

Environmental Pollution Bioindication Based on *Ficus benjamina* L. Leaf Reflectance in the City of Abidjan, Côte d'Ivoire

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Abstract

The air in African cities is increasingly polluted mainly due to human activities. A bioindication technical of urban air quality based on active remote sensing might be an alternative to existing physico-chemical methods. Reflectance measurements in the visible spectrum have been carried out at the adaxial and abaxial sides of *Ficus benjamina* L. leaves in the city of Abidjan, Côte d'Ivoire, with a precision digital camera. Leaves were collected in industrial zones and in parks. The impact of air pollution on leaf physiological as well as structural characteristics in these two contrasts urban environments was determined by Dorsiventral Leaf Reflectance Correlation (DLRC) and dorsiventral leaf reflectance asymmetry quantitatively defined with Normalized Dorsiventral Asymmetry Index (NDAI). Species leaf susceptibility to air pollution from season to season was determined by NDAI seasonal variation. Leaf reflectance measurements allowed the estimation of environmental stress level among industrial areas and parks. NDAI and DLRC were significantly higher in industrial zones compared to parks. NDAI values were found significantly higher for major rainy season compared to major dry season, indicating probably that *F. benjamina* leaf structure changes were increased from one season to another. Thereby, assessment of urban air quality can be done using leaves reflectance in the visible spectrum.

Keywords

Bioindication, Air Pollution, Reflectance, Visible Spectrum, Côte d'Ivoire

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1. Introduction

Urbanized and industrialized areas are known to be subjected to high concentrations of air pollutants [1]. Atmospheric pollution is the main result of different components such as heavy metals, polycyclic aromatic hydrocarbons (PAHs), particulate matter ($PM_{0.1}$, $PM_{2.5}$ and PM_{10}) and organic components, which can originate from various sources.

The main sources in the urban environment are road traffic and industrial activity [2]. Air pollution is a serious threat to human health [3] such as cardiovascular effects and respiratory tract problems [4].

Current physico-chemical techniques for the determination and monitoring of air quality are expensive for developing cities [5] like Abidjan, Côte d'Ivoire, where air quality strategies are ineffective [6]. Alternatively, methods based on biomonitoring approach are cheap [7]. The plants can be used as passive bio-monitors in the urban environment to indicate the environmental quality [8] [9].

Atmospheric pollutants can induce changes in leaf structure and physiology [10]. This variation in structural and physiological leaf properties affects leaf reflectance, transmittance and absorptance [11] [12].

The use of spectral and remote sensing techniques for detection of plant stress is based on the assumption that stress factors interfere with leaf structural and physiological properties, influencing the spectral leaf characteristics that can be detected in the different regions of the electromagnetic spectrum [13] [14].

The objective of this study is the evaluation of leaf reflectance parameters in the visible spectrum as an indicator for assessing urban environment quality and the investigation of species leaf susceptibility to air pollution from one season to another.

2. Material and Methods

2.1. Study Area and Experimental Set-Up

The city of Abidjan ($5^{\circ}00'N$ - $5^{\circ}30'N$, $3^{\circ}50'W$ - $4^{\circ}10'W$), Côte d'Ivoire, with distinct land use classes and spatially varying habitat quality [6], was chosen as the study area. The study area has a tropical climate with a major rainy season (May-July), a minor rainy season (September-November), and two dry seasons in between.

Abidjan is characterized by a high level of industrialization and urbanization. The city has a significant growing old automobile park, of which 70% are secondhand vehicles. Many parks and green spaces were preserved in the city, but these parks disappear quickly due to urbanization. Actually, Abidjan is one of the most polluted cities of sub-Saharan Africa.

Samples were collected in areas with contrasting environment quality. We sampled in industrial zones (2 sites) and in parks (2 sites) which were relatively far away from traffic and industrial activity. Sampling locations were determined based on the availability of plant species tested.

2.2. Study Species

We have chosen *Ficus benjamina* L. as a study species because of its homogeneous spatial distribution in the city. *F. benjamina* is a tropical dicotyledonous belonged to the family Moraceae. It is an evergreen tree with several spreading branches from the base, with oval and glossy leaves of 2 - 5 cm wide. This tree species is hypostomatous (*i.e.*, it only have stomata on the abaxial leaf surface) with dorsiventral structure (*i.e.*, the adaxial surface is adjacent to the palisade parenchyma layer, while the abaxial surface is adjacent to the spongy mesophyll layer). Moreover, the leaves of *F. benjamina* have abaxial wax and smooth cuticle.

2.3. Sampling and Leaf Reflectance Measurements

Two sampling locations were selected at each site according to accessibility and geographical distribution criteria. At each sampling location 2 trees were selected. We sampled 9 mature and undamaged leaves from each tree at a height between 1.5 m and 2.5 m above ground level. Sampling campaign was conducted during six month on the same trees species. A first sampling campaign was conducted from February until April 2012 for major dry season. The mean air temperature during this period was $28.3^{\circ}C$, air humidity was 83%, precipitation was 43.18 mm and mean wind speed was $11.7\text{ m}\cdot\text{s}^{-1}$. A second campaign was organized from May until July 2012 for major rainy season. The mean air temperature during this period was $26^{\circ}C$, while air humidity was 89.2%. Precipitation and mean wind speed were respectively 375.91 mm and $12\text{ m}\cdot\text{s}^{-1}$. Overall, we sampled 432 leaves.

The field level leaf reflectance measurements were performed under standard illumination conditions using a field illumination set up as described by Khavanin Zadeh *et al.* [15]. A Canon EOS 550D, RGB reflex camera equipped with a zoom lens (EF-S 18 - 55 mm f/3.5-5.6 IS) was used and mounted on the field illumination setup. The inner side of the setup is coated with non-reflecting black paint, intended to avoid indirect illumination (stray light) of the target as much as possible, thus avoiding multiple scattering conditions as much as possible. The camera is locked on top of the leaf reflectance measurement setup with a camera mounting cylinder. The setup allows keeping the distance between the camera objective and the target leaf surface constant throughout all measurements. In setup, the illumination of a leaf is performed with two warm-white LED light sources (Ostar, 2005) which, during field campaigns, can be fed with the electric power of a fully charged battery. The maximum peak wavelength for the red, green and blue of LEDs is 645, 550 and 490 nm, respectively. In the device, a removable grid made of transparent thin nylon wire, is used to flatten as well as fix a leaf at the bottom of the setup. Immediately after sampling of the leaves from the sample trees, imagery was acquired from both the adaxial and abaxial sides of a sampled leaf. The measurement time for tree leaf harvesting and the imaging of both leaf sides, takes less than 5 minutes. We converted leaf imagery after transfer to a computer platform, into R (Red), G (Green) and B (Blue) 1-byte grey images. Leaf reflectance has been computed for both the upper (adaxial) and lower (abaxial) leaf surfaces. By superimposing a polygon on the leaf surface, a leaf mask is obtained. We define this polygon as the leaf region of interest (ROI) for reflectance determinations by using ENVI 4.4 (Research Systems, Boulder, Colorado, USA). Leaf reflectance (in percentage) is computed for the R, G, and B spectral bands using the leaf R, G and B digital numbers ($R_{l,\lambda}$) and the white reference target panel digital number ($R_{r,\lambda}$) according to Equation (1) [15].

$$R_{l,\lambda} = 100 \times R_{r,\lambda} \times \frac{DN_{l,\lambda}}{DN_{r,\lambda}} \quad (1)$$

$R_{l,\lambda}$ and $R_{r,\lambda}$ are, respectively, leaf and white reference target reflectance for band λ (nm), with λ indicating the R, G or B spectral band. $DN_{l,\lambda}$ and $DN_{r,\lambda}$ are leaf ROI digital number and white reference target (range: 0 - 255), respectively.

Leaf reflectance variation can indicate environmental stress difference between land use classes studied.

2.4. Dorsiventral Leaf Reflectance Correlation (DLRC) and Normalized Dorsiventral Asymmetry Index (NDAI)

DLRC expresses the relationship between the adaxial and abaxial leaf reflectance which is characterized by the slope and the coefficient of determination obtained from the linear regression analysis between abaxial and adaxial leaf reflectance.

Dorsiventral leaf reflectance asymmetry can be quantitatively defined with NDAI, which is defined as a linear combination of the leaf reflectances at both the adaxial and abaxial leaf sides [15] [16] as defined in Equation (2):

$$\text{NDAI} = \frac{R_{l,ab} - R_{l,ad}}{R_{l,ab} + R_{l,ad}} \quad (2)$$

$R_{l,ab}$ and $R_{l,ad}$ indicate abaxial (*ab*) and adaxial (*ad*) leaf reflectance for the same spectral band, respectively.

NDAI as well as DLRC can be affected both by changes in leaf structural as well as physiological characteristics caused by environment quality.

2.5. Species Leaf Susceptibility to Air Pollution from Season to Season

Species leaf susceptibility to air pollution from season to season was determined by NDAI variation from major dry season (February-April) to major rainy season (May-July) in industrial zones and parks in different visible spectral bands.

2.6. Statistical Analysis

All data were analyzed using Statistica software, version 7.1 (StatSoft Inc., 1984-2005). The normality of the

data was tested with a Shapiro-Wilk test. Descriptive statistics, including mean and standard deviation were calculated for leaf reflectance parameters. Means were compared by using a one-way analysis of variance (ANOVA) procedure and a Tukey-HSD test. Linear regression analysis was performed to identify the relationship between adaxial and abaxial leaf reflectance. Species leaf susceptibility to air pollution from season to season was assessed using the Student-*t*-test. Differences were considered significant at $p < 0.05$.

3. Results and Discussion

3.1. Leaf Reflectance

F. benjamina leaf reflectance values for both leaf sides are higher in Parks (P) in comparison with industrial zones (IZ) for all spectral bands according to **Table 1**. This tendency is confirmed by the observations of Khananin Zadeh et al. [15] for *Carpinus betulus* L. in two contrasting urban environments (suburban green and Industrial habitats) in the city of Gent, Belgium. It might be related to structural responses of the leaves to air pollution in IZ. Indeed, Dineva [17] reported in the industrial district Kremikovtzi, Bulgaria, a decreasing of *Acer platanoides* L. leaf thickness. Carter [18] demonstrated a correlation between leaf reflectance and leaf thickness for the wavelength range between 400 and 700 nm. We observed a statistically significant difference (Tukey-HSD test: $p = 0.035$) in leaf reflectance among IZ and P. This difference was pronounced at adaxial side in the blue band as shown in **Table 1**. An explanation for this one might be the higher exposure of adaxial leaf surfaces (see **Figure 1**) to urban air pollution [19] that can lead to epidermal erosion of adaxial leaf side [20], consequently

Table 1. Mean (\pm standard deviation) of leaf reflectance (%) in industrial zones and parks in the spectral bands studied. Mean values on each column in the same spectral band followed by different letters are significantly different ($p < 0.05$).

Land use class	Spectral band	Adaxial leaf reflectance	Abaxial leaf reflectance
Industrial zones	Red	25.03 \pm 5.23 ^a	36.46 \pm 4.27 ^b
	Green	38.91 \pm 5.57 ^a	49.83 \pm 4.01 ^b
	Blue	18.19 \pm 3.76 ^a	23.79 \pm 3.91 ^b
Parks	Red	29.34 \pm 2.49 ^a	37.15 \pm 2.44 ^b
	Green	42.67 \pm 1.51 ^a	51.72 \pm 2.96 ^b
	Blue	22.27 \pm 1.64 ^b	26.53 \pm 1.62 ^b



Figure 1. Leaf morphology for *Ficus benjamina* L.

decreasing leaf thickness. Arriaga *et al.* [21] used epidermal characters as bioindicators of environmental pollution and stated that width of upper epidermal cells of *F. benjamina* were higher in periurban area and lower in urban area in the metropolitan area of Buenos Aires, Argentina. On the other hand, several authors [22]-[24] report that leaf reflectance for the blue band is affected by leaf internal structure, and less so by chlorophyll content [25]. On the other hand it could be that also the waxy leaf surface properties of *F. benjamina* [26] affect abaxial leaf reflectance which might lead to an increase in specular and a decrease diffuse leaf reflectance [27] resulting in lower differences in abaxial leaf reflectance between P and IZ (see **Table 1**). Khavanin Zadeh *et al.* [16] came to a similar conclusion for the adaxial leaf side for non-hairy *Tilia* sp. (non-hairy referred to the absence of trichomes on the leaf surface) in polluted compared to less polluted urban environment.

3.2. Normalized Dorsiventral Asymmetry Index (NDAI)

Table 2 shows that NDAI is higher in industrial zones (IZ) compared to parks (P) for all spectral bands. These values are statistically significant between land use classes for the blue and red bands (Tukey-HSD test: $p < 0.05$).

For *F. benjamina*, NDAI showed a different behavior than leaf reflectance in land use classes. It significantly decreased in P and increased in IZ for the red and blue bands. Khavanin Zadeh *et al.* [16] observed NDAI significantly increased in green area compared to industrial area for hairy *Tilia* sp. (hairy referred to the presence of trichomes on the leaf surface), while the reverse was observed for *C. betulus* (with a ridged leaf surface) by Khavanin Zadeh *et al.* [15] for the same study area. These authors related the different results for the NDAI index to different morphological and physiological response of the leaves against air pollution in an urban area. They observed that specific leaf area (SLA, area of leaf divided by its dry mass) significantly increased (considering that normally higher SLA indicates lower leaf thickness) in *C. betulus* because of air pollution, while it does not change for hairy *Tilia* sp. Inversely, they observed that relative chlorophyll content significantly decreased in polluted area for hairy *Tilia* sp. while it does not change for *C. betulus*. The authors concluded that NDAI seems to be species-dependent. Taylor *et al.* [28] reported that *Tilia* sp. are relative sensitive to air pollution, while *C. betulus* identified as a relative insensitive species. This trend for *C. betulus* seems to be similar for *F. benjamina*. Indeed, according to Tra Bi *et al.* [29], *F. benjamina* was categorized as intermediate tolerant species to air pollution. The identification and categorization of plants into sensitive and tolerant groups indicates that they differently react to air pollution [30]. Considering results of abovementioned studies, we can suggest that NDAI seems to be more sensitive to leaf structural changes than to biochemical changes for *F. benjamina* induced by air pollutants.

3.3. Dorsiventral Leaf Reflectance Correlation (DLRC)

The linear regression between abaxial and adaxial leaf reflectance show strong and higher correlation ($R^2 > 0.50$, $p < 0.05$) in industrial zones (IZ) compared to parks (P) for all spectral bands, while the slopes of the linear relation are lower in P than IZ except for green band (**Figure 2**).

As illustrated in **Figure 2**, DLRC representing R squared and the slope of the linear relation was significantly highest in IZ land use class. This result is in agreement with the findings of Khavanin Zadeh *et al.* [16]. These

Table 2. Mean (\pm standard deviation) of Normalized Difference Asymmetry Index (NDAI) in industrial zones and parks in the spectral bands studied. Mean values followed by different letters are significantly different ($p < 0.05$) between industrial zones and parks in the same spectral band.

Land use class	Spectral band	NDAI
Industrial zones	Red	0.19 \pm 0.05 ^b
Parks	Red	0.12 \pm 0.04 ^a
Industrial zones	Green	0.13 \pm 0.04 ^a
Parks	Green	0.10 \pm 0.02 ^a
Industrial zones	Blue	0.14 \pm 0.04 ^b
Parks	Blue	0.09 \pm 0.02 ^a

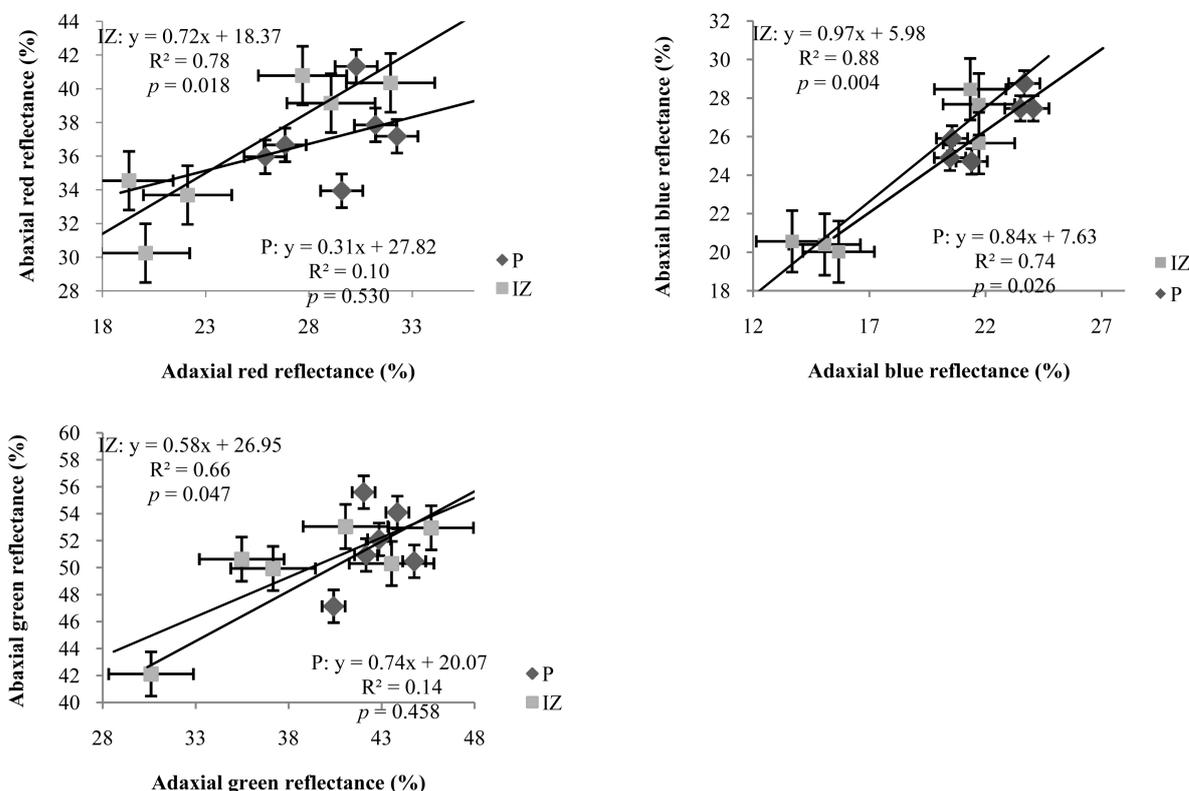


Figure 2. Dorsiventral Leaf Reflectance Correlation (DLRC) in industrial zones (IZ) and parks (P) for the spectral bands studied. Error bars represent standard errors on the individual points.

authors recorded that DRLC shows a more stable variation than NDAI in polluted sites for the species considered, and that it seems to be species-independent. DLRC increasing in blue and red bands in IZ might be a direct reflection of environmental pollution impact on the abaxial versus the adaxial leaf reflectances [15].

According to Dineva [17] and Brakke *et al.* [31], dorsiventral leaf properties in many leaves is an important characteristic in photosynthesis and gas exchange, and can be affected by air pollution. Indeed, gaseous pollutants mostly enter through the stomata [32], which for *F. benjamina* are located on the abaxial side [33]. Therefore, it seems that spongy parenchyma is more sensitive to gas exchange than palisade parenchyma, and consequently it is more susceptible to damage from gaseous pollutants [17]. This susceptibility is confirmed by an increasing abaxial reflectance leading to an increased DLRC, which might be explained by a decreasing chlorophyll content and anatomical changes in the spongy parenchyma such as decreased intercellular spaces and a thinner layer of spongy parenchyma [34]-[38]. For instance, Johnson *et al.* [39] reported that under adaxial illumination, palisade mesophyll acts to propagate light into the spongy mesophyll and that light propagation is inhibited when illuminated from the leaf side opposite the palisade layer (under abaxial illumination). Thereby a decreasing in intercellular spaces and a thinner layer of spongy parenchyma leads to a reduction in the light path length through this layer, causing the abaxial leaf reflectance to become more affected by the palisade than the spongy parenchyma layer [16], consequently increasing abaxial reflectance. Moreover, Carter and Knapp [11] demonstrated that a lower leaf chlorophyll content leads to a higher leaf reflectance, and thus an increase of DLRC.

3.4. Species Leaf Susceptibility to Air Pollution from Season to Season

Species leaf susceptibility to air pollution from season to season was determined by NDAI seasonal variation. The results show that NDAI values are higher for major rainy season (MRS) compared to major dry season (MDS) in parks (Figure 3(a)) and industrial zones (Figure 3(b)) for all spectral band. The difference were more pronounced in parks in the red band (Student-*t*-test: $t_{0.05} = -8.537$, $p = 0.001$) (Figure 3(a)).

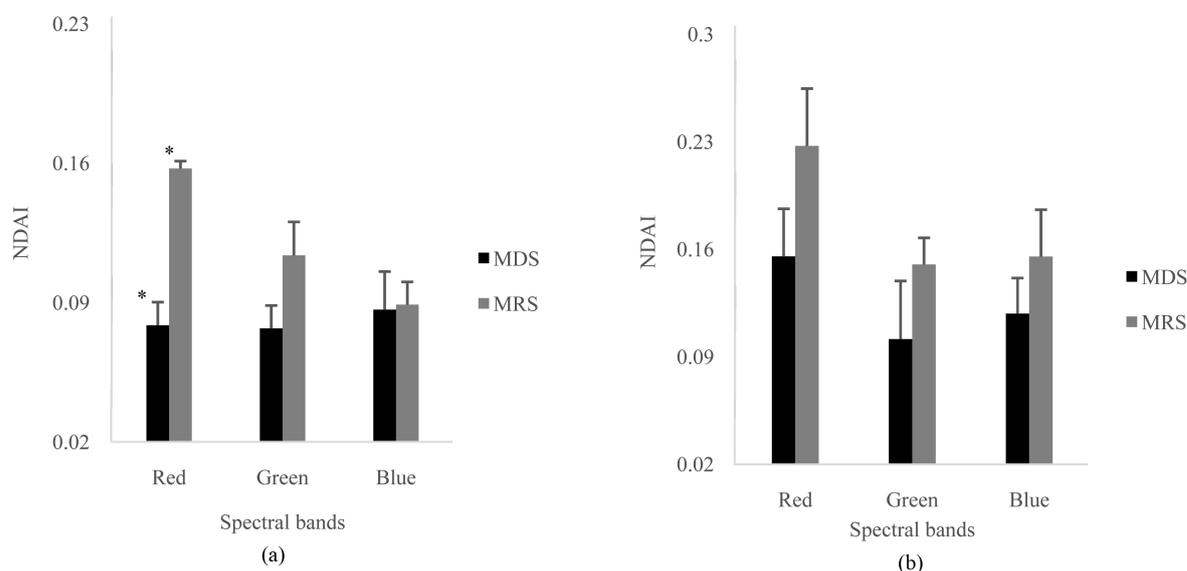


Figure 3. Seasonal variation on the air quality determined through Normalized Dorsiventral Asymmetry Index (NDAI) values in parks (a) and industrial zones (b) in the spectral bands studied. MDS: Major dry season, MRS: major rainy season, *: Significant difference, $p < 0.01$. Error bars are standard deviation.

NDAI seasonal showed same trends from one land use class to another, higher in the MRS and lower in the MDS, but significantly in parks as observed in (Figure 3(a)). These observations might be probably due to leaf surface characteristic and habit. Urvat *et al.* [40] demonstrated that rainfall cannot remove all deposited PM, as particles can be permanently trapped in the surface wax layer during wax development. This process might be important for *F. benjamina*. Moreover, this species is an evergreen species, keeping its leaves during several years, resulting in a longer time increase in leaf SIRM, as was observed by Lehndorff *et al.* [41] for *Pinus nigra*. This might increase changes in leaf structure from one season to another.

4. Conclusion

Leaf reflectance as well as NDAI and DLRC in the visible spectral bands can be good indicators to estimate differences in urban environment quality. Results revealed effects of air pollution on anatomical and physiological leaf characteristics in IZ in comparison with P. NDAI significant increasing during the major rainy season probably indicated that *F. benjamina* leaf structure changes were increased from one season to another, and was probably related to species. These results make it possible to consider an operational approach for assessing and monitoring of urban environment quality based on biomonitoring technique and an active remote sensing. However, the suitability of these results still requests some further research such as anatomical studies in tropical area.

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