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USING SPECTROMETRY TO MONITOR REACTIONS OF *CITRUS* × *LIMON* AT DIFFERENT TIMES OF IRRIGATION DURING WINTERIZING

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The aim of this work is to monitor changes of *Citrus* × *limon* leaves spectrometry at different times of irrigation and the exposure to stress caused by drought or excessive watering during winterizing. The changes were monitored at plants at different times of irrigation (daily, once in two days, once in three days, and once a week). The changes are monitored in the visible spectrum and compared with shed leaves in order to monitor changes in spectrometry, choose the correct time of irrigation and compare them with stress-exposed plants.

Keywords: spectrometry, irrigation, time, *Citrus* × *limon*, winterizing

Citrus × *limon* has been becoming a more and more popular plant of casual growers in Slovakia, and not only those focused exclusively on tropical or subtropical plants. The main reasons behind its cultivation are not only the aesthetic elements of the plant and its pleasant odour, but also the possibility to grow the plants at home and consume it, with no use of chemical treatments.

The main objective of the paper is to identify the most suitable interval between the times of irrigation based on the reactions of plants, measured by spectrometers. The times of irrigation need to be determined according to the plant's needs, and not only the soil humidity as the times and volume of irrigation cannot be set on general basis, on the contrary, it needs to take into account several different factors, such as air temperature, relative air humidity and properties of soil substrates.

Material and methods

Citrus × *limon*

Citrus are subtropical plants. It is very difficult to create one, universal handbook to citrus cultivation in Slovakia as there are various microclimatic conditions. To prevent from failure, it is necessary to know the environment and its conditions and to answer the question if the grower is able to provide the plant with suitable conditions. In natural climatic conditions, the citrus grow periodically and do not require dormancy.

Ideal growth conditions

Light

Citrus × *limon* requires intensive but dispersed light. When replacing from interior to exterior and vice versa, it is necessary that the plant could gradually adapt itself to the changed light conditions and does not skip growth period out of the light shock. It is convenient for the plants to be shaded by other plants, nets etc. In apartments, they need proper lightning, ideally by windows. They should be placed by south-eastern or south-western window, however, it would be ideal to place them at 1 meter distance from the interior glass.

Temperature

The *Citrus* × *limon* requirements change throughout a year:

- -3 to -5 °C critical temperature,
- 0 to 10 °C forced dormancy (November to February),
- less than 13 °C stopped growth,
- 14 to 20 °C blooming, fruits ripening, winterizing by light,
- 20 to 28 °C ideal growth in 2–3 growth periods (March – October),
- 30 to 38 °C slowed growth,
- over 38 °C stopped growth,
- over 50 °C critical temperature.

Under natural conditions, air temperature changes within 6 °C throughout a day. That is why if the plant is placed in a well ventilated room, it does not reach sufficient growth intakes through intensive nocturnal breathing of substances created by assimilation throughout a day. The temperature in apartments is usually suitable from spring to autumn, however, it is quite problematic in winter as the plant needs a bright room with the temperature of about 10 °C.

Water

The plant requires proper quality and volume of irrigation. The nutrients solved in water are most consumed during the period of intensive growth. Lower temperature means lower irrigation requirements. The plant should be watered when the substrate surface is drie dup, ideally in morning, with

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the volume of water of about 1/10 of its pot so that the 70% water saturation of substrate is reached. If water flows out to pot plant saucer, it should be poured off to prevent constant soaking of the substrate which could result in insufficient access to air and roots rotting.

Rain water or drinking water can be used for irrigation, however, it should stay at least one day or be cooked. Hard water from wells is not suitable. The water temperature should be 3–4 °C higher than air temperature. If roots are supercooled, the plant experiences shock, and if there is a rise in air temeprature at the same time, the plant will shed its leaves to prevent evaporation.

Air humidity

Citrus × *limon* requires proper air humidity. By 20 °C, the suitable air humidity is about 60%. Rising air temperature means lower air humidity and vice versa. Negative values are reached in so-called pannel apartmenrs with humidity of 20–30% during winter when heating up to 23 °C. The proper values are reached either by lower temperatures or by dewing (only temporarily).

Plant placement

Plant 0 is placed on a window sill oriented to south/southwest, which provides the plant with enough sunlight during the winter months. In the room there is a relatively stable temperature of 18–20 °C and a relatively high air humidity, which has not been measured but it has been monitored in form of dewing from the inner side from sunset to sunrise.

The capacity of the yellow ceramic pot is 3.5 dl in order to prevent the plant from overheating its roots through sunshine.

The plant itself is 27 cm high, with 4 principal branches with altogether 38 leaves before the start of measuring.

Field spectrometer

The ASD FieldSpec® HandHeld™ 2 is a portable spectroradiometer enabling quick, precise, non-destructive and contactless measuring. HandHeld 2 provides extremely precise and quick data based on reflectance and radiation in the whole range of the spectrum. It is equipped with a full-colour LCD display, showing calculations, it has got a great internal memory, aiming laser and it is GPS compatible. The device is portable, equipped with a battery so that it could be used both outside as well as under laboratory conditions (ASD, 2010).

Spectral data collected by HandHeld 2 might be imported into an external computer with HH2 Sync. Aplication, which is a software designed for HandHeld 2, setting the file title system and file exporting.

There are two ways the HandHeld 2 device manages and collects data

- Independent mode – the mode is used for internal control and ability to collect data
- Connecting mode – the mode combines the functions of HandHeld 2 with those of an external computer.

Methods

1. Selection of irrigation intervals with the volume of 10% of the total pot capacity. In case there is any water left at the bottom, it will be poured off:
    a) Every day.
    b) Every three days.
    c) Every five days.
    d) Once a week.
2. Daily data collection between 1–2 p.m. as there are the best ligh conditions then.
3. Data collection through the Field spectrometer HandHeld2:
    a) Test of light based on a white referential pannel, if reflectance of the spectre from the white referential pannel is too departed, optimization takes place.
    b) Measurement of current by dark conditions and the white reference.
    c) Regular updating and optimization in case the light conditions vary a lot.
    e) Measurement and saving the spectrum.
4. A set of spectral pictures is collected from each leaf, the ViewSpecPro programme selects a representative picture to represent the given leaf in the best possible way. Selection from several samples minimizes the probability of the picture's deformation.
5. A spectral area is identified in the samples, which is monitored – a green one in the visible spectrum, and parts of the yellow and red spectrum in order to monitor the leaf ageing.
6. In this measurement, also shed leaves are taken into account.
7. The spectrums are compared as well as changes in reflectance measured at different irrigation times. The samples from drought period (shed leaves) are also taken into comparison, also in the time respect.
8. The best irrigation interval is selected based on the spectrometry results.

Results and discussion

Measurements

In order to reach the planned objective of the spectroradiometry, the measurements on 10 selected leaves of the plant was carried out so that the leaves differed in age and the optical and physico-chemical properties could be provided (colour, brightness, growth imperfections, imperfections caused by pests). The data collection was conducted using the HandHeld 2 spectroradiometer, operating in the spectrum of 325–1,075 nm, with the sample collection interval of 1.5 nm.

The spectroradiometer was installed on a stand in order to preserve the distance of 30 cm between plants and the lens and to provide stable conditions regarding leaves illumination's angle. Before a measurement itself, we focused on the white referential area, and if there was significant ripple at the extreme values of the spectrums or the values were not approaching 1 (saturation), optimization was carried out; if the conditions were suitable and it was not necessary to optimize the device, the data collection of dark spectrum and white reference were conducted. Then, the data collection itself was performed, collecting samples from each leaf, analyzing them and selecting the most suitable ones. Measurement errors may have included unpredictable issues such as sudden shading, deformation of results

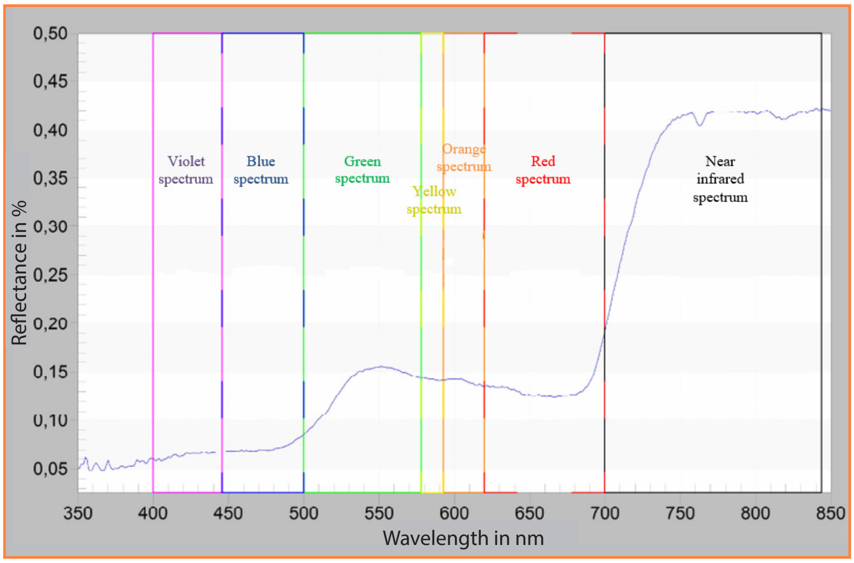

Figure 1 Graphic section of visible spectrum and close infrared spectrum
1 – Violet spectrum: 0.4–0.446 µm; 2 – Blue spectrum: 0.446–0.500 µm; 3 – Green spectrum: 0.500–0.578 µm; 4 – Yellow spectrum: 0.578–0.592 µm; 5 – Orange spectrum: 0.592–0.620 µm; 6 – Red spectrum: 0.620–0.7 µm

caused by laser radiation, movements of leaves, improper technique of sampling, or slight depositioning of the spectroradiometer (although on the stand), which may have taken a wrong sample of a leaf.

It is necessary to set proper reflective and transmittive spectra for leaves as a function of light absorption aided by chlorophylls, carotenoids, water, cellulose, proteins etc. (Newnham and Burt, 2001; Dangel et al. 2003). Each plant has got different spectral resolution based on its phenological cycle (Gouranga and Harsh 2005; McCloy 2010; Papadavid et al. 2011).

The domain of optical monitoring starts from 400 nm in visible spectrum and electromagnetic spectrum and ends with shortwave, infrared spectrum. Strong light absorption by photosynthetic pigments prevails in green spectrum (400–700). Leaf chlorosis causes increased reflection and transmission. NIR (700–1,100 nm) is a part of the spectrum where there is limited biochemical absorption and which is limited to compounds present particularly in dry leaves, sucha s cellulose, lignin and structural carbohydrates (Wang et al., 2005).

Blue, green and red spectra are considered the primary colours or wave lengths of the visible spectrum, because through their combination, all other colours of the visible spectrum can be composed. The visible spectra can be seen within spectra, when sunshine falls on the edge and defragmented into light with different properties based on the wave length.

The chemical compound present in leaves, chlorophyll, strongly absorbs red and blue wave length radiation, but reflects green wave length, that is why we consider leaves the greenest in summer, when there is the maximum of chlorophyll. In autumn, when the level of chlorophylls is lower, there is also lower absorption and greater reflection in the red spectrum, so that leaves or red or yellow (yellow is a combination of red and green wave lengths). The internal structure of healthy leaves is an excellent difusion reflector of close infrared wave length. If our eyes are sensitive to reflection of close infrared spectrum, we consider trees very bright under these wave lengths. Measurement and monitoring of NIR reflectance is one of the ways to assess the health status of vegetation.

Leaves of healthy *Citrus* × *limon* contain high levels of chlorophylls, which is responsible for their specific green colour. Therefore, there is only low reflection in blue and red part of spectra and great reflectance in the green part of the visible spectrum and NIR. If the leaves are under strain or damaged, the content of chlorophyll decreases together with changed structure of the leaves. Subsequently, this results in lower reflectance in the green part of spectrum and NIR. These reactions provide early warning, stating plant stress. The rate of the reflected NIR to red spectrum is an excellent index of vegetation health, known as the NDVI (normalized differential vegetation index). The index has high values for healthy plants because of high reflectance in NIR and low one in red spectrum. It is possible to set stress conditions visually, based on the basic shades of green colour. Properly irrigated plants are brighter; if there is lack of water, leaves become darker.

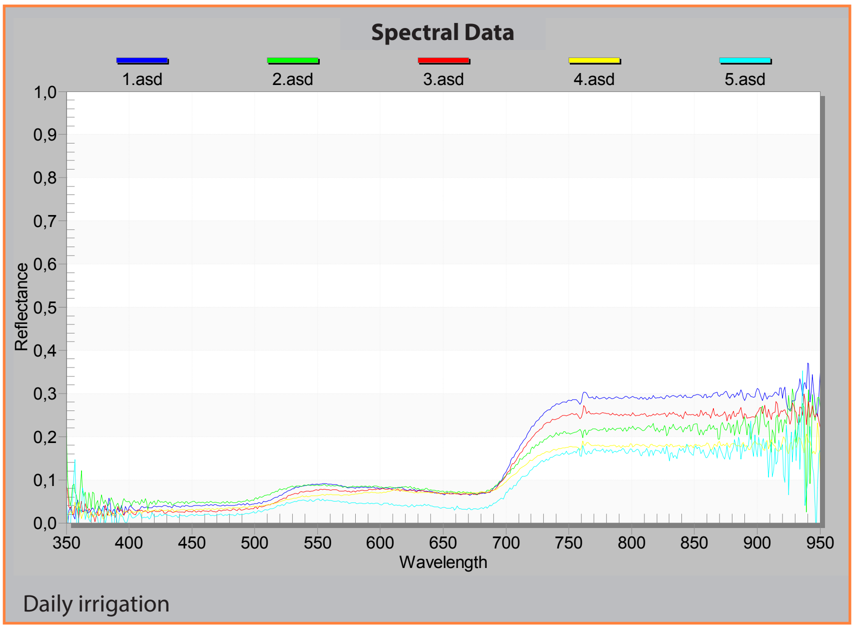

Figure 2 Graphic section of visible spectrum and close infrared spectrum of daily irrigation for 5 consecutive days

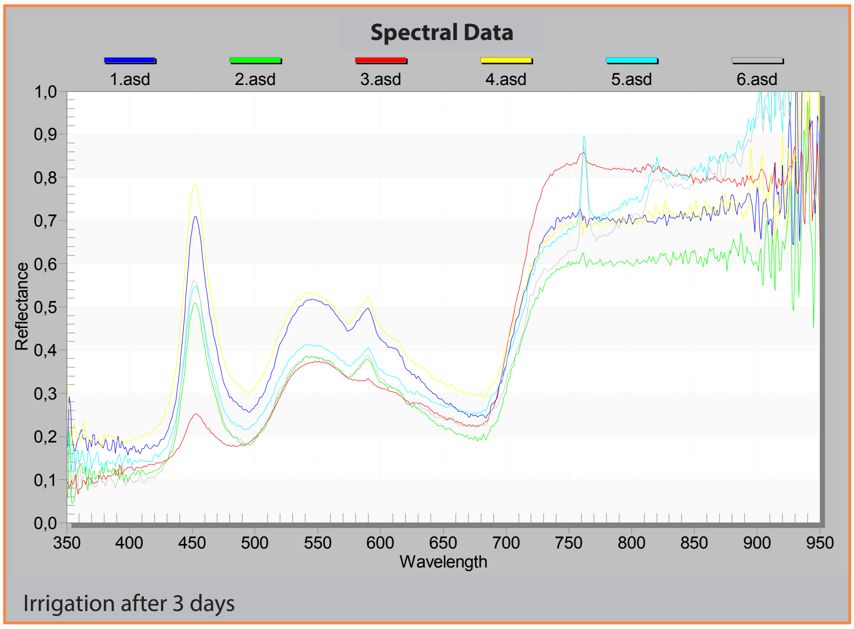

Figure 3 Graphic section of visible spectrum and close infrared spectrum of irrigation for once in three days

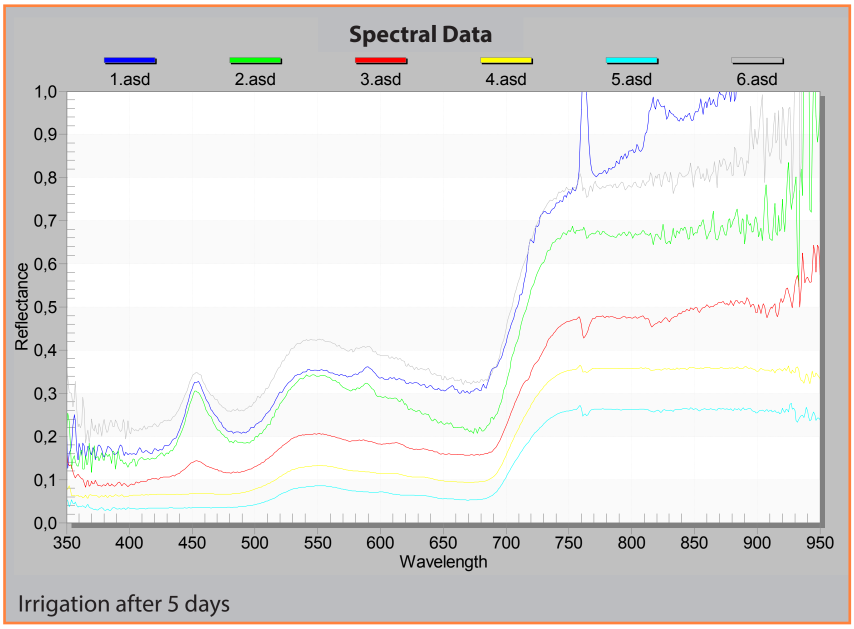

Figure 4 Graphic section of visible spectrum and close infrared spectrum of irrigation for once in five days

Spectrometry data

The spectrometry data were always collected 24 hours after watering, i.e. 24 hours apart so that there could be created a coherent set of data.

The first two day were deliberately omitted, because plants could be watered more times, especially as there was a great number of plants. Therefore, only the data from the next days are depicted, reflecting permanent watering. During the 24 hours Ing interval, there was marked a gradual decrease of reflection. *Citrus* × *limon* reacts on overwatering by chlorosis, which causes lower reflectance in green spectrum and higher one in yellow spectrum, with no significant difference between the two spectra. Throughout the days, reflectance becomes lower, which is considered a negative phenomenon. This was visibly expressed by shedding leaves.

There were not marked significant changes, neither visually, nor by measurements in 24 and 48 hours. On the third day, there were marked changes in the measured data (axis 3, red colour), when reflection decreased in green spectrum. In the trial, we watered the plant after the third day, and after 24 hours, the values (axis 4, yellow colour) were similar to those measured 24 hours after irrigation in the previous irrigation period.

There were not marked significant changes after 24 and 48 hours after irrigation, however, after 72 hours, spectrometry detected more significant changes, which could not be identified visually. Generally, there was a decrease in the whole section of visible light. On the fourth and fifth day after irrigation, the changes were identifiable visually, which was confirmed by spectrometry data. Leaves' colours changed significantly, from green to hints of yellow, which was caused by lack of water and also nutrients, which are supplied to plants through water. Regarding the balance of green and yellow spectra, they became more similar, which caused leaves turning yellow. After irrigation (axis 6, grey colour), the measured data showed almost the same data as after the previous irrigation. The 5-days interval was proved a more acceptable one by lower temperatures than the 3-days one.

In the first two days after irrigation, no significant changes were marked, the overall course was similar to the ones by 3-days and 5-days intervals. By raising number of days without irrigation, the overall reflection decreased. After irrigation (axis 7, black colour), reflection increased, but it did not reach the values comparable with those after the previous irrigation.

Based on the measurements we concluded that the most suitable irrigation intervals are once in three days by stable conditions of 18 °C, in a well-ventilated room, with higher air humidity. The period might by one day shorter, if it is necessary to increase the air temperature to 20–23 °C. If the temperature was 15 °C, the interval could be prolonged to 5 days, as there are no significant differences in first three days of the cycle, and the reactions on the decreasing supply of water in leaves can be detected on the fourth day only. It is not suitable to require daily irrigation, not only

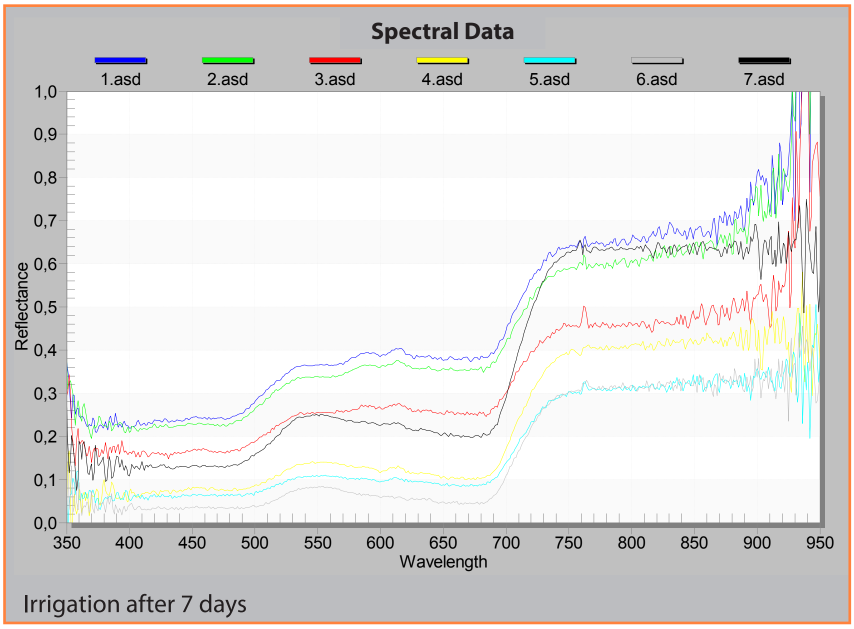

Figure 5 Graphic section of visible spectrum and close infrared spectrum of irrigation for once in seven days

because of the measured data, but also due to the plant requirements. On the other hand, in 7-days interval, there is a deficiency of water in plants.

Based on literature as well as experience, spectrometry can be used not only in agriculture, but also in plant cultivation considered a free time activity, as there is no universally acceptable literature on the cultivated plants and it is necessary to treat plants based on their reactions.

Throughout the past decades, spectrometry progressed a lot, which means that it is not a problem to acquire necessary device for a relatively low price, which can return to growers in short time, either in the form of finance, or through the good feeling motivated by high-quality, home-made crops.

Conclusion

The use of spectrometry provides many benefits as the health conditions of plants are monitored. Firstly, it is a non-invasive, non-destructive method. The method can be used both in interior and exterior premises, as it provides trustworthy information on plants' healt status under stable conditions. Particularly, green and NIR spectra provide valuable data on water requirements as well as other information, i.e. about chlorosis (caused either by biotic or by abiotic features), and thus prevent the unwanted phenomena. This method is not difficult regarding the use of IT as the HadHeld2 and the related software is easy to use, the results are very clear and suitable to use in practice.

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