



Spectral responses of soybean leaflets crops to different water conditions and tillage systems

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ABSTRACT

The state of Rio Grande do Sul is one of the greatest Brazilian soybean producers, which justifies the use of remote sensing techniques for monitoring areas occupied by this crop. The purpose of this work was to characterize throughout the crop cycle the variability of spectral responses of soybean leaflets, subjected to different conditions of soil tillage and water supply. The experiment was carried out in a 0.5 ha area, located in Eldorado do Sul, Rio Grande do Sul State, southern Brazil, in two systems of soil tillage (no-tillage and conventional tillage) and two levels of water supply (irrigated and non-irrigated). The cultivar Fepagro RS-10 was sown in a row spacing of 0.40 m and in a population of 400,000 plants per hectare. An integrating sphere of a spectroradiometer LI-COR, model LI-1800 was used for measuring the absorbance, reflectance, transmittance on soybean leaflets. The results showed that the pattern of the incident radiation partitioning in the reflectance, transmittance and absorbance components is influenced by the crop phenological stage and by the tillage system. Despite this, there is stability on the reflectance of soybean leaflets in the red and infrared portions of the electromagnetic spectrum, throughout the crop cycle. The inversely proportional relation between absorbance and reflectance of soybean leaflets revealed viability on the reflectance data, in monitoring of agricultural crops.

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Introduction

Considering the economic and social importance of the soybean crop and its high yield variability in the State of Rio Grande do Sul, Brazil, the development of techniques and methods for monitoring the conditions of development of plants is relevant. Due to the large areas

occupied by soybeans, in the State, one of the alternatives that have contributed in this sense is the use of orbital remote sensing techniques, which allow monitoring crops, on a regional scale and with adequate frequency. Several studies developed in this theme have shown the contribution of satellite images in crop forecasting systems (Zhu et al., 2016; Gusso et al., 2017). For soybean

cultivation in Rio Grande do Sul, images have been used for crop area estimation (Santos et al., 2014; Mengue et al., 2015) and to monitor plant growth and development conditions (Schirmbeck et al., 2017) and to estimate crop yields (Rizzi et al., 2006; Melo et al., 2008).

Several efforts have been developed to generate information to support agricultural monitoring systems. In this sense, however, it is necessary to develop basic studies, using terrestrial radiometry, that allow to establish useful relations between the spectral responses and plant growth parameters (Fonseca et al., 2002; Monteiro et al., 2012; Pinto et al., 2017; Junges et al., 2017).

The expression "spectral response of vegetation" can be used to represent the spectral properties of the leaves, branches, isolated plant, or set of plants, which depend on the surface characteristics of the leaf as well as other characteristics of the vegetation and of the canopy (Ponsoni et al., 2012). Reaching the surface, the solar radiation incident on the vegetation can be reflected, absorbed or transmitted. By the law of energy conservation, the sum of reflected, absorbed and transmitted radiation is equal to the radiation incident on the leaves. In general, these components are expressed in relative terms, using the variables referred to as absorbance (α), transmittance (τ) and reflectance (ρ), whose sum must equal 1 or 100% (Formaggio and Sanches, 2017).

The values of reflectance and transmittance have similar variations over the electromagnetic spectrum (Jensen, 2009). On the other hand, it is known that the biomass production is highly related to the total amount of energy absorbed by the leaves (Hall e Rao, 1980). Since the values of absorbance are almost inverse to those of reflectance, and considering the possibility of measuring reflectance values through remote sensors, it is therefore feasible to establish relations between reflectance and biomass parameters. Quantification of these flows may be useful in differentiating the growth conditions of a given crop.

In the visible portion of the electromagnetic spectrum, as a consequence of the high absorption rate by the pigments present in leaf chloroplasts, there is a low reflection of the radiation (Jensen, 2009). The reflectance in this range is, on average, 10% of the incident radiation. The average amounts of pigments in higher plants are: 65% chlorophyll, 6% carotene and 29% xanthophylls (Hall e Rao, 1980). Chlorophyll gives a green color to plants. Chlorophyll a is blue-green and is present in all organisms that release O₂. Chlorophyll b is yellowish green and is present in the leaves of higher plants and in green algae. Several factors may alter these proportions of pigments, such as leaf senescence and mineral malnutrition. Reductions in the content of chlorophyll allow the manifestation of auxiliary pigments, previously masked, in a phenomenon called

chlorosis.

The reflectance is higher in the near infrared region, in comparison to that in the visible portion, due to the intense scattering of the radiation in the mesophyll. From 700nm a marked gradient occurs, indicating the transition to high reflectance in this portion of the electromagnetic spectrum, which corresponds to approximately 40-60% of the incident radiation. Up to about 1,300 nm, the reflectance remains high and is related to internal structural characteristics of the leaves, where multiple reflections and refractions occur, associated with discontinuities of refractive indices (Jensen, 2009).

This work assumes that management practices applied to soybean in Rio Grande do Sul, which influence the growth of plants, may promote changes in their spectral responses, whose detection by remote sensors allows the quantification of biophysical parameters of the crop. In this context, the objective of this work was to characterize the variability in spectral responses of soybean leaflets, during the cycle of soybeans, when submitted to different soil tillage systems and to different levels of water availability.

Material and Methods

The data presented are part of an extensive set of measurements obtained in a field experiment carried out during the 2003/2004 cropping season, in an area of 0.5 ha, cultivated with soybeans, at the Agronomic Experimental Station of the Universidade Federal do Rio Grande do Sul, in Eldorado do Sul, Rio Grande do Sul State, in southern Brazil (30° 05'S, 51° 40'W and 46m).

The climate of the region is humid subtropical, belonging to the type Cfa, according to the classification of Köppen. The average monthly air temperature is 25°C in January and February, and 13°C in June and July (Bergamaschi et al., 2003). The soil of the experimental area is a typical Dystrophic Red Argis soil, with textural B horizon. Its granulometry is composed of 49, 22 and 29% sand, silt and clay, respectively, in no-tillage, and 42, 27 and 31%, respectively, in conventional tillage (Rojas, 1998).

The soybean cultivar was the late-cycle Fepagro-RS10, which was sown on November 20th, 2003, with 0.40m of line spacing and a population of 300,000 plants ha⁻¹. The experimental design was a three-factor randomized blocks, in strips for the soil tillage systems and irrigation levels, with the time (evaluation periods) in subplots. The treatments were: non-irrigated no-tillage (DNI), irrigated no-tillage (DI), irrigated conventional tillage (IC), and non-irrigated conventional tillage (CNI). Further information on the conduct of this experiment can be found in Martorano (2007).

During the crop cycle, the partitioning of the radiation incident on the soybean leaflets, in the components:

transmittance, reflectance and absorbance, was determined. The measurements were made on January 7, February 9, March 2, 9, 24, 30 and April 15 and 20, 2004, which corresponded to 41, 74, 95, 102, 117, 123, 139 and 144 days after the emergency (DAE), respectively. In each evaluation period, leaflets were randomly collected from the last expanded leaf, using three replicates for each of the four treatments. The determinations were performed in the laboratory, and the leaves were collected and stored, for a short time, in plastic bags to avoid dehydration. Measurements were performed in laboratory, immediately, after sampling the leaflets of each treatment.

The equipment used was a LI-COR spectroradiometer, model LI-1800, with a spectral resolution of 2nm, at wavelengths between 300nm and 1,100nm. Using the integrator sphere of the spectroradiometer, the incident energy (reference), the reflected energy, and the transmitted energy by the soybean leaflets were measured. The illumination was done by an external source of halogen light (lamp), present in a cone with a diameter of 1.14cm, that restricts the lighting in the sheet. The sphere has five entrances; the entrance where the leaf sample is placed is 1.45cm in diameter, and the entry for observation is 0.64cm in diameter. The inner part of the sphere is covered with barium sulphate, as well as the reference plate used.

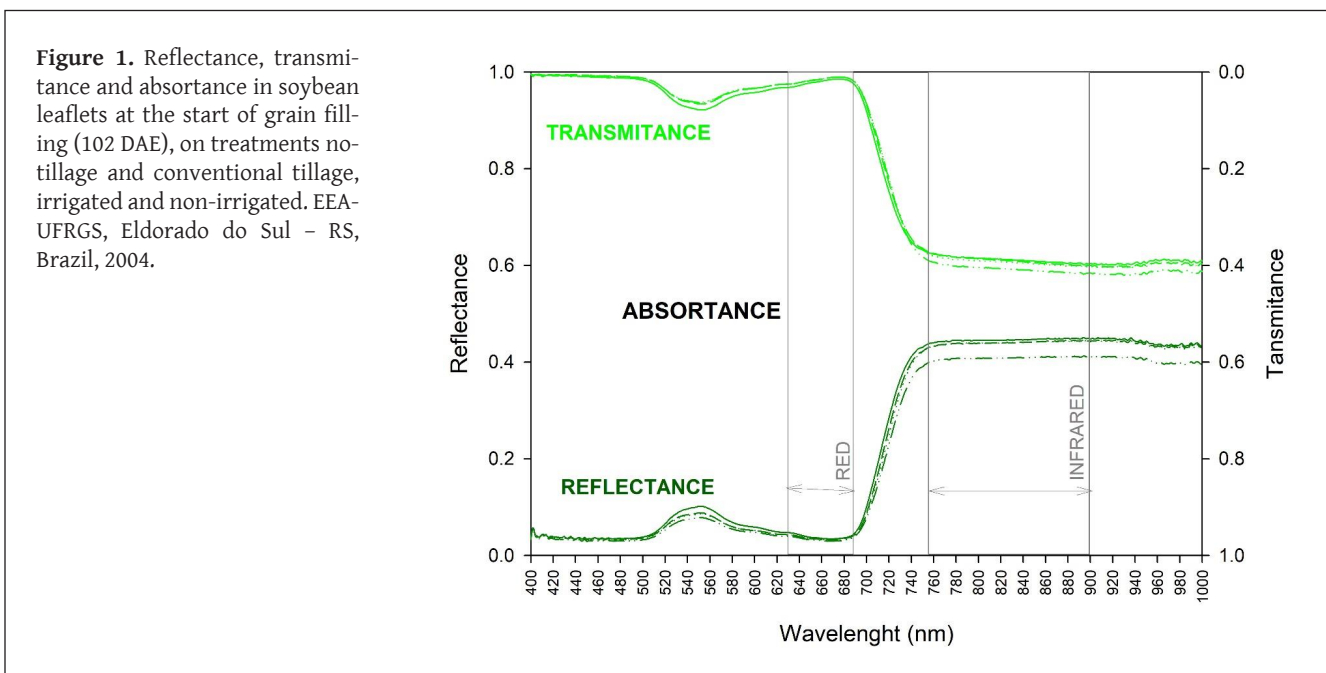
After the measurements, the following components were determined: transmittance (τ_λ - ratio between the transmitted and incident energy), and reflectance (ρ_λ - ratio between the reflected and incident energy). The values of the absorbance component (α_λ - ratio between the absorbed and incident energy) were obtained through the equation:

$$\alpha_\lambda = 1 - (\rho_\lambda + \tau_\lambda) \quad (1)$$

The data of absorbance, reflectance and transmittance along the measured electromagnetic spectrum were used for plotting curves at three representative dates: start of cycle (41 DAE), start of grain filling (102 DAE) and end of cycle (139 DAE). In the subsequent analysis, the pattern throughout the time (January to May) of the reflectance in the visible and infrared portions was characterized, since these are the wavelengths most commonly used to calculate vegetation indices. Finally, the Pearson correlation analysis ($p < 0.05$) was performed, between the reflectance and absorbance components, in these same portions of the electromagnetic spectrum.

Results and Discussion

The typical spectral response of developed green leaflets (Formaggio e Sanches, 2017) can be observed in Figure 1, for March 9th, at 102 days after plant emergence (102 DAE). The reflectance and transmittance were similar, but with lower values in the visible spectrum and higher in the near infrared, compared to the absorbance, which presented an inverse pattern. In the visible portion of the electromagnetic spectrum, high absorption is a consequence of the action of foliar pigments, especially chlorophylls, whose absorption peaks of chlorophyll a in ether occur in 430 and 660nm and of chlorophyll b, with peaks at 453 and 643nm in length wave (Hall e Rao, 1980). In this same experiment, Almeida (2008) found a coefficient of determination of -0.99 and -0.98 between red reflectance



and chlorophyll content a and b, respectively. The peak of reflectance occurred around 500nm, corresponding to the green region, which explains the green coloration of the plants.

In this typical pattern, however, some differences were observed in the partitioning of radiation incident on soybean leaflets, throughout the crop cycle, and among the applied treatments. The partitioning of the incident radiation at three representative moments: vegetative growth (41 DAE), grain filling initiation (102 DAE), and end of cycle (139 DAE) is shown in Figures 2 and 3. In general, at the beginning of the crop cycle (41 DAE), young leaves with small thickness were characterized, by intermediate values of

reflectance, transmittance and absorbance in the visible portion of the electromagnetic spectrum, in comparison to the other measurement days. At the beginning of grain filling (102 DAE), with well structured leaflets, the reflectance and the transmittance decreased, and the absorbance had the largest magnitude. In the end of cycle (139 DAE), with a consequent beginning of the collapse of cellular structures, a pattern similar to the beginning of the cycle was observed, but with a great increase in reflectance.

By analyzing separately the component reflectance (Figures 2 and 3, A and B), a similar pattern of variation over time is observed in the visible portion of the four treatments. The highest values were observed at the end of

Figure 2. Reflectance (A and B), transmittance (C and D) e a absorbance (E and F) in soybean leaflets at 41, 102 e 139 days after emergence (DAE) for no-tillage non-irrigated treatment (A, C and E) and for no-tillage irrigated treatment (B, D and F). EEA-UFRGS, Eldorado do Sul - RS, Brazil, 2003/04.

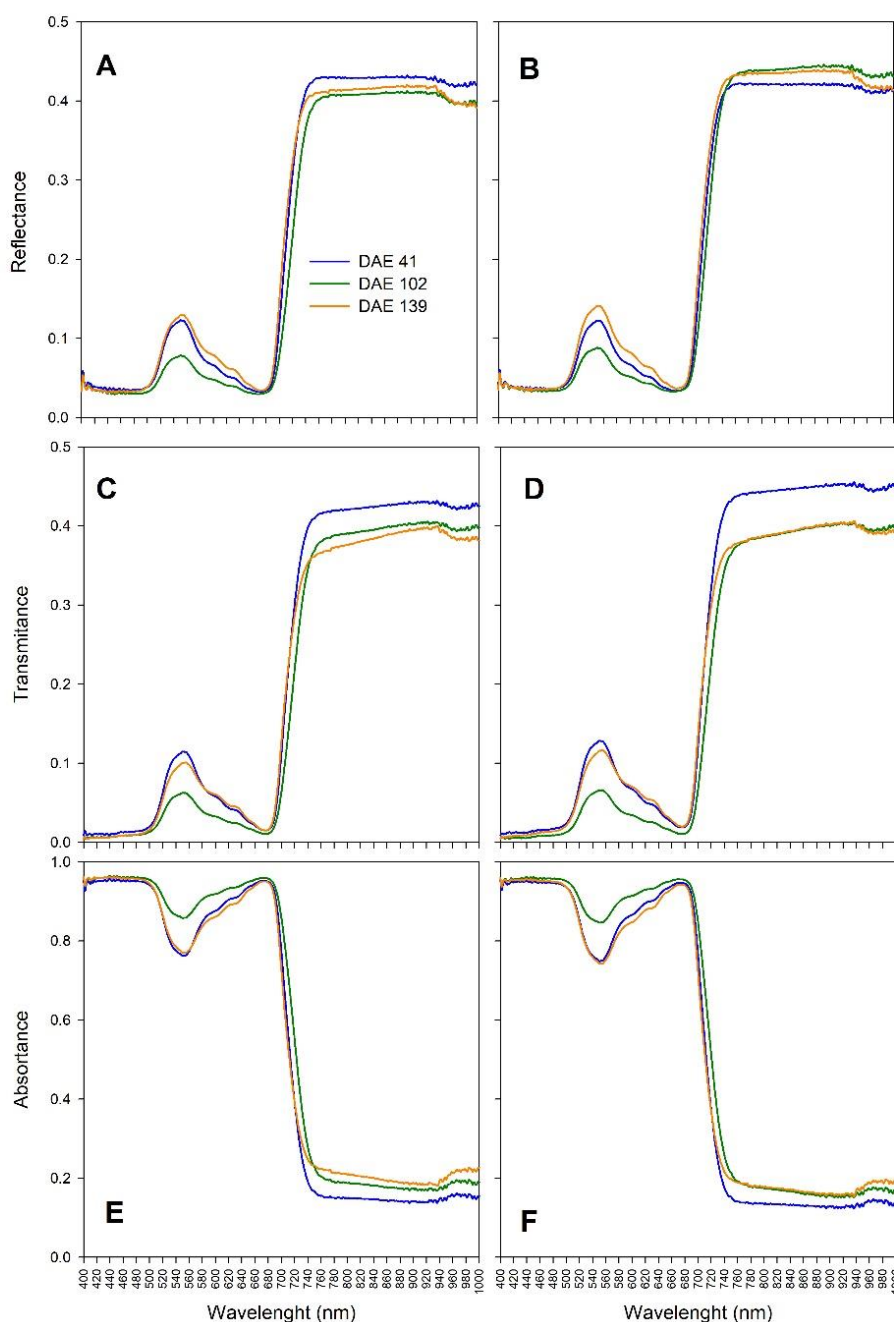
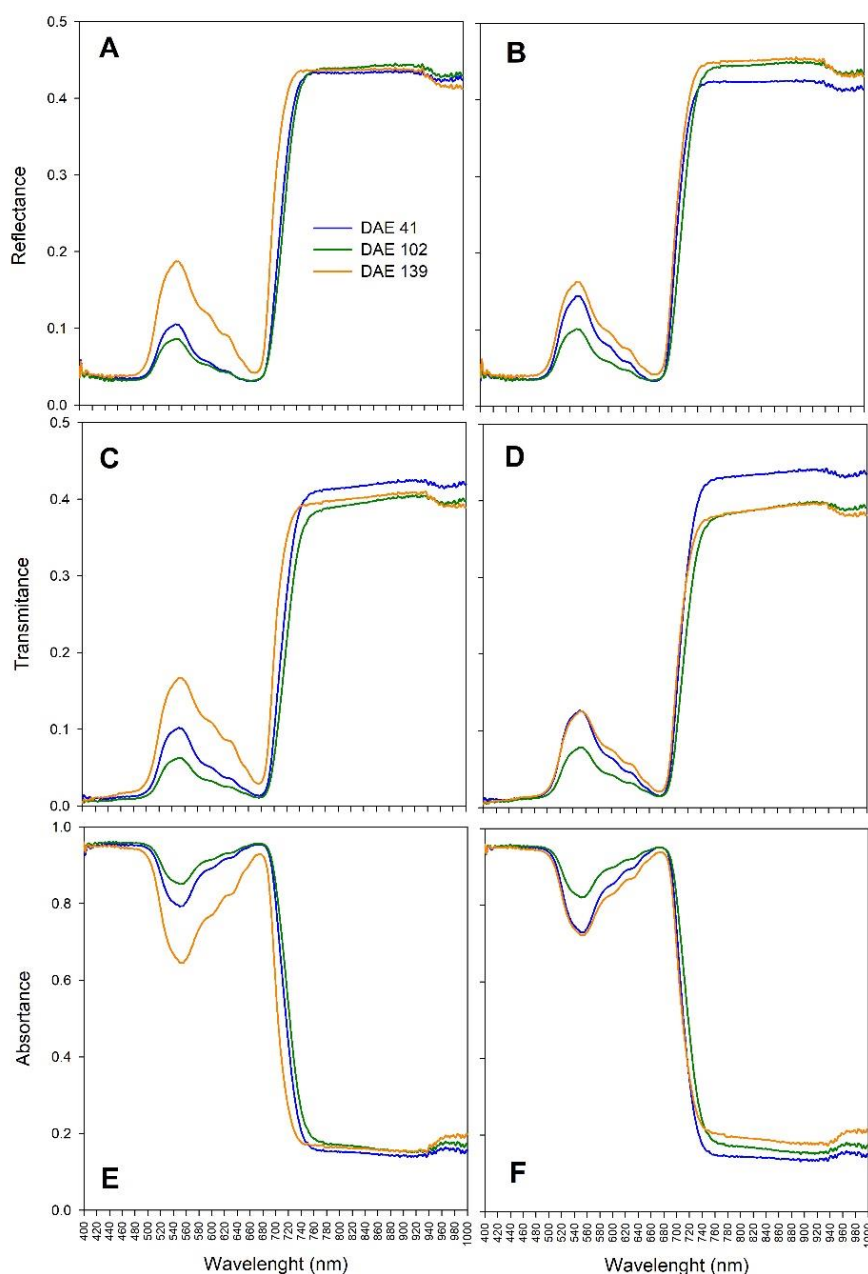


Figure 3. Reflectance (A and B), transmittance (C e D) and absorbance (E and F) in soybean leaflets at 41, 102 e 139 days after emergence (DAE) for conventional tillage non-irrigated treatment (A, C and E) and for conventional tillage irrigated treatment (B, D and F). EEA-UFRGS, Eldorado do Sul – RS, Brazil, 2003/04.



the crop cycle (139 DAE), while the lowest values occurred at the beginning of grain filling (102 DAE). Despite this similarity in terms of temporal pattern, there were differences in the reflectance magnitude between the treatments. In the conventional tillage system (Figure 3A and B), the variations between dates were higher than those observed in no-tillage (Figure 2A and B) and even more accentuated in the conventional tillage without irrigation (Figures 3A).

When the analyzed component was the transmittance (Figures 2 and 3, C e D), also in the visible spectrum, it is verified that the lowest values occurred at the beginning of grain filling (102 DAE), in all the treatments. Probably, the lower transmittance was associated to variations in leaflet

pilosity or accessory pigment levels, such as xanthophylls and carotenoids. This is only a hypothesis, since no measurements have been made at the time to prove it. But the most significant changes did not occur in the blue or red portion, where chlorophyll modulates the response, but in the green portion of the spectrum. On the other hand, the highest transmittance values had a variable response, depending on the treatment. In the no-tillage treatments (Figures 2 C and D), the highest transmittances were observed in vegetative growth (41 DAE), while the conventional tillage treatments (Figures 3 C and D) had the highest transmittance values at the end of the cycle (139 DAE).

In the visible spectrum, the absorbance was higher (Figures 2 and 3, E and F) at the beginning of grain filling (102 DAE). The lowest values occurred at the end of the crop cycle (139 DAE), in particular in the conventional tillage system. According to Valeriano (2003), leaf senescence and mineral malnutrition may reduce the chlorophyll content, allowing the increase of accessory pigments (carotenes and xanthophylls) previously masked, in a phenomenon called chlorosis.

The differences observed in the reflectance, transmittance, and absorbance of the soybean leaflets were probably caused by different conditions to which the plants were submitted, throughout the cycle, as a consequence of the treatments used (Martorano, 2007). However, it is important to point out, once again, that if measurements were taken on canopies rather than on individual leaflets, the differences between treatments would be better evidenced, especially in infrared portion, expressing variables associated with their structure, as demonstrated by Almeida (2008).

In the infrared portion of the spectrum, differences between dates and between treatments were smaller for the three components compared to those in visible portion. The greatest differentiation occurred for transmittance at 41 DAE.

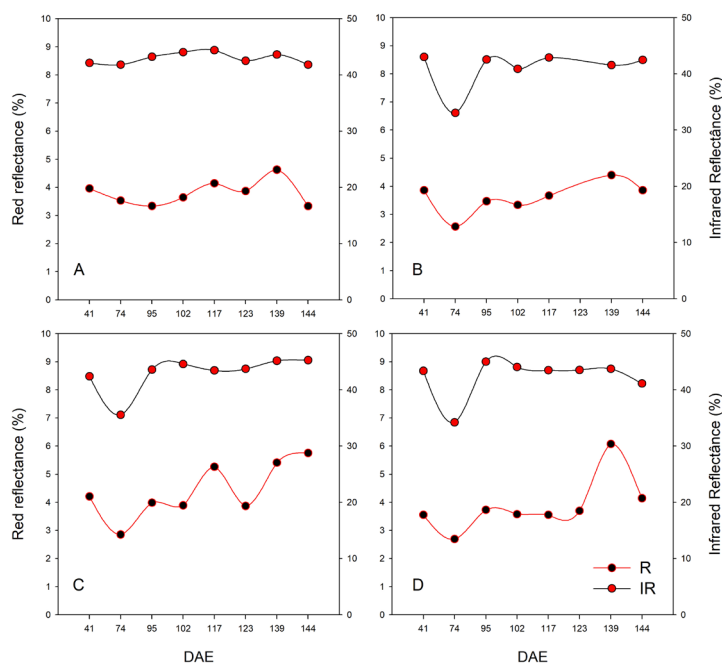
In the subsequent analysis, the reflectance pattern was characterized for the visible and infrared portions, along the crop cycle (Figure 4). This analysis is justified by the use of these regions of the radiation spectrum in calculating several vegetation indices, for different types of vegetation cover, such as *Paspalum notatum* (Fonseca et al., 2002), soybeans (Monteiro et al., 2012), rapeseeds (Pinto et al., 2017), grapevines (Junges et al., 2017), among

others, and their use in biomass estimation models.

Low values of reflectance were observed in the red portion of the spectrum, while high values were observed in the infrared portion, as expected (Formaggio e Sanches, 2017). In the visible portion of the electromagnetic spectrum, reflectance was around 4%, with small fluctuations over the evaluation period. These variations can be attributed to water restrictions, even in irrigated treatments, since there were some temporary problems in the application of irrigation treatments, as described by Martorano (2007). Also, reductions in reflectance values in the red portion of the electromagnetic spectrum can be attributed to the finalization of the plant cycle, as observed in the 144 DAE, in three of the treatments (Figure 4 A, B, and D). At this time, the plants were already in the end of cycle and, consequently, their leaflets already showed some degree of tissue disruption, altering their spectral response (Formaggio e Sanches, 2017).

Except for the others treatments, in conventional tillage without irrigation (Figure 4 C) the crop presented high values of reflectance, since the plants still had some green leaves, even at the end of the cycle. Periods of significant water deficits were observed during the experimental period. In this way, plants that did not receive irrigation were submitted to severe water stress, compromising the formation of grains. This caused foliar retention at the end of the crop cycle, which is a typical reaction of soybean plants to the lack of grains and pods, which can drain reserves from the vegetative tissues. Plants under no-tillage system, whose soil may conserve more water than in tilled soil, even without receiving irrigation, did not present this reaction. According to Martorano (2007), this different response reinforces the premise that plants in a conven-

Figura 4. Reflectance in soybean leaflets in Red and Infrared portions related to days after emergence (DAE). A, B, C and D represents treatments no-tillage non-irrigated, no-tillage irrigated, conventional tillage non-irrigated and e conventional tillage irrigated, respectively. EEA-UFRGS, Eldorado do Sul – RS, Brazil, 2004.



tional system tend to maintain leaves longer at the end of the cycle, while no-till plants lose leaves, draining their reserves for formation of grains.

In the infrared portion of the spectrum, a similar stability was observed in values of reflectance along the crop cycle, but higher than those observed in the red portion, which is characteristic of the spectral response of the vegetation and unique in relation to other natural targets (Jensen, 2009). Very often, the infrared portion of the spectrum is used to monitor plant growth, given its sensitivity in detecting the number of layers of leaves, which is explained by the additive reflectance phenomenon (Valeriano, 2003). Although the stability found in the present study appears inconsistent, it is due to the measurements being made on individual leaflets. On the same measurement days, near-infrared reflectance evidenced differences in growth, caused by the different treatments. Especially on days 102 and 139, the infrared reflectance measured on the canopies was much higher than that observed on the leaflets (Almeida, 2008).

Finally, the correlation analysis showed an inversely proportional relationship between values of reflectance and absorbance, for the red and infrared portions of the electromagnetic spectrum (Table 1). The coefficients were highly significant in both treatments (no-tillage and conventional tillage), all above -0.94. The absorbance is the parameter of interest, since it comes from the absorbed energy that plants perform photosynthesis (Hall e Rao, 1980). However, because the absorbance is not measurable with remote sensors, there remains the search for parameters that can be obtained by remote sensors, such as reflectance. Therefore, given the high correlation between absorbance and reflectance, it is consistent to use reflectance measurements in crop monitoring.

Conclusions

The radiation partitioning pattern on soybean leaflets, in the reflectance, transmittance and absorbance components, is influenced by the crop phenological stage and type of management applied to plants. More significant differences are observed in the visible portion of the spectrum and in the treatment of non-irrigated conventional tillage. At the beginning of grain filling the absorbance is higher, with lower reflectance and transmittance, in relation to the beginning and end of the cycle.

Despite this, there is stability of reflectance in the red and infrared portions, throughout the crop cycle, which shows that the greatest variations in the spectral responses of this crop are associated to the structure and arrangement of the canopy and not to changes in properties of individual leaflets.

Table 1. Correlation coefficients between Red (R) and Infrared (IR) reflectances and absorbances for soybean in conventional tillage and no-tillage treatments. EEA-UFRGS, Eldorado do Sul – RS, Brasil, 2004.

Treatment	Parameter	Absorbance R	Absorbance IR
Conventional tillage	Reflectance R	-0.97*	
	Reflectance IR		-0.97*
No-tillage	Reflectance R	-0.98*	
	Reflectance IR		-0.99*
Both	Reflectance R	-0.94*	
	Reflectance IR		-0.94*

* significant difference at 1% level of significance.

The inversely proportional relation between absorbance and reflectance of soybean leaflets evidences the viability of the use of reflectance measurements in agricultural crop monitoring.

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Respostas espectrais de folíolos de soja a diferentes condições hídricas e de preparo do solo

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RESUMO

O estado do Rio Grande do Sul é um dos maiores produtores brasileiros de soja, o que justifica o uso de técnicas de sensoriamento remoto para monitorar áreas ocupadas por essa cultura. O objetivo deste trabalho foi caracterizar a variabilidade ao longo do ciclo das respostas espectrais de folíolos de soja, submetidos a diferentes condições de preparo do solo e suprimento de água. O experimento foi conduzido em uma área de 0,5 ha, no município de Eldorado do Sul, no sul do Brasil, em dois sistemas de preparo do solo (plantio direto e preparo convencional) e dois níveis de suprimento de água (irrigado e não irrigado). A cultivar Fepagro RS-10 foi semeada com espaçamento de 0,40 m e população de 400.000 plantas por hectare. Utilizou-se uma esfera integradora de um espectrorradiômetro LI-COR, modelo LI-1800, para medir a absortância, refletância e transmitância em folíolos de soja. Os resultados mostraram que o padrão de repartição da radiação incidente nos componentes de refletância, transmitância e absortância é influenciado pelo estágio fenológico da cultura e pelo sistema de preparo do solo. Apesar disso, há estabilidade na refletância de folíolos de soja nas porções vermelha e infravermelha do espectro eletromagnético, ao longo do ciclo da cultura. A relação inversamente proporcional entre absortância e refletância de folíolos de soja revelou viabilidade nos dados de refletância, no monitoramento de culturas agrícolas.

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