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Analyzing the Relation between Loss-on-Ignition and Other Methods of Soil Organic Carbon Determination in a Tropical Cloud Forest (Mexico)

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The objective of this study was to analyze the relationship between soil organic carbon content, determined by dry combustion (%OC_{LECO}) and the Walkley–Black method (%OC_{WB}), and loss on ignition (LOI). Soil samples were collected from noncalcareous O and A1 horizons within a tropical cloud forest. Linear regression equations were developed to estimate organic carbon from LOI. The applicability of the predictive equations was evaluated by comparison of measured and predicted organic carbon data for independent soil samples. The results showed that the LOI method produced a better linear relationship with the %OC_{LECO} ($R^2 = 0.96$, $P < 0.001$) than with the %OC_{WB} ($R^2 = 0.88$, $P < 0.001$) method. These results also showed that %OC_{WB} and %OC_{LECO} prediction equations underestimate and overestimate soil organic carbon by 0.74% and 0.56%, respectively. This study suggests that LOI may be a good estimator of soil organic carbon for noncalcareous O and A1 horizons in a tropical cloud forest.

Keywords Dry combustion, loss on ignition, soil organic carbon, tropical cloud forest, Walkley–Black method

Introduction

The assessment of organic carbon in soils is an important topic. The interest of scientists and policy makers in the overall global carbon budget has grown dramatically in recent years because of concerns about an increase in levels of carbon dioxide (CO_2) in the atmosphere and its potential for causing global climate change (Cheng and Kimble 2001). Understanding carbon cycling processes in the environment can help assess the impact of increasing CO_2 emission into the atmosphere and the mitigating effect on the carbon status in various environmental compartments (Cheng and Kimble 2001). Carbon stored in soil organic matter is a substantial part of the global carbon cycle, estimated at about 1500–2000 Pg (1 Pg = 10^{15} g), more than is found in the biota (400–600 Pg) or atmosphere (785 Pg) (Schimel 1995; Janzen 2004; Smith et al. 2008). Key attributes of soil organic matter are its functions as a nutrient source, nutrient retainer, and a contributor to structural stability in soil. In addition, organic matter is an energy source for organisms that have a pivotal role in recycling nutrients for plant growth (CAST 2004). According to Shukla, Lal, and Ebinger (2006), soil organic carbon is probably the most widely used soil quality indicator, but land use change and many agricultural practices have made soils release CO_2 into the atmosphere, contributing to global warming (Rosenberg, Izaurralde,

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and Malone 1999). Present soil carbon stocks reflect land use and a management history in accordance with a certain soil type and hydrological regime under prevailing climatic conditions (Ellert, Janzen, and McConkey 2001; Shrestha and Singh 2008). Assessment of soil carbon stocks is a basic step in evaluating the carbon sequestration potential of an ecosystem for the implementation of the Kyoto Protocol of the United Nations Framework Convention on Climate Change (Shrestha and Singh 2008).

Measurements of soil carbon under different agricultural treatments, soil types, climatic conditions, and land-use histories are keys to establishing relationships between these variables and soil carbon (Westman, Hytönen, and Wall 2006). Methods for measuring soil carbon have been documented extensively, and there are well-developed standards (Nelson and Sommers 1996; Carter and Gregorich 2007). The most commonly used methods for determining soil carbon are wet digestion and dry combustion (Nelson and Sommers 1996; Kimble, Lal, and Follett 2001).

In soil-testing laboratories, the Walkley–Black (1934) method has been the most widely used routine procedure for organic-matter determination because it is easy to conduct and requires minimum equipment (Lettens et al. 2007). Wet combustion or oxidation methods most commonly use potassium dichromate in concentrated sulfuric acid to oxidize soil organic carbon under diverse temperature regimes and heating times, but this method generates waste containing strong acids and chromium (Schulte and Hopkins 1996; Zhang, Li, and Stoffella 2005). Organic-matter content in soil is commonly estimated by measuring dichromate reduction after organic carbon oxidation, calculated by assuming 58% carbon in organic matter and an absence of other oxidizable species (Nelson and Sommers 1996). However, the traditional factor of 0.58 is considered too high (Read and Ridgell 1922; Jain, Graham, and Adams 1997). Soil organic matter is not a homogenous entity, and its carbon concentration may vary with the age of the organic matter, quality of litter, and decomposition conditions (Read and Ridgell 1922; Westman, Hytönen, and Wall 2006). On the other hand, dry combustion methods, using automated carbon (C) analyzers, are based on thermal oxidation of soil carbon in the medium temperature range ($\sim$1000 °C) in an oxygenated environment (purified O_2 and metal oxides), usually in the presence of a metal combustion accelerator to convert all carbon into CO_2 (Allison 1965; Nelson and Sommers 1996). Dry combustion, measuring CO_2 evolved from organic matter oxidized in a high-temperature furnace, is considered the most precise and accurate procedure today, but the high cost of dry-combustion instruments is a limitation for many laboratories (Konen et al. 2002).

The high cost of dry-combustion instruments and environmental hazards associated with the Walkley–Black technique have prompted the search for alternative methods for estimating soil organic carbon (Schulte and Hopkins 1996). The weight loss on ignition (LOI) method is a promising alternative for quantification of soil organic carbon (Schulte and Hopkins 1996). This method measures the weight loss from a dry soil sample subjected to high-temperature ignition, as weight loss is proportional to the amount of soil organic matter in the sample (Konen et al. 2002; Zhang, Li, and Stoffella 2005). Heating times and temperatures used in the LOI method have been reported to range from 2 to 24 h and from 360 to 600 °C, respectively (Schulte, Kaufmann, and Peter 1991). LOI is an accurate and labor-efficient method for determination of soil organic carbon that requires only a muffle furnace, ceramic crucibles, and an analytical balance; thus, analysis can be conducted rapidly at low cost (Konen et al. 2002; Westman, Hytönen, and Wall 2006). Although studies suggest that LOI has great potential for easily and accurately estimating soil organic carbon (David 1988; Cambardella et al. 2001; Konen et al. 2002), several important considerations apply to LOI (Abella and Zimmer 2007). Loss on ignition overestimates the amount of soil organic matter due to the removal of hygroscopic and intercrystalline water from clay minerals and allophane, water loss from hydroxyl groups in sesquioxides, and CO_2 release from carbonates (Howard and Howard 1990; Goldin 1987). Because of problems associated with the wide range of factors used to convert soil organic carbon to soil organic matter, and more recently because of interest in the quantification of soil carbon, researchers have suggested that LOI should be used to estimate soil organic carbon rather than soil organic matter (Nelson and Sommers 1996; Schulte and Hopkins 1996; Konen et al. 2002).

The objective of this study was to investigate the relationship among LOI, dry combustion, and Walkley–Black methods for estimation of soil organic carbon content in a tropical cloud forest landscape.

Materials and Methods

Site Description

This study was carried out in two small adjacent watersheds covered with a tropical cloud forest. Found in the middle part of the eastern side of the Cofre de Perote Volcano, they are located between 19° 29′ 28.8″ N, 97° 02′ 23.9″ W and 19° 29′ 36.2″, 97° 02′ 40.8″ W. Their size is approximately 24.7 and 52.8 ha, and the altitude there ranges from 2169 to 2059 m above sea level, respectively. Each watershed includes a small perennial stream fed by base flow. The tropical cloud forest is the most diverse type of vegetation in Mexico because it occurs on less than 1% of the territory but harbors 2,500 plant species that grow in this type of forest (Rzedowski 1996). Eighteen percent of the plant species are trees, more than 30% are epiphytes, and around 20% are ferns (Rzedowski 1996). Based on the work of Castillo-Campos (1991), the arboreal species in tropical cloud forest are *Liquidambar macrophylla* Oerst., *Carpinus caroliniana* Walter, *Ulmus mexicana* (Liebm.) Planch., *Platanus mexicana* Moric., *Clethra macrophylla* M. Martens and Galeotti, *Quercus xalapensis* Bonpl., and *Quercus germana* Schltdl. The three most common tree fern species are *Alsophila firma* (Baker) D. S. Conant, *Lophosoria quadripinnata* (J. F. Gmel.) C. Chr., and *Sphaeropteris horrida* (Liebm.) R. M. Tryon (Bernabe, Williams-Linera, and Palacios-Rios 1999). The climate is humid subtropical with frequent fog, mainly during autumn and winter; the mean monthly temperature is 19.3 °C. January is the coldest month (15.8 °C), and May is the hottest (22.2 °C). Total annual rainfall is 2081 mm. The soil is umbric Andosol (FAO 1988) and originated from volcanic ash.

Sampling and Measurement

To conduct this research, three typical slopes were chosen in small watersheds. Slopes have gradients of 40°, 25°, and 20° and lengths of 194, 254, and 105 m, respectively. Slopes were divided into three positions according to their topography: shoulder (SH), backslope (BS), and footslope (FS). All slopes transects extended to base flow. At the center of each slope position, soil was sampled randomly. A total of 55 soil samples were taken from O ($n = 33$) and A1 ($n = 22$) horizons. The O horizon was sampled in total (Oi, Oe, Oa) and had at average depth of 8.0 cm. A PVC (polyvinyl chloride) cylindrical corer (10.5 cm in diameter and 10.0 cm high) was used to take samples of the O layer. Macroscopic living material and roots were discarded from the samples. The A1 horizon soil samples were taken from soil profiles. Also, to evaluate soil carbon linear regression models, 32 soil samples (37% of the total soil samples) were taken at random from different slope positions. The samples obtained covered a wide range of soil carbon content, slope features, and typical environmental conditions of tropical cloud forest.

The samples were air dried. Prior to analysis, organic material samples were pulverized using a rotor mill (Arthur H. Thomas CO., Scientific Apparatus, Philadelphia, Penn.) to powder ($\leq$ 0.85 mm), and mineral soil samples were crushed and passed through a 2-mm sieve. Loss on ignition was determined on a 2-g oven-dry soil sample, dried at 105 °C, weighed in a porcelain crucible, and combusted in a muffle furnace (Barnstead/Thermolyne, model FA 1850-1, Dubuque, Iowa) at 550 °C for 4 h (Bhatti and Bauer 2002). Then, ignited samples were allowed to cool in a desiccator to room temperature, and the weight loss percentage was determined from the following formula:

$$\text{LOI}, \% = \frac{\text{Weight}_{105} - \text{Weight}_{550}}{\text{Weight}_{105}} \times 100 \tag{1}$$

where Weight_{105} is that of the soil sample after heating at 105 °C and Weight_{550} is that of the soil sample after ignition at 550 °C.

Organic carbon was determined in all samples by the standard Walkley–Black method (Walkley 1947; Nelson and Sommers 1996). The total carbon concentration of all samples was measured with a dry-combustion analyzer (LECO CN-2002, St. Joseph, Mich.). Sample quantity was between 0.15 and 0.2 g. Total carbon was presumed to equal organic carbon because all the soils sampled were noncalcareous with a water pH of less than 4.4.

Statistical Analysis

Regression analysis was used to describe the relationship between soil organic carbon content and LOI. In this study, LOI was used as an independent variable in regression equations to estimate soil organic carbon, which had been measured on test samples using the Walkley–Black method and a dry combustion analyzer (LECO CN-2002, St. Joseph, Mich.).

Simple linear regression was performed with SigmaStat 3.5 (2006). The resulting models were also checked for statistical consistency by analysis of residuals and outliers. Goodness of fit was evaluated by the coefficient of determination (R^2).

Evaluation of Predictive Equations

Predictive equations were applied on an independent soil data set (but representative for the same region and tropical cloud forest), and their predicted soil carbon contents were compared with measured soil carbon contents. The prediction quality was determined by statistical indices, which evaluate several aspects of the prediction (Moreels et al. 2003). I used the mean predicted error (MPE), standard deviation of the prediction error (SDPE), and root mean square prediction error (RMSPE), calculated as follows:

$$\text{MPE} = \frac{1}{n}\sum_{i=1}^{n}(y_i - \hat{y}_i) \tag{2}$$

$$\text{SDPE} = \sqrt{\frac{1}{n-1}\sum_{i=1}^{n}\left[(y_i - \hat{y}_i) - \text{MPE}\right]^2} \tag{3}$$

$$\mathrm{RMSPE} = \sqrt{\frac{1}{n}\sum_{i=1}^{n}(y_i - \hat{y}_i)^2} \tag{4}$$

where y_i is measured soil carbon content of the ith soil sample, $\hat{y}_i$ is predicted soil carbon content of the ith soil sample, and n is the total number of observations. According to De Vos et al. (2005), MPE allows the evaluation of a positive or negative bias of a predictive equation, indicating an average tendency for overestimation or underestimation, respectively, whereas SDPE shows the random variation of the predictions after correction for the global bias. RMSPE is a measure of the overall error of the prediction (De Vos et al. 2005).

Results and Discussion

Prediction of Soil Organic Carbon

Strong linear relationships between LOI and soil organic carbon determined by dry combustion ($\%OC_{LECO}$) and the Walkley–Black method ($\%OC_{WB}$) were observed for sample sets [Eqs. (5) and (6)].

$$\%OC_{LECO} = -2.901 + 0.547\,(\mathrm{LOI}), \quad R^2 = 0.962,\ P < 0.001 \tag{5}$$

$$\%OC_{WB} = -3.924 + 0.453\,(\mathrm{LOI}), \quad R^2 = 0.886,\ P < 0.001 \tag{6}$$

The LOI explained 96% and 88% of the variation in soil organic carbon content determined by dry combustion and the Walkley–Black method, respectively, indicating that the relationship was improved with the use of the dry-combustion analyzer. The t-values for the intercepts of the regression equations for $\%OC_{LECO}$ ($P = 0.002$) and $\%OC_{WB}$ ($P = 0.005$) were significantly different from zero and therefore could not be omitted. The slope in Eq. (5) (0.547) was closer to the ratio that is generally accepted and used worldwide (0.58; De Vos et al. 2007). However, a number of studies have shown that the proportion of carbon in soil organic matter is highly variable under a range of soil environmental conditions and that there is no factor appropriate for all soils (Nelson and Sommers 1996). These authors mention that carbon is the chief element present in soil organic matter, comprising 48 to 58% of its total weight.

Scatterplots (Figures 1 and 2) of the standardized residuals in relation to soil organic carbon content show that the regression equations describe the data correctly, since a pattern forms when the residuals are evenly distributed across the regression line. Thus, plots do not suggest violations (regarding the prerequisite of homoscedasticity) of the assumptions of zero means and constant variance in the random error. On the other hand, upon checking individual observations, Cook's distance is acceptable because none of the coefficients were more than 1. This means that the models are correctly specified, predictions are accurate, and therefore LOI allows a good estimation of soil organic carbon for O and A1 horizons in tropical cloud forest.

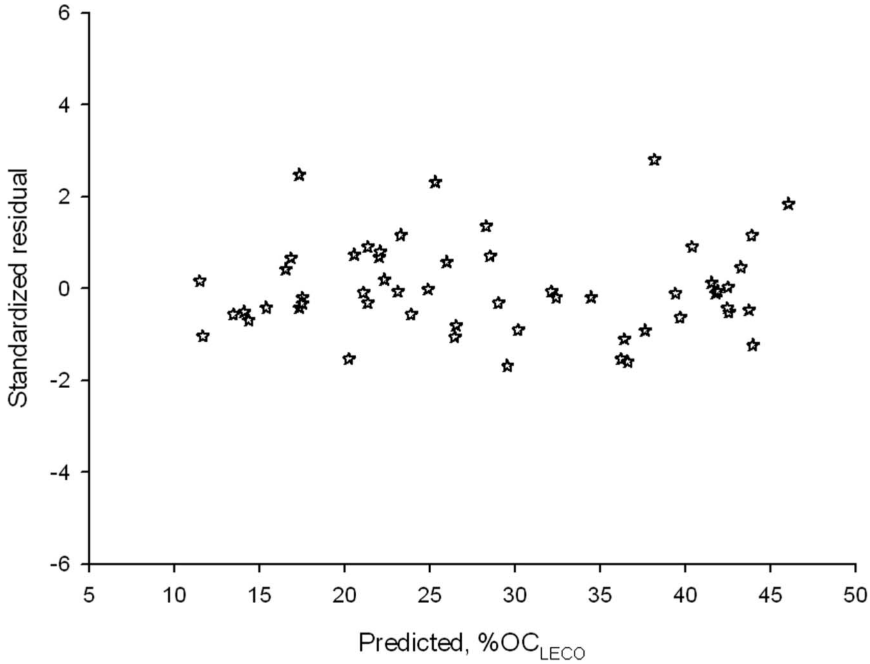

Figure 1. Scatter plot of the standardized residual in relation to predicted $\%OC_{LECO}$.

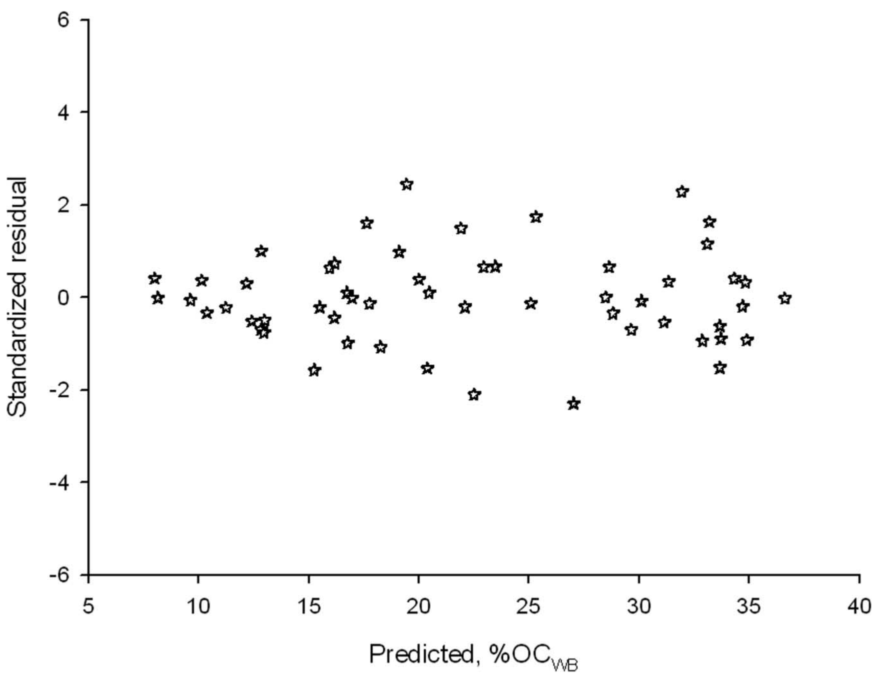

Figure 2. Scatter plot of the standardized residual in relation to predicted $\%OC_{WB}$.

Table 1
Predictive equations of soil organic carbon content and error analysis for evaluation of the equations

	Evaluation (n = 32)		
Equations	MPE (%)	SDPE (%)	RMSPE (%)
$\%OC_{WB} = -3.924 + 0.453\ (LOI)$	−0.745	2.622	2.234
$\%OC_{LECO} = -2.901 + 0.547\ (LOI)$	0.557	2.013	2.050

Evaluation of Predictive Equations

The predictive equations were evaluated (Table 1) using an independent soil organic carbon content data set. Equation (6) underestimates soil organic carbon content by 0.745%, whereas Eq. (5) has a positive bias and overestimates soil organic carbon content by 0.56%. Mikhailova, Noble, and Post (2003) also found that the Walkley–Black method underestimates soil organic carbon more than the dry-combustion method. The use of the latter method to test soil organic carbon appears more accurate; however, it is only valid in soils with no carbonates (Mikhailova, Noble, and Post 2003). In this study, a slightly greater accuracy (lower MPE) was found with Eq. (5) than with Eq. (6). Equation (5) also showed the least deviation of the prediction error (SDPE), indicating a slightly greater precision than Eq. (6). The RMSPE and MPE results showed that Eq. (6) was associated with slightly greater prediction inaccuracies and bias than Eq. (5). This means that LOI was more closely related to the dry-combustion analyzer, considered the most accurate procedure to determine total carbon content, but the high cost of this instrument is a limitation for many laboratories (Konen et al. 2002). These authors recommend that unique predictive equations be developed for individual soil–geographic regions. In any case, these results support the use of LOI to estimate soil organic carbon for the O and A1 horizons in a tropical cloud forest.

Conclusions

Regression equations developed for the prediction of soil organic carbon content from LOI for O and A1 horizons of a tropical cloud forest achieved statistical consistency. The results of the present study showed that LOI is highly correlated with total organic carbon, explaining 96% of variation. Compared to Eq. (6), Eq. (5) was less biased, more precise, and more accurate. The statistical results of the study indicate that for prediction of soil organic carbon in a tropical cloud forest based on the LOI, the linear regression model $\%OC_{LECO} = -2.901 + 0.547LOI$, $R^2 = 0.962$, can be recommended. The LOI method would be of particular value for restoration ecology research and for determining the impact of deforestation and anthropogenic disturbance on the soil organic carbon pool in a tropical cloud forest.

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