

## Relationship Between Color Characteristic and Reflectance Index by Ground-based Remote Sensor for Tobacco Leaves

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To determine the critical level for optimum maturity of flue-cured tobacco leaves (KF118) at the stalk position from cutter to tips, the reflectance index using ground-based remote sensors and chlorophyll meter were investigated. The sensors estimated were Crop Circle™ (Holland Scientific), Green Seeker™ (Ntech Industries), Spectroradiometer (LICOR, LI-1800), Chlorophyll meter (SPAD502, Minolta), and Field Scout™ Chlorophyll meter (CM-1000, Spectrum). The L, a, b values and greenness for flue-cured leaf were measured and estimated for correlation with sensor's measurement of harvested leaf. On a reflectance curve of 340nm~1100 nm, the reflectance peaks on 550nm and 675 nm for the harvested leaf were lowered as change from light green to darker green. Darker green leaf harvested produced darker flue-cured leaf. The reflectance at 675 nm for flue-cured leaf decreased as greenness increased in the harvested leaf. This result means that the red edge band of 675 nm wavelength is related to the absorbance of chlorophyll for photosynthesis. The greenness of flue-cured leaf showed significantly positive correlation with the entire reflectance indexes for harvested leaf while the L value by colorimeter showed negative correlation with greenness of cured leaf. The critical level for optimum maturity of harvested leaf were less than 22, 135, and 0.43 for SPAD reading, CM-1000 reading, and gNDVI by Crop Circle™, respectively. Consequently, ground-based remote sensing providing a non-destructive real-time assessment of plant greenness could be a useful tool in the selection of optimum maturity of flue-cured tobacco leaves in relation to high quality of flue-cured tobacco

**Key words:** Flue-cured tobacco, Optimum maturity of leaves, Reflectance index, ground-based remote sensor, Greenness

### Introduction

The spectral characteristics of radiation reflected, transmitted, or absorbed by leaves can provide specific information of physiological response to growth conditions and plant adaptations to the environment. The use of the ratio spectra has allowed the identification of reflectance bands corresponding to the absorption bands of specific pigments.

The pigments making the greatest contribution to light absorption in the photo-synthetically active radiation region are those which are intimately involved in photosynthesis, namely, chlorophyll a and b, and the carotenoid. The absolute and relative concentrations of these photosynthetic pigments dictate the photosynthetic potential of the plant, that is, its efficiency in the

utilization of photo-synthetically active radiation for bio-synthetic purposes (Chappelle et al., 1992).

Many indices computed as differences, ratios, or linear combinations of reflectance in visible and NIR wavebands have been used for evaluation of various vegetation parameters (Deering et al., 1975; Richardson and Wiegand, 1977; Tucker, 1979; Jackson, 1983; Wiegand et al., 1991).

The most common indices utilize red and near-infrared canopy reflectance in the form of ratios such as normalized difference vegetation index (rNDVI, Tucker, 1979), where  $rNDVI = (NIR-Red)/(NIR+Red)$ , or in linear combination such as perpendicular vegetation index (Richardson and Wiegand, 1977).

These indices have been found to be well correlated with various vegetation parameters including green leaf area, biomass, percent green cover, productivity, and photosynthetic activity (Hatfield et al., 1993; Sellers,

1985). The green normalized difference vegetation index (gNDVI), where the green band is substituted for the red band in the rNDVI equation, was proved to be more useful for assessing canopy variation in green crop biomass related to nitrogen fertility in soils (Gitelson et al., 1996; Shanahan et al., 2001).

Remote sensing techniques, particularly multi-spectral visible and near infrared (NIR) reflectance, can provide instantaneous, non-destructive, and qualitative assessment of the crop biomass. The goal of this study was to compare the possibility for estimation of the optimum maturity of tobacco leaves by remote sensing technique.

## Materials and Methods

**Sampling of flue-cured tobacco green leaves harvested** Green leaves of flue-cured tobacco (KF118) were sampled from cutter and tips stalk position of tobacco plant based on the greenness classified by 5 level of maturity at the 68 and 75 days after transplanting, respectively. The reflectance index of each green leaf harvested was measured using all the remote sensors estimated. The leaves were then processed in bulk curing system for making flue-cured tobacco. The reflectance index of each cured leaf was measured the same remote sensors. The degree of greenness was examined with the naked eye and L, a, b values by colorimeter were also determined.

**Sensors estimated** Plant measurements were made using 1) chlorophyll meter (Minolta, SPAD 502), 2) Fieldscout™ (Spectrum Technologies, CM1000), 3) GreenSeeker™ (GS, Ntech Industries), 4) Crop Circle™ (CC, Holland Scientific), and 5) spectroradiometers (SR, LICOR Inc. LI-1800). The SPAD meter uses two light-emitting diodes (650 and 940 nm) to determine the amount of light transmitted through a 2x3-mm leaf section. The Fieldscout™ chlorophyll meter commercialized by Spectrum Technologies, Inc. uses ambient and reflected light at 700 and 840 nm to estimate chlorophyll content. The GreenSeeker™ as commercialized by Ntech Industries Inc. was the only active sensor used in this study and measures red and near infrared (NIR) reflectance to generate rNDVI. The field of view for this sensor is 1 x 60 cm with an effective depth of field from 80 to 120 cm (32 to 48 inches) from the target. The Crop Circle™ sensor made by Holland

Scientific is a multi-waveband sensor designed for measuring light reflecting from crop canopies. Crop Circle™ is a passive sensor with a conical field of view (20 degree) that measures four-band reflectance; green (550 nm), amber (600 nm), red (680 nm), and near infrared (NIR, 820 nm) allowing the calculation of red NDVI, green NDVI, and amber NDVI. The spectroradiometer can measure reflectance from visible to near infrared wavebands: 340~1100 nm (2-nm interval) for LI-1800 in 2004.

**Reflectance measurement** Canopy reflectance measurements with the GreenSeeker™ and Crop Circle™ sensors and spectroradiometers (LI-1800) were made on the same date as the SPAD and CM1000 measurements.

All leaf reflectance measurements were made during midday under incident solar radiation from an angle perpendicular to the canopy. To minimize the background effect on reflectance measured with GreenSeeker™, Crop Circle™, and spectroradiometers, a black board was installed under the leaf. In case of GreenSeeker™ measurement, sensor was slowly scanned over the tobacco leaf keeping about 100 cm distances from top canopy to the sensor head. For the outdoor measurement of Crop Circle™, the sensor was installed above the fixed leaf position having 80~90 cm distance from the sensor head to the leaf surface. The rNDVI, gNDVI and aNDVI were used for evaluation of color characteristics of tobacco leaf. After collecting reflectance measurements, two leaf discs were taken from the upper-most expanded leaf using a cork borer (1-cm dia.) for determination of chlorophyll content. Leaf disc samples were homogenized in 80% acetone solution and the mixture was centrifuged at 2000 rpm for 2 minutes. Thereafter, the optical density of supernatant was measured at 645 and 663 nm. Chlorophyll content was calculated using the Arnon method (1949).

**Vegetation Indices** Various vegetation indices, based on passive solar reflectance, have been developed to diagnose and evaluate plant health. These indices involve reflectance data from several wavebands and are generally preferred over single wavelength because they compensate for short- and long- term changes in solar irradiance and atmospheric conditions. The most common vegetation index is NDVI.

$$NDVI = (NIR - Red) / (NIR + Red)$$

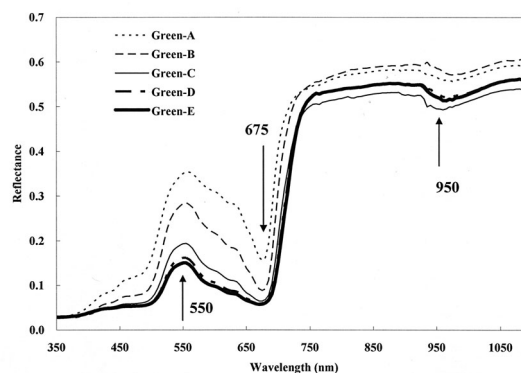
Where, NIR represents reflectance from the near infrared portion of the electromagnetic spectrum (760-900 nm) and is most sensitive to the living plant tissue. Reflectance from the red portion of the spectrum (630-690 nm), like much of the visual portion of the electromagnetic spectrum, is sensitive to chlorophyll. Both GreenSeeker™ and Crop Circle™ software provide NDVI output. Active sensors essentially eliminate the need to deal with temporal changes in solar irradiance and atmospheric conditions.

Therefore, the use of multi-waveband vegetation indices like NDVI may be sacrificing some of the differentiating power of single wavebands. However, if sensor height above the soil is not well controlled, some type of normalized vegetation index is recommended to correct for the strong sensitivity of active sensor output to distance from the target (especially <60 cm).

The NDVI was originally developed to correlate leaf area index of trees with reflectance patterns. Since then, it has been widely used to quantify the amount of living biomass of many targets. As noted above, the effectiveness of NDVI is questionable for canopies containing high levels of chlorophyll because red reflectance remains consistently low. Other indices (Gitelson, 2004) have been proposed to estimate specific parameters.

## Results and Discussion

Reflectance from 350nm to 1100nm for a harvested green leaf affected by the greenness classified by 5 level of maturity is shown in Figure 1. The reflectance pattern of tobacco leaf was similar to that of regular plant showing the little peak in green band, minimum in red band, and maximum in near infrared(NIR) band, respectively (Hatfield, 1993). The reflectance of green



**Fig. 1.** Reflectance of harvested leaf selected by greenness on the stalk position of cutter at the 68th day after planting. Harvested leaf was classified by 5 groups from light green to dark green symbolized from Green A to Green E.

and red band decreased as the degree of greenness increased (Schepers, 1998).

The correlation coefficient between chlorophyll content and reflectance indices for green leaf harvested is shown in Table 1. Chlorophyll content of harvested leaf was significantly correlated with all the reflectance indices estimated especially chlorophyll meter, SPAD 502 and CM 1000, gNDVI and aNDVI by Crop Circle™ and spectroradiometer showing more than 0.9 of correlation coefficients (Cater et al., 2002; Hong, 2003).

Figure 2 shows the reflectance from 350nm to 1100nm for a flue-cured leaf classified by 5 levels of maturity. The reflectance pattern of flue-cured leaf has a tendency to increase with longer wavelength from visible to NIR band showing different pattern from that of green leaf plotted in Figure 1. However the reflectance of green band for cured leaf decreased with immaturity, having a similar tending to that of the green leaf. This result was thought to be correlated to the more residual greenness in cured leaf due to the immaturity of tobacco leaves (Gausman 1977; Grant, 1987; Slaton et al., 2001;

**Table 1.** Correlation coefficient between chlorophyll content and reflectance indices by remote sensors (n=74).

Factor	Chlorophyll content	Chlorophyll meter	
		SPAD reading	CM1000 reading
SPAD reading	0.962		
CM1000 reading	0.965	0.965	
rNDVI(SR)	0.661	0.708	0.716
gNDVI(SR)	0.931	0.950	0.933
aNDVI(SR)	0.925	0.966	0.935
rNDVI(GS)	0.820	0.899	0.840
rNDVI(CC)	0.771	0.841	0.772
gNDVI(CC)	0.926	0.928	0.936
aNDVI(CC)	0.918	0.950	0.929

r= 0.295 at p<0.01

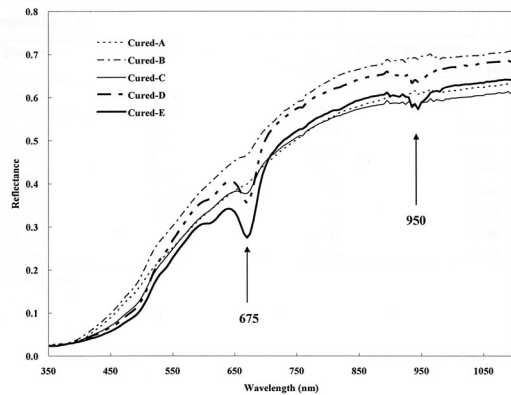


Fig. 2. Reflectance of flue-cured leaf selected by greenness on the stalk position of cutter at the 68th day after planting. Flue-cured leaf symbolized as Cured A to Cured E correspond to 5 groups of harvested leaf classified from Green A to Green E.

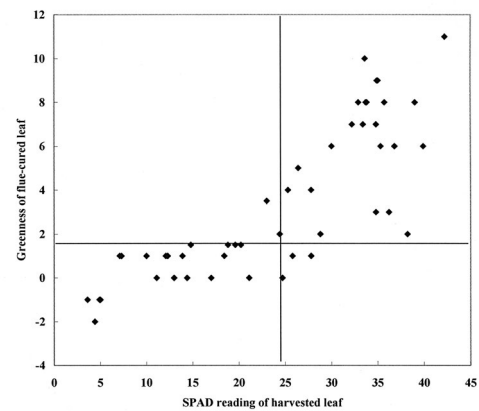


Fig. 3. Relationship between greenness of flue-cured leaf and SPAD reading of harvested leaf on the stalk position from cutter to tips.

Chappelle et al., 1992).

Interrelationship between green and cured leaf classified by 5 levels of maturity for color and chlorophyll characteristics, and reflectance indices by ground-based remote sensors was compared in Table 2. The degree of greenness of cured leaf was significantly correlated with the b value of colorimeter while chlorophyll characteristics and reflectance indices of the green leaf harvested showed significant negative correlation with L value of colorimeter. From the above result, the reflectance indices by ground-based remote sensors could be useful criteria of optimal maturity of tobacco leaves.

In Figure 3, the interrelationship between the degree of greenness of cured leaf and the SPAD reading of green leaf harvested by chlorophyll meter was plotted by Cate-Nelson graphical method, that is by dividing the Y-X

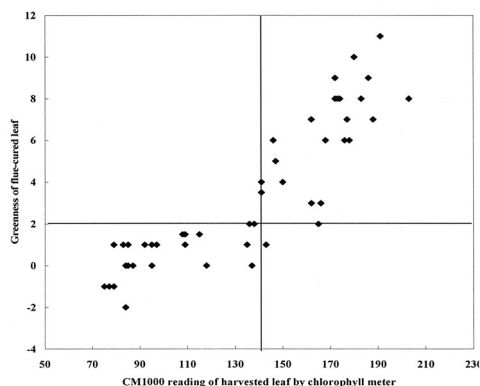
scatter diagram into four quadrants and maximizing the number of points in the positive quadrants while minimizing the number in the negative quadrants (Cate and Nelson, 1971). From the above graphical method, the criteria of SPAD reading for keeping less than 1.5 of the degree of greenness by the naked eye as the optimal quality of flue-cured tobacco was estimated using 24 of the SPAD reading. This result shows that more than 24 of SPAD reading for the harvested green leaf may be due to harvesting of immature tobacco leaves.

By the same estimation method as that of Figure 3, interrelationship between the degree of greenness of cured leaf and CM-1000 reading by another chlorophyll meter as well as between the degree of greenness of cured leaf and reflectance index, gNDVI by Crop Circle<sup>TM</sup> are shown in Figures 4 and 5, respectively. From the estimation in Figure 4, the criteria of CM-1000

Table 2. Correlation coefficient among greenness and L, a, b value of flue-cured leaf and chlorophyll content and reflectance indices of harvested leaf (n=50).

Factor		Flue-cured leaf			
		Greenness	L-value	a-value	b-value
Flue-cured leaf	L-value	-0.758			
	a-value	-0.092	-0.144		
	b-value	0.714	-0.386	0.095	
Harvested leaf	Chlorophyll	0.709	-0.677	0.319	0.685
	SPAD	0.703	-0.653	0.347	0.761
	CM1000	0.752	-0.766	0.354	0.655
	rNDVI(SR)	0.728	-0.857	0.642	0.512
	gNDVI(SR)	0.714	-0.698	0.411	0.707
	aNDVI(SR)	0.719	-0.662	0.423	0.783
	rNDVI(GS)	0.658	-0.517	0.357	0.822
	rNDVI(CC)	0.663	-0.373	0.208	0.791
	gNDVI(CC)	0.710	-0.641	0.306	0.632
	aNDVI(CC)	0.706	-0.275	0.320	0.727

r=0.354 at p<0.01



**Fig. 4.** Relationship between greenness of flue-cured leaf and CM1000 reading of harvested leaf on the stalk position from cutter to leaf (n=50).

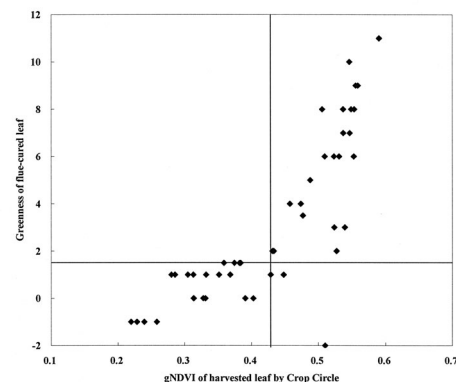
reading for keeping less than 1.5 of greenness degree by the naked eye as the optimal quality of flue-cured tobacco was estimated by 140 of the CM-1000 reading. The criteria of gNDVI by Crop Circle<sup>TM</sup> for keeping less than 1.5 of greenness degree by the naked eye as the optimal quality of flue-cured tobacco was estimated by 0.43 of the gNDVI as the reflectance index by ground-based remote sensor.

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### Summary

To determine the critical level for optimum maturity of flue-cured tobacco leaves (KF118) at the stalk position from cutter to tips, the reflectance index using ground-based remote sensors and chlorophyll meter were investigated. On a reflectance curve of 340nm~1100nm, the reflectance peaks on 550nm and 675nm for the harvested leaf were lowered as change from light green to darker green. Since harvested darker green leaf produced darker green flue-cured leaf, the reflectance on the wavelength of 675nm for flue-cured leaf decreased as greenness of harvested leaf increased. This result means that the red edge band of 675nm wavelength was related to the absorbance of chlorophyll for photosynthesis. The greenness of flue-cured leaf showed significantly positive correlation with the entire reflectance index for harvested leaf while the L value by colorimeter showed negative correlation with greenness of cured leaf. The critical level for optimum maturity of harvested leaf were less than 22,



**Fig. 5.** Relationship between greenness of flue-cured leaf and gNDVI of harvested leaf on the stalk position from cutter to tips (n=50).

135, and 0.43 for SPAD reading, CM-1000 reading, and gNDVI by Crop Circle<sup>TM</sup>, respectively. Consequently, ground-based remote sensing providing a non-destructive real-time assessment of plant greenness could be a useful tool for selection of optimum maturity of flue-cured tobacco leaves in relation to the high quality of flue-cured tobacco.

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## 연초 엽의 색 특성과 원격탐사 반사율지표의 상호관계

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지상원격탐사 센서를 이용한 황색종의 적숙엽 판단기준을 평가하기 위하여 성숙기에 중엽과 상엽부위에 대하여 수확엽의 녹색도를 5개 수준으로 구분하여 여러 가지 센서지표들의 상호관계를 평가하였다. 건조엽의 색 특성은 색차계(Colorimeter, CR-300)를 이용하여 L, a, b값을 측정하였고 건조엽의 청색도는 육안관찰에 의한 분포정도를 수치화하여 비교 검토하였다. 수확엽의 녹색도에 따른 반사율은 550nm와 675nm에서 녹색도가 증가할수록 감소되는 특성을 보였다. 건조엽의 반사율은 미숙된 엽이 건조된 후 녹색이 잔류되었기 때문에 미숙엽의 675nm 반사율이 더 낮아졌다. 그 결과 동일한 엽위에서 생엽과 건조엽의 반사율지표는 직선적인 정의 상관을 보였다. 또한 건조엽의 청색도는 모든 센서지표들과 유의성 있는 정의 상관을 보였고 명도를 나타내는 색차계 L값은 센서지표들과 유의성 있는 부의 상관을 보였다. 따라서 건조엽의 청색도로 평가된 센서 종류별 적숙엽의 기준은 엽록소 측정치 SPAD 값은 22 이하, 엽록소 측정치 CM-1000 값은 135 이하, 그리고 원격탐사센서 Crop Circle의 gNDVI 값은 0.43 이하로 평가되었다.