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Spectral response of grapevine (*Vitis vinifera* L.) cultivar Riesling to foliar applications

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ABSTRACT

This study investigates the effects of different foliar applications on the spectral characteristics of grapevine (*Vitis vinifera* L.) cv. Riesling leaves. In addition to untreated control plants, experimental plots were treated with natural bioactivator extracts and microelements. Leaf samples were collected at the beginning of July and September 2024, referring to E-L33-34 and E-L38 Eichhorn-Lorenz phenological stages. Reflectance measurements were performed on leaf discs of uniform size ($r = 10$ mm; $A = 314.2$ mm^2) using a CI-710 Leaf Spectrometer. Spectral data were analyzed using principal component analysis (PCA), and selected wavebands were utilized to compute vegetation indices, including NDVI (Normalized Difference Vegetation Index), EVI (Enhanced Vegetation Index), CI-G (Chlorophyll Index Green), MCARI (Modified Chlorophyll Absorption Ratio Index), NDRE (Red-Edge Normalized Difference Vegetation Index), VARI (Visible Atmospherically Resistant Index), and PRI (Photochemical Reflectance Index). PCA revealed that the first principal component (PC1) accounted for 54.3% of the total variance, while PC2 and PC3 explained 35.5 and 5.5%, respectively. The highest loading for PC1 was observed at 725 nm, whereas for PC2, it was at 505 nm. Analysis of variance (ANOVA) indicated that the applied treatments had a

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statistically significant effect on the vegetation indices. These findings highlight the impact of foliar applications and corroborate previous results demonstrating that leaf chlorophyll content and morphology are influenced by different treatments.

KEYWORDS

vegetation index, reflectance, leaf, nutrient supply

INTRODUCTION

Grapevine (*Vitis vinifera* L.) is produced on more than 7.3 million hectares under various environmental conditions resulting in several different abiotic stress factors such as drought, flood, uneven rainfall, high UV radiation, which force the growers and researchers to find effective solutions to enhance plant hardiness (OIV, 2025; Naulleau et al., 2021).

Grapevine leaves show high morphological variability among genotypes, while certain traits are stable between individuals of a cultivar that makes leaf morphological investigation suitable for cultivar identification. At the same time, other leaf traits can vary highly in the canopy. The diversity concerns the size, shape, anatomy and plant physiology of the leaves. This is due to the position and age of the leaves. The leaves on the lower parts of the shoots/canopy are the oldest, while those on the upper parts of the foliage are the youngest (Chitwood and Mullins, 2022; Bodor-Pesti et al., 2023). Consequently, leaf age has a significant effect on the morphology of the lamina.

Foliar applications are widespread in viticulture to support plant development and increase stress tolerance, while certain nutrients are also possible to be supplied through canopy such as boron (B), zinc (Zn), iron (Fe) and manganese (Mn). According to several literature sources, grapevine is not considered to be a highly nutrient-demanding crop, although one of the keys to balanced yield and high quality is adequate nutrient supply (Jackson, 2020). Foliar treatments are usually grouped as nutrients, bioactivators and elicitors applied to mitigate stress. The physiological effects of biostimulants and bioactive substances are diverse while several studies are dealing with these general effects. For example, Jindo et al. (2022) reviewed different groups of biostimulants and their effects on viticulture, including microorganisms, protein hydrolysates, humic acids, pyrogenic materials, and seaweed extracts. The role of salicylic acid in plant protection and yield enhancement was investigated in several studies such as Maleck et al. (2000), Király (2004), Reymond et al. (2004), De Vos et al. (2005), Wodnicka et al. (2017). Babaei et al. (2014) investigated the effect of salicylic acid on photosynthesis in two grape varieties under arid growing conditions. They observed that salicylic acid increased leaf thickness, transpiration, stomatal conductance and net photosynthetic activity. It was concluded that salicylic acid could reduce the drought stress effect on grapevine growth. Peng-Fei Wen et al. (2008) found that salicylic acid also plays a role in increasing drought tolerance and in protecting against heat stress by inducing phenylalanine ammonia-lyase. Abdel-Salam (2016) examined the effect of certain microelements and salicylic acid on the ripening processes and chemical content of grape berries, and concluded that the microelements increased the weight and content values of clusters and berries, while salicylic acid treatment delayed the ripening of the fruit

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by two weeks. Our former study verified that foliar applications have noticeable effect on leaf thickness (Bodor-Pesti et al., 2025).

The supply or demand of the nutrients could cause significant influence on the leaf morphology, in more particular coloration, texture, lobature, thickness, anatomy. Sun et al. (2018) showed that according to the plant nutrition mechanism, leaf characteristics display different changing trends under nitrogen (N), phosphorus (P), and potassium (K) nutrition stress. Lopes et al. (2024) investigated the effect of N dose and shade on the morphogenetic, structural, and leaf anatomical characteristics of *Megathyrsus maximus* cv. Tamani and Quênia.

To overcome the nutrient deficiency of plantations, foliar fertilization is an increasingly accepted and applied method in agriculture. The application of biostimulants to the foliage can improve the uptake and efficient use of nutrients. Cangi et al. (2006) found that the application of humic acid affected the soluble solids content, total nitrogen and iron content of grape leaves. In addition, it can reduce the stomatal function and transpiration of leaves, thereby reducing water consumption, improving plant water supply, ensuring normal plant growth and development under drought conditions, and enhancing drought tolerance.

Remote and proximal sensing of leaves can provide valuable information about the physiological status of plants, through for example the chlorophyll content, water status, and moreover nutrient content. In this context, RGB (R: red, G: green, B: blue), multispectral and hyperspectral sensors are commonly involved. Within the visible spectrum nutrient deficiency often shows characteristic patterns, for instance, magnesium deficiency causes light green and later yellow discoloration starting between the veins, while iron deficiency results chlorotic pattern. In addition to the raw visible red, green, blue spectrum (RGB) data, there are several vegetation indices to monitor leaf discoloration such as Soil-Adjusted Vegetation Index (SAVI), Visible Atmospherically Resistant Index (VARI). For multispectral image analysis the Normalized Difference Vegetation Index (NDVI) is one of the most widespread vegetation indices, which is involved in many studies to define plant nutrient status. One major benefit of the hyperspectral analysis is that, beside analyzing the spectral pattern, it allows us to calculate diverse kinds of vegetation indices (Debnath et al., 2021; Chancia et al., 2021). Recently several studies reported leaf nutrient content analysis according to spectrometric data. These studies reported that nutrient deficiency causes significant change in the spectrum, while the vegetation indices are also influenced. Osborne et al. (2002) compared the effect of four rates of nitrogen and phosphorus on corn (*Zea mays* L.). Spectral radiance measurements range from 350 to 1,000 nm at different phenological stages, and the authors defined those regions and wavelengths that correlated with plant nutrient concentration. Li et al. (2006) used the whole visible spectrum (380–780 nm) to detect pre-visual spectral pattern for Fe and P deficiency. Later Jin et al. (2022) applied a near-infrared spectrometer (900–1,700 nm) to define the effect of nitrogen, phosphorus and potassium deficiency on the pear leaf reflectance spectra.

Our former study emphasized that foliar applications cause significant alteration on the leaf morphological traits of the grapevine (*V. vinifera* L.) cultivar Riesling (Bodor-Pesti et al., 2025). We found that leaf thickness, chlorophyll content and light transmittance are influenced by the treatment of different foliar applications, therefore this study aims to determine whether different bio-based and conventional foliar treatments result in measurable reflectance differences.

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MATERIALS AND METHODS

Plant material and treatments

The investigation was carried out at the Research Station for Viticulture and Oenology Kecskemét of the Hungarian University of Agriculture and Life Sciences. The investigated grapevine (*V. vinifera* L.) cultivar was Riesling. Leaf samples were collected on 11th of July (EL33-34) and 3rd of September 2024 (EL38). Following the recommendations of the OIV Descriptor List (2009) 15 leaves from the middle third of several shoots from both sides of the canopy from at least 10 plants were randomly collected from each treatment. To reduce the possible spectral variability caused by the age (ontogeny effect) and position (heteroblastic effect) of the leaves, samples were collected from the same vertical position of the shoots. Samples were pooled rather than treated as replications, as this approach allowed direct comparison with leaf nutrient content data, where samples were also pooled for destructive analysis. For spectral measurements, the leaves were stored in plastic zip-bags in a cooling box and transported immediately to the nearby laboratory (approximately 300 m from the experimental plots).

The following treatments were applied:

- AS (Active start) - Active start technology: Activstart is a bioactivator that consists of natural antioxidants (e.g., flavonoids), small amounts of essential microelements, activators (e.g., enzymes) and complexing agents (e.g., pyroglutamate). With proper use, it is possible to effectively prevent stress effects (drought, temperature, UV radiation, etc.) during cultivation, and if necessary, to achieve rapid regeneration and harmonious development of the grapevine.
- DrG - The research field plantation has been subjected to biological control since 2016. The carefully formulated composition of the fertilizer has a protective, regenerative, and stress-relieving effect on the crop and significantly influences among others: supplementing deficiencies and limiting the negative effects of their occurrence, enhanced resistance to thermal stress (low/high temperatures) and biotic and abiotic factors, such as diseases caused by pathogens, or damage associated with pest feeding, regulation of processes related to the water management of plants.
- PLHU (PlanTonic and Humic acid) - The research field plantation has been subjected to biological control since 2016. The "PlanTonic" plant conditioning preparation contains only plant substances (nettle aqueous extract + willow twig oily extract), to which we have added humic acid + microelement treatment as a supplement to increase the yield and quality. It strengthens, enhances and activates the plant's natural defenses.
- Control: In the control parcels no nutrient supplementation was applied according to the rules of Agri-environment program without (CAP project nr: KAP-RD19a-1-24).

In all treatments, the conditioning/biostimulant preparations were applied at the same time (E-L12; E-L19; E-L31-33; E-L35). In the case of PLHU, one more treatment was applied at the time of flowering (E-L27-28).

Meteorological parameters

The 2024 vintage was characterized by high temperatures, atmospheric drought and very low amount of precipitation in the growing season (Fig. 1). The minimum temperature was −1.83 °C, while the maximum was 38.97 °C with an average 18.63 °C. The annual precipitation

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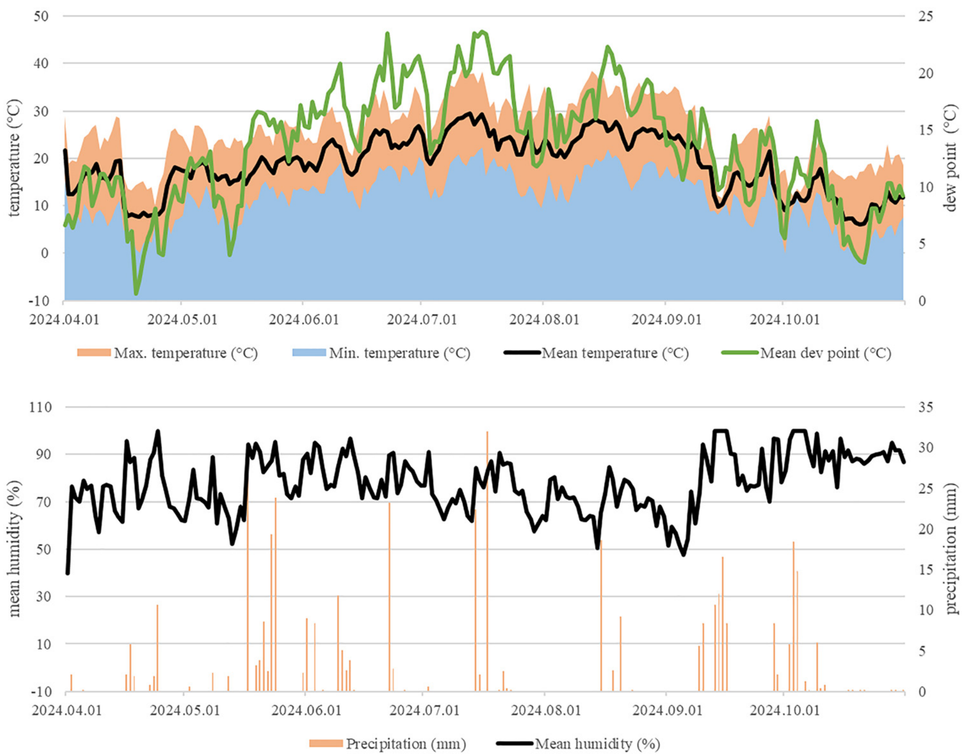

Fig. 1. Temperature and precipitation conditions in vegetation period of the year 2024

was 399.2 mm. In April and early May, spring frosts caused significant frost damage to the early-bud breaking vines. The vegetation started 2–3 weeks earlier than the long-term average. The harvest began in July for the early maturing varieties, and by the end of August, harvest of all varieties was finished in the Hungarian Great Plain. In other Hungarian wine regions, the harvest was finished by mid-September. Bud break started very early on 6th March 2024. In March, the max. temperature was 26 °C, there was hardly any precipitation. April started with summer heat, the max. temperature reached 28.9 °C, but night frosts were also present (−4.53 °C). On April 24, the shoots reached a size of 15–20 cm, which had frozen over a large area of the country because of the delivered frost. There was still no precipitation. The flowering started on 27th April 2024. In May, the rate of shoot growth slowed down significantly due to the night (4.25 °C) and daytime cold. The maximum temperature reached 28.1 °C. Flowering of grape varieties began on 17th May 2024. The beginning of June was moderately warm, with cool night temperatures. The amount of precipitation was 67 mm. The maximum temperature in June was 35.16 °C. July and August were very hot and dry. Powdery mildew infection continued. The maximum temperature was 38.97 °C. In August the conditions for fungal infections (downy mildew, powdery mildew, black rot) were not favorable due to the high UVB radiation. September started with heat, then after the 13th there was a slight cooling with precipitation (72 mm), the drought eased.

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Leaf nutrient content

Leaf nutrient content was analyzed two times during the vegetation period at flowering (E-L19-23) and ripening (E-L35-38). Fifty leaf samples were collected from the opposite side of the bunches from the treated plots and nutrient content was investigated. The chemical sample preparation of plant materials was carried out in accordance with the Hungarian standard MSZ-08-1783-1:1983 section 3.3.2. and 4.3., which is functionally equivalent to AOAC 965.09 (Wet Ashing of Plant Tissue) and EN 15505:2008 (Pressure Digestion) (Table 1) (for more details see On-line Szabványkönyvtár).

Spectral investigation and vegetation indices

Fifteen leaves per treatment were washed, dried with filter paper and 3 leaf discs of equal size ($r = 10$ mm; $A = 314.2$ mm^2) from each leaf were cut and investigated with a SpectraVue CI-710 Leaf Spectrometer (Cid-Bioscience, Camas, WA, USA). Based on spectral information the following vegetation indices were calculated: Normalized Difference Vegetation Index (NDVI), Enhanced Vegetation Index (EVI), Chlorophyll Index Green (CI-G), Modified Chlorophyll Absorption Ratio Index (MCARI), Red-Edge Normalized Difference Vegetation Index (NDRE), Visible Atmospherically Resistant Index (VARI), Photochemical Reflectance Index (PRI) (Table 2) according to Daughtry et al. (2000), Rossini et al. (2015), Gitelson et al. (2003) and the Index Database (2025).

Statistical analysis

To remove the noise from the lower wavelengths raw spectra were preprocessed with different methods: PCA denoising, Gaussian smoothing, and standardization (Z-transformation). Although we applied these steps to reduce noise, we found that the outcomes of the principal component analysis (PCA) did not change, and these preprocessing methods did not modify the PCA scatterplot. This indicated that the variance highlighted by PCA was predominantly driven

Table 1. Nutrient content of the investigated plots obtained at flowering (E-L19-23) and ripening (E-L35-38)

Date	EL19-23 (27th May 2024)				EL35-38 (27th August 2024)			
Nutrient content/Treatment	CO	AS	DrG	PLHU	CO	AS	DrG	PLHU
Nitrogen content (%)	2.77	2.81	3.17	2.53	1.82	1.72	1.58	1.81
Phosphorus content (%)	0.32	0.2	0.21	0.2	0.12	0.15	0.12	0.15
Potassium content (%)	1.29	1.17	1.36	1.12	0.66	0.74	0.89	0.82
Sodium content (mg kg^{-1})	98.7	112	91.9	139	217	155	136	205
Calcium content (%)	1.77	2.12	2.38	2.15	4.22	3.63	4.37	4.32
Magnesium content (%)	0.34	0.41	0.35	0.37	0.72	0.68	0.65	0.74
Ferrum content (mg kg^{-1})	96	93.3	121	86.9	149	102	135	155
Manganese content (mg kg^{-1})	69.6	81.3	40.6	61.4	85.5	73	63	79.1
Copper content (mg kg^{-1})	82.69	103	162	285	86.2	82.9	507	952
Zinc content (mg kg^{-1})	136	136	74.6	175	57.8	55	33.4	82
Boron content (mg kg^{-1})	63	84.6	49.4	0.23	30.1	32.5	16.2	35.5

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Table 2. The effect of the applied treatments and sampling dates on the mean values and ANOVA of the vegetation indices and reflectance at 725 and 505 nm

Date	Treatment	NDVI $(R_{800} - R_{680})/(R_{800} + R_{680})$ mean ± st.dev.	EVI $2.5 \times (R_{800} - R_{680})/(R_{800} + 6 \times R_{680} - 7.5 \times R_{473} + 1)$ mean ± st.dev.	CI-G $(R_{800}/R_{560}) - 1$ mean ± st.dev.	MCARI $((R_{700} - R_{670}) - 0.2 \times (R_{700} - R_{550}))/(R_{700}/R_{670})$ mean ± st.dev.	NDRE $(R_{750} - R_{705})/(R_{750} + R_{705})$ mean ± st.dev.	VARI $(R_{560}\text{-}R_{670})/(R_{560} + R_{670} - R_{473})$ mean ± st.dev.	PRI $(R_{531} - R_{570})/(R_{531} + R_{570})$ mean ± st.dev.	725 nm mean ± st.dev.	505 nm mean ± st.dev.
July, 2024	Control	$0.8000 \pm 0.02_{cA}$	$0.7416 \pm 0.05_{c}$	$3.1943 \pm 0.46_{ab}$	$0.1974 \pm 0.05_{bB}$	$0.4656 \pm 0.03_{aA}$	$0.5793 \pm 0.05_{b}$	$0.0461 \pm 0.01_{a}$	$36.1228 \pm 4.00_{c}$	$5.1480 \pm 1.01_{bB}$
	AS	$0.8123 \pm 0.02_{cA}$	$0.7434 \pm 0.03_{c}$	$3.7204 \pm 0.65_{cA}$	$0.1747 \pm 0.05_{bB}$	$0.4995 \pm 0.04_{bA}$	$0.5668 \pm 0.05_{b}$	$0.0571 \pm 0.01_{b}$	$35.1595 \pm 2.61_{c}$	$4.5442 \pm 0.65_{a}$
	DrG	$0.7259 \pm 0.05_{aA}$	$0.5511 \pm 0.09_{aA}$	$2.8003 \pm 0.66_{a}$	$0.0820 \pm 0.02_{aA}$	$0.4687 \pm 0.04_{a}$	$0.4620 \pm 0.05_{a}$	$0.0610 \pm 0.01_{cB}$	$25.8340 \pm 4.06_{a}$	$5.0764 \pm 0.54_{b}$
	PL + HU	$0.7658 \pm 0.04_{b}$	$0.6523 \pm 0.08_{bA}$	$3.5141 \pm 0.66_{bcA}$	$0.0931 \pm 0.03_{a}$	$0.5078 \pm 0.03_{bA}$	$0.4668 \pm 0.06_{a}$	$0.0643 \pm 0.01_{cB}$	$29.3445 \pm 3.97_{b}$	$5.1336 \pm 0.68_{bB}$
September, 2024	Control	$0.8336 \pm 0.01_{cB}$	$0.7656 \pm 0.03_{b}$	$4.8296 \pm 0.67_{b}$	$0.1351 \pm 0.04_{bA}$	$0.5425 \pm 0.04_{bB}$	$0.5206 \pm 0.05_{b}$	$0.0517 \pm 0.02_{b}$	$33.5523 \pm 2.46_{b}$	$4.2403 \pm 54_{aA}$
	AS	$0.8272 \pm 0.02_{bc}$	$0.7487 \pm 0.02_{b}$	$4.9520 \pm 0.59_{bB}$	$0.1162 \pm 0.04_{abA}$	$0.5545 \pm 0.04_{bB}$	$0.4960 \pm 0.05_{ab}$	$0.0574 \pm 0.01_{b}$	$32.4676 \pm 2.14_{b}$	$4.2585 \pm 0.62_{a}$
	DrG	$0.7876 \pm 0.04_{aB}$	$0.6626 \pm 0.08_{aB}$	$3.1017 \pm 0.75_{a}$	$0.1769 \pm 0.07_{cB}$	$0.4560 \pm 0.06_{a}$	$0.5728 \pm 0.05_{c}$	$0.0345 \pm 0.02_{aA}$	$32.1575 \pm 3.97_{b}$	$4.7858 \pm 0.73_{b}$
	PL + HU	$0.8106 \pm 0.04_{bB}$	$0.7004 \pm 0.08_{aB}$	$4.6167 \pm 0.86_{bB}$	$0.0939 \pm 0.03_{a}$	$0.5477 \pm 0.04_{bB}$	$0.4753 \pm 0.06_{a}$	$0.0511 \pm 0.02_{bA}$	$29.8591 \pm 3.58_{a}$	$4.4054 \pm 0.75_{abA}$

Where different letters indicate significant ($P < 0.01$) differences between the treatment (small letters) and sampling date (capital letters):
aA = treatment differences/temporal differences.

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by biological differences (foliar application treatments) rather than noise. As the results did not change, we were using the original spectra for the PCA without preprocessing. Data evaluation of the spectra, calculation of the mean values and the PCA were conducted with Orange 3.38 (Demsar et al., 2013). We used PCA to visualize the overall spectral variability at the two sampling dates and to determine which wavelength ranges drive the main differences among samples. The principal components highlighted the spectral regions with the highest discriminative potential. These two bands that showed high loading on the PC1 and PC2 were selected for further statistical evaluation. Two-Way ANOVA of the vegetation indices and spectral bands (505 and 725 nm) and Pearson correlation between the vegetation indices and nutrient content data was carried out with the PAST (Hammer and Harper, 2001).

RESULTS

In this study 3 foliar treatments were compared with untreated control samples obtained from a Riesling grapevine (*V. vinifera* L.) cultivar. We compared the nutrient content at the two sampling dates (Fig. 2) and found that calcium (%) increased, especially in CO (+138.4%) and PLHU (+100.9%) treatments. Magnesium (%) also showed a large increase in all treatments, the largest in CO (+111.8%). Iron content (mg kg^{-1}) increased in PLHU treatment (+78.4%), while copper content increased drastically in DrG and PLHU treatments, especially in PLHU (+234.0%). We observed some negative changes too. Zinc content decreased in all treatments, especially in AS (−59.6%). Boron (mg kg^{-1}) decreased in all treatments except for PLHU, where it produced an extreme increase. Among the different treatments, the salicylic acid + humic acid (PLHU as a biostimulant) treatment showed the greatest change in the ratio of nutrients that play a role in photosynthetic activity and influence chlorophyll formation.

Spectral comparison of the average reflectance spectra calculated from at least 40 individual samples per treatment and sampling date showed noticeable differences between 520 and

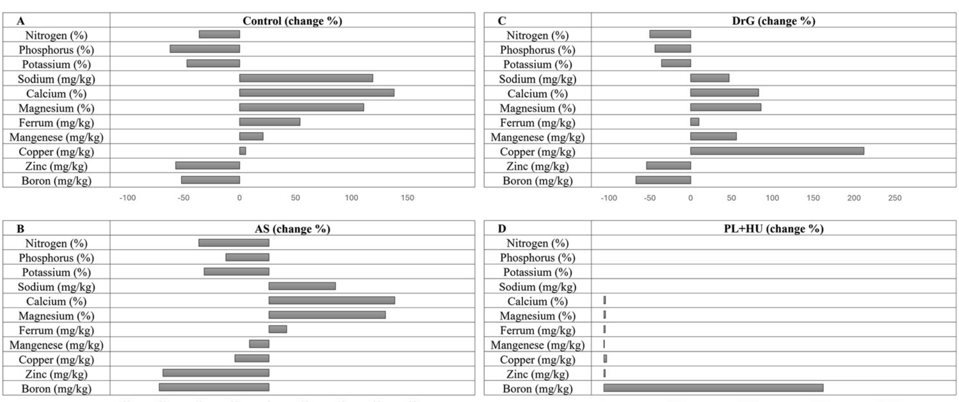

Fig. 2. Percentage change of each nutrient over time for the four different treatments (CO (a), AS (b), DrG (c), PLHU (d))

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640 nm, moreover from 740 to 1,000 nm corresponding to the near-infrared range. In the blue region of the visible spectrum, the reflectance was the highest in the case of the PLHU and DrG in July, while the lowest in the case of the Control in September. At 550 nm corresponding to the green reflectance peak the reflectance was the highest in the case of the Control samples in July, while the lowest in the case of the PLHU in September. In the red region of the spectrum, the highest values were obtained in the case of the Control samples collected in July while the lowest values were recorded in the case of the Control samples collected in September. Above the visible part of the spectra in red edge region (680–750 nm) the samples' reflectance was changing compared to the red region. Highest reflectance was in the case of the Control samples collected in July, while the lowest reflectance was obtained in the case of the DrG samples. In the near-infrared region above 740 nm the lowest values were recorded in the case of the DrG treatment collected in July, while the highest values were recorded on Control leaves sampled in September (Fig. 3).

Principal component analysis

Principal component analysis of the spectra showed that PC1 explained 54.3% of the total variance, while PC2 and PC3 explained 35.5 and 5.5% of the total variance respectively. The loading of PC1 was the highest at 725 nm suggesting that red-edge reflectance was a key factor in treatment discrimination that could be linked to the physical structure of the leaf. The lowest reflectance in this wavelength was obtained in the case of the DrG collected in July (25.83%), while the highest in the case of the Control samples obtained in July (36.12%). The loading for PC2 was the highest at 505 nm. Control samples collected in September showed the lowest reflectance (4.24%) while the samples from July showed the highest reflectance (5.14%) at this wavelength. PCA scatterplot in July and September showed grouping of the samples along the two represented principal components (Fig. 4). During flowering samples

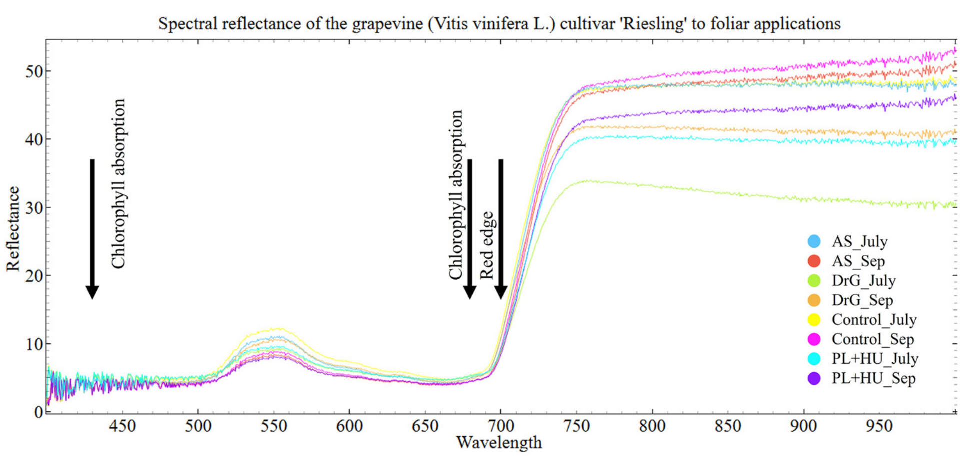

Fig. 3. Spectra of leaf reflectance obtained from Control, AS, DrG and PLHU treatments at two phenological stages

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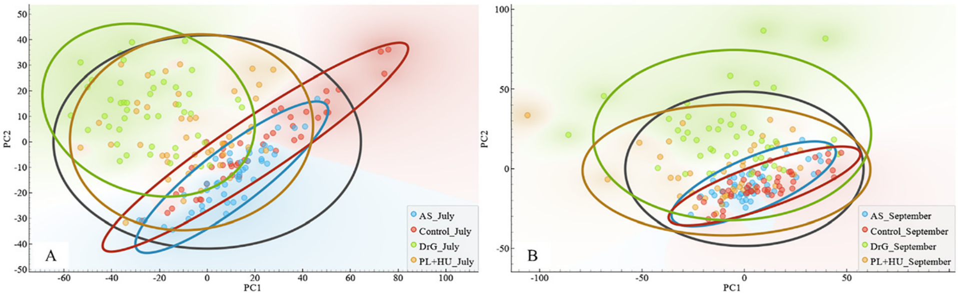

Fig. 4. Scatter plot of the principal component analysis (PCA) to summarize spectral descriptors based on the first two principal components (PC1 and PC2) of the samples obtained in July (A) and September (B)

treated with DrG formed a distinct group with only a slight overlap with Control and AS samples. Later sampling showed less distinctiveness, while it suggests that Control and AS samples have a lower variability than the DrG and PL + HU samples.

VEGETATION INDICES AND SPECTRAL BANDS

Two-way ANOVA showed a significant effect of both treatment and time on several vegetation indices (Table 2). The two-way ANOVA revealed significant effects of both treatments ($F = 83.58$, $P < 0.001$) and date ($F = 143.1$, $P < 0.001$) on the NDVI values. Additionally, a significant interaction between treatment and date was observed ($F = 9.30$, $P < 0.001$), indicating that the effect of treatment varied depending on the date. These results suggest that both treatment and temporal factors, as well as their interaction, contributed significantly to the observed variation of the NDVI values. For EVI, both treatments ($F = 101.0$, $P < 0.001$) and time ($F = 47.02$, $P < 0.001$) and their interaction ($F = 11.43$, $P < 0.001$) had significant effects. Likewise, CIG values were significantly affected by treatment ($F = 74.92$, $P < 0.001$), date ($F = 230.2$, $P < 0.001$) and interaction ($F = 15.79$, $P < 0.001$). For MCARI, treatment ($F = 50.82$, $P < 0.001$) and interaction ($F = 72.53$, $P < 0.001$) were significant, while the effect of date was not ($F = 2.37$, $P = 0.125$). The NDRE showed a significant effect of treatment ($F = 50.64$, $P < 0.001$), date ($F = 86.15$, $P < 0.001$) and their interaction ($F = 19.75$, $P < 0.001$). For VARI, significant treatment ($F = 36.78$, $P < 0.001$) and interaction effects ($F = 56.24$, $P < 0.001$) were found for the date and treatment but not for the sampling time ($F = 0.28$, $P = 0.599$). PRI was significantly influenced by treatment ($F = 12.08$, $P < 0.001$), date ($F = 30.14$, $P < 0.001$) and their interaction ($F = 22.33$, $P < 0.001$). Reflectance at 725 nm showed a significant effect of treatment ($F = 66.51$, $P < 0.001$) and interaction ($F = 34.09$, $P < 0.001$), but not of date ($F = 1.08$, $P = 0.299$). Reflectance at 505 nm, showed significant difference for all the three factors: treatment ($F = 8.88$, $P < 0.001$), date ($F = 54.37$, $P < 0.001$) and their interaction ($F = 4.43$, $P < 0.01$). Taken together, these results highlight that vegetation indices and specific reflectance bands responded differentially to both experimental treatments and temporal variation, with several indices showing strong interactions.

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CORRELATION BETWEEN THE NUTRIENT CONTENT AND SPECTRAL CHARACTERISTICS

To investigate the relationship between nutrient content of the leaves obtained from the different treatments and vegetation indices, a linear regression analysis was performed. Although nutrient content data were available only as one mean value per treatment per phenological stage, these values were assigned to all replicate vegetation index measurements, resulting more 360 data points. This approach enabled statistical testing but has to be interpreted with caution, as the nutrient values do not vary within treatments. According to the correlation analysis most vegetation indices, for example NDVI, CI-G and NDRE, showed significant correlation with nutrient content (Fig. 5). NDVI negatively correlated with N, P, K (-0.4608, $P < 0.01$; -0.2202, $P < 0.01$; -0.53482, $P < 0.01$ respectively), CI-G (N: -0.4901, $P < 0.01$; P: -0.39937, $P < 0.01$; K: -0.64312, $P < 0.01$) and NDRE (N: -0.33632, $P < 0.01$;

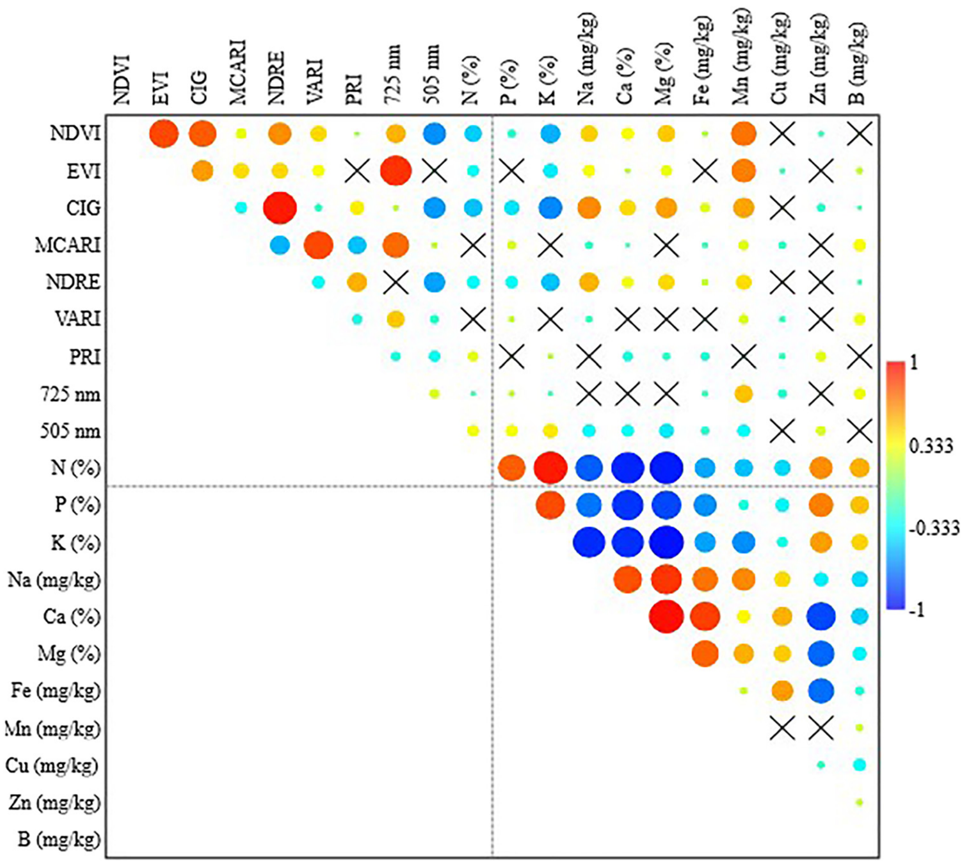

Fig. 5. Pearson correlation among the investigated vegetation indices, wavelengths and the leaf nutrient content

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P: −0.32517, $P < 0.01$; K: −0.49088, $P < 0.01$) showed similar results. VARI index showed a weak positive correlation with P, Na, Mn and Zn.

DISCUSSION

Spectral characteristics of the leaves are important indicators of plant physiological and nutrient status, moreover, the presence of pests and diseases. In this study we investigated the reflectance of the Riesling grapevine cultivar from 400 to 1,000 nm evaluating the visible part of the spectra, the red edge and NIR region according to the spectrum and calculated vegetation indices. We found that both the sampling date and the applied treatments have a significant effect on the reflectance. In the blue part of the visible spectrum the reflectance was the highest in the case of PLHU and DrG in July, while the lowest values were detected in the case of the Control samples in July. At the same time, it should be noted that the spectral differences in this region were negligible compared to later regions. High reflectance in this region indicates a lower chlorophyll concentration or a stress-induced reduction. In accordance with these results the highest reflectance in the green part of the spectrum was obtained in the case of the Control samples collected in July, while PLHU provided the lowest values in September. Later samples were probably already in the physiological aging that could induce chlorophyll reduction or pigment change. In the red region where the chlorophyll absorption is the strongest the Control samples showed the highest and lowest reflectance in July and September respectively that could be explained with the aging of the leaves. The red-edge region of the spectrum and its characteristics are indicative of chlorophyll content and leaf structural properties. The Control samples collected in July showed the highest reflectance, while the DrG treatment exhibited the lowest values. The NIR region's reflectance mainly depends on internal leaf structure and water content. According to the low values of the DrG in this region we can emphasize stress or lower turgor.

Nutrient status of the plants has a significant role in vegetative growth, yield and quality, therefore regular inspection and tissue analysis is required to avoid deficiency. According to Jackson (2020) these deficiency symptoms are often distinct to be diagnostic. Therefore, remote sensing is widely applied to get information about the nutrient status. Verdenal et al. (2021) introduced hand-held instruments for the leaf nutrient content measurements and showed that macroelements, in more particular nitrogen, can influence the biomass and yield of the grapevine, and that genotypes have different nutrient demand that must be taken into consideration during the annual cultivation practice. Debnath et al. (2021) were using a hyperspectral camera to compare leaf samples obtained from N, K and Mg deficient and control groups to determine differences among the samples.

In this study we applied a SpectraVue CI-710 Leaf Spectrometer to get the reflectance spectra of samples. While the instrument provides both reflectance, transmittance and absorbance data, we analyzed only the reflectance spectra in this study. This instrument is widely applied in agricultural research, for example in rice (Székely et al., 2024), sorghum (Craigie et al., 2024), moreover on mosquito fern (*Azolla pinnata*) (Vinanthi Rajalakshmi and Paari, 2024) and Indian sandalwood (Madhuvanthi et al., 2024). Hariadi et al. (2024) predicted nitrogen, phosphate, and potassium contents of oil palm leaf with the instrument between 400 and 900 nm and developed a prediction model. They found that the red-edge region of the spectra was relevant and had a

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high correlation with the nutrient content. In this study we also found that the red-edge region showed a high variability among the samples obtained from the different treatments and phenological stages.

Evaluating the nutrient demand of grapevines is essential for high-quality production, therefore regular monitoring of macro- and micronutrient status is necessary. Several methods are available for this purpose, including soil analysis, destructive laboratory-based plant analysis and visual inspection of deficiency symptoms. Proximal and remote sensing methods based on UAV, drones or on-the-go solutions provide a non-destructive way of evaluating nutrient status that could be more representative than point sampling. For this reason, identifying those single wavebands or vegetation indices that show a high correlation with the content of each nutrient is very important. Peng et al. (2022) involved 11 vegetation indices to predict the nitrogen, phosphorus and potassium contents of Hutai No. 8 grapevine cultivar at different phenological stages based on UAV multispectral remote sensing and selected the optimal spectral variable combinations for the nutrient content estimation. They found that the B, Structure-Intensive Pigment Index (SIPI), G, False Color Vegetation Index (FCVI), Optimized Soil Adjusted Vegetation Index (OSAVI) and Normalized Difference Vegetation Index (NDVI) as input prediction variables at veraison and maturity stage provided the best prediction results. In this study we found significant differences among the vegetation indices influenced by both the treatments and the phenology. Correlation analysis showed that NDVI like CI-G and NDRE negatively correlated with N, P, K. While VARI index showed a weak positive correlation with P, Na, Mn and Zn.

Significant changes in the spectral characteristics might be linked to the leaf morphology affected by the treatments. Our former study showed that foliar applications have effect not only on chlorophyll concentration but on leaf thickness and light transmittance (Bodor-Pesti et al., 2025). Correlation analysis of the parameters investigated (chlorophyll concentration, leaf thickness and light transmittance) showed treatment-specific relationship, that underline the specific effect of the foliar applications on the leaf spectral characteristics. It has to be highlighted that not only the treatments but age of the leaves e.g., phenological stage would have a significant effect on the reflectance.

CONCLUSION

The nutrient supply and conditioning of grapevine is crucial for the health, yield and stress tolerance of the plant (Spring et al., 2003). Humic, fulvic and salicylic acid play a significant role in this process, which supports the development of the plant and the uptake and utilization of nutrients in different ways. With the different nutrient treatment combinations, our goal was to increase the condition and stress tolerance of the grape plant (drought, disease resistance) during the vegetation period. Humic acids improve the nutrient uptake of the root system, especially in the case of difficult-to-absorb microelements (e.g., ferrum, zinc, copper), while fulvic acids increase chlorophyll production, thereby improving photosynthesis. Based on the leaf analysis results, it can be seen that in the case of treatments containing humic, fulvic and salicylic acid (PLHU) and microcrystalline nutrients (DrG), there is a significant difference in the uptake of copper, ferrum and boron, while the treatment consisting of plant extracts, amino acids and micronutrients (AS) shows a positive shift only in the case of iron. In the control

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(CO), the nutrient balance shifts towards sodium, which was primarily the consequence of the unbalanced nutrient uptake during the drought period in July–August.

Results of this study showed that the reflectance spectra of the Riesling grapevine cultivar are significantly influenced by the applied foliar applications. Control samples generally exhibited lower reflectance in the blue and red regions, but higher values in the red-edge and NIR regions. This indicates higher chlorophyll content and a more intact leaf structure. In contrast, the DrG and PLHU treatments exhibited spectral traits indicative of physiological stress, such as reduced chlorophyll absorption and lower internal reflectance. According to the PCA we identified two bands: 725 and 505 nm, which had the highest loading on the PC1 and PC2 respectively. Statistical evaluations showed that both treatments and phenological stage of the sampling have a significant effect on the vegetation indices and the two selected wavebands. We found that NDVI, CI-G and NDRE had a significant negative correlation with N, P, and K content of the samples, while positive correlation with Na, Mg, Ca and Fe content. Mn content had a positive and significant correlation with NDVI, EVI, CI-G, NDRE and VARI. As we had limited numbers of samples for the comparison of the vegetation indices and nutrient status we consider the results as preliminary observations, that need to be extended with a larger sample set.

Grapevine is a woody perennial crop, which means that it takes more than a year for the fruit to develop due to bud differentiation. In addition, shoot development occurs before bud break, which emphasizes the long-term effect of biostimulants and foliar fertilizers and the need for long-term analysis of their effects. Our current research, after several years of repeating treatments and spectral analyses, can provide confirmation of the effects of the treatments. Results obtained in this study need further verification to get more data on the leaf nutrient content and spectral characteristics of the grapevine.

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