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Leaf Nitrogen Determination Using Non-Destructive Techniques -- A Review

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ABSTRACT

The optimisation of plant nitrogen-use-efficiency (NUE) has a direct impact on increasing crop production by optimising use of nitrogen fertiliser. Moreover, it protects environment from negative effects of nitrate leaching and nitrous oxide production. Accordingly, nitrogen (N) management in agriculture systems has been major focus of many researchers. Improvement of NUE can be achieved through several methods including more accurate measurement of foliar N contents of crops during different growth phases. There are two types of methods to diagnose foliar N status: destructive and non-destructive. Destructive methods are expensive and time-consuming as they require tissue sampling and subsequent laboratory analysis. Thus, many farmers find destructive methods to be less attractive. Non-destructive methods are rapid and less expensive but are usually less accurate. Accordingly, improving the accuracy of non-destructive N estimations has become a common goal of many researchers, and various methods varying in

complexity and optimality have been proposed for this purpose. This paper reviews various commonly used non-destructive methods for estimating foliar N status of plants.

Keywords

non-destructive, nitrogen, image processing

INTRODUCTION

The measurement of leaf chlorophyll contents and nutrient status in crop plants is crucial for agronomic studies of crop growth and yield quality. Various macro and micro nutrients are essential for plant metabolism, and their deficiency can severely limit crop yield. Nitrogen (N) plays an important role in key growth processes such as cell division, protein synthesis and enzyme production. If cell division is inhibited, leaf area expansion is similarly inhibited, and the plant thereby loses its potential to produce high yield. Despite of the importance of N supply to plant growth, excessive supply of fertiliser is costly and excess N that runs off arable land can negatively impact the environment. Thus a mismatch between N supply and crop requirement can hamper crop growth and harm the environment, resulting in low N-use-efficiency (NUE) and economic losses. Thus increasing NUE can reduce additional fertiliser application and protect the environment. However, for the best N application rate, the farmers and advisers must have the most up-to-date information on the crop and soil N status (Li et al., 2010).

There are two commonly used techniques for estimating tissue nutrient concentration; destructive and non-destructive. Ramirez (2010) showed that plant N status can be accurately estimated using a destructive technique; in which foliar samples are analysed using laboratory procedures. This technique is generally laborious, time consuming and expensive (Sui et al., 2005). In contrast, non-destructive methods are rapid and less expensive but are generally less accurate. There are a number of non-destructive methods available that vary in complexity and optimality. These include hand held methods such as the use of leaf colour chart (LCC), which relies on visual comparison between leaf colour and a colour chart to assess the N status of

certain plants. One of the most widely used digital tools is the chlorophyll meter (SPAD-502). This hand held device estimates the chlorophyll content of leaves and hence gives an indication of leaf N contents, as leaf chlorophyll level is closely correlated to leaf N concentration.

Recently, digital imaging has been investigated in the agriculture industry for plant colour analysis. Digital cameras or scanners in combination with computers and appropriate software can collect images of leaves and evaluate their colour with relative ease and at a reasonable cost. Hand held devices are quite appropriate for small fields. For large fields, tractor -- mounted systems sensors to detect N status are used that save time and effort for large scale N application. The three most commonly used commercial technologies that measure plant N status in real-time: Yara N-Sensor, Crop-Circle, and GreenSeeker (Samborski et al., 2009).

Remote sensing (RS) techniques have also been used for estimating nutrient contents in growing crops using a single wavelength or combination of wavelengths (Osborne et al., 2002). Due to their influence on leaf chlorophyll and photosynthesis, a strong relationship between leaf nutrient contents and spectral reflectance, particularly in visible absorption is expected. The RS techniques have been mainly used in natural resources management for land cover and biomass estimation, and to note changes in land usage (Sala et al., 2000, Kogan et al., 2004, Henebry et al., 2005). In the last decade, some successful efforts have been made to apply this approach to commercial agriculture. Below is a description of available techniques to estimate N content in plants.

MATERIALS AND METHODS

Handheld Meters

Non-digital Tools

Leaf colour is a good indicator of plant health and nutrition. Different types of stress may cause different symptoms, and a comparative analysis can yield information about the type of stress. N deficient leaves turn to pale or yellowish green rather than dark green and farmers generally prefer to dark green leaves of the crop. The standardised LCC used for estimation leaf nutrient contents contains four colours (Figure 1 a). The six- panel LCC, which was an improved version of the standard LCC (Thind and Gupta, 2010, IRRI, 1996) and the 8 -panel LCC (UCD-LCC), was recently developed by the researchers at University of California (Figure 1c).

The LCC has mainly been applied for rice crop. Despite the fact that it has made some improvements in NUE for rice growers, the LCC is not an optimal tool to assess foliar N content, because it is largely affected by a number of factors, such as ambient lighting condition and variations amongst the cultivars in leaf colour. Accordingly, the development of a more reliable tool for more accurately detecting the inception of N stress before it becomes visible to human eye would be valuable, especially if it can be used across a large range of species. Consequently the development of leaf chlorophyll meters has received much commercial interest (Debaeke et al., 2006).

Digital Tools

The SPAD meter

Minolta Co. (Japan) has developed the chlorophyll meter (SPAD-502 Figure 2), which effectively measures the relative greenness or chlorophyll content of leaves (Turner and Jund, 1994). Because leaf chlorophyll content is closely related to leaf N concentration (Balasubramanian et al., 1999), this meter has commonly been used to assess foliar N content. The SPAD meter estimates the relative chlorophyll concentration in a leaf by measuring the differential transmittance of light through it. Within a small chamber (2 - 3 mm) in which part of a leaf is held, the meter emits light from two diodes, one producing a peak wavelength near 650 nm (red), which is absorbed by chlorophyll and the other, a peak near 940 nm wavelength (near infra-red, NIR) is transmitted through leaves, and serves as an internal reference to compensate for leaf thickness and moisture content (Shapiro et al., 2006). More red light is absorbed by leaves when more chlorophyll is present. Thus, the chlorophyll concentrations of leaves are correlated with SPAD meter values. Understanding the basic theory of electromagnetic spectrum will give us good idea about the basic functional theory of SPAD.

The electromagnetic spectrum covers a wide range of wavelengths and photon energies, which is broken up into a number of different categories, each of which shares certain properties (Figure 3). Energy reaching to the surface can be either transmitted, absorbed or reflected (Figure 4). The physical properties of a body and the precise wavelength of radiation determine the degree of each, thus obtaining profiles of the surface reflectance (Figure 5). The spectral response of the plants mostly relies on the plant leaf structure. Layers of diverse types of cells make up the leaf (Figure 6). Each cell contains chlorophyll pigment in specialised structures called chloroplasts, which are responsible for the healthy green living vegetation. Every colour in the electromagnetic radiation spectrum is absorbed by chloroplast except green, which reflected

back. Approximately 60% of the NIR radiation from this leaf layer is reflected by the mesophyll cells resulting in the healthy green vegetation, and give a brighter and higher NIR response than in the spectrum for green. The leaf loses its green pigment upon senescence. The dying leaves have a brown and yellow appearance as red and blue is no longer used, so these reflect back green spectrum light. Also, the NIR wavelengths are completely absorbed by the leaf as they can no longer be reflected giving a dark or black appearance in the NIR (Gobson, 2000, Havránková, 2007)

Basic functional mechanism of SPAD

SPAD meter determines relative chlorophyll concentration by measuring the variance of light being transmitted through the leaf. Fairly large nonlinear relationship between leaf chlorophyll content and chlorophyll meter values allows calculation of a unitless “SPAD” value (on an index from 1 to 100). The SPAD values are calculated from the difference between optical density of red and infrared (IR) wavelengths, which is detected by a photodiode situated in the leaf chamber (Minolta Camera Co, 1989) As a result, the values calculated by SPAD meter reflect leaf chlorophyll concentrations. Since leaf chlorophyll concentration are positively correlated with leaf N concentration (as chlorophyll is made up of most of N containing enzymes and organic matter), a regression equation can be used to link the SPAD estimates to the foliar N status, and therefore, it can be used as a rapid diagnostic tool for measuring leaf N concentrations (Chapman and Barreto, 1997). The following equation is used for SPAD meter;

$$\text{SPAD} = A \left[\log \left(\frac{\text{RCo}}{\text{RC}} \right) - \log \left(\frac{\text{IRCo}}{\text{IRC}} \right) \right] + B$$

Where:

A and B are Constants

RC and IRC are Currents from red and IR detectors, respectively, that pass through the chamber that contains leaf sample.

RC₀ and IRC₀ are the Currents from red and IR detectors, respectively, without leaf sample.

Farmers often use the crop colour as an indicator, by performing a subjective visual inspection, to determine the time when N fertiliser is required (Wells and Turner, 1984, Furuya, 1987), and they prefer dark green colour of their crop, which leads to the overuse and inefficiency of N fertiliser. (Thind and Gupta, 2010) found that if the farmers used spectral properties in a more rational manner, they could be guided in a need based N application. Use of the chlorophyll meter allows early detection of the onset of N stress before it is visually obvious (Schepers et al., 1992) and assist in correcting N deficiencies and minimising yield losses (Shapiro et al., 2006). Thus chlorophyll meter is more reliable than the simple leaf colour chart visual assessments (Debaeke et al., 2006).

Broad testing has been done with the SPAD in wheat (Reeves, 1993, Fox et al., 1994), rice (Peng et al., 1993, Turner and Jund, 1994), cotton (Wu et al., 1998), tall fescue (Kantety et al., 1996), fruit trees (Li et al., 1998, Peryea and Kammereck, 1997) and maize (Chapman and Barreto, 1997). Use of the SPAD meter in predicting N status or foliar chlorophyll has been documented in over 200 publications (Uddling et al., 2007). A reading using the SPAD meter may be performed in 2 seconds, without destroying the plant tissue sample, and it will save space,

resources and time. The two main limitations of SPAD are; (1) high initial cost, which is approximately US\$ 1350 per unit, where even farmers in the USA (Turner and Jund, 1994) are reluctant to accept, and (2) relatively small measuring area (12.57 mm²) which may not reflect the true value of leaf chlorophyll and lead to fluctuating readings (Netto et al., 2002)

Estimation of Leaf Nutrients Using the SPAD Meter

In 1963, the original SPAD meter was designed to diagnose leaf N status of rice (*Oryza sativa* L.) in Japan (Takebe and Yoneyama, 1989), which included new models called the SPAD-501 and SPAD-502 (Uddling et al., 2007). When taking N readings from a crop, it is essential to ensure that plants samples are representative of the whole crop. It is necessary to have an average of approximately thirty readings to get a representation of the whole crop, and each individual reading may be different but anomalies should be recognised. Thirty individual readings are automatically stored, and the average is calculated by the Minolta SPAD 502. To achieve this goal, 30 individual samples must be collected and averaged from both the reference field and the bulk field, and then compared (Note: the first fully expanded leave from the top of plant must be used for each reading).

A guide to in-season N fertilisation, using the chlorophyll meter, is based on a sufficiency index, which is calculated as follows (Varel, 1997);

$$\text{Sufficiency index} = \frac{\text{Average Chlorophyll meter readings of unknown area}}{\text{Average Chlorophyll meter readings of well - fertilised area}} \times 100 \%$$

N is applied when the sufficiency index < 95%.

Balasubramanian et al. (1999) determined the SPAD threshold value in SPAD reading, which shows the N deficiency that could cause yield loss if not corrected (the crop is suffering N deficiency). Upon reaching this SPAD level, the farmers are able to take an immediate action (apply N), to limit possible yield reduction (Swain and Sandip, 2010). Nevertheless, Schepers et al. (1992) showed that cultural practices, stage of growth and crop variety being grown all affect the critical SPAD value. Critical SPAD value for different crop species varies, each crop has a specific critical SPAD value under specific growing environment (Huang et al., 2008). Balasubramanian et al. (1999) recommended SPAD threshold as 35 for dry season transplanting of rice, where 32 SPAD value is suggested for wet-seeded rice transplantations in cloudy weather or low radiation in Philippines, Maligaya and Nueva Ecija as 32. The critical SPAD value of 37.5 was determined by (Kyaw, 2003) for rice in Pakistan to receive need based N top-dressing. In West Bangal, (Maiti et al., 2004) the critical value was 37 for rice cultivar IET-4094 for high yields and less fertiliser N application to the tune of 27.5 to 45.5 kg N ha⁻¹ rather than a blanket dose of 150 kg N ha⁻¹.

Factors Affecting Leaf Chlorophyll Content

Various environmental and plant growth factors such as temperature, moisture stress and sunlight (Schepers et al., 1992), nutrient deficiencies other than N (Turner and Jund, 1991), varietal variations (Minotti et al., 1994, Hoel, 2003), plant genotype (Villa et al., 2000) and growth stages (Ramesh et al., 2002, Swain and Sandip, 2010), could influence leaf chlorophyll content or greenness. Variation in leaf chlorophyll concentrations, in turn affect the SPAD readings, varietal differences are generally the most common reason for variable SPAD meter

reading, some hybrids in crops such as tomato, sorghum and corn have darker green colour compared with varieties. Villa et al. (2000) found that SPAD readings could be easily affected by any factor changing leaf chlorophyll contents. Due to such a large range of factors influencing the plant chlorophyll, it is impossible for a meter to accommodate all crops for accurately measuring N sufficiency. In order to make the meter effective, it must be calibrated to the specific variety of the crop being grown as well as other environmental factors. To accomplish this several small areas (strips or spots) are over-fertilised with N, so a calibration may be taken using these spots as reference points for rest of the field (Murdock et al., 1997).

Limitations

There are mainly two limitations which restrict the use of the SPAD meter for indirect N measurement in plant the tissue (Rostami et al., 2008). The stress-induced sampling errors may influence chlorophyll contents in the plants (Villa et al., 2002), and the varietal / species based difference that may lead to variable results from different plant species using the same SPAD meter (Murdock et al., 1997). The latter problem could be avoided by calibrating SPAD meter for specific crop variety being grown. Furthermore, this method is not helpful for detecting luxury N uptake as maize plants will achieve maximum chlorophyll content regardless of the level of over-fertilization (Hawkins et al., 2007).

Other Digital Chlorophyll Meters

The Hydro N Tester or HNt (Yara International ASA, Oslo, Norway) is another type of chlorophyll meter (Figure 7) that uses two electromagnetic spectrum wavelengths (940 nm NIR

and red 650 nm) but the digital readout range for the chlorophyll index is from 0 to 800 (HNt). According to Richardson et al. (2001) the chlorophyll content meter (CCM-200, Opti-Sciences, Tyngsboro, Massachusetts, USA) weighs 180 g, 2 measurement area of 0.71 cm and based on the absorption measurements at 660 and 940 nm calculates a chlorophyll content index (CCI). In fact, both the SPAD and N-Tester have some common limitations

Another type of chlorophyll meter is the Spectrum CM1000 or R meter (Spectrum Technologies, Inc. 2009), which is a chlorophyll reflectance meter and hand-held working on parallel principles to the SPAD meter. The Spectrum meter works on the fine-leaved turf stands canopy. This allows larger area assessment, and integrates many leaf surfaces. At 30 cm from the plant leaf, it integrates 1.27 cm diameter area; it gives consistent readings, and is simple to use. At the pull of trigger chlorophyll, measurements may be made from a standing or walking position. The sun angle and heavy overcast conditions restrict the use of the R-meter, as it is dependent on ambient sunlight. Figure 8 shows, how the sunlight intensity, meter angle to wheat canopy, sun angle and sunlight intensity influence the R-type meter readings

Recent work based on the use of transportable NIRs systems have been published (Ecartot et al., 2013, Serbin et al., 2012). Since these NIR systems are not affected by genotype or environmental factors, these can present a better alternative for developing leaf N content calibration at species scale. Edrees et al. (2013) used the HandHeld 2 Portable Spectroradiometer to detect chlorophyll content from remotely sensed spectral data for optimising N doses without reducing wheat yield. The comparisons between these handheld tools are shown in Table (1).

Digital Image and Colour Analysis in Agriculture

Plant nutrition and vigour are closely associated with foliar colour and therefore, changes in leaf colour indicate changes in plant health (Graeff et al., 2008). Some scientists suggested the computer automated digital image analysis is an alternative, unbiased, more precise and consistent method for analysing leaf nutrition (Turner et al., 2004, Mirik et al., 2006). It is a non-invasive and non-destructive method for capturing, processing and analysing information from leaf images (Richardson et al., 2001, Karcher and Recharadson, 2003). This method allows simultaneous collection and analysis of hundreds of images at convenience (Diaz-Lago, 2003). Currently, the equipment for this process are inexpensive that makes it an attractive method of data collection and storage. Digital imaging analysis has been effectively used in crop studies for quantifying water deficiency, nutrient status, and disease, insect and stress-induced damage (Karcher and Recharadson, 2003, Richardson et al., 2001, Adamsen et al., 2000).

Use of Digital Image and Colour Analysis for Plant Growth and Yield Estimation

Various applications have been considered for the RGB (red, green and blue) based image analysis in agriculture i.e. weed recognition (Ahmad et al., 2006), weed and crop mapping (Tillet et al., 2001), weed identification (Hemming and Rath, 2000), seed colour test for identification of commercial seed traits (Dana and Ivo, 2008), quantitative analysis of specially variable physiological process across leaf surface (Aldea et al., 2006), quantification of turf grass colour (Karcher and Recharadson, 2003) and weed / crop discrimination (Aitkenhead et al., 2003). Various RGB and HSI (H = Hue, which in turn measures the permeated colour, S = saturation, the colour permeated with white colour and I = light intensity)-based methods have been used for discriminating between plant and soil have been described by Georg and Bockisch (1992) that guided the development of an automatic seedling transplanting machine (Lin et al., 1994).

These studies suggested higher efficacy of digital imaging analysis for quantifying biophysical plant parameters, especially when dealing with leaf diseases such as rust and tan spot in wheat (*Triticum aestivum* L.). However, the impact of image size, format, quality and sample size on digital image analysis results was determined covering a range of disease intensity (Steddom et al., 2004). They concluded that digital image analyses for disease quantification, even using the low quality JPEG (joint photographers expert group) images, is extremely desirable due to robustness, low-cost and commercial availability of equipment. This technique is emerging as a favourable tool for crop yield management due to the ability to detect crop stress situations before visual symptoms appear and adverse effects established. This technique also allows for the digital images to be archived for future use, therefore, maximum data is available for future users. Colour parameters of the digital images may be easily interpreted by the computer processing system and evaluated using different colour systems. Erickson et al. (1988) reported that the RGB colour values could be effectively used for analysing and describing colour images.

To study the influence of illumination on the quality analysis of apple using image processing, Truppel et al. (1998) used three different colour spaces RGB, HSI and L^*a^*b (where L = lightness component, a = determines the degree from green to red, and b = determines the degree of blue to yellow). They concluded that both the selected colour space and illumination were very important for developing good quality images. Using various colour spaces [(HIS, Luv, (CIE 1976, L^* , u^* , v^* color space), Lab, (CIE 1976 L^* , a^* , b^* color space)], Chapron et al. (1999) developed a system that efficiently recognises the weeds in maize fields if foliar overlapping was less than 5%. To improve the separation of soil and vegetation they tested different colour spaces like HSI and Lab. Similarly aerial images with high resolution IR were

used for detecting N stress in maize Gopala Pillai et al. (1998). They determined that canopy reflectance in the red channel a good predictor of maize yield, as the canopy reflectance was closely correlated to the applied N. For assessing N status of bush bean plants, Thai et al. (1998) used spectral video images utilising two handpass filters and distinguished different N application level using two selected bands. Minimum of light deviation, maximum resolution and image distortion are important parameters for developing good quality scanned images (Cometti et al., 2003). The video / image camera are the most practical approach for determining the leaf chlorophyll and nutrient status (Chappelle et al., 1992, Ferns et al., 1984, Curran et al., 1991). Without making any contact with the plants, digital camera images can be acquired from a point in time. This method allows for a more accurate and reliable measurement of plant growth and no plant harvesting is required.

Use of Digital Imaging for Plant N Estimation

Using colour imaging technique Luna et al. (2010) successfully examined the tomato leaf polyphenolic and chlorophyll content, and correlated them with leaf N status. Many other field studies suggested the use of video camera and computer based image analysis as an effective method for estimating chlorophyll content of and nutrient status of crop plants. Jia et al. (2004) indicated that level of N fertilisation can be detected using digital photography, while Yadav et al. (2010) showed that real time predictions of chlorophyll content of plants could be achieved by image analysis method using the three primary colours, red (R), green (G) and blue (B). A pixel in a digital leaf image is represented as a combination of the three primary colours, which can be used to develop a mathematical formula that reflects correlation with the chlorophyll content of

the plant (Su et al., 2008). Pagola et al. (2009) developed a new technique for measuring leaf greenness using the RGB components of a colour image and established a greenness index, which estimates barley yield and N requirements. They compared the values estimated by new method with the value given by the SPAD-502 chlorophyll meter observed that RGB-based greenness index gives an equal or better prediction to that of SPAD.

The normalised variance (red-blue) / (red+blue) estimated by portable video camera was found the most relevant variance for data collection under variety of meteorological conditions (Kawashima and Nakatani, 1998). Using variable algorithms of RGB values obtained from video images, various plant growth parameters have been estimated for example (Iwaya and Yamamoto, 2005) used $(R-G)/(R+G)$ equation for measuring water content of wheat panicle (Suzuki et al., 1999) used $G/(R+G+B)$ value for measuring chlorophyll content in broccoli, Cai et al. (2006) used $R/(R+G+B)$ and G/R and $R/(R+G+B)$ for estimating leaf chlorophyll and carotenoid contents respectively of cucumber leaf. Researching wheat senescence, Adamsen et al. (1999) observed a linear correlation between the G/R and SPAD values, and the G/R efficiently responded to the changes in both leaf chlorophyll concentrations and leaf number. The image colour analysis has also been applied to studies on nutrition deficiency of plants (Xu et al., 2002) and flower number detection (Adamsen et al., 2000).

Despite the fact that all analysis performed by researchers was carried out under different environmental conditions, with different plant materials and selected optimum indices, most of them analysed the RGB image by ratio values, while concentrating on plants colour variances and more particularly on plant canopy chlorophyll evaluation (Cai et al., 2006). The provision of

N estimates can be quickly conducted using RGB image analysis. On the contrary, a SPAD chlorophyll meter was unable to estimate real time leaf chlorophyll content of regenerated plants enclosed in a culture vessel (Yadav et al., 2010). Regenerated plants chlorophyll estimates could be much better estimated utilising RGB based image analysis.

Whilst researching cucumber plants grown in a greenhouse under various levels of nutrients, Qin and Zhang (2005) developed a method to employ a special image sampler to take leaf images and determine correlation between the image property and leaf N content. The system involved a platform, eight lamps and a window for fixing the camera. Images were taken by placing the leaves on the platform, around which the lamps were arranged. RGB and HSI modes were used to analyse the correlation between leaf N content and leaf images. The use of the camera proved more efficient and faster for obtaining the highest correlation for cucumber leaf N content. A strong correlation was achieved for N status of broccoli plants when digital images were taken under unchanging light conditions Graeff et al. (2008). By minimising the image taking limitations such as use of automatic camera setting, the higher width camera angle and using a 100 W lamp to control light, Luna et al. (2010) developed a method of taking images of tomato leaves grown in a greenhouse (Figure 9). Red and blue colour of images was used for estimating and analysing N status of tomato seedlings. With R^2 above 0.89, it was shown that red and blue colours yielded a good N predication. Thus this method could quickly estimate more and efficiently the N deficiencies in tomato seedlings.

A recently developed hand-held crop fluorescence sensor is Multiplex®, which detects leaf chlorophyll contents using four wavelengths i.e. Blue (450 nm), Green (530 nm), and Red (630

nm). This optical sensor is more efficient as compared with the chlorophyll meter as it can distinguish N treatments under any irradiance level such as shade or full sunlight and can be used at any time during of the day. However, the short distance (approximately 10 cm plant) requirement for measuring leaf chlorophyll contents and small covering area (50 cm²) limits its use for larger areas (Muñoz-Huerta et al., 2013).

Evaluation of LCC, SPAD and Image Processing-Based N Estimation Meters

Various factors influence the suitability and applicability of different N estimation meters. LCCs are the cheapest, and have been widely used in rice, maize and sugarcane. However, as this approach is based on visual inspection of leaf colour and the accuracy is not guaranteed, especially for different lighting conditions. SPAD on the other hand, is less sensitive to lighting conditions, but has not shown consistent performance across all species. The fluctuation of SPAD performance is influenced by its small measuring area (around 12.57 mm²). The relatively high cost of SPAD makes it less appealing for small farmers, especially, from developing countries. Similar to LCC, image processing techniques are also affected by the environmental conditions, as these have to be set appropriately to produce reliable results across the different species. Despite being only applied to limited number of species, image processing showed good potential compared to SPAD and LCC. Image processing technique may require calibration, but their cost is generally less than that of SPAD. In general, each of these methods may have their own advantages and disadvantages. However, the image processing technique is yet to reach its full potential. Table 2 shows a comparison between LCC, SPAD and image processing based N estimators.

Tractor - Mounted Systems

Handheld meters are more suitable for a limited area and most farmers use them in small farms. For the large farms, tractor mounted systems are more appropriate to detect leaf chlorophyll and N contents in plants. These systems are quite fast and labour efficient. The compact size and low weight design allow easy adaptation of these devices to pole-mounted, and the information produced by sensors is utilized to quantify the crop N deficiency. Currently, there are three main commercially available technologies used on board to assess the plant N status in real-time to drive fertiliser spreaders and apply variable N rates to crops: Yara N-Sensor / FieldScan, Crop-Circle, and GreenSeeker (Samborski et al., 2009). According to (Kim et al., 2000) tractor-mounted systems are based on sensors that measure crop characteristics saves time and avoids any delay between the assessment of needs and the actual N application time.

(Philippa et al., 2012) conducted a series of experiments to compare the performance of 3 visible / near infrared (VIS/NIR) sensors such as Greenseeker™ from Trimble, Crop Circle ACS-470™ from Holland Scientific and CropSpec™ from Topcon for N fertiliser estimation for crop production. Despite of variation in the sensing footprint, wavelengths used and indices the data collected from all these sensors had a common management purpose. Even the tractor-mounted systems also have some limitation, as it cannot directly estimate crop N requirement (Mulla, 2013). Thus scientists developed N fertiliser response functions by comparing the sensor readings with readings in reference strips from the crops receiving adequate N supply that helped to overcome this issue (Scharf et al., 2011). Using different active sensors (GreenSeeker RT100, Holland Scientific CropCircle ACS 470, YARA N-Sensor ALS), Kipp et al. (2014) studied how

environmental variation such as light intensity and temperature, and measuring distance influence the accuracy of spectral reading. Depending on the type of sensor used, measuring distance (between sensor and target surface) was found the main determinant of the accuracy of spectral readings. Optimum measuring distances from crop canopy to sensor was set from 10 to 200 cm that enabled stable sensor outputs. Depending on the sensor and spectral index, device temperature variably affected the spectral readings but light conditions had little or no effect on the performance of these sensors.

GreenSeeker

GreenSeeker[®] is an incorporated system of optical sensor and application system for optimising N application (Figure 10). This unit emits light in two wavelengths and the light reflectance from target (plants in the soil) is measured. The GreenSeeker^{®TM} active lighting optical sensor uses high intensity light emitting diodes (LEDs) that radiate light at 780 nm (NIR) and 600 nm (red) as light sources. These LEDs are pulsed at high frequencies. The photodiode detector measures the magnitude of light reflected off the role (Figure 11). Background illumination is eliminated by electronic filters. The magnitude of the filtered signal is measured by a multiplexed A/D converter (Convert the signal from analog-to-digital). Measurements are collected and averaged across the treatments and sensing distance of 0.61 m. The normalised difference vegetation index (NDVI) is calculated from the NIR and red values by the computer. GreenSeeker[®] sensors have a 0.6 m field of view and 0.8 -- 1.2 m above the plant is the optimal sensing height. The number of sensors used and their spacing affect the percentage of area covered by the mapping system. The sensor analysis application rates and works out the mix of the three valves required to have this

rate applied. The computer then forwards the data to valve control module computer, and that controls the valves (Industries, 2005).

As previously stated, the GreenSeeker[®] ascertains crop status by calculating NDVI using IR and red light. During photosynthesis, the red light is absorbed by the plant chlorophyll as an energy source. Plants are healthy when they are reflecting more NIR and absorbing much of the red light. Varying light conditions have very little impact on the GreenSeeker[®] measurements as it is an active sensor (Jones, 2004). GreenSeeker[®] sensors have been used in several published studies for detecting crops N status (Arnall et al., 2006). Many producers have been reported to improve N fertilisation of cereal crops by an efficient GreenSeeker[®] application (Mullen et al., 2003). Raun et al. (2002) observed 15% improvement in NUE of wheat crop using the GreenSeeker

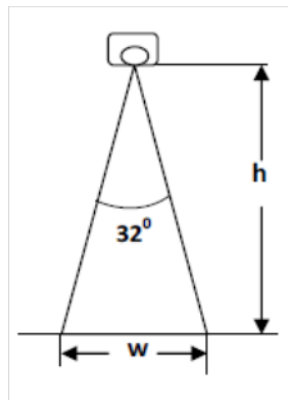
Crop-Circle

The Crop-Circle sensor ACS-210 ('Crop Circle', Holland Scientific Inc., NE, USA) (Figure 12) can measure plant reflectance on-the-go using light sensor (Havránková, 2007). It can either be hand-held or mounted on any type of vehicle. The ACS-210 uses PolySource[®] technology containing a single LED light source which emits NIR and visible light simultaneously. There are two sensor models, which use yellow/NR (590 and 880 nm) and red/NIR (650 and 880 nm) sensing capabilities. Spectrally sensitive photo sensors capture portion of light reflected back to the sensor from plant canopy. The ACS-210 minimises its dependency on other light sources by distinguishing its own light source which is a modulating (rapid pulsing many times per second).

In this way regardless of the light conditions (artificial lighting, complete darkness, cloudy skies, full-sun etc.), the ACS-210 remained efficient and unaffected.

Sensor can run from 25 cm to 213 cm. The width of the projected beam when the sensor mounted above a target is defined by the following equation;

$$w = 2.h.\tan\left(\frac{\theta}{2}\right) \approx 0.57.4$$



Where:

θ = Angular FoV (field of view) in degrees (≈ 32 degrees for the ACS-210)

w = Projected beam width

h = Height of the sensor above target

The output capacity of Crop-Circle sensor ranges between 1 to 20 samples per second. The samples are collected using a standard RS-232 interface with a laptop, personal digital assistant (PDA) or other data acquisition device, and stored using a comma-delimited format. Basic

reflectance and the classical vegetative index are provided by the sensor (HollandScientific, 2004). However, recently a new model of Crop-Circle sensor ACS-470® (Holland Scientific Inc., Lincoln, Nebraska), which can be spectrally customised measures 20 samples per second. The sensor measures reflectance from the crop canopy at 0.8 m distance and have a footprint of 0.1×0.5m.

The working process of ACS-470® is similar to that of GreenSeeker system, and it has been used to detect biomass and leaf area of many cereals (Shearman et al., 2005, Trotter et al., 2008_ENREF_36). It is a three channel instrument that has a range of interference filters in the VIS and NIR range, the filters are placed in the respective channel, usually one filter is in the NIR range (Channel 2) and normally the other two in the VIS range. Each channel gives an actual value and then computing ratios or indices it finally give five outputs. The ACS-470 is an active sensor, which has optimal sensor-to- canopy distance is 30 to 36 inches (750 to 900 mm). This system has more flexibility in terms of programming and use of sensors in the range of 440 to 800 nm. Each sensor contains six filters as a standard set, these include, 450, 550, 650, 670, 730 and 760 nm.

Yara N-Sensor

The Yara passive N-Sensor / Fieldscan (Yara International ASA, Oslo, Norway) is a tractor-roof-mounted multispectral scanner (Zebarth et al., 2003, Berntsen et al., 2006), which utilises a two diode array spectrometers (Figure 2.13). With two spectrometers on either side of the vehicle, the spectrometer utilising four lenses with an oblique view (64° with the zenith, solar azimuth effects are largely avoided by the special viewing geometry), to analyse the light reflectance from crop.

This allows simultaneous measurements from both left and right side of the tractor if desired. Approximately 25% of the total area is scanned at a 24 m working width. The second spectrometer measures the ambient light for permanent correction of the reflectance signal and guarantee constant measurements under variable irradiance conditions (Reusch et al., 2002, Zillman et al., 2006). The N status of crop may be gauged by determining crop reflectance characteristics using selected wavebands from 450 to 900 nm, and then using algorithms to calculate optimal N application rates either from NDVI or from other vegetation indices. The data is transmitted to spreader equipment which controls the N fertiliser application rate on-the-go (Reusch et al., 2002). Link and Reusch (2005) presented N-Sensor ALS (active light source) in 2005 that have 24 hours per day working capacity due to its own light source and had an on-the-go capacity to vary N application rates irrespective of ambient light conditions. The passive N-Sensor is comparable with effective geometry and spectral channels and has been efficiently used for N management in various crops such as potato, maize, wheat, barley, onion and oil seed rape (Yara, 2006, Havránková, 2007). In order to minimise the errors caused cloud cover and low light, a new version of Yara-N with active sensor is introduced, which improved the overall performance of the sensor (Link and Reusch, 2006).

A dealer of the N sensor in Czech Republic, Company Leading Farmers reported approximately 5% increase in crop yield when N sensors were used (Farmers, 2006). Keeping in view of the optimal yield potential of individual fields, Ebertseder et al. (2005) recommended that the technology should be improved by taking the soil conditions into account whilst using the sensor.

Crop Reflectance

Crop reflectance is the ratio of the amount of radiation reflected by an individual leaf or leaf canopy to the amount of incident radiation (Schröder et al., 2000). Chappelle et al. (1992) found that dark green plants exhibit relatively low light reflectance and transmittance into the visible spectrum due to pigmentation and photosynthetic tissue. Visible light is selectively absorbed by the pigments involved in photosynthesis (chlorophyll a and b). Also, the green wavelength is reflected, whilst the red and blue wavelengths of the visible spectrum are absorbed. Hence a good indication of how green the crop is can be measured at these wavelengths using crop reflectance.

Chlorophyll contents in plants is the most important feature that influence leaf reflectance in certain parts of the spectrum, and is the most advantageous tissues in leaves containing sufficient N contents (Gausman, 1974). Sunflower leaves suffering N stress were found to by to have higher reflectance in the visible part of electromagnetic spectrum and a lower reflectance in the NIR (Filella et al., 1995). The NDVI and the total forage N update were found highly correlated (Stone et al., 1996). Using an assortment of spectral indices and optical techniques Filella et al. (1995) determined N status of wheat crop, which looked promising. Similarly Zhao et al. (2005) found foliar N deficiency significantly reduced of the leaf chlorophyll content, which subsequently reduced the leaf reflectance of approximately 550 and/or 710 nm spectrum (Carter and Estep, 2002).

On the basis of multispectral images, various ways have been investigated for assessing crop N stress (Yang and Anderson, 2000, Noh and Zhang, 2003). Leaf chlorophyll contents are tested in

various ways to increase the sensitivity; some NDVI are defined by a ratio, a variance, or a difference / ratio combination of the measured reflectance at different wavelengths (Gitelson et al., 1996). Chapman and Barreto (1997) obtained low chlorophyll content data, and confirmed it through field testing the results suggested that NDVI approach was more sensitive than the direct reflectance approach. Multispectral reflectance analysis is now used on RS images to give a synoptic view of crop N status to producers with spatial variability (Moran et al., 1997, Lee et al., 1999, Thai et al., 1998). Further investigation has shown that whilst RS can provide producers with a comprehensive overview of crops and greatly assist with variable rate N management, the technical difficulties requiring professional suppliers and associated costs e.g. accessibility to satellite / aerial images, calibration and registration of images (Zhang et al., 2002, Noh et al., 2005).

Reflectance and NIR area has been related to crop N status in various ways. Crop leaves which give stronger spectral reflectance in particular NIR and blue wavebands have higher N contents. Alternatively, crop leaves containing lower chlorophyll and N contents reflect more spectral in red region and less in NIR region (Serrano et al., 2000). N deficiencies in plant tissues have been detected using various reflectance indices and ratios (Plant et al., 2000, Lukina et al., 2000).

Vegetation Indices

Vegetation indices are the values generated using reflectance measurements from two or more spectral wavelengths. Reflectance is the ratio of the total amount of radiation (energy) reflected by a surface to the total amount of radiation incident on the surface. The correlation of vegetation indices with biomass, leaf area index (LAI), N status or with yield depends on the index used

(Thorp and Tian, 2004). Some of the most commonly used vegetation indices are ratio vegetation index (RVI), difference vegetation index (DVI), green vegetative index (GVI), and land perpendicular vegetation index (PVI) (Ramirez (2010). NDVI is one of the most widely used vegetation indices that provide a simple answer for vegetation health. It can be calculated by the following simple formula;

$$NDVI = \frac{(NIR - RED)}{(NIR + RED)}$$

Where NIR is the reflectance in the near infrared region (770 ± 15 nm) and RED is the reflectance in the red region (650 ± 10 nm) of the electromagnetic spectrum. Reflectance in NIR region is positively correlated with the leaf area, while reflectance in the red region is negatively correlated with the green leaf area (Knipling, 1970). Likewise, vegetation reflectance is governed by the contribution of stems and leaf orientation to canopy reflectance (Carter and Estep, 2002). The NDVI value shows a discrepancy between -1.0 and +1.0. NDVI values for vegetation typically range between 0.2 and 0.7. Practically, a value less than 0.2 usually mean unhealthy vegetation and a value close to +1.0 (more than 0.6) means very healthy green leaves or very dense vegetation, whereas, zero or negative values means no vegetation. Nevertheless, real wavelengths and bandwidths for NDVI calculations varied with different sensors and applications.

A good relationship has been found among the SPAD readings, sap nitrate concentration, aboveground biomass and total tissue N concentration, and the spectral indices DVI, GNDVI, NDVI and RVI (Flowers et al., 2003, Lina et al., 2007). A typical reflectance spectrum was

suggested by Kumar and Silva (1973), which identifies healthy and stressed plants using the reflectance ratio of the energy reflected from an object to the energy incident on an object (Figure 2.14). The spectral reflectance of a crop significantly varies within the electromagnetic spectrum in the NIR range ($\lambda = 700\text{-}1300$ nm), and in the visible red range ($\lambda = 550\text{-}700$ nm). Plants identified as green due to low reflectance in the red and blue wavelength range, high chlorophyll absorption, and a high reflectance rate of green wavelength. In case of NIR radiant energy, various leaf attributes such as cellular structure and the air-cell wall-protoplasm-chloroplast interfaces determine the level of reflectance from plant surface (Kumar and Silva, 1973). Considering the difference between soil and vegetation in the NIR and red regions as maximum, Adamsen et al. (1999) suggested that the spectral reflectance data could be effectively used to calculate a diverse range of vegetative indices, which are well-correlated with biophysical and agronomic parameters connected to plant productivity. Since the vegetative indices utilise both red light and NIR, Carlson et al. (1990) stated that NDVI is effective in forecasting photosynthetic activity. Chlorophyll content determine the level of photosynthetic activity of a plant, and are associated with leaf N (Chapman and Barreto, 1997). Thus estimating leaf chlorophyll contents could help to determine vegetation nutrient level

Near-real-time and real-time in field nutrient management can be enhanced with the use of Remote Sensing (RS) technology. Negative environmental impact and costs can be minimised by monitoring the changing conditions in the field that gives the producers an advantage of adjusting fertilisers and / or chemicals application rates and times accordingly (Godwin et al., 2003). Depending on the platform and type of RS system, the use of RS is widespread in precision farming technology i.e. it helps to determine crop yield (Freeman et al., 2003, Aparicio

et al., 2000), weed detection, water stress, and crop N deficiency (Scotford and Miller, 2005, Yang et al., 2005, Goel et al., 2003).

Nusz (2009) showed that the RS is the science and art of collecting information about an object, area, or phenomenon through analysing the data acquired by a device that is not directly in contact with that specific object, area, or phenomenon (Lillesand and Keifer, 2000). Henebry et al. (2005) found that natural resources management in the area where RS was extensively used have exhibited significant improvements in land use, biomass estimation and land cover estimates. Some success has been seen in the last decade with implementing this system into commercial agriculture.

Several studies confirmed a close relationship among crop N status, chlorophyll contents and spectral reflectance that propose a high potential of RS application in the precision agriculture (Barnes et al., 2003, Pinter et al., 2003). However, the adoption of RS by farmers is limited due to lack of training, high costs and time consumption in imagery (Robert, 2002). Within the precision agriculture, there is a great capacity of RS applications for improving crop production and quality (Barnes et al., 2003, Pinter et al., 2003). RS has long been known as a non-destructive method, and subsequently after its first introduction to agriculture in early 1920s, when aerial photography was used to assist the soil mapping it have been successfully implemented for various agricultural operations such as rangeland surveys, yield forecasting, crop identification and monitoring for disease or insect pressures in crops (Riedell et al., 2004).

Many field studies have witnessed the RS and vegetation indices application as a successful technique for detecting various stresses in crop plants. The first use of aerial photography for

detecting plant viruses in crops was conducted by Bawden (1933) in potato (*Solanum tuberosum* L.) and tobacco (*Nicotiana tabacum* L.). Following this, stress detection using spectral data has been used for wheat (Jones, 2004), barley (*Hordeum vulgare* L.) (Nilsson, 1995), soybean (*Glycine max* L.) (Adamsen et al., 2000), maize (*Zea mays* L.) (Kim et al., 2000), sugar beet (*Beta vulgaris* L.) (Laudien et al., 2004), alfalfa (*Medicago sativa* L.) (Guan and Nutter, 2002), watermelon (*Citrullus lanatus* Schrad.) (Blazquez and Edwards, 1986) and tomato (*Lycopersicon esculentum* L.) (Zhang et al., 2003). Moreover, Alchanatis and Schmilovitch (2005) mentioned its successful application for crops, predominantly the grain crops like barley and winter wheat. The use of RS in precision farming technology is wide; it depends on the platform and system of RS used. RS systems are used to for assessing crop yield N deficit, weeds, water stress (Yang et al., 2005).

RS is now essential in many fields such as, mining, urban planning and forestry but its application in agriculture industry has been slow. In fact, there are many factors within agriculture that influence the rapid acceptance of RS in the agricultural context such as distrust of the farming community on RS for example low profit margins compared to high cost of imagery, limited spatial resolutions and timeliness of data (Robert, 2002).

Types of Remote Sensing Systems

Havránková (2007) indicated two basic types of RS systems i.e. active or passive (Figure 2.15). Passive system measures naturally emitted energy from the sun or earth. According to Gobson (2000), the process of the solar radiation interacting with the surface (e.g. reflection) and the sensing equipment enable to measure the energy that is reflected. The electromagnetic radiation

source in active RS equipment is carried on-board. The electromagnetic radiation is reflected off the surface and then a record is made of that scattered back from the surface (Gobson, 2000).

Remote Sensing Platforms

Satellite Platform

A satellite has capacity to collect images covering a much greater area than that of a plane. However, the quality of obtained images could be affected by various factors such as nutrient disorders, which are not related to N status of the crop, soil water content, weather conditions and soil background reflectance (Gerard et al., 1997, Shou et al., 2007). Multi-spectral images are now commercially available from satellites, and the future might bring higher spatial and spectral resolution images. Satellite based radar systems (SAR) have been effectively used for studying soil (Leconte et al., 2004) and crop moisture contents (Griffiths and Wooding, 1996).

New opportunities for distinguishing spatial details have been offered by the successful launch of high-resolution satellite sensors i.e. QuickBird and IKONOS. The QuickBird's spatial resolution imagery data is 0.61 m for panchromatic and 2.44 m for multispectral images at nadir (The point on the ground, which is directly below the QuickBird spacecraft) (DigitalGlobe, 2002). Yoder and Pettigrew-Crosby (1995) observed that it was imperative to discriminate between field N deficiency and differences between nutrient and water stress. Therefore, the QuickBird's imagery, which uses high spatial resolutions, and NIR (760-900 nm), red (630-690 nm) and green (520- 600 nm) wavelength bands, provides an essential support for this. However, making timely management decisions using QuickBird technology may not necessarily possible due to

slow turnaround of image processing, clear sky conditions, high cost of commercial satellite images and potential cloud interference during image acquisition (Weiss et al., 2000, Wu et al., 2007).

Airborne Platform

This technique provides high spatial resolution and possibly short revisit time, but its application is limited by high operational complexity, costs and inconvenience to control in windy conditions. The potential use of airborne data for cereal production has already been demonstrated (Wood et al., 2003). Digital cameras or radiometers could obtain images, while an electrical signal proportional to the light energy exposed is produced by radiometers. Hence, the radiometer is exposed only to the specific wavelengths required by filtering the light. Scotford and Miller (2005) observed that an optical band pass filter may be fitted onto a digital camera, whereas Morris (2006) evaluated the case for aerial images being used;

- The adaptability of flight schedules, for example as required by husbandry considerations.
- Images have a high resolution.
- Economical and relatively rapid capacity to map large areas.
- Turnaround time is feasible e.g. a few days.
- The producer can be involved with the system.
- If repeating the imaging is required, turnaround time is short.

While negative aspects of aerial image use:

- Clear weather conditions are required for images collection
- It is uneconomical for only a small area due to the cost of running the aircraft
- Images need to be ground calibrated.

A substantially large area can be surveyed in short time using remote sensing from aerial platforms (Shou et al., 2007). Aerial photography was successfully used to vary in-season N application rate, reproductive growth phases of crop, prediction of side-dressing N rates for assisting with growth by utilising green or blue colour spectrum photography (Scharf and Lory, 2002). Aerial photographs taken from a helium filled balloon were used to monitor growth status of winter wheat (Jia et al., 2004). According to (Quilter and Anderson, 2000), aerial photography from radio controlled model aircraft could deliver high-resolution imagery at a relatively low cost. Hunt et al. (2002) advised that model-aircraft photographic imagery has considerably lower costs spatial resolution associated compared with satellite and airborne sensors. Model aircraft NDVI by colour IR film with a low-cost automatic camera was found equivalent to NDVI from the advanced sensors. (Hunt et al., 2003) used an Olympus D40 4.1-megapixel colour digital camera (Olympus, Inc., Melville, NY, USA), and found it useful for precision agriculture and obtained high spatial resolution at relatively low cost that was comparable with the NDVI from advanced sensors. However, they experienced various complications such as overexposure of the film, lack of radiometric and spectral calibration, and the requirement of long landing and take-off areas during the initial work.

In order to use aerial photography for predicting crop responses at early vegetative growth stages, high resolution digital cameras and/or lower flight elevation are recommended to improve image resolution and remove non-crop reflectance (Williams et al., 2010). According to Hunt et al. (2003), airborne and satellite sensors can provide either high cost/high resolution imagery or low cost/low spatial resolution for use in precision agriculture.

CONCLUSION

N estimation is crucial for obtaining higher productivity at economical costs. Out of two techniques for N estimation, destructive technique is expensive and laborious, whereas, non-destructive methods are rapid and less expensive. There are a number of non-destructive methods available that vary in complexity and optimality, which include handheld meters like LCC which has found to be widely adopted in developing countries due to its low cost and relative advantage. Digital meter devices, such as SPAD and N-Tester are usually more accurate than LCC, but more expensive as well. Image processing techniques have started to attract the agriculture scientists due to their promising results and possibly moderate cost. However, they are still to be commercially available. For the large farms, tractor-mounted systems are more appropriate for detecting crop nutrient requirements. These are less labour extensive, and their compact size and low weight design allow to easy adaptation to pole-mounted. Since its development, 40 years ago, the RS has been successfully applied in a huge numbers of fields due to its efficiency and wide coverage but still have limitations like clear weather conditions are required for images collection, specialist software for data analysis and professional operators

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Table 1 Comparison between popular handheld instruments

Instrument	Wavelength	Measurement area	Reference
SPAD	650 nm in the red and 940 nm in the near-infrared	12.57 mm ²	(Netto et al., 2002)
N Tester	650 nm in the red and 960 nm in the near-infrared	2×3 mm ²	(Richardson et al., 2001, Goffart and Olivier, 2004)
CCM-200	660 nm in the red and 940 nm in the near-infrared	0.71 cm ²	(Richardson et al., 2001)
R meter (CM 1000)	ambient and reflected light at 700 and 840 nm	1.27 cm ²	(Murdock et al., 1998, Carter and Spiering, 1999)
Handheld 2 Portable Spectroradiometer	Spectral range (wavelength range) 325-1075 nm	Precise data over wide 25° field-of-view	(Edrees et al., 2013) http://www.asdi.com/products/fieldspec-spectroradiometers/handheld-2-portable-spectroradiometer

Table 2 Effect of different factors on LCC, SPAD and image processing-based nitrogen estimators

Factors	LCC	SPAD	Image processing technique	Reference
Applicability to wide range of species	More relevant for cereals such as rice, maize and sugarcane	Has been applied to a number of species, but did not produce accurate results for others. For example, not suitable for leaves of regenerated plants.	As this is a relatively new technique for N estimation, it has been applied to limited number of species. However, it has the potential to be applied to a wide range of species.	(Nagappa et al., 2002, Yadav et al., 2010)
Effect of environment on readings	As direct sunlight affects leaf colour readings, it is recommended to take the reading in the shade.	Environment has no or very little effect on readings.	Similar to LCC, sunlight has an effect on the colour of the acquired image.	(IRRI, 1996)
Accuracy	Accuracy is not guaranteed, as	Can be quite accurate. However, due to the	Has the potential to produce accurate reading	(IRRI, 1996,

	reading depend on visual assessment.	small measuring area of the device (around 12.57 mm ²), constancy in performance is not guaranteed	when images are collected in optimal environment/setting.	Netto et al., 2002)
Potential for improvement	Limited improvement only, through increasing the number of panels (green shades)	Improvement in the underlying technology. The latest version is SPAD 502	Has the potential for further improvement by enhancing the image processing technique.	(IRRI, 1996, Yang et al., 2003)
Calibration	Does not require calibration	Does not require calibration	Digital cameras may require calibration	
Price	USD \$1	USD \$1300-1400	Depends on the type of camera, but costs usually lower than SPAD	(Balasubramanian et al., 2000)

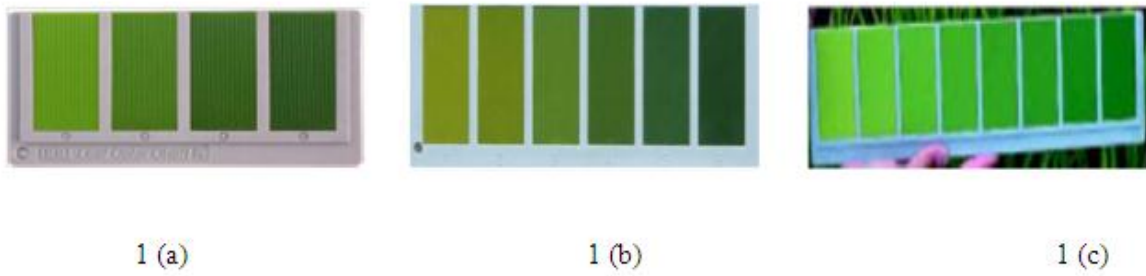


Figure 1 Various commonly used leaf colour chart (LCC)



Figure 2. Close-up of SPAD Meter

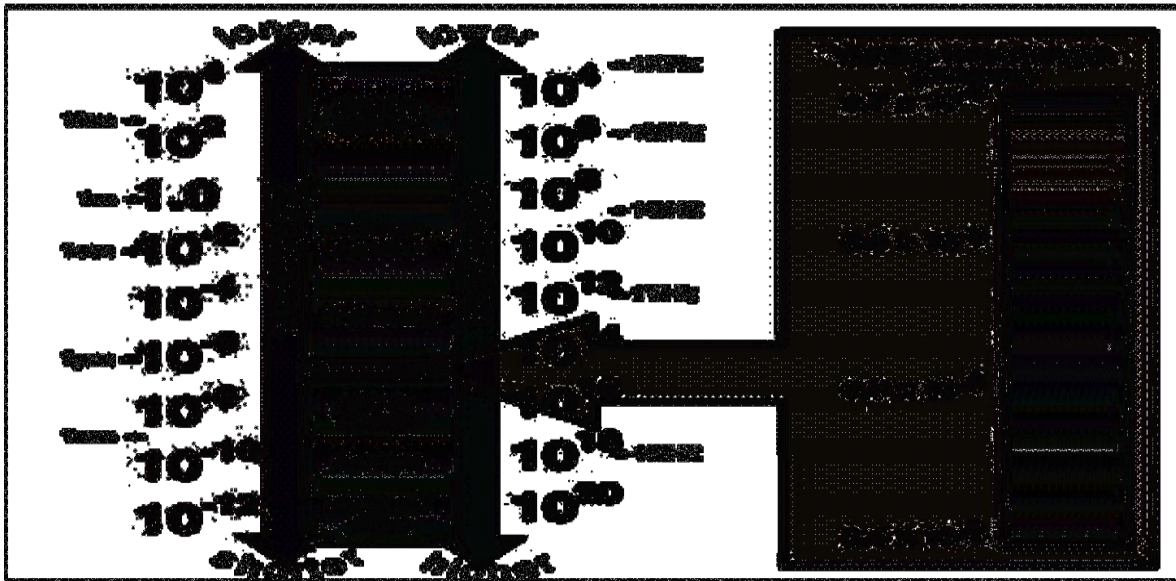


Figure 3: Electromagnetic spectrum (Casady and Palm, 2002)



Figure 4: Incident (I), reflected (R), absorbed (A) and transmitted (T) energy from a leaf surface

(Havránková, 2007)

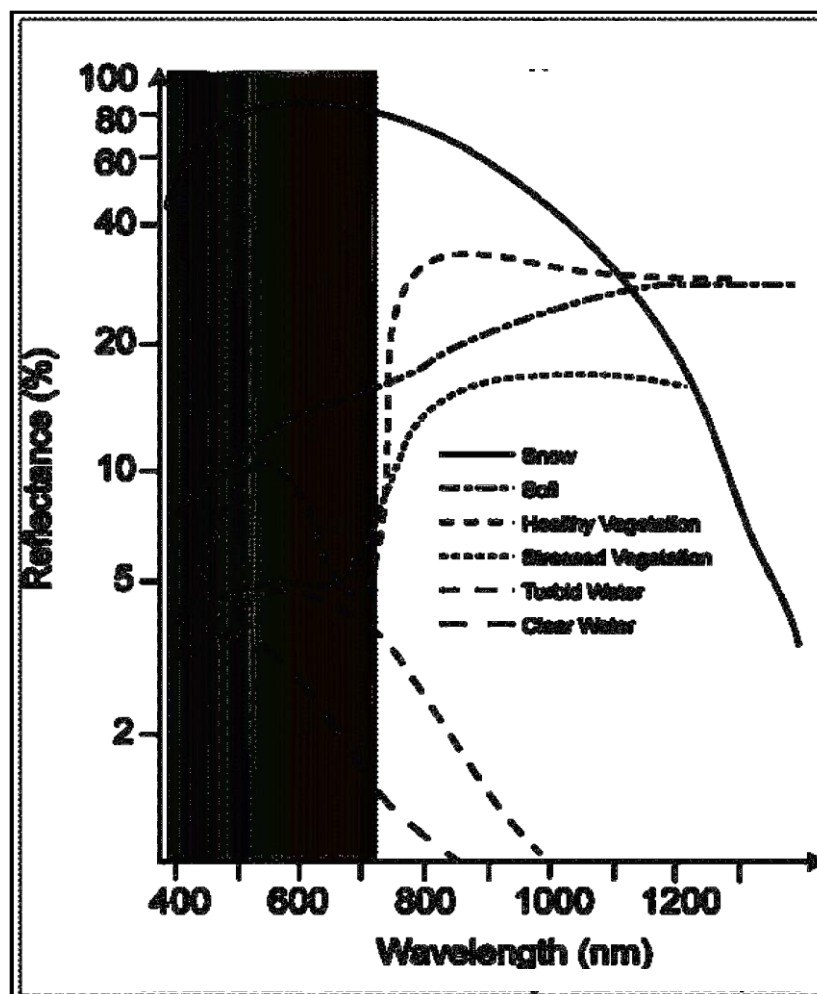


Figure 5: Light reflectance curves from different objects (Keiner and Cilman, 2007)

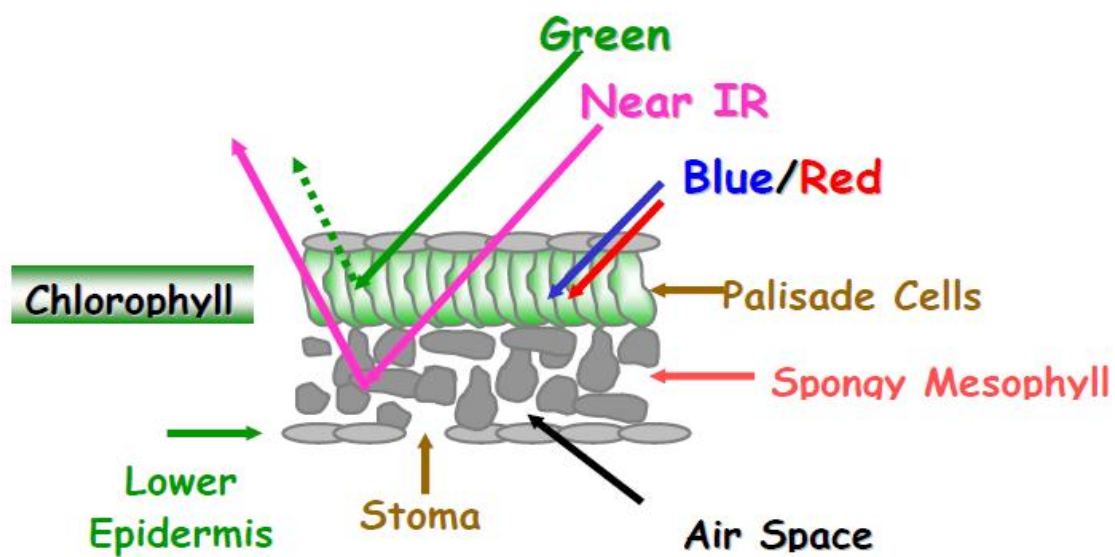


Figure 6: Leaf inner structure and light absorbance (Scheppers, 2005)



Figure 7: Hydro N-Tester http://www.arablefarmer.net/uploads/media/mineral_fertil.pdf

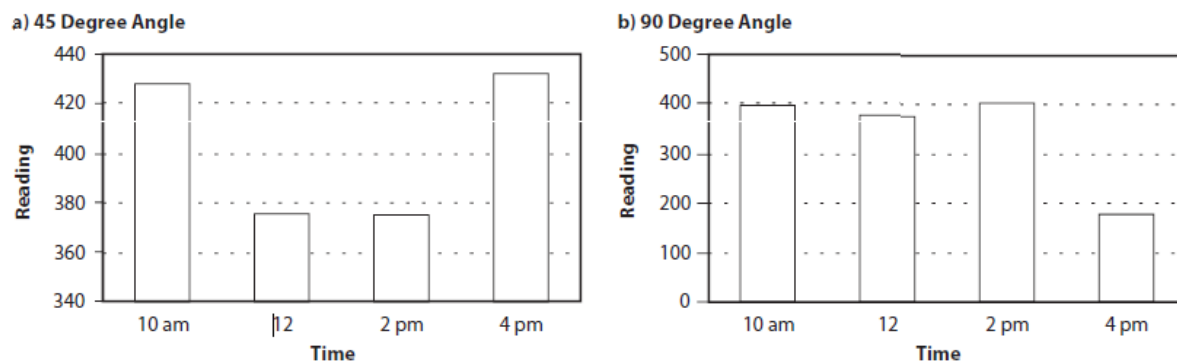


Figure 8: Effect of the time of day on CM 1000 readings at 45 and 90 degrees to the canopy surface

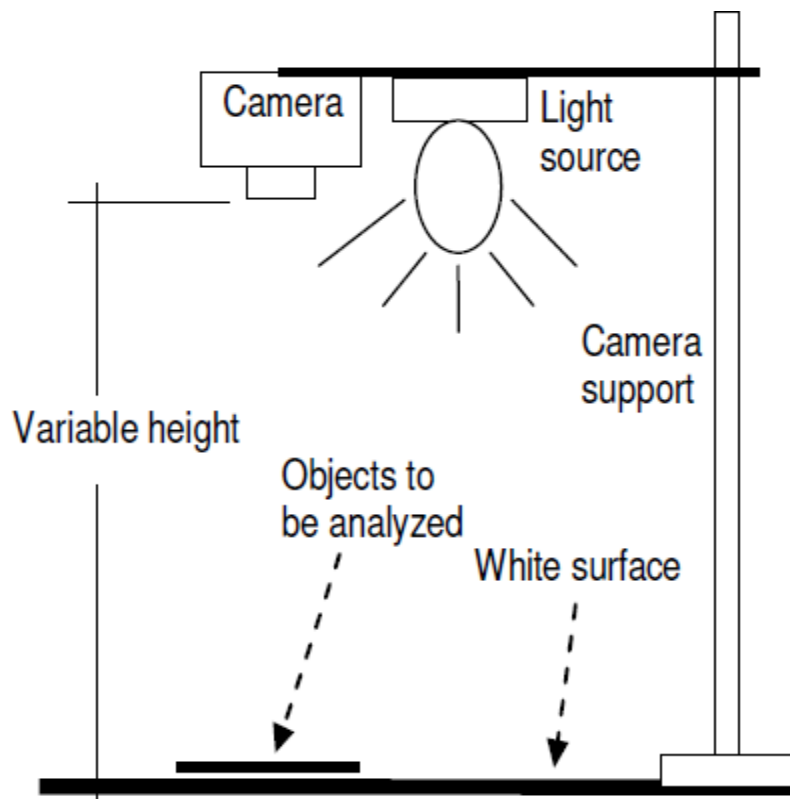


Figure 9: Camera set up for estimating leaf N status (Luna et al. 2010)

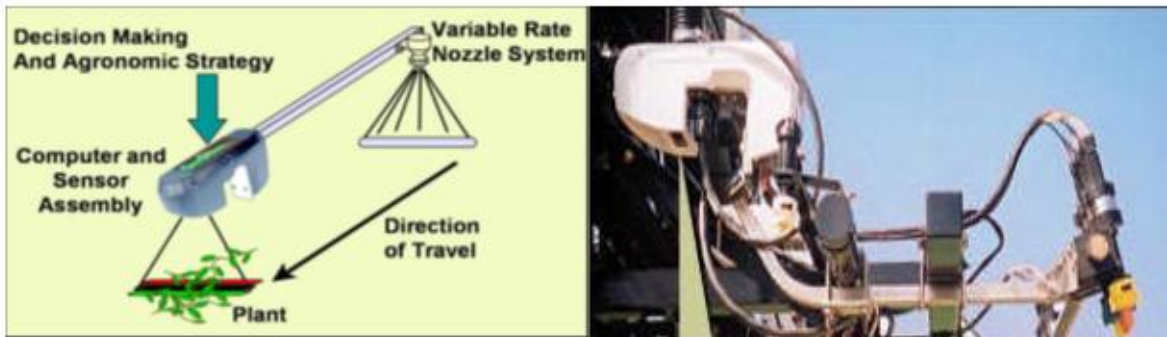


Figure 10: Fertilising systems using the GreenSeeker (Havránková, 2007).[®]

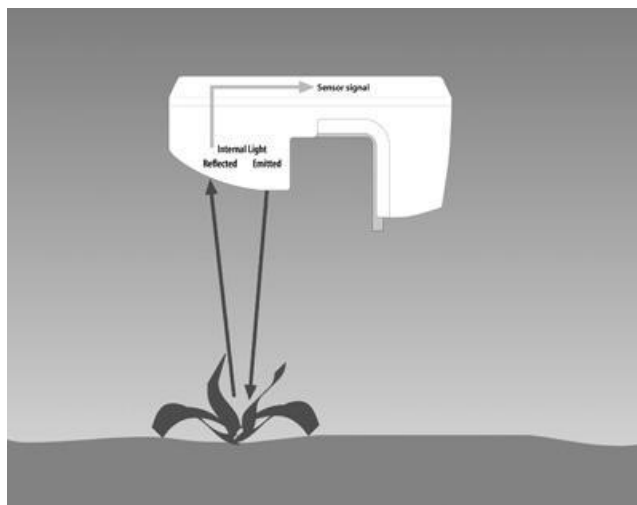


Figure 11: Illustration of GreenSeeker's light emission and reflectance measurement system

(Image was taken from <http://www.lesspub.com>)

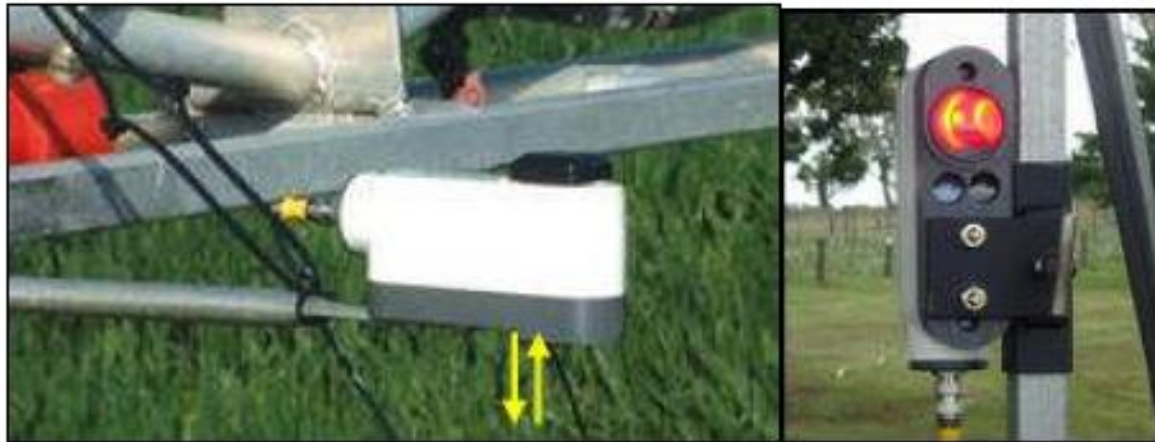


Figure 12 : Crop-Circle sensor (Havránková, 2007)



Figure 13: Yara N sensor (Image was taken from <http://www.agricon.com>)

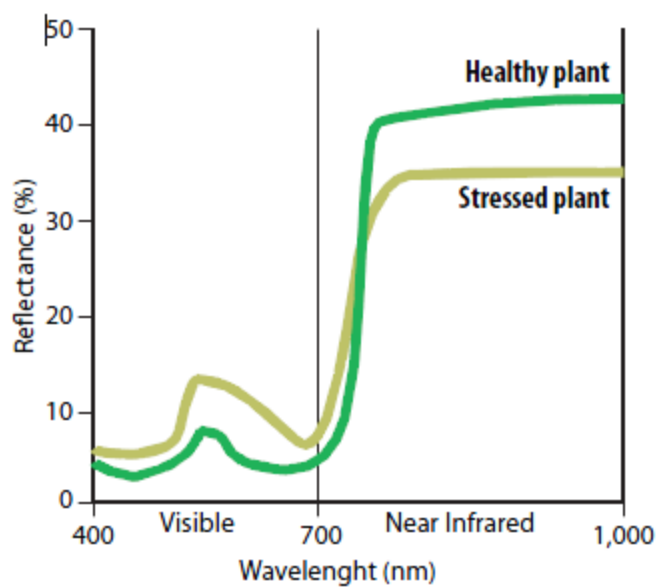


Figure 14: Typical reflectance spectrum of a healthy and a stressed plant (Govaerts and Nele, 2010)

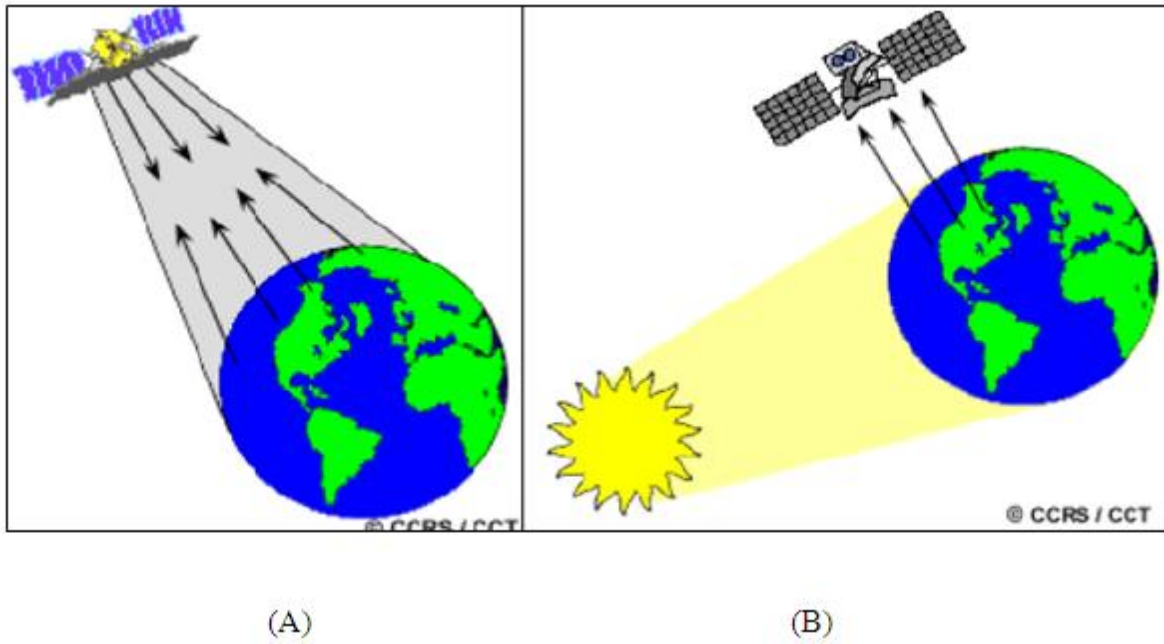


Figure 15: Active (A) and passive (B) remote sensing system (CCRS 2006)