

Leaf reflectance response to deficit irrigation in olive trees

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Abstract

Multispectral signatures from leaves at different spectral bands are increasingly used for characterize plants and their response to biotic and abiotic environmental factors. As the increase of water scarcity and consequent adoption of deficit irrigation treatments, this type of analysis can be useful for monitoring the water status of the plants. Therefore, the aim of this work is to assess the influence of different irrigation treatments on reflectance to VIS-NIR radiation of olive leaves (*Olea europaea* L, 'Cobrançosa'). The study was conducted in an olive orchard located in Alfândega da Fé (northeast of Portugal), under five irrigation treatments: full-irrigated (FI) with 100% of estimated Evapotranspiration (ET), sustained deficit irrigation (SDI60 and SDI30 of ET) and regulated deficit irrigation (RDI100 and RDI60 of ET). Reflectance (RVIS-NIR) was evaluated using a near infrared spectroscopy (400-1000 nm) in 15 leaves from five random olive trees of each irrigation strategy. The measurements were performed in the middle of June, before irrigation cut off; in cut off irrigation period (OFF; during pit hardening); after shift irrigation (third week of August) and in late irrigation period (LI; early autumn). The results showed that in all sampling dates, leaves from FI had lower RVIS-NIR than deficit irrigation treatments with differences found in VIS spectrum (540-590nm) and in the near infrared spectrum (740-820 nm). However, no significant differences were observed in radiation wavelength absorbed by photosynthetic pigments (420-500 and 610-720 nm), except in OFF and LI periods in which SDI30 and RDI60 had higher RVIS-NIR at 610-720 nm. The most discriminatory reflectance difference between FI and deficit irrigations was verified in SDI30 and RDI at OFF and LI irrigation, where SDI30 had an increase of 28% at 760 nm when compared with FI values. In these periods, the relative water content (RWC) was 92, 86, 76, 85 and 70% for FI, SDI60, SDI30, RDI100 and RDI60 respectively. We also observed a relationship between mean reflectance at different wavelength with RWC and Ψ_{MD} , especially in the mean green reflectance. Thus, multispectral reflectance appears as useful and potential strategy to monitor water status and a promising tool for irrigation management.

Keywords: irrigation management, precision agriculture, spectroscopy

INTRODUCTION

Water is an essential natural resource for maintenance of adequate food supply and a productive environment for the human population. As human population grow, global freshwater demand has been increasing rapidly (Gleick, 2003). However, due to global warming that increasingly triggered drought and long seasons, this natural resource faces the scarcity and has a strong impact on the agriculture and food production (Porkka et al., 2016). Thus, sustainable strategies to increase crop water productivity are gaining importance in arid and semi-arid regions, where, in particular case of olive orchards, the irrigation has a large impact on the productivity (Moriani et al., 2003; Fernandes-Silva et al., 2010).

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In order to increase water productivity, many researchers recommended the adoption of deficit irrigation treatments (DI) in different crops such as maize, vineyard, almond trees, apricot trees and olive trees (Domínguez et al., 2012; Fernandes-Silva et al., 2010; Lipan et al., 2019; Pérez-Pastor et al., 2009). DI is a water-saving strategy that allows to preserve water resources minimizing the impact on yield reduction, in which crops are exposed to a certain level of water stress, either during a particular period or throughout the whole growing season (English, 1990). In particular case of olive trees in the region of “Terra Quente Transmontana” Fernandes-Silva et al. (2010) concluded that this strategy had a very beneficial effect compared to rainfed conditions, with a yield reduction only 25% compared to Full-Irrigation, while saving 60% of applied water. However, the adoption of DI implies appropriate knowledge of crop evapotranspiration (ET), crop response to water deficit and economic impacts of yield reduction (Pereira et al., 2002). Thus, it is extremely crucial to monitor the crop under this strategy and to analyze its response through water status indicators (Fernandes-Silva et al., 2016) such as relative water content (RWC) and leaf water potential (LWP), being these methods laborious, time-consuming and destructive.

Therefore, new methodologies are emerging, such as spectral reflectance, which showed its potential applicability in different crops. Some authors have investigated the potential use of aerial thermal infrared imagery retrieved from Unmanned Aerial Vehicles (UAV) for water stress monitoring in different crops such as vineyard (Bellvert et al., 2016), palm trees (Cohen et al., 2012) and peach trees (Wang and Gartung, 2010). However, this type of tool has a high associated cost, it is dependent from the atmospheric conditions and requires an operator with experience to control the UAV. Therefore, the aim of this study is to analyze the feasibility of using spectroscopy to extract leaf $R_{VIS-NIR}$ in order to evaluate water status of olive trees under different irrigation treatments.

MATERIAL AND METHODS

Study area description

This study was carried out in a commercial olive orchard (*Olea europaea* L., ‘Cobrançosa’) located at Alfândega da Fé, (41°19’54.4”N; 7°02’33.6”W; altitude: 243 m) a typical olive growing area in the northeast of Portugal. The olive orchard area is ~2 ha with olive trees spacing 6×6 m apart, drip irrigated, and was submitted to five irrigation treatments: full-irrigated (FI) with 100% of estimated evapotranspiration (ET), sustained deficit irrigation (SDI) 60 and 30% of FI, and regulated deficit irrigation (RDI) 100 and 60% of FI (Figure 1). While SDI is a continuous deficit throughout irrigation season, in RDI watering was interrupted during the fruit pit hardening (~1 month). Crop water requirements was estimated according the approach described in Fernandes-Silva et al. (2010).

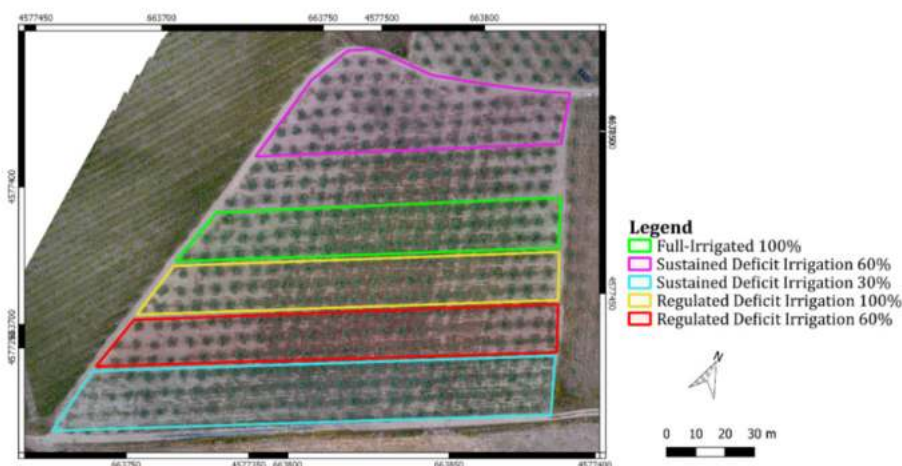


Figure 1. Aerial overview of the olive orchard with the defined irrigation treatments.

Data acquisition and analysis

For the leaf $R_{\text{VIS-NIR}}$ extraction, three fully expanded and healthy leaves of the year from five random olive trees, located in the central line of each irrigation treatment, were detached and putted into a plastic bag, which was sealed and placed in a cold handled container and transported to the laboratory. $R_{\text{VIS-NIR}}$ was measured by the infrared spectroscopy Ocean Optics HR4000 (Ocean Optics, Rostock, Germany) in the wavelength range from 400 to 1000 nm, being the sensor placed in the central part of the leaf next to the main vein. To assign one multispectral signature per irrigation treatment, it was calculated the mean reflectance (400 to 1000 nm) of the 15 leaves.

Measurements of midday shoot water potential (Ψ_{MD}) and relative water content (RWC) were used to evaluate tree water status. For the Ψ_{MD} measurement, a young leafy shoot was collected, from sunny position at the crown, from three olive trees per irrigation treatment. After cutting, the small leafy shoot was immediately enclosed in a plastic bag to avoid any loss of water and quickly placed into the pressure chamber (model PMS 1000, Oregon, Corvallis, USA).

Concerning RWC measurements, for each selected tree, three leaves of the year were removed and placed in a glass tube, which was sealed, placed in a cold container and transported to the laboratory. The sample was weighed on a precision balance to obtain the fresh mass (FM). Afterwards, cold distilled water was put into the glass and stored in the dark at 4°C during 48 h, and, after that, the leaves were again weighed to obtain the turgid mass (TM). Finally, the leaves were placed in a ventilated oven-drying at approximately 70°C until 48 h and weighed again and dry mass (DM) was obtained. The RWC was calculated as shown in Equation 1.

$$RWC = 100 \times \frac{(FM-DM)}{(TM-DM)} \quad (1)$$

Temporal behavior of leaf $R_{\text{VIS-NIR}}$ and water status indicators in response to water deficit along irrigation season was evaluated in four dates: June 14th (DOY 165) – before irrigation cut off in RDI_{100} and RDI_{60} ; August 20th (DOY 232) – during cut off irrigation period (OFF; during pit hardening) in the previous irrigation treatments; August 28th (DOY 240) – after shift irrigation; and October 11th (DOY 284) – late irrigation period (LI; early Autumn).

The statistical treatment of the data (ANOVA and Tukey HSD test) was performed with the SPSS program (IBM SPSS Statistics 26).

RESULTS AND DISCUSSION

The results of plant water status (Ψ_{MD} and RWC) during the irrigation season are shown in Table 1. On August 28th (DOY 240) no Ψ_{MD} measurements were made due to logistic reasons.

On DOY 165 (June 14th), all treatments showed values of RWC higher than 90% and no differences were observed in Ψ_{MD} . According to Fernandes-Silva et al. (2016), the olive trees were in well-watered conditions. The mean values between FI and deficit treatments were compared using Tukey test, and on this date, for both RWC and Ψ_{MD} slight differences were found but they were not statistically significant ($p > 0.9$). This homogeneity is probably related with a rainfall event (11 mm) that occurred in the week before the data collection. However, three weeks (August 20th, DOY 232) after irrigation cut off in RDI_{60} and reduced to 5% in RDI_{100} , the water stress indicators (Table 1), showed significant differences between irrigation treatments, in which SDI_{30} and RDI_{60} had the lowest values of RWC and Ψ_{MD} . The low values of RWC (70%) and Ψ_{MD} (-6.2 MPa) observed in RDI_{60} indicate that the plants experienced a severe water stress (Fernandes-Silva et al., 2016).

Table 1. Mean values of midday shoot water potential (Ψ_{MD}) and relative water content (RWC) for full irrigation (FI), sustained deficit irrigations (SDI₃₀ and SDI₆₀) and regulated deficit irrigations (RDI₁₀₀ and RDI₆₀) treatments during the irrigation season ($n=3$).

Irrigation treatment	June 14 th		August 20 th		August 28 th		October 11 th	
	RWC (%)	Ψ_{MD} (MPa)	RWC (%)	Ψ_{MD} (MPa)	RWC (%)	Ψ_{MD} (MPa)	RWC (%)	Ψ_{MD} (MPa)
FI	94±1.5	-2.2±0.2	92±1.8	-2.4±0.3	92±1.9	-	89±1.7	-2.8±0.2
SDI ₆₀	93±2.6	-2.3±0.1	86±0.8	-3.4±0.1	89±2.3	-	77±1.6	-5.0±0.3
SDI ₃₀	91±1.5	-2.6±0.3	76±1.1	-4.8±0.5	85±4.4	-	62±2.1	-5.9±0.2
RDI ₁₀₀	93±1.1	-2.1±0.2	85±2.6	-3.9±0.8	90±1.8	-	84±3.1	-3.5±0.3
RDI ₆₀	93±1.4	-2.4±0.1	70±3.8	-6.2±0.3	89±2.4	-	72±1.7	-4.9±0.4

On DOY 232 (August 20th), discriminatory differences were found between FI and others treatments. Regarding SDI₃₀ and RDI₆₀ treatments, the mean values of RWC ($p<0.05$) and Ψ_{MD} ($p<0.01$) were statistically different from those observed on DOY 165 (June 14th). According to Fernandes-Silva et al. (2016), the plants from the two deficit treatments were in severe water stress with RWC values as low as 76 and 70% and Ψ_{MD} of -4.8 and -6.2 MPa respectively. Concerning SDI₆₀ and RDI₁₀₀, the mean values of RWC were 86 and 85% and the mean values of Ψ_{MD} were -3.4 and -3.9 MPa, respectively, thus, showing a slight water stress (Fernandes-Silva et al., 2016).

On DOY 240 (August 28th), it was possible to verify a recovery of water status, which can be related with a rainfall event (14 mm) that occurred two days before. As verified on DOY 165 (June 14th), there was no statistical difference between FI and deficit treatments ($p>0.3$). However, on DOY 284 (October 11th), all deficit irrigation treatments were statistically different from FI (RWC with $p<0.05$ and for Ψ_{MD} $p<0.01$). The intensity of the water stress was increased in all deficit irrigation treatments, where, SDI₃₀ showed low values as 62% for RWC and -5.9 MPa for Ψ_{MD} .

Moreover, linear correlations between mean leaf $R_{VIS-NIR}$ at different wavelengths and water status indicators were performed, with exception of August 28th (due to logistic reasons). In the next sections, the main conclusions and correlations obtained on the different dates are discussed.

Before irrigation cut of (June 14th)

As previously described, on this date, no differences were observed in the water status in all treatments, which caused low correlations between mean $R_{VIS-NIR}$ in different wavelength with water status indicators. Moreover, the maximum reflectance for green (570 nm) and near infrared wavelength (800 nm) was 10 and 70%, respectively (Figure 2).

Cut off irrigation period (August 20th)

On DOY 232, considerable differences were pronounced in the green, red and near infrared wavelength, being more discriminatory between FI, SDI₃₀ and RDI₆₀ irrigation treatments (Figure 3). Although the mean blue reflectance did not show statistically difference between FI and deficit treatments ($p>0.9$), the mean green reflectance of FI showed differences with SDI₃₀, RDI₁₀₀ and RDI₆₀ ($p<0.01$). Moreover, the mean red reflectance was only statistical different in SDI₃₀, whereas, the mean near infrared reflectance (NIR) was discriminatory between FI, SDI₃₀ and RDI₆₀ ($p<0.01$). Furthermore, in the green and NIR wavelength, it was possible to verify that FI had the lowest maximum reflectance, 14 and 70%, respectively, while SDI₃₀ and RDI₆₀ showed the highest peak (20% for green and 76% for NIR). Thus, SDI₃₀ and RDI₆₀ reveals an increase of 43 and 8.5% in the green and NIR spectrum respectively, when compared to FI reflectance values.

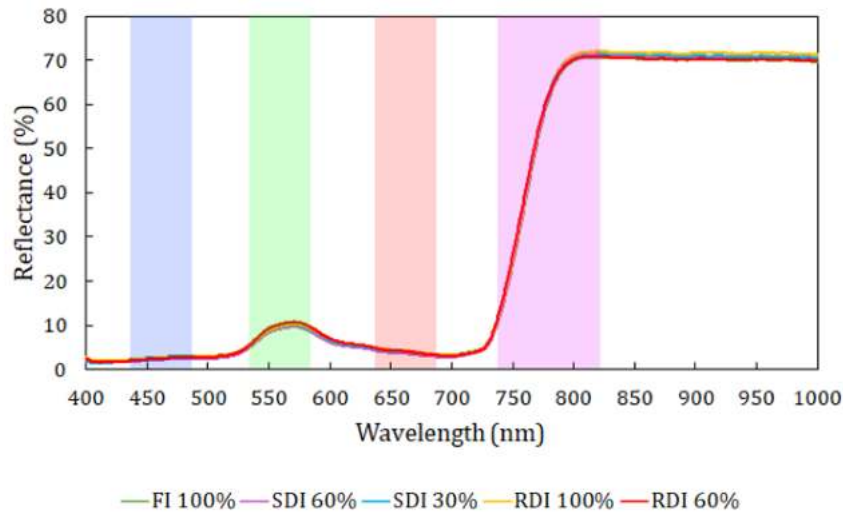


Figure 2. Mean leaf reflectance in different irrigation treatments in June 14th, day of the year 166. FI = control; SDI = sustained deficit irrigation and RDI = regulated deficit irrigation.

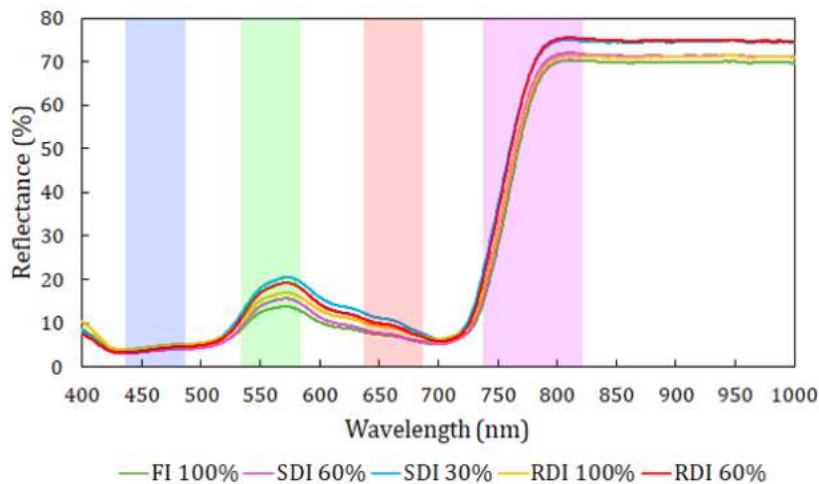


Figure 3. Mean leaf reflectance in different irrigation treatments in August 20th, day of the year 232. FI = control; SDI = sustained deficit irrigation and RDI = regulated deficit irrigation.

Comparing these results with the previous data, it was observed that all irrigation treatments had a $R_{VIS-NIR}$ increase of 40, 60, 100, 70 and 90% for FI, SDI₆₀, SDI₃₀, RDI₁₀₀ and RDI₆₀, respectively, in the 570 nm. Regarding to the peak of reflectance in NIR wavelength (800 nm), no changes were observed in FI and neglecting in RDI₁₀₀ (≈ 1), while in deficit irrigation treatments was observed an increase of 3, 7 and 9% in SDI₆₀, SDI₃₀, and RDI₆₀, respectively. This behavior is consistent to the high differences observed in plant water status indicators (Table 1). Therefore, it is possible to state that the more the plants are stressed, the higher is the leaf $R_{VIS-NIR}$. Thus, it seems that the leaf pigments that absorb radiation in VIS region were more affected by water stress than the leaf structure that absorbs in NIR regions which corroborates with Marino et al. (2014).

Furthermore, correlations were made between the mean green, red and near infrared

reflectance with RWC and Ψ_{MD} where some relationships were found (Figure 4). However, as the green wavelength was more affected by the water stress (Figure 3), it reached the highest correlation for RWC and Ψ_{MD} when compared with the other wavelengths. Thus, the results suggest that this region consists of the most sensitive water stress spectrum.

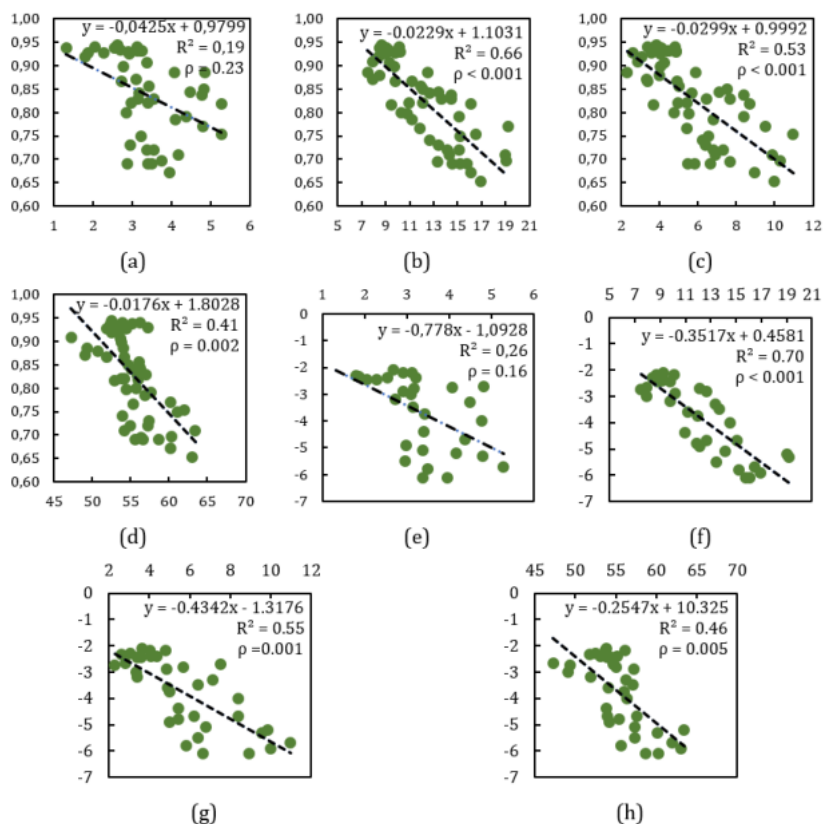


Figure 4. Relationship between mean reflectance and water status indicators: (a) blue vs. RWC; (b) green vs. RWC; (c) red vs. RWC; (d) near infrared vs. RWC; (e) blue vs. Ψ_{MD} ; (f) green vs. Ψ_{MD} ; (g) red vs. Ψ_{MD} ; (h) near infrared vs. Ψ_{MD} . $n=55$ and 35 for RWC and Ψ_{MD} .

After shift irrigation (August 28th)

A week after the reestablishment of the irrigation in RDI₁₀₀ and RDI₆₀ treatments, a recovery of water stress was observed as the values of RWC are close to 90% and very close to those observed in olive trees of FI. Surprisingly, a recovery of water stress was also observed in SDI₃₀ and SDI₆₀ which may be explained by an event of precipitation (14 mm) just two days before leaves collection. As a consequence of this recovery, it was observed that the mean reflectance values decreased in all irrigation treatments, reaching similar values to those recorded on June 14th (Figure 5). This homogeneity induced to the absence of statistical differences ($p > 0.3$) between FI and deficit treatments in the mean blue, green, red and near infrared values.

Late irrigation period (October 11th)

On this date (Figure 6), higher $R_{VIS-NIR}$ differences between irrigation treatments were again observed as in August 20th in the green, red and near infrared wavelength. While green and near infrared reflectance showed statistical differences ($p < 0.05$) between FI and deficit treatments, the mean red reflectance was only discriminatory between FI, SDI₃₀ and RDI₆₀ ($p < 0.01$). The lowest values were observed in FI treatment whether in the peak of the green (8%) and in the near infrared wavelength (69%). The highest reflectance was reached in SDI₃₀

treatment with 15 and 74% in the peak of the green and near infrared spectrum respectively. Surprisingly, leaves of the RDI₆₀ only showed similar reflectance to SDI₆₀ in the near infrared spectrum, while in the green spectrum, leaves showed values of reflectance similar to SDI₃₀.

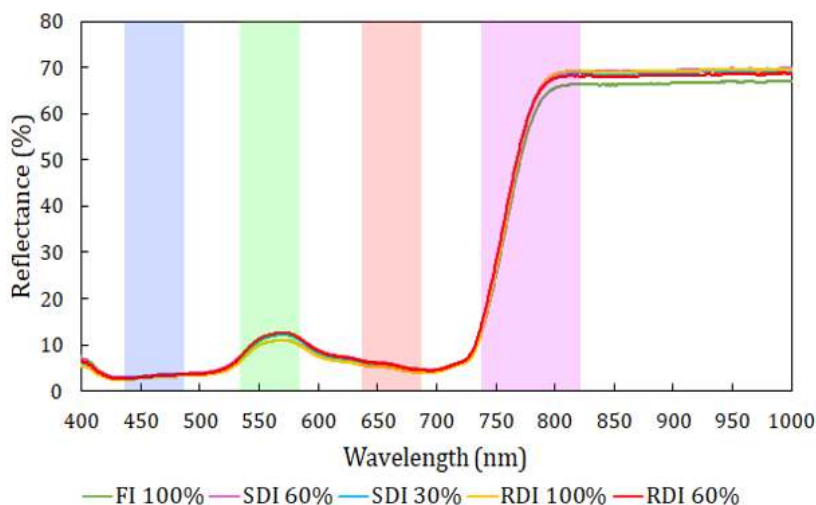


Figure 5. Mean leaf reflectance in different irrigation treatments in August 28th, day of the year 240. FI = control; SDI = sustained deficit irrigation and RDI = regulated deficit irrigation.

Temporal overview

In general, the results showed that in all sampling dates, leaves from FI had lower $R_{VIS-NIR}$ than deficit irrigation treatments, where, higher differences were found in VIS spectrum (540-590 nm) and in the near infrared spectrum (740-820 nm) as illustrated in Figures 3 and 6. This $R_{VIS-NIR}$ behavior was also observed by Rallo et al. (2014).

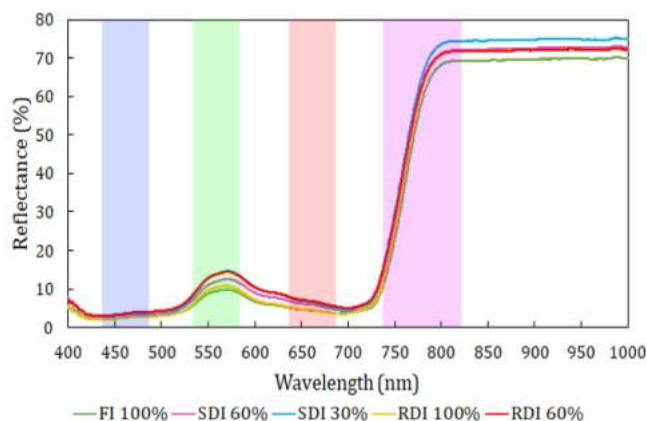


Figure 6. Mean leaf reflectance in different irrigation treatments in October 11th, day of the year 284. FI = control; SDI = sustained deficit irrigation and RDI = regulated deficit irrigation.

Concerning the correlations between mean red, green and near infrared reflectance with water status indicators, some relationships were found (Figure 6). When combining the data from all sampling dates, the mean green reflectance had the highest correlation with water status indicators, with $r^2=0.66$ and $r^2=0.7$ for RWC and Ψ_{MD} , respectively.

Between the mean $R_{VIS-NIR}$ analysis, the R_{NIR} had the lowest correlation with RWC and Ψ_{MD} , corroborating this way the conclusion achieved in particular dates of August 20th and October 11th, where leaf pigments (400-700 nm) were very sensitive to water stress.

CONCLUSIONS

In this study, we analyzed the leaf $R_{VIS-NIR}$ response to deficit irrigation in olive trees using a spectroscopy. In general, it was possible to verify that deficit irrigation treatments had higher $R_{VIS-NIR}$ values than the well irrigated (FI). The differences were more pronounced in the green and near infrared wavelength, indicating that the leaf photosynthetic pigments were very sensitive to water stress.

Among the analyzed dates, the differences between the deficit irrigation treatments and FI, were observed in the cut off irrigation period and in late irrigation period (October 11th), where, SDI_{30} and RDI_{60} irrigation treatments differed the most from FI. According to Ψ_{MD} and RWC values measured on these dates, these treatments had the lowest values, thus, presenting consistency with the leaf $R_{VIS-NIR}$ retrieved from the spectroscopy. Moreover, some correlations observed between mean $R_{VIS-NIR}$ at different wavelength with RWC and Ψ_{MD} , especially in mean green reflectance. On other hand, results indicated that leaf reflectance can detect a fast recovery of an olive leaf water status.

It was also possible to verify that the $R_{VIS-NIR}$ of the olive tree leaves is relatively sensitive to the rain, since its reflectance after this natural rain event has decreased. This behavior was also confirmed in water stress indicators such as Ψ_{MD} and RWC, since an increase in their values was registered when compared to previous dates.

Thus, the assessment of leaf $R_{VIS-NIR}$ by spectroscopy may be promising in estimating the leaf water status and appears to be a useful and potential tool for irrigation management.

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