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Herbivory Rate on Woody Species of the Caatinga and NDVI as Indicators of Plant Stress

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R E S U M O

Espécies de plantas distribuídas em uma paisagem são submetidas a um mosaico de condições abióticas que podem ter efeito negativo sobre o desenvolvimento (stress geração) e expô-las à predação por herbívoros. Esse estresse pode causar adicionalmente assimetria foliar e uma redução na produção primária. A taxa fotossintética, relacionada com a produtividade da planta, pode ser medida por índices espectrais, tais como o NDVI (índice de vegetação da diferença normalizada), calculado a partir de imagens de satélite. No presente trabalho, testou-se a hipótese de que ambientes com baixa produtividade primária (NDVI baixo) irá possuir maior assimetria foliar e maiores taxas de herbivoria. Os resultados mostram que na região de Caatinga semi árida de Pernambuco, Brasil, a folha de assimetria diminui com valores mais elevados de NDVI, indicando uma estreita relação entre esta medida da planta e o índice espectral. Por outro lado, a correlação entre herbivoria e produção primária ou assimetria foliar não foi significativa, sugerindo que os herbívoros vão além da simples seleção de indivíduos mais estressados.

Palavras-Chave: Assimetria flutuante, herbivoria, NDVI

Taxa de Herbivoria em Espécies Arbóreas da Caatinga e o Uso do Índice de Vegetação por Diferença Normalizada (NDVI) como Indicador de Estresse em Planta

A B S T R A C T

Plant species distributed on a landscape are submitted to a mosaic of abiotic conditions that may have a negative effect on development (generating stress) and expose them to predation by herbivores. This stress can cause additionally leaf asymmetry and a reduction on primary production. The photosynthetic rate, related to plant productivity, can be measured by spectral indexes, such as the NDVI (normalized difference vegetation index), calculated from satellite images. In the present work, we test the hypothesis that environments with low primary productivity (low NDVI) will possess larger leaf asymmetry and higher herbivory rates. Our results show that in the Caatinga semi-arid region of Pernambuco, Brazil, the leaf asymmetry reduces with higher NDVI values, indicating a close relationship between this plant measure and the spectral index. On the other side, the correlation between herbivory and primary production or leaf asymmetry was not significant, suggesting that herbivores go beyond just selecting more stressed individuals.

Keywords: Leaf asymmetry, NDVI, herbivory

1. Introduction

Several studies show that there is a high correlation between part of the incident solar radiation on plants and their physiologic state (Méndez-Barroso *et al.*, 2008; Wessels *et al.*

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2004). Normally vegetation has low reflectance on the visible red part of the spectrum, and high on the green and near infrared - as the wavelength of the former is absorbed by plants for photosynthetic activity (Weier and Herring, 1999). In this way, the fraction of the photosynthetic active radiation (PAR) captured by the photosynthesizing tissues is, among other factors, related to the productivity of a vegetated area (Hill and Donald, 2003).

The ability to translate spectral data to meaningful biological variables is a key step in the increase of use and value of information collected by satellites (Paruelo *et al*., 1997). Based on this statement, several indexes were developed involving algebraic operations on satellite image bands. There is substantial evidence that PAR is related to some of these vegetation indexes, although in several cases pixel heterogeneity, atmospheric distortions and solar angle can degrade the quality of the data (Myneni and Williams, 1994). The most used of these indexes is the Normalized Difference Vegetation Index (NDVI), which was developed and proven to be indicator of photosynthetic activity, even for heterogeneous areas (Gamon *et al*., 1995), different scales (Stefanov and Netzband, 2005) and different ecosystems (Myneni and Williams, 1994; Paruelo *et al*., 1997).

The NDVI has been used as a sensible measure for disturbed area identification (Weier and Herring, 1999), varying in relation to the aridity of the environment (Barbosa *et al*. 2006), and according to some authors (White, 1974; Rhoades, 1979), plants that suffer stress of any sort must be more susceptible to predation by herbivores than less stressed individuals, although higher productivity (an aspect of a low stress plant) can lead to higher herbivory rates (Carmo and Penedo, 2004). Stress is defined as any environmental factor that can cause potential damage to the biological system (Hoffman and Parsons, 1991).

One of the indicators of plant stress and thus vulnerability to herbivory (e.g., Moller, 1995; Wiggins, 1997; Zvereva *et al*., 1997) is the level of leaf asymmetry (LA), which represents a small random deviation from the bilateral symmetry in a morphologically symmetrical leaf (Van Valen, 1962; Palmer and Strobeck, 1986; Bjorksten *et al*., 2000).

The present work was carried out in an area of Caatinga (semi-arid scrub-forest formation, Northeast Brazil), an excellent environment to test our hypothesis, for it has natural vegetation on a landscape mosaic of abiotic factors. We have as objectives: a) evaluate herbivory on nine species of woody plants; b) verify if the NDVI can be used as a measure of plant stress. For this, we observed if in individuals of the same species NDVI, LA, and herbivory have correlations between themselves.

We hypothesize that the leaves of individuals under stress conditions at a local scale present a high LA, high herbivory, and low NDVI, indicating an environment low on productivity and high on vulnerability (Figure 1).

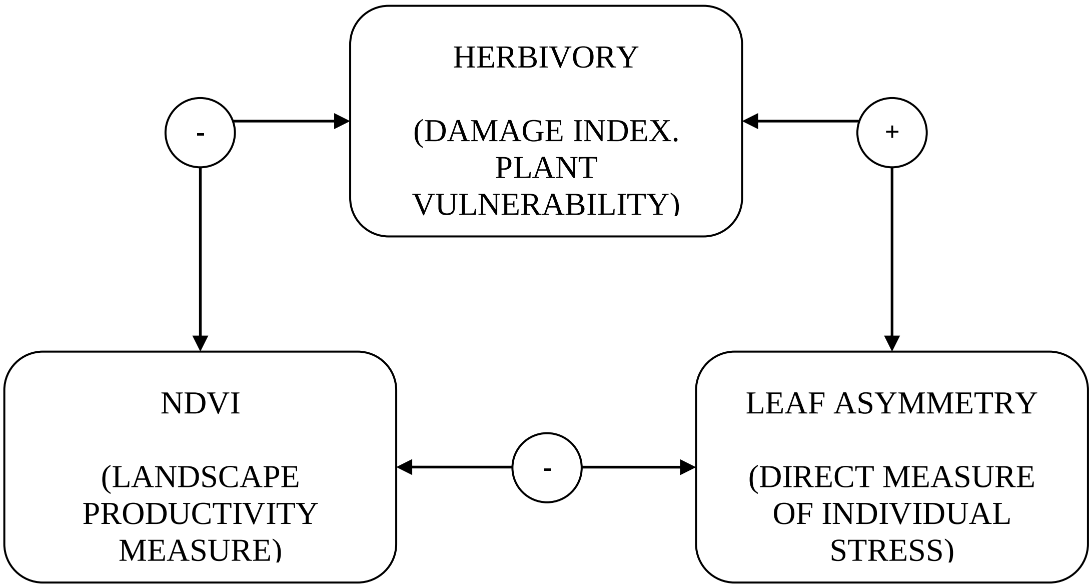

Figure 1. Hypothesis system of this work. The connectors with (-) indicate a negative expected correlation, and connectors with (+) indicate a positive expected correlation.

2. Materials and Methods

2.1 Study Area

The study was carried out between the 15 and 16th of march 2007, in a Caatinga area of the Catimbau Valley National Park (8º 32' 27''S; 37º 14' 51''W), municipality of Buíque, Pernambuco state, Brazil, situated 285km from Recife (Figure 2). The closest meteorological station of the area is located on the town of Buíque and recorded an annual average air temperature and rainfall of 25ºC and 1,095.9 mm, respectively, with the highest rainfall rates occurring during the months of April to June (SUDENE, 1990).

2.2 Data Sampling

In the study area, two trails were sampled, 2.6 km apart from each other. The first (930 m average altitude) with 569 m long, and the second (840 m average altitude) 924 m long.

Plant species with different habits were selected: four scrub species (*Croton sonderianus* Müll.Arg. N=18, Euphorbiaceae; *Couratea hexandra* (Jacquin) K. Schumann. N=19, Rubiaceae; *Croton campestris* St.Hil. N=20, Euphorbiaceae; *Tocoyena formosa* K.Schum. N=12, Rubiaceae), three tree species (*Bauhinia pentandra* Vog. ex D.Dietr. N=9, Leguminosae; *Erythroxylum revolutum* Mart. N=9, Erythroxylaceae; *Anacardium occidentale L*. N=10, Anacardiaceae) and two climber species *(Passiflora luetzelburgii* Harms N=7, Passifloraceae; *Ipomoea subincana* Meisn. N=19, Convolvulaceae). For the abundant species, 20 individuals were sampled, and for the less frequent, the number of sampled individuals varied according to the abundance in the sampling area. From each individual, ten leaves were removed for herbivory rate and LA estimation.

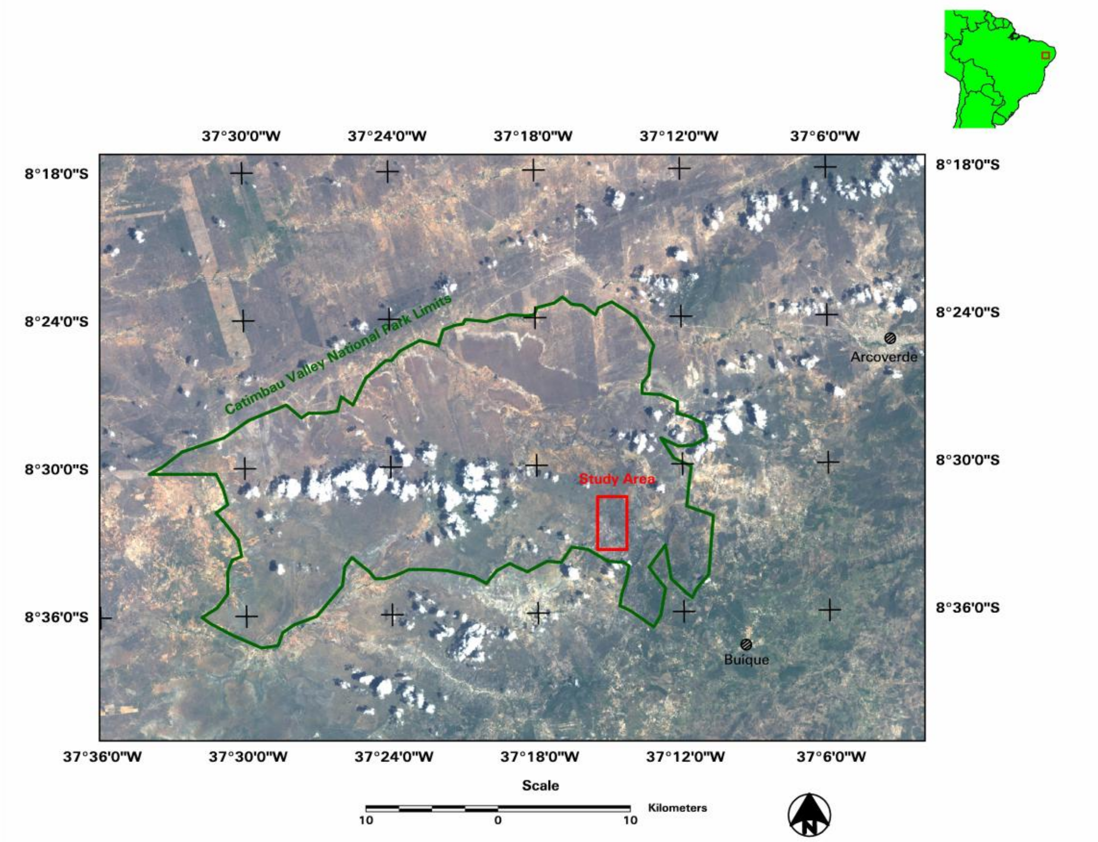

Figure 2. Delimitation of the Catimbau Valley National Park (green), Pernambuco, Brazil, with emphasis to the study area (red), and the Landsat 7 ETM+ image from 12/01/2001 in the background. The map units are UTM, zone 24 south, georreferenced to the South America 69 datum.

2.3 Digital Database

At each individual a GPS point was marked, and from this point with the use of the ARCVIEW 3.2a software, a circle of 20 m radius was generated. A NDVI average was calculated inside the circle, and associated to the landscape vegetation productivity (Figure 3).

The NDVI is obtained through the combination of vegetation reflectance or radiance values in two wavelengths, in the case of the LANDSAT ETM+ Sensor, visible red (630-690 nm) and infrared (770-900 nm). In the visible spectrum there is great absorption of incident radiation by plant chlorophyll, whereas in near infrared there is great reflectance on the leaf mesophyll. The contrast between these two wavelengths emphasizes vegetation, allowing for its clear identification and evaluation of some properties. The NDVI values span from -1 to +1, corresponding to vegetation stress, high reflectance in visible red, and low primary production (towards 0) and an exuberant vegetation, low visible red reflectance and high primary production (towards +1). The index's equation is presented in (1) (Myneni and Williams, 1994):

$$NDVI = \frac{NIR - VR}{NIR + VR} \qquad (1)$$

NIR: Near Infrared. VR: Visible Red.

For the NDVI calculation, we used the ERDAS 8.4 software, with a LANDSAT 7 ETM+ sensor satellite image (orbital parameters: p215 r066, data: 12/01/2001), that was acquired from the Global Landcover Facility (http://glcf.umiacs.umd.edu/). This image was selected for its availability, lack of clouds and for having precision georeferencing (ortho level). The image was corrected for atmospheric interferences, converted to reflectance, and had the vegetation index value obtained using the SEBAL procedure (Allen et al. 2002).

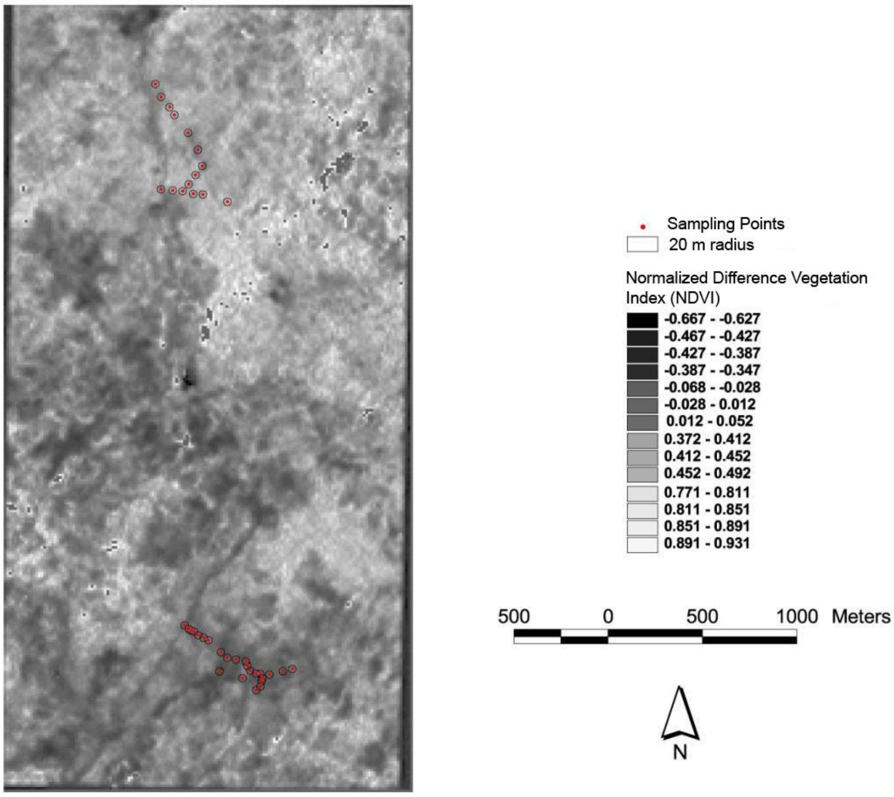

Figure 3. NDVI processing of the study area, indicating the sampling points through the trails. The trail 1 was sampled on 03/15/2007, and trail 2 in the next day. The bright areas indicate an index closest to +1 (highest photosynthetic activity) and the dark areas indicate an index close to -1 (lowest photosynthetic activity). The images on the lower half of the figure were taken from Google Earth (http://earth.google.com) for illustrative purposes

2.4 Herbivory Rates

The herbivory rate in each leaf was determined by the Garcia-Guzmán and Dirzo (2001) method, which consists in visualizing the leaves and estimating the leaf area lost to herbivores. Therefore, five damage classes were defined (0 = 0% damaged area; 1 = 1-5% damaged area; 2 = 6-25% damaged area; 3 = 26-50% damaged area; 4 = 51-100% damaged area) and then an index was calculated for each individual, with the formula $\sum(n_i.i)/N$.

2.5 Leaf Asymmetry

With the help of a precision ruler of

0,1mm, measures were executed on the wider part of the leaves, from the central nerve to the edge on the right and left sides. To determine the Leaf Asymmetry (LA) of each plant an index was calculated according to (2), along with a correction, in case the asymmetry is size-dependent (Cornelissen and Stiling, 2005).

$$LA = \frac{\sum \left(\frac{|Ri - Li|}{(Ri + Li)/2} \right)}{N} \quad (2)$$

Ri: Right leaf measure. Li: Left leaf measure.

2.6 Data Analysis

We verified the existence of natural LA in the plant species. This step is necessary because many plants exibit natural leaf asymmetry. We applied the Kolmogorov-Smirnov test comparing right and left measure averages in each species (Palmer and Strobeck, 1986). The hypothesis is only viable to test on plants that do not exhibit natural asymmetry.

Next, we tested using Kruskal-Wallis and post-hoc Dunn tests to verify differences in asymmetries and average herbivory rates between species. This step was taken to test for natural differences on herbivory and asymmetry between species.

Finally, using the Spearman correlation test, we verified if the NDVI, LA and herbivory who correlated among themselves.

3. Results

Herbivory caused by chewing insects differed significantly between plant species (Kruskal-Wallis, H=44.717; p=0.001) (Table 1 and Figure 4). In crescent order, *Tocoyena formosa* and *Bauhinia pentandra* demonstrated higher rates of herbivory, while *Passiflora luetzelburgii* and *Croton campestris* suffered a lower herbivory. In the study area, 62% of the sampled leaves were undamaged, and 22% had up to 5% of damage (Figure 5).

Table 1. Sampled plant species in a Caatinga area, Catimbau Valley National Park, Pernambuco, Brazil. Averages of herbivory and LA indexes, observed between the 14th and 15th March 2007

Family	Species	Type	N	[1] Average – Herbivory index	[2] Average – LA index
Anacardiaceae	*Anacardium occidentale*	Tree	10	0.31	0.1045
Caesalpineaceae	*Bauhinia pentandra*	Tree	9	0.7889 b	0.0657
Convolvulaceae	*Ipomoea subincana*	Climber	19	0.3474	0.0724
Erythroxylaceae	*Erythroxylum revolutum*	Tree	9	0.2667	0.0967
Euphorbiaceae	*Croton campestris*	Shrub	20	0.075 abc	0.0939
	Croton sonderianus	Shrub	18	0.2389	0.0999

continuação					
Passifloraceae	*Passiflora luetzelburgii*	Climber	7	0.0286 abc	0.1
Rubiaceae	*Couratea hexandra*	Shrub	19	0.5842 a	0.0964
	Tocoyena formosa .	Shrub	12	0.7083 c	0.1023

[1]Averages without letters do not differ statistically in the column. Averages followed by the same letter differ by the Dunn test at 5%.

[2] Averages do not differ significantly by the Kruskal-Wallis test at 5%.

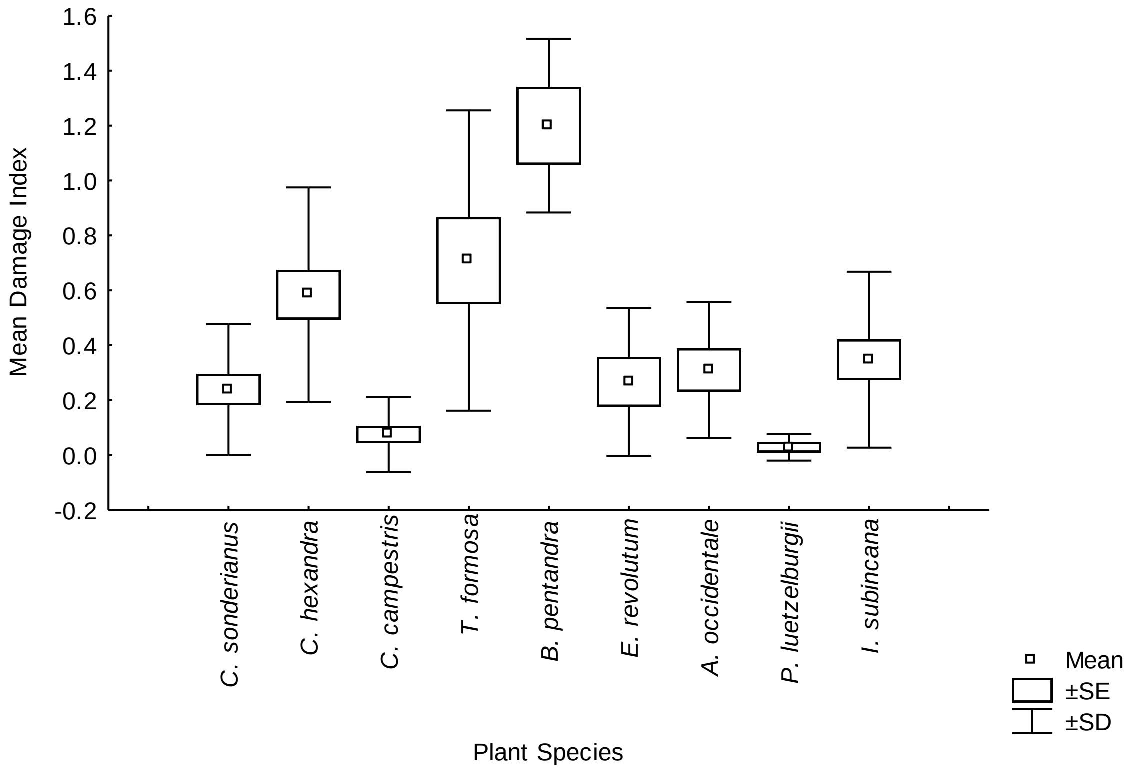

Figure 4. Mean Damage Index (Garcia-Gusman & Dirzo, 2001) of plant species in a Caatinga area, Catimbau Valley National Park, Pernambuco Brazil

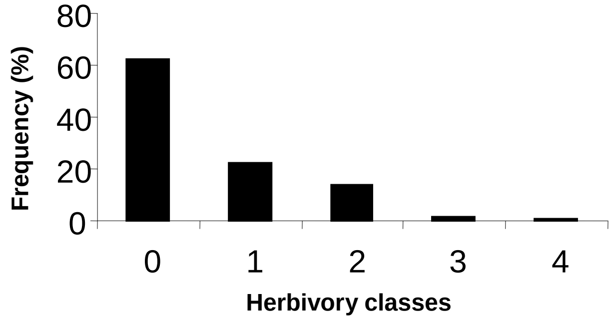

Figure 5. Frequency of the herbivory classes (0 = 0% damaged area; 1 = 1-5%; 2 = 6-25%; 3 = 26-50%; 4= 51-100%), of all the studied plant species (N=8; Sampled leaves = 1230) in an area of Caatinga, Catimbau Valley, Pernambuco, Brazil

The test for LA showed a slight tendency for natural asymmetry in the analyzed plant species. Even with this result, we still thought it would be interesting to test LA with the correlations. The LA index between species did not differ significantly (Kruskal-Wallis, H=9.4961, p=0.3022) (Table 1). Next, we performed correlation analysis (5% significance) comparing two on two the indexes (NDVI, LA and Herbivory). In the correlation test between NDVI and LA, we found that out of the nine analyzed species, only *Tocoyena formosa* and *Bauhinia pentandra* were not significantly correlated (Figure 6 and Table 2). The inverse was found when we correlated Herbivory and NDVI – the two cited species where the only ones to correlate significantly.

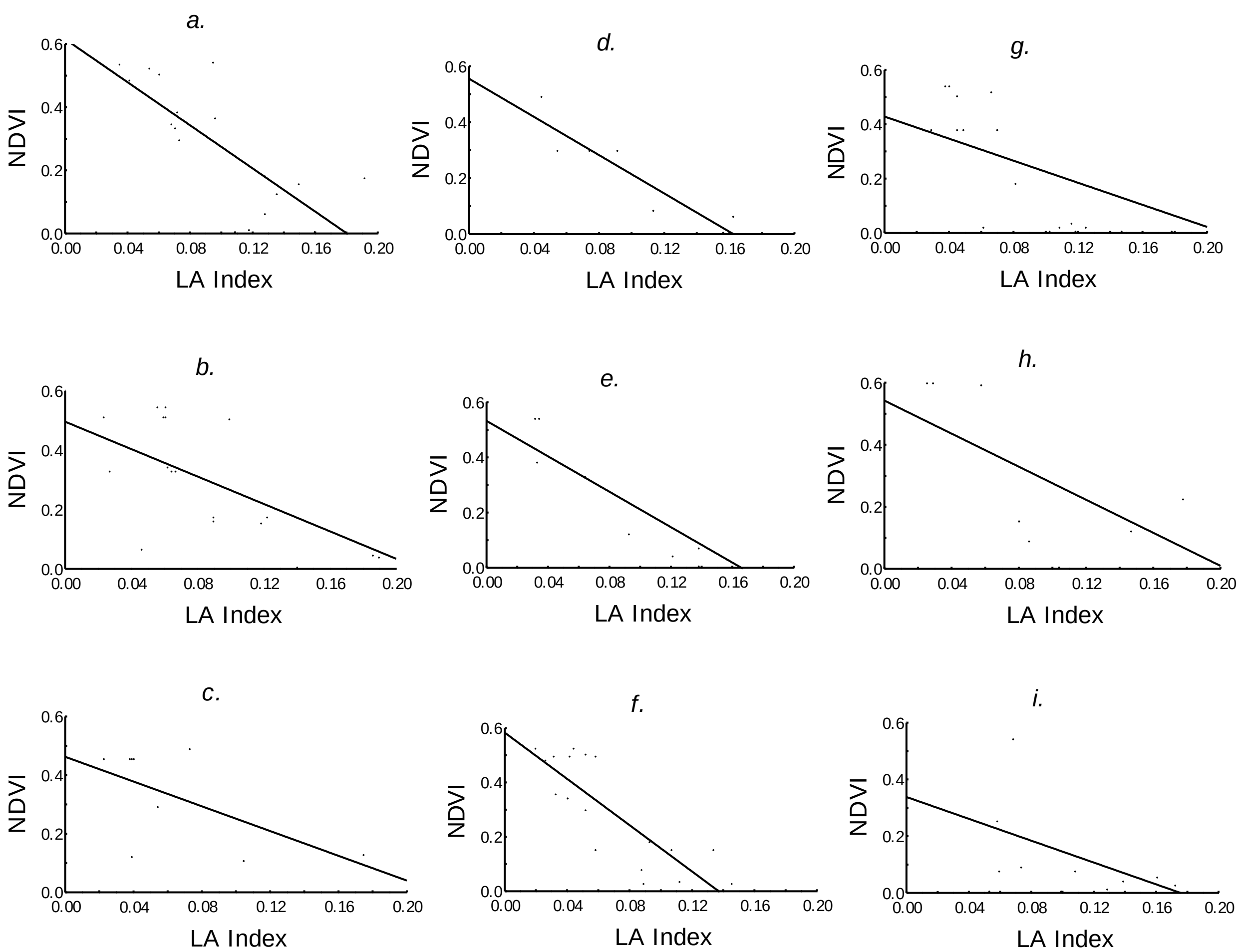

Figure 6. Correlation plots of NDVI vs. LA index for the species a. *Croton sonderianus*, b. *Couratea hexandra*, c. *Bauhinia pentandra*, d. *Passiflora luetzelburgii*, e. *Erythroxylum revolutum*, f. *Ipomoea subincana*, g. *Croton campestris*, h. *Anacardium occidentale* e i. *Tocoyena formosa* in an area of Caatinga, Catimbau Valley, Pernambuco, Brazil.

Table 2. Correlations between the NDVI and LA index in an area of Caatinga, Catimbau Valley, Pernambuco, Brazil. N.S.: Not significantly correlated.

Species	N	Spearman R	P
Anacardium occidentale	9	-0.728040	0.026154
Bauhinia pentandra	9	-0.409082	ns
Couratea hexandra	19	-0.700900	0.000800
Croton campestris	20	-0.589232	0.006260
Croton sonderianus	18	-0.777491	0.000146
Erythroxylum revolutum	9	-0.937247	0.000192
Ipomoea subincana	19	-0.813026	0.000023
Passiflora luetzelburgii	7	-0.926562	0.002697
Tocoyena formosa	12	-0.284212	ns

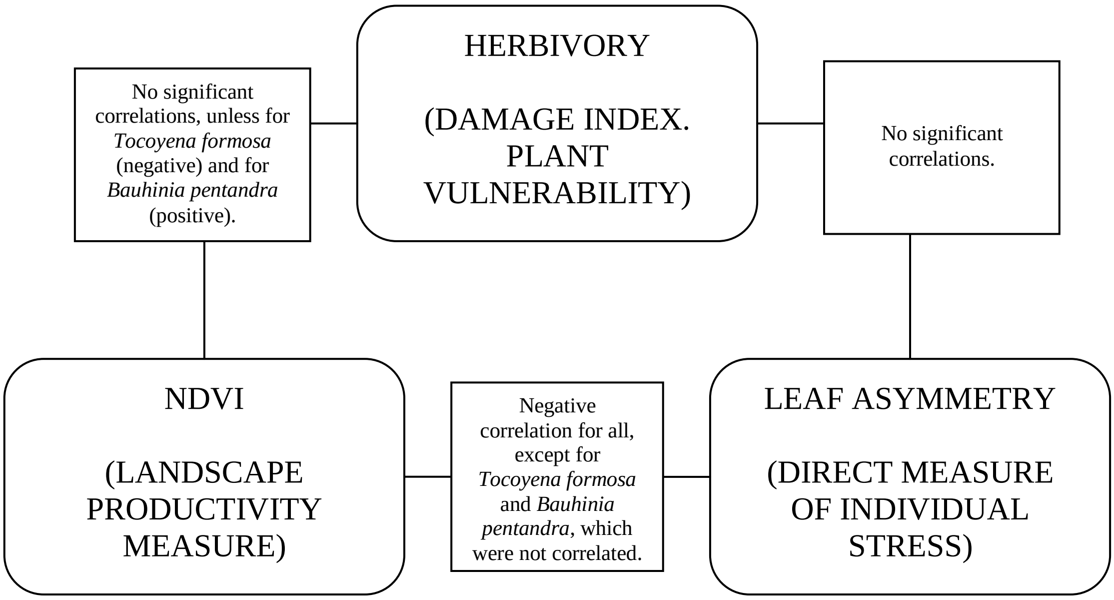

Figure 7. Result of the hypothesis tests for this study.

4. Discussion

Out of the nine analysed species in the transect, seven presented significant correlations of NDVI with LA. The NDVI is known to be an indicator of photosynthetic activity (Myneni and Williams, 1994), with good efficiency for analysis in drier climates (Liesenberg *et al*., 2007) and the correlations indicated that individuals with higher LA values were located in areas with lower photosynthetic rates (as measured by the NDVI).

Therefore, using the NDVI index as a reference, we were able to identify environments less favourable to plant growth.

Although several studies suggest the LA index as one of the measures of plant stress and thus vulnerability to herbivory, (i.e., Moller, 1995; Wiggins, 1997; Zvereva et al., 1997), this relation is not so evident, and each species can present specific responses (Bañuelos *et al.*, 2004).

Another explanation for the lack of significant differences between the studied species is that they respond in a similar manner to environmental conditions.

Our hypothesis that LA and herbivory index were positively correlated between themselves and negatively correlated with NDVI was only partially corroborated (Figure 7). The relationship between LA and the herbivory index was inconclusive, and the correlation between the herbivory index and NDVI was only significant for one species, *Tocoyena formosa*. The statistics tests for the correlation between NDVI and LA were significant for 7 out of the 9 species, making this element of the hypothesis system the most likely to be correlated in future studies.

Besides from differing in relation to other species with respect to NDVI-LA correlation, the *Tocoyena formosa* and *Bauhinia pentandra* were also the taxons with highest rates of herbivory. This fact may shed a light in this distinctive pattern – the LA increases its value in very high peaks of herbivory (Bañuelos *et al.*, 2004). In this way, high values of herbivory could mask the relationship between LA and NDVI. Another possibility is that there is not in fact any relation between LA and stress in these two species, as found in some recent studies (Lens *et al.*, 2002; Siikamäki *et al.*, 2002). More detailed studies are needed to elucidate these patterns.

In the studied Caatinga area, the plants differed significantly in relation to herbivory rate. As this ecosystem has a wide variety of biotic (species richness, population density and distribution) and abiotic (light, temperature, humidity, precipitation, soil, relief, etc.) factors, this result can stem from the large number of specific interactions between plants and herbivorous insects (Lowman, 1985; Coley and Aide, 1991).

In this study the NDVI provided a rather interesting quantification of the environment productivity, being probably related to plant stress. The results suggest the NDVI as a long range habitat stress indicator, in this way assisting natural reserve management and the definition of priority areas for conservation.

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