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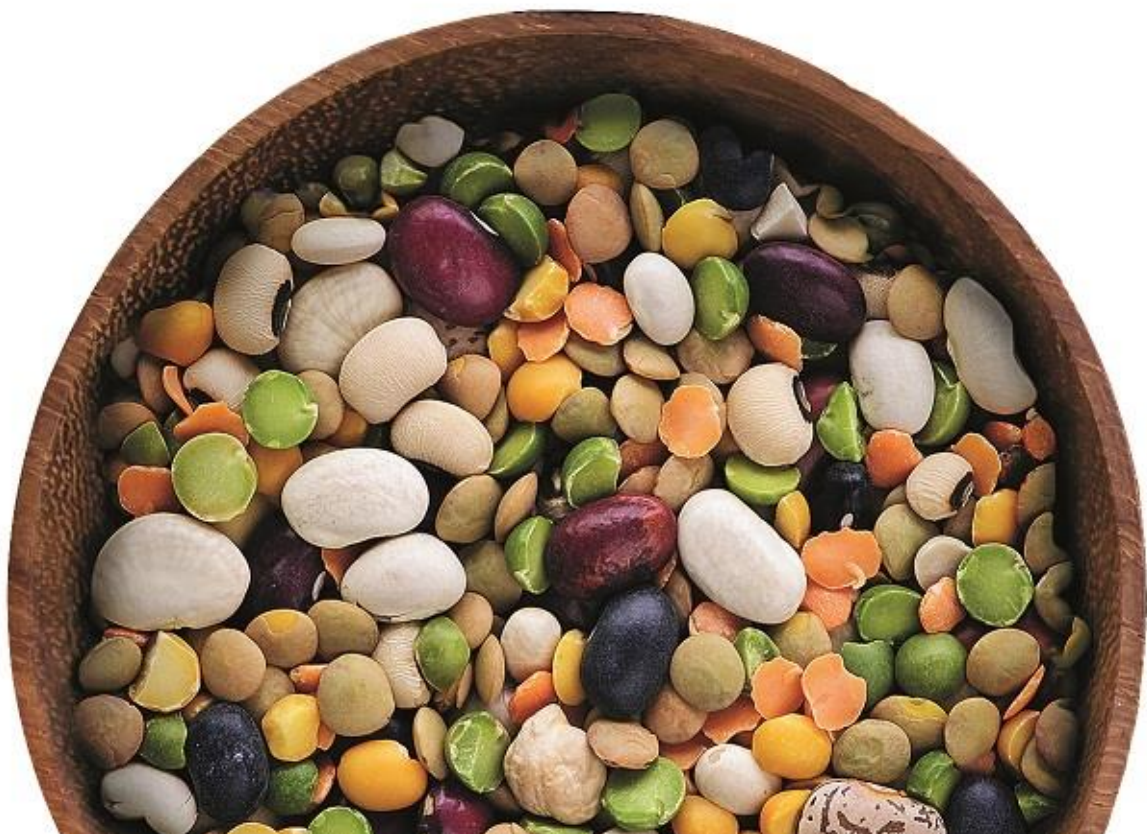
TRansition paths to sUstainable
legume-based systems in Europe

Soil Organic Carbon (OC)

Date: September, 30th 2018

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legume-based systems in Europe

Content

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1 Summary Information

1.1 Partner Summary

SOP Code	EU_TRUE_SOP_026
TRUE Partner Acronym	AUA
Primary Author	Savvas, Dimitrios (*dsavvas@aua.gr*)
Other Authors	Karavidas I., Ntatsi G.
Linked Reference and Hyperlink (if available)	Walkley, A. and I.A. Black. 1934. An examination of the Degtjareff method for determining soil organic matter, and a proposed modification of the chromic acid titration method. Soil Sci.37:29-38 *http://dx.doi.org/10.1097/00010694-193401000-00003*
Associated files to use with the SOP [and function]	*Not applicable*

1.2 SOP Summary

Title

Soil Organic Carbon (OC)

Brief description

Soil organic carbon improve soil structure, water retention, soil biodiversity, buffering capacity and the cycling and storage of plant nutrients. In addition, the mineralization of soil organic carbon provides plant nutrients, especially nitrogen, phosphorus and sulfur (S), as well as with micronutrients. Therefore, the measure of soil organic carbon levels is necessary to determine the soil fertility. The Walkley-Black method was chosen as it is a simplified method, which provides accurate measurements on soils with high organic carbon content.

Application and Principle

The Walkley-Black (WB) method (Walkley and Black, 1934) determines soil organic carbon (OC) by oxidising carbon with acidic dichromate ($K_2Cr_2O_7$). The oxidation step is followed by titration of excess dichromate with ferrous sulfate. The OC is calculated from the the difference between the total dichromate added and the amount of dichromate left unreacted after the OC oxidation. The method provides an estimate of soil Organic Matter from OC determination assuming that the fraction of soil OC reacted and the amount of OC in soil OM. This method was widely considered a standard method for OM after its development, but the advance of thermal decomposition instruments has resulted in its limited use. Instead loss on ignition and thermal decomposition methods are more widely used due to their ability to analyse samples in a more time efficient manner.

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2 Protocol Steps

Equipment and Apparatus

1. Analytical balance with 0.01 g resolution
2. 250-mL wide mouth graduated Erlenmeyer flasks
3. Repipetter to deliver 10 mL volume
4. Fume Hood
5. Titration stand and burette
6. Stir plate with light
7. Stirring rods
8. Weighing vessel

Reagents

1. **Potassium dichromate ($K_2Cr_2O_7$)**, 0.167 M, 1 N
   Dissolve 98.08 g of oven-dried potassium dichromate in approximately 1500 mL of deionized water and dilute to 2 L. After preparation, transfer to a clean glass bottle for use with a repipetter to deliver 10 mL. Do not mix old potassium dichromate solution with the new solution.

2. **Ferrous sulfate ($FeSO_4.7H_2O$)**, 0.5 M, 0.5 N
   Dissolve 278.02 g of ferrous sulfate in approximately 1500 mL of deionized water. Carefully add 30 mL of concentrated sulfuric acid, mix, cool, and dilute to 2 L. After preparation, this solution may be transferred to a clean 8-L plastic carboy. Do not mix old ferrous sulfate solution with the new solution. The tubing, stopcock, and attachments to the burette should be rinsed three times with new ferrous sulfate solution before titrating any blanks or samples. Prepare a new solution every 30 days.

3. **Ferroine** (1,10-Phenanthroline-ferrous sulfate-complex) indicator solution, 0.025 M
   Available from chemical supply companies.

4. **Sulfuric acid** (H_2SO_4)
   Concentrated and not less than 96%.

Procedure

1. Titrate blank samples without soil before proceeding with any unknown samples. Add 10 mL of potassium dichromate into a 250 mL Erlenmeyer flask. Titrate with ferrous sulfate. Titrant volume should be approximately 20 mL. Record the exact volume of titrant to the nearest 0.1 mL. If the titrant volume of the two blanks is not within 0.2 mL, clean the burette and associated tubing. Reanalyse two more blanks.

2. Weigh 1.00g of processed mineral soil (dried, <2 mm) into a 250-mL wide mouth graduated Erlenmeyer flask. If soil weight is not exactly 1.00 g, record weights for later calculation.

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3. Pipette 10 mL of potassium dichromate into each flask and mix carefully by rotating the flask to wet all of the soil.

4. Under a fume hood, carefully add 20 mL of sulfuric acid to each flask and mix gently.

5. Allow flasks to stand for 5 min under the fume hood.

6. Add deionized water to each flask so the final volume is approximately 125 mL. Mix by swirling gently.

7. Allow the samples to cool and return to room temperature. If volume is less than 125 mL after 30 min, add more deionized water to achieve 125 mL volume.

8. Add 5 or 6 drops of ferroine indicator solution and immediately titrate with ferrous sulfate. Use a mixing bar to thoroughly stir the sample as it is titrated. As the titration proceeds, the solution will take on a green colour that will change abruptly to reddish-brown when the endpoint of the titration is reached.

9. Record each volumetric reading to the nearest 0.1 mL.

Calculations

1. Soil organic C (OC) is determined from the m_{eq} of $K_2Cr_2O_7$ (in mL) reacting with the soil to oxidize organic C, which is the difference between total m_{eq} of $K_2Cr_2O_7$ (in mL) added to soil and the m_{eq} of $FeSO_4$ that titrated the remaining $K_2Cr_2O_7$ after the reaction.

   OC (%) = (m_{eq} $K_2Cr_2O_7$ - m_{eq} $FeSO_4$) × [0.003 (g.C.m_{eq}^{-1})] × cf ÷ g soil × 100

   The oxidation state of C in organic matter is assumed to be 0. The C in organic matter is oxidised to CO_2 with an oxidation state of +4. Thus, the equivalent weight of C is 3 g (12 g C ÷ mole × mole C ÷ 4 eq C = 3 g C_{eq}^{-1} = 0.003 g C m_{eq}^{-1}). An oxidation correction factor (cf) is required since not all the organic C in soil is oxidized with room temperature oxidation. Correction factors can range from 1.14 to 1.32.

2. The m_{eq} of $K_2Cr_2O_7$ reacting with soil to oxidise OC is determined from the following equation.

   (m_{eq} $K_2Cr_2O_7$ – m_{eq} $FeSO_4$) = (mLB – mLS) ×0.5 N

   with **mLB** = average volume of ferrous sulfate to titrate two blanks of potassium dichromate without soil (in mL)
   mLS = the volume of ferrous sulfate to titrate the sample (in mL)
   0.5 = the normality of the ferrous sulfate titrant

3. Substitution into the equation for OC results in the following.

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$$\text{OC (\%)} = (mLB{-}mLS) \times 0.5 \times [0.003 \ (g.C.m_{eq}^{-1})] \times cf \div g \ soil \times 100$$

Using a correction factor (cf) of 1.3 results in:

$$\text{OC (\%)} = (mLB{-}mLS) \times 0.195 \div g \ soil$$

4. Since there is approximately 58% C in soil organic matter, soil OM can be determined as:

$$\text{OM, \%} = (mLB{-}mLS) \times 0.195 \div g \ soil \times (100 \ g \ OM \div 58 \ g \ OC)$$
$$= (mLB{-}mLS) \times 0.336 \div g \ soil$$

Analytical Performance

Range and Sensitivity

1. This method is appropriate for mineral soils with OM content less than 6%. Soils with more than 6% OM should be analysed using a quantity of soil lower than 1 g to avoid difficulties in determining the colour change during the titration and consumed all the potassium dichromate, which oxidises the OM.

2. Each volumetric reading is recorded to the nearest 0.1 mL. This titrant volume change corresponds to 0.03% OM when 1 g of soil is used.

Precision and Accuracy

1. To ensure good precision, you must be able to view the reddish-brown colour change at the endpoint to stop the titration. The aid of a stirring bar and a well-lit stirring plate can improve the ability to view the end point.

2. Results from 151 North American Proficiency Testing samples from 1999 through 2008 show median interlaboratory precision of 10.6% ranging from 4.6 to 28.4% (median absolute deviation/median x 100) with median soil OM of 2.0% ranging from 0.5 to 8.1%.

3. Intralaboratory precision from the University of Florida Extension Soil Testing Laboratory on control samples, analysed 15 times, averaged 1.09 ± 0.074% (standard deviation/average x 100) for soils with OM ranging from 0.85% to 1.14%.

Interferences

1. Ferrous iron and chloride in soil can result in positive errors in OM as these constituents can result in a reduction of $K_2Cr_2O_7$. However, ferrous iron should not be a problem in soils thoroughly air-dried since this step ensures almost complete oxidation of ferrous iron to ferric iron. Chloride should not be a concern in well drained soils devoid of any recent fertiliser application. Options for treating soils with high chloride can be found in Nelson and Sommers (1996).

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2. Manganese oxide (MnO_2) can compete with $K_2Cr_2O_7$ in oxidizing soil OM resulting in negative errors in OM. This is a rare occurrence and only occurs with freshly precipitated MnO_2.

Interpretation

Interpretation of OM concentrations regarding soil fertility can be found In Magdoff *et al*. (1996).

Effects of Storage

1. Air-dried soil can be stored indefinitely without affecting these measurements.

2. Potassium dichromate and ferrous sulfate solutions should be prepared fresh every day

Safety and Disposal

Potassium dichromate is defined as a hazardous chemical by the ResourceConservation and Recovery Act due to toxicity (USEPA, 1980a). A laboratory generating more than 100 kg of material in a month is considered a hazardous waste generator that needs to follow hazardous waste disposal protocols defined by the US Environmental Protection Agency (USEPA, 1980b).

Additional Reference

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Walkley, A. and I.A. Black. 1934. An examination of the Degtjareff method for determining soil organic matter, and a proposed modification of the chromic acid titration method. Soil Sci.37:29-38

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3 Linked SOPs

SOP Code	SOP Function
EU_TRUE_SOP_025	Soil nutrient analyses

4 Disclaimer

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