbance at 760 nm (Model No. 31). The concentration of ascorbic acid in the leaf was determined using a standard graph that included absorbance and ascorbic acid concentrations and was created using the same method.

Air Pollution Tolerance Index (APTI)

Plant APTI values were obtained using the Datta and Sinha (1995) equation.

$$APTI = A\ (T + P) + R/10$$

Where A =Ascorbic Acid Content of the leaf ($mg.g^{-1}$)

P = pH of leaf extract

R = Relative Water Content of leaf (%)

T = Total Chlorophyll level of leaf extract (mg/g)

The APTI category described by Lakshmi et al. (2009) is shown in Table 1. The APTI values were calculated using each sampling area's four biochemical parameters. The plant species' Air Pollution Tolerance Index (APTI) was then compared to the standard values in the table below to determine how the *G. arborea* tree species responded to air pollution.

Table 1: APTI category described by Lakshmi et al. (2009).

Species	APTI computed values
Tolerant species	30-100
Intermediate tolerant species	17-20
Sensitive species	1-16
Very sensitive species	< 1

Statistical Treatment of Data

With a significance level of $p = 0.05$, the Kruskal-Wallis test was used to determine whether there are significant differences in the APTI values of *G. arborea* trees collected within all three study locations in the Caraga Region, Philippines.

RESULTS AND DISCUSSION

This section discusses the susceptibility level of *G. arborea* tree to air pollutants in terms of the following biochemical parameters for the air pollution tolerance index

Relative Water Content

The study of Gonzalez et al. (2001) defined relative water content as the amount of water in a leaf at the time of sampling in relation to the maximum amount of water a leaf can hold. They also stated that it is an important parameter in air pollution tolerance index relation studies (Jitin 2014) that have evaluated the osmotic potential at full turgor within the leaf to determine the plant's current health. When pollution and transpiration rates stress plants are high, this characteristic also refers to the water present in the plant, which helps it maintain its physiological equilibrium, and plants with higher water content are more drought resistant (Rai et al. 2013). Gholami et al. (2016) noted that in the occurrence of contaminated air in the area, the transpiration rates of plants are typically high, which can contribute to desiccation. Pigment analysis in *G. arborea* can be used as a physiological indicator of plant responses to water scarcity because they provide information about the stress event and because there is an inverse relationship between chlorophyll and carotenoid levels as water stress increases (Rojas et al. 2012).

Table 2 shows the relative water content (RWC) in each sampling area, which includes the fresh, dry, and turgid weights of the leaf samples based on related studies that also conduct tolerance indices with plants (Kaur & Nagpal 2017, Roy et al. 2020, Wolf et al. 2022). As cited by Roy et al. (2020), the location of a tree species' growth could significantly affect the relative weight of its leaves, with a nearby canopy covering inhibiting a greater loss of water

Table 2: Relative water content in *G. arborea* leaf samples.

Sampling areas	Fresh Weight [g]	Dry Weight [g]	Turgid Weight [g]	FW-DW	TW-DW	RWC [%]
Butuan	8.63	1.5	9	7.13	7.50	95.07
Cabadbaran	7.30	1.4	7.9	5.90	6.50	90.77
Bayugan	5.05	1	6.3	4.05	5.30	76.42

content within the leaves. This corresponds with the fact that the Butuan City sample has the highest fresh weight of the three sampling sites, at 8.63g, dwarfing the samples collected from Cabadbaran City (7.30 g) and Bayugan City (5.05 g), because it is located near a building that has the potential to shade it from sunlight at certain times of the day, rather than being within the sidewalk of the main road with little to no cover from the sun. Navarrete et al. 2017 found that the relative water content and subsequent water absorption rate rise as pollution levels rise. They explained why the samples gathered in Bayugan City gained the most weight (1.5 g) after 24 h of soaking in distilled water, then the samples gathered from Cabadbaran (0.6 g) and Butuan (0.37 g), due to its location that is found near a garbage bin. After drying in an oven for 2 h at 105°C, Butuan lost approximately 8.5 g with a dry weight of 1g, whereas Bayugan lost only 5.3g with a dry weight of 1g, and Cabadbaran lost 6.5 g with a dry weight of 1.4 g. For the RWC, the *Gmelina arborea* leaf sample in Butuan had the highest average relative water content (95.07%), Cabadbaran (90.77%), while the area around Bayugan had the lowest (76.42%). RWC in the *G. arborea* tree leaf sample is lower in the Bayugan area. This could be due to some factors, such as the condition of the sample species, which is located near burning garbage, and the tree's age (Shafiq & Iqbal 2012).

The findings of this test complement the study done by Mahecha et al. (2013), where they stated that a plant's high-water content could help it maintain its physiological balance under stress conditions such as air pollution, where transpiration rates are often high. According to Dash and Dash (2017), transpiration rates are relatively high, and a greater volume of water is required to maintain physiological equilibrium in a plant body against contaminants. They also stated that meteorological conditions, humidity, and the availability of moisture content in soil influence relative water content. A decrease in RWC reduces stomatal conductance, and therefore, according to Amulya (2015), CO_2 assimilation Net CO_2 exchange, CO_2 assimilation, and photosynthetic potential all reach zero at very low RWC (around 40%), indicating that the plant species are sensitive to a sudden change in air quality within its area. Plant transpiration rates may increase under stressful conditions, such as exposure to air pollution. Plants with a higher RWC are more resistant to air pollution.

Total Chlorophyll Content

Giri et al. (2013) explained that the amount of chlorophyll in the leaf determines the spectral variation in visible bands during the photosynthetic process. Chauhan (2010) extrapolated its significance because a decrease in the leaf chlorophyll content reduces the amount of solar radiation that can be absorbed, limiting the efficiency of corresponding photosynthetic processes and thus lowering primary photosynthetic production. As a result, an accurate estimation of the plant leaf chlorophyll content is a critical foundation in assessing the plant's tolerance to air pollution.

In accordance with Rai (2019), Roy et al. (2020), and Kousar (2014), stated that high amounts of air pollutants in the urban environment could degrade chlorophyll molecules into pheophytin by replacing Mg++ ions with two hydrogen atoms and increasing the activity of the chlorophyllase enzyme. These findings provide the reason why Table 3 shows that the Cabadbaran City sample has the highest chlorophyll a and b content. Still, the Butuan City sample got the highest total chlorophyll content. Similarly, Notman et al. 2006 explained that a tendency of low chlorophyll content in urban roadside plant leaves could result from constant exposure to the waste, which can be observed in the Bayugan City sample, which has the lowest levels of chlorophyll a, b, and total chlorophyll. These findings reveal a significantly low level of chlorophyll content in the leaf sample collected from Bayugan City compared to those

Table 3: Total chlorophyll content of *G. arborea* leaf samples.

Sampling areas	Chlorophyll content (mg.g^{-1} of fresh leaves)		
	Chlorophyll a	Chlorophyll b	Total Chlorophyll
Butuan City	0.637 ± 0.003	1.15 ± 0.01	1.29 ± 0.01
Cabadbaran City	0.643 ± 0.003	1.29 ± 0.01	1.16 ± 0.01
Bayugan City	0.408 ± 0.001	0.740 ± 0.002	0.863 ± 0.001

collected within Butuan City and Cabadbaran City. The result could be due to the exposure of the Bayugan *G. arborea* tree to the pollutants within its vicinity. This is supported by related studies, which stated that higher chlorophyll content plants are more pollution-resistant (Salimi & Aghdash 2019). Moreover, the decline in chlorophyll concentration appears to be linked to rising pollution levels (Gharge & Menon 2012).

However, all tree samples in all sites are not within the range with standard controlled *G. arborea* tree species found in other research (Dash & Dash 2017) but are higher than those found within heavily polluted sites (Glenn et al. 2018). According to one study, drought stressors significantly negatively influenced physiological parameters and total chlorophyll content, significantly reducing selected growth features. In contrast, proline content was elevated (Sachan et al. 2019, Nikolaeva et al. 2010).

Leaf-Extract pH

As illustrated in Fig. 3 below, there is a substantial difference in pH levels of *G. arborea* leaf samples across the three sites, with the leaf sample from Butuan City having the lowest pH level of 5.95, indicating acidity, and the leaf sample from Bayugan having the highest pH level of 6.51, indicating close to neutral. The acidic leaf extract pH of 6.80 indicates that the leaf extract is more susceptible to air contaminants (Kuddus et al. 2011). High pH has been shown to increase plant tolerance to pollution (Akande et al. 2021).

Stomatal sensitivity is affected by pH, and leaves with a low pH are more susceptible to pollution, while those with a neutral pH are more tolerant (Krishnaveni 2013). According to Uka and Chukwuka (2014), the pH of the leaf filtrates in the sites indicates the presence of gaseous air pollutants, specifically SO_2 and NO_2. However, Ter et al. (2020) found that in all plant species, the pH value of leaf extract in contaminated sites was lower than in control sites. Additionally, the researchers observed that the high pH of leaf extract at the control site and the low pH at the polluted site could be due to the high pollution levels at the polluted sites.

The graph also illustrates that even if the leaf sample from the Butuan site is more acidic than the Cabadbaran and Bayugan sites, the test showed that the Butuan site has the highest total chlorophyll content ascorbic acid content out of the 3. Thus, it is expected that compared to the other species studied (Glenn 2018), the total chlorophyll content and ascorbic acid content should be relatively lower.

Ascorbic Acid Content

Ascorbic acid is a significant redox buffer that also serves as a cofactor for enzymes that regulate photosynthesis, hormone production, and the regeneration of other antioxidants (Gallie 2012). In this study, Fig. 4 displayed a significant difference between the ascorbic acid levels of *G. arborea* leaf samples in Butuan and Cabadbaran from the sample collected in Bayugan. It shows a 0.0179mg/g difference between the ascorbic acid content from Butuan and Bayugan. Cabadbaran has a closer ascorbic acid to Butuan, which has only a 0.0007mg/g difference. The study found that ascorbic acid levels in plant leaves fell considerably in polluted areas with visible rubbish near the Bayugan sample site compared to

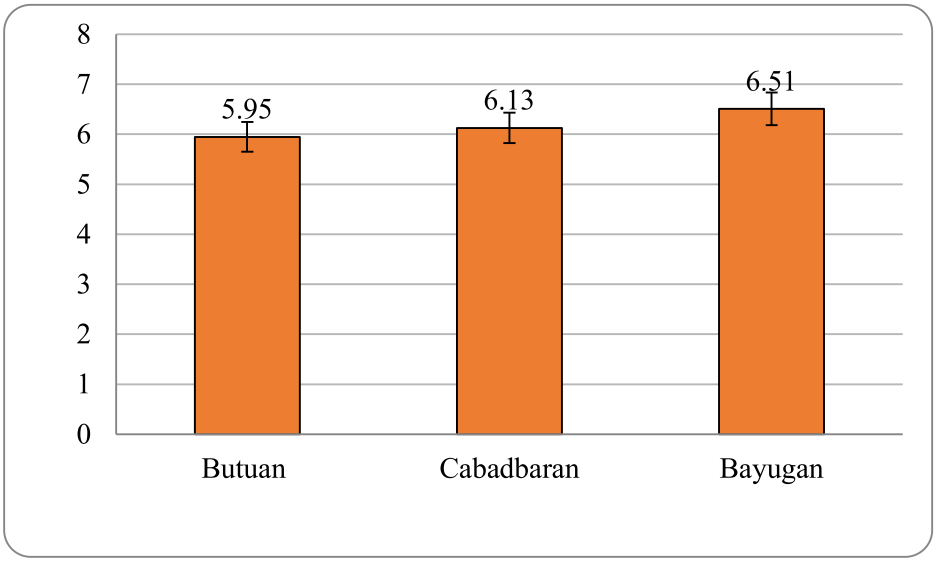

Fig. 2: The relative pH level graph of *G. arborea* leaf samples.

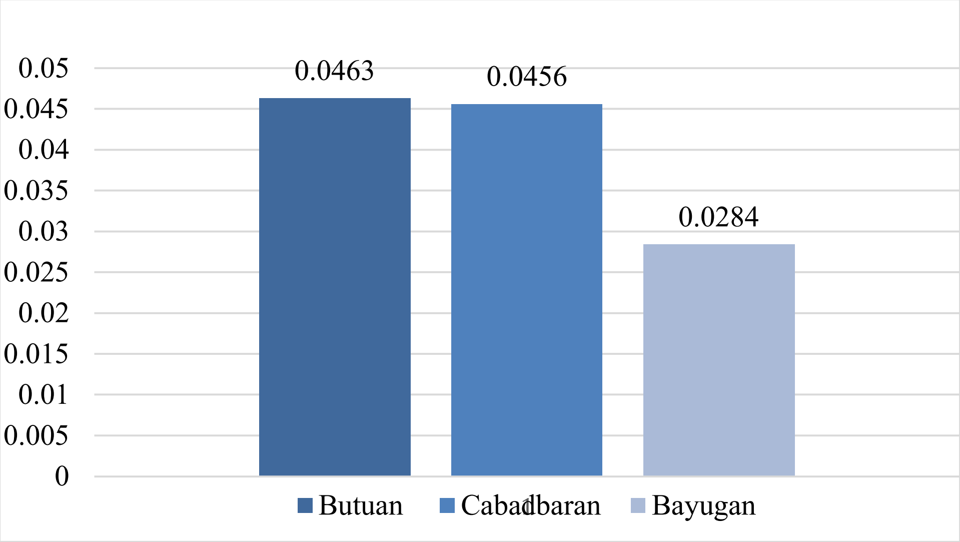

Fig. 3. Ascorbic Acid Content of *G. arborea* leaf samples.

Cabadbaran and Butuan sample sites. Consistently, the study of Falusi et al. (2015) reported that pollution of dumpsite habitats is linked to the release of various air pollutants, including acidic gases like SO_2, NO_x, and H_2S. As a result, the drop in ascorbic acid concentration found in *C. odorata* and *M. lucida* gathered from the four dumpsites compared to the control site could be related to acidic gas deposition on the surface of their leaves.

The researchers also discovered that the overall tree health in Cabadbaran and Butuan was better than in Bayugan, although the sample in Butuan is more exposed to vehicular emissions. This discovery is consistent with Conklin's (2001) and Aguiar-Silva et al. (2016) findings that increased ascorbic acid concentration in plants implies resilience to air pollution. *G. arborea* species in the experimental site had the highest ascorbic acid levels (Ogboru et al. 2021). Because its reducing power is related to its concentration, plants that retain a high ascorbic acid content even under polluted environments are thought to be tolerant of air pollutants (Ogunrotimi et al. 2017). Various biochemical parameters acted differently in the tested plant species, but the ascorbic acid level was discovered to be the most important element in providing plants with air pollution resistance (Bharti et al. 2017).

According to Rahul & Jain (2014), the air pollution tolerance index (APTI) describes plants' inherent ability to tolerate air pollution. It is among the most important factors when choosing traffic barrier plant species. Plant APTI has been described using four biochemical parameters: total chlorophyll, relative water content (RWC), ascorbic acid, and leaf extract pH (Nadgórska–Socha et al. 2017). Pollution-induced changes in a single parameter may not provide a complete picture. As a result, four biochemical parameters are considered to obtain an empirical value representing plant APTI.

Table 4 shows the results of the Air Pollution Tolerance Index (APTI) of *G. arborea* trees identified in three cities, as well as biochemical characteristics such as relative water content (RWC), total chlorophyll content (TCC), leaf-extract pH, and ascorbic acid concentration (AA), all of which are required for multiple studies to successfully determine APTI values of any plant species. One sample revealed a significant difference when comparing the APTI values of the three Caraga Region sites. It was discovered that Bayugan samples have a lower APTI value than Butuan and Cabadbaran samples. The reason for the Bayugan City sample *G. arborea*'s low APTI value in the Bayugan City sample may have a low pH value because a higher pH value

Table 4: APTI values of *G. arborea* tree leaves in the 3 cities.

Sampling areas	RWC (%)	TCC (mg/g)	pH	AA (mg/100g)	APTI
Butuan	95.07	1.29 ± 0.01	5.95	4.63 ± 0.07	9.54
Cabadbaran	90.77	1.16 ± 0.01	6.13	4.56 ± 0.07	9.11
Bayugan	76.42	0.86 ± 0.001	6.51	2.84 ± 0.00	7.66

Table 5: The Kruskal-Wallis test between the three sites within the Caraga Region

Factor	Statistics	df	p-value*	Remark
Caraga Region Cities	6.489	2	0.039	Reject H_0

* Tested on 0.05 significant difference

increases plant tolerance to air pollution (Ninave et al. 2001). Despite the wide range of APTI values, the final assessment of labeling all of the trees is sensitive (Nadgórska–Socha et al. 2017, Gulia et al. 2015, Li et al. 2018). Guillermo and Mallarpe (2016) found that the mean APTI value of the *G. arborea* individuals studied was 11.45, putting them in the sensitive category. Furthermore, Viradia et al. (2020) reported that *G. arborea* is a sensitive species, with APTI values of 3.079 and 7.192, respectively, on their sites 1 and 2.

The findings of this study are highly consistent with those of Molnar et al. (2020) and Pandey et al. (2015), who discovered that plant species with lower APTI values are particularly valuable as bioindicators of air pollution and proxies for urban health. Furthermore, APTI determination is a reliable method for screening many plants for their response to air contaminants (Deepalakshmi et al. 2013). However, *G. arborea* species were classified as tolerant in the study of Carillo and Ocampo (2016). Ogunrotimi & Adereti (2017) conducted an earlier study in which *G. arborea* had APTI values, indicating it was an intermediately tolerant species within their respective areas for sample collection. This may lead to the initial assumption that G. arborea species found within the sites are tolerant as the samples tested in previous research (Wang et al. 2011, Ugulu et al. 2015, Liu et al. 2012). As a result, APTI values can be used to select pollution-tolerant plants for urban greening or green belt development (Mondal et al. 2011).

Table 5 shows the Kruskal-Wallis Test in JASP v.0.16.2.0 which is used to answer the hypotheses if significant differences exist between the Air Pollution Tolerance Indices (APTI) of each study site within the Caraga region, which is tested on 0.05 significant difference. The results indicate a significant difference between the APTI values of trees within the 3 cities in the Caraga Region (*p*=0.039). This indicates at least 1 significant difference within all study sites in Caraga Region. It is supported in the study by Dash and Dash and Dash (2017) that low APTI levels were susceptible to air contaminants in general and vice versa. Sensitive plants can serve as indicators, while tolerant plants can serve as a barrier for pollutants in the air (Gholami et al. 2018). Tolerant species are bio accumulators, can be efficient roadside vegetation, and can help with the absorption and screening of air pollution (Kunwar et al. 2016). According to Berame et al. (2021), significant progress has been made due to biomonitoring efforts relating to industrial, agricultural, and domestic pollution affecting ecosystems in the Philippines. Therefore, plants with high APTI values have the potential to grow under air pollution conditions, while low APTI valued (sensitive) plants can be utilized to detect the current state of pollution indices are recommended to satisfy the concept of the green-belt development program (Dash & Dash 2017).

CONCLUSION

Based on the APTI computed values of the tree leaves determined in all sites, it is concluded that *G. arborea* species can be used as a bioindicator of air pollution classified as sensitive. This could be attributed to the high concentrations of air contaminants at each study site. The large disparity in APTI values obtained across all three sites is attributable to the significant difference in APTI values measured between Bayugan City and Butuan City. Even though the study indicates that all samples are sensitive to air pollution, there is still a disparity between the samples. Thus, rejecting the null hypothesis means that there is a significant difference in the APTI values of the *G. arborea* tree leaves within the three cities in the Caraga Region. Further studies about *G. arborea* as a bioindicator in other areas in the Philippines should be done since it is a sensitive species suitable for identifying the air pollution tolerance of trees. Multiple tree species found in polluted-ridden areas, not specifically about air pollution but also land and water pollution, must be gathered throughout all study sites to ensure a more robust outcome. It is suggested to use various tree species that are tolerant to air pollution as they serve as bioaccumulators of air pollution. Thus, they could be planted along national highways as part of the greening development in the Philippines. Time intervals between collecting samples should be done in comparison to the pollution levels throughout the day.

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