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Estimation of Apple Tree Leaf Chlorophyll Content Based on Machine Learning Methods

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Abstract: Leaf chlorophyll content (LCC) is one of the most important factors affecting photosynthetic capacity and nitrogen status, both of which influence crop harvest. However, the development of rapid and nondestructive methods for leaf chlorophyll estimation is a topic of much interest. Hence, this study explored the use of the machine learning approach to enhance the estimation of leaf chlorophyll from spectral reflectance data. The objective of this study was to evaluate four different approaches for estimating the LCC of apple tree leaves at five growth stages (the 1st, 2nd, 3rd, 4th and 5th growth stages): (1) univariate linear regression (ULR); (2) multivariate linear regression (MLR); (3) support vector regression (SVR); and (4) random forest (RF) regression. Samples were collected from the leaves on the eastern, western, southern and northern sides of apple trees five times (1st, 2nd, 3rd, 4th and 5th growth stages) over three consecutive years (2016–2018), and experiments were conducted in 10-20-year-old apple tree orchards. Correlation analysis results showed that LCC and ST, LCC and vegetation indices (VIs), and LCC and three edge parameters (TEP) had high correlations with the first-order differential spectrum (FODS) (0.86), leaf chlorophyll index (LCI) (0.87), and $(SD_r - SD_b)/(SD_r + SD_b)$ (0.88) at the 3rd, 3rd, and 4th growth stages, respectively. The prediction models of different growth stages were relatively good. The MLR and SVR models in the LCC assessment of different growth stages only reached the highest R² values of 0.79 and 0.82, and the lowest RMSEs were 2.27 and 2.02, respectively. However, the RF model evaluation was significantly better than above models. The R² value was greater than 0.94 and RMSE was less than 1.37 at different growth stages. The prediction accuracy of the 1st growth stage (R²=0.96, RMSE = 0.95) was best with the RF model. This result could provide a theoretical basis for orchard management. In the future, more models based on machine learning techniques should be developed using the growth information and physiological parameters of orchards that provide technical support for intelligent orchard management.

Keywords: leaf chlorophyll content; hyperspectral remote sensing; RF; SVR

1. Introduction

The Leaf chlorophyll content (LCC) represents the photosynthetic rate, nitrogen content and health status of crops; it is also an indicator of the nutrient status of crop plants and the degree of senescence [1]. It has significance for crop growth monitoring, nutrient content monitoring, quality evaluation and yield estimation [2]. LCC is essential because the nutritional status of crops determines yield and food safety. In the past, traditional methods of its calculation were time-consuming, laborious, and destructive to leaves [3]. Therefore, the accurate estimation of LCC is an important challenge that urgently needs to be addressed.

In recent years, hyperspectral remote sensing (HRS) has become a major development trend in monitoring the chlorophyll content of vegetation due to its high spectral

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Copyright: © 2021 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (http://creativecommons.org/licenses/by/4.0/). resolution, simplicity, effectiveness, and non-destructiveness [1,4,5]. This method provides an effective means for research on the chlorophyll content of crops and the implementation of precision agriculture. The spectral indices captured by hyperspectral sensors are considered to be quantitative indicators of vegetation vigor [6], and continuous narrow bands have the potential to identify regions sensitive to specific crop parameters. It is well known that the spectral properties of crop leaves are controlled by surface characteristics, internal structure, and concentration of biochemical constituents [7]. Vegetation indices (VIs) have been derived from different mathematical combinations (simple ratios and normalized difference, etc.) of spectral bands to help indicate plant features. Vegetation indices have been used to detect the seasonal variability of LCC, which has been widely used in precision agriculture and crop phenotyping [8–10].

Recently, machine-learning techniques have been widely used to build predictive models with improved prediction accuracy. Shah et al. reported that the RF method could better reduce the root mean square error (RMSE) than standard linear regression [8]. Li et al. showed that support vector machine regression (SVMR) methods could better estimate chlorophyll content than the back-propagation neural network (BPNN) method [11]. There is not only a linear relationship between crop physiological parameters and the spectrum but also a nonlinear relationship. Machine learning technology has been extensively applied for crop LCC [8], leaf nitrogen concentration (N) [12], and leaf area index (LAI) [13] estimation.

Apple trees are perennials, and their phenotypic structure and growth characteristics are different from those of field crops, causing large differences in spectral response characteristics. Apple (Malus pumila Mill.) is a kind of deciduous tree, which generally begins to bear fruit 3–5 years after planting, but only apple trees with a fruit age of 10–20 years are in the full fruit period. The chlorophyll content of apple tree leaves varies with changes in different apple phenology phases, leading to different leaf photosynthetic rates at different growth stages, which causes differences in the accumulation of organic matter, which may have an impact on apple yield. According to the annual cycle and law of nutrient transportation, the leaves of the plants gradually turn yellow after apple harvest, so dynamic monitoring the chlorophyll content of apple leaves from the flowering period to the harvest period has great significance [11,14]. Many researchers have performed LCC evaluations with hyperspectral remote sensing technology for a variety of crop species (such as winter wheat, rice and maize) [15–17], but estimation of LCC in apple trees is limited, and especially the dynamic monitoring of the chlorophyll content of apple tree leaves in different phenological periods is even rarer. However, China's apple planting area has grown to account for 42.24% of the world's total, and it has become the world's largest apple producer [18]. Shaanxi Province is one of the main apple production areas in China. Apple trees play an indispensable role in the local economy, and nondestructive measurement technology is urgently needed to measure the biochemical parameter indices of orchards. In this context, the accurate estimation of LCC for apple orchards can enable government management departments to formulate appropriate orchard management measures, such as optimizing regional planting structures and fertilization practices. It is imperative, therefore, to find a suitable method for estimating LCC in apple orchards.

In this study, LCC was measured and assessed at five growth stages (1st, 2nd, 3rd, 4th and 5th growth stages) with apple trees of 10–20-year old orchards. The objectives of this study were as follows: (1) to select spectral parameters suitable and applicable for apple tree LCC estimation and (2) to determine the suitability and general applicability of the methods for measuring apple tree LCC.

2. Materials and Methods

2.1. Study Area and Experimental Setup

The study was conducted during the 2016–2018 growing seasons (from May to October) in orchards located in Xinglin town, Fufeng County, Shaanxi Province, China (W 107°45′–108°03′, N 34°12′-34°37′) (Figure 1). This region has a temperate, continental, semiarid climate. The average annual precipitation and average annual temperature are 591.9 mm and 12.4 °C, respectively.

The average height of the apple trees was 2.5 m, the average crown width was 2.5 m, and the spacing was 4 m × 3 m in the apple orchard, which ensured good light conditions and suitable growth temperatures throughout the year. The soil in the apple orchard was a Loess agricultural soil. The basic fertility level of the cultivated soil layer was as follows: the average content of organic matter was 18.65 g/kg, the alkali hydrolyzed nitrogen was 76.68 mg/kg, the available phosphorus was 35.19 mg/kg, and the quick-acting potassium content was 114.91 mg/kg.

From May to October 2016–2018, 17–20 apple trees were selected in 10–20-year-old apple orchards and collected five times (the final flowering stage, fruiting stage, fruit expansion stage, fruit coloring growth stage, and fruit ripening stage) each year. Due to the weather, data from only four growth stages were collected in 2016 (Table 1). Five healthy apple leaves were collected evenly from the upper layer to the lower layer of the canopy in the four directions of the apple tree. A total of 20 apple leaves were collected from each apple tree. The $17 \times 20 = 340$ (in 2016) and $20 \times 20 = 400$ (in 2017 and 2018) samples were placed into a freshness protection package, and the packages were then placed in an ice box.



Figure 1. Study area in this study.

2.2. Hyperspectral Data Acquisition

A spectroradiometer SVC HR1024i (Spectra Vista Crop., Poughkeepsie, NY, USA) with 1024 spectral channels and a spectral range of 350–2500 nm was used to measure the spectral reflectance of apple leaves. Spectral resolutions of 3.5, 9.5 and 6.5 nm with spectral ranges of 350–1000, 1000–1890 and 1890–2500 nm [19], respectively, were used. Because the chlorophyll response band to the leaf spectrum is mainly in the visible and reflected infrared bands, the 350–1000 nm band was primarily used for the analysis, and the spectrum was resampled to 2 nm. The spectra of leaves were measured at three randomly selected sample points. Six measurements were conducted for each sample leaf, and 30 spectral measurements (5 \times 6) were averaged to represent the reflectance of the leaves. The numbers of spectral samples in this study are shown in Table 1.

Growth Stages	2016	2017	2018	Total
1st (1)		68	80	148
2nd (2)	68	80	80	228
3rd ⁽³⁾	68	80	80	228
4th (4)	68	80	80	228
5th ⁽⁵⁾	68	80	80	228
All (6)	272	388	400	1060

Table 1. Different growth stages of the samples used in this study.

Note: ⁽¹⁾ 1st is the final flowering stage, ⁽²⁾ 2nd is the fruiting stage, ⁽³⁾ 3rd is the fruit expansion stage, ⁽⁴⁾ 4th is the fruit coloring growth stage, ⁽⁵⁾ 5th is the fruit ripening stage, and ⁽⁶⁾ All represents all growth stages combined.

2.3. Leaf Chlorophyll Content Measurement

In this study, leaf LCC was determined by collecting samples from point leaves (corresponding to spectral sampling) via a hand-held chlorophyll meter (SPAD-502, Minolta Osaka Company Ltd., Tokyo, Japan) in the laboratory. The chlorophyll meter mainly uses leaf transmittance in the central band of 650 to 940 nm to determine the chlorophyll content [20], and the SPAD value can better reflect the change in leaf greenness. Each sample was collected from the same location on the leaf where the spectral data were sampled. Six measurements were conducted for each sample leaf. The 30 value measurements (5 × 6) were averaged to represent the SPAD values of the leaves.

2.4. Methods

2.4.1. Spectral Transformation (ST)

In this study, the spectral response in Vis-NIR infrared bands at 350–1000 nm was used to estimate the apple tree LCC. All reflectance spectra were resampled at 2 nm spectral intervals [21]. In addition, widely used preprocessing methods were employed in this study, and labeling of the original spectrum (OS) was performed. Then, three types of common STs were used, including a reciprocal transformed spectrum (RS), first-order differential spectrum (FODS) and continuum removal spectrum (CRS) [19,22,23] (Table 2).

2.4.2. Vegetation Indices

The construction of a vegetation index involves the combination of different sensitive bands to eliminate the impact of environmental background effects (such as non-vegetation target soil, water body, etc.) to a certain extent. The vegetation indices evaluated in this study are presented in Table 2. NDVI [24] and mSR₇₀₅ [25] were selected because they are the earliest and simplest VIs, and their narrowband versions have great potential to improve LCC estimation accuracy. The leaf chlorophyll index (LCI) is a vegetation index involving the red-side band, that uses the spectral reflectance characteristics of the red-side band and the near-infrared band to show differences in chlorophyll content [26]. The ratio vegetation index (RVI) is based on the red and near-infrared bands [27].

2.4.3. Three Edge Parameters

To further study the relationship between spectral data and LCC, the preprocessing of spectral data is required. The "three edge" parameters refer to the relevant variables based on the characteristics of the spectral position, namely, red edge, blue edge and yellow edge [28]. The "three edge" parameters can better reflect the spectral characteristics of green vegetation and are sensitive to LCC changes. Therefore, these parameters can be used as a diagnostic characteristic parameter for LCC. The relevant definitions of the "three edge" parameters are shown in Table 2.

Parameters	Explanation	References	
ST ⁽¹⁾			
OS	Original spectrum	[19]	
RS	Reciprocal transformed spectrum	[19]	
FODS	First-order differential spectrum	[19]	
CRS	Continuum removal spectrum	[19]	
VIs ⁽²⁾			
Normalized difference Vegetation index (NDVI)	$(R_{800} - R_{670})/(R_{800} + R_{670})$	[24]	
Ratio vegetation index (RVI)	R765/R720	[27]	
Green ratio vegetation index (GRVI)	$(R_{620} - R_{506})/(R_{620} + R_{506})$	[29]	
Photochemical reflectance Index (PRI)	$(R_{570} - R_{531})/(R_{570} + R_{531})$	[30]	
Normalized pigment chlorophyll index (NPCI)	$(R_{642} - R_{432})/(R_{642} + R_{432})$	[31]	
Modified red edge simple ratio index (mSR705)	$(R_{750} - R_{445})/(R_{705} - R_{445})$	[25]	
Plant pigment ratio (PPR)	$(R_{503} - R_{436})/(R_{503} + R_{436})$	[32]	
Structure intensive pigment index (SIPI)	$(R_{800} - R_{445})/(R_{800} - R_{680})$	[33]	
Normalized difference spectral index (NDSI)	$(R_{813} - R_{763})/(R_{813} + R_{763})$	[34]	
Leaf chlorophyll index (LCI)	$(R_{850} - R_{710})/(R_{850} - R_{680})$	[26]	
TEP (3)			
	First-order differential spectrum maximum in	[25]	
b	the wavelength range of 490~530 nm	[55]	
D	First-order differential spectrum maximum in	[25]	
Dy	the wavelength range of 560~640 nm	[55]	
D-	First-order differential spectrum maximum in	[35]	
	the wavelength range of 680~760 nm	[55]	
SD	First-order differential spectral integration in	[35]	
	the wavelength range of 490~530 nm	[55]	
SD.	First-order differential spectral integration in	[35]	
	the wavelength range 560~640 nm	[88]	
SD:	First-order differential spectral integration in	[35]	
	the wavelength range of 680~760 nm	[88]	
SDr/SDb	Ratio of the red edge area to the blue edge area	[35]	
SDr/SDv	Ratio of the red edge area to the yellow edge	[35]	
	area	[88]	
$(SD_r - SD_h)/(SD_r + SD_h)$	Normalized value of the red edge area and the	[35]	
	blue edge area	[20]	
$(SD_r - SD_v)/(SD_r + SD_v)$	Normalized value of the red edge area and the	[35]	
	yellow edge area	[00]	

Table 2. Spectral parameters evaluated in this study.

Note: ⁽¹⁾ spectral transformation (ST); ⁽²⁾ vegetation indices (VIs); ⁽³⁾ three edge parameters (TEP).

2.4.4. Linear Regression Analysis

To date, ULR and MLR are the most popular statistical models for estimating LCC [36–39]. Some regression models (linear, logarithmic, and exponential models) were chosen based on the highest coefficient of determination (R²) and the lowest root mean square error (RMSE). Even though regression analysis is simple to perform, fast to model, and especially useful when the variable region is not particularly complicated, other more advanced data analytics methods likely required to take advantage of highly multiplex and multidimensional hyperspectral datasets.

2.4.5. Random Forest (RF) Regression

RF regression is based on the decision tree method and bagging method with an additional layer of randomness in the bagging process [40]. The random forest regression model is a nonparametric regression technique. In the process of model establishment, the assignment of *ntree* and *mtry* is automatically adjusted by Python 3.6.0. The RF algorithm is as follows: first, a bootstrap sample is drawn *ntree* times from the original dataset, and each bootstrap sample is used to build a tree; then, an unpruned tree is created for each bootstrap sample, and only randomly selected *mtry* predictors are used for each tree. Finally, predictions are made by aggregating the *ntree* tree prediction results, where the aggregation strategy is generally based on the majority of votes for classification and the average for regression. *ntree* and *mtry* are the two key parameters that control the performance and complexity of RF modes. In this study, *mtry* was set to 1/3 of the number of independent variables, and *ntree* was set to 500, as suggested by Breiman [40].

2.4.6. Support Vector Regression (SVR)

SVR is a machine learning method based on statistical learning theory. It maps input space (ST, VIs, and TEP) to high-dimensional space through nonlinear transformation, analyses the data and builds estimation models. Small samples and nonlinear aspects have advantages that traditional learning methods do not have and use more rigorous mathematical theoretical foundations [41].

An SVR model mainly achieves the fitting of nonlinear functions by selecting different inner product kernel functions. In practical applications, commonly used kernel functions are linear functions, polynomial kernel functions, and radial basis kernel functions. The key to success with an SVR model is to choose the penalty parameter *C* and the kernel parameter γ . The penalty parameter *C* controls the degree of punishment for the sample beyond the error and determines the impact of the empirical risk generated by the training sample on the performance of the model, the structural risk and the empirical risk relationship. The kernel parameter g controls the regression error of the model and affects the complexity of the distribution of the sample data in the high-dimensional feature space. Therefore, this paper used the grid search method to select the parameters to obtain the best estimation results. *C* and γ were optimized within [10⁻², 10⁻¹, 1, 10, 100] and [10⁻⁴, 10⁻³, 10⁻², 10⁻¹, 1, 10], respectively.

2.5. Data Analysis

Data from three consecutive years (2016–2018) were collected and then randomly divided into a calibration dataset (2/3) and a validation dataset (1/3). The total of each growth stage of the calibration and validation datasets is shown in Table 3. In the analyzed LCC, the coefficient of variation (CV) of the calibration datasets was 11.49–13.63%, the CV of the validation datasets was 11.70–13.40%, and the CV of both datasets exhibited variability (Table 3).

Crowth Stagos	Calibration Datasets					Validation Datasets			
Growth Stages	n	Range	Mean ± SD	<i>C</i> _v (%)	n	Range	Mean ± SD	<i>C</i> _v (%)	
1st	99	33.20-54.15	41.94 ± 4.87	11.61	49	32.25-53.20	42.65 ± 4.85	11.38	
2nd	152	33.05-58.20	45.93 ± 5.28	11.49	76	31.55-59.50	46.64 ± 5.46	11.70	
3rd	152	33.50-61.22	48.27 ± 5.88	12.17	76	34.65-58.70	48.38 ± 5.67	11.72	
4th	152	32.10-63.40	46.22 ± 6.30	13.63	76	31.35-59.05	45.87 ± 5.86	12.79	
5th	152	25.45-61.50	45.25 ± 5.57	12.31	76	31.25-59.80	44.46 ± 5.60	13.40	
All	707	25.45-61.50	45.74 ± 6.01	13.14	353	32.05-63.40	45.93 ± 5.79	12.62	

Table 3. Summary of LCC measurements for each growth stage.

2.6. Calibration and Validation

The scikit-learn library [42,43] was used to establish models for the estimation of LCC using two common machine learning methods: RF regression and SVR. Ten-fold cross-verification and grid searching were used to identify the optimal parameters during model development. The coefficient of determination (R²) and root mean square error (RMSE) were used to measure the predictive performance of each estimation model by different methods. Higher values of R² and lower values of RMSE indicate better dependability and accuracy of the regression model in predicting LCC.

3. Results

3.1. Descriptive Analysis of Measured LCC

The range of LCC was 25.45 to 63.40, as shown in Table 3. The LCC increased in young expanding leaves, reached the highest value at maturity, and then decreased significantly during senescence. The measured LCC increased gradually from the 1st growth stage to the 3rd growth stage and decreased to a minimum at the 5th growth stage (Figure 2).



Figure 2. Measured LCC in this study.

3.2. Selection of Sensitivity Parameters

Table 4 shows that the outcomes of the correlation analysis between the four STs and LCC were extremely significant. The CRS had the highest significant correlation with LCC in the 1st and 5th growth stages and all stages together, with values of 0.85, 0.84 and 0.74, respectively. At the 2nd, 3rd and 4th growth stages, FODS had the highest significant correlation with LCC, with correlation coefficients of 0.81, 0.86 and 0.85, respectively. All the sensitive wavebands of STs were selected in the visible band: CRS at 750 nm, FODS at 730 nm, FODS at 732 nm, FODS at 514 nm, CRS at 710 nm, and CRS at 718 nm at the 1st to 5th and All growth stages, respectively (Figure 3).

The relationships between the NDSI and LCC at the 1st growth stage had the highest correlation, with a correlation coefficient of 0.80. Except for the 1st growth stage, the correlation between LCI and LCC was highest at the 2nd, 3rd, 4th, 5th and All growth stages, and the correlation coefficients were 0.79, 0.87, 0.86, 0.84, and 0.75, respectively. As shown in Table 4, SDr/SDb, SDr/SDy, and (SDr – SDb)/ (SDr + SDb) had the best correlation with LCC at different growth stages.



Figure 3. Correlation coefficients between LCC and spectral transformation at the different growth stages. (a) 1st growth stage; (b) 2nd growth stage; (c) 3rd growth stage; (d) 4th growth stage; (e) 5th growth stage; (f) All growth stages. OS: original spectrum; RS: reciprocal transformed spectrum; FODS: first–order differential spectrum; CRS: continuum removal spectrum.

Tal	le 4. Correlation relationship between LCC and different variables.

Parameters	1st	2nd	3rd	4th	5th	All
ST (1)						
OS	0.75**	0.65 **	0.80**	0.82 **	0.77 **	0.67**
RS	0.78**	0.66 **	0.79**	0.83 **	0.77 **	0.67 **
FODS	0.81**	0.81 **	0.86**	0.85 **	0.78 **	0.73 **
CRS	0.85**	0.80 **	0.59**	0.84 **	0.84 **	0.74 **
VIs ⁽²⁾						
NDVI	0.33**	0.04	0.42**	0.68 **	0.63 **	0.37 **
RVI	0.74**	0.74 **	0.85 **	0.83 **	0.83 **	0.73**
GRVI	0.70**	0.03	0.56**	0.70 **	0.61 **	0.52**
PRI	0.57**	0.23 **	0.23**	0.55 **	0.50 **	0.34 **
NPCI	0.29**	0.67 **	0.63 **	0.69 **	0.67 **	0.53 **
mSR	0.69**	0.60 **	0.79**	0.85 **	0.79 **	0.66 **
PPR	0.05	0.67 **	0.51**	0.57 **	0.51 **	0.39 **
SIPI	0.28**	0.55 **	0.34 **	0.30 **	0.27 **	0.23 **
NDSI	0.80**	0.13	0.41 **	0.64 **	0.46 **	0.43 **
LCI	0.69**	0.79 **	0.87**	0.86 **	0.84 **	0.75 **
TEP (3)						
D_{b}	0.78**	0.76 **	0.68**	0.84 **	0.75 **	0.72**
D_{y}	0.74**	0.64 **	0.20 *	0.70 **	0.54 **	0.58 **
Dr	0.24**	0.15	0.09	0.03	0.11	0.13

SDb	0.73**	0.74**	0.83**	0.84 **	0.75 **	0.70 **
SD_{y}	0.77**	0.72**	0.82**	0.81 **	0.70**	0.68**
SDr	0.05	0.46 **	0.27 **	0.10	0.03	0.09
$SD_{ m r}/SD_{ m b}$	0.79**	0.81 **	0.86**	0.87 **	0.78 **	0.73**
$SD_{ m r}/SD_{ m y}$	0.81**	0.79**	0.87**	0.86 **	0.77 **	0.73**
$(SD_{\rm r}-SD_{\rm b})/(SD_{\rm r}+SD_{\rm b})$	0.68**	0.81 **	0.87**	0.88 **	0.82**	0.73**
$(SD_r - SD_y)/(SD_r + SD_y)$	0.65**	0.78 **	0.85 **	0.86 **	0.78 **	0.72**

Note: Significance level: * p < 0.05, ** p < 0.01. ⁽¹⁾ spectral transformation (ST); ⁽²⁾ vegetation indices (VIs); ⁽³⁾ three edge parameters (TEP).

3.3. ULR

ULR was performed to identify the three most appropriate and sensitive parameters (ST, VIs, and TEP) for LCC estimation using spectral datasets. The three parameters (ST, VIs, and TEP) were set as the independent variables, LCC was set as the dependent variable, and the optimal fitting models were used. In Table 5, a linear model was established by correlation analysis to obtain the R² and RMSE of the calibration and validation datasets. The R² values of the calibration datasets were all greater than 0.60 at every growth stage. A single ST had the best LCC performance at the 1st growth stage, and the R² and RMSE were 0.75 and 2.44, respectively. A single VI had the best LCC performance at the 3rd growth stage, and the R² and RMSE were 0.75 and 2.96, respectively. A single TEP had the best LCC performance at the 3rd growth stage, and the R² and RMSE were 0.74 and 2.98, respectively. All these relationships were significant at p < 0.01.

The ST, VIs, and TEP were established by the regression models for the prediction of LCC at different growth stages, which were validated using the validation datasets, and the results are shown in Figure 4. The R² and RMSE of the validation datasets under different growth conditions and the R² of the validation datasets were all greater than 0.50.

Growth		ST			VIs			TEP	
Stages	Model	R^2	RMSE	Model	R^2	RMSE	Model	R^2	RMSE
1st	L	0.75	2.44	L	0.62	3.00	L	0.66	2.83
2nd	Е	0.63	3.23	Е	0.60	3.36	Ln	0.63	3.22
3rd	Ln	0.72	3.11	L	0.75	2.96	L	0.74	2.98
4th	Ln	0.74	3.23	L	0.71	3.37	Е	0.74	3.11
5th	L	0.69	3.08	Ln	0.70	3.06	L	0.66	3.25
All	Е	0.67	3.39	L	0.69	3.32	L	0.66	3.52

Table 5. Regression analysis and curve fitting results of three parameters versus LCC.

Note: L: linear function; E: exponential function; Ln: logarithmic function. ST: spectrum translation; VIs: vegetation indices; TEP: three edge parameters.



Figure 4. R² and RMSE of validation datasets at different growth stages. Spectrum translation (ST), vegetation indices (VIs), and three edge parameters (TEP).

3.4. MLR

In this section, the three best parameters (ST, VIs, and TEP) were selected and combined to predict the LCC at different growth stages. The MLR analysis results indicated that R² was 0.66–0.79 and RMSE was 2.27–3.40 (Table 6). The estimation model at the 3rd growth stage had the highest R² (0.79) and lowest RMSE (2.46) compared with the other growth stages. These MLR models performed better than models based on a single parameter in terms of R² and RMSE. In addition, the R² of the validation datasets for each growth stage was greater than 0.60, and the RMSE was low (Figure 5).

This suggested that multiparameter modeling was feasible at each growth stage. Based on this result, these parameters were used to build a machine learning algorithm model and to reveal the impact of the complex algorithm on LCC inversion in the multivariate case.

	Growth Stages	Models	R ²	RMSE
1st		$y = 1286.55 - 1245.17 \times x_1 + 487.67 \times x_2 + 0.16 \times x_3$	0.77	2.27
2nd		$y = 22.22 + 2612.22 \times x_1 - 4.87 \times x_2 + 1.19 \times x_3$	0.72	2.82
3rd		$y = 18.3 + 1532.21 \times x_1 + 26.22 \times x_2 - 0.66 \times x_3$	0.79	2.46
4th		$y = -21.96 + 2296.78 \times x_1 + 37.9 \times x_2 + 56.55 \times x_3$	0.79	2.92
5th		$y = 29.05 - 15.16 \times x_1 + 48.11 \times x_2 - 3.76 \times x_3$	0.66	3.18
All		$y = 68.08 - 52.86 \times x_1 + 7.97 \times x_2 + 13.94 \times x_3$	0.67	3.40

Table 6. Multiple variate linear regression analysis model in calibration datasets.

Note: 1st growth stage: x_1 is CRS₇₅₀, x_2 is NDSI, and x_3 is SD_r/SD_y ; 2nd growth stage: x_1 is FODS₇₃₀, x_2 is LCI, and x_3 is SD_r/SD_b ; 3rd growth stage: x_1 is FODS₇₃₂, x_2 is LCI, and x_3 is SD_r/SD_y ; 4th growth stage: x_1 is FODS₅₁₄, x_2 is LCI, and x_3 is $(SD_r - SD_b)/(SD_r + SD_b)$; 5th growth stage: x_1 is CRS₇₁₀, x_2 is LCI, and x_3 is $(SD_r - SD_b)/(SD_r + SD_b)$; All growth stages: x_1 is CRS₇₁₈, x_2 is LCI, and x_3 is $(SD_r - SD_b)/(SD_r + SD_b)$; All growth stages: x_1 is CRS₇₁₈, x_2 is LCI, and x_3 is $(SD_r - SD_b)/(SD_r + SD_b)$; All growth stages: x_1 is CRS₇₁₈, x_2 is LCI, and x_3 is $(SD_r - SD_b)/(SD_r + SD_b)$.



Figure 5. Measured and predicted LCC along the 1:1 line in the standalone validation dataset of the MLR models. (**a**) 1st growth stage, (**b**) 2nd growth stage, (**c**) 3rd growth stage, (**d**) 4th growth stage, (**e**) 5th growth stage, and (**f**) All growth stages.

3.5. Machine Learning Models

The parameters of the machine learning methods (SVR and RF) were optimized by the grid search method to obtain the best parameters and the calibration dataset coefficients R² and RMSE, as shown in Table 7. Moreover, the R² values of RF were higher than SVR for LCC estimation. For RF, the R² values were all greater than or equal to 0.94. However, for SVR, the R² values were rarely higher than 0.80, with the highest being approximately 0.82.

The validation results showed that both the SVR and RF models performed best at the 4th growth stage (Figures 6 and 7, respectively). To estimate LCC, the RF models consistently performed better than the SVR models at every growth stage.

Matria	Madala	Growth Stages						
Metric	widdels	1st	2nd	3rd	4th	5th	All	
D2	SVR (1)	0.82	0.80	0.81	0.78	0.72	0.67	
K ²	RF (2)	0.96	0.94	0.96	0.95	0.95	0.94	
DMCE	SVR	2.02	2.30	2.46	2.78	3.08	3.38	
RMSE	RF	0.95	1.21	1.18	1.27	1.33	1.37	

Table 7. R² and RMSE of calibration datasets in the machine learning models.



Note: ⁽¹⁾ SVR is supper vector regression; ⁽²⁾ RF is random forest regression.

Figure 6. Measured and predicted LCC along the 1:1 line in the standalone validation datasets of the support vector regression (SVR) model. (a) 1st growth stage, (b) 2nd growth stage, (c) 3rd growth stage, (d) 4th growth stage, (e) 5th growth stage, and (f) All growth stages.



Figure 7. Measured and predicted LCC along the 1:1 line in the standalone validation datasets of the random forest (RF) model. (a) 1st growth stage, (b) 2nd growth stage, (c) 3rd growth stage, (d) 4th growth stage, (e) 5th growth stage, and (f) All growth stages.

4. Discussion

4.1. Selected Optimized Spectral Indices

Hyperspectral RS is commonly used to monitor seasonal changes in the LCC of crops. In this study, a correlation analysis between three spectral variables (ST, VIs, and TEP) and LCC is shown in Table 4. The results of this study indicate that FODS had the best sensitivity to LCC at the 2nd, 3rd, and 4th growth stages and that CRS had the best sensitivity to LCC at the 1st and 5th growth stages and in All growth stages combined. The most relevant VI of the 1st growth stage was the NDSI, the most relevant VI of other growth stages was the LCI, and the correlation coefficients were all greater than 0.70. In this study, a correlation analysis between TEP and LCC showed that SD_r/SD_b , SD_r/SD_y , and $(SD_r - SD_b)/(SD_r + SD_b)$ had the best correlation with LCC; the correlation coefficients were 0.81, 0.81, 0.87, 0.88, 0.82, and 0.73 at the 1st, 2nd, 3rd, 4th, 5th and All growth stages, respectively. These results may be attributed to the reflectance in the red-edge region (680–760 nm) being closely related to the chlorophyll content in apple trees, which has always been considered important in relationships between biochemical or biophysical parameters [44].

In summary, the first-order differential spectrum (FODS) and $(SD_r-SD_b)/(SD_r+SD_b)$ are generally the most sensitive to apple leaf chlorophyll content. In previous research, the first derivative was closely related to LCC in wheat crops [45], which was designed to eliminate background signals or noise and to resolve overlapping spectral features. In the future, we will use the spectral data after spectral transformation for models, and spectral transformation could improve the accuracy of the model. The vegetation index has begun to be used more and more in research evaluating crop parameters, but some of the previously researched vegetation indices may not be suitable for future research. Therefore, the vegetation index will be developed or optimized in future research.

4.2. Comparison of Estimation Models with LCC

In this study, LCC was estimated using ST, VIs and TEP alone at different growth stages. The SLR results indicate that ST, VIs and TEP alone best predicted LCC at the 1st, 3rd, and 4th growth stages, and the highest R² values for these stages were 0.75, 0.75 and 0.74, respectively (Table 5). The MLR regression results indicated that the combination parameter had the best estimated LCC at the 3rd growth stage, at which the R² value was 0.79 (Table 6). These results demonstrate that using ST, VIs and TEP predicted LCC to have seasonable features. In addition, farmers can perform measurements and use appropriate amounts of fertilizer at the 3rd growth stage. Apple tree leaves had the highest LCC at the 3rd growth stage (Figure 2).

The U/M linear regression models can provide only linear estimations, while ML methods can determine nonlinear relationships. Machine learning (RF and SVR) methods provided better estimations than methods based on linear regression. The machine learning algorithms, especially the RF models, which had the best estimation capacity, all resulted in higher calibration than the U/M linear regression models; however, for the validation datasets, the RF and SVR models performed unevenly at different growth stages. These results could indicate that ML modeling has an overfitting issue [46].

However, multiple variable regression models encompass linear models and machine learning models (SVR and RF) that were used in this study. More research has demonstrated that machine learning models can predict apple tree leaf chlorophyll content [8,47]. The RF regression algorithm is an ensemble-learning algorithm that combines a broad set of regression trees. A regression tree represents a set of conditions or restrictions that are hierarchically organized and successively applied from the roots to the leaves of a tree [48]. The SVM algorithm is based on statistical learning theory and can be regarded as the same type of network, which can also be used for both classification and regression problems [49]. ML models all achieved better results than linear regression models with respect to calibration, but the accuracy of the RF models was lower than that of the SVR models in the validation analysis. A possible reason for such results is that the RF model often results in an overfitting phenomenon, and the robustness and generalization ability of RF are stronger than those of the other SVR methods [46]. Although linear regression models performed slightly worse than ML models in this study, these approaches possessed an extremely fast processing speed; this attribute indicates that these methods comprise a promising technique that can be integrated into crop monitoring systems.

4.3. Challenges and Future Research

In this study, MLR models and machine learning models (SVR and RF) were established using indoor measurement spectral data to assess LCC data at different growth stages. Machine learning methods can improve the prediction accuracy of leaf chlorophyll content at different growth stages. However, this research found that MLR models and machine learning (SVR and RF) models showed decrease sensitivity various at growth stages. In the future, we need to optimize the model to further improve prediction accuracy and stability. In addition, an increasing number of advanced technologies, such as unmanned aerial vehicles (UAVs) and satellite RS images, are now used to retrieve crop growth parameters and physiological parameters [50,51]. We will use these advanced technologies in future research and develop more machine learning methods to evaluate the growth information and physiological parameters of orchard fruit trees and provide technical support for intelligent orchard management.

5. Conclusions

Experiments undertaken using machine learning and statistical regression methods included an analysis of ST, VIs, and TEP. Notably, using the RF algorithm significantly

improved the predictability and accuracy of the model in terms of the R² and RMSE values. Our results also showed that the prediction models of different growth stages were better in their prediction accuracy when using the RF model (R^2 =0.96) and that the prediction accuracy for the 1st growth stage was the best with the RF model. The results indicated that the predicted LCC follows seasonal patterns. Furthermore, evaluating the 1st growing season using the RF model can provide reasonable accuracy for each growth stage.

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