

## INVESTIGATION OF THE RELATIONSHIP BETWEEN SPECTRAL VEGETATION INDICES AND CHLOROPHYLL CONTENT IN THE LEAVES OF SYRAH CULTIVAR VINES

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### Abstract

*Chlorophyll is a vital component of vine leaves, offering insights into their physiological state. It is the catalyst for photosynthesis, a process that drives the vegetative growth of vine plants. The present study investigates the correlation between chlorophyll content in vine leaves, which were simultaneously monitored by ground-based measurements and Sentinel 2 high-resolution vegetation indices (VIs) NDVI, CVI, CLGR through multispectral imaging. The experiment was conducted on Syrah grapevines over a three-year period, from 2020 to 2022. The vineyard was in full fruiting at the time of the study. In situ measurements of the photosynthetic activity of the vines were performed twice during the vegetative phase, before and after the flowering phase, in real conditions with a mobile apparatus (CCM-300). The remote data and imageries were processed using SNAP and QGIS. This study is of significant value to the field of precision viticulture, particularly in the context of grape production, as it provides insights into the potential of vegetation indices based on in situ measurements and VIs data from satellites.*

**Key words:** Chlorophyll, precision viticulture, remote sensing, vegetation index, vine.

### INTRODUCTION

Precision agriculture is dependent on several factors, one of which is remote sensing, which allows the health and condition of vegetation to be monitored. Many researchers in the field have analysed plant monitoring applications, research and scientific methodologies in this area (Giovos et al., 2021). The concept of precision viticulture is beginning to impact the wine sector, with several monitoring studies having been conducted in viticulture using a combination of vegetation indices, different remote sensing methods and in situ measurements (Arnó Satorra et al., 2009). The spatial variability of vineyard vigour has been assessed using the Normalized Difference Vegetation Index (NDVI) (Santesteban et al., 2013). Vegetation indices are primarily employed for the purpose of monitoring vine conditions, with the objective of correlating them with yield and other parameters such as chlorophyll concentration in leaves, nitrogen content, and water content in relation to canopy (Caruso et al., 2017; Gil-Pérez et al., 2010; Romero et al., 2018). Chlorophyll estimates

derived from remote sensing are utilised in precision viticulture. It has been established that chlorophyll has a correlation with vegetation spectral indices estimated according to digital image analysis (Bodor-Pesti et al., 2023). However, the results of the correlation of pigments with chlorophyll concentration do not always coincide (Romero et al., 2018). The studies carried out with the chlorophyll concentration in the leaves of the vine variety 'Hárslevelű' (*Vitis vinifera*) examined the relationship of different vegetation indices. The authors of the study concluded that the deficiency of mobile elements is evident in older leaves, while immobile elements manifest symptoms in young organs such as the shoot tip (Bodor-Pesti et al., 2023).

Chlorophyll is the molecule in the chloroplasts of all green-pigmented parts of plants that act as a catalyst for photosynthesis. It absorbs the strong blue spectrum of light as well as red, and converts sunlight into chemical energy. As a result, chlorophyll content is reported to directly affect crop growth and yield. The ability to detect changes in chlorophyll content facilitates the distinction of physiological

variations in plants through phenotyping crops and monitoring crop characteristics (Jayne et al., 2018). Chlorophyll content is of particular significance in the context of reduced fertilizer application, improved yield estimation, and, most crucially, precision agriculture (Naqvi et al., 2021).

The necessity to improve the quality and profitability of agricultural production is driving farmers to introduce new technologies. This is a useful step in the evolution of agricultural management systems through the introduction of new technologies such as multispectral remote sensing cameras (Whelan & McBratney, 2012). The use of VIs in agriculture is beginning to evolve rapidly and is significantly expanding its range of applications (Boiarskii & Hasegawa, 2019). The aim of this work is to investigate the relationship between vegetation indices, which refer to the content of chlorophyll in leaves, and in situ measurements in vines in different clones of the cultivar Syrah.

## MATERIALS AND METHODS

### Study area

The experiment was carried out on the training experimental field of Agricultural University in the town of Kuklen, Plovdiv district situated in Central-South Bulgaria (Figure 1).

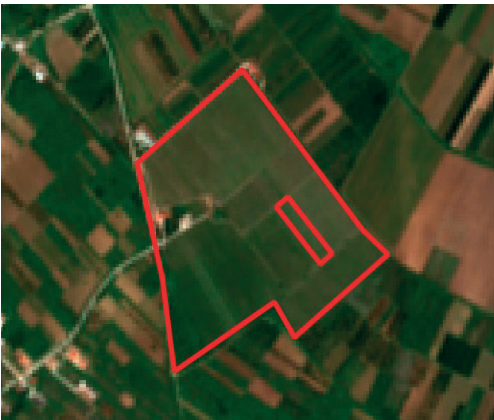


Figure 1. Location of study area (Sentinel - 2 image 23.05.2021)  
L 42° 3'29.71"N B 24°47'0.18"E

## Experimental design and treatments

The images from Sentinel -2 were obtained from Copernicus browser and Vis were calculated by the formulas in Table 1. <https://browser.dataspace.copernicus.eu/>

Table 1. Formulas for Vis for chlorophyll contents in crops

Index name	Formula	Formula Sentinel-2	References
Chlorophyll Index Green (CLGR)	$CLGR = (NIR/GREEN) - 1$	$CLGR = (B8/B3) - 1RED$	(Gitelson et al., 2005)
Chlorophyll Vegetation Index CVI	$CVI = (NIR/GREEN) * (RED/GREEN)$	$CVI = (B8/B3) * (B4/B3)$	(Vincini et al., 2008)
Normalized difference vegetation index NDVI	$NDVI = (NIR - RED) / (NIR + RED)$	$NDVI = (B3 - B4) / (B3 + B4)$	(Rouse JW, 1974)

NIR - Near infrared light from electromagnetic spectrum

GREEN - Green light from electromagnetic spectrum

RED - Red light from electromagnetic spectrum

The vegetation indices are calculated from Sentinel-2 bands images and sample raster values were processed by SNAP and QGIS 3.10.

The correlations between the studied variables were obtained by regression analysis in Excel Microsoft 365 and were valid within the time range studied.

The present study utilised clones of the variety Syrah, namely numbers 100, 174, 470 and 524, which were grafted onto the rootstock Berlandieri x Riparia SO4. These clones were planted in April 2011 in the experimental training field of the Agricultural University in Plovdiv. *In situ* data collection was conducted between 2020 and 2022.

Experimental scheme includes the following 4 options:

V1 - Syrah variety, clone 174;

V2 - Syrah variety, clone 470;

V3 - Syrah variety, clone 524;

V4 - Syrah variety, clone 100.

Each variant includes 60 vines (4 repetitions x 15 vines).

The climate data for temperature and precipitation has been recorded by the climate station in the plantation during the period April - June. The determination of total chlorophyll content in plants was conducted using a portable apparatus, designated as CCM-300. Measurements were performed during the active growth stage of vines under controlled conditions, on vine shoots of uniform size, load, exposure and illumination.

The BBCH scale was utilised to ascertain the phenology of the vines, with the decimal coding for determining the phenological stages of vine growth being identified as follows: the first digit indicates the main growth stage, and the second digit indicates the secondary growth stage, corresponding to a sequential number or percentage value.

The growth stages in the research period are based on DOY (Table 2).

Table 2. BBCH codes and growth stages

BBCH codes	Growth stages	DOY 2020	DOY 2021	DOY 2022
1:16	1: Leaf development 16 - sixth leaves are open	131	130	130
5:57	5: Flower display 57- inflorescences fully developed, flowers separated	146	143	140
7:71	7: Fruit development 71-budding, young fruit beginning to set, remnants of flowers falling off	156	168	158
7:75	7: Fruit development 75- Clusters of grains are pea-sized	181	180	180

## RESULTS AND DISCUSSIONS

### *In situ data*

#### *Climate data*

The pivotal elements of climate that exert an influence on the vital functions of the vine plant are air temperature and precipitation. It has been demonstrated that heat conditions have the capacity to influence the normal course of physiological and biochemical processes during the vegetation of the vines.

During the study period, daily mean temperatures and precipitation were recorded and presented in ten daily periods.

The average temperatures recorded during the study period, over the course of several years, did not exhibit significant variations. However, the total precipitation showed considerable differences (Figure2).

In a temperate climate, the soil is adequately supplied with water when the rainfall is 400-600 mm/m<sup>2</sup> and is distributed relatively evenly throughout the year.

The average annual precipitation in the vineyard in 2020 was 572.4 mm, with the highest recorded rainfall occurring in April at 106.8 mm. In 2021, the mean annual rainfall for the study area was approximately 200 mm higher than the previous year, at 755.4 mm. The mean annual precipitation in 2022 for the area was 572.4 mm, with June being the rainiest month (103.8 mm).

The total precipitation recorded during the 2020 growth stage BBCH 5:57 was 0.4 mm, while the post-flowering BBCH 7:71 precipitation was 17.4 mm.

A comparative analysis of the precipitation data from 2021 and 2022 with that from 2020 reveals that the respective amounts were 13.6 and 41.4 mm and 13.4 and 41.4 mm (Figure 3).

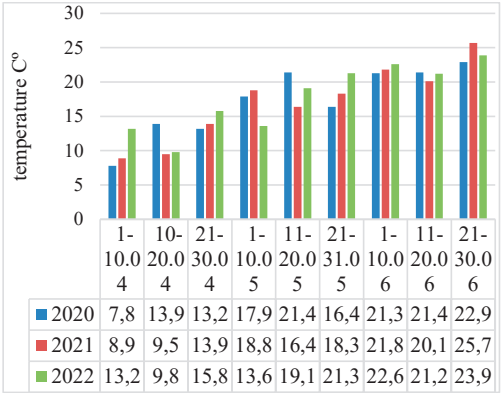


Figure 2. Average temperatures by ten-day periods from April to June

### *Chlorophyll measurements*

The Syrah clones, numbers 100, 174, 470 and 524, were selected based on their identity, biological and production potential (ENTAV-INRA-ENSA-MONIVINS, 1995).

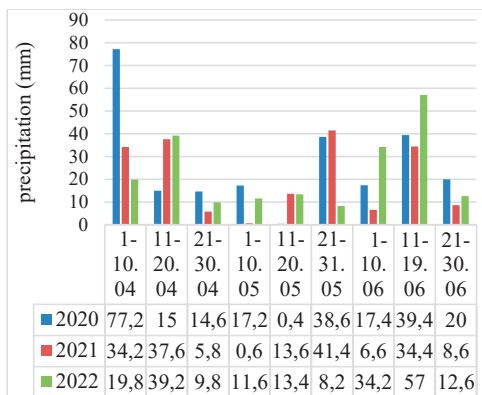


Figure 3. Precipitation totals in mm by ten-day periods from April to June

The content of plastid pigments (chlorophyll) in the tissues before the flowering stage of the vines is highest in the 0.470-0.366 g/m<sup>2</sup> clone, which also shows the highest net photosynthetic rate. The other three clones included in our study had lower chlorophyll content, with the lowest being at 100-0.316 g/m<sup>2</sup>.

Following growth stage flowering, total chlorophyll levels remained relatively stable across the different vines, maintaining the same order as before flowering. Clone 470 exhibited the highest chlorophyll content of 0.271 g/m<sup>2</sup>, which corresponded to a high transpiration intensity of 1.34 mmol H<sub>2</sub>O m<sup>-2</sup> s<sup>-1</sup>. The other three clones displayed lower chlorophyll contents, with the lowest recorded in clone 100, at 0.246 g/m<sup>2</sup> (Table 3).

#### *Vegetation indices relevant to chlorophyll*

The chlorophyll leaf index CVI was proposed by Vincini et al. (2008) as a means of estimating the chlorophyll concentration in crop leaves. The leaf pigment of plants provides important information on their health status.

(Gitelson et al., 2005) developed a Chlorophyll Index Green CLGR to estimate the chlorophyll content in crops.

Plant reflectance values obtained by specialised methods are used to measure the content of, for example, chlorophyll, carotenoids and other nutrients (Gitelson et al., 2005). Chlorophyll,

being capable of absorbing light energy, contributes to the photosynthetic process, whilst carotenoids protect photosynthetic systems from damage.

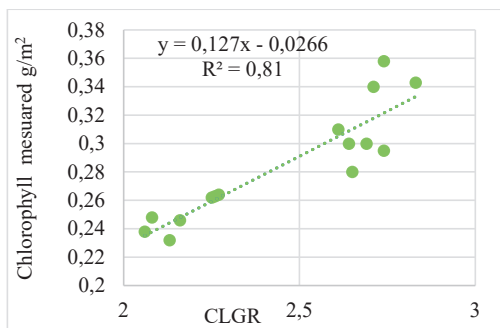
Table 3. Chlorophyll measured

BBCH CODE	V1 - Clone 174 Chlorophyll g/m <sup>2</sup>	V2 - Clone 470 Chlorophyll g/m <sup>2</sup>	V3 - Clone 524 Chlorophyll g/m <sup>2</sup>	V4 - Clone 100 Chlorophyll g/m <sup>2</sup>
<b>2020</b>				
<b>1:16</b>	0.247	0.264	0.262	0.263
<b>5:57</b>	0.343	0.358	0.340	0.295
<b>7:71</b>	0.238	0.246	0.248	0.232
<b>7:75</b>	0.310	0.300	0.280	0.300
<b>2021</b>				
<b>1:16</b>	0.250	0.268	0.270	0.270
<b>5:57</b>	0.326	0.335	0.335	0.287
<b>7:71</b>	0.248	0.248	0.235	0.233
<b>7:75</b>	0.300	0.330	0.320	0.300
<b>2022</b>				
<b>1:16</b>	0.313	0.298	0.282	0.270
<b>5:57</b>	0.356	0.405	0.374	0.367
<b>7:71</b>	0.263	0.321	0.273	0.274
<b>7:75</b>	0.284	0.290	0.276	0.343

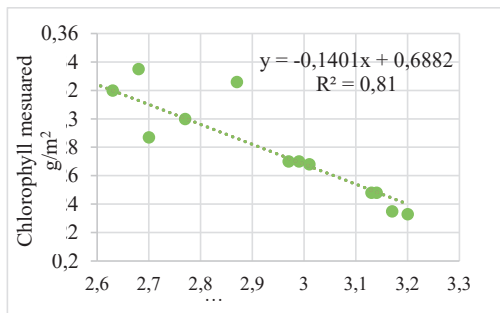
The Normalized Difference Vegetation Index (NDVI) is a widely used metric for assessing vegetation health and density. It measures the difference between near-infrared (which vegetation strongly reflects) and red light (which vegetation absorbs).

The study aimed to investigate the relationships between actual measured chlorophyll values and vegetation index CLGR, CVI and NDVI values obtained from Sentinel 2 imagery. The dates of the images obtained were as close as possible to the in-situ measurements.

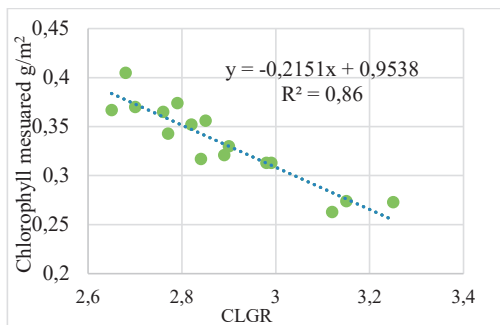
The regression analyses performed for the three-years study were processed between the CLGR, CVI and NDVI indices and the real in situ data of chlorophyll in the leaves of the vine cultivar clone Syrah are demonstrated in the figures given below (Figure 4, Figure 6, Figure 7).



a) 2020



b) 2021



c) 2022

Figure 4. Relationship between CLGR value from satellite and *in situ* data

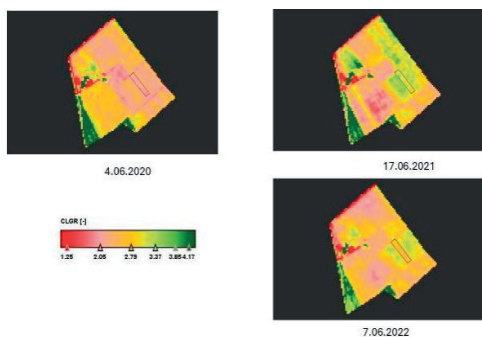
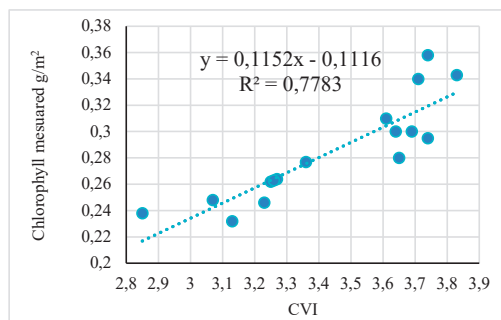


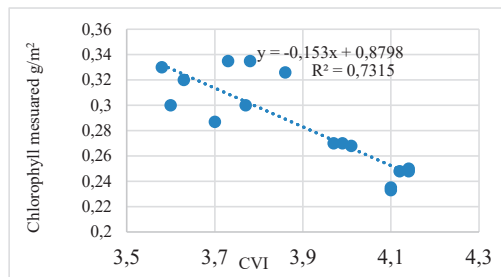
Figure 5. CLGR index from Sentinel 2 - BBCH 7:71 Fruit development

The range of CLGR index values across the experimental periods is from 2.1 to 3.3, with values in the first year of the period (2020) differing from those in the other two (2021, 2022). This is due to the higher temperatures and lack of or few amounts of precipitation in the period close to the chlorophyll measurements. The index ranges were from 2 to 2.9 in 2021 and from 2.6 to 3.3 in 2022. The multiple correlation coefficient is high for all three reporting years,  $R^2 = 0.81$  and  $R^2 = 0.86$ .

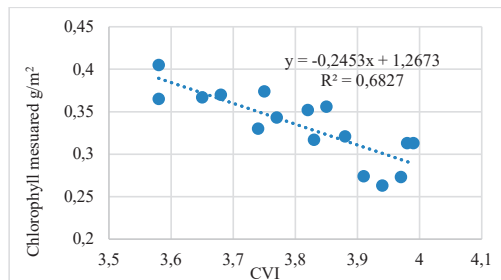
The scatter of values was observed in May during of the three years in growth stage BBCH 5:57 in clone 470 Syrah, and the best relationship was observed in phenological phase BBCH 7:71 (Fruit development) (Figure 5).



a) 2020



b) 2021



c) 2022

Figure 6. Relationship between CVI value from satellite and *in situ* data

The range of CVI index values for the three-year period under investigation is 2.8 to 4.2. The range of values for the chlorophyll vegetation index compared to the CLGR is largest in 2020.

In comparison, in 2021 and 2022 the index ranges from 3.5 to 4.2 and 3.5 to 4, respectively.

The strongest multiple correlation coefficient is reported in 2020, with a value of  $R^2 = 0.778$ . (Figure 6).

NDVIs are presented in a growth stage BBCH 7:71 Fruit development (Figure 8).

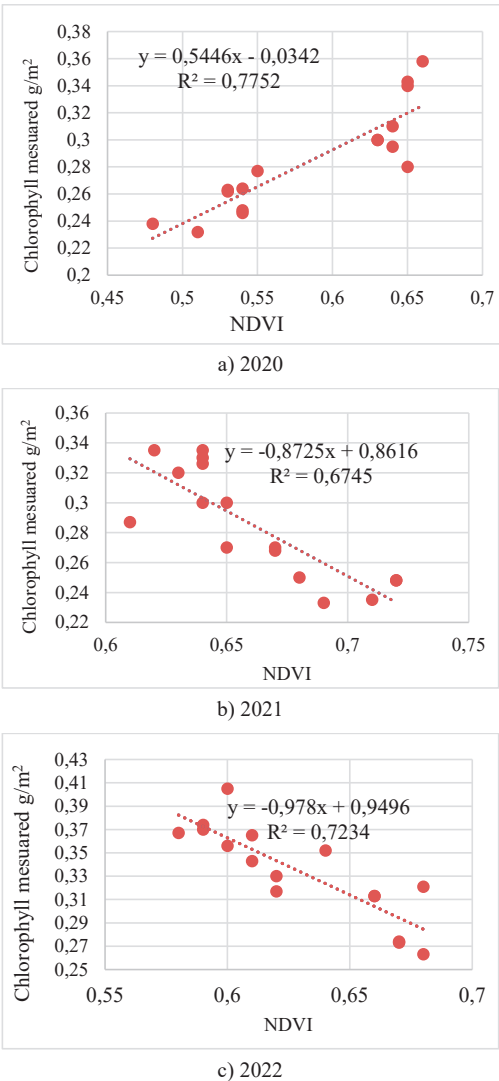


Figure 7. Relationship between NDVI value from satellite and in situ data

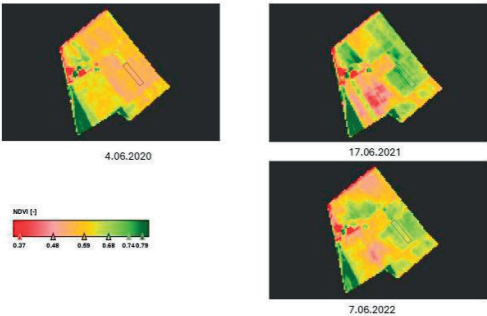


Figure 8. NDVI from Sentinel 2 - BBCH 7:71 Fruit development

The Normalised Difference Vegetation Index (NDVI) is the most common index for assessing plant vigour. Studies conducted during the experimental period demonstrated a very good multiple correlation coefficient of  $R^2 = 0.67$  to  $R^2 = 0.77$  for 2020. The widest range of values was reported for 2020, from 0.45 to 0.67, but in 2021, higher values of the index were reported (Figure 7).

This discrepancy can be attributed to the higher precipitation levels that occurred prior to the measurement of chlorophyll values and the acquisition of the image.

## CONCLUSIONS

The application of the index comparison process resulted in the identification of a robust relationship and a high multiple correlation coefficient for CLGR, with values of  $R^2=0.81$  and  $R^2=0.86$ , respectively.

The multiple correlation coefficient for NDVI is reported to be the lowest, with the smallest value recorded in 2020,  $R^2 = 0.67$ .

Despite the variations in the regression equations' values from those studied, the correlations proved to be of a very good degree.

The regression equations derived represent the relationships with *in situ* data and can be used to fit the values of satellite vegetation indices to analytical ones. This is extremely useful for estimating chlorophyll values with a high degree of probability and for making accurate predictions.

The primary advantage of CLGR, CVI and NDVI for practical applications is that they enable the use of RED, Green and NIR bands a



with a high spatial resolution (HR) of 10 m for the non-invasive monitoring of vine plantations, facilitating the assessment of physiological and biophysical parameters.

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