

**Evaluation of Two
Field Methods to Estimate
Soil Organic Matter
In Alberta Soils**

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ABSTRACT

Soil quality can be briefly defined as how well a soil is able to function within an ecosystem. One way to assess soil quality is to use a quantitative method such as the United States Department of Agriculture-Agricultural Research Service (USDA-ARS) Soil Quality Test Kit. The kit measures various biological, physical, and chemical indicators but does not include a test for determining soil organic matter. The objectives of this study were to test two field methods for determining soil organic matter using soils from 41 sites across Alberta and to compare to measured soil organic matter and light fraction carbon obtained through conventional laboratory analyses. The first field method determines active carbon utilizing a deep purple solution of potassium permanganate, which is reduced to a lighter color when it reacts with soil organic carbon. This color change is quantitatively measured using a colorimeter. The second field method visually estimates soil organic matter and involves a solution of Basic Ethylenedinitro Tetraacetic Acid. The color of this solution is dependent upon the amount of soil organic matter in each sample. Results indicate that the method for determining active carbon showed a strong relationship between measured soil organic matter and active carbon ($R^2=0.82$), but a weak relationship between light fraction carbon and active carbon ($R^2=0.18$). A relationship was found between the visually estimated soil organic matter values and conventional laboratory results ($R^2=0.67$). Based on our findings both methods have the potential to become an integral part of the USDA-ARS Soil Quality Test Kit for measuring soil organic matter in Alberta.

1.0 INTRODUCTION

Soils are dynamic living systems whose quality is dependent on various attributes encompassing the physical, chemical and biological realms. The quality of a soil is best defined in relation to the functions it performs within natural or agro-ecosystems. The basic definition and a broader interpretation of soil quality is “the capacity of a specific kind of soil to function, within natural or managed ecosystem boundaries, to sustain plant and animal productivity, maintain or enhance water and air quality, and support human health and habitation” (Karlen et al. 1997).

Soils are the storehouses for water and nutrients, they regulate water flow, can be sources or sinks of carbon dioxide, and can store and degrade substances that can become pollutants. Therefore, soils have considerable direct and indirect impacts on water quality, the global climate and agricultural systems (NRC 1993), making the measurement of soil quality multifaceted.

Since soil quality cannot be determined by measuring only one parameter, it is necessary to rely on evaluation of a range of indicators. Indicators such as texture, permeability, depth, biological activity, the extent to which soil can store water and nutrients, and the amount of organic matter it contains are essential characteristics used to determine the quality of a soil (NRC 1993). An assessment of soil quality provides information about the functional status of a soil at a specific point in time (USDA 2001a). The information acquired from the evaluation can be used to help identify problem areas, areas of special interest, or to compare the effects of varying management systems.

Various methods are available to evaluate soil quality. One such method is the Alberta Soil Quality Card (AAFRD 2003), which is utilized in the field to qualitatively describe and measure farm level indicators. It is a non-technical procedure in which various indicators, such as drainage, crusting, and residue cover, are ranked either low, medium or preferred (unhealthy, impaired, or healthy) based on visual observations of the conditions in the field. No quantitative measurements are taken.

Another method to assess soil quality is the Soil Quality Test Kit (USDA 1999) developed by the United States Department of Agriculture-Agricultural Research Service (USDA-ARS). This easy to use, low-cost kit includes components that quantitatively measure the physical, chemical, and biological soil indicators *in situ*. These tests include bulk density, infiltration rate, aggregate stability, pH, electrical conductivity, and soil respiration. The indicators included in the kit are meant to provide quick results to determine differences in management systems, detect changes in soil quality over time, and diagnose possible soil problems. While the kit encompasses a variety of indicators to evaluate soil quality it leaves out a direct assessment of an essential indicator, soil organic matter. The kit utilizes proxy measures or derivative traits of soil organic matter like infiltration, aggregate stability and slaking.

Soil organic matter, or soil organic carbon (SOC) as it is often reported, is described as being the single most important indicator of soil quality and productivity (NRC 1993). It affects several critical soil functions including aggregation, infiltration, compaction, moisture holding capacity, resistance to erosion, bulk density, and nutrient retention (AAFRD 1985, USDA 2001b). Soil

organic matter is influenced by land management practices and it is vital in agricultural settings. Soil organic matter is composed of a variety of components. These include raw plant residues and microorganisms, well decomposed residues that are considered to be stable or resistant, often referred to as humus, and the active or labile portion (AAFRD 1985, Bowman and Peterson 1997). Measuring soil organic matter provides important information, however, because of the high proportion of recalcitrant carbon, slight changes in soil organic matter due to alterations in soil management are often difficult to detect. The active fraction of soil organic matter is the most highly influenced component and is most directly related to important biological processes in the soil (NRC 1993). Changes in this component may provide a better indication of management impacts on soil quality. The active carbon component consists of microbial biomass carbon, particulate organic matter, and soil carbohydrates (Weil et al. 2003). One way to measure the active carbon pool is to isolate the light fraction portion, which is derived primarily from plant residues but also includes significant amounts of microfaunal and microbial debris (Janzen et al. 1992).

Traditionally, the analysis of soil organic matter in analytical laboratories has been conducted by either dry combustion or wet digestion procedures (Carter 1993, Bowman et al. 1991). Until recently a method that is quick, accurate, inexpensive, and can be carried out in a field setting had not been developed. Two field methods have been proposed to attempt to meet the above criteria, one is a method developed by Weil et al. (2003) to determine the amount of active carbon and the other, developed by Bowman (1997), visually estimates the amount of soil organic carbon.

1.1 Background to Field Methodologies

In a study by Weil et al. (2003) a methodology was developed to test soil samples in the field for active carbon content (Appendix 1.1). The purpose of the study was to test and develop changes to methodology created by Blair et al. (1995) in order to generate a quantitative procedure that was simple, repeatable, and provided reliable results from which to base management decisions. The authors tested various aspects of the procedure outlined by Blair et al. (1995) including molarity of the potassium permanganate solution, shake time, soil-drying properties related to organic matter and tested the repeatability and reliability of the procedures by comparing their results to laboratory results. A 0.2M solution of potassium permanganate (KMnO_4) was used, as portions of the soil organic carbon will react with the KMnO_4 to reduce the deep purple color of the solution to a lighter shade depending on the amount of oxidizable carbon in the soil. Potassium permanganate is a good indicator and safe for use in the field. The color change of the KMnO_4 was measured by a hand-held colorimeter (generic 550 nm colorimeter, Hach[®] Company, Boulder, CO) (Figure 1).

Weil et al. (2003) determined that the procedure was easy to follow, repeatable, and suitable for use in the field as all the components could be readily transported and employed. The authors found the simplified methodology provided results that were similar to those obtained by using more complex laboratory procedures. Weil et al. (2003) concluded that the newly developed procedure was more sensitive to management effects and related to soil productivity and soil properties, such as respiration, aggregation, and microbial biomass, better than procedures based on measurements of total organic carbon.

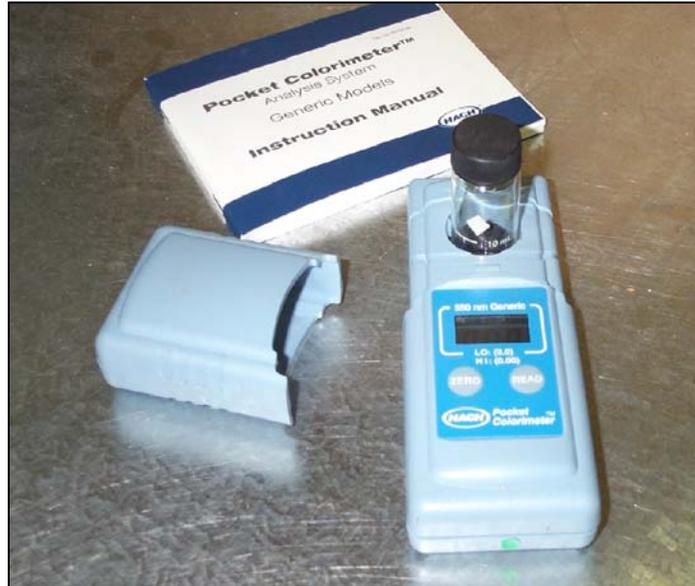


Figure 1. Hand-held colorimeter used for methodology of Weil et al. (2003)

A second methodology developed by the United States Department of Agriculture (USDA), estimates the amount of soil organic matter using a solution of Basic Ethylenedinitro Tetraacetic Acid, or EDTA (Bowman 1997). Basic EDTA is comprised of sodium hydroxide (NaOH) and EDTA disodium salt (Na_2EDTA) and is described as being relatively safe to handle. The NaOH works by solubilizing the organic carbon and the EDTA chelates metal cations to increase the efficiency of the soil organic matter extraction (Bowman and Moir 1993). The release of soil organic matter is directionally proportional to the intensity of the color of the filtrate. The protocol involves basic steps that are designed to be easy to follow and requires that standards are generated from soils in the general area of study with varying, but known degrees of soil organic matter content (Appendix 1.2). The method relies on qualitative visual comparisons of the colors of standards to the filtrate from each of the soil-EDTA solutions.

2.0 OBJECTIVES

This study has three main objectives:

- Test two field methods using soil samples from across Alberta which vary in management treatments, soil-landscape patterns, and are representative of the agricultural areas of Alberta
- Compare the results of the methodologies to standard laboratory analysis of the same soil to determine reliability
- Determine if the methodologies are realistic for use in the field (i.e. easy to use, applicable in various field conditions, repeatable and if either would be a reasonable addition to the USDA-ARS Soil Quality Test Kit for use in Alberta).

3.0 MATERIALS AND METHODS

3.1 Samples

Soil samples used for the purposes of this study were taken from 41 benchmark sites throughout Alberta. The benchmark sites (Leskiw et al. 2000, Cannon 2002) are located throughout the cultivated areas of Alberta and represent the soil-landscape patterns and agronomic practices of 41 ecodistricts in the province (Figure 2). The benchmark sites are unique in that they encompass a wide array of soil, climatic and vegetative zones. The management systems within the network of benchmark sites vary considerably and include continuous cropping and fallow systems, no-till and cultivation, and various crop rotations, including forages, and have a wide range of organic matter content.

Alberta Agriculture, Food and Rural Development (AAFRD) Regional Conservation teams collected the soil samples used for this study after harvesting had occurred in the fall of 2003, prior to fall fertilization and before freeze-up as part of the annual Soil Quality Benchmark soil sampling. Using DGPS equipment, the benchmark sampling locations are relocated each year. Five to ten cores within 2 meters of the marker at the specific landscape position were taken using a hand sampler. The cores were then bulked, mixed, bagged and labeled. Sub-samples from the upper (0-15 cm) depth of the mid-slope position were taken for this study. The samples were kept frozen until they could be analyzed (Appendix 2). Analysis of the samples for organic carbon was completed at Norwest Labs. The light fraction C analysis was done at the University of Alberta. Prior to analysis, the soils were air-dried and ground to <2 mm diameter (pass through a 20 mesh sieve).

Analysis of the samples for active C content and percent organic matter according to the field methods was completed at the AAFRD Bonaventure shop located in Edmonton, AB. The tests were performed in the main room at the shop and ran from January 13 to March 5 2004. Three replicates of each test were performed on 41 samples, for a total of 123 samples.

3.2 Active C Field Method

We tested specific aspects of the procedure followed by Weil et al. (2003) for variability and made modifications to the procedure based on our findings. These aspects included: readings of standard solutions, the ten-minute settling time, the effects of light and temperature on standard readings, the measurement of soil, and the drying time (Appendix 3).

The procedure we followed was nearly identical to that of Weil et al. (2003). Deviations from the protocol included: testing greater numbers of soils at one time, using 3.91 grams of soil instead of 5 grams, letting the soil-KMnO₄ solutions sit for a maximum of 17 minutes, and including a fourth standard solution of 0.015M. For a complete list of materials needed and the detailed protocol refer to Appendix 4.1 and 4.2. Calculations used to determine active C are shown in Appendix 4.3.

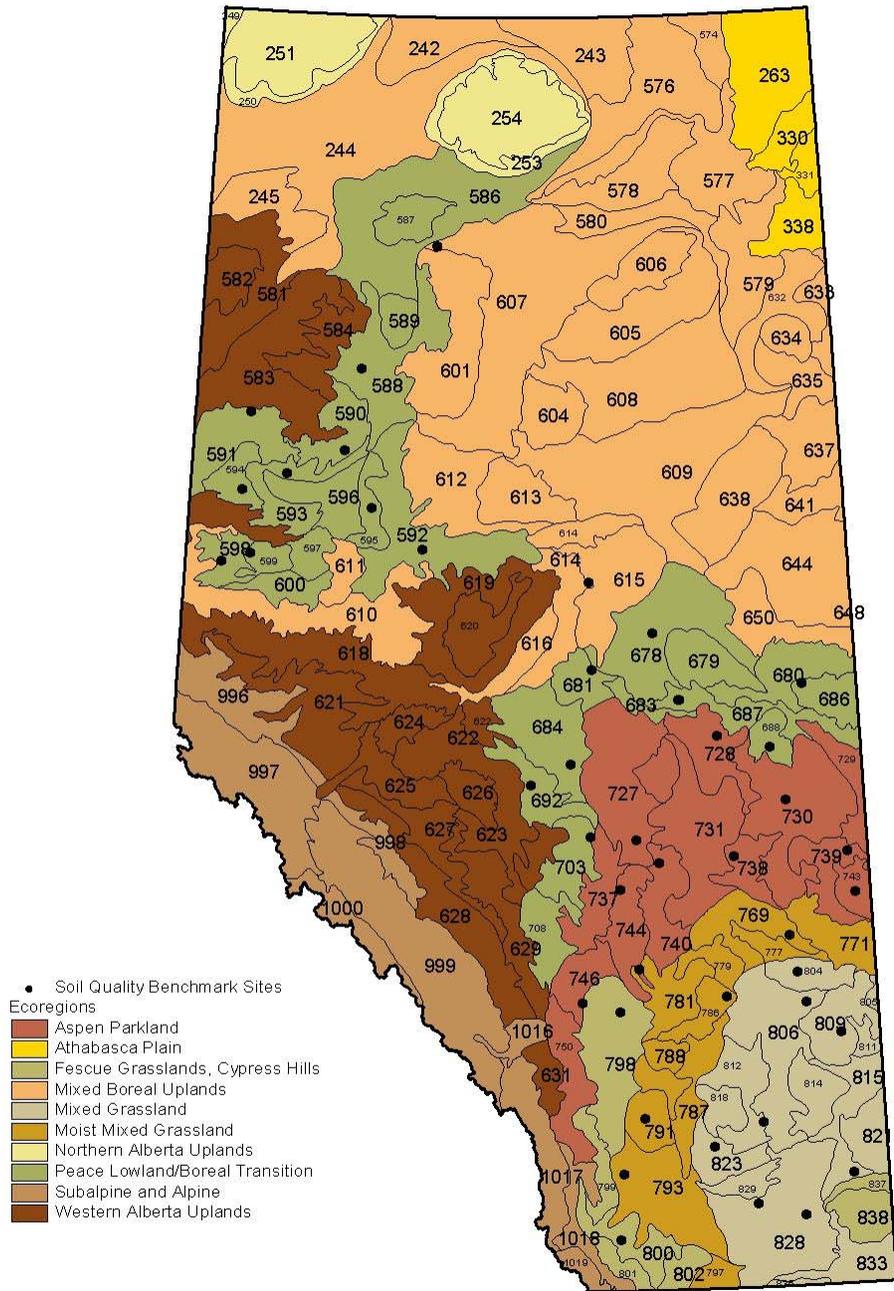


Figure 2. Location of benchmark sites and corresponding ecoregions and ecodistricts in Alberta

3.3 Basic EDTA Field Method

The protocol followed and materials used during this study varied only slightly from the procedure outlined in Bowman (1997). See Appendix 5.1 and 5.2. Samples to be used for the standards were chosen from the 41 benchmark soils. The percent organic matter values from 2001 to 2003 lab results were compared for all samples. Those soils with the values closest to the necessary standard values were selected (Appendix 5.3). We chose to increase the number of standards used for comparison. Instead of preparing four standards (<1%, 2%, 3%, and 4%), we prepared ten (0%, ~1.5%, 2%, 3%, 4%, 6%, 7%, 9%, 10%, and 12%) to encompass the range of organic matter values of the soil samples from Alberta. We also conducted a test to determine the accuracy of the measuring scoop used for this procedure (Appendix 5.4), like the one utilized to test the procedure of Weil et al. (2003).

4.0 RESULTS

4.1 Active C Field Method

An example of a characteristic standard curve based on the standard readings is shown in Figure 3. This standard curve is used to relate the absorbance readings to the amount of active carbon in each of the samples.

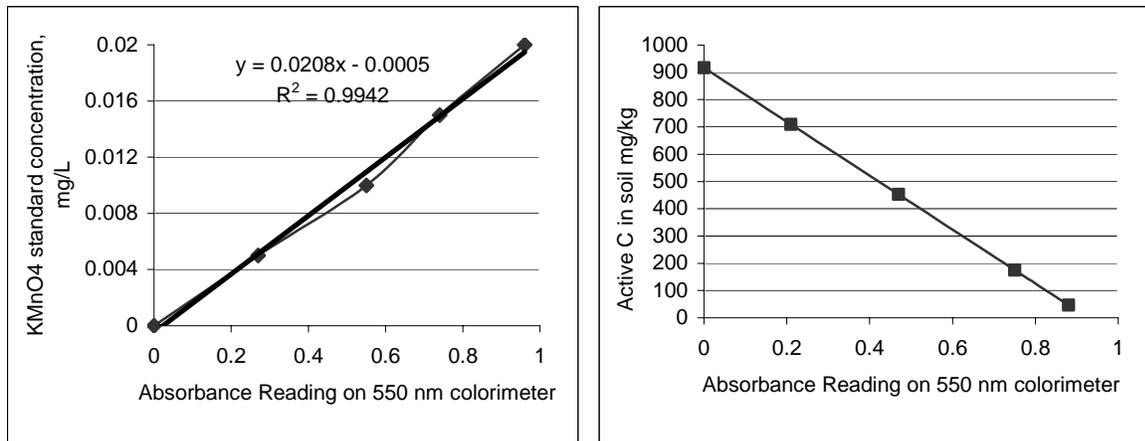


Figure 3. Characteristic standard curve generated using the 550 nm colorimeter (left). A sample of the use of the standard curve to relate absorbance readings to active C in the soil sample (right).

Descriptive statistics were calculated for each of the 41 benchmark sites active C values (Appendix 6). Active C values ranged from 194 to 964 mg carbon per kg soil. Of the 41 sites included in this study 17 had high variances of 1000 or more. Fourteen of these sites were located within the mixed grassland and Peace Lowland/Boreal transition ecoregion and the remaining three occurred in the Athabasca Plain ecoregion.

We compared the estimated active C obtained by the field method of Weil et al. (2003) to the light fraction carbon (LFC) determined by the University of Alberta (Figure 4).

Although there was a positive correlation between the amount of LFC and active C in the samples, the two factors were not closely related ($R^2=0.18$).

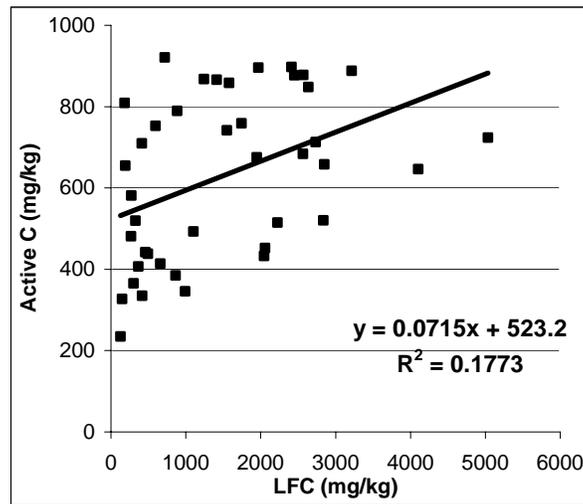


Figure 4. Relationship between active C (mg/kg) and LFC (mg/kg).

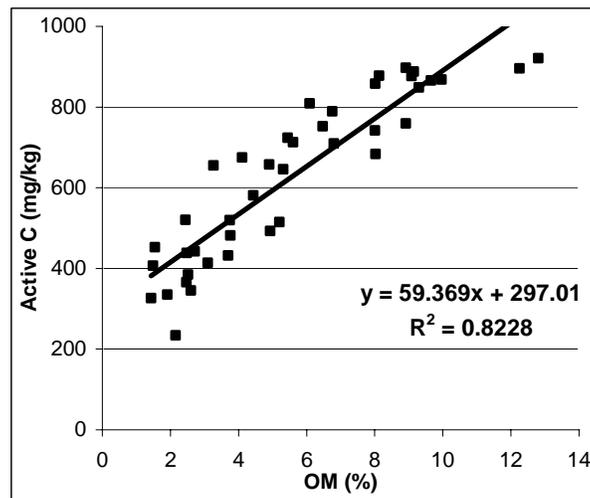


Figure 5. Relationship between active C (mg/kg) and organic matter (%).

We then compared the OM% values obtained from Norwest Labs to the estimated active C measured (Figure 5). The linear association between the percent of organic matter and active C was positive and had a high correlation ($R^2=0.82$). It appears that there may be a ceiling effect occurring for soils having higher values of organic matter determined by the lab since none exceed 964 mg/kg active carbon. However, further investigation is necessary to verify the relationship between active carbon and organic matter and between active carbon and light fraction carbon.

We expected to see a stronger relationship between the active C and the amount of light fraction carbon as both measure the active or labile fractions of soil organic matter. However, the strong correlation between the active C and the percent organic matter suggests that this method could be used in the field to successfully determine the amount of soil organic matter.

4.2 Basic EDTA Field Method

The descriptive statistics were calculated for the estimated organic matter for each of the 41 benchmark sites (Appendix 7). The values ranged from a low of 1.5% organic matter to a high of 12% organic matter. The variance between reps reached a high of 3%, and occurred in samples in which it was necessary to differentiate between 6% and 9% organic matter. We found it easy to distinguish up to the 6% standard, but after this it was more difficult.

The correlation of estimated organic matter (%), by the method of Bowman (1997), and the lab percent organic matter (Figure 6) was positive and indicated that the two values were related ($R^2=0.67$).

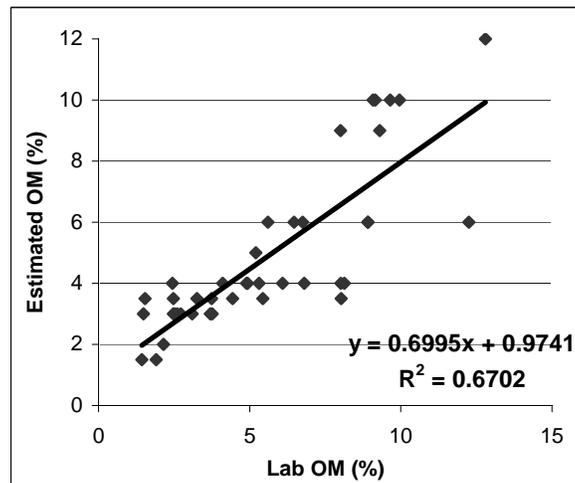


Figure 6. Correlation of lab OM (%) values and estimated OM (%)

To determine the ability of the test to distinguish between higher and lower values of organic matter we separated the data into two groups based on lab values over or under 6% organic matter. We found that the correlation was higher ($R^2=0.52$) when differentiating between 0% and 6% organic matter (Figure 7) and then decreased ($R^2=0.37$) when attempting to differentiate between the higher levels of organic matter (Figure 8).

This corresponded to our observations while carrying out the procedure. The standards created for the purposes of comparison were easy to distinguish at 0, ~1.5, 2, 3, 4 and 6%. However, when the standards were created for the range of 6 to 12% it was harder to visually discern the filtrates from each other (Figure 9).

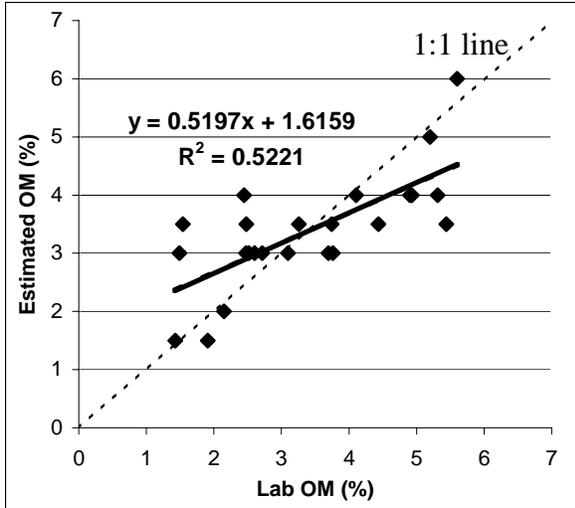


Figure 7. Correlation of known lab OM (%) values under 6% and estimated OM (%)

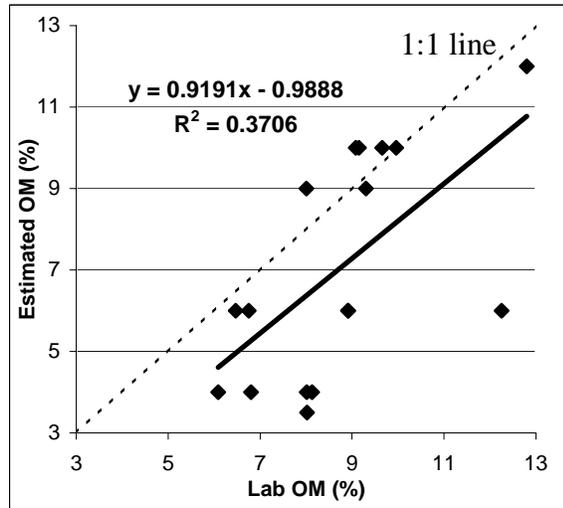


Figure 8. Correlation of known lab OM (%) values over 6% and estimated OM (%)

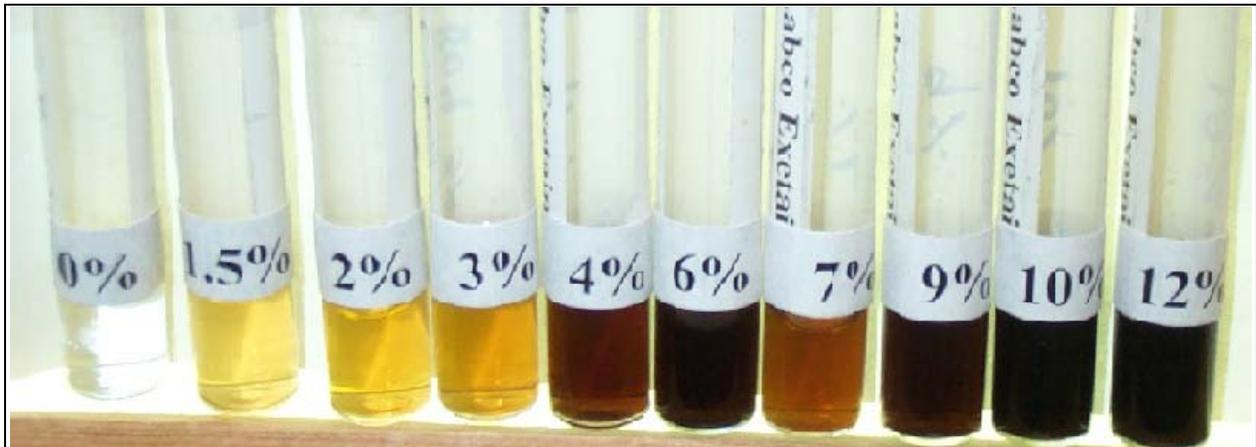


Figure 9. Organic matter standards for Basic EDTA method

The standards for 6 and 9% were impossible to distinguish from each other and this was the same for the standards of 10 and 12%. There was a marked visual difference between the 9% standard and the 10% standards. In creating the 7% standard we found that the filtrate was lighter than the 6 and 4% standard, and darker than the 3%. To deal with this anomaly we recorded any filtrate that visually matched this color as 3.5% organic matter to correspond with the gradient of filtrate occurring between 0 and 6%.

5.0 DISCUSSION

The objectives outlined by Weil et al. (2003) included developing a method that was simple, quick, reproducible, and safe. In Bowman (1997) these same objectives are reiterated, along

with meeting the goals of cost effectiveness and accuracy. After testing the two methodologies on soils found throughout Alberta we now must determine if either has met any or all of the proposed goals.

5.1. Active C Field Method

The methodology described by Weil et al. (2003) was easy to follow. Creating the standard solutions involved calculations to determine the quantity of chemical needed to ensure the correct molarity. To make up the standard curves and determine the amount of active carbon in the samples required the use of a method of least squares means (Mendenhall 1983). Both these aspects required referring to information sources outside of the directions provided by the authors, which may not be readily available to others. In order to make this process as simple as possible for the potential user it would be preferred to provide the molarity calculation information within the procedure or provide the solutions already prepared. Weil et al. (2003) found that the standard solutions maintained consistent concentrations when retested after three years. Similarly, we found no statistically significant difference between the readings of the standard solution after a two-month period to readings obtained at the beginning of the study. Therefore, using a generalized standard curve that relates the absorbance readings to the amount of active C would clarify this process even further. This would eliminate both the need to keep standard solutions in the kit and concerns regarding maintaining standard solutions and supplying them to users.

Due to the large quantity of samples in our study the process took longer than would be expected for someone employing this technique in the field with minimal samples to process. Weil et al. (2003) required that their samples be dried for 15 minutes and then analyzed. However, when comparing all samples it is preferred that the samples are of equal dryness in order to achieve minimum variability (Weil et al. 2003). If this method were to be included in the Soil Quality Test Kit for use in Alberta it is recommended that the samples be air dried for 24 hours or more to achieve minimum variability. This longer drying time may be a more convenient option for the user as the respiration and aggregate stability tests included in the kit already require a 16-24 hour waiting period. Therefore, allowing the soil to dry for a minimum of 24 hours and then proceeding with analysis would not increase the time necessary to complete the tests beyond what is already required. After the ten-minute settling time had elapsed the processing of each sample went very quickly, taking an average of 1 minute per sample. Being able to analyze more than one sample at once is an advantage of this procedure.

All of the chemicals used in the procedure were relatively safe to use. It was necessary to wear safety glasses and use gloves to protect hands from the KMnO_4 solution, which can be an irritant to skin. Since the solution permanently stains clothing brown it is also recommended that coveralls be worn.

In terms of cost effectiveness, this procedure requires the use of a colorimeter. A hand-held colorimeter costs \$650 (Cdn). This together with the cost of the Soil Quality Test Kit (\$500 U.S.) is a major hindrance to including this procedure in the Soil Quality Test Kit.

There are numerous factors to consider when following the procedure of Weil et al. (2003). As we showed in our tests of various methodologies, including settling time, weight of soil used,

and dryness of the soil; all affect the accuracy of the results and introduce variability. Although we were able to ensure that dryness and settling time were not an issue, the weight of the soil was still a factor (Appendix 3.5). The coefficient of variation varied from 2.53 to 9.72 and affects the outcome for the calculated active C values (Table 4). Therefore, it would be best to provide a standardized measuring scoop to all users in order to decrease variability caused by differences in the amount of soil used.

Our results indicate that this method was not able to accurately determine the amount of light fraction carbon in the soils of Alberta. However, it was able to predict the amount of soil organic carbon very well. It was a quantitative procedure that reduced the bias of the user as compared to the qualitative procedure proposed by Bowman (1997).

5.2. Basic EDTA Field Method

The method outlined by Bowman (1997) was simple to follow. The procedure involved making the Basic EDTA reagent, which was a straightforward combination of two chemicals. No calculations were necessary in order to determine the amount of organic carbon. Acquiring the soils with known standard values is a downfall to this protocol. We were able to easily create the standard filtrates because the samples had been analyzed in a lab prior to this study to determine their organic matter contents. However, those employing the method created by Bowman (1997) would not have the same advantage. Therefore, if this method were to be included in the kit an extensive amount of soil samples and information regarding the organic matter contents of soils throughout Alberta would have to be acquired. Providing standard filtrates to the user that accurately represent the area of study would be a costly and time-consuming process. It is also not known how long the filtrates would remain stable and if the color would degrade over time. In order to overcome these issues it may be possible to use Munsell color chips instead of filtrates to represent the standards for estimating soil organic matter. The unknown filtrates could then be compared to the color chips, avoiding issues of degradation and supplying standard solutions to users.

Creating the standards and the sample filtrates took up to 1 hour to analyze 21 samples (7 samples x 3 reps). The longest part of the procedure is the filtration process. For some soils, especially those higher in clay content, filtering took up to 30 minutes in order to collect enough filtrate to compare to the standards. Like the procedure of Weil et al. (2003) we found that more than one sample can be analyzed at once which speeds up the process.

This procedure was safe to perform and, like the active C method, the only safety requirements were gloves and safety goggles.

This method did not require the utilization of expensive equipment. Costs to provide glass vials, tubes, a mortar and pestle, and various other items in the Soil Quality Test Kit would be minimal.

The Basic EDTA method was developed in the Central Great Plains region of the United States (Bowman and Peterson 1997). Here, the soil organic matter ranges from 1 to 6% with soils typically ranging between 1 and 2% for cultivated soils and between 1.5 and 3% for native grassland. This differs from soils in Alberta where soil organic matter can range from 1-10% for virgin soils (AAFRD 1985) and between 1 and 10% for cultivated soils within the Soil Quality

Benchmark sites (Leskiw et al. 2000). Applying the procedure of Bowman (1997) to Alberta soils allowed us to determine how well the test could distinguish between higher levels of organic matter.

When creating the standards we found that it was difficult to distinguish between levels of organic matter greater than 6%. A visual difference could be detected between the 6% and 9% standards and the 10% and 12% standards by holding the glass vials up to a light source. However, distinguishing between 6% and 9% was very difficult and the determination of a sample as 9% only occurred when it was identified as being lighter than 10%, yet darker than 6%. Based on these findings the methodology may have a limited application in Alberta. However, a high degree of accuracy over a 6% organic matter level may not be necessary as soils higher in organic matter (8%) are not fundamentally more fertile or productive than those with less organic matter (5%) (AAFRD 1985).

When creating the 7% standard we found that the color of the filtrate did not correspond to the color gradient of the other standards. It is not known whether the texture or color of the soil affected the resulting filtrate, however, if either of these variables caused the anomaly then the reliability of this test would be reduced.

This method resulted in a reasonably strong relationship between the estimated organic matter and the laboratory determined organic matter values. As the need for determination of higher levels of organic matter may not be necessary this method provides an acceptable level of accuracy for the purposes of the field kit. This procedure is qualitative, but the clear distinction between the standards ranging from 0-6% reduces the user bias.

6.0 CONCLUSIONS

The AESA Soil Quality Program tested two field methods for determining soil organic matter using soils from across Alberta. The objectives of this study were to test the methods proposed by Weil et al. (2003) and Bowman (1997) on Alberta soils to determine the reliability of the procedures by comparing them to results obtained through lab analyses and to determine if either method would make a significant contribution to the Soil Quality Test Kit.

We found that the method developed by Weil et al. (2003) was able to accurately detect soil organic matter when compared to lab results. However, the results exhibited a weak relationship between light fraction carbon and active carbon. The procedure outlined by Bowman (1997) provided results showing a reasonably strong relationship between the soil organic matter values obtained through lab analysis and the estimated soil organic matter values.

Based on the findings of this study either field method could potentially make a positive contribution to the USDA-ARS Soil Quality Test Kit for use in Alberta if used by those familiar with soil analyses. For those unfamiliar with procedures related to soil analyses, both field methods required the use of materials, including chemicals and measuring devices that could not easily be utilized without the aide of an experienced individual. Even with the chemicals and calculations provided, the amount of materials and background knowledge needed for each test may be daunting to an inexperienced user.

Therefore, both procedures would need to be adapted to ensure that the methodologies are as simplified as possible. The Weil et al. (2003) method to determine active C would need to be altered so that a generalized standard curve would be provided to the user to eliminate extensive calculations. The method proposed by Bowman (1997) would need to be changed so that standards are provided. Both methods involve making chemicals, which could introduce inconsistency if measured incorrectly or could be a discouraging aspect for the user. To eliminate these concerns it would be best to provide the user with the necessary chemical already mixed. Pre-prepared solutions also reduce the number of materials necessary to carry out both procedures in the field, increasing the ease of operations. Although the method to determine active C was more costly, it provided accurate results when measuring soil organic matter ($R^2=0.82$) and was quantitative, which reduces user bias. The qualitative basic EDTA method was less expensive and, although not as accurate as the active C method, still provided results consistent with those of the lab ($R^2=0.67$).

Both field methods have the capability to become an integral part of the USDA-ARS Soil Quality Test Kit for use in Alberta to measure soil organic matter. Further testing and modifications of both methods are necessary to ensure that they are straightforward and effective for the potential user.

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8.0 APPENDICES

Appendix 1. Methodologies for Field Tests

Appendix 1.1 Active C Methodology

In their study, Weil et al. (2003) used a small subsample (approximately 20 grams) of field moist soil which was crumbled gently onto a piece of black paper and left to air-dry, preferably in direct sunlight, for 15 minutes.

A stock solution consisting of 0.2M KMnO_4 adjusted to a pH of 7.2 using NaOH was prepared, along with three standard solutions. The standard solutions consisted of the 0.02M KMnO_4 diluted using three volumes of distilled water (1.25, 2.50, and 5.00 ml) to produce three standards with molarities of 0.005, 0.01, and 0.02, respectively. Standard solutions were made in order to create a standard curve based on the absorbance readings from the 550 nm colorimeter. This curve was then used to determine the amount of active C in the soil- KMnO_4 solutions.

Once the soil was dry, 2.0 ml of the 0.2M KMnO_4 was placed in a centrifuge tube using a bulb pipette and distilled water added to the 20 ml mark on the tube. The tube was shaken to ensure the solution was mixed thoroughly. One level scoop (approximately 5 grams) was then placed in the solution and the tube capped and shaken vigorously for two minutes.

The tube was left to sit for a ten minute settling period, during which time the standard solutions could be tested for absorbance using the colorimeter. This involved diluting 0.05 ml of each standard in a centrifuge tube containing 50 ml of distilled water. Approximately 15 ml of the diluted standard was then put in the glass cuvette. The outside of the cuvette was wiped to ensure no particles interfered with the absorbance reading and the cuvette was placed in the colorimeter well. The reading was recorded and a standard curve was created.

After the ten minute settling time, 45 ml of distilled water was added to a clean centrifuge tube. Using a bulb pipette, 0.05 ml from the test sample was added to the tube containing 45 ml of distilled water. Distilled water was then added to the 50 ml mark and the tube capped and shaken. Fifteen milliliters of this solution was then transferred to the glass cuvette. The cuvette was wiped down and placed in the colorimeter well and the absorbance recorded.

Appendix 1.2 Basic EDTA Methodology

In this study 5 to 10 grams of dry field sample is placed into a mortar and pestle and pulverized (Bowman 1997). One scoop (approximately 0.5 g) of each soil sample to be used for the standards and one scoop of each of the unknown samples are then placed in separate labeled vials or tubes. This procedure requires that four soils with known soil organic matter (SOM) contents of <1%, 2%, 3%, and >4% be used as the standards. Twenty milliliters of the basic EDTA, consisting of equal parts NaOH and Na_2EDTA , is added to each of the containers and the mixture is shaken vigorously for 30 seconds. A clean vial is then prepared by placing a funnel lined with filter paper on top and the mixture of soil and basic EDTA is transferred to the funnel. The color of the clear filtrate that results from the unknown samples is then compared with the color of the known standards and the estimated percent SOM recorded.

Appendix 2. Lab Procedures for LFC and OM (%)

The Norwest lab values for the percent organic matter were obtained using a loss on ignition method (McKeague 1978).

The University of Alberta lab values for the light fraction carbon (LFC) were obtained using a loss on ignition method (University of Alberta 2002). The carbon and nitrogen content of the light fraction material was obtained through a dry combustion process using Carlo Erba instrumentation.

Calculation of the LFC (mg/kg) was then completed as outlined below (Figure 10).

Example of Calculation of Light Fraction Carbon			
Used data from B.H. Prairie, Site #586			
From Lab:			
LF (Wt) g = 0.1776 per 20 grams soil			
% Carbon as LF material = 28.918			
Therefore:			
$\frac{0.1776 \text{ g LF}}{20 \text{ grams soil}}$	x	$\frac{28.918\% \text{ Carbon as LF}}{100}$	equals $\frac{0.00256792 \text{ g LFC}}{1 \text{ g soil}}$
To convert to mg/kg:			
$\frac{0.00256792 \text{ g LFC}}{1 \text{ g soil}}$	x	$\frac{1000 \text{ mg}}{1 \text{ g}}$	x $\frac{1000 \text{ g}}{1 \text{ kg}}$ equals $\frac{2567.92 \text{ mg LFC}}{\text{kg soil}}$

Figure 10. Calculation of light fraction carbon (LFC)

Appendix 3. Tests for Variability in Methodology of Weil et al. (2003)

Appendix 3.1 Comparison of Standard Readings in Morning and Afternoon

There was a statistically significant difference between the readings of the 0.005M standard from the morning to the afternoon (Table 1). Therefore, in order to decrease the variability that might result from preparing and reading the standards once a day we decided to prepare the standard solutions twice a day, in the morning and afternoon. We then applied the resulting standard curve to those samples tested at the corresponding time period, which allowed us to test a greater quantity of samples per day. This differed from the procedure followed by Weil et al. (2003) who completed readings of the standards for each batch of three samples.

Table 1. The Descriptive Statistics and Results of Paired T-test for Standard Readings

Standard (M)	Morning Reading			Afternoon Reading		
	Mean	Variance	Standard Deviation	Mean	Variance	Standard Deviation
0.005*	0.22*	0.0004	0.019	0.30	0.0005	0.022
0.010	0.46	0.0002	0.015	0.50	0.0032	0.056
0.015	0.71	0.0022	0.047	0.70	0.0020	0.045
0.020	0.87	0.0034	0.058	0.94	0.0045	0.067

* significant difference at $p < 0.05$ between morning and afternoon readings

This was based on five readings, in both the morning and afternoon, for the 0.005, 0.01 and 0.05M standards and three readings for the 0.015M standard.

Appendix 3.2. Settling Time

To test the effects of settling time on the readings we used 12 soil specimens from samples taken during fall 2002 Soil Quality benchmark sampling, which varied across textural classes and management practices. We prepared the samples according to the protocol followed by Weil et al. (2003) and obtained the absorbance readings after five time periods, 10, 15, 20, 30 and 60 minutes, had elapsed. We found there to be a statistically significant difference in the readings taken at 10 minutes to those taken at 20, 30 and 60 minutes (Table 2). Therefore, in order to maximize the number of samples that could be tested at one time and to keep variability as low as possible, we decided that no more than 10 samples could be analyzed at once to ensure none exceeded a 17 minute settling time.

Table 2. Absorbance Readings and Results of Paired T-tests on 2002 Soils Across Varying Textural Classes

Sample ID	Reading at:				
	10 min	15 min	20 min*	30 min*	1 hr*
116	0.46	0.47	0.46	0.45	0.38
20	0.34	0.31	0.29	0.28	0.23
72	0.02	0.02	0.02	0.01	0.01
9	0.12	0.11	0.12	0.11	0.08
59	0.05	0.05	0.04	0.04	0.04
32	0.25	0.24	0.21	0.19	0.18
80	0.02	0.05	0.03	0.03	0.03
114	0.66	0.6	0.6	0.61	0.49
66	0.25	0.26	0.23	0.21	0.17
92	0.45	0.44	0.41	0.44	0.38
132	0.57	0.51	0.52	0.49	0.49
52	0.53	0.52	0.51	0.44	0.46

* significant difference at $p < 0.05$ when compared to readings taken

Appendix 3.3. Light

Exposure of the KMnO_4 solution to light can increase the rate of decomposition (Weil et al. 2003). We tested the readings of standard solutions at various locations of differing light exposure at Bonaventure and found no statistically significant differences between locations (Table 3).

Table 3. Readings of Standards at Various Locations

Date 1/14/2004		Readings of standards			Means		
Location	Rep Number	0.005M	0.01M	0.02M	0.005M	0.01M	0.02M
Coolers	1	0.24	0.49	0.84	0.26	0.51	0.81
	2	0.26	0.51	0.76			
	3	0.27	0.53	0.83			
Grinders	1	0.25	0.52	0.86	0.25	0.54	0.80
	2	0.26	0.53	0.81			
	3	0.24	0.57	0.73			
Scale bench	1	0.24	0.46	0.9	0.24	0.50	0.87
	2	0.23	0.52	0.85			
	3	0.24	0.53	0.85			
In direct sun	1	0.24	0.46	0.96	0.24	0.49	0.91
	2	0.25	0.51	0.93			
	3	0.24	0.49	0.85			
Range of readings		0.23-0.27	0.46-0.57	0.73-0.96			

Appendix 3.4. Temperature

We recorded the temperature of the shop once in the morning and again in the afternoon. We found that it was generally cooler in the morning than the afternoon. The average ambient temperature was 20.17 °C and the temperature ranged by 1.8 °C.

Appendix 3.5. Accuracy of Measuring Scoop

The protocol followed by Weil et al. (2003) uses a scoop to measure out the five grams of soil required for testing. To assess the accuracy of our measuring scoop to this requirement we weighed 11 soil samples with varying soil properties using a metal five-milliliter scoop.

We ran the test according to the following procedure:

- 1) Take the scoop and place on scale, tare scale.
- 2) Take scoop of dry soil and level.
- 3) Place scoop with soil on scale and weigh, record the weight of the soil.
- 4) Take soil that was in scoop and place in separate container.
- 5) Repeat steps 1-4 four more times for the sample.
- 6) After one sample has been weighed 5 times clean off the scoop and re-weigh the scoop and tare it again if needed.
- 7) Proceed to the other samples repeating steps 1-6.

We found that the scoop weights of the soils ranged from 3.08 grams to 5.07 grams and the mean weight of all the soil measured using the scoop was 3.91 grams (Table 4). In order to decrease the variability caused by the inaccuracy of the scoop we substituted the value of 0.005 kg with 0.00391 kg into the equation for calculating the active C (mg/kg) (Appendix 4.3).

Table 4. Results of Measuring Accuracy Test for Weil et al. (2003) Procedure

Sample I.D	Rep Number					Mean (g)	Range (g)	Variance	Standard Deviation	Coefficient of Variation	
	1	2	3	4	5						
DAPP	3.22	3.67	3.48	3.63	4.14	3.63	0.92	0.11	0.34	9.27	
Carstairs	3.08	3.14	3.24	3.28	3.22	3.19	0.2	0.01	0.08	2.53	
Youngstown	5.02	4.85	4.89	5.07	4.75	4.92	0.32	0.02	0.13	2.64	
Tilley	4.71	4.45	4.4	4.25	4.57	4.48	0.46	0.03	0.17	3.89	
Enchant	3.83	3.83	4.17	4.1	3.78	3.94	0.39	0.03	0.18	4.54	
Boyle	3.43	3.52	3.32	3.43	3.19	3.38	0.33	0.02	0.13	3.75	
Beiseker	3.57	4.11	4.11	3.96	4.34	4.02	0.77	0.08	0.28	7.09	
Dunmore	3.56	3.7	4.07	3.95	4.46	3.95	0.9	0.12	0.35	8.86	
Warspite	4.02	3.99	4.22	3.89	3.56	3.94	0.46	0.06	0.24	6.15	
Lacombe	3.41	3.5	3.59	3.62	4.14	3.65	0.73	0.08	0.28	7.80	
Etzicom	4.21	4.25	4.2	3.37	3.76	3.96	0.88	0.15	0.38	9.72	
Overall Mean						3.91					

Appendix 3.6. Drying Times

Our study utilized 41 field moist samples taken from AESA Soil Quality benchmark sites across Alberta. We decided to test the effects of air-drying all the samples for a period of 24 hours (or until all samples were of equal dryness) as compared to drying the field moist samples for the 15-minute drying time suggested by Weil et al. (2003).

A sub-sample (5 scoops) of each of the 11 moist soil samples was taken and crumbled gently and allowed to dry in an aluminum tray. The sample was evenly spread out in the tray and left to dry until all samples were of equal dryness. The samples were then tested according to the procedure outlined in Appendix 4.2 and replicated three times.

Meanwhile, another sub-sample of 5 scoops was taken from the 11 samples. Each of these was crumbled gently and laid out to dry on a black rubber mat. The samples were spread out evenly and thinly and placed in direct sunlight when possible. The soils were allowed to dry for 15 minutes and were mixed three times to help ensure that they dried as evenly as possible. When the drying time was up the samples were tested based on the procedures in Appendix 4.2 and replicated three times.

We found that in four of the 11 sites there was a statistically significant difference between soils dried for 15 minutes and those dried for 24 hours (Table 5). Higher standard deviations, standard errors and variances were recorded for soils dried for 15 minutes, with the exception of one site. In order to minimize this variability we decided to dry the soils for 24 hours.

Table 5. Readings and Descriptive Statistics of 24 hr and 15-Minute Dried Soils and the Results of a T-test

Site Name	Readings		15 Minute Drying Time				24 Hour Drying Time			
	15 min	24 hrs	Mean	Standard error	Standard deviation	Variance	Mean	Standard error	Standard Deviation	Variance
Dapp	0.46	0.28	0.48	0.0841	0.1457	0.0212	0.32	0.0203	0.0351	0.0012
	0.34	0.35								
	0.63	0.32								
* Carstairs	0.32	0.08	0.31	0.0291	0.0503	0.0025	0.07	0.0186	0.0321	0.0010
	0.26	0.09								
	0.36	0.03								
Youngstown	0.51	0.52	0.58	0.0636	0.1102	0.0121	0.57	0.0404	0.0700	0.0049
	0.53	0.54								
	0.71	0.65								
Tilley	0.63	0.57	0.62	0.0186	0.0321	0.0010	0.67	0.0491	0.0850	0.0072
	0.58	0.7								
	0.64	0.73								
* Enchant	0.61	0.45	0.65	0.0633	0.1097	0.0120	0.44	0.0296	0.0513	0.0026
	0.56	0.48								
	0.77	0.38								
* Boyle	0.48	0.36	0.52	0.0536	0.0929	0.0086	0.34	0.0252	0.0436	0.0019
	0.46	0.37								
	0.63	0.29								
Beiseker	0.37	0.33	0.47	0.0612	0.1060	0.0112	0.33	0.0376	0.0651	0.0042
	0.45	0.39								
	0.58	0.26								
Dunmore	0.54	0.52	0.68	0.0872	0.1510	0.0228	0.57	0.0677	0.1172	0.0137
	0.66	0.7								
	0.84	0.48								
Warspite	0.38	0.41	0.55	0.1041	0.1804	0.0325	0.42	0.0406	0.0702	0.0049
	0.54	0.49								
	0.74	0.35								

* indicates a statistically significant difference between the 15 minute and 24 hour dried soils at $p < 0.05$

Table 5. cont'd.

Site Name	Readings		15 Minute Drying Time				24 Hour Drying Time			
	15 min	24 hrs	Mean	Standard error	Standard deviation	Variance	Mean	Standard error	Standard Deviation	Variance
* Lacombe	0.24	0.14	0.28	0.0219	0.0379	0.0014	0.13	0.0176	0.0306	0.0009
	0.31	0.16								
	0.3	0.1								
Etzikom-Irrig	0.44	0.47	0.59	0.0801	0.1387	0.0192	0.47	0.0549	0.0950	0.0090
	0.63	0.57								
	0.71	0.38								

* indicates a statistically significant difference between the 15 minute and 24 hour dried soils at $p < 0.05$

Appendix 4. Procedure for Active C Test

Appendix 4.1. List of Materials Needed for Active C Test

Materials for Active C Test	
Aluminum trays to dry soil samples	Metal scoop to level off measuring spoon
Large brown glass bottle (>1L)	Timer
3 smaller brown glass bottles	Centrifuge tubes
pH meter	Lab tissues
Small scoop (5 ml aluminum measuring spoon)	Plastic basin
Weigh paper	2 glass cuvettes
CaCl ₂ (500g container)	10 % bleach solution
550 nm colorimeter	Labels
KMnO ₄ (500 g container)	Distilled water
0.1M NaOH (1 liter bottle)	Plastic cup
Bulb pipettes	Stir stick
Funnel	Graduated cylinder
Felt marker	

Appendix 4.2. Revised Protocol for Active C Field Method

Mixing the stock solution:

- 1) Combine 147.01 grams CaCl₂*2H₂O into 1 L distilled water in a dark glass bottle
- 2) Add 31.6 grams KMnO₄ to mixture described above
- 3) Add small amounts of NaOH with a bulb pipette and after each addition test the solution until a pH of 7.2 is reached (between 5 and 10 drops in 1 L of solution, test after each drop).

To test pH we immersed the pH meter in the solution corresponding to the basic or acidic reading needed and then rinsed the meter with distilled water. We placed approximately 15 ml of the stock solution in a plastic cup and tested the pH. If the ideal pH had not been reached we rinsed the pH meter and the cup with distilled water. We then added another drop of the NaOH solution and re-tested until the desired pH was reached.

- 4) Place a label on the bottle and using a marker, note the contents for safety.

The standard solutions were prepared two times a day, in the morning and afternoon. This procedure differs from that of Weil et al. (2003) in that we added a fourth standard solution, 0.015M, in order to create a more accurate standard curve.

- 1) Take 1.25 ml (29 drops) of the stock solution and place it in a clean brown bottle. Dilute the solution to the 50 ml mark with distilled water, using the graduated cylinder to measure. Label the bottle 0.005M.
- 2) Take 2.5 ml (58 drops) of the stock solution and place in clean brown bottle. Dilute the solution to the 50 ml mark with distilled water, using the graduated cylinder to measure. Label the bottle 0.01M.

- 3) Take 3.75 ml (86 drops) of the stock solution and place in clean brown bottle. Dilute the solution to the 50 ml mark with distilled water, using the graduated cylinder to measure. Label the bottle 0.015M.
- 4) Take 5 ml (115 drops) of the stock solution and place in a clean brown bottle. Dilute the solution to the 50 ml mark with distilled water, using the graduated cylinder to measure. Label the bottle 0.02M.
- 5)
 - a) Fill a clean glass cuvette with distilled water. Wipe the outside of the vial with a tissue. Place vial in the colorimeter (generic 550 nm colorimeter, Hach[®] Company, Boulder, CO) well and put the cover in place. Press the zero button and after a few seconds the LED should read 0.00. Remove the cuvette and rinse it with distilled water (If the reading is higher after 10 to 20 determinations clean the cuvette with a 10% bleach solution).
 - b) Add 49.5 ml of distilled water to a *clean* centrifuge tube. Using the disposable bulb pipette specifically for the standard solution of 0.005M KMnO₄ place 0.5 ml (12 drops) of the standard into the tube. Place the cap on the tube and shake for a count of 15.
 - c) Pour approximately 15 ml of the diluted standard into a 20 ml glass cuvette, filling the cuvette and emptying it three times with the diluted standard, wipe the outside with a tissue and place in colorimeter well. Put the cover in place and press the read button. Record the absorbance displayed.
 - d) Rinse the cuvette thoroughly with distilled water
 - e) Repeat steps 5 (a-d) using 0.5 ml of the 0.01M, 0.015M, and 0.02M standard solutions. Record the absorbance for each standard solution.
- 6) Create standard curve using readings.

To create the standard curves we first plotted the absorbance reading for each of the standard solutions. Using the method of Least Squares (Mendenhall 1983) we obtained the standard curve equation (Appendix 4.3). We then used these results in the equation provided by Weil et al. (2003) to calculate the active C content of all soil samples (Active C (mg/kg) = (0.02Mol/L – (a+b*absorbance))*(9000mg C/Mol)*(0.02L soln/0.005kg soil). Please note that based on our findings we used 0.00391 kg soil in place of the 0.005 kg used in the above equation by Weil et al. (2003).

To test the soil samples we followed the procedure described below:

- 1) Soil samples were laid out to air dry in aluminum tins for a period of 24 hours. After this time each sample was checked to ensure it was dried thoroughly. If any sample was still wet it was left to dry for longer.
- 2) Before the tests were begun the temperature of the lab was taken in the morning and afternoon using a thermometer (Checktemp-1 pocket thermometer, Hanna[®] instruments, Laval, Quebec) and recorded.
- 3) Using a pipette place 2.0 ml (46 drops) of the stock solution in a *clean* 50 ml centrifuge tube. Add 20 ml of distilled water to the tube, using the graduated cylinder to measure the correct amount of water. Cap the tube, and using a sharpie mark the site name on the lid. Swirl the tube for a count of 15 to mix the solution thoroughly.
- 4) Add one level scoop of uniformly dry soil to the tube and cap it.

- 5) Shake the tube vigorously (~ 100 strokes/min) for 2 minutes, set the timer to ensure that each tube is shaken for a consistent amount of time.
- 6) Stand tube in rack and set the timer for 10 minutes to allow the soil to settle. Ensure the rack is not in direct sunlight.
- 7) After the ten minute settling time is up add 49.5 ml of distilled water to a *clean* centrifuge tube labeled with the corresponding site name of the first tube.
- 8) Using a clean bulb pipette take 0.5 ml (fill pipette) of the liquid from the upper 1 cm of the soil-KMnO₄ solution (first tube) while avoiding floating debris, and place 12 drops (0.5ml) in the tube with distilled water.
- 9) Cap the tube and shake it for a count of 15.
- 10) Pour approximately 15 ml of the diluted solution from the second tube into a clean glass cuvette swirl and dump contents and repeat two times. Fill cuvette with the diluted solution.
- 11) Wipe the outside with a lab tissue and place in the colorimeter well. Put the cover in place and press read.
- 12) Record the absorbance for the solution on the datasheet (if all the KMnO₄ has reacted, i.e. no detection on the colorimeter, repeat steps 1-5 and 7-13 using only 0.25 g of dry soil).

In order to speed up the process a maximum of seven tubes were done at once. This was achieved by placing fourteen tubes in the rack and filling seven with 20 ml of distilled water and the other seven with 49.5 ml of distilled water. We used a graduated cylinder to measure out the volumes of distilled water to add to the centrifuge tubes to reduce the variability caused by measuring based on the markings on the tubes.

Forty-six drops (2 ml) of the stock solution were then added to the tubes with 20 ml of water, the lids labeled and then the tubes capped and shaken. A scoop of the corresponding soil sample was placed in the appropriate tube, ensuring that the scoop and the metal leveler were wiped after each sample to avoid cross-contamination. When the 10-minute settling period was up the procedure outlined above was followed for each tube. Two glass cuvettes were used during this process. One was filled with distilled water and was used to zero the colorimeter after every five samples (a reading of this cuvette was also taken after five samples to ensure it read as 0.00 on the colorimeter). The second glass cuvette was used specifically for the soil-KMnO₄ solutions. It was not rinsed with distilled water after each sample as each solution was rinsed through the cuvette three times to ensure that no solution from the previous sample remained. At the end of each day all the bottles containing the standard solutions and the scoop were cleaned with distilled water and set out to dry. The glass cuvettes were rinsed and cleaned with a 10% bleach solution and set out to dry.

Appendix 4.3. Calculations used to Determine Standard Curve Equation

Example From Readings Taken in the Morning of January 30, 2004

y = Standard Solutions	0M	0.005M	0.01M	0.015M	0.02M
x = Corresponding Absorbance Readings	0	0.24	0.48	0.66	0.91

Using the Method of Least Squares:

	y	x	x ²	x*y	y ²
	0	0	0	0	0
	0.005	0.24	0.0576	0.0012	3E-05
	0.01	0.48	0.2304	0.0048	0.0001
	0.015	0.66	0.4356	0.0099	0.0002
	0.02	0.91	0.8281	0.0182	0.0004
SUM	<i>0.05</i>	<i>2.29</i>	<i>1.5517</i>	<i>0.0341</i>	<i>0.0008</i>

To obtain the least-squares prediction equation use:

$$\hat{y} = B_0 + B_1x \quad \text{where} \quad B_0 = \text{y-intercept}$$

$$B_1 = \text{slope of the line}$$

Least squares estimators of B_0 and B_1

$$B_1 = SS_{xy}/SS_x$$

$$B_0 = y(\text{mean}) - B_1x(\text{mean})$$

$$SS_x = (1.5517) - (2.29)^2/5 = 0.50288$$

$$y(\text{mean}) = (0+0.005+0.01+0.015+0.02)/5 = 0.01$$

$$SS_{xy} = 0.0341 - (2.29)(0.05)/5 = 0.0112$$

$$x(\text{mean}) = (0+0.24+0.48+0.66+0.91)/5 = 0.458$$

$$B_1 = SS_{xy}/SS_x = 0.0112/0.50288 = 0.022272$$

$$B_0 = y(\text{mean}) - B_1x(\text{mean}) = 0.01 - (0.022272)(0.458) = -0.0002$$

Therefore equation of the line is:

$$\hat{y} = -0.0002 + 0.022272x$$

Substitute this into the equation for Active C (Weil et al. 2003)

$$\text{Active C (mg/kg)} = (0.02 \text{ Mol/L} - (-0.0002 + 0.022272 \cdot \text{abs})) \cdot (9000 \text{ mg C/Mol})$$

$$\quad \quad \quad \cdot (0.02 \text{ L soln} / 0.00391 \text{ kg soil} \cdot \cdot)$$

** based on our measurements substituted 0.00391kg instead of 0.005kg

So the Active C (mg/kg) in a sample with an absorbance reading of 0.24 is:

$$\text{Active C (mg/kg)} = (0.02 \text{ Mol/L} - (-0.0002 + 0.022272 \cdot 0.24)) \cdot (9000 \text{ mg C/Mol})$$

$$\quad \quad \quad \cdot (0.02 \text{ L soln} / 0.00391 \text{ kg soil} \cdot \cdot)$$

$$\text{Active C (mg/kg)} = 683.8755$$

Appendix 5. Procedure for Basic EDTA Test

Appendix 5.1. List of Materials Needed for Basic EDTA Test

Materials for Basic EDTA Test	
EDTA disodium salt (4L)	Centrifuge tubes
Sodium hydroxide (2 x 1L)	Test tube holder for standards
Measuring cup (500 ml)	Timer
Label	Funnels
Large plastic container	Filter paper
Scoop (1 ml aluminum measuring spoon)	Felt marker
Metal scoop to level off measuring spoon	Graduated cylinder
Aluminum trays to dry samples	Glass vials to place standards and unknown
Mortar and pestle	filtrates in

Appendix 5.2. Revised Protocol for Basic EDTA Field Method

To prepare the Basic EDTA:

- 1) Add 1 part EDTA disodium salt (0.05M) to 1 part Sodium hydroxide (0.25M) into a large container. Mix solution thoroughly using magnetic stirrer.
- 2) Label the contents on the container

To prepare the standards:

- 1) Place each standard soil sample in mortar and pestle and crush.
- 2) Take 0.5 g of sample and place in a centrifuge tube and label the cap with the site name.
- 3) Using a graduated cylinder, add 20 ml of the basic EDTA to the tube. Cap and shake the tube for 30 seconds.
- 4) Prepare a funnel by lining it with filter paper and place it in a clean centrifuge tube.
- 5) Transfer the soil-basic EDTA solution to the funnel, making sure to pour slowly to ensure the solution does not seep over the filter paper.
- 6) When filtrate has collected in tube, dispose of filter paper and clean the funnel. Transfer the filtrate to a glass vial, cap the vial and mark with the appropriate percent value of organic matter.
- 7) Repeat steps 1-6 for soils corresponding to all standards, ensuring to wipe down the mortar and pestle after each use in order to avoid cross-contamination.

To prepare the samples for comparison to the standards the same procedure was followed as outlined above. We tested a maximum of 7 samples at one time, with 3 replicates for each sample, in order to process as many samples as possible. We set up 42 centrifuge tubes in test tube racks. Twenty milliliters of the basic EDTA was added to 21 of the tubes. The remaining 21 tubes were prepared by placing a funnel lined with filter paper in them. Using a 1 ml measuring scoop, 0.5 g of leveled soil from one of the samples was placed in a tube containing the basic EDTA solution. This was repeated two more times for this sample. The three tubes with the soil-basic EDTA mixture were then capped with lids labeled with the samples site and rep number (e.g. site 812 rep 1). They were shaken for 30 seconds and then each solution poured

into three funnels lined with the filter paper. While these samples were filtering a new sample was prepared.

When all the filtrate had been collected for all 7 samples each filtrate was transferred to a glass vial, ensuring that the vial contained the same volume of filtrate as in the standard vials. The filtrate was compared to the standards and the estimated organic matter (%) recorded on a datasheet.

Appendix 5.3. Table of Sites Used for Standards in Basic EDTA Test

Sites chosen to be standards and their corresponding organic matter values are presented in Table 6.

Table 6. Standard Soils and Corresponding OM (%) Lab Values

Standard	Site Name	Site #	Lab Values		
			OM (%) 2001	OM (%) 2002	OM (%) 2003
0%	Blank	/	/	/	/
~1.5 %	Tilley	812	1.73	1.4	1.6
2%	Chinook	806	2.23	1.9	2.1
3%	Claresholm	793	3.8	3.4	3.3
4%	B.B. Mountain	594	4.15	3.6	3.8
6%	Beiseker	798	6.64	6.5	6.4
7%	Tomahawk	692	6.9	7	8
9%	Sexsmith	599	8.76	9.2	8.9
10%	Carstairs	746	9.96	11.6	10
12%	Wetaskiwin	727	12.43	12.8	12.5

Appendix 5.4. Accuracy of the Measuring Scoop for Procedure of Bowman (1997)

We determined the accuracy of the measuring scoop for the Bowman (1997) method according to the following procedure:

- 1) Take the scoop and place on scale, tare scale.
- 2) Take scoop of dry soil and level
- 3) Place scoop with soil on scale and weigh, record the weight of the soil.
- 4) Take soil that was in scoop and place in separate container.
- 5) Repeat steps 1-4 four more times for the sample
- 6) After one sample has been weighed 5 times clean off the scoop and re-weigh the scoop and tare it again if needed.
- 7) Proceed to the other samples repeating steps 1-6.

Results for the 11 soils used in this test indicate that on average the scoop measured 0.66 g of soil (Table 7).

Table 7. Results of Measuring Accuracy Test for Bowman (1997) Procedure

Sample I.D.	Rep Number					Mean (g)	Range (g)	Variance	Standard Deviation	Coefficient of Variation
	1	2	3	4	5					
Three Hills	0.68	0.72	0.67	0.65	0.64	0.67	0.08	0.001	0.03	4.63
Pincher Creek	0.6	0.58	0.67	0.58	0.57	0.60	0.10	0.002	0.04	6.77
Grimshaw	0.54	0.67	0.64	0.63	0.61	0.62	0.13	0.002	0.05	7.88
Oyen	0.71	0.68	0.67	0.81	0.69	0.71	0.14	0.003	0.06	7.97
Hanna	0.76	0.72	0.76	0.76	0.74	0.75	0.04	0.000	0.02	2.39
Wetaskiwin	0.6	0.53	0.5	0.55	0.59	0.55	0.10	0.002	0.04	7.51
Veteran	0.72	0.72	0.66	0.81	0.65	0.71	0.16	0.004	0.06	8.96
Carvel	0.55	0.62	0.75	0.65	0.73	0.66	0.20	0.007	0.08	12.40
Chinook	0.78	0.81	0.81	0.74	0.83	0.79	0.09	0.001	0.04	4.42
Lacombe	0.61	0.62	0.59	0.68	0.57	0.61	0.11	0.002	0.04	6.77
Carstairs	0.57	0.6	0.68	0.62	0.53	0.60	0.15	0.003	0.06	9.35
Overall Mean						0.66				

Appendix 6. Descriptive Statistics for Results of Active C Method Based on Three Repeated Measures

Site Name	Site #	Position	Lab OM (%)	Lab LFC (mg/kg)	Active C (mg/kg)	Active C				
						Mean	Sample Variance	Standard Deviation	Standard Error	Range
B.H. Prairie	586	M	8.13	2567.92	878.01	854.91	424.66	20.61	11.90	39.60
					848.31					
					838.42					
Manning	588	M	8.02	1576.99	858.21	861.51	32.67	5.72	3.30	9.90
					868.11					
					858.21					
Grimshaw	590	M	4.90	2850.83	657.80	690.70	1082.82	32.91	19.00	65.81
					723.61					
					690.70					
Worsley	591	M	12.25	1967.22	896.09	892.88	30.98	5.57	3.21	9.64
					886.45					
					896.09					
High Prairie	592	M	8.92	2411.17	897.81	897.81	0.00	0.00	0.00	0.00
					897.81					
					897.81					
Fairview	593	M	6.75	883.24	789.42	811.36	842.19	29.02	16.76	54.84
					844.27					
					800.39					
B.B. Mtn	594	M	3.76	266.65	481.57	468.71	495.64	22.26	12.85	38.56
					481.57					
					443.01					
Fahler	595	M	8.01	1547.88	741.85	716.14	588.57	24.26	14.01	48.20
					693.65					
					712.93					
Sexsmith	599	M	8.91	1742.30	759.22	831.82	4344.55	65.91	38.05	128.69
					887.91					
					848.31					
Smith	615	M	5.44	5033.70	723.61	749.20	521.36	22.83	13.18	43.87
					756.52					
					767.48					

Site Name	Site #	Position	Lab OM (%)	Lab LFC (mg/kg)	Active C (mg/kg)	Active C				
						Mean	Sample Variance	Standard Deviation	Standard Error	Range
Boyle	678	M	5.31	4105.46	646.12	652.57	31.20	5.59	3.23	9.68
					655.80					
					655.80					
Bonnyville	680	M	5.60	2734.11	712.64	719.95	1965.12	44.33	25.59	87.75
					767.48					
					679.73					
Dapp	681	M	4.11	1949.12	675.15	623.55	2090.63	45.72	26.40	87.08
					607.42					
					588.07					
Carvel	684	M	1.54	2055.99	452.65	455.86	1889.61	43.47	25.10	86.76
					500.85					
					414.09					
Warspite	687	M	2.44	2835.22	520.35	581.62	4711.71	68.64	39.63	135.45
					655.80					
					568.72					
Beauvallon	688	M	9.65	1411.99	866.20	888.14	360.94	19.00	10.97	32.91
					899.11					
					899.11					
Tomahawk	692	M	8.02	2565.56	684.01	677.58	123.91	11.13	6.43	19.28
					664.73					
					684.01					
Westerose	703	M	2.52	861.77	384.89	371.99	218.42	14.78	8.53	29.03
					375.22					
					355.87					
Wetaskiwin	727	M	12.80	720.63	921.05	946.64	521.36	22.83	13.18	43.87
					953.95					
					964.92					
Hairy Hill	728	M	9.08	2447.68	876.81	867.17	92.93	9.64	5.57	19.28
					867.17					
					857.53					
Killam	738	M	5.20	2222.80	515.20	537.14	360.94	19.00	10.97	32.91
					548.11					
					548.11					

Site Name	Site #	Position	Lab OM (%)	Lab LFC (mg/kg)	Active C (mg/kg)	Active C				
						Mean	Sample Variance	Standard Deviation	Standard Error	Range
Chauvin	739	M	2.48	492.67	438.42	456.70	280.73	16.76	9.67	32.91
					460.36					
					471.33					
Bashaw	740	M	9.30	2637.38	848.31	861.51	228.66	15.12	8.73	29.70
					878.01					
					858.21					
Provost	743	M	4.44	271.06	581.02	555.42	761.99	27.60	15.94	54.84
					559.08					
					526.17					
Lacombe	744	M	9.15	3214.64	887.91	884.61	32.67	5.72	3.30	9.90
					878.01					
					887.91					
Carstairs	746	M	9.96	1242.79	868.11	874.71	32.67	5.72	3.30	9.90
					878.01					
					878.01					
Veteran	769	M	4.93	1101.24	492.92	499.30	122.25	11.06	6.38	19.15
					492.92					
					512.07					
Three Hills	781	M	6.80	411.83	709.72	696.52	2580.60	50.80	29.33	98.99
					739.42					
					640.43					
Hanna	786	M	3.70	2044.03	432.54	465.54	816.64	28.58	16.50	49.50
					482.04					
					482.04					
Vulcan	791	M	3.74	326.04	519.83	571.09	1997.36	44.69	25.80	82.02
					591.60					
					601.85					
Claresholm	793	M	3.26	190.89	655.09	622.96	2261.34	47.55	27.46	86.76
					645.45					
					568.33					
Beiseker	798	M	6.47	594.67	752.55	726.75	592.86	24.35	14.06	48.38
					704.18					
					723.53					

Site Name	Site #	Position	Lab OM (%)	Lab LFC (mg/kg)	Active C (mg/kg)	Active C				
						Mean	Sample Variance	Standard Deviation	Standard Error	Range
Pincher Creek	800	M	6.09	180.92	809.33	764.34	1889.61	43.47	25.10	86.76
					761.13					
					722.57					
Youngstown	804	M	2.60	991.76	345.53	325.02	1997.36	44.69	25.80	71.77
					355.78					
					273.76					
Chinook	806	M	2.15	129.09	234.55	237.85	1992.61	44.64	25.77	89.09
					284.05					
					194.96					
Oyen	809	M	2.71	460.35	442.44	392.94	2449.93	49.50	28.58	98.99
					343.45					
					392.94					
Tilley	812	M	1.43	148.87	326.84	262.34	3494.78	59.12	34.13	116.10
					249.44					
					210.74					
Dunmore	815	M	1.91	417.59	335.27	277.17	3188.77	56.47	32.60	112.78
					273.76					
					222.49					
Enchant	823	M	2.47	303.13	365.54	394.57	842.49	29.03	16.76	58.05
					394.57					
					423.59					
Etzikom-Dry	828a	M	1.49	366.84	407.04	345.53	2943.48	54.25	31.32	102.53
					325.02					
					304.51					
Etzikom-Irrig	828b	M	3.10	657.66	413.92	368.77	3026.73	55.02	31.76	106.43
					384.89					
					307.49					

Appendix 7. Descriptive Statistics for Results of Basic EDTA Method Based on Three Repeated Measures

Site Name	Site #	Position	Lab OM (%)	Estimated OM (%)	Estimated OM (%)				
					Mean	Variance	Standard Deviation	Standard error	Range
B.H. Prairie	586	M	8.13	4	4	0	0	0	0
				4					
				4					
Manning	588	M	8.02	4	4.67	0.33	0.58	0.33	1
				5					
				5					
Grimshaw	590	M	4.90	4	4	0	0	0	0
				4					
				4					
Worsley	591	M	12.25	6	7	3	1.73	1	3
				9					
				6					
High Prairie	592	M	8.92	6	8	3	1.73	1	3
				9					
				9					
Fairview	593	M	6.75	6	6	0	0	0	0
				6					
				6					
B.B. Mtn	594	M	3.76	3	3	0	0	0	0
				3					
				3					
Fahler	595	M	8.01	9	9	0	0	0	0
				9					
				9					
Sexsmith	599	M	8.91	6	6	0	0	0	0
				6					
				6					
Smith	615	M	5.44	3.5	3.5	0	0	0	0
				3.5					
				3.5					
Boyle	678	M	5.31	4	4	0	0	0	0
				4					
				4					
Bonnyville	680	M	5.60	6	8	3	1.73	1	3
				9					
				9					
Dapp	681	M	4.11	4	4	0	0	0	0
				4					
				4					
Carvel	684	M	1.54	3.5	3.5	0	0	0	0
				3.5					
				3.5					

Site Name	Site #	Position	Lab OM (%)	Estimated OM (%)	Estimated OM (%)				
					Mean	Variance	Standard Deviation	Standard error	Range
Warspite	687	M	2.44	4	4	0	0	0	0
				4					
				4					
Beauvallon	688	M	9.65	10	10	0	0	0	0
				10					
				10					
Tomahawk	692	M	8.02	3.5	3.5	0	0	0	0
				3.5					
				3.5					
Westeros	703	M	2.52	3	3	0	0	0	0
				3					
				3					
Wetaskiwin	727	M	12.80	12	12	0	0	0	0
				12					
				12					
Hairy Hill	728	M	9.08	10	10	0	0	0	0
				10					
				10					
Killam	738	M	5.20	5	5	0	0	0	0
				5					
				5					
Chauvin	739	M	2.48	3.5	3.5	0	0	0	0
				3.5					
				3.5					
Bashaw	740	M	9.30	9	9.67	0.33	0.58	0.33	1
				10					
				10					
Provost	743	M	4.44	3.5	3.5	0	0	0	0
				3.5					
				3.5					
Lacombe	744	M	9.15	10	10	0	0	0	0
				10					
				10					
Carstairs	746	M	9.96	10	10	0	0	0	0
				10					
				10					
Veteran	769	M	4.93	4	4	0	0	0	0
				4					
				4					
Three Hills	781	M	6.80	4	4.67	1.33	1.15	0.67	2
				4					
				6					
Hanna	786	M	3.70	3	3.5	0.25	0.5	0.29	1
				4					
				3.5					

Site Name	Site #	Position	Lab OM (%)	Estimated OM (%)	Estimated OM (%)				
					Mean	Variance	Standard Deviation	Standard error	Range
Vulcan	791	M	3.74	3.5	3.5	0	0	0	0
				3.5					
				3.5					
Claresholm	793	M	3.26	3.5	3.5	0	0	0	0
				3.5					
				3.5					
Beiseker	798	M	6.47	6	7	3	1.73	1	3
				9					
				6					
Pincher Creek	800	M	6.09	4	4	0	0	0	0
				4					
				4					
Youngstown	804	M	2.60	3	3	0	0	0	0
				3					
				3					
Chinook	806	M	2.15	2	2	0	0	0	0
				2					
				2					
Oyen	809	M	2.71	3	3.17	0.08	0.29	0.17	0.5
				3					
				3.5					
Tilley	812	M	1.43	1.5	1.83	0.08	0.29	0.17	0.5
				2					
				2					
Dunmore	815	M	1.91	1.5	1.83	0.08	0.29	0.17	0.5
				2					
				2					
Enchant	823	M	2.47	3	3	0	0	0	0
				3					
				3					
Etzikom-Dry	828a	M	1.49	3	3	0	0	0	0
				3					
				3					
Etzikom-Irrig	828b	M	3.10	3	3	0	0	0	0
				3					
				3					