Assessment of Fv/Fm absorbed by wheat canopies employing in-situ hyperspectral vegetation indexes

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Chlorophyll fluorescence parameter of Fv/Fm, as an important index for evaluating crop yields and biomass, is key to guide crop management. However, the shortage of good hyperspectral data can hinder the accurate assessment of wheat Fv/Fm. In this research, the relationships between wheat canopy Fv/Fm and in-situ hyperspectral vegetation indexes were explored to develop a strategy for accurate Fv/Fm assessment. Fv/Fm had the highest coefficients with normalized pigments chlorophyll ratio index (NPCI) and the medium terrestrial chlorophyll index (MTCI). Both NPCI and MTCI were increased with the increase in Fv/Fm. However, NPCI value ceased to increase as Fv/Fm reached 0.61. MTCI had a descending trend when Fv/Fm value was higher than 0.61. A piecewise Fv/Fm assessment model with NPCI and MTCI regression variables was established when Fv/Fm value was ≤ 0.61 and > 0.61, respectively. The model increased the accuracy of assessment by up to 16% as compared with the Fv/Fm assessment model based on a single vegetation index. Our study indicated that it was feasible to apply NPCI and MTCI to assess wheat Fv/Fm and to establish a piecewise Fv/Fm assessment model that can overcome the limitations from vegetation index saturation under high Fv/Fm value.

Photosynthesis was the most important biological process on earth. It was the unique approach by which plants gained energy from the environment. There were three basic effects when light struck a leaf surface: absorption, reflection and transmission. The major part of light was absorbed by the chlorophyll used for photosynthesis, and only a small proportion was de-excited via emission with a longer wavelength as fluorescence, or dissipation as heat. Chlorophyll fluorescence emissions occurred in the red and far-red regions of the plant spectrum (650–800 nm). Changes in chlorophyll fluorescence parameters of plant leaves could reflect the changes of environmental factors and their effects on plant photosynthetic physiology to a certain extent. In many chlorophyll fluorescence parameters, Fv/Fm was used to characterize the conversion efficiency of the light energy of the PS II reaction center, and its numerical changes were of special significance. However, conventional methods of assessing Fv/Fm from field observations, that involved site-specific complicated parameterizations and calculations, made it difficult to apply over large agricultural areas. These shortcomings could be overcome through the complementary use of hyperspectral measurements of crops, which had several advantages - non-destructive, uniform, could be performed rapidly, and no complicated parameterizations were necessary.

Assessment of Fv/Fm from vegetation indexes (VIs) derived from hyperspectral data, especially remote sensing data, have been reported by several studies. For instance, some researchers compared the performance of VIs to assess Fv/Fm of legume crops and reached the conclusion that out of the nine kinds of VIs with a close relationship with Fv/Fm, modified soil adjusted vegetation index (MSAVI) performed best. If ground cover was significant, the impact of the background significantly reduced, and Fv/Fm could be better estimated using normalized difference vegetation index (NDVI). Re-normalized difference vegetation index (RDVI) showed an approximate linear relation to Fv/Fm regardless of ground cover. Hyperspectral remote sensing is an important technique to fulfill real-time monitoring for crop growth status based on its superior performance in acquiring vegetation canopy information rapidly and non-destructively. However, the regression analysis was based on only five points making it statistically uncertain. Other scientists used radiative transfer models to estimate Fv/Fm and found that a linear model based on NDVI produced the best estimate results.

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Recently, modeled Fv/Fm products based on MODIS have been reported in several studies. Coops et al. investigated the increasing availability of time series of Fv/Fm data from MODIS and reported the three dynamic habitat index components varied significantly in their magnitude, principally because of MODIS Fv/Fm estimates being larger than those observed by medium resolution imaging spectrometer (MERIS) Fv/Fm. In some previous studies, some researchers compared MODIS Fv/Fm with the measurements for sites in the US and found MODIS Fv/Fm was higher than the ground-measured Fv/Fm. Winkel et al. compared Fv/Fm from MODIS with in-situ measurements for a tropical rainforest in Brazil and concluded that MODIS Fv/Fm was reliable for Fv/Fm assessment. There was a need for an investigation of the performance of VIs in different vegetation ecosystems.

Models based on linear Fv/Fm-NDVI relationships suffered from a major flaw—NDVI saturated at high leaf area index values and, thus a linear model tended to be insensitive to Fv/Fm changes in such cases. Another issue that needed to be recognized was the scarcity of data for boreal ecosystems. The majority of the above-cited studies presented empirical evidence suggesting a functional relationship between Fv/Fm and hyperspectral VIs, and these were mostly focused on forests, grasses (prairies), and some crop types such as rice, wheat and cotton. There were only few reports on quantitatively estimating Fv/Fm for wheat canopies using VIs from remote sensing data. Besides, VIs-Fv/Fm relationships differed from one ecosystem type to another ecosystem due to the influences of vegetation type, strong background signals, canopy structure and spatial heterogeneity. Existing remote sensing-based Fv/Fm products lacked adequate ground validation, which was critical for establishing the uncertainty and accuracy of such products so that they could be used for guiding crop production practices.

This study is motivated by the above-mentioned issues and focuses on exhaustive statistical analyses of Fv/Fm-VIs relationships for wheat canopies, using in-situ hyperspectral data collected from a series of field experiments, and aims at determining a practical methodology for estimating Fv/Fm of wheat canopies.

**Results**

**Changes in wheat canopy Fv/Fm with growth stage.** Fv/Fm revealed the progressive increase as the growth of wheat crops at different growth stages (Fig. 1). An initial significant increase in Fv/Fm by about 23.1% was observed corresponding to crop development from turning stage to jointing stage. However, the further changes in Fv/Fm from booting stage to the milk stage were not significant (2.4%, 0.12%, 1.31% and −2.59%, respectively). Until the blooming stage, Fv/Fm began to increase and reached its maximum value of 0.68. From the blooming stage to the milk stage, Fv/Fm tended to slow, that was saturated.

**VIs-Fv/Fm relationship.** Statistically significant correlations between Fv/Fm and VIs were observed in 51 cases out of 56 VIs considered (Table 1), and these were both positive and negative. Positive correlations between VIs and Fv/Fm were generally stronger than the negative ones. Fv/Fm was most strongly correlated to NPCI, MTCI and NDVI[900, 680]—correlation coefficients (r) are 0.891, 0.886 and 0.879, respectively. Thus, NPCI, MTCI and NDVI[900, 680] could be identified as three common VIs relatively well correlated to wheat canopy Fv/Fm, and these were the post probable VIs of choice for estimating Fv/Fm.

**Establishing the Fv/Fm assessment model based on VIs.** A total of 10 VIs were considered for modeling Fv/Fm based on a threshold on VI-Fv/Fm correlation (i.e. r > 0.82 in Table 1). These non-linear Fv/Fm assessment models were best represented as exponential functions and were evaluated using their predictive (R²) and error statistics (RRMSE) (Table 2). Among them, Fv/Fm had the closest exponential relation with NPCI, and had a closer exponential relation with MTCI and NDVI[900, 680], and the models based on NPCI, MTCI and NDVI[900, 680] were capable of estimating the Fv/Fm with R² of 0.874, 0.859 and 0.834, respectively, with RRMSE of 0.109, 0.116 and 0.126, respectively, with the assessment accuracy of 89.1%, 88.4% and 87.4%, respectively. Furthermore, according to comparisons of R², RRMSE and assessment accuracy, it was more suitable to assess wheat canopy Fv/Fm by NPCI and MTCI than by NDVI[900, 680].

![Figure 1. Fv/Fm absorbed by for wheat canopies at different growth stages.](image-url)
Saturation analysis of VIs. All three VIs in Fig. 2, i.e. NPCI, MTCI and NDVI [900, 680], which had the strongest relationship to \( F_v/F_m \), were increased progressively as \( F_v/F_m \) was increased to about 0.61. Beyond this point, NPCI and NDVI [900, 680] values started leveling off at 0.65 and 0.61 respectively, which was also known saturation. On the other hand, MTCI displayed a different tendency when \( F_v/F_m \) value was higher than 0.61. Based on this information, a reliable \( F_v/F_m \) model could be constructed with NPCI as the regression variable before the saturation point sets in (\( F_v/F_m \leq 0.61 \)), and with MTCI as while MTCI as the regression variable after the saturation point (\( F_v/F_m > 0.61 \)).
Based on the aforementioned research results, the piecewise hyperspectral assessment model of $F_v/F_m$ was built according to the range of $F_v/F_m$ value in Fig. 3. Namely, if $F_v/F_m \leq 0.61$, NPCI should be used to assess $F_v/F_m$, and the assessment model was $y = 1.0616x + 0.076$, $R^2 = 0.929$ ($p < 0.01$); if $F_v/F_m > 0.61$, MTCI should be used to assess $F_v/F_m$, and the assessment model was $y = 5.5259e^{-3.529x}$, $R^2 = 0.835$ ($p < 0.01$).

**Evaluation of VIs-based $F_v/F_m$ model.** A total of 60 samples observed from the experiments in 2017 were used to test the hyperspectral VIs-based assessment model of $F_v/F_m$. The estimated and measured $F_v/F_m$ cross-resistance almost coincided with 1:1 relation line shown in Fig. 4 for comparison. At low $F_v/F_m$ values, estimated value might be underestimated. As $F_v/F_m$ increased, estimated values were closer to the measured values. The $R^2$, RRMSE and assessment accuracy values of the piecewise $F_v/F_m$ model were 0.9278, 0.084 and 91.6%, respectively. Compared with the assessment models based on only NPCI, MTCI and NDVI [900, 680] in Table 3, the assessment accuracy values of the piecewise $F_v/F_m$ model in different ranges of the $F_v/F_m$ increased by 11.3%, 13.9% and 16.4%, respectively. In conclusion, the piecewise model based on NPCI and MTCI, used to assess $F_v/F_m$, can not only improve the assessment accuracy, but also solve the saturation problems that occurred in NPCI and NDVI [900, 680].
| VIs | Abbreviation | Algorithm |
|-----|--------------|-----------|
| Simple ratio 1 | SR[787, 765] | R<sub>787</sub>/R<sub>765</sub> |
| Simple ratio 2 | SR[415, 710] | R<sub>415</sub>/R<sub>710</sub> |
| Simple ratio 3 | SR[415, 695] | R<sub>415</sub>/R<sub>695</sub> |
| Simple ratio 4 | SR[750, 705] | R<sub>750</sub>/R<sub>705</sub> |
| Simple ratio 5 | SR[900, 680] | R<sub>900</sub>/R<sub>680</sub> |
| Simple ratio 6 | SR[801, 670] | R<sub>801</sub>/R<sub>670</sub> |
| Simple ratio 7 | SR[672, 550, 708] | R<sub>800</sub>(R<sub>750 * R<sub>680</sub>) |
| Optimized vegetation index 1 | V<sub>opt1</sub> | R<sub>660</sub>/R<sub>550</sub> |
| Optimized vegetation index 2 | V<sub>opt2</sub> | 100 * (lnR<sub>545</sub> − lnR<sub>670</sub>) |
| Pigment specific simple ratio 1 | PSSR[800, 680] | R<sub>800</sub>/R<sub>680</sub> |
| Pigment specific simple ratio 2 | PSSR[800, 635] | R<sub>800</sub>/R<sub>635</sub> |
| Pigment specific simple ratio 3 | PSSR[800, 470] | R<sub>800</sub>/R<sub>470</sub> |
| Zarco-Tejada & Miller | ZTM | R<sub>550</sub>/R<sub>750</sub> |
| Red-edge model index | R-M | (R<sub>720</sub> − R<sub>600</sub>) |
| Difference index | DI | R<sub>660</sub> − R<sub>550</sub> |
| Difference vegetation index | DV1 | R<sub>660</sub> − R<sub>550</sub> |
| Pigment specific normalized difference 1 | PSND[800, 635] | (R<sub>800</sub> − R<sub>635</sub>)/(R<sub>800</sub> + R<sub>635</sub>) |
| Pigment specific normalized difference 2 | PSND[800, 470] | (R<sub>800</sub> − R<sub>470</sub>)/(R<sub>800</sub> + R<sub>470</sub>) |
| Modified simple ratio index 1 | mSRI1 | (R<sub>800</sub> − R<sub>670</sub>)/(R<sub>670</sub> + R<sub>800</sub>) |
| Modified simple ratio 2 | mSRI2 | (R<sub>800</sub> − R<sub>670</sub>)/(R<sub>670</sub> + R<sub>800</sub>) |
| Normalized difference index | NDI | (R<sub>800</sub> − R<sub>670</sub>)/(R<sub>670</sub> + R<sub>800</sub>) |
| Modified normalized difference index | mNDI | (R<sub>800</sub> − R<sub>670</sub>)/(R<sub>670</sub> + R<sub>800</sub>) |
| Plant senescence reflectance index | PSRI | (R<sub>800</sub> − R<sub>670</sub>)/(R<sub>550</sub>) |
| Re-normalized difference vegetation index | RDVI | (R<sub>800</sub>)/(R<sub>660</sub>) |
| Simple ratio pigment index | SRPI | R<sub>660</sub>/R<sub>550</sub> |
| Ratio vegetation index | RVI | (R<sub>545</sub>)/(R<sub>670</sub>) |
| Normalized pigments chlorophyll ratio index | NPCI | (R<sub>800</sub> − R<sub>635</sub>)/(R<sub>800</sub> + R<sub>635</sub>) |
| Normalized phaeophytinization index | NPIQ | (R<sub>430</sub> − R<sub>680</sub>)/(R<sub>430</sub> + R<sub>680</sub>) |
| Structure intensive pigment index | SIPI | (R<sub>800</sub> − R<sub>635</sub>)/(R<sub>800</sub> − R<sub>670</sub>) |
| Medium terrestrial chlorophyll index | MTCI | (R<sub>800</sub> − R<sub>650</sub>)/(R<sub>650</sub> − R<sub>800</sub>) |
| Modified chlorophyll absorption in reflectance index | MCARI | [(R<sub>800</sub> − R<sub>650</sub>) − 0.2 * (R<sub>550</sub> − R<sub>650</sub>)] * (R<sub>700</sub>/R<sub>800</sub>) |
| Green normalized difference vegetation index | GNDVI | (R<sub>800</sub> − R<sub>670</sub>)/(R<sub>670</sub> + R<sub>800</sub>) |
| Modified transformed vegetation index | MTVI | 1.2 * [1.2 * (R<sub>800</sub> − R<sub>650</sub>) − 2 * R<sub>550</sub>] |
| Photochemical reflectance index | PRI | (R<sub>800</sub> − R<sub>670</sub>)/(R<sub>550</sub> + R<sub>670</sub>) |
| Transformed vegetation index | TVI | 0.5 * [120 * (R<sub>16</sub> − R<sub>13</sub>) − 200 * (R<sub>810</sub> − R<sub>800</sub>)] |
| Temperature condition index | TCI | 1.2 * (R<sub>700</sub> − R<sub>650</sub>) − 1.5 * (R<sub>700</sub> − R<sub>635</sub>) * SQRT(R<sub>900</sub>/R<sub>700</sub>) |
| Double difference index | DDI | (R<sub>550</sub> − R<sub>670</sub>) − (R<sub>670</sub> − R<sub>550</sub>) |
| Normalized difference vegetation index 1 | NDVI1 | (R<sub>670</sub> − R<sub>550</sub>)/(R<sub>670</sub> + R<sub>550</sub>) |
| Normalized difference vegetation index 2 | NDVI2 | (R<sub>550</sub> − R<sub>480</sub>)/(R<sub>550</sub> + R<sub>480</sub>) |
| Normalized difference vegetation index 3 | NDVI3 | (R<sub>670</sub> − R<sub>550</sub>)/(R<sub>670</sub> + R<sub>550</sub>) |
| Normalized difference vegetation index 4 | NDVI4 | (R<sub>800</sub>)/(R<sub>670</sub> + R<sub>800</sub>) |
| Normalized difference vegetation index 5 | NDVI5 | (R<sub>800</sub>)/(R<sub>670</sub> + R<sub>800</sub>) |
| Ratio between TCI and OSAVI | TCI/OSAVI | TCI/OSAVI |
| Ratio between MTVI and MSAVI | MTVI/MSAVI | MTVI/MSAVI |
| Ratio between DDI and MSAVI | DDI/MSAVI | DDI/MSAVI |
| Ratio between MCARI and OSAVI | MCARI/OSAVI | MCARI/OSAVI |
| Ratio between TCARI and OSAVI | TCARI/OSAVI | TCARI/OSAVI |

Table 3. Definition of hyperspectral VIs evaluated in the study.30
Discussion

Ft/Fm was primarily controlled by ground cover and leaf area 24. Before the jointing stage, Ft/Fm increased significantly (Fig. 1), which was characterized by strong absorption of incoming Ft/Fm as wheat crops grew vigorously, adding leaf area driven by nitrogen fertilization. This was followed by a lower rate of crop growth (and leaf area expansion), which was captured by the lower rate of Ft/Fm increase. According to agronomic principle of wheat, although the research was lack of Ft/Fm data after the milk stage, it was still available to conclude that leaves started to turn yellow and gradually litter, as the growth period went, and Ft/Fm declined in the combination of wheat's photosynthetic physiological characteristics. Until full-ripe stage, Ft/Fm was close to 0, because leaves took off green and became withered and died so that they were unable to absorb light energy and the accumulation of dry matter had stopped 25.

Significant efforts were presently focusing on the use of VIs in general, and NDVI in particular, for estimating vegetation canopy Ft/Fm. Furthermore, many studies indicated that VIs were better correlated to Ft/Fm than the reflectance in single wavebands 26, 27, which could be plausibly explained by the fact that VIs could minimize the influence of atmospheric scattering and soil background and enhanced the information of the sensitive wavebands 28. Similarly, this study found Ft/Fm to be strongly correlated to the majority of VIs (49 out of 56), with NPCI, MTCI and NDVI [900, 680] being the best performing VIs. This result is helpful to provide an important technique for the establishment of perfect wheat photosynthetic groups, the improvement of sunlight energy efficiency and the implementation of cultivation control.

As compared to the previous studies with NDVI, NPCI and MTCI for estimating the Ft/Fm gave the lower RMSE and higher assessment accuracy than NDVI proposed in several studies. Future research should focus on evaluating the performance of the proposed model over wheat crops grown under a variety of conditions, different wheat varieties, as well as other crop types. This will help in refining the model as a useful tool for informing crop management practices. Efforts should also be made to test this model with data from different sources – field-based spectral measurements, as well as current and future satellite data.

Conclusion

VIs, like NDVI, were often plagued with saturation at high biomass areas, which was a major disadvantage for VIs-Ft/Fm models. We have addressed this issue by employing the differences in sensitivity of different VIs to Ft/Fm, i.e. Ft/Fm had the highest coefficients with NPCI and MTCI. Both NPCI and MTCI were increased with the increase in Ft/Fm. However, NPCI value ceased to increase as Ft/Fm reached 0.61. MTCI had a descending trend when Ft/Fm value was higher than 0.61. A piecewise Ft/Fm assessment model with NPCI and MTCI regression variables was established when Ft/Fm value was ≤ 0.61 and >0.61, respectively. The model increased the accuracy of assessment by up to 16% as compared with the Ft/Fm assessment model based on a single vegetation index. Our study indicated that it was feasible to apply NPCI and MTCI to assess wheat Ft/Fm and to establish a piecewise Ft/Fm assessment model that can overcome the limitations from vegetation index saturation under high Ft/Fm value.

Materials and Methods

Experimental design.

Four varieties of wheat - Yangmai 13, Yangmai 15, Yangmai 16 and Ningmai 9 were used in a field experiment conducted from March to May during the three wheat seasons of 2015, 2016 and 2017 on the Experimental Farm of Yangzhou University, China (119°18′, 32°26′N). The former crop in the field was rice. The soil is yellow brown soil (Alfisolsin U.S. taxonomy), containing 2.23 g kg⁻¹ organic matter, 121.3 mg kg⁻¹ available nitrogen, 25.9 mg kg⁻¹ available phosphorus and 83.7 mg kg⁻¹ available potassium in the 0–30 cm soil layer. Canopy spectral parameters were recorded alongside with the quasi-simultaneous measurement of Ft/Fm upon the growing wheat canopies. In order to highlight variations in wheat growth due to biochemical composition changes, three different levels of nitrogen fertilization as urea were implemented, including non-nitrogen fertilization, adequate nitrogen fertilization (450 kg ha⁻¹) and heavy nitrogen fertilization (900 kg ha⁻¹). There are three replicates for each nitrogen level. The plot size was 4 m × 4 m. Local standard wheat cropping management practices pertaining to water, pest, disease and weed were followed. Training data consisted of 95 and 87 samples in 2015 and 2016, respectively, and test data consisted of 60 samples in 2017.

Canopy hyperspectral reflectance data.

In 2015, six spectral measurements were carried out in the wheat turning green stage (March 7), jointing stage (March 20), booting stage (April 9), blooming stage (April 25), 15 days after blooming stage (May 9), and milking stage (May 18), respectively. All canopy spectrometry determinations were taken at a vertical height of 1.6 m over the canopy under the cloudless or near cloudless condition between 11:00 and 14:00, using an ASD FieldSpecPro spectrometer (Analytical Spectral Devices, USA) fitted with 25° field of view fiber optics, operating in the 350–2500 nm spectral region with a sampling interval of 1.4 nm between 350 nm and 1050 nm, and 2 nm between 1050 nm and 2500 nm, and with spectral resolution of 3 nm at 700 nm, 10 nm at 1400 nm, selecting the representative, growth-uniform, pest-free plants, placing the probe of sensor down in measuring. A 40 cm × 40 cm BaSO₄ calibration panel was used for calculation of hyperspectral reflectance. Vegetation and panel radiance measurements were taken by averaging 20 scans at optimized integration time, with a dark current correction at every spectrometry determination.

In 2016, four spectral measurements with 87 test samples were carried out in the wheat turning green stage (March 9), jointing stage (March 22), blooming stage (April 23), and milking stage (May 20), respectively. In 2017, total three spectral measurements with 60 test samples were carried out in the wheat booting stage (April 11), blooming stage (April 22), and 15 days after blooming stage (May 12), respectively. The other practices in 2016 and 2017 were as same as those in 2015.
Spectral smoothing. Spectral smoothing process was performed in order to remove high frequency noise and the random errors caused by spectral measuring instruments, which enhanced signal to noise ratio. A five-point weighted smoothing method was used to process the raw spectral data\(^\text{29}\). Five-point weighted smoothing method is carried out using Equation (1):

\[
n = \left( \frac{m_2}{4} + \frac{m_{-1}}{2} + \frac{m_1}{2} + \frac{m_2}{4} \right) / 25
\]

Here, \(n\) is the weighted average of the intermediate data points in the filter window, namely the smoothed spectrum value, and \(m\) is the value of unsmoothed data points, namely the original spectral value.

\(F_{\text{v}}/F_{\text{m}}\) measurement. The chlorophyll fluorescence parameters of wheat leaves were determined by modulated fluorescence OS1-FL (Opti-Sciences, Tyngsboro, MA, USA) after the completion of each spectrum. First, the dark adaptation clamp was used to adapt the blade to 10 min, and then the initial light energy conversion efficiency of photosystem II (PS II) \(F_{\text{v}}/F_{\text{m}}\) was measured, and the calculation was repeated 9 times each time. The formula is as follows:

\[
F_{\text{v}}/F_{\text{m}} = (F_{\text{m}} - F_{\text{c}})/F_{\text{m}}
\]

Here, the \(F_{\text{c}}\) is the basic fluorescence value under the dark adaptation condition; the \(F_{\text{m}}\) is the maximum fluorescence value under the dark adaptation condition; the \(F_{\text{v}}\) is the fluorescence value under the variable condition.

Hyperspectral VIs. In reference to previous studies, based on spectral characteristics of wheat and combined with the physical meaning of spectral index, a total of 56 VIs were considered (Table 3)\(^\text{30}\) related to \(F_{\text{v}}/F_{\text{m}}\) leaf area index and chlorophyll (known as an important influence on \(F_{\text{v}}/F_{\text{m}}\) absorbed by green vegetation) as the independent variables for establishment of remote sensing assessment models of wheat canopy \(F_{\text{v}}/F_{\text{m}}\). Data from the field experiment in 2015 (95 samples) and 2016 (87 samples) were used to develop the regression models, and data from the field experiments in 2017 (60 samples) were used to evaluate the models.

Statistical analysis. VIs-\(F_{\text{v}}/F_{\text{m}}\) relationships were analyzed using a variety of regression models - linear, exponential, logarithmic, and quadratic. Models were ranked based on statistically significant (\(p < 0.05 \) or \(0.01\)) correlation coefficients (\(r\) in case of linear models) and coefficients of determination (\(R^2\) in case of non-linear models). Finally, by plotting the relation figure under the scale 1:1 between estimated and measured \(F_{\text{v}}/F_{\text{m}}\) values, the performance of the model was evaluated through the coefficient of determination (\(R^2\)) and relative root mean squared error (RRMSE) for the assessment of in-situ measured \(F_{\text{v}}/F_{\text{m}}\). The higher the \(R^2\) and the lower the RRMSE, the higher the accuracy of the model to assess the \(F_{\text{v}}/F_{\text{m}}\). The RRMSE and assessment accuracy are calculated using Equations (3) and (4), respectively:

\[
\text{RRMSE} = \left[ \frac{1}{n} \sum_{i=1}^{n} (y_i - \hat{y}_i)^2 \right]^{1/2}
\]

\[
\text{Assessment accuracy} = 1 - \text{RRMSE}
\]

Here, \(y_i\) and \(\hat{y}_i\) are the measured values and predicted values of wheat canopy \(F_{\text{v}}/F_{\text{m}}\), respectively. \(n\) is the number of samples.

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Author Contributions
C.W.T. and W.S.G. conceived the research. C.W.T., Y.D. and J.Z. designed and performed the experiments. C.W.T. and D.L.W. prepared and revised the manuscript. All the authors have reviewed the manuscript and agreed the submission and publication.

Additional Information
Competing Interests: The authors declare no competing interests.

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