Diagnostic Nutrient Testing

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Abstract
Recommendations made for nutrient applications have been traditionally focused on economic yield and quality. Current day testing procedures and recommendations are however required to simultaneously ensure both economical and environmental sustainability of all agricultural production systems. Soil test is a calibrated index to determine crop response to applied nutrients. Any application rate devoid of an economical response in yield or quality is deemed unnecessary. Therefore a soil test becomes the first step in any nutrient BMP development, implementation and monitoring activity. Certain significant areas in Florida such as calcareous soils require development of calibrated soil tests rather urgently. Nutrient sufficiency of perennial crops and deficiency diagnostics can be gauged through in-season plant tissue testing. However nutrient delivery through foliar sprays is not always effective and may require multiple applications. Spectral reflectance methods show significant promise as an alternative to traditional wet chemistry analyses with regard to ease, costs, and speed with wider range of applications including natural resources. Additional research is however needed to develop this technology for field-scale applications. Current research is focusing on environmental nutrient management to include nutrient sources, application rates and timing, nutrient uptake efficiency, retention capacity of soils, estimating and minimizing nutrient losses to the environment, etc. Nutrient loss assessments tools such as the Florida Phosphorus Index and Bahia and Citrus tests for Phosphorus are now being made possible in Florida through integration of soil and tissue testing methods. Development and improvements of such analytical methods and tools specific to Florida to include other nutrients, heavy metals, soil capacity, eco-sensitive regions, is vital to ensure sustainability to state’s tourism, agriculture, and urban-rural balance.

Introduction
High quality diagnostic soil, plant and water testing programs are required to achieve optimum agricultural production and to minimize negative environmental impacts. While soil fertility assessments and field calibrated nutrient recommendations have been tremendously successful during the past half-century resulting in self-reliance in food production and food security, the current day diagnostic testing programs are becoming extremely sophisticated to simultaneously ensure environmental sustainability of nutrient cycling through rural and urban agricultural sectors and natural ecosystems.

The primary objective of traditional soil and plant testing programs is to make estimate the nutrient supplying capacities of various soil types and fertilizer management is to formulate such protocols so as to maintain long-term agricultural production efficiency through the sustainable and rational utilization of soil resources. Soils vary considerably from year to year in their capacity to meet nutritional needs of plants. A systematic approach involves determination and elimination of soil nutrient constraints for balanced
supply of all deficient essential nutrients for sustainable economic yields. In addition, soil tests are used to monitor the impact of past fertility practices on changes in the soil nutrient status. The recommendations are made based on the results from analytical tests done on soil, plant tissue and sap and subsequent crop response. The main disadvantage of a fertilizer management program is that it is very crop specific and hence cannot be extrapolation to other situations for improvement of micronutrient recommendations. This major drawback had led to the use of remote sensing techniques which estimate micronutrient availability on a fundamental basis so that fertilizer management recommendations can be used on different soils types in different regions.

**Soil Testing**
Soil testing is a multistep process starting with collection of a sample. The sample has to be a true representative of the area and the test results and recommendation will be only as good as the sample. The need for an appropriate sample can never be overemphasized. Then at the lab the first step is extraction or digestion of the sample. The method employed is specific to the nutrients, geographic region as well as physiographic and mineralogical nature of the sampling site. Therefore multiple methods exist. In Florida, we have four methods for three major groups of soils. Interpretation of the test results for the intended application is the second and most important step in the entire process. State laboratories backed by nutrient management and environmental quality research information make the process credible. In the state of Florida, over 80 years worth of research on soil fertility has made the soil testing very credible (SERIA-IEG-6, 2007).

Once the test results are interpreted, nutrient recommendations are formulated based on the field-calibrated crop response. A separate soil pH and lime requirement determination is also made to ensure effective soil pH management as per the target pH specific to the crop being grown. In addition to the soil test results and recommendations, several footnotes are also included in the reports, providing tips on the split doses, rates, timing, irrigation, other management practices and those footnotes form an integral part of the recommendations.

While extent of adoption of soil test recommendations is probably governed by socioeconomic factors, there are some limitations to the traditional diagnostic soil testing process. Soil testing is time consuming and in certain cases, expensive. A soil test cannot prevent poor crop growth due to drought, disease, insects, too much water, pesticides residues, etc. Additionally in the absence of regular field calibrations of recommendations for new varieties, new crop species, new nutrient sources, micronutrients, altered management practices, new analytical methods and reagents etc., the viability of soil testing will be limited. Lack of a reliable soil test for estimating nitrogen availability is a serious disadvantage, given the fact that the plant uptake efficiency of applied fertilizer N is only 30-50% (Raun and Johnson, 1999). Sandy soils in Florida combined with high temperature and rainfall increase the potential for N loss significantly. Integration of nutrient and irrigation management recommendations is the critical need of the hour, particularly for our soils and intensive production practices.
Plant Tissue Testing

Plant analysis is a proven and effective means of predicting fertilizer needs for many crops, especially perennial crops, although it does not however completely replace a soil test. Soil and plant diagnostics are complementary and serve different purposes. Plant tissue tests help monitor the nutrient status of plants throughout the growing season to determine whether each nutrient is present in sufficient concentration for optimum growth characteristics. The tissue tests confirm suspected nutrient deficiency symptoms, reveal hidden hunger and verify toxicities. Plants may not show any visible symptoms, but the nutrient content maybe insufficient enough to reduce the yields. In-season corrections to nutrient deficiencies can be made through foliar applications of specific deficient nutrients. Tissue tests also indirectly aid in evaluating the efficiency of applied fertilizers.

Why test both soil and plant tissue testing?

Soil testing indirectly estimates the amount of nutrients present and in turn the portions of those soil nutrients that will be available to the crop during the growing period. Leaf diagnosis shows the nutrient status of the plant at the particular time of sampling. However, leaf diagnosis alone may not give an accurate picture of nutrient requirements. Soil nutrient levels are only one of many factors which determine the nutritional status of a plant. Other factors such as temperature, water availability, management factors also influence the soil fertility status, nutrient balance, etc. Due to such complexity, leaf analysis has to be integrated with soil analysis. In Florida, two such new tests have been developed and implemented- for Bahia pastures (Mylavarapu et al., 2007) and for commercial Citrus. In both tests, phosphorus requirement is determined by a combination of both soil and a plant tissue test and deficiency confirmation.

Collection of foliar samples for nutrient analyses is critical to success of nutrient management program. Each sample should be collected so leaflets come from representative trees over the entire sample area (SERA-IEG-6, 2000). Sampling should be done from the proper plant part at the proper growth stage. For young seedlings, the entire plant is sampled 2.5 cm above the soil level. For larger plants, the most recent fully developed and mature leaf is the best indicator of nutritional status. For certain crops, including corn, the best indicator of nutritional status is the leaf adjacent to the uppermost fruit (ear leaf), when they approach flowering stage. When unfamiliar with sampling protocol for a specific crop, it is generally acceptable to select the most recent mature leaf as the best indicator of nutritional status. Insect and disease damaged leaflets, and leaflets showing severe deficiency symptoms, should be avoided or should be sampled separately (Herrera, 2005)

Plant Sap Testing

Analysis of plant sap measures mobile nutrients in the plant system, thus providing a different view of plant nutrition than the traditional dry tissue analysis. Sap testing is
more sensitive and therefore can detect minor or temporary deficiencies more easily. An accurate diagnosis of plant stress is possible and thus effective management decisions to correct nutrient imbalances or deficiencies can be made. Sap testing is useful for monitoring the effectiveness of fertilizer application, plant stress due to N and K, etc.

Temperature and time of the day are shown to influence plant sap nitrate content. Taking readings consistently between 9 a.m. and 4 p.m. will yield the most consistent results (Hochmuth, 2003). Sap samples are usually taken from petioles of most-recently-matured leaves. Although three or four petioles may produce a sufficient amount of sap for testing, additional plants must be sampled to ensure that the sap sample is representative of the field or area being tested. In a uniform field, about 20 leaves should be samples to adequately represent a 5- to 10-acre area. The petioles should be chopped and mixed, and a subsample of the chopped petiole pieces used for the final sample to crush (Hochmuth, 2003). Vegetable crops grown in Florida under intensive management can benefit from sap testing and nutrient monitoring. The frequency and consistency required for sampling, and the validity of the procedure for only mobile nutrients such as N and K result in limited application of the procedure.

**Remote sensing Techniques: Near Infra Red Spectroscopy (NIRS)**

Near-infrared (NIR) spectroscopic technique is a multi-step process (Fig 1) that enables rapid and nondestructive analytical correlation of diffusely reflected NIR radiation with chemical and physical properties of soil and plant components. It identifies chemical compounds based on the differential absorption of infrared light at different wavelengths depending upon the nature of the particular chemical functional group. When white light strikes the surface of soil/plants, it is reflected in wavelengths that have a characteristic frequency and energy based on the chemical composition of soil/plants. The reflected light is from the sample surface is measured by a spectrophotometer. While the current technology allows for the use of this technology in the laboratory, sensors are being developed for on-the-go measurement of soil properties with the potential to provide benefits from the increased density of measurements at a relatively low cost (Adamchuk, 2004).

**Why Should NIRS Replace Wet Chemistry Analysis?**

With the advancement of technology, now many soil and plant analyses can be carried out rapidly using automated laboratory equipments, such as an autoanalyzer and inductively coupled plasma (ICP) spectrometer. Each property or concentration has a specific level of energy and the instruments capture the wavelength of that specific light energy, similar to the NIR technique. Determinations are done indirectly, calibrated and validated and interpreted through indices.

Analysis of soil and plant tissue samples using NIRS can provide compositional information of every sample within the time frame and cost of a single determination. The NIRS technique is rapid, instantaneous, non-destructive, cheap, portable and has a wide variety of applications such as routine soil analysis, soil property classification, soil survey and mapping, precision agriculture, diagnosis of soil problems, contaminated site characterization, input data for models, decision support, etc.
The rays are not merely reflected from the outside surface, but actually penetrate the sample. Each time a chemical bond is encountered that does not absorb the particular wavelength, the rays are scattered and reflected in all directions. These scattered beams may then be absorbed or reflected by other chemical bonds until a portion of the rays eventually exits the sample in all directions. The reflected rays then are concentrated onto a measuring cell. The number of reflections at different wavelengths are measured and then converted to analytical results by a microprocessor.

Fig 1. Reflectance measurement, data processing and analysis

Three major research projects in Florida have been funded to develop fundamental relationships between soil properties from representative soil orders in Florida and the spectral characteristics of the soil samples, as a preliminary step toward development of a real-time soil property sensor for use in site specific crop management. The sensor system could eventually contribute to efficient water and nutrient management for economically viable agricultural production and natural resources protection and preservation. Soil pH, organic matter, and nutrient content were studied in 90 soil samples each collected from Alfisols, Entisols and Ultisols (Lee et al., 2003). The predicted concentrations of soil P and Ca using PLS regression are depicted in Figs. 2 and 3 for Alfisols, Entisols and Ultisols of Florida.
Fig 2. Prediction of P with the samples from three representative soil orders (Alfisol, Entisol, and Ultisol) by PLS regression.

Fig 3. Prediction of Ca with the samples from three representative soil orders (Alfisol, Entisol, and Ultisol) by PLS regression.
In another study (Cohen, et al., 2007), a set of 1933 samples, representative of major soil orders in Florida, was assembled from samples submitted to the IFAS Extension Soil Testing Laboratory for routine testing during 2004-05. High-resolution diffuse reflectance spectra of each soil in the visible and near infrared regions were used to predict observations using standard laboratory analytical procedures for soil pH, Mehlich-1 extractable phosphorus, potassium, calcium, magnesium, copper, manganese and zinc, percent organic matter (OM), and saturated hydraulic conductivity ($K_{sat}$). The predicted values using PLS and GBT statistical techniques for certain soil physical and chemical properties studied are shown in Fig 4. Following results highlight some of the conclusions from the study:

- M-1 Al, Ca, Mg and P were predicted more effectively from spectra, and generally have higher concentrations in the soils that were analyzed.
- Analytes with higher concentration were more predictable than those with lower concentrations.
- The relatively high prediction efficiency for Mg is presumably due to its correlation with Ca.
- The relatively high RPD for P might have been a result of correlation with Al and Ca, both of which were effectively predicted.
- We observed model efficiency to be positively associated with mean analyte concentration
- Categorical modeling (threshold-based classes) was successful for pH, M-1 P, Mg, and $K_{sat}$
- In addition, categorical condition for analytes where continuous prediction was insufficient (M-1 Cu, Mn) was successfully diagnosed
- Predictions of M-1 K and Zn categories were statistically significant but of insufficient accuracy to be of diagnostic utility

In an on-going third study, a sample set consisting of 1,000 soil and corresponding 1,000 tissue samples is being collected from throughout the state (Mylavarapu, et. al. 2007). Commodities targeted are forages, vegetables, peanuts and citrus (Ridge, Flatwoods and Indian River). Soil and leaf tissue reflectance will be correlated. A subset of soil samples was used to determine the influence of multiple moisture levels. Soil texture will also be determined for a subset of soil samples.

The NIR technique is very promising and with the availability of new robust statistical techniques, the calibration with wet-chemistry test results can be significantly improved. However, the real-time sensors and field applications of the technology is a few years away.
Fig 4. Predicted values of soil physical and chemical properties using 33% of samples for validation.
Diagnostic Testing and Environmental Assessment
This is the most critical current need in the area of nutrient management. The need to
generate new knowledge to bridge the gap between diagnostic testing for agronomic
production and environmental impact assessment is immense. Traditional soil fertility
research has been revamped into Nutrient Management to accomplish this goal. Typically
multiple factors influence the fate of the nutrients and nutrient cycling in the natural and
agricultural systems. Anthropogenic factors only enhance the cycling rates. In Florida,
such tools have been developed and are being fine-tuned. For example, a simple routine
soil test P is adequate to estimate the plant available phosphorus in the soils. Other factors
that affect the phosphorus movement, adsorption, retention and release in soils have been
compiled along with site management factors into a tool called the Florida Phosphorus
Index (PI) (Hurt and Mylavarapu, 2006). Although originally intended for sites that
receive organic sources of nutrients, the PI can be applied to any situation where
nutrients, particularly are applied. The potential for offsite movement of applied
phosphorus can be qualitatively determined. Use of a combined soil and plant tissue test
for determining the need for phosphorus application to agricultural fields for bahia
pastures and commercial citrus producers have been developed. Similar, models for
nitrogen and heavy metals can be developed.

Priorities

- Develop, calibrate and establish a valid soil extraction procedure for nutrient
  recommendations for calcareous soils, a major soil category for commercial and
  urban landscape horticulture in the state
- Validate the Florida P-Index, a critical tool for phosphorus management, particularly
  when organic sources are used and for various sensitive ecosystems in the state
- Develop a tool for nitrogen management to assess the plant uptake efficiency and
  environmental loss
- Determine the feasibility of complementary soil and tissue analyses for phosphorus
  management for perennial crops and landscape plants and grasses
- Integrate recommendations for irrigation management with nutrient recommendations
  for commercial horticultural crops
- Further develop VNIR techniques and continue field calibration for wider diagnostic
  applications
- Develop newer diagnostic techniques for environmental impact assessment.

References


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