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# Spatial-temporal variability of leaf chlorophyll and its relationship with cocoa yield

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#### ABSTRACT

The objective of this study was to evaluate the spatial-temporal variability of leaf chlorophyll index and its relationship with cocoa yield. The experiment was carried out in an experimental area of cocoa production located in Ilhéus, Bahia State, Brazil. Leaf chlorophyll content was measured in September, October, January, February, March and April in the 2014/2015 season, at each sampling point of a regular grid by using a portable chlorophyll meter. Under the same conditions, yield was evaluated and the data were submitted to descriptive statistics and a linear correlation study. Geostatistical analysis was used to determine and quantify the spatial and temporal variability of leaf chlorophyll index and yield. Leaf chlorophyll index varied over the period evaluated, but the months of February, March and April showed no spatial dependence in the study area, indicating absence of temporal stability. Cocoa monthly yield, except in January, presented high spatial variability. Under the conditions of this study, it was not possible to establish a relationship between leaf chlorophyll index and cocoa yield.

Palavras-chave:

*Theobroma cacao* geoestatística mapas temáticos

## Variabilidade espaço-temporal da clorofila foliar e sua relação com a produtividade do cacaueiro

#### RESUMO

Objetivou-se neste estudo avaliar a variabilidade espaço-temporal do índice foliar de clorofila e a relação com a produtividade do cacaueiro. O experimento foi realizado em uma área experimental de produção de cacau localizada no município de Ilhéus, BA. O índice foliar de clorofila foi medido nos meses de setembro, outubro, janeiro, fevereiro, março e abril na safra 2014/2015 em cada ponto amostral de uma malha regular, utilizando-se um medidor portátil de clorofila. Nas mesmas condições, a produtividade foi avaliada e os dados foram submetidos à estatística descritiva e estudo de correlação linear. A análise geoestatística foi utilizada para determinar e quantificar a variabilidade espacial e temporal do índice foliar de clorofila e da produtividade. O índice foliar de clorofila variou ao longo do período avaliado, porém os meses de fevereiro, março e abril não apresentaram dependência espacial na área em estudo, indicando ausência de estabilidade temporal. A produtividade mensal do cacaueiro, exceto no mês de janeiro, apresentou elevada variabilidade espacial. Nas condições estudadas, não foi possível estabelecer uma relação entre o índice foliar de clorofila e a produtividade do cacaueiro.

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#### **INTRODUCTION**

As with other agricultural crops, cocoa cultivation has transformed into a specialized branch, which aims at differentiation and maximum added value to the final product. Silva et al. (2014) comment that the differentiation of agricultural products encompasses the adoption of integrated technologies that aim to obtain as much information as possible about the production systems, allowing the adoption of new management strategies.

Using technological models and different sensors for data collection has led to higher efficiency in decision-making, ensuring higher accuracy in the managements and lower uncertainties in agricultural production (Valente et al., 2012). Partial replacement of conventional methods of soil fertility and plant nutritional status evaluation using sensors has been widely studied in precision agriculture (Godoy et al., 2008; Silva et al., 2014).

Some equipment, such as chlorophyll meters, after correlation with traditional methods, have been satisfactorily used in different agricultural crops to recommend foliar supplementation of nutrients (Barbieri Júnior et al., 2012), but this information is still scarce for the cocoa crop.

Joint use of sensors in indirect measurement of plant nutrition and the knowledge and evaluation of spatial and temporal variability of yield and the involved factors constitutes an important tool to define rational management strategies for agricultural areas (Valente et al., 2012). Silva et al. (2010a) claim that the knowledge on the variation of attributes from the soilplant system is important for the management of agricultural practices and, in certain situations, can ensure the success of the agricultural enterprise.

Given the above, this study aimed to adopt precision agriculture tools in cocoa cultivation and evaluate the spatial-temporal variability of leaf chlorophyll index and its relationship with cocoa yield.

#### MATERIAL AND METHODS

The study was carried out in an area belonging to the Executive Commission for the Cocoa Farming Plan – CEPLAC, of the Ministry of Agriculture, Livestock and Supply. The area is located in the southern region of Bahia, municipality of Ilhéus (14° 47' S; 39° 16' W). The climate, according to Köppen's classification (Alvares et al., 2013) is Af, humid tropical, with mean annual rainfall of 1830 mm and mean annual values of relative humidity and air temperature of 80% and 23.5° C, respectively. The soil, according to the Brazilian Soil Classification System - SiBCS (EMBRAPA, 2013), was classified as eutroferric Haplic Nitosol.

The experimental area was installed in 2003 and has been cultivated in the agroforestry system, shaded by Erythrina. Cocoa is cultivated at spacing of  $3.0 \times 1.5$  m and Erythrina at spacing of  $24 \times 24$  m. In 2013, a regular grid with 120 sampling points was built in the area, and each point was composed of only one cocoa tree. Coordinates of the points were materialized using the local Cartesian coordinate system, with maximum distance of 9.5 m (X-axis) and minimum distance of 6.6 m (Y-axis).

Leaf chlorophyll index was evaluated at each sampling point of the grid, along the 2014/2015 season, in the months of September, October, January, February, March and April. Leaf chlorophyll index was measured using a portable chlorophyll meter (ClorofiLOG<sup>\*</sup> - Model CFL 1030), operated according to instructions of the manufacturer and as suggested by Godoy et al. (2008). Leaf chlorophyll index was determined by measurements taken in the four quadrants of the plant composing each point (lower third, middle third, upper third and apex). In each quadrant, two readings were taken (one on the adaxial face and the other on the abaxial face), totaling 16 values of leaf chlorophyll per point. Thus, the mean value of the 16 readings was attributed to the sampling point.

Cocoa yield was evaluated at the same sampling points of the leaves in the same months of chlorophyll measurement. All healthy fruits apt for harvest were quantified at each sampling point. Healthy fruit data were converted to wet beans by multiplying the number of healthy fruits of each point by the weight of wet beans per fruit. Results of dry beans production per plant were converted to yield (kg ha<sup>-1</sup>).

The results were subjected to descriptive statistical analysis to determine the measures of position, dispersion and form of dispersion. Data normality was evaluated by the Shapiro-Wilk test (W) at 0.05 probability level. The relationship between leaf chlorophyll index and cocoa yield was verified by Pearson's linear correlation analysis at 0.05 probability level.

After that, geostatistical analysis was performed to verify the existence and, in this case, quantify the spatial dependence degree, by fitting theoretical functions to the models of classical experimental variograms, based on the assumption of stationarity of the intrinsic hypothesis, according to the following Eq. 1:

$$\gamma^{*}(h) = \frac{1}{2N(h)} \sum_{i=1}^{N(h)} [z(x_{i}) - z(x_{i} + h)]^{2}$$
(1)

where:

N(h) - number of experimental pairs of observations Z(xi), Z(xi+h), separated by a vector h.

A mathematical function was fitted, and its parameters are known as: nugget effect ( $C_0$ ), corresponding to the value of the intersection in the semivariances axis; sill ( $C_0 + C_1$ ), approximately equal to the data variance value; and range (a), which represents the distance at which the semivariogram reaches the sill value and the region of spatial dependence between the samples.

In the fit of theoretical functions to the experimental variograms, spherical, exponential, Gaussian and linear with sill models were tested. The models were selected based on the least squares criterion, opting for choosing models with higher  $R^2$  (coefficient of determination), lower RSS (residual sum of squares) and higher  $R^2$  of the cross-validation ( $R^2$ -CrVd) (Isaaks & Srivastava, 1989).

After spatial dependence was confirmed, ordinary kriging interpolation method was used to estimate values in sites not measured for leaf chlorophyll index and cocoa yield. This interpolator uses a minimum-variance linear unbiased estimator, which considers the structure of spatial variability found for the variable.

#### **RESULTS AND DISCUSSION**

Based on Table 1, yields in the sampled months and leaf chlorophyll index measured in February did not exhibit normal distribution according to the Shapiro-Wilk test. The other variables showed similar central tendency values (mean and median), with variations of 0.43, 0.20, 0.61, 0.06 and 0.20, respectively, for September, October, January, March and April, indicating symmetrical distribution, according to the asymmetry value close to zero. For Cressie (1991), data normality is not required in geostatistics, provided that the normal distribution curve does not have very elongated tails (high amplitude).

The coefficient of variation, according to the classification proposed by Warrick & Nilsen (1980) (low for CV < 12%, intermediate for 12% > CV < 60%, and high for CV > 60%), was high for the yields in all months. Santos et al. (2017) comment that cocoa yield in the 2014/2015 season, in an experiment in Southern Bahia, also showed high CV, onseason and off-season, with 93 and 75.6%, respectively. Leaf chlorophyll indices in September, January and February showed intermediate variation, while in October, March and April it showed low variation. In months in which the CV was low, rainfall was 71, 73 and 41 mm, respectively. This low variability confirms the hypothesis of Godoy et al. (2008), who claim that, in periods with an increase in light incidence and low rainfall, there is less need for chlorophyll production by plants, which explains the less intense green color and low mean value of leaf chlorophyll index (Table 1).

Leaf chlorophyll indices were similar in all months, with mean value of 56. This result is close to that observed in the literature for the cocoa crop. Dantas et al. (2012), working with cocoa trees in agroforestry system, under similar agroclimatic condition to the present study, found mean chlorophyll value of 52.25. Although the mean value corroborates with the study of Dantas et al. (2012), an analysis of maximum and minimum values demonstrate that the months of September and January show greater amplitude, which justifies the higher CV values for these months.

For the yields, high amplitudes of the data were observed in all months, leading to high coefficients of variation. Silva et al. (2014) claim that CV values, highly influenced by the amplitude of the distribution, are the first indications of data spatial variability. According to the results of Pearson's correlation between yield and chlorophyll values (Table 2), the mean leaf chlorophyll indices relative to the months were not correlated with any value of cocoa yield in the same periods evaluated.

This behavior may be related to the high variability between crop yield data, and may have been influenced by genetic factors or unfavorable environmental conditions (Leite et al., 2012), as well as issues related to the adopted management (Silva et al., 2010b). According to Silva et al. (2010a), these aspects compromise studies of correlation, since it is difficult to establish distribution patterns. Another factor is related to the large genetic diversity between plants cultivated in the area, because of the system of progenies used. Dantas et al. (2012) comment that this form of cultivation is still predominant in Bahia, where most farmers have in the same area a large variety of genotypes with different capacities to tolerate biotic stresses and, consequently, with different production potentials.

In the geostatistical analysis (Table 3), the readings of chlorophyll index in February, March, April and yield in March do not exhibit spatial dependence. Absence of spatial dependence does not characterize inexistence of variability in the phenomenon, it only indicates that the variation occurs randomly (Silva et al., 2013).

The other variables showed spatial dependence, indicating that the behavior of leaf chlorophyll index between plants is not random, but depends on the distance between sampling points. The fitted variograms showed well-defined sills, which indicates that the intrinsic stationarity hypothesis was met and that the variables do not exhibit trend of variation with the directions.

Variables	Mean	Median	Minimum	Maximum	CV	Cs	Ck	W
Chl_Sept <sup>1</sup>	58.04	57.59	26.50	83.25	20.46	-0.20	-0.47	Ns
Chl_Oct <sup>1</sup>	58.61	58.41	45.80	69.78	9.24	-0.18	-0.25	Ns
Chl_Jan <sup>1</sup>	55.29	55.90	29.00	74.85	17.87	-0.38	-0.32	Ns
Chl_Feb <sup>1</sup>	54.96	54.38	32.48	68.40	14.69	-0.52	-0.24	*
Chl Mar <sup>1</sup>	55.70	55.76	45.18	65.36	7.43	-0.09	-0.27	Ns
Chl_Apr <sup>1</sup>	53.82	53.62	45.90	62.58	6.45	0.22	-0.23	Ns
Yield_Sept <sup>2</sup>	428.56	200.89	0.00	4921.78	173.00	3.12	12.41	*
Yield_Oct <sup>2</sup>	417.68	100.44	0.00	7533.33	209.76	5.28	37.27	*
Yield_Jan <sup>2</sup>	453.67	200.89	0.00	4921.78	160.86	3.26	13.43	*
Yield_Feb <sup>2</sup>	415.17	200.89	0.00	4520.00	152.43	3.43	16.03	*
Yield_Mar <sup>2</sup>	657.91	301.33	0.00	4620.44	134.13	2.52	7.28	*
Yield Apr <sup>2</sup>	474.60	241.07	0.00	5303.47	151.20	3.59	17.89	*

Table 1. Descriptive statistics of leaf chlorophyll indices and cocoa yield in different periods

<sup>1</sup>Dimensionless; <sup>2</sup>kg.ha<sup>-1</sup>; M - Mean; Md - Median; s – Standard deviation; CV – Coefficient of variation; Max. – Maximum values; Min. – Minimum values; Cs - Coefficient of asymmetry; Ck – Coefficient of kurtosis; w (\*) – Non-normal distribution by the Shapiro-Wilk test at 0.05 probability level; w (ns) – Normal distribution by the Shapiro-Wilk test at 0.05 probability level; w (ns) – Normal distribution by the Shapiro-Wilk test at 0.05 probability level; w (ns) – Normal distribution by the Shapiro-Wilk test at 0.05 probability level; w (ns) – Normal distribution by the Shapiro-Wilk test at 0.05 probability level; w (ns) – Normal distribution by the Shapiro-Wilk test at 0.05 probability level; w (ns) – Normal distribution by the Shapiro-Wilk test at 0.05 probability level; w (ns) – Normal distribution by the Shapiro-Wilk test at 0.05 probability level; w (ns) – Normal distribution by the Shapiro-Wilk test at 0.05 probability level; w (ns) – Normal distribution by the Shapiro-Wilk test at 0.05 probability level; w (ns) – Normal distribution by the Shapiro-Wilk test at 0.05 probability level; w (ns) – Normal distribution by the Shapiro-Wilk test at 0.05 probability level; w (ns) – Normal distribution by the Shapiro-Wilk test at 0.05 probability level; w (ns) – Normal distribution by the Shapiro-Wilk test at 0.05 probability level; w (ns) – Normal distribution by the Shapiro-Wilk test at 0.05 probability level; w (ns) – Normal distribution by the Shapiro-Wilk test at 0.05 probability level; w (ns) – Normal distribution by the Shapiro-Wilk test at 0.05 probability level; w (ns) – Normal distribution by the Shapiro-Wilk test at 0.05 probability level; w (ns) – Normal distribution by the Shapiro-Wilk test at 0.05 probability level; w (ns) – Normal distribution by the Shapiro-Wilk test at 0.05 probability level; w (ns) – Normal distribution by the Shapiro-Wilk test at 0.05 probability level; w (ns) – Normal distribution by the Shapiro-Wilk test at 0.05 probability level; w (ns) – Normal distribution by the Shapiro-

Tab	le 2. P	'earson'	s I	inear	corre	lation	between	yield	d and	ch	lorop	bhy	/11	leve	els
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Variables	Yield_Sept	Yield_Oct	Yield_Jan	Yield_Feb	Yield_Mar	Yield_Apr
Chl_Sept	-0.06	0.02	-0.06	-0.10	-0.07	0.11
Chl_Oct	-0.01	0.05	-0.02	-0.02	-0.09	0.08
Chl_Jan	0.04	0.09	-0.09	-0.12	-0.03	0.08
Chl_Feb	0.06	0.07	-0.07	-0.10	-0.04	0.10
Chl_Mar	0.03	0.01	-0.08	-0.13	-0.02	0.25
Chl Apr	0.01	0.05	-0.07	-0.10	-0.05	0.14

Variables	Model	Co	<b>C</b> <sub>0</sub> + <b>C</b>	Α	R <sup>2</sup>	SDI	R2-CrVd
Chl_Sept	Spherical	5.81	121.80	16.00	83.00	95.00	23.10
Chl_Oct	Exponential	6.00	48.66	19.00	86.00	87.70	24.50
Chl Jan	Gaussian	0.10	89.40	10.00	94.00	98.00	29.80
Chl_Feb	PNE	-	-	-	-	-	-
Chl_Mar	PNE	-	-	-	-	-	-
Chl_Apr	PNE	-	-	-	-	-	-
Yield_Sept	Spherical	0.18	0.93	13.00	93.00	81.00	27.00
Yield_Oct	Spherical	0.20	0.88	13.00	83.00	77.00	21.80
Yield_Jan	Exponential	0.29	1.32	33.00	89.00	78.00	25.00
Yield_Feb	Spherical	0.17	0.94	14.00	91.00	82.00	23.00
Yield_Mar	PNE	-	-	-	-	-	-
Yield_Apr	Spherical	0.21	0.91	11.00	89.00	77.00	29.00
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Table 3. Parameters of the variograms for yield and chlorophyll levels

Cn – Scaled nugget effect; Cn + C – Scaled sill; a - Range; SDI – Spatial Dependence Index; R<sup>2</sup> - Coefficient of determination; CrVd – Cross-validation; PNE – Pure nugget effect

For variables that showed spatial dependence, the geostatistical analysis demonstrates that the theoretical models that fitted best to the experimental variograms were the spherical, exponential and Gaussian. According to Silva et al. (2007), the presence of spatial dependence observed for the studied variables indicates that management practices based only on mean values are not much reliable, because they do not consider spatial variability, possibly leading to values that reduce the efficiency of the systems.

For Cambardella et al. (1994), spatial dependence is considered as strong when the spatial dependence index (SDI) is lower than 25%, moderate from 25 to 75% and weak above 75%. Based on the respective values of SDI, the dependence was weak for all variables, with SDI >75%. SDI informs that regionalized variables do not have random behavior and that the distance between points, defined in the sampling grid, satisfactorily demonstrated the spatial variability existing in the field (Dalchiavon et al., 2012). In terms of management, this indicates that the practices adopted to apply the fertilizers must be carried out at varying rates, considering the spatial behavior of the variable, i.e., management strategies performed based on mean doses may lead to unsuccess or unsatisfactory results.

The highest range of the variograms occurred for the variable of yield in January (33 m), contrary to the chlorophyll index in the same month with lower range (10 m). The high value of range (33 m) confirms that, within this interval, the variable showed low spatial variability, because its area of influence will be larger, suggesting a greater similarity between the close sampling points, i.e., leading to a behavior of high spatial continuity. Range represents the limit of spatial dependence and, from this value on, the samples behave randomly and serve as indication of the interval between the mapping units (Grego & Vieira, 2005).

Except for chlorophyll indices obtained in September and October, values of nugget effect  $(C_0)$  for all variables were very close to zero. Since  $C_0$  is the value of semivariance for the distance zero and represents the spatial variability component that cannot be related to a specific cause (random variability), the lower its value, i.e., the lower the random variation, the more precise the estimate, through kriging (Silva et al., 2010b).

In the thematic maps of yield (kg ha<sup>-1</sup>), interpolated by ordinary kriging, considering the spatial dependence of September (Figure 1A), October (Figure 1B), January (1C), February (Figure 1D) and April (Figure 1E), it is observed through the spatialization of yield a large portion of the studied area with values from 0 to 400 kg ha<sup>-1</sup> and in lower proportion, above 1200 kg ha<sup>-1</sup>, in all months evaluated. Genetic and environmental factors may interfere with the yield of a plant and, when different genotypes are evaluated in agroforestry system at different planting densities and solar radiation, there is a direct influence on the photosynthetic rate of the plants, making such difference in yield even more accentuated (Leite et al., 2012).

Deheuvels et al. (2012), evaluating samples of cocoa in different agroforestry systems and planting densities, observed that the yield estimated in bean dry matter varied along the year. According to these authors, different cultivation systems resulted in different yields among the plants; hence, the variation expressed within and between systems is influenced by variations existing in the soil, plant and in the covering canopy.

The maps interpolated by ordinary kriging for leaf chlorophyll index obtained using a chlorophyll meter, considering spatial dependence, are presented in Figures 2A, B and C for September, October and January, respectively. The differences within the area and along the months are evident in the maps and may result from the differences in light intensity because, even in an agroforestry system, there are some clearings that allow greater passage of light. Dantas et al. (2012) concluded that chlorophyll index variability between cocoa plants is related, among other factors, to morphological adaptations of the different genotypes in response to variation of luminosity in the agroforestry environment.

Plants from the largest portion of the area show intermediate to high indices, considering the three classes of distribution adopted in the construction of the maps. Variations in chlorophyll indices within a same species are correspondent to the morphological adaptations occurred in response to the variations of luminosity in the environment (Dantas et al., 2012). With an increase in light availability, there is less need for chlorophyll production (Godoy et al., 2008).

Leaves in the shade exhibited higher chlorophyll content than leaves exposed to sunlight, because this pigment is constantly synthesized and destroyed by photo-oxidation in the presence of light (Siebeneichler et al., 2008). Leaves exposed to the sun prioritize the biochemical production of photosynthesis, so that N is preferentially allocated in chloroplast enzymes, such as nitrate reductase, RuBisCO, among others, in detriment of the light-harvesting system (chlorophylls), deferred under low irradiance (Wang et al., 2012).



Figure 1. Thematic maps of cocoa yield (kg ha<sup>-1</sup>) in different periods: September (A), October (B), January (C), February (D) and April (E)



Figure 2. Thematic maps of chlorophyll levels (Chl) in cocoa trees in September (A), October (B) and January (C)

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#### Conclusions

1. Leaf chlorophyll index showed temporal variability, but the months of February, March and April did not exhibit spatial dependence.

2. Cocoa monthly yield, except in January, showed high spatial variability.

3. There was no relationship between leaf chlorophyll index and cocoa yield.

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