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Determination of tomato quality with hyperspectral imaging

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Abstract. Tomatoes (*Solanum lycopersicum* L.) are a widely used vegetable in the human diet throughout the year, both fresh and in various processed products. Tomatoes contain compounds important to human health and are an important source of vitamins, antioxidants, and mineral elements. Performing biochemical analyses is an expensive, environmentally unfriendly and time-consuming process; therefore, a way to determine the biochemical composition of tomatoes using non-destructive methods is being sought. The study includes 45 varieties of tomatoes with different colors - red, pink, orange, brown, yellow, and bicolor tomato fruits. The content of dry matter, soluble dry matter, titratable acidity, lycopene, β-carotene, total phenol, and flavonoids was determined by standard biochemical procedure. Reflectance spectrums of tomato fruits were obtained with Remote Sensing Portable Spectroradiometer RS-3500 (Ltd. Spectral Evolution, Haverhill, MA, USA) at the wavelength 350–2,500 nm with a 1 nm interval. In order to determine the content of various biochemical parameters in tomatoes, the vegetation indices found in the literature were used, and new ones were developed. The research demonstrated that the developed vegetative indices allow to detect lycopene and β-carotene content non-destructively. For the determination of the dry matter, soluble solids and phenolic content, indices designed for detecting water content can be used, but their correlation coefficients with chemical methods are moderately high - 0.65, 0.56 and 0.57, respectively. It was found that the best correlation between biochemically detected parameters and vegetation indices is for lycopene > β-carotene > dry matter> total phenols = titratable acidity ≥ soluble solids > taste index > flavonoids.

Key words: lycopene, taste index, total phenols, Vis-NIR, β-carotene.

INTRODUCTION

Tomato (*Solanum lycopersicum* L.) is one of the most important and widely grown vegetables in the world with large health promoting potential: a rich source of antioxidants, dietary fibres, phenolic compounds, flavonoids, mineral elements and vitamins (Tolasa et al., 2021). Global consumption of fresh and processed tomatoes is estimated at 6.4 million tons per year (FAO, 2021). More than 10,000 tomato varieties are available in the world market, differing in their fruit size, shape and colour (Moore, 2021).

Measurement of fresh tomato fruit quality is challenging. Performing biochemical analyses is an expensive, environmentally unfriendly and time-consuming process; therefore, a way to determine the biochemical composition of tomatoes using non-destructive methods is being sought. However, the destructive methods cannot be applied in high volume measurements. Among the quality indicators, the characteristics of tomato fruits, such as degree of ripeness, lycopene content and firmness, are most often explained (Vursavus & Kesilmis, 2016). In recent years, the possibilities of using non-destructive methods in the determination of carotenoids, chlorophylls, phenols, flavonoids, sugar content, acidity and aroma have also been explained (Hussain et al., 2018). Different scientific methods are used: machine vision, electronic nose technology, nuclear magnetic resonance technology, x-ray transmission, acoustic transmission and near infrared spectroscopy (NIRS), Raman imaging, hyperspectral and fluorescence imaging laser light backscattering etc. (Hussain et al., 2018; Brito et al*.,* 2022). The Visible-Near infrared (Vis-NIR) spectroscopy is increasingly common because it is a convenient, non-destructive and fast analytical method that requires minimal sample preparation (Brito et al., 2022; Duckena et al., 2023).

Spectroscopy is a widely used non-destructive testing method for fruit analysis. Near-infrared (NIR) spectroscopy is one of the most popular techniques to determine indices of postharvest vegetable quality. The latest developments in optics and sensors allow the creation of hyperspectral images with wavelength-specific measurements over a large part of the spectrum.

Hyperspectral imaging (HSI) has been widely used for non-destructive testing in various fields, including horticultural products (Huang et al., 2017), and they are effective for the quality analysis of fruits. Rahman et al. (2017) use hyperspectral imaging to estimate water content, but Rahman et al. (2018) fit the sweetness and firmness of tomatoes. Therefore, hyperspectral imaging techniques can effectively measure or classify fruit and vegetable products. HSI shows considerable promise for non-destructive analysis of horticultural and food products, and it is ideally suited to the requirements of the agro-food industry in terms of quality control (Mo et al., 2017).

Hyperspectral vegetation indices are an essential tool to monitor tomato growth, estimate the yield and analyze the quality parameters of obtained fruits. The use of vegetation indices has been demonstrated to provide high-speed analysis of fruit quality in comparison to biochemical methods (Aldubai et al., 2022).

The aim of the study is to develop vegetation indices for the determination of lycopene, β-carotene, dry matter, total phenols, titratable acidity, soluble solids, taste index, and flavonoids by using the visible and near-infrared reflectance spectra.

MATERIALS AND METHODS

Plant material

The study included 45 tomato varieties grown in the 2022 growing season in the soil in polyethylene film greenhouses without additional heating during the summer season. The fruits were harvested in the phase of full ripeness. The study included 15 red cultivars ('Aurea F1', 'Bellastar F1', 'Borsalina F1', 'Berberana F1', 'Cocktail Crush F1', 'Encore F1', 'Gourmandia F1', 'Honey Moon', 'Lancelot F1', 'Panekra F1', 'Red Trilly', 'Roseta', 'Sakura F1', 'Strabena F1', 'Sunstream F1'), eight pink cultivars ('Cassarosa F1', 'Fuji Pink F1', 'Gusto Pink F1', 'Kongo F1', 'Pink Oxheart', 'Pink Rock F1', 'Rhianna F1', and 'Rosa Star F1'), eight orange/yellow ('Apressa F1', 'Beorange F1', 'Bolzano F1', 'Kinkanstar F1', 'Orange Queen', 'Organza F1', 'Santorange F1', 'Yellow Trilly', and 'Yellow Oxheart'), three brown ('Black Cherry F1', 'Chocostar F1', 'Chocomate F1'), and ten bicolor tomato fruit cultivars ('Ananas', 'Bucanero', 'Gargamel', 'Melange', 'Santa Rosa', 'Patio Rosa', 'Gaglvando', 'Flins', 'Tayo' and 'Orange Oxheart').

Depending on the variety and fruit size, 3–12 fruits were taken for analysis. After obtaining reflectance spectrums, the same fruits were homogenized, and biochemical analyses were performed.

Biochemical analyses

In homogenized tomato mass dry matter content, soluble dry matter content, titratable acidity content, lycopene, carotene, phenol, and flavonoid content was determined. A taste index was calculated using the content of soluble solids and the content of titratable acidity. The description of the methods used is given in the article Alsina et al. (2022).

Acquisition of reflectance spectra and calculation of vegetation indices

Spectro-radiometer RS-3500 was used to obtain reflectance spectra from the surface of tomato fruits in 12 replicates from each cultivar. The reflectance was read with 1 nm resolution in the 350–2,500 nm range.

To obtain (calculate) the Vegetation indices from reflectance spectra of tomato fruits, were used:

1. Equations available in the literature (Table 1),
2. Absorption/reflection maxima and minima available in the literature (Huang et al., 2018; Hussain et al., 2018; Brito et al., 2022; Najjar & Abu-Khalaf, 2021; Xiang et al., 2022; Shao et al., 2022),
3. Values of the reflection spectra obtained in our experiments.

Table 1. Vegetation indices (VI) calculated from tomato fruit reflectance by using published equations

Vegetation index	Abbreviation	Equation	Reference
Carotenoids Index	CRI1	$\frac{1}{\mathrm{W510}} - \frac{1}{W550}$	Gitelson et al., 2001
Structure Intensive Pigment Index 1	SIPI 1	$\frac{W445 - W800}{\mathrm{W670} - \mathrm{W800}}$	Peñuelas & Filella, 1998
Lycopene Index	LYC	$\frac{W630 - W570}{\mathrm{W630} + \mathrm{W570}}$	Alsiņa et al., 2019

Table 1 (continued)

β-carotene Index	CAR	$\frac{W445 - W800}{\mathrm{W670} - \mathrm{W800}}$	Alsiņa et al., 2019
Flavonoid reflectance Index	FRI	$\left(\frac{1}{\mathrm{W410}} - \frac{1}{W460}\right) W800$	Skoczowski et al., 2021
Flavonoid reflectance Index		$lg\frac{W635}{W370}$	
Lichtenthaler index 1	LIC1 (NDVI)	$\frac{\mathrm{W800} - \mathrm{W680}}{\mathrm{W800} + \mathrm{W680}}$	Lichtenthaler, 1996
Normalized Difference Vegetation Index	NDVI	$\frac{\mathrm{W760} - \mathrm{W670}}{\mathrm{W760} + \mathrm{W670}}$	Padilla, 2017
Plant Senescence Reflectance Index	PSRI	$\frac{\mathrm{W678} - \mathrm{W500}}{\mathrm{W750}}$	Merzlyak et al., 1999
Water use efficiency Index	WBI3	$\frac{\mathrm{W950}}{\mathrm{W900}}$	Peñuelas et al., 1993
Water Index	WBI2	$\frac{\mathrm{W970}}{\mathrm{W900}}$	Peñuelas et al., 1997
Disease-Water Stress Index 2	DSWI-2	$\frac{\mathrm{W1660}}{\mathrm{W550}}$	Apan et al., 2004
Disease-Water Stress Index 5	DSWI-5	$\frac{\mathrm{W800} - \mathrm{W550}}{\mathrm{W1660} + \mathrm{W680}}$	Apan et al., 2004
Healthy Index	HI	$\frac{\mathrm{W534} - \mathrm{W698}}{\mathrm{W534} + \mathrm{W698} - 0.5\mathrm{W704}}$	Mahlein et al., 2013

In order to find the wavelengths sensitive to each investigated parameter, a correlation analysis was performed between the biochemical parameters and the average wavelength of the reflection spectrum of the respective sample biochemical parameters. Reflectance (W) with the highest correlation coefficients were used in index development. Vegetation indices were created according to the normalized index creation algorithm (Eq. 1). As base (Wbase) reflectance at the maximum/ minimum points was used or where the less fluctuations were observed.

$$VI = \frac{W - Wbase}{W + Wbase} \quad (1)$$

Totally 63 vegetation indices were generated. After calculating the vegetation indices, a correlation analysis was performed between each index and each studied parameter. The most relevant indices have been determined. To improve the correspondence to the specific parameter, a regression analysis has been performed using the two best indices with the lowest correlation between them. The obtained data were described in the text and figures as predicted values.

The statistical analysis of the data was conducted using Microsoft Office Excel 2021.

RESULTS AND DISCUSSION

The obtained reflectance spectra from tomato fruits show that visible light is the most informative zone in these spectra. For tomatoes of different colors, the largest differences are observed in the green and yellow light range (500−600 nm) (Fig. 1).

Reflectance maxima were observed at 634, 1,060, and 1,290 nm, and minima at 480, 970, and 1,410 nm. The smallest fluctuations were observed at wavelengths of 830 and 1,030 nm. Reflectance at these wavelengths has been used as a base for calculating vegetation indices.

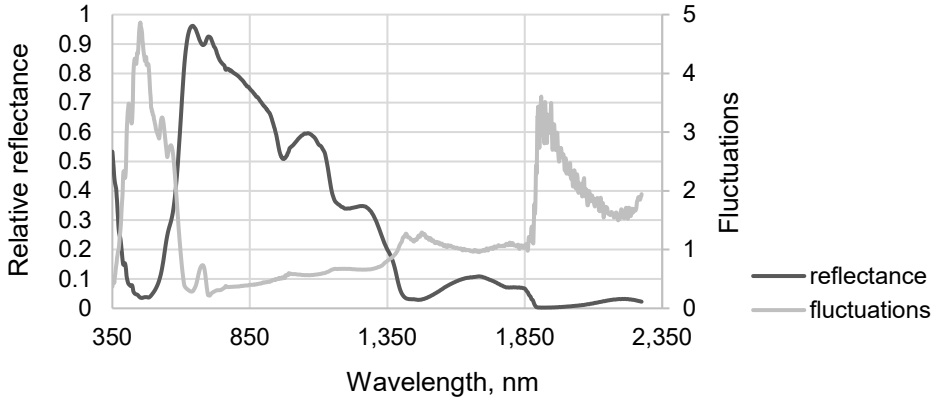

Figure 1. Average reflection spectra from tomato fruits and its fluctuations at different wavelengths.

By performing a correlation analysis between the biochemical parameters and the reflectance spectrum at each nanometre, it was found that the highest correlation coefficients for the absorption spectrum were with lycopene content. There, at the wavelength of 569 nm, the correlation coefficient with the biochemically determined lycopene content was >|0.8|. One significantly ($p \leq 0.05$) higher value of the correlation coefficient was also found for carotene - at 551 nm, the correlation coefficient was -0.74. Reflectance at these wavelengths has also been used to create vegetation indices. For the rest of the studied parameters, no distinct correlation peaks were observed between the absorption spectra and the biochemically determined parameters, but the wavelengths with the highest correlation are used to determine the vegetation indices (Fig. 2).

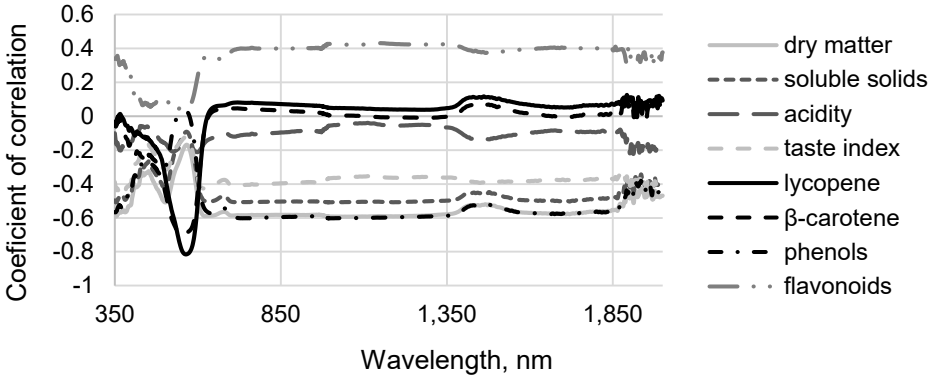

Figure 2. Variation of the correlation coefficient between reflectance and biochemically determined tomato fruits' parameters depending on the wavelength.

Dry matter

The best results for non-destructive dry matter detection were obtained using wavelengths 900 nm, 950 nm, 970 nm, 1,060 nm, 1,180 nm, 1,196 nm and 1,300 nm. Using these wavelengths in the calculation of vegetation indices, the correlation coefficient is $r = |0.5{-}0.6|$. The best results were obtained with the Water Index (WBI2) (Fig. 3, A).

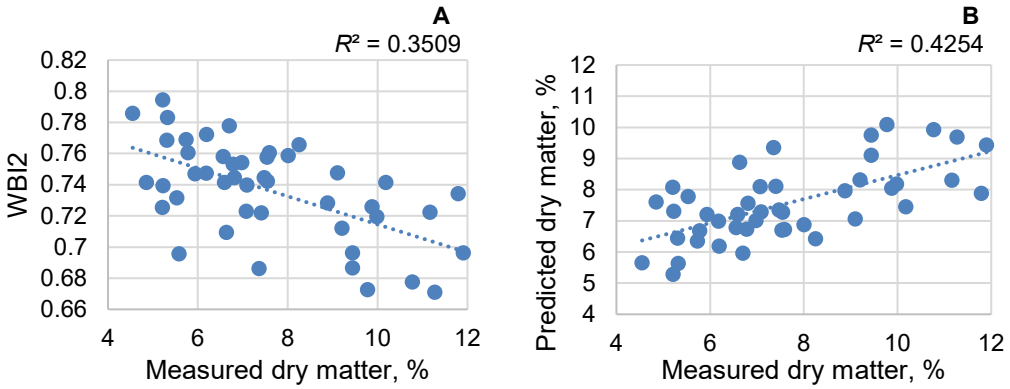

Figure 3. Dependence of the vegetation index (WBI2) and the determined dry matter (A) and relationships between predicted and measured dry matter content (B) in tomato fruits.

By using two vegetation indices to calculate dry matter, we managed to improve the correlation of parameters. Using Eq. 2, the correlation coefficient between the measured and calculated (predicted) amount of dry matter is $r = 0.652$ (Fig. 3, B).

$$VI = 31.09 - 34.2\frac{W970}{W900} - 19.33\frac{W1{,}196 - W1{,}060}{W1{,}196 + W1{,}060} \quad (2)$$

The literature mentions higher correlation coefficients for determining the amount of dry matter (Najjar & Abu-Khalaf, 2021, $R^2 = 0.7$, Brito et al., 2022, $R^2 = 0.595$).

Soluble solids

The content of soluble solids and titratable acidity is the fruit quality indicators. For non-destructive soluble solids detection the best results are obtained using wavelengths 900 nm, 950 nm, 970 nm, 995 nm, 1,060 nm, 1,180 nm, and 1,196 nm. Using these wavelengths in the calculation of vegetation indices, the correlation coefficient was $r = |0.45{-}0.57|$. Similar to dry matter, the best results were obtained with the Water Index (WBI2) (Fig. 4, A).

The use of several vegetation indices for the determination of soluble dry matter practically did not change the correlation. Using equation 3, the correlation coefficient between the measured and predicted amount of soluble solids is $r = 0.569$ (Fig. 4, B).

$$VI = 31.04 - 33.53\frac{W970}{W900} + 2.36\frac{W1{,}180 - W1{,}060}{W1{,}180 + W1{,}060} \quad (3)$$

Other researchers have reported higher correlations: Radzevičius et al. (2016) $R^2 = 0.815$, Najjar & Abu-Khalaf (2021) $R^2 = 0.97{-}0.98$, Huang et al. (2018) $R^2 = 0.642$, Brito et al. (2022) $R^2 = 0.79{-}0.99$, Xiang et al. (2022), $R^2 = 0.577$, with comments that accuracy depends on used model and data set.

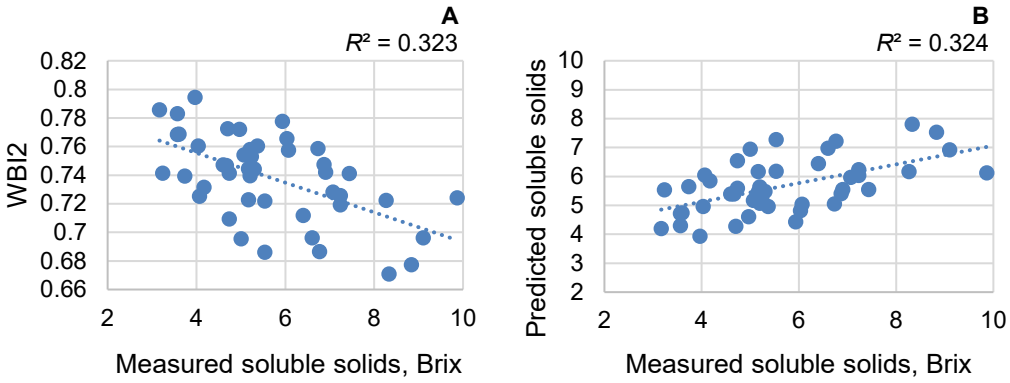

Figure 4. Dependence of the vegetation index (WBI2) and the determined soluble solids content, Brix (A) and relationships between predicted and measured soluble solids content (B) in tomato fruits.

Titratable acidity

The best wavelengths for titratable acidity assessment were 365 nm, 710 nm, 1,060 nm, 1,440 nm and 1,460 nm. Using these wavelengths in the calculation of vegetation indices, the correlation coefficient $r = |0.45{-}0.54|$. The best correlation between titratable acidity and the established vegetation index (VI_53) was 0.536. (Eq. 4, Fig. 5, A).

$$VI\_53 = \frac{W365 - W1{,}060}{W365 + W1{,}060} \quad (4)$$

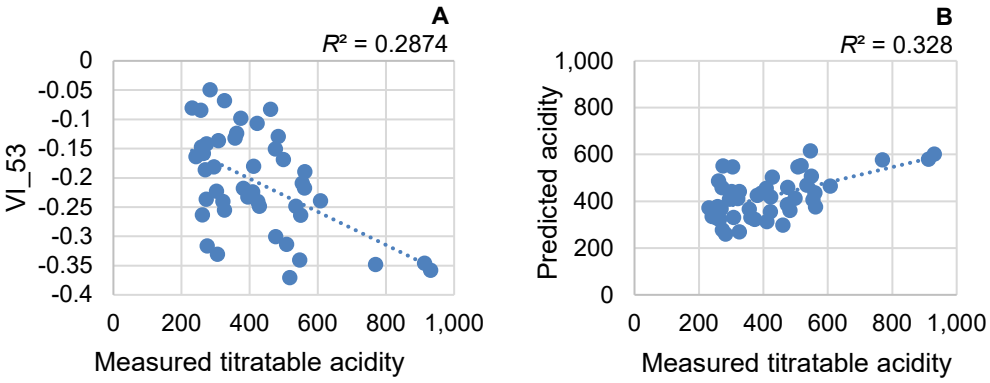

Figure 5. Dependence of the vegetation index (WVI_53) and the determined titratable acidity, mg of citric acid per 100 g of fresh tomato weight (A) and relationships between predicted and measured titratable acidity (B).

By using two vegetation indices, it was possible to significantly improve the correlation between the parameters (Eq. 5, Fig. 5, B), but unfortunately it was not possible to obtain such high results as mentioned in the literature: Huang et al. (2018), $R^2 = 0.656$ (pH was determined), Najjar & Abu-Khalaf (2021), $R^2 = 0.91{-}0.98$, Brito et al. (2022), $R^2 = 0.70{-}0.98$.

$$VI = -166 - 748\frac{W365 - W1{,}060}{W365 + W1{,}060} + 2015\frac{W1{,}060 - W1440}{W1{,}060 + W1440} \quad (5)$$

Taste index

Organic acids, soluble sugars, and pigments are other compounds that contribute to the taste development of tomatoes. In our study, the taste index was determined as the ratio of soluble solids and titratable acidity and for the non-destructive assessment of taste index and the best wavelengths for its non-destructive determination were 365 nm, 370 nm, 635 nm, 900 nm, 995 nm, 970nm, 1,060 nm, 1,440 nm and 1,460 nm. Using these wavelengths in the calculation of vegetation indices, the correlation coefficient $r = |0.42–0.51|$. The best correlation between the taste index and the established vegetation index (VI_53) was 0.512 (Eq. 4, Fig. 6, A).

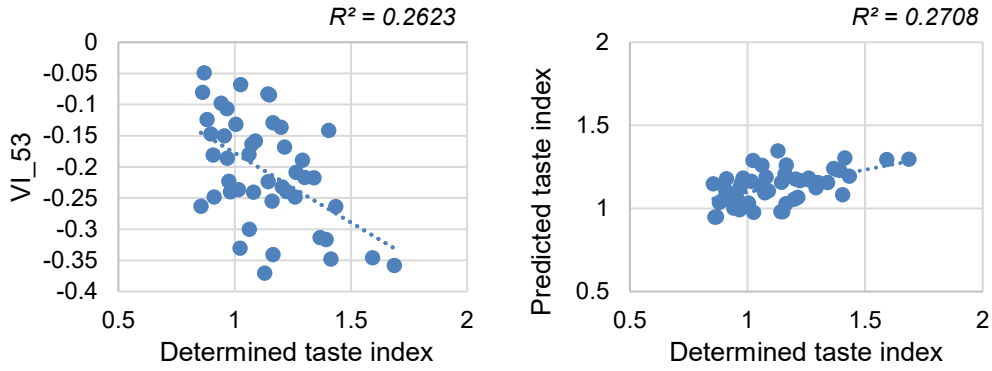

Figure 6. Dependence of the vegetation index (VI_53) and the determined tomato taste index (A) and relationships between predicted and measured taste index (B).

The use of two vegetation indices allows for improvement in the determination of the taste index (Eq. 6, Fig. 6, B).

Correlation indices between measured and predictable taste indices mentioned in the literature are Huang et al. (2018), $R^2 = 0.656$ (pH was determined) Najjar & Abu-Khalaf (2021), $R^2 = 0.91–0.94$, Sun et al. (2021), $R^2 = 0.702–0.917$ (sensory detected taste).

$$VI = 0.789 - 1.04\frac{W365 - W1{,}060}{W365 + W1{,}060} + 0.12\lg\frac{W635}{W370} \quad (6)$$

Lycopene

Lycopene is the pigment responsible for the deep-red colour of ripe tomatoes. For the determination of lycopene content in tomato fruits, non-destructive methods proved to be the most suitable. Eight out of 63 vegetation indices showed a correlation, where the coefficient was higher than $r = |0.9|$. The most sensitive wavelengths used in detecting lycopene were 551 nm, 560 nm, 564 nm, 570 nm, 630 nm, 634 nm, 734 nm, 990 nm and 1,060 nm. Most of them are in the yellow-orange part of the visible light spectrum. This shows that, in fact, the whole part of the yellow spectrum can be used to determine the lycopene content. The highest correlations were obtained using Eq. 7. The use of two vegetation indices (Eq. 8) does not justify itself, as the correlation coefficients change little (Fig. 7).

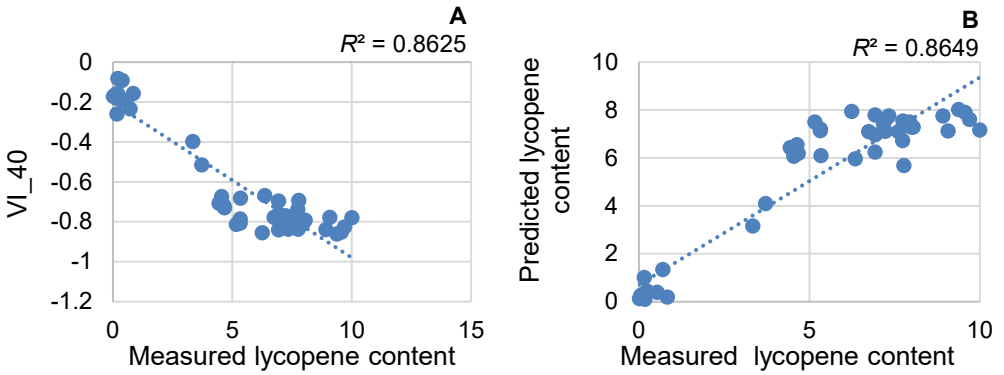

Figure 7. Dependence of the vegetation index (VI_40) and the determined lycopene content, mg 100 g^{-1} (A) and relationships between predicted and measured lycopene content (B) in tomato fruit.

$$VI\_40 = \frac{W551 - W725}{W551 + W725} \tag{7}$$

$$VI = -1.37 - 7.35\frac{W551 - W725}{W551 + W725} + 3.52\frac{W643 - W564}{W643 + W564} \tag{8}$$

In the scientific articles published by other authors, the lycopene content is a parameter whose biochemically determined content correlates well with various non-destructively determined ones. Brito et al. (2022) reports $R^2 = 0.96$, Saad et al. (2017) $R^2 = 0.88$, Tilahun et al. cited from Brito et al. (2022) $R^2 = 0.89$, Alenazi et al. (2020) $R^2 = 0.864$.

β-carotene

Similar to the lycopene, also for β-carotene, the best correlation between the biochemically determined amount and the non-destructive method can be observed in the visible light spectrum, unfortunately the correlation coefficients are on average 8–12% lower. The best correlation ($r = 0.854$) was obtained with the reflectance at 538 and 710 nm. (Eq. 9, Fig. 8, A).

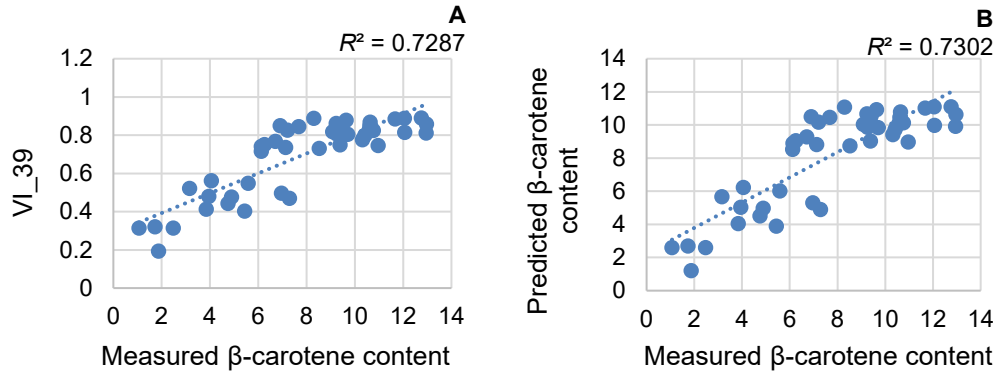

Figure 8. Dependence of the vegetation index (VI_39) and the determined β-carotene content, mg 100 g^{-1} (A) and relationships between predicted and measured β-carotene content (B) in tomato fruits.

$$VI\_39 = \frac{W710 - W538}{W710 + W538} \quad (9)$$

$$VI = -2.01 + 14.43\frac{W710 - W538}{W710 + W538} - 0.32\frac{W643 - W552}{W643 + W552} \quad (10)$$

The use of two vegetation indices did not significantly improve the correlation between the determined and predicted carotene content (Eq. 10, Fig. 8, B).

Similar to lycopene, visible light is also used to detect β-carotene. Correlation coefficients in other authors' publications were Saad et al. (2017) $R^2 = 0.91$ (total carotenoids $R^2 = 0.85$), Ibrahim et al., cited from Brito et al. (2022) $R^2 = 0.92$, 0.938, Alenazi et al. (2020) $R^2 = 0.708$.

Phenols

Phenolic compounds are an important group of organic molecules with high radical scavenging, antimicrobial, anti-inflammatory, and antioxidant properties (Razem et al., 2022). The non-destructive determination of the phenolic content is problematic, as it was not possible to find indices developed for the determination of these compounds in the literature. The best correlation was found in those wavelengths that can also be used to determine dry matter (water content) and soluble solids content, and it is 900 nm, 970 nm, 995 nm, 1,060 nm, 1,180 nm, and 1,300 nm. Correlation coefficients of the vegetation indices created by reflectance at these wavelengths with chemically detected are in the range of $r = |0.47–0.56|$. The highest correlation was observed with VI_32 (Eq. 11, Fig. 9).

$$VI\_32 = \frac{W900 - W970}{W900 + W970} \quad (11)$$

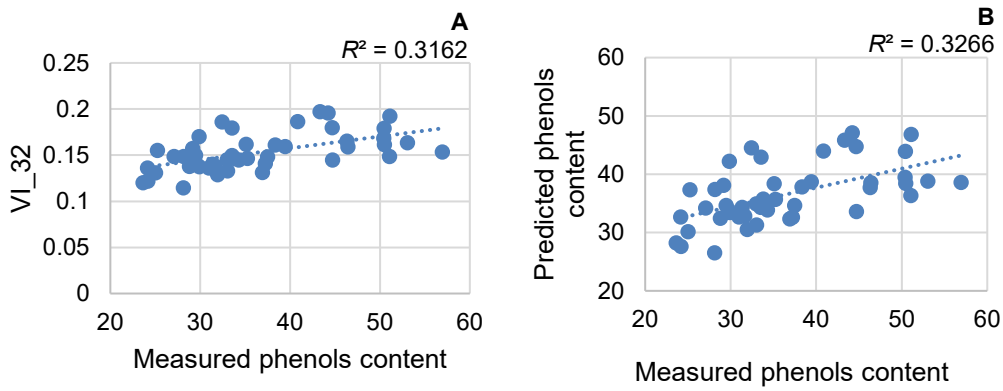

Figure 9. Dependence of the vegetation index (VI_32) and the determined phenols content, gallic acid equivalent (GAE) per 100 g of fresh tomato mass (A) and relationships between predicted and measured phenols content (B) in tomato fruits.

$$VI = 0.754 + 204.7\frac{W900 - W970}{W900 + W970} - 277.3\frac{W995 - W1{,}060}{W995 + W1{,}060} \quad (12)$$

The use of two vegetation indices improves the correlation from $r = 0.562$ to 0.571. Better phenol determination accuracy was reported in the study by Alenazi et al. (2020) $R^2 = 0.834$, and Szuvandzsiev et al. (2014) $R^2 = 0.81$.

Flavonoids

Flavonoids are secondary metabolites found in plants. The direct correlation between flavonoid intake and decreased risk of cardiovascular disease, cancer, and age-related diseases is reported. Unlike phenols, non-destructive detection methods have been developed for flavonoids, but in most cases, they involve the use of UVa radiation. Reflection in the blue-violet part of the visible light spectrum is also used. It was not possible to achieve a correlation coefficient higher than $r>|0.4|$ with the indices found in the literature, nor with the self-generated ones. Correlation coefficients between 0.35 and 0.4 were found for five indices, the best of which is VI_62, where reflectance at wavelengths 1,180 nm and 1,060 nm were used (Fig. 10, A & B).

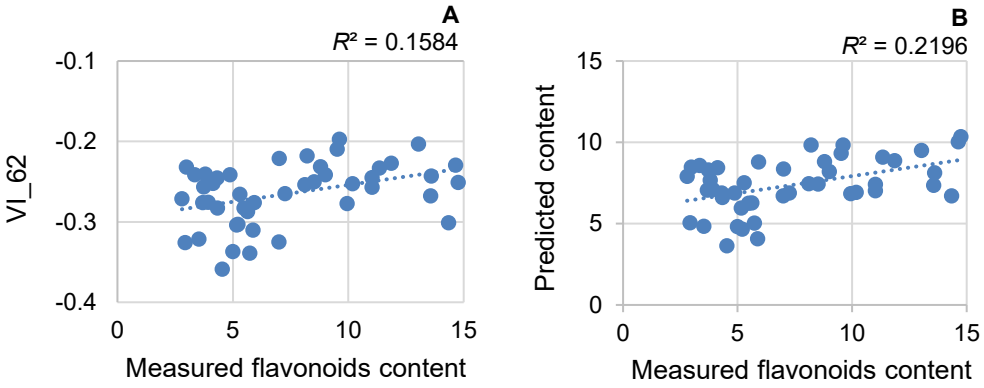

Figure 10. Dependence of the vegetation index (VI_62) and the determined flavonoids content, amount of quercetin equivalents (QE) per 100 g of fresh tomato mass (A) and relationships between predicted and biochemically measured flavonoids content (B).

$$VI = 18.1 + 32.3\frac{W1{,}180 - W1{,}060}{W1{,}180 + W1{,}060} - 24.6\frac{W620 - W1{,}060}{W620 + W1{,}060} \quad (13)$$

Although using the two vegetation indices, the correlation coefficient increased from 0.398 to 0.469, unfortunately it is insufficient; therefore, further research is required.

There is little information in the scientific literature on the non-destructive determination of flavonoid content in tomato fruits. Alenazi et al. (2020) reports a relatively high correlation between the amount of flavonoids determined biochemically and that obtained by NIR spectrometry $R^2 = 0.790$.

Both our data and those of other authors show that non-destructive methods are suitable for determining the color of tomatoes. Good results were obtained in the non-destructive determination of lycopene and carotene. However, other authors have managed to obtain a significant correlation also between soluble solids and phenolic content. Our results are not so conclusive. This can be explained by the selection of the tomato sample. In the works of most other authors, tomatoes of several varieties or different degrees of ripeness of the same variety are used. This allows to reduce data dispersion. In order to increase the reliability of the results, future studies should develop indices for tomatoes with different fruit color. Although NIR wavelengths exceeding 1500 nm are mentioned in various literature sources for the determination of sugars,

dry matter, titratable acidity, and phenolic content, in our research the reflectance at these wavelengths proved to be of little information. For the determination of these parameters, the wavelengths corresponding to the absorption/reflection spectra of water molecules turned out to be the most informative. Determination of flavonoid content with the spectro-radiometer at our disposal was problematic because both the wavelengths indicated in the literature and those we found did not give a correlation whose coefficient would be higher than |0.4|. The solution could be the development of models or the use of hardware that uses UV radiation. A good correlation found between biochemical measurements and the Vis/NIRs (500–750 nm) allows us to predict that the evaluation of tomato quality indicators using these wavelengths could become a routine practice.

CONCLUSIONS

1. The research demonstrated that the developed vegetative indices allow to detect lycopene and β-carotene content non-destructively with good prediction accuracy, and their correlation coefficients with biochemically measured ones were 0.93 and 0.85, respectively.
2. For the determination of the dry matter, soluble solids and phenolic content, vegetation indices designed for detecting water content can be used, but their correlation coefficients with chemical methods are moderately high - 0.65, 0.56 and 0.57, respectively.
3. It was found that the best correlation between biochemically detected parameters and vegetation indices is for lycopene > β-carotene > dry matter > total phenols = titratable acidity ≥ soluble solids > taste index > flavonoids.

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