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Near Infrared Hyperspectral Imaging Spectroscopy Applied to Investigation of Plant Leaves: A Brief Review

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Abstract: ***World agricultural production is the focus of worldwide attention because increasing productivity is imperative today and in the near future. Increasing productivity not only requires soil management, use of pesticides or biological agents to obtain best vintages, but also the improved seed and grains quality and other intrinsic factors. Many technical advances were implemented in recent years in agriculture. The plant leaves study can be provide important information about nutrition, soil condition and integrated pest management. Near infrared hyperspectral imaging spectroscopy (NIR-HIS) combines spectroscopy and imaging, providing information about the chemical properties of a material and their spatial distribution and is a relatively new technique. It represents an advance of traditional Near Infrared (NIR) spectroscopy. NRI-HSI is a highly flexible form of analysis that can be applied to a wide range of analytical applications. Long a basic technology in remote sensing, NIR imaging has become popular as an economical tool for chemical analysis. NIR-HIS is used to analyze multiple constituents in a single scan and to identify a non-destructive or non-invasive analysis method. This paper aims to present a brief review of the principal aspects of the applicability of NIR-HIS in agriculture related to investigation of plant leaves in the last two decades.***

Keywords: Hyperspectral chemical imaging, NIR spectral imaging, agriculture, plant leaves

1. Introduction

World agricultural production is the focus of worldwide attention because increasing productivity is imperative today and in the near future. Increasing productivity not only requires soil management, use of pesticides or biological agents to obtain best vintages, but also the improved seed and grains quality and other intrinsic factors. Many technical advances were implemented in recent years in agriculture and are denominated of precision agriculture.[1] The plant leaves study can be provide important information about nutrition, soil condition and integrated pest management. For this, *in vivo* studies of plant leaves are vital to obtain data that can be used to better productivity, including fertilization and adequate use of pesticide / herbicide and for this proposed the near infrared hyperspectral imaging spectroscopy (NIR-HIS) is a useful technique.[2]

Near infrared (NIR) is a type of vibrational spectroscopy that corresponds to the wavelength range of 750 to 2,500 nm (wavenumbers: 13,300 to 4,000 cm^{-1}). The analytical methods resulting from the use of the NIR spectroscopic region reflect its most significant characteristics, such as: fast (one minute or less per sample), non-destructive, non-invasive, with high penetration of the probing radiation beam, suitable for in-line use, nearly universal application (any molecule containing C-H, NH, S-H or O-H bonds), with minimum sample preparation demands. In NIR-HIS sample absorbance is recorded at each wavelength. NRI-HSI is a highly flexible form of analysis that can be applied to a wide range of analytical applications. Long a basic technology in remote sensing, NIR imaging has become popular as an economical tool for chemical analysis. NIR-HSI is used to analyze multiple constituents in a single scan and to identify a non-destructive or non-invasive analysis method. The principal advance in the technological applications of this technique was possible by computation development and the new discipline of Chemometrics.[3]–[5].

Chemometrics involves the application of statistical and mathematical methods, as well as those based on mathematical logic, to chemical analysis, providing the tools for gathering information and its wise use.[6] [7]

This paper aims to present a brief review of the principal aspects of the applicability of NIR-HIS in agriculture related to investigation of plant leaves in the last two decades and related to agricultural productivity. For clarity in our explanation, we have divided the review into four periods: 2000 – 2005, 2006 – 2010, 2011 – 2015, and 2016 – 2020. The articles are described in chronological order, as follows.

NIR-HIS in the investigation of plant leaves

Years 2000 – 2005

In situ Analyses of the correlation between hyperspectral reflectance and pigment content including chlorophyll-a, chlorophyll-b and carotenoid of leaves in different sites of rice were reported. The chlorophyll-a, chlorophyll-b and carotenoid contents in rice leaves in rice fields to which different levels of nitrogen were applied were measured. The chlorophyll-a content of upper leaves was well correlated with the spectral variables. The study showed that the most suitable estimated model of chlorophyll-a of upper leaves was obtained by using some hyperspectral variables.[8] Kobayashi et al. studied the ratio of rice reflectance for estimating leaf blast severity with a multispectral radiometer and observed that as disease severity increased, reflectance also increased in the 400 – 500, 570 – 700, and 900 – 2000 nm regions but decreased in the 500 – 570 and 700 – 900 nm regions. Rice reflectance ratios were evaluated as indicators of leaf blast severity. Two reflectance ratios, R550/R675 (550 nm divided by 675 nm), and R570/R675 quantified the significant disease severity. The authors concluded that the variation in ratios must be considered when they are used to estimate leaf blast severity.[9]

The kinetics of water desorption from olive leaves stored at different humid conditions was studied using a NIR multispectral imaging spectrometer. The kinetics of water desorption from olive leaves show that rate of water desorption is strongly dependent on the environment in which the leaves were stored. Water desorbed from leaves faster when leaves were stored under dry conditions. The rate for leaves stored in 0% humidity environment is 1.5x faster than those stored in 50% humidity.[10] Kakani et al. investigated the effects of UV-B radiation and atmospheric carbon dioxide concentrations ([CO_2]) on leaf senescence of cotton by hyperspectral reflectance. Plants were grown in controlled-environment growth chambers. Photosynthesis, chlorophyll, carotenoids and phenolic compounds along with leaf hyperspectral reflectance were measured on three leaves aged 12, 21 and 30 days in each of the treatments. The study revealed that cotton leaves senesced early on exposure to UV-B and leaf hyperspectral reflectance can be used to detect changes caused by UV-B and leaf ageing.[11]

Years 2005 – 2010

The diagnosis of rice growth and nutrient status is critical for prediction of rice yield and grain quality and prescription of nitrogen topdressing at panicle initiation stage. The results of the study showed that Partial Last Square Regression (PLS) model using hyperspectral canopy reflectance data to predict four plant variables (leaf area index, leaf dry weight, leaf nitrogen concentration, and leaf nitrogen density) produced an acceptable model precision and accuracy. The most important reflectance as judged by factor loading in PLS model for rice leaf characterization was at various bands as NIR and visible (355, 420, 524-534, 583 687 nm) and red edge (707 nm) region.[12] Botha et al. carried out one study using inverted PROSPECT radioactive transfer model to predict leaf chlorophyll and N (as protein) contents in potato leaves. The results showed that chlorophyll content was predicted with reasonable accuracy. However, protein content could not be predicted with any degree of accuracy by the model because of the small percentage of protein in the total leaf biomass and the masking effect of water absorption.[13] Li et al. used an ASD spectrometer to derive the hyperspectral data of cotton leaves in canopy in north Xinjiang. The integral area variables of red edge were used to estimate the total nitrogen (TN). The results showed that there is a significant positive correlation between the chlorophyll content and the TN content and the data of chlorophyll content can be used to estimate the TN content in single cotton leaves. The authors considered that there is an applying potential to use the integral area variables of red edge for estimating the TN content in cotton leaves in canopy.[14] In order to compare the predictive ability of propagation neural network (BPN), the artificial neuron network (ANN), to that of the multivariate linear regression models (MLR) models in estimating the content of pigments in rice leaves and panicles a study was performed. Results showed that all BPN models gave higher coefficients of determination (R2), lower absolute errors, and root mean squared errors than the corresponding MLR models, in both calibration and validation tests. Further significance tests by paired t tests and bootstrapping algorithms indicated that most of the BPN models outperformed the MLR models.[15] In the same year, Zhang et al. [16] using hyperspectral instrument, and the inverted PROSPECT radioactive transfer model, quantified the leaf chlorophyll content and optical properties of 255 overstory and understory leaf samples through the growing season mature sugar maple (*Acer Saccharium*) stand. The method used was capable of deriving the seasonal and canopy-height-related in leaf chlorophyll content from leaf reflectance and transmittance spectra.

A nitrogen sensor was developed to predict nitrogen concentrations in orange leaves. Based on chlorophyll and proteins spectral absorption bands, the sensor´s wavelength ranges were chosen to be 620 – 950 nm and 1400 – 2500 nm. Test results showed that the nitrogen sensor had good linearity and stability. The system was able to predict nitrogen content with a good mean square difference. Using the nitrogen sensor developed unknown leaf samples could be classified into low, medium, and high nitrogen levels with good accuracy.[17] Yang analyzed hyperspectral canopy reflectance spectra of two rice cultivars with different susceptibilities to bacterial leaf blight (BLB) to establish spectral models for assessing disease severity for future management. The results indicated that wavebands from 757 to 1039 nm were the most sensitive region of the spectrum. The author found good relations between the severity disease and spectral reflectance.[18] Wu et al. selected wavelengths at the red edge of the vegetation spectrum (705 and 750 nm) to test vegetation indices using space borne hyperspectral Hyperion data for the estimation of chlorophyll content and leaf area index in different canopy structures. The results demonstrated possibilities for analyzing the variation in chlorophyll content and leaf area index using hyperspectral Hyperion data with bands from the red edge of the vegetation spectrum.[19] Grishman et al. employed the hyperspectral remote sensing to detecting *sugarcane yellow leaf virus* (SCYLV) infection in asymptomatic leaves. Infection was determined by reverse transcriptase-polymerase chain reaction analysis. The results showed that leaf reflectance was effective at predicting SCYLV infection in more of 70 % of cases and demonstrated that the hyperspectral remote sensing is a good method to detection of SCYLV.[20] Zhou et al. compared the applicability of the single leaf and the leaf positional difference in the vegetation index between in detecting nitrogen (N)-overfertilized rice plants. The hyperspectral reflectance (350 – 2500 nm) and the chlorophyll concentration were measured at different stages. The results of the study showed that the N-overfertilized rice plants can be effectively detected with the leaf positional difference.[21]

The estimation of nitrogen (N) and silicon (Si) is very important because these nutrients are one of the factors that influence the prevalence of the *Eldana saccharina* Walker borer (Lepidoptera: Pyralidae) in sugar cane agriculture. The hyperspectral remote sensing was used to estimate the concentration of N and Si in the leaves and their proportion in sugar cane. The conclusion was that hyperspectral remote sensing has the potential to be used in estimating the N: Si ratio and that potential infestations by *E. saccharina*.[22]

Yao et al. presented a new method to extract hyperspectral spectrum information to developed regression models for estimating leaf N accumulation per unit soil area (LNA) in winter wheat. The hyperspectral range 350 – 2500 nm was investigated and normalized different spectral indices, ratio spectral indices and their relationships with LNA were quantified. It was concluded that the hyperspectral parameters could be used for estimation LNA in winter wheat.[23] Xiaobo et al. presented the identification of the desirable information using independent component analysis for hyperspectral imaging at visible and short NIR region to estimate the pigment content in cucumber leaves. When the calibration models was applied to an independent validation set, chlorophyll content was reasonably well predict with high correlation (r^2 = 0.774). The results obtained showed that the technique could be used with good accuracy to analysis *in situ* of living plant.[24] Pacumbaba et al. performed an experiment to study if hyperspectral reflectance could be used to detect nutrient stress in *Lactuca sativa* L. cv. Black Seeded Simpson. Nutrient stress containing nitrogen, phosphorous, potassium, cadmium, and magnesium was applied. Spectral reflectance varied with some factors and spectral responses of lettuce leaves under macronutrient deficient condition showed an increase reflectance in red, near red, and infrared wavelengths ranges. The study concluded that the spectral reflectance could be promissory in predicting deficiencies in general.[25]

Years 2011 - 2015

Other authors also investigated the leaf nitrogen content (LNC) in wheat by using hyperspectral imaging. The conclusion of the work was that nitrogen concentration is accessible from reflectance spectra in the range of 400 – 1000 nm for both dried leaves and fresh samples; the spectral results in natural conditions are similar those laboratory and, finally, there are good reasons to believe the method is effective for non-destructive nitrogen content determination.[26] Mukoyama et al. used of a trial observation equipment using hyperspectral sensor mounted on a radio-controlled helicopter to determinate chlorophyll meter in rice cultivation. Therefore, it is an *in situ* and *in vivo* measure. It was possible conclude that the path to practical application of the technology can be considered.[27] Bauer et al. described three methods of automatic classification of leaf diseases based on high-resolution multispectral stereo images. It was took stereo images of single sugar beet leaves with two cameras in a laboratory under well-controlled illumination conditions. The leaves were either healthy or infected with the leaf spot pathogen *Cercospora beticola* or the rust fungus *Uromyces betae*. The medians of pixelwise classification rates achieved in the experiments were 91% for *Cercospora beticola* and 86% for *Uromyces betae*.[28] Hardin et al. used a SPAD meter (Konica Minolta, Osaka, Japan – that is a handheld device that measures light intensity at wavelengths of 650 and 940 nm transmitted through a 2 x 3-mm area of a leaf clamped in the instrument) to determine pecan foliar N *in situ*. The results indicated that the method have potential to provide useful indications of pecan leaf N concentration.[29] Huang et al. suggested that the hyperspectral reflectance has potential to detect rice leaf folder damage severity in rice, because the results encountered showed the reflectance from rice leaves significantly decreased in the green (530 – 570 nm) and NIR (700 – 1000 nm) regions and increase in the blue (450 – 520 nm) and red (580 – 700 nm) regions as the leaf-roll rate of rice increase.[30]

LNC is, also, an important indicator of tobacco quality and is used in prediction of tobacco field. The study identified the hyperspectral reflectance central band that sensitive to tobacco LNC. The optimum band combinations for ratio vegetation index and normalized different vegetation index were (590 and 1980 nm) and (1970 and 650 nm) respectively. The results indicated that the method, with stepwise multiple linear regression and back-propagation neural network models, is enable of monitoring LNC.[31] Zhang et al. investigated of potential of (Vis-NIR) hyperspectral imaging system for determination and estimation of nitrogen, phosphorus, and potassium in oilseed rape leaves. Hyperspectral images were acquired between 380 – 1030 nm and partial least square regression and least-square support vector machines were use. The results revealed the procedure is useful to detect macronutrients non-destructively and could be used to *in situ* determination in living plants.[32] Some of these authors (Zhang and He), and also work with oilseed rape leaves, presented a new partial least square regression (PLS) using relevant wavelengths (543, 686, 718, 741, 824 and 924 nm) in the visible and near infrared region for predicting seed weights of individual plants. Th results demonstrated that hyperspectral imaging system is promising to predict the seed yield in oilseed rape based on its leaves in early growing stage.[33] Zarco-Tejada et al. presented method for leaf carotenoid content estimation in vineyards using high-resolution hyperspectral imagery acquired from an unmanned aerial vehicle. The good results demonstrated the feasibility of the method of mapping leaf carotenoid concentration at the pure-vine level from high-resolution hyperspectral imagery.[34]

It is important knowledge of vegetation water condition because can contribute to drought assessment. In the study, all two-band combinations (350 − 2500 nm) in the ratio type of vegetation index and normalized difference type of vegetation index were performed on cotton leaf raw spectra reflectance and first derivate reflectance. Bands with center in near infrared region from 950 – 1100 nm, and 1650 – 1750 nm were represented almost all selected bands. Despite the good results, the authors claim that further studies are needed to test the selected bands. [35] Singh et al. employed chemometric models and hyperspectral features to measured canopy reflectance and leaf pigments (chlorophyll and carotenoids pigments) in soybean. The analytical and transformations methods employed could be useful to develop models or extract features from spectral reflectance spectra relative to other leaf concentration or canopy characteristics.[36] Ordónez et al. applied two functional prediction models (functional linear regression and functional nonparametric methods), to the prediction of the chemical characteristics (moisture, dry mass, and concentrations of nitrogen, phosphorus, potassium, calcium, iron, and magnesium) of vine leaves, using electromagnetic reflectance between 350 and 2500 nm as the input. The results obtained showed different levels of correlation between reflectance and the predicted data and non-parametric methods presented the best results.[37]

LNC also is a key indicator of nitrogen status and can be used to evaluate nitrogen nutrient levels and improve fertilizer regulation in fields. The authors proposed a new method with hyperspectral measurements to estimate LNC in barley by introducing optimal combination (OC) principle. The results indicated that hyperspectral measurements displays good potential to effectively estimate LNC in barley, and the OC method presented adaptability and accuracy due to the optimal selection of spectral parameters responding LNC.[38] Kong et al. examined the possibility of fast detection of peroxidase activity in tomato leaves infected with *Botrytis cenerea* using hyperspectral imaging data. Genetic algorithm-partial least square was applied to select optimal wavelengths (576, 578, 579, 600, 601, 603, 618, 619, 620, 621, 741, 742, 746, 747, 748, 752, 753, 777, 893, 895, 896 nm). It was developed a new fast neural algorithm named extreme learning machine (ELM) and employed as multivariate analytical tool. The results showed that the method is useful to detection of *Botrytis cenerea* in tomato leaves and the selected wavelengths could be potential resources for instrument development.[39] Zhao et al. assessed the effect of severity of stripe rust (*Puccinia striiformis*) on the hyperspectral reflectance of wheat by analysis of samples with the disease. The spectra of the adaxial and abaxial surfaces of the leaf samples were taken using an ASD Leaf Clip. The results indicated that the pathogen caused changes in foliar water and chlorophyll, and those changes made it feasible to assess disease severity using *in situ* hyperspectral measurements.[40]

The detection of early blight and late blight diseases on tomato leaves was studied. Healthy leaves, early blight and late blight diseased leaves were selected to obtain hyperspectral images between 380 to 1023 nm. An extreme learning machine (ELM) classifier model was established based on full wavelengths. Successive projections algorithm (SPA) was used to identify the most important wavelengths (442, 508, 573, 696 and 715 nm). The results demonstrated that hyperspectral imaging has the potential as a non-invasive method to identify early blight and late blight diseases on tomato leaves.[41] Guo et al. investigated the hyperspectral reflectance images of healthy and diseased leaves infected with *Citrus tristeza virus* (CVT). The spectra were transformed with 15-point Savitzky Golay second derivative. Then principal component analysis was performed on the transformed data in order to reduce the dimension of data. The optimal wavelengths: (405, 424, 920, 947, 957, 972, 978, 980, and 998 nm) selected by stepwise regression showed great potential in CTV diagnosis. Despite of good results, further study is required to under field conditions.[42] Bergsträsser et al. introduced a tailor-made hyperspectral absorption-reflectance-transmittance imaging (HyperArt) system that uses the reflectance and transmittance images to calculate absorption, to assessing the *Cercospora beticola* in sugar beet leaves. The results showed that the chlorophyll content could be accurately estimated and monitored both in different fertilization regimes during growth and fungal symptoms.[43]

Years 2016 – 2020

A new nitrogen indicator based on direct link between spectral index and chlorophyll content in winter wheat through quantitative relationships between LNC and ground-based multi-angular remote sensing hyperspectral reflectance in field experiments was developed. The results suggested that the model is useful to LNC determination in wheat and also indicated the importance of taking into account angle effects when analyzing optimum vegetation indices.[44] Kong et al. investigated the feasibility of hyperspectral imaging with 400 – 1000 nm to detect malondialdehyde (MDA) content in oilseed rape leaves under herbicide stress. MDA distribution map was successfully achieved by partial least squares (PLS) model with competitive adaptive reweighted sampling (CARS). The study indicated that hyperspectral imaging technology provided a fast and non-destructive solution for MDA content detection in plant leaves.[45] Wu et al. [46], like Kong et al. [39] previously mentioned, established a genetic algorithm (GA) – partial least squares (PLS) correction model as a module conducted band selection for the application of hyperspectral imaging to non-destructive testing of corn seedling leaves. Twelve bands selected by statistic test in genetic algorithm were 484, 485, 499, 500, 501, 533, 705, 1051, 1052, 1056, 1057, and 1058 nm). The results showed that GA-PLS model is better than PLS model established in full spectrum and experience-based selected bands.

The micro structural changes of mulberry was studied by scanning electron microscope and, then, NIR-HIS was used to predict the distribution of pesticide residues in mulberry leaves with the aid of gas chromatography. SPA, stepwise regression, and regression coefficients were utilized to extract the characteristic wavelengths. Maximum residue limit was applied to build prediction models based on spectral feature in characteristic wavelengths. The study demonstrated that pesticide residues distribution map, obtained by NIR-HIS, clearly showed the distribution of pesticide residues in mulberry leaves.[47] Liu et al. (2017) carried out a study, in which they used multifactorial statistical regression methods and neural networks of Back Propagation (BP), of hyperspectral data based on unmanned aerial vehicle (UAV) and the corresponding LNC data on winter wheat in several growth stages. The results showed that the spectrum sampling of UAV-based hyperspectral data is reliable in the determination LNC.[48] Mo et al. developed multispectral imaging algorithms using visible-near infrared (VNIR) and NIR-HSI techniques to detect worms in fresh-cut lettuce. The optimal wavebands that can detect worms in fresh-cut lettuce were investigated for each type of HSI using one-way ANOVA. The results demonstrated that the technique have the potential to detect worms in fresh-cut lettuce.[49] Yang et al. studied NIR-HSI applied to investigation of nitrate content in spinach (*Spinacia oleracea L.*). PLS analysis revealed that there was a high correlation between the reference data and NIR spectra. The results showed that the nitrate content in spinach leaves was successfully mapped at a high spatial resolution, clearly displaying its distribution in the petiole, vein, and blade.[50] Xie et al. in another study used the hyperspectral technique to classify healthy and gray mold diseased tomato leaves. Hyperspectral images of diseased samples were collected at various times after inoculation and healthy samples were taken in the wave range of 380 – 1023 nm. Principal component analysis (PCA) was firstly used to compress spectral reflectance data, and then it used to again to compress hyperspectral images. The good results allowed concluding that hyperspectral images have potential for early detection of gray mold disease on tomato leaves.[51] Lu et al. also working with tomatoes, investigated the possibility of discriminating tomato yellow leaf curl disease by a hyperspectral imaging technique. A hyperspecral imaging system collected hyperspectral images of both healthy and infected tomato leaves. The reflectance spectra, first derivative reflectance spectra, and absolute reflectance difference spectra in the wavelength range of 500 – 1000 nm of both background and the leaf area were analyzed to select sensitive wavelengths and band ratios. It was demonstrated that multispectral images at 560, 575 and 720 nm have a potential for detecting tomato yellow leaf curl virus infection in field applications.[52] Sytar et al. investigated way of use methodology of non-destructive detection with hyperspectral reflectance imaging together with wet chemistry quantitative analysis based on correlation analysis of receiving data in experimental buckwheat cultivars. It was found that high total polyphenols content is related with high values of hyperspectral indices, which characterize chlorophyll concentration, and parameters of vegetation.[53]

A comparative assessment of different modeling algorithms (simple and non-parametric modeling algorithms alongside the physical model retrieval method) for winter wheat LNC estimation using UAV was proposed. A five-band multispectral camera (490 nm, 550 nm, 671 nm, 700 nm, and 800 nm) was mounted on a UAV to acquire canopy images across five critical growth stages. Good results were obtained to performing vegetation index.[54] Also working with UAV and hyperspectral analysis and spectral indices applied to LNC estimation for winter wheat, Zhu et al. encountered that several LNC spectral indices are effective in quantitatively inversing the LNC of winter wheat.[55] Liu et al. aiming at detecting common invertebrate pests on corps in natural farming fields, developed a multispectral 3D machine vision systems (MVS). The proposed method could be used as a sensor for pest mapping or on-the-go pesticide spraying.[56] Wang et al. predicted chlorophyll a, chlorophyll b, total chlorophyll, and carotenoid content in tea leaves under different levels of nitrogen treatment using HSI in combination with variable selection algorithms. The authors concluded that the method is rapid and accurate for predicting the content of pigments in tea plants.[57] Huang et al. developed a hyperspectral imaging system in visible and NIR region for the rapid identification of *Diaphania pyloalis* larvae and its damage in mulberry trees. The extracted spectra of five region of interests, leaf vein, healthy mesophyll, slight damage, serious damage, and *Diaphania pyloalis* larva at 400 – 1000 nm (visible range) and 900 – 1700 nm (NIR range), were used to establish a partial least squares discriminant analysis (PLS-DA) and least-squares support vector machines models (LS-SVM). The results showed that the visible and near infrared hyperspectral imaging could distinguish *Diaphania pyloalis* larvae and their damage from leaf vein and healthy mesophyll in a rapid and non-destructive way.[58]

Accurate monitoring of the LNC in maize can provide a fundamental basis for effective N management. The authors realized a study to verify the predictive ability of the published vegetation indices (VIs), the PLS regression and the two-band optimal combinations algorithms, and to determine the most accurate method for assessing the LNC of unsynchronized growth stages in maize. It was concluded that the best 2-band VIs (680 – 760 nm) and the PLS regression based on selected first-derivative reflectance wavelengths provide a useful explorative tool for estimating LNC of maize across years, ecological areas, and unsynchronized growth stages.[59] Zhou and Jiang proposed an inversion method of maize leaf area index based on UAV hyperspectral remote sensing. The improved Snake model was used to achieve coarse convergence of target image contour after denoising. Through particle swarm optimization iterative algorithm, the optimal image segmentation point was found and the image segmentation was achieved. Based on the results the expressions of modified chlorophyll absorption ratio, normalized difference spectral index, and ratio-type spectral index were obtained. Experiments showed that the proposed method has a comprehensive performance, and has a strong advantage over the current method.[60] Ji'An et al. investigate the utility of hyperspectral images for the detection of oilseed rape water logging stress. Six optimal wavebands of 529, 641, 698, 749, 856, and 979 nm were used as input for SPA classification and analysis. The results showed that hyperspectral imaging technology is feasible and useful for the detection of oilseed rape waterlogging stress.[61] Zhou et al. applied hyperspectral imaging to early detection of *magnaporthe oryzae*-infected barley leaves. The results indicated that linear discriminant analysis (LDA) coupled with competitive adaptive reweighted sampling (CARS) achieved good results. The method could be used both early infection period identification and lesion visualization.[62] Dan-dan et al. modeled the total leaf N concentration (TLNC) in winter wheat constructed from hyperspectral data by considering the vertical N distribution (VND). The results showed that the LNC model with VND provided an accurate and non-destructive method to monitor N levels in the field.[63] Li and Hu developed kinetic models to predict peroxidase (POD) activity in potato leaves. Infection-induced changes in potato leaves stored in an artificial climate chest at 25 °C were analyzed using hyperspectral imaging. Four prediction models were developed by using PLS and nonlinear support vector machine (SVM) methods based on the full spectrum and effective wavelengths. The effective wavelengths (540, 680, 863, 897, and 941 nm) were selected by the successive projection algorithm (SPA). In the study, the prediction model developed by means of SPA-SVM method obtained the best performance.[64]

Recently, attention has also been given to the quantification of inorganic constituents in the leaves by NIR-HSI. Lu et al. studied the potassium levels and plant varieties between 340 groups of leaf samples with different K contents. The samples were used to examine the relationship between reflectance spectra (350 – 2500 nm) and leaf K content (LKC). The correlation between LKC and with random bands from 350 to 2500 nm were determined for the published K vegetation indices in rice. Results showed that the spectral reflectance, R, of the shortwave infrared (1300–2000 nm) region was sensitive to the K levels and significantly correlated with rice LKC.[65] Sun et al. conducted a study to estimate cadmium content in lettuce leaves using hyperspectral imaging in the range of 431 – 961 nm. A deep brief network optimized by particle swarm optimization was proposed to predict cadmium content in lettuce leaves. Traditional models such as "support vector regression based on successive projections algorithm" were also established as comparison. The results showed that is possible to realize rapid and non-destructive detection of cadmium content in lettuce leaves.[66] Some of the authors of this last described article, Zhou et al., studied the validity and reliability of Vis–NIR hyperspectral imaging for the determination of lead concentration in lettuce leaves. A method involving wavelet transform and stacked auto-encoders (WT-SAE) was proposed to decompose the spectral data in the multi-scale transform and obtain the deep spectral features. The best prediction performances for detecting lead (Pb) concentration in lettuce leaves was obtained from raw data set The results of the study indicated that WT-SAE can effectively select the optimal deep spectral features and Vis–NIR hyperspectral imaging has great potential for detecting lead content in lettuce leaves.[67]

2. Conclusion

Agricultural productivity is directly related to two main factors: fertilization and occurrence of pests. For the first, the quantification of nitrogen is of fundamental importance and, for the second, the detection of microorganisms that cause disease. Fast, non-destructive and *in situ* methods are therefore essential. As seen in the articles described here, NIR-HSI, and chemometrics, applied to leaf analysis has been showing effectiveness for this purpose. In addition, contaminants, such as heavy metals, and other inorganic constituents, can be detected in plant leaves. Both the nitrogen content and the content of other elements provide soil conditions. Thus, a punctual fertilization can be carried out, minimizing costs and maximizing agricultural production. The detection of microorganisms in plant leaves through the NIR-HSI helps in the selected application of pesticides / herbicides, resulting in a gain for producers.

The future of the NIR-HSI technique applied to leaf research is promising and we believe that it, coupled with unmanned aerial vehicles or harvester machines, will open an unprecedented path in agriculture worldwide.

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