

Current and Emerging Protocols for Carbon Measurement in Agricultural Soils¹

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Introduction

The concentration of carbon dioxide (CO₂) in the atmosphere has been steadily increasing over the past several decades, contributing significantly to the greenhouse effect and climate change. Therefore, to mitigate climate change strategies such as carbon (C) sequestration (the capture and storage of atmospheric CO₂) and reductions in CO₂ emissions are essential to address these challenges effectively. Soils have the capacity to function as a sink of atmospheric CO₂ (i.e., sequestration) and are crucial for climate regulation. Globally, the soil stores around 1.5 to 2.4 trillion metric tons of C annually (Ciais et al. 2014) and can store three times more C than the atmosphere and four times more than plants (Griggs and Noguier 2002). These massive C sinks have the potential to reverse soil degradation, mitigate climate change, and enhance food security (Lal 2019) by increasing soil health and fertility, which in turn can lead to increased crop yields and quality. It is therefore essential to monitor the global C cycle by accurately measuring the amount of soil C in various ecosystems, including highly managed agricultural fields. For accurate soil C stock (or pool) calculations, common standards and procedures for different ecosystems worldwide are required. One of the most difficult aspects of estimating C is obtaining an accurate picture of the soil C stock. Because C stocks can vary greatly even within a single field, it is difficult to obtain a representative sample of soil for testing. Additionally, the selection of sampling methods, sampling design, baseline selections, depth of soil, and use of proper analytical methods are prerequisites for accurately estimating the soil C stocks and getting an overall picture of the area's C levels at different scales.

This article summarizes current and emerging analytical methods for measuring C on different spatial scales, providing valuable information to extension agents, researchers, and other professionals. Other end-users in

sectors such as forestry, ranch and cattle management, and ecosystem services such as wetlands, may also benefit from the information provided here. For more information on the role of growers in the C economy, including topics such as C credits and agriculture practices for C sequestration, please refer to EDIS publications [#AE573](#) and [#AE582](#).

Protocols for Quantifying Carbon

Measuring soil C is crucial for understanding soil health, carbon sequestration potential, and overall ecosystem functioning. Various methods (Figure 1) have been developed to quantify soil C at different scales, ranging from laboratory analyses to remote sensing techniques. These methods provide valuable insights into the spatial and temporal distribution of soil C, enabling scientists, researchers, and land managers to make informed decisions regarding sustainable land management practices and climate change mitigation strategies. Below, we delve into a comprehensive discussion of the prevalent methods used to measure soil C, exploring their principles, methodologies, and applications.

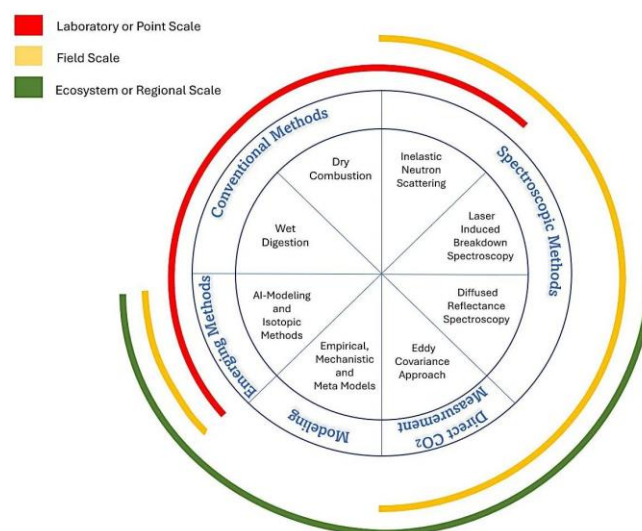


Figure 1. Methods for measuring soil carbon at different scales. Credit: Suraj Melkani and Jehangir H. Bhadha, UF/IFAS

Conventional Methods

Wet digestion and dry combustion are conventional methods, widely used for regular laboratory analysis of C. These methods are the standard procedures available for measuring soil C. These procedures involve analyzing soil samples in a controlled environment to accurately determine the amount of C present. Typically, soil samples are taken using soil augers, which are cylindrical tools used to collect soil samples from a specific depth. The samples are then taken back to the laboratory for analysis. Soil C is measured at the point scale because it involves measurements representative of fields at specific locations and times.

Wet Digestion

The Walkley-Black dichromate oxidation method is a widely used laboratory method for determining C (Walkley and Black 1934), first by oxidizing C in soil with potassium dichromate and sulfuric acid. After the oxidation, the remaining unreacted potassium dichromate is titrated or reacted with a reducing agent like Ferrous Ammonium Sulfate along with an indicator like Diphenyl Amine. This titration process helps determine how much of the potassium dichromate was consumed during the oxidation process. The change in color of the solution, typically from orange to green, due to the reduction of potassium dichromate to chromium ions, plays a crucial role in determining the amount of unreacted potassium dichromate. This change in color is easily observable and serves as an endpoint indicator. The amount of unreacted potassium dichromate can then be related to the amount of carbon present in the original soil sample. This technique is extensively used worldwide because it is easy and rapid, while involving minimum apparatus. However, the wet digestion method may partially oxidize labile soil C, resulting in the loss of some labile carbon compounds during the digestion process. This is a concern because labile carbon is a major portion of soil C. To address these concerns, there is a need for correction factors when interpreting the results obtained through wet digestion methods. These correction factors are designed to account for the potential loss of labile carbon during digestion, ensuring that the estimated carbon content in the soil reflects the true carbon content, including both stable and labile carbon fractions (Neal and Younglove 1993). Labile C refers to the fraction of soil C that is more easily decomposable and can change rapidly in response to environmental factors. Despite being

widely used to measure C concentration around the world, the wet digestion method has limitations, like the effects on labile C, because of variable C recovery percentage. However, the accuracy of C measurement using the wet digestion method can be improved through the use of exogenic heat during digestion and the development of site-specific correction factors.

Dry Combustion

The dry combustion method is a standard and precise laboratory technique for analyzing soil C concentration, which involves incineration of the soil C at a high temperature and measurement of the resulting CO₂ using one of two methods:

1. **Loss-on-Ignition Method (LOI)** (Matus et al. 1997) involves measuring the difference in mass of pre- and post-incinerated dry soils after achieving dry combustion in a high-temperature incinerator (Figure 2A). The variable sample size can be a source of error, and content of C measured with LOI reportedly decreases significantly with an increase in sample weight (Schulte et al. 1991).
2. **Dry Combustion with Elemental Analyzer (EA)** (Atkins and Jones 1991) is the procedure for measuring C using EA, based on the collection of emitted CO₂ after dry oxidation of C (Figure 2B). Dry oxidation involves heating the sample in a combustion chamber containing pure oxygen gas, which oxidizes the C in the sample to CO₂ gas. The resulting CO₂ gas is then collected and measured to determine the amount of C in the sample. However, it should be noted that carbonates do not completely oxidize in both LOI and EA methods, which can introduce variation when measuring C.



Figure 2. Incinerator for measuring C (left) and Dry Combustion Elemental C Analyzer (right).

Credit: Suraj Melkani and Jehangir H. Bhadha, UF/IFAS

The wet digestion and dry combustion methods for measuring soil C are accurate but require laboratory facilities, making them time-consuming and expensive. Therefore, there is a need for portable, rapid, accurate, and cost-effective methods to measure soil C, which would benefit researchers who investigate C cycling and global change processes. As a response to these needs, recent developments have led to the use of spectroscopy methods to improve the efficiency of soil C analysis and stock estimation.

Spectroscopic Methods

The spectroscopic methods can be quick and efficient ways to measure and monitor C. These methods can be used for laboratory as well as in-situ measurements via platforms such as aircraft, satellites, and unmanned aerial vehicles. Spectroscopic methods include methods like diffused reflectance spectroscopy, laser-induced breakdown spectroscopy (LIBS), and inelastic neutron scattering (INS).

Diffused Reflectance Spectroscopy Techniques, or Infrared Spectroscopy

This technique works on the diffuse reflectance property of the soil, which depends on the composition of the soil, particle size distribution, organic matter, and soluble salts within the soil (Ben-Dor et al. 1999). When infrared radiation falls on a sample, it causes electronic transitions in atoms along with vibrational stretching and bending of atomic bonds in crystals (Wetterlind et al. 2010). The amount of C present in the soil can be determined by comparing the sample spectrum to an existing library of spectra (Figure 3).

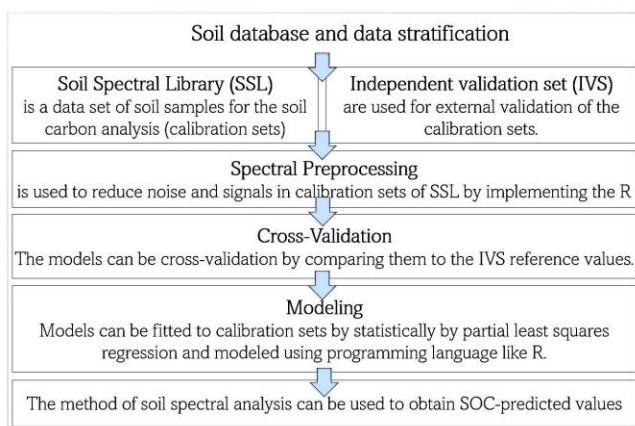


Figure 3. Diffused Reflectance Spectroscopy Techniques for C estimation.

Credit: Diffused Reflectance Spectroscopy Techniques for C estimation.

Depending on the wavelength range of infrared radiation used in diffused reflectance spectroscopy, different structural C properties like C activity, functional groups, aromaticity, arrangement of molecules, etc. can be determined.

Laser-induced Breakdown Spectroscopy (LIBS)

LIBS (Figure 3) is an atomic emission spectroscopy method which uses a high energy laser pulse focused on a soil sample. This causes ionization (i.e., the process of forming ions), excitation (i.e., the process of raising atoms to higher energy levels), evaporation, and atomization (i.e., particle division) of atoms and results in the formation of a high-temperature plasma (Hahn and Omenetto 2010). After cooling, the excited ionic, atomic, and molecular fragments generated within the plasma emit radiations indicative of the elemental composition of the volatilized matter (i.e., matter that has been converted into a vapor or gas state due to high temperature). The individual peaks in the spectrum (Figure 4A) correspond to the elements found in the sample. For C analysis, the strong emission line at the peaks were chosen for calibrating and testing LIBS data with those obtained by laboratory analysis (Figure 4B and 4C). However, there are some known problems with plasma formation as well as unknown problems with how soil properties, such as soil structure and composition, affect LIBS accuracy in estimating C (Senesi and Senesi 2016).

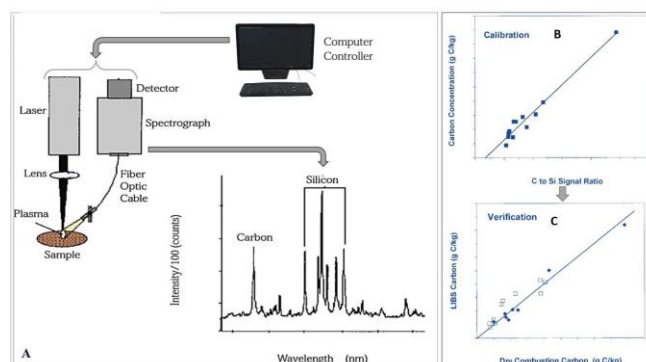


Figure 4. (A) LIBS with a calibration curve reproduced from Chatterjee et.al. (2009) (B) Calibration curve for the detection of total soil carbon using LIBS (C) Correlation between carbon concentration predicted by LIBS and determined by dry combustion to create the carbon prediction model.

Credit: (A) Adapted from Chatterjee et al. (2009); (B) and (C) adapted from Chatterjee et al. (2009)

Inelastic Neutron Scattering (INS)

Inelastic Neutron Scattering (INS) is a method (Figure 5) that uses a low energy neutron beam to measure C atoms

in the soil (Wielopolski and Carayannis 2011). This method can detect both inorganic and organic C by measuring the total number of C atoms present in the scanned area. However, INS faces some challenges, including high costs, lack of optimization and calibration, and the absence of field models ready for research and testing (Nayak et al. 2019). The INS requires further development, such as calibration in larger areas and different soil types, system optimization, and more efficient models to reduce measurement errors.

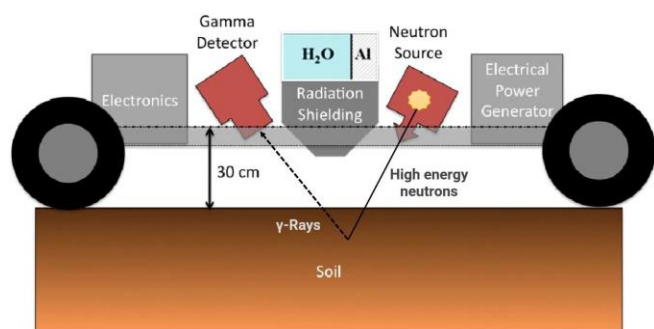


Figure 5. This is a simplified depiction of the Brookhaven INS device based on Wielopolski et al. (2008). The principal components of the device are a 14-MeV-neutron source and multiple gamma ray detectors, mounted on a four-wheeled cart, which would be capable of traversing arable land.

Credit: Adapted from Wielopolski et al. (2008)

Spectroscopic methods can be used for measuring and monitoring C on a field as well as on a regional scale. But, since they are indirect methods for measuring soil C, their current precision is low. Soil C can also be measured on a large scale by directly measuring the ecosystem C exchange.

Direct CO₂ Exchange Measurement: Eddy Covariance Approach

An alternative to measuring changes in soil C stocks is to measure all the inputs and output of C at a large geographical scale. Ecosystem CO₂ exchange is dominated by photosynthesis and respiration. All these CO₂ exchange components can be measured directly with the Eddy covariance flux approach (Figure 6B). This method can effectively measure C and direct, greenhouse gas emissions (Burba et al., 2013).

Eddy covariance measurements of CO₂ exchange include that of vertical wind velocity using a sonic anemometer, in addition to measuring fluctuations in CO₂ mixing ratio using an open- or closed-path gas analyzer. C flux is commonly used to track long-term changes in land C as

well as substantial CO₂ fluxes, which refer to the exchange of CO₂ between the atmosphere and the earth's surface. They occur due to natural processes and human activities such as burning fossil fuels and deforestation (Nayak et al. 2019).

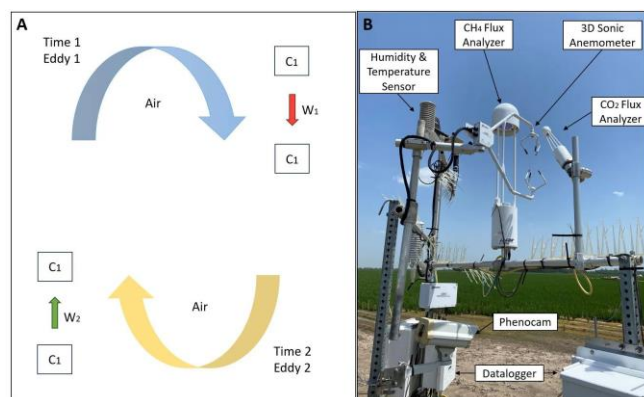


Figure 6. (A) Two eddies on the tower showing CO₂ with parcel of air C1 moving down at speed W1 and parcel of air C2 moving down at speed W2 (B) Eddy Covariance Flux Tower

Credit: Suraj Melkani and Jehangir H. Bhadha, UF/IFAS

Challenges with this method of measuring C include inherent uncertainties with the flux estimation and biomass sampling, as well as the proper development of hardware that can rapidly and accurately analyze turbulent flux data (Rinne and Ammann 2012). Eddy covariance approaches also require long-term studies and reduction of uncertainties associated with CO₂ flux estimation. On a regional or global scale, C can also be predicted with the use of C models.

Soil C Modelling Approaches

Soil C models can be used to simulate C and estimate soil C budget components such as: (a) CO₂ emissions from soil caused by the decomposition of organic C compounds in soil; (b) the C pool soil; and (c) changes in the C pool of soil over time. Soil C models can be classified as empirical models, mechanistic- or process-based models, or meta (fusion) models.

Empirical Models

Empirical models use linked mathematical equations to express conceptual understanding of ecosystem processes.

Mechanistic- or Process-based Models

Process-based models have been established and utilized to simulate biogeochemical processes in the plant-soil-ecosystem (Wang et al. 2017). Table 1 provides

information about some process-based models that can effectively simulate soil C.

Meta Models (Fusion Models)

These models combine the features of both empirical and process-based models to achieve greater accuracy. These models are also less complex than process-based models.

The modelling approach requires less time and saves resources used in destructive methods. However, accounting for soil C change using C models needs reliable databases from different agriculture ecosystems. One of the other limitations of process-based models is that they are available for only a few crops and sometimes do not respond to all environmental and management factors. To counter some of the limitations, new techniques for studying C dynamics are emerging.

Emerging Methods

Using C Isotopes to Determine C Decomposition and Turnover Rate

C isotopes can be particularly useful in determining the organic matter decomposition rate and dynamics, rather than actual soil C values in real time. Several methods involve the use of C-13 and C-14 isotopes to determine and measure the decomposition or turnover of C (Balesdent, Wagner, and Mariotti 1988; Paul et al. 1997). The turnover rate of an element in a pool is defined as the balance of the element's inputs (I) and outputs (O) to and from the pool (Six and Jastrow 2002). The obtained decomposition, or turnover value, can be used to calculate soil C:

Soil C = C input – C loss (calculated from turnover rate)

Using Machine Learning for Modeling Soil C Change

Soil C modeling using machine learning is becoming a powerful tool for understanding soil C dynamics. When compared to other modeling approaches, these artificial intelligence generated soil C models perform better in predicting soil C components and other global C cycle properties (Grunwald 2022).

Soil C artificial intelligence models use environmental covariates as inputs, representing different parameters associated with soils, topography, ecology, parent material or lithology, climate, hydrology, organisms, and human activities (Grunwald, Thompson, and Boettinger

2011; Thompson et al. 2012). Table 2 presents commonly applied artificial intelligence algorithms to model soil C.

Artificial intelligence modeling offers sophisticated abilities to estimate C stocks. These artificial intelligence models have potential for avoiding human error at all scales, even though they are currently data-restricted in their ability to explain the spatial variability of soil C storage within landscapes.

Conclusion

Soils have a great capacity to sequester C. However, data on their C storage is limited. Accurate soil C measurements are therefore critical for quantifying soil C pools and inventories, as well as for monitoring the inherent spatial heterogeneity of soil C content and changes that may occur because of various sustainable management practices. Therefore, accurate assessment of soil C quantity would make a significant contribution to assessing disturbance impacts, global climate change, and land use change.

Regardless of the underlying principles, each method has its distinct advantages and limitations. All these methods are effective, but they need to be more accurate at all geographical scales. Therefore, for more efficient quantification of soil C, further research on the existing and emerging methods is required.

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Table 1. Process-based Models for Soil C Simulation: A Review of Popular Tools.

Model	Relevance/ Importance to Professionals	Links/References
DSSAT (Decision Support System for Agrotechnology Transfer)- CENTURY Model	Helps extension agents and researchers with decision-making in agriculture by simulating crop growth and yield under different conditions.	https://dssat.net/
DayCent (Daily Century) Model	Helps researchers and professionals better understand and model the C, nitrogen, and water cycles in terrestrial ecosystems, including croplands and forests.	https://www.nrel.colostate.edu/projects/century/
DNDC (Denitrification-Decomposition) Model	Helps researchers and professionals understand and model the biogeochemical cycles of C, nitrogen, and phosphorus in agricultural and natural ecosystems, as well as predict their response to different management practices.	https://www.dndc.sr.unh.edu/
Comet-Farm Model	Helps farmers, ranchers, and land managers evaluate and compare the environmental and economic impacts of different land management practices, including the reduction of greenhouse gas emissions.	https://comet-farm.com/

Table 2. Artificial Intelligence Algorithms for Soil C Modeling.

Artificial Intelligence Algorithm	Relevance and uses
Classification and Regression Trees	This algorithm is useful for predicting soil C components and other global C cycle properties and can help extension agents and researchers to better understand soil C dynamics.
Bagged Regression Trees	This algorithm is useful for identifying patterns in large datasets and can help researchers and other professionals to analyze complex soil C data.
Boosted Regression Trees	This algorithm is useful for modeling nonlinear relationships and can help researchers and Extension agents to predict soil C dynamics under changing environmental conditions.
Random Forest	This algorithm is useful for identifying important predictors of soil C dynamics and can help researchers and Extension agents to prioritize management interventions for soil C.

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