AG : BOT/85/011
FIELD DOCUMENT 18

Soil Mapping and Advisory Services

Botswana

LABORATORY PROCEDURES

REVISED MANUAL

FOOD & AGRICULTURE
ORGANIZATION OF THE
UNITED NATIONS

UNITED NATIONS
DEVELOPMENT
PROGRAMME

REPUBLIC OF
BOTSWANA

GABORONE, DECEMBER 1989

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LABORATORY PROCEDURES

REVISED MANUAL

based on the work of:

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edited by Bas Schalk

Food and Agricultural Organization of the United Nations
United Nations Development Programme
Republic of Botswana

Gaborone, 1989

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INTRODUCTION

The first version of this manual was written by Dr. R. Breitbart and published as part 2 of Field Document 3.
In view of the revision of some of the procedures and the introduction of additional methods for routine analysis, it became necessary to publish a revised version.

Bas Schalk,

Gaborone, December 1989

SAMPLE RECEPTION, GRINDING AND STORAGE

Sample reception:

Incoming samples must always be accompanied by a list of all samples in duplicate. Check the samples against this list immediately after arrival.

Mark any irregularities on the list and report to the soil chemist.

The duplicate of the list is returned to sender together with any remarks or questions there may be.

Labnumbers are assigned as follows :
the last two figures of the year in which assignment takes place
+ a numerical order number

For example 89-336 is the 336th sample received in 1989.

Labnumbers are thus assigned regardless of the origin of the sample. Assignment is done as follows:
If the list with samples is correct and all samples on that list are present, the sample identifications are entered into the computer. The program assigns the labnumbers.
The samples are entered in the sample register with these labnumbers. If samples are not completely dry, they should be spread out on a tray to allow drying immediately after registration.

Sample grinding:

Before grinding most samples first have to be dried.

Spread each sample on a tray, together with the sample bag and sample number card. Place in a wind protected spot until dry.

Write lab numbers on the sample containers with a permanent marker. Arrange sample trays and sample containers in serial order to avoid any possibility of mixing up samples.

If analysis requiring clods has to be done on the sample, such will be indicated on the request form. In that case at least about 10 clods have to be returned to the sample bag.

Using the "dynacrush" sample grinder :
Mix the sample well in the tray and fill an appropriate amount of soil from the sample tray into the 'Dynacrush' sample grinder. After about 1 minute, switch off. Fill the fine earth fraction which has passed into the receiver through the 2 mm sieve at the bottom of the grinder into the sample container and discard the remainder.
Return the remainder of the sample from the tray to the sample bag.

Using crushing by hand :
Mix the sample well in the tray, return the excess to the sample bag, keeping enough sample to fill the sample box. Crush the sample in the tray by pressing with a stone. Pass the crushed sample through the 2 mm sieve into the collecting bowl. Apart from hard gravel, all the sample should pass the sieve. If necessary crush again in the tray with the stone.

<u>NOTE</u>: Some samples, especially those very rich in clay, form extremely hard aggregates when dry. These aggregates <u>cannot</u> be ground successfully with the Dynacrush grinder, they might even destroy it. Crush the aggregates first with a stone or a hammer before filling into the grinder. Be careful not to crush any stones.

Store the samples in the sample store room with ascending lab numbers.

pH-DETERMINATION

Equipment:

Two 25 litre drums for water and $CaCl_2$-solution.

One pH-meter with electrode. (Zeiss pH-meter 300 with plastic protected 'Orion' electrode 91-55).

Two "AD 1-3 three volume one aliquot" dispensers.

Reagents:

For pH-H_2O: distilled water

For pH-$CaCl_2$: 0.01m $CaCl_2$ solution
(0.01m $CaCl_2$ = 1.11 g $CaCl_2$ per litre or
27.75 g $CaCl_2$ per 25 liter)

Buffer solutions: Use pH 7-, pH 4-, pH 9.2-buffer tablets. Prepare according to instructions on the packet.

All water is distilled or deionised water.

Procedure:

1. Switch the pH-meter to "stand by" position for warming up.

2. Of each sample weigh two times 20 g into 100 ml plastic bottles. (One set of bottles is marked 'pH-H_2O', the other 'pH-$CaCl_2$')

3. To the samples in the bottles marked 'pH-H_2O' add 50 ml H_2O dist. with the dispenser connected to the water drum.
To the samples in the bottles marked 'pH-$CaCl_2$' add 50 ml $CaCl_2$-solution with the other dispenser.

4. Close bottles tightly, put on shaker in horizontal position and shake for two hours.

5. Switch the pH-meter into 'measuring' mode.
Place the electrode in the pH 7 buffer solution. Wait for stable reading.
(Less than 30 seconds should be required - if it takes much longer the electrode might require cleaning.)
Set the meter to 7.00 with the calibration control.

6. Rinse the electrode with distilled water. Place it in the pH 4 buffer solution (for acid soils), respectively in the pH 9.2 buffer solution (for alkaline soils). Set the meter reading with the slope control.
(The slope should be greater than 92% - if not the electrode might require cleaning)

7. After calibration immerse electrode into upper part of sample suspension.
   Do not stir the suspension.
   Wait for a stable reading.
   A change of 0.01 units in 10 seconds is considered a stable reading. Note the value, rinse the electrode with distilled water from a wash bottle and continue with the next sample.

8. After pH-H_2O measurement also measure electrical conductivity of the extract.
   (To decide whether or not preparation of saturated paste extracts for electric conductivity and/or soluble salts determination is required)

9. Storing and printing of soil analytical data is done with the Soil Laboratory Database system on a IBM compatible micro computer.

SATURATED PASTE EXTRACT

Equipment:

24-place mechanical vacuum extractor cups for extractor.

large beakers (800-1000ml)

burette (100 ml)

spatula

Reagent:

distilled water

Procedure:

Preparation of saturated soil paste:

1. Weigh 200 g of soil into beakers
2. With the burette add few ml of water and mix with spatula.
3. Slowly add more water and mix until soil paste glistens as it reflects light, flows slightly when the container is tipped and the paste slides freely and clean off the spatula (for all soils but the soils with high clay content).
4. After mixing allow the soil to stand for one hour or more. Then check saturation criteria again:

   If the paste stiffens or loses its glistening appearance remix with water.

   Free water should not collect on the surface. If the paste is too wet, add additional dry soil.
5. If soluble salts are to be determined record total amount of water and soil.
6. Cover beakers with watch glasses and let stand for an appropriate time.

Preparation of the saturated paste extract:

For conductivity determination the extraction can be made a few minutes after preparing the saturated paste.
If the soil contains gypsum the conductivity can increase by 1 or 2 mS/cm upon standing.

Therefore if gypsum is present allow the saturated paste to stand several hours (4-6 hours) before extracting the solution.

If the solution is to be analyzed for its chemical constituents, the saturated paste should stand for 12 hours or more (over night).

Place 9 cm fine porous filter paper (i.e Whatman no 42,44 or 50) in extraction cups. Moisten with few drops of water and press it on the bottom of cups.

Hang cups into extractor and connect syringes. Transfer samples into cups. Set extractor to about 1 hour extraction time and start extraction.

ELECTRICAL CONDUCTIVITY

Electrical conductivity is measured in the dispersion prepared for pH measurement. If the conductivity is higher than 1 mS/cm, a saturated paste extract is prepared in order to get a proper value for electrical conductivity.

Procedure:

1. Transfer saturated paste extract into small narrow vials. The cell constant of the conductivity electrode, presently in use is 1.00. If another electrode is used check cell constant (usually either engraved on electrode or on the connecting cable).
2. Switch the range of the Zeiss con 602 conductivity meter on 'cal' and set the display to 1000 with the 'cal' knob (calibration of cell constant)
3. Record the temperature of the solutions.
4. Switch 'range' into the lowest of the three **mS/m** positions (do **not** use the **µS/m** range).
5. Insert electrode into extract.

   NOTE: The two platinum electrodes inside the measuring chamber must be completely immersed in the solution. If the extract is not enough prepare a new saturated paste extract of this sample and combine the two extracts. (this can happen with soils that are very rich in clay).
6. Record the reading and continue with the next sample. If the conductivity is out of the set range (display shows 1), switch to a higher range.

There are five range settings :

1. 0-2000 micro S/m

2. 0-20.00 mS/m
3. 0-200.0 mS/m
4. 0-2000 mS/m

5. 0-20.00 S/m

As already mentioned above only the three mS/m ranges (2-4) should be used.

If otherwise, it must be clearly recorded, so that the correct unit can be defined to the data processing program.

Calculations and data storage:

Calculation, storing and printing of soil analytical data is done with the Soil Laboratory Database system on a IBM compatible micro computer.

Data are reported in mS/cm.

To convert mS/m to mS/cm the data processing program of the laboratory database system divides the reading by 100.

mS/cm = mS/m/100.

Temperature correction:

Use temperature correction table to find correction factor
Figure 1 page 7 (US. Salinity laboratory staff, 1954).

The correction factor also can be calculated with the formula:

corr. factor = 2.581+(-0.493)*LOG (TEMP).

(Logarithmic regression curve calculated from temperature correction table.)
This formula is used in the data processing program

SOLUBLE SALTS DETERMINATION

Equipment, reagents and standards:

Use same equipment and same chemicals as for exchangeable cations determination.

Take combined stock solutions:

200 meq/l Ca + 50 meq/l Mg
50 meq/l K + 50 meq/l Na

Prepare diluted stock solutions and standard series in the same way as for exchangeable cations determination, only that

the volumetric flasks are filled up with distilled water and not with ammonium acetate solution.

Procedure:

Pipette 1 ml saturated paste extract into 100 ml volumetric flask. Fill up with distilled water and shake well to mix.

<u>NOTE</u>: If absorbance readings of samples are too low with this dilution other ratios can be chosen.
The '1. dilution' then is calculated as follows :

Size of volumetric flask : ml saturated paste extract

Further procedure is exactly as for exchangeable cations determination.

Calculations:

If linear regression curve is calculated concentration **C** of soluble cations in measuring solution is:

$$C = a * E + b$$

a, b = regression coefficients
E = Extinction (absorbance reading)

If no linear regression curve is calculated concentration **C** of soluble cations in measuring solution is:

$$C = E / e$$

E = Extinction (absorbance reading)
e = Extinction coefficient

e is calculated from standard curve:

$$e = (E1/C1 + E2/C2 + E3/C3 + \dots + En/Cn) / n$$

E1, E2, E3, ... , En = Absorbance readings of standards
C1, C2, C3, ... , Cn = Concentrations of standards
n = Number of standards

Concentration **K** of soluble salts in extract **(meq/l)** is:

K = C * 1.dilution * 2.dilution

Concentration **K** of soluble salts in soil **(meq/100 g soil)** is:

K = C * extract / weight * 1.dilution * 2.dilution / 10

Extract = ml water used for saturated paste
Weight = g soil used for saturated paste
1. dilution = 100 (1 ml extract in 100 ml volumetric flask)
2. dilution = additional dilution

AVAILABLE PHOSPHORUS DETERMINATION
(Bray and Kurtz I&II)

Equipment:

Two 25 litre drums for extracting solution and distilled water.

Multi-funnel rack for 33 funnels of approx. 5 cm diameter.

Filter paper Whatman 44 or 42, 9 cm diameter.

Colorimeter, preferably with flow through cell.

One AD 1-3 'three volume one aliquot' dispenser
or
One AD 3 'three volume three aliquot' dispenser

One SCD 1 'combination diluter-dispenser'

One multisample stirrer

Reagents:

Ammonium Fluoride	NH_4F	(MW=37.05)
Hydrochloric acid conc.	HCl	(36%)
Ammonium hepta-molybdate	$(NH_4)_6Mo_7O_4$* 4 H_2O	
Antimony Potassium tartrate	$K(SbO)C_4H_4O_6$* 0.5 H_2O	
Sulphuric acid conc.	H_2SO_4	(98%)
Ascorbic acid		
Potassium dihydrogen phosphate	KH_2PO_4	

Preparation of solutions :

Extracting solution for Bray and Kurtz I: 0.03 m NH_4F in 0.025 m HCl

Dissolve 27.78 g NH_4F in approximately 500 ml distilled or deionized water. Add 53.6 ml HCl conc. (36%).

Transfer to 25 liter drum and make to 25 liters with distilled/deionized water. Close the drum and mix thoroughly.

Extracting solution for Bray and Kurtz II: 0.03 m NH_4F in 0.1 m HCl

To the dissolved 27.78 g NH_4F add 214.5 ml HCl conc. (36%).

Proceed as for the Bray and Kurtz I extracting solution.

Color reagent A:

1. Dissolve in a beaker 30 g of Ammonium hepta-molybdate, in about 250 ml H_2O dist.

2. Dissolve in a second beaker 0.727 g Antimony Potassium tartrate.

3. Fill about 500 ml H_2O dist. into a 2000 ml volumetric flask.
4. Add **slowly** 340 ml conc. H_2SO_4.
5. Cool in a waterbath to 20° Celsius.
6. Transfer Molybdate and Tartrate solutions.
7. Fill up to the mark with distilled water and transfer into 5 litre bottle.
8. With a measuring cylinder add 3 liters distilled water. Mix well.
9. Store cool and dark.

Color reagent B:

Dissolve 1 g Ascorbic acid per 100 ml of reagent A. (Approximate requirement for 33 samples plus 5 standards is 250 ml.)

This solution keeps only for one day and has to be prepared new just before use.

Stock solution 1000 ppm P/litre:

Dry Potassium dihydrogen phosphate at 80° Celsius in an oven.

Cool in a desiccator.

Weigh 4.393 g and dissolve in 1000 ml volumetric flask with distilled water.

Diluted stock solutions:

a. 100 ppm P/l: Pipette 50 ml of 1000 ppm stock solution into 500 ml volumetric flask. Fill with distilled water until about 1 cm below mark. Temperate to 20° Celsius in waterbath and fill up to mark.

b. 10 ppm P/l : Pipette 50 ml of 100 ppm solution into 500 ml volumetric flask. Proceed as for 100 ppm.

Standard series:

1. Pipette 10, 20, 30, 40, 50 ml

   of the 10 ppm solution into 500 ml volumetric flask.
2. Proceed as for 100 ppm P.
3. The resulting standard series has the concentrations:

   0.2, 0.4, 0.6, 0.8, 1.0 ppm P/l

Settings:

A soil/extractant ratio of 1:7 is used.

Set the dispenser for the extractant to 35 ml. Using a AD 3 'three volume-three aliquot dispenser, it must be made sure that all three syringes dispense precisely the same volume.

Set the SCD 1 'combination diluter-dispenser' to dispense 5 ml color reagent, 15 ml dist. water and take up 20 ml extract.

The resulting dilution of 1:2 for samples and standards is found convenient.

Procedure:

1. Prepare the multi funnel rack. Fold double layers of filter paper and staple the edges that they hold their form. Put the filters into the funnels and place a tray of 33 bottles under the rack.

2. Prepare the color reagent B. For one batch take 250-300 ml of reagent A. Add 2.5 - 3 g Ascorbic acid and stir with a magnetic stirrer until dissolved.

   NOTE: Color reagent B is stable only for a few hours and must always be prepared just before the determination.

3. Weigh 5 g of the samples into the bottles of a tray. The last sample should be a standard sample which is repeated with every batch.

4. Quickly dispense 35 ml extracting solution to each of the samples.

5. Stir for 1 minute with the multisample stirrer. (If the Bray and Kurtz II extractant is used stir for 40 seconds.)

6. **Immediately** after stirring filter by pouring simultaneously the contents of 11 bottles (one bottle rack) onto one row of filters. If the filtrates are not clear, filter again using the same filters.

7. Put the tube connected to the syringe calibrated to 5 ml of the combination diluter-dispenser into the color reagent B solution. (The syringe calibrated to 15 ml is permanently connected with the water drum.) Pump at least 5 or more times and discard solution to make sure that all tubes are filled with fresh solution.

8. Place a third bottle rack tray next to the one with the filtrates. Put the double plastic tips of the diluter-dispenser into extract. By pressing the lever take up 20 ml extract, 5 ml color reagent and 15 ml water. Transfer this into the bottles of the third tray by releasing the lever. Dip the tips into a beaker with distilled water to clean the outside.

9. For the standards proceed in the same way. Fill a little more than 20 ml of the standards and extracting solution (for the blanc) into a row of bottles and transfer with the diluter-dispenser into a second row.

10. After 30 minutes to 1 hour the blue color of the molybdate complex is fully developed. Determine the absorption of standards and samples at 670 nm wavelength on the spectrophotometer. Use blanc for zero.

NOTE: The standard curve is adapted to the relatively phosphorus-poor soils most widespread in Botswana. Exceptions are diluted with a SD 1 diluter. If the P-concentration is generally higher the standards and the initial dilution (here 1:2) should be changed.

Calculations:

Calculation, storing and printing of soil analytical data is done with the Soil Laboratory Database system on a IBM compatible micro computer.

The correlation factor for linear regression that the computer calculates
should be at least 0.9930, otherwise new standards have to be prepared and measured together with the sample solutions.

If the computer is not used the concentration of phosphorus in soil in ppm P is calculated as follows:

Concentration C of P in solution is:

$$C = E / e$$

E=Extinction (absorption)
e=Extinction coefficient

The Extinction coefficient is calculated from the standard curve. Concentration C of the standards is known. Extinction E is read from the spectrometer.

$$e = (E1 / C1 + E2 / C2 + E3 / C3 + \dots + En / Cn) / n$$

E1, E2, E3,..., En = Extinction of standard 1, 2, 3,..., n
C1, C2, C3,..., Cn = Concentration of standard 1, 2, 3,..., n
n = Number of standards

ppm P in soil is calculated:

$$\text{ppm P} = C * \text{dilution} * \text{ml Extract} / \text{Weight}$$

NOTE: Dilution does not refer to the initial dilution with the diluter-dispenser because initially the standards are diluted too; it only applies to samples that are diluted because of high absorbance of the first reading.

AVAILABLE PHOSPHORUS DETERMINATION
(Olsen method)

Equipment :

reciprocal shaker
spectrophotometer or colorimeter with a 880 nm filter
racks with filter funnels + filter (whatman 42)
racks with beakers
dispensers

Reagents :

sodium bicarbonate	$NaHCO_3$
sulphuric acid	$H2SO_4$
ammonium hepta-molybdate	$(NH4)_6Mo_7O_{24}$
potassium antimony tartrate	$KSbOC_4H_4O_6$
ascorbic acid	
all water is distilled or deionized water	

Preparation of solutions :

0.5 M Sodium bicarbonate pH 8.5 (extracting solution)

Dissolve 42 g $NaHCO_3$ in 1 liter of water. Adjust to pH 8.5 by adding 1 M NaOH. In case of overshooting, add some $NaHCO_3$. After storage the pH should be checked and if necessary adjusted.

Color reagent A

1. Add 224 ml of sulphuric acid to 600 ml of water in a 1 liter beaker. After cooling make to 1 liter.
2. Dissolve 12 g of ammonium hepta-molybdate in 300 ml water.
3. Dissolve 0.275 g of potassium antimony tartrate in 100 ml of water.
4. Transfer these three solutions to a 2.5 litre brown bottle and mix well.
5. Store in a cool and dark place.

Color reagent B

Dissolve 1.75 g of ascorbic acid in 100 ml of water.
Prepare fresh daily.

Mixed reagent

mix the following in a 500 ml polythene bottle :

- 70 ml of color reagent A
- 30 ml of color reagent B
- 200 ml of water

This solution has to be prepared fresh daily.

Stock solution 1000 ppm P

1. Dry potassium dihydrogen phosphate in the oven at 80° Celsius. Cool in a desiccator.
2. Weigh 4.393 g and dissolve in distilled water in a 1000 ml volumetric flask. Make to volume.

Stock solution 100 ppm P

Pipette 50 ml of 1000 ppm P into a 500 ml volumetric flask and make to volume with distilled water.

Stock solution 10 ppm P

Pipette 50 ml of 100 ppm P into a 500 ml volumetric flask and make to volume with distilled water.

Standard series 0.2 0.4 0.6 0.8 1.0 ppm P

Pipette 10, 20, 30, 40 and 50 ml of the 10 ppm solution into 500 ml volumetric flasks and make to mark with extracting solution.

Procedure:

1. Weigh 2.00 g of sample into a 100 ml polythene shaking bottle. Do not forget to include 2 blanks and the standard sample.
2. Add 40 ml of extracting solution.
3. Shake for 30 minutes in the reciprocal shaker.
4. Filter through a whatman 42 filter paper into a sample beaker.
5. If the filtrate is dark colored (like tea), decolorize by shaking 20 ml of the filtrate with 0.02 g phosphate free active carbon for 30 minutes.
   Also treat a blank and the standard solutions with active carbon. The active carbon is separated from the extract by filtration over a whatman 42 filter paper.
6. Pipette into test tubes 10 ml of the standard series, the blanks and the sample extracts.
7. Add 10 ml of the mixed reagent and homogenize.

8. Leave the solutions for 1 hour to allow for color development.

9. Measure the absorbance on the spectrophotometer at 880 nm.

Calculation :

Calculation, storing and printing of soil analytical data is done with the Soil Laboratory Database system on a IBM compatible micro computer.

The correlation factor for linear regression that the computer calculates should be at least 0.9930, otherwise new standards have to be prepared and measured together with the sample solutions.

The phosphorus content of the soil is calculated as follows :

ppm P in soil = (ppm P in extract) * 40/2

40 = ml extract
2 = gm soil

Reference :

Labex method L8903-52

ORGANIC CARBON DETERMINATION
(modified Walkley-Black)

Equipment:

Conical flasks 50 ml
Drying oven with temperature regulation
Colorimeter with 570 nm filter or a spectrophotometer.
acid-proof dispenser 0 - 25 ml
5 litre plastic container for digestion solution
25 liters drum for Flocculent solution
AD 1-3 'three volume one aliquot' dispensers
SD 1 diluter
bottle rack trays with bottles

NOTE: For dispensing chromic acid the AD 1-3 dispenser proved to be not suitable because the rubber of the plunger was dissolving after some time and caused wrong results.

Reagents:

Sodium dichromate	$Na_2Cr_2O_7$
conc. Sulphuric Acid	H_2SO_4
Glucose	$C_6H_{12}O_6$
Superfloc 127	

Preparation of solutions:

Sodium dichromate in approximately 10 N H_2SO_4 (digestion solution)

1. In a 5000 ml beaker weigh 745 g of sodium dichromate. Add about 2500 ml of deionized water.
2. Very slowly and constantly stirring with a magnetic stirrer add 1360 ml conc. H_2SO_4.
3. After cooling to 20° Celsius fill up to the 5000 ml mark with water and stir.

Standard solutions:

Dissolve: 1.250 g, 2.500 g, 5.000 g, 10.000 g

glucose in 100 ml volumetric flasks with water.

Superfloc 127 Flocculent solution:

Superfloc 127 Flocculent is a nonionic very high molecular weight polyacrylamide. It is highly effective as flocculent in acid, neutral and alkaline suspensions.

Superfloc has been found very effective at a rate of 1 gram per 10 liters of solution. For 25 l solution weigh 2.5 g Superfloc.
First prepare a concentrated solution in a 1000 ml beaker.

1. Stir 800 ml of water with a magnetic stirrer so that a positive vortex is formed.
2. Add the dry powder very slowly to the edge of this vortex. (Care must be taken to prevent formation of clods or lumps which then require a very long time to dissolve.)
3. Continue stirring for one or two hours until all flocculent is dissolved. Stir at less than 400 rpm to prevent breakage of the long Superfloc molecules which would reduce its flocculation power.

Settings:

Set : - the dispenser for digestion solution to 10 ml
- the AD 1-3 dispenser for flocculent to 35 ml
- the SD 1 diluter to dilution ratio of 1:15.(3 ml uptake and 42 ml dispensing.)

Procedure:

1. From each sample weigh 1 g of soil into 50 ml conical flasks.
2. Pipette 1.0 ml of the glucose standard solutions into 50 ml conical flasks. For the blanc take one empty flask where only digestion solution will be filled.
3. Add 10 ml digestion solution.
4. Put the flasks into the oven preheated to 140° Celsius.
5. After 30 minutes take them out and add 35 ml of the Superfloc 127 flocculent solution with the AD 1-3 dispenser.
6. Shake well and let stand for about one hour to allow the particles to settle.
7. With the SD 1 diluter take an aliquot from the clear supernatant solution and transfer into a bottle of the bottle rack tray. (Take care not to stir the sediment.) Also transfer standards and blanc.
8. Measure absorbance with a colorimeter or spectrophotometer at 570 nm.

Calculations:

Calculation, storing and printing of soil analytical data is done with the Soil Laboratory Database system on a IBM compatible micro computer.

The correlation factor for linear regression that the computer calculates
should be at least 0.9930, otherwise new standards have to be prepared and measured together with the sample solutions.

When **1 g** of soil were weighed the standard curve corresponds with:

0.50, 1.00, 2.00, 4.00 % org. C

Example standard 2:

Glucose contains 40 wt% C => 2.50 g glucose/100 ml = 1 g C / 100 ml

1 ml standard = 0.01 g C => 0.01 g C * 100 / 1 g = 1.00 % org. C

NOTE: If the org. C contents of a sample exceeds 3 %, it must be repeated with **0.5 g** of soil.

The standard curve corresponds then with:

1.0, 2.0, 4.0, 8.0 % org. C

For samples with more than 8% org C different methods (for example dry combustion method) must be applied.

CEC-DETERMINATION

Equipment:

Automatic Extractor, 24 place,"Concept Engineering,Inc." with:

Syringes,disposable,polypropylene,60 ml,'Monoject' 'Reservoir syringes' with rubber stopper attached to fit into 'Sample syringes'
Rubber tubing cut in short lengths for connecting syringe barrels

Analytical filter pulp, i.e. Schleicher and Schuell, No.289 or grade 32-17 from Centurion Int.

For heavy soils: 'Celite', analytical filter aid or acid washed quartz sand.

Kjeltec distilling unit with storage tank for concentrated NaOH-solution

Automatic titration unit (Schott TR 156)

Digestion tubes, 250 ml
Conical flasks, 250 ml
Volumetric flasks, 100 ml

Reagents:

Ammonium acetate
Acetic acid
Ammonia solution
Boric acid
Sodium hydroxide flakes
Volumetric solution of sulphuric- or hydrochloric acid
Ethanol 96% (or methylated spirit)
Ethanol 48% (or methylated spirit)
Potassium chloride
Hydrochloric acid 36%
All water is distilled or deionised water

Preparation of solutions:

1 normal, neutral ammonium acetate :

1. Dissolve 1926.8 g ammonium acetate in 5 liters water. Make up to 25 liters.
2. Adjust pH to 7.0 with either acetic acid (if pH>7.0) or ammonia solution (if pH<7.0).

1 normal, acidified potassium chloride :

1. Dissolve 1863.9 g potassium chloride (analytical grade) in about 5 l water.
2. Add 125 ml 36% Hydrochloric acid and make up to 25 liter.

4% boric acid solution:

1. Add 200 g of boric acid powder to about 1.5 liters water.
2. Heat to near boiling and stir until dissolved.
3. Transfer hot through a large polyethylene funnel into a 5 litre polyethylene bottle containing about 2 liters of water.
4. Add a few ml of 'mixed indicator', make up to 5 liters and mix well.

Mixed indicator solution :

Dissolve 0.5 g bromcresol green and 0.1 g methyl red in 100 ml 96% alcohol

50% sodium hydroxide solution:

1. Weigh 2500 g sodium hydroxide flakes in a 5 litre beaker.
2. **Carefully (!)** add water up to a volume of 5000 ml.
3. Stir until dissolved and let cool.
4. Fill into storage tank of Kjeltec distilling unit.
5. Repeat points 1 - 4. (Tank has 10 liters storage capacity.)

NOTE: Fume cupboard, coat, gloves and goggles are imperative!

Sulphuric or hydrochloric acid 0.01 n:

Dilute a volumetric solution accordingly.

(If volumetric solution is 1 normal pipette 10 ml into a 1 l volumetric flask and fill with water to about 2 cm below the mark. Put flask in waterbath at 20° C for 30 minutes. Then fill up to mark.)

Ethanol 48 % :

Mix equal amounts of ethanol 96 % and water.

Ammonium chloride solution 50 meq / l :

Weigh 1.59 g of dry ammonium chloride into a 500 ml volumetric flask and make to mark with 1 N KCl solution (used for leaching).

Ammonium chloride solution 5 meq / 1 :

Pipette 50 ml of the 50 meq ammonium chloride solution into a 500 ml volumetric flask and make to mark with 1 N KCl solution (used for leaching).

Procedure:

1. Prepare sample extraction tubes by placing approximately 1 g of filter pulp formed into a ball in a syringe barrel.
2. Force it down tightly with plunger.
3. Weigh 2.5 g of soil sample. If sample is rich in clay and/or saline, mix well with one 10 ml scoop sand or celite. Fill mixture into tube.

   NOTE: A blank should be prepared with every batch. This is imperative if samples have been mixed with sand or celite because both adsorb a certain amount of ammonium which must be subtracted from the result.
4. Place sample tubes in upper disk of extractor and connect to inverted syringe. The plunger of this syringe is inserted in slot of stationary disk of extractor.
5. If the sample contains soluble salts (el. conductivity > 0.5 mS/cm), a prewashing with 48 % ethanol should be done : same procedure as for ammonium acetate.
6. Fill sample tube to the 15 ml mark with ammonium acetate. Stir carefully to release all enclosed air bubbles from the sample. Rinse the stirring rod and the sides of the tube with ammonium acetate from a wash bottle not exceeding the 25 ml mark.
7. Set extractor to 2 hours extracting time.
8. Add about 45 ml ammonium acetate solution to reservoir tube, start extractor.
9. At the end of extraction the extractor stops automatically by a micro switch. Switch extractor off and pull plungers down as far as extractor will allow.
10. Disconnect syringes from sample tubes (leaving rubber connector on sample tube) and fill contents into numbered 100 ml volumetric flasks.
11. Fill syringes again to 35-45 ml with ammonium acetate solution from a beaker and fill also into volumetric flask. Fill up to volume with ammonium acetate from a wash bottle. Mix well.
12. Keep the extracts for exchangeable cations determination.
13. Return upper 2-disc unit to starting position.
14. Attach syringes to sample tubes, rinse sides of sample tubes with ethanol and fill to 20 ml mark.

15. Place reservoir tubes in sample tubes. Fill to about 45 ml mark with ethanol (The total volume of ethanol should be just a little less than 60 ml).

16. After extractor has stopped, turn it off, pull plungers down, remove syringes and discard ethanol wash.

17. Fill syringes with acidified 1 N potassium chloride solution and extract. Collect extract in 100 ml volumetric flasks. Fill up to mark with potassium chloride solution and shake well to mix.

18. Pipette a 50 ml aliquot KCl extract to digestion tubes.

19. Remove sample tubes and transfer samples to the digestion tubes of the distilling unit.

20. Fill 25 ml of the boric acid solution into 250 ml conical flasks.

21. Switch Kjeltec I distilling unit on.

22. Place an empty digestion tube and a beaker in position.

23. Close valve of steam generator outlet at the back of the unit.

24. Open steam valve. Open tap until water level reaches the electrodes in steam generator. Close tap.

25. When water starts boiling open tap again. Regulate water current such that the back flowing water (coming from condenser as cooling water) is just hand warm (4-5 liters per minute).

26. Distill about 150 ml into beaker. Close steam valve.

27. Place a tube containing 50 ml of 5 meq/l ammonium chloride and the first conical flask with boric acid in position. Add 50 % NaOH by pulling the lever of the alkali pump.

28. Open steam valve and distill 4-5 minutes (collect 125-150 ml distillate). Close steam valve. Remove tube and conical flask.

29. Place tube with first sample and second conical flask with boric acid in position. Add 50% sodium hydroxide solution by pulling the lever of the 'alkali pump' etc. etc. etc.

30. During second distillation titrate first sample with 0.01 N H_2SO_4 or HCl. If the result of titration is not close to 12.5 ml, the test has to be repeated until a satisfactory result is obtained.

31. At the endpoint of the titration the green color disappears completely. The solution looks grey. One drop more gives pink. (If an automatic titrator is used, set the end point to pH 4.0.)

Calculations:

Calculation, storing and printing of soil analytical data is done with the Soil Laboratory Database system on a IBM compatible micro computer.

CEC in milliequivalent per 100 g soil is calculated as follows:

CEC = (t-b)*N*extract/weight* extract/aliquot

t = ml acid used for titration
b = ml blanc titration
N = normality of acid (= 0.01)
extract = 100 ml
weight = 2.5 g
aliquot = 50 ml

=> CEC [meq/100g soil] = (t-b)*0.01*100/2.5 * 100/50

=> **CEC [meq/100g soil] = (t-b) * 0.8**

EXCHANGEABLE CATIONS DETERMINATION

Equipment:

100 ml volumetric flasks
SD-1 diluter
50 ml polyethylene containers or similar
Varian Atomic Absorption Spectrometer

Reagents:

Strontium chloride	$SrCl_2*6H_2O$
or	
Lanthanum chloride	$LaCl_3*7H_2O$
Calcium carbonate	$CaCO_3$
Magnesium sulfate	$MgSO_4*7H_2O$
Sodium chloride	NaCl
Potassium chloride	KCl

Interelement effects during Ca and Mg determination:

Interelement effects due to aluminum and silicon and ionization of calcium in the flame might cause problems in the determination. Ionization of calcium and influence of aluminum in the Mg-determination can be reduced by adding strontium- or lanthanumchloride to standard- and measuring solutions (or by measuring in a nitrous oxide - acetylene flame).

Preparation of solutions :

100 000 ppm Sr or La concentrated stock solution

Dissolve 1521.5 g $SrCl_2*6H_2O$ in 5 liters dist. water.(For better solubility add some conc. HCl). ($SrCl_2*6H_2O$=32.9% Sr)

or

Dissolve 1338.7 g $LaCl_3*7H_2O$ in 5 liters dist. water. (For better solubility add some conc. HCL). ($LaCl_3*7H_2O$=37.4% La).

2000 ppm working solution

Pipette 100 ml of concentrated stock solution into a 5 litre polyethylene bottle and dilute to 5 liters with distilled water.

Combined stock solution 200meq/l Ca + 50meq/l Mg

1. Dry $CaCO_3$ and $MgSO_4*7H_2O$ at 100° Celsius in drying oven.

2. Weigh 10.090 g $CaCO_3$
   and 6.162 g $MgSO_4*7H_2O$

   into a 1000 ml volumetric flask.

3. Fill about half with dist. water.

4. Add just enough HCl to dissolve the calcium carbonate.

5. Fill up to 2 cm below mark and adjust temperature to 20° C in a waterbath.

6. After 30 minutes fill up to mark and shake well.

Diluted Ca/Mg stock solution (1:5)

1. Pipette 100 ml stock solution into 500 ml volumetric flask.

2. Fill to about 2 cm below mark with 1 N ammonium acetate solution (from CEC determination).

3. Adjust temperature to 20° celsius in waterbath.

4. After 30 minutes fill up to mark, stopper flask and shake well.

   This solution contains: <u>40 meq Ca/l and 10 meq Mg/l</u>.

Ca/Mg standard series

1. Pipette: 5, 10, 20, 25, 50 ml

   of the diluted stock solution into 100 ml volumetric flasks.

2. Fill to about 1 cm below the mark with 1 N ammonium acetate solution.

3. Adjust temperature to 20° C in waterbath.

4. After 30 minutes fill up to mark, stopper flasks and shake well.

   These solutions contain:

   1st. Standard 2 meq/l Ca and 0.5 meq/l Mg
   2nd. Standard 4 meq/l Ca and 1.0 meq/l Mg
   3rd. Standard 8 meq/l Ca and 2.0 meq/l Mg
   4th. Standard 10 meq/l Ca and 2.5 meq/l Mg
   5th. Standard 20 meq/l Ca and 5.0 meq/l Mg

Combined stock solution 50 meq/l K + 50 meq/l Na

1. Dry KCl and NaCl at 100° Celsius in drying oven.
2. Weigh 3.7278 g KCl
   and 2.9220 g NaCl

   into a 1000 ml volumetric flask
3. Fill up to about 2 cm below mark with dist. water.
4. Adjust temperature to 20° C in waterbath.
5. After 30 minutes fill to mark, stopper flask and shake well.
6. Store in polyethylene bottle.

Diluted K/Na stock solution (1:25)

1. Pipette 20 ml of stock solution into 500 ml volumetric flask.
2. Fill with 1 n ammonium acetate solution (used for CEC determination) to about 2 cm below mark.
3. Adjust temperature to 20° C in waterbath.
4. Fill to mark after 30 minutes, stopper flask and shake well.

   The solution contains: 2 meq/l K and 2 meq/l Na

K/Na standard series

1. Pipette: 5, 10, 20, 25, 50 ml

   of diluted stock solution into 100 ml volumetric flasks.
2. Fill to about 1 cm below mark with 1 N ammonium acetate solution (used for CEC determination)

The solutions contain:

1. Standard 0.1 meq/l K + Na
2. Standard 0.2 meq/l K + Na
3. Standard 0.4 meq/l K + Na
4. Standard 0.5 meq/l K + Na
5. Standard 1.0 meq/l K + Na

Ca/Mg and K/Na with Atomic Absorption Spectrometer:

In order to avoid too high absorbencies dilute samples and standards:

Ca/Mg-determination by factor 50
K /Na-determination by factor 10

Connect the SD-1 diluter set to a dilution ratio of 1:50 to the 2000 ppm lanthanum chloride solution. (1 ml sample uptake + 49 ml dispension.)

Normally Potassium and Sodium should be measured on a flame photometer. Only when this instrument is not available or when concentrations are to low for detection (which is not the case for soils) should the AAS be used.

Connect the diluter set to a ratio of 1:10 to a drum with distilled water. (5 ml sample uptake + 45 ml dist. water.)

In the literature a 1000 ppm cesium chloride solution is recommended to suppress partial ionization of K and Na. Tests showed that the effect lies within the error range of the determination. The costs for the very expensive chemically pure cesium chloride can be saved.

Dilute ammonium acetate extracts from the CEC determination and standards into polyethylene containers. Use dilute ammonium acetate solution as a blank. Swirl containers to mix. Then fill solutions into vials fitting the automatic sample changer.

Preparation of the Varian AA1275 Atomic Absorption Spectrometer:

Setting of acetylene gas pressure:

a. Slowly open main valve of gas cylinder.

b. Check that main pressure is more than 700 kpa. (If pressure is below exchange gas cylinder because acetone may be formed and can come into the system.)

c. Check that gas pressure into system is 70 kpa. (At lower or higher pressure flame might not get started)

Operating modes:

The Varian Atomic Absorption Spectrometer can be operated in several modes.

a. Fully automatic with automatic sample changer and the Hewlett Packart HP86B micro computer as controller and receiver of the data.

b. Half automatic without sample changer but controlling and data handling by the computer.

c. Half automatic with sample changer but without computer.

d. Manual without sample changer and computer.

a. Full automatic operating mode:

Switch system on in the following sequence:

1. Uninterruptable power supply
2. Spectrometer
3. Exhaust fan (and compressor, if there is no compressed air)
4. Sample changer
5. Printer (make sure that printer is connected to spectrometer and **not** to computer !)
6. Monitor
7. Disc drive
8. Insert HP86/AA1275 - system disc into drive 0 and data disk into drive 1
9. computer

The 'Autostart' program will start up computer. For the further procedure follow instructions displayed on monitor.

In the 'Master Index' section press '4' to enter sample labels. Then press '2' to operate system.

In the 'Optimize Instrument' section select lamp and set wavelength of element to measure.

Set: Ca-wavelength(422.6 nm)
Mg-wavelength(285.2 nm)
K -wavelength(766.5 nm)
Na-wavelength(589.0 nm)

NOTE: Lamp alignment must be checked regularly by senior technician.

Enter 'Y' at the question 'Do you want auto set up?'.
Enter '1' to have recommended standard settings for spectrometer and sample changer programmed. (In this programme section the keyboards of spectrometer and sample changer are unlocked so that both could also be programmed manually.)

Recommended standard settings:

Spectrometer:	Sample changer:
Lamp 1 (Ca/Mg): 5 mA	No. standards :5
Lamp 2 (K /Na): 10 mA	Rinse rate : 0
Integration time: 1 sec	Rinse time : 5
	Delay time : 5
Absorbance mode (ABS)	Multiples : 4
Integration hold	Reslope rate : 6

Continue following instructions displayed on monitor.

Press 'FLAME ON'. Flame must be light blue to colorless with a sharp light blue rim at the burner. If there are edges in the flame switch off and clean burner slit with a brass blade.

Check whether highest standard is within measuring range. If not, tilt burner until needle moves into left most field of peakmeter. (This is necessary for Magnesium using the combined Ca/Mg-standards).

Start automatic run. The end of a run is signalled by a continuous beep from the spectrometer. The system automatically resets. Prepare determination of next element.

NOTE: The setting of the system parameters does not have to be repeated. Press 'N' or 'Spacebar' at the question in the 'Optimize Instrument' section.

After determination of all elements go to 'Master Index' section. Press '6' (transfer data programm) to transfer the data into the soil survey data base.

b. Half automatic operating mode without sample changer but with computer as controller:

The same software which controls the full automatic operating mode also handles the half automatic mode without sample changer. The setting of the spectrometer is done either manually or by software using the same recommended parameters as above.

Change samples by hand. To take readings press 'Spacebar', to proceed to next sample press 'CONT'.

Transfer data into soil survey data base the same way as above.

c. Half automatic operating mode without computer but with sample changer:

Make sure that sample changer is in 'Auto sample' mode and not in 'Single sample' mode.

Loop through parameters listing pressing repeatedly 'Programm set up', change parameters using numeric keyboard.

Set spectrometer to recommended parameters. (Spectrometer must be in 'ABS'-mode.)

Load sample changer with samples.

Press 'Start' to start run.

d. Manual operating mode without computer and sample changer:

Operating the spectrometer manually offers a variety of measuring modes:

Readings are displayed and printed either in absorbance or in concentration mode. In concentration mode three standards are allowed from which the concentration of the samples is calculated.

The samples are measured either in 'Integration hold' or in 'Run mean' mode. In 'integration hold' mode impulses are integrated over a certain set time. In 'run mean' mode an accumulative mean is calculated of readings taken between start reading and pressing the 'read' key a second time to end.

For further information refer to the operating handbook for the spectrometer.

Calculations:

In case only absorbancies have been measured and no linear regression curve of the standards can be calculated concentrations in meq/100 g soil are calculated as follows:

Dimension of standards is : **meq / liter**

The concentration **C** in meq/l of an element in a sample is:

$$C = E / e$$

E = Extinction (absorption)
e = Extinction coefficient

The extinction coefficient is calculated from the standard curve:

$$e = (E1/C1 + E2/C2 + E3/C3 + \ldots + En/Cn) / n$$

E1, E2, E3, ... , En = Absorbance readings of standards
C1, C2, C3, ... , Cn = Concentrations of standards
n = Number of standards

The concentration K of a sample in meq / 100 g soil is:

$$K = C * \text{dilution} * \text{extract} / \text{weight} / 10$$

Extract = 100 ml
Weight = 2.5 g

$$\mathbf{K = C * dilution * 4}$$

K/Na determination with 'CORNING 400' flamephotometer:

The emission/concentration relation is linear only up to a certain concentration. Therefore it is advisable to dilute samples and standards for the potassium determination by factor 5.

The concentration of the Na-standards is low enough to stay in the linear range of the emission/concentration function.

Set the SD-1 diluter to a dilution ratio of 1:5 (8 ml solution + 32 ml dist. water).

1. For K-determination dilute ammonium acetate extracts from CEC determinations and standards into plastic containers. For blank dilute ammonium acetate solution.
2. For Na-determination transfer the extracts and standards without dilution into the containers. For blank use undiluted ammonium acetate solution.
3. Switch on power of the Corning 400 flamephotometer.
4. Open the gas tap. Switch 'Fuel - on'. Open window to the burner chamber and press 'ignition' until flame is burning.
5. Set the filter to 'K' (for potassium).
6. Insert tube into blank and set to 0 with 'blank' regulator. Then insert tube into highest standard and set to 100 with 'sensitivity - coarse - fine'. Repeat blank setting, repeat sensitivity setting until readings are constant.
7. Measure standard curve, then measure samples. Record the values on the appropriate form.
8. When all samples are determined set filter to 'Na' (for sodium) and calibrate the instrument as above (point 6).
9. Measure standard curve and samples and record the values.
10. For closing down first close gas tap, then switch 'fuel - off', then 'power-off'.

CARBONATES DETERMINATION

Equipment:

5 litre conical flask
wide mouth shaking bottles + cap
reciprocal shaker
100 ml erlenmeyer flasks
burette (50 or 100 ml)
100 ml pipette or dispensor

Reagents :

Hydrochloric acid	HCl
Sodium hydroxide	NaOH
Phenolphthalein	

standardized HCl <u>or</u> sodium hydrogen phthalate

All water is distilled or deionized water

Preparation of solutions :

Hydrochloric acid 1 M

1. Add approximately 4 liters of water to a 5 liter conical flask.
2. Slowly add 425 ml of concentrated HCl under constant stirring.
3. Cool and add water to the 5 liter mark.

Sodium hydrogen phthalate 2 meq

1. Weigh 0.7 g of sodium hydrogen phthalate into a beaker and dry for 2 hours in an oven at 120 ° Celsius.
2. Cool in a desiccator and weigh 0.448 g (= 2 meq) into a 100 ml erlenmeyer. Dissolve in approximately 20 ml of water.

Sodium hydroxide 0.500 M standard solution

1. Dissolve 20 g NaOH pellets in 1 liter water.
2. Cool and standardize immediately before use against 1 M HCl standard solution from an ampoule or against sodium hydrogen phthalate.
   with 1 M HCl : normality NaOH = ml HCl / ml NaOH
   with phthalate : normality NaOH = 2 / ml NaOH

Phenolphthalein indicator solution

1. Dissolve 100 mg of phenolphthalein in 100 ml ethanol 96 %.

Procedure :

1. Weigh 5.00 g of sample into a shaking bottle. Do not forget to include 2 blanks and the standard sample for carbonates.
2. Add 100 ml of the approximately 1 M HCl with a pipette or dispenser and swirl.
3. Loosely screw the lid on (do not tighten, to allow CO_2 to escape). Swirl occasionally and let stand overnight.
4. Next morning, indent the bottle by hand, tighten the lid and shake for 2 hours.
5. Allow the suspension to settle and pipette 10 ml of the supernatant solution into a 100 ml erlenmeyer flask. Add 25 ml of water and 3 drops of phenolphthalein.
6. Titrate with 0.5 M NaOH to a permanently pink endpoint.

Calculation :

Calculation, storing and printing of soil analytical data is done with the Soil Laboratory Database system on a IBM compatible micro computer.

$$\% \ CaCO_3 = 50 * \text{normality NaOH} * (a - b) / s$$

where : 50 = molecular weight of $CaCO_3$
a = ml NaOH used for the blank
b = ml NaOH used for the sample
s = grams of sample used

Reference

Labex method L8703-41-1

EXCHANGEABLE ACIDITY

Equipment :

100 ml volumetric flasks
drum for KCl solution
extractor
filter pulp
burette or automatic titrator

Reagents :

Potassium Chloride	KCl
Sodium hydroxide	NaOH
Phenolphthalein	

Preparation of solutions :

1 Normal Potassium Chloride (extracting solution)

Dissolve 373 g KCl in water and make to 5 liter.

0.1 % phenolphthalein indicator solution

Dissolve 100 mg phenolphthalein in 100 ml ethanol 96 %.

0.025 Normal Sodium hydroxide

a. pipette 25 ml of a 1 N NaOH standard solution and make to 1 liter.

or

b. dissolve approximately 4 g of NaOH pellets in water and make to 1 liter. Standardize against 0.05 N HCl.

Procedure :

1. Place approximately 1 g of filter pulp into each extraction tube.
2. Weigh 10.0 g of sample and transfer to the extraction tube. Do not forget the standard sample and 2 blanks.
3. Assemble syringes and plungers.
4. Add approximately 10 ml of 1 N KCl to each sample.
5. Place a reservoir tube above each extraction tube.
6. Fill the reservoir tubes with approximately 45 ml KCl.

7. Set the extractor time for about 2.5 hours and start the extractor.

8. After completion of the extraction, transfer the extract to 100 ml volumetric flasks.

9. Rinse the syringes with 1 N KCl and make up to volume.

10. Pipette 25 ml of the extract into a 150 ml erlenmeyer and add 2 - 3 drops of phenolphthalein.

11. Titrate with 0.025 N NaOH until the color turns just permanently pink.
    (when using the automatic titrator, set the endpoint to PH=7.80). Sometimes the pink color can be very weak. In that case add another drop of phenolphthalein.

Calculation :

Calculation, storing and printing of soil analytical data is done with the Soil Laboratory Database system on a IBM compatible micro computer.

The exchangeable acidity is expressed in milliequivalent per 100 g of soil.

It is calculated as follows : Exac = (a - b) * 0.025 * 4 * 10

in which : a = ml NaOH needed for sample
b = ml NaOH needed for blank
0.025 = normality of Na
4 = factor for titrating 25 ml out of 100
10 = factor for taking 10 g of soil instead of 100

Reference :

Labex method L8703-26-1

PARTICLE SIZE DETERMINATION

Equipment:

1000 ml or 800 ml beakers
watch glasses fitting beakers
rubber policemen for cleaning beakers
1000 ml plastic bottles with tight closing lid
50 ml measuring cylinder
500 ml measuring cylinder
1000 ml measuring cylinders without spout
Stopper to close cylinders
3 Soil hydrometers

Waterbath with thermostat controlled cooler/heater
Sample shaker
2-decimal place balance
Stopwatch

Sieves 53, 106, 250, 500, 1000 µm
Sieve 53 µm for wet sieving
Sieve shaker
Soft brushes for cleaning sieves
A2 paper with smooth surface

Reagents:

Sodium acetate	$NaC_2H_3O_2*3H_2O$
Acetic acid glacial	CH_3-COOH
Hydrogen peroxide 30%	H_2O_2
Sodium citrate	$Na_3C_6H_5O_7*2H_2O$
Sodium bicarbonate	$NaHCO_3$
Sodium dithionite	$Na_2S_2O_4$
Sodium chloride	$NaCl$
Acetone	
Sodium hexa-metaphosphate	$(NaPO_3)_6$
Sodium carbonate	Na_2CO_3

Preparation of solutions:

1 normal sodium acetate solution pH 5.0:

Dissolve 3400 g sodium acetate and dilute to 25 liters in drum. Adjust to pH 5.0 with acetic acid.

0.3 normal sodium citrate solution:

Dissolve 1305 g sodium citrate and dilute to 25 liters in drum.

1 normal sodium-bi-carbonate solution:

Dissolve 2100 g sodium bicarbonate and dilute to 25 liters in drum.

Sodium hexa-metaphosphate solution (dispersing solution) :

Mix 165 g dry sodium hexa-metaphosphate with 35 g sodium carbonate powder in 5000 ml beaker. Fill up with water and stir until dissolved. Then transfer into storage bottle.

Pretreatment of samples:

NOTE: Removal of carbonates and free iron oxides is optional.

Removal of carbonates (if present):(pH>6.8)

1. Weigh depending on clay content 50-100 g soil into 800 ml beaker.
2. Add 250 -500 ml 1 N Na-acetate solution.
3. Heat on water bath to boiling. After effervescence has stopped, add increments of about 25 ml until it does not recur.
   (In case of very high carbonate contents 5 ml increments of glacial acetic acid can be used instead of buffer. In this case monitor the pH with indicator paper.)
4. Add 500 ml of water and add a few ml saturated NaCl solution for precipitation.
5. Let stand over-night and siphon off or centrifuge.

Removal of soluble salts (if present: pH>8.2 and EC>4 mS/cm)

If no carbonates are present but the electrical conductivity exceeds 4 mS/cm, respectively 400 mS/m soluble salts should be washed out:

1. Add approximately 200 ml Water to the samples, let stand for about two hours, stir occasionally.
2. For precipitation add few ml saturated sodium chloride solution or acetone.
3. Centrifuge excess water, then continue with normal procedure.

Oxidation of organic matter (if present: org. C > 0.3 %)

Only if the organic Carbon content exceeds 0.3 % must organic matter be removed.

1. Add 50 ml water and 15 ml of H_2O_2 (30%) (In case of buffer pretreatment less or no water need to be added). Let stand overnight.

2. Place on hotplate and warm to about 50° Celsius. When the frothing has calmed down, gradually increase the temperature of the hotplate to boiling point.

3. If necessary, add small increments (2-5 ml) of 30% hydrogen peroxide until decomposition of organic matter is completed. The soils of Botswana generally contain low organic matter contents, so it should not be necessary to add much extra peroxide. In doubt check the organic carbon content that was determined.

4. Too strong frothing can be tempered by removing from hotplate (cooling in cold water) and/or adding some water.

5. In general supernatant solution looks clear and colorless after decomposition of organic matter (although for example iron in gleysols can cause a yellow or green color).

6. To remove the remaining hydrogen peroxide add about 100 ml of water and 5 drops of ammonia. Boil for about half an hour without cover.

7. If no iron coatings are to be removed, transfer samples into shaking bottles.

Removal of iron oxides:

1. Add 500 ml 0.3 n Na-citrate and 70 ml 1 n Na-bicarbonate solution.

2. Warm the suspension to 80° Celsius ; then add 2-10 ml scoops of solid sodium dithionite.

3. Stir suspension constantly for 1 minute and occasionally for 15 minutes. (Avoid heating above 80° Celsius because iron sulfite (FeS) forms.)

4. Add 50 ml saturated NaCl solution to flocculate.

   (If suspension fails to flocculate, add 50 ml acetone.)

5. Siphon supernatant solution off, or centrifuge.

6. Wash with Na-citrate (add NaCl or acetone if necessary for flocculation.)

7. Siphon or centrifuge again, then transfer samples into shaking bottles.

Procedure:

Dispersion of soil particles:

1. After transfering samples into 1 l shaking bottles add 50 ml sodium hexa-metaphosphate solution with a measuring cylinder. Make up volume to about 400 ml with water. (Shaking bottles must be half full to ensure proper dispersion. Do not over fill.)

2. Put samples on shaker and shake for about 16 hours (overnight) at a speed of about 125 strokes per minute.

Clay and silt determination by hydrometer method:

1. Transfer samples quantitatively from shaking bottles into 1 l measuring cylinders. Fill up to 1 l with water.

2. **a.** place in water bath (temperature at 20° Celsius). If necessary let stand for about 30 minutes to 1 hour to equilibrate temperature.
   or
   b. regulate the room temperature to about 20° Celsius. If the sample temperature is not more than 1° Celsius higher or lower than the room temperature, the sedimentation process can be started.

3. Take hydrometer readings for a particular sample after:

   40 seconds, 4 minutes, 4 hours

4. Close the first cylinder with rubber stopper. Shake vigorously end over end for about 1 minute. Make sure that the sample is completely dispersed.

5. Put cylinder down, start stopwatch and insert hydrometer carefully. Take first reading 40 seconds after sedimentation has started.

6. Close second cylinder, shake and put down after precisely 2 min total time on stopwatch. Insert hydrometer and read after 2 min 40 sec total time.

7. At 4 minutes take second reading of first sample. At 6 minutes take second reading of second sample.
   During waiting shake third cylinder already. Put cylinder down after 8 minutes total time. Take first reading after 8 min 40 sec.

8. Shake fourth cylinder and put down at 10 min, read at 10 min 40 sec, wait until 12 min, take 2nd reading of third sample, wait until 14 min, take 2nd reading of fourth sample, and so on.

9. Take the third reading after 4 hours settling time.

10. For starting / reading times for 24 samples see table 2.

Separation of sand fraction from clay and silt:

1. Place a 53 micron test sieve in a large funnel into a sink equipped with a 'soil trap' to avoid blockage of the drainage pipes.
   NOTE: Check sieve regularly for holes and tightness along the rim.

2. Transfer sample from measuring cylinder on to sieve and rinse thoroughly with water until all clay and silt fraction is washed out. (The water jet should not be too strong to avoid loss of sand fraction due to splashing).

3. Wash sand fraction from sieve into beaker.

4. Place beaker on hot plate or drying oven to evaporate water.

Dry sieving of sand fraction:

1. Stack five test sieves 53, 106, 250, 500, 1000 micro m mesh size and a receiver pan (mesh size decreasing from top downwards).

2. Transfer sand fraction onto the top sieve (1000 micro m) and put onto sieve shaker.

3. Shake for about 5 min at an amplitude of 2.5-3 (amplitude scale is on sieve head).

4. Brush the fractions on separate large sheets of paper with smooth surface.

5. Clean sieves thoroughly. (Do not use brass or steel brushes for sieves with mesh size smaller than 500 micron).

6. Tare beaker then add fractions starting with receiver, then 53 micro m, 106 micro m, ...

7. Record each weight in hundredths of a gram.

Calculations:

Sand Fraction:

weight 53 micron fraction = weight 53 - receiver
weight 106 micron fraction = weight 106 - weight 53
weight 250 micron fraction = weight 250 - weight 106
weight 500 micron fraction = weight 500 - weight 250
weight 1000 micron fraction = weight 1000 - weight 500

weight % of fraction = weight of fraction x 100 / total weight

Total % sand = sum weight % of all sand fractions

(receiver fraction is considered to be coarse silt !)

Clay and silt fraction:

Corrected reading = reading - blank + (temp-20) * 0.36

(Temperature correction: 0.36 is an empiric factor used in literature. It is valid from soils of an average composition only. If temperature of waterbath is kept at 20° Celsius, temperature correction is 0).

(Total clay + silt) % = corr. first reading * 100 / total weight.

A correction factor can be calculated to make:

% Sand + % clay + % silt= 100 %

Let:
SS = % sand from sieving
HY = (clay + silt) %

Corr. factor = 100 / (HY + SS)

This correction factor is based on the assumption that all fractions contribute equally to the error in the determination. It equalizes minor reading or rounding errors since it is difficult to read a hydrometer more precisely than one unit, it accounts for loss of the sand fraction during sieving and the fact that the actual moisture content need not be close to zero.

If the result is rejected because of to high a correction factor, it may be due to the moisture content of the sample. Especially samples high in organic matter and/or clay may have considerable moisture contents. Moisture content should than be determined for these samples.

If the fraction remaining in the pan is important (more than 1 gram) this may lead to bad results. (there is not supposed to be any silt and clay among the fraction that is sieved, however it is possible that sand size particles break up and pass the 53 micron sieve). The result may than be recalculated as if the fraction in the pan is 0.

The correction factor using soil hydrometers should be in the range of 0.94 to 1.06, the computer will otherwise automatically reject the result.

Values outside this range indicate errors in the determination.

Let: cr1 = corrected first reading (40 seconds)
cr2 = corrected second reading (4 minutes)
cr3 = corrected third reading (4 hours)
wt = sample weight
vol = volume of the dispersion
wtr = weight % receiver fraction

% coarse silt = ((cr1 - cr2) * vol / wt) * corr. factor / 10.

% fine silt = ((cr2 - cr3) * vol /wt) * corr.factor / 10.

% clay = (cr3 * vol / wt) * corr. factor / 10.

MOISTURE DETERMINATION

Equipment :

numbered aluminum containers with lid
drying oven
2 decimal place balance

Procedure :

1. Record the weight of the empty containers.
2. Add approximately 25 grams of sample to each container and record the weight of container + sample.
3. Allow to dry in the oven at 105° C overnight.
4. The next day close the lid, remove from the oven and allow to cool down for 10 - 15 minutes.
5. Record the weight of container + sample after drying.

Calculation :

The moisture content will be calculated by the computer under the function for input of particle size data.

it is calculated as follows :

$$m = 100*(b-c)/(b-a)$$

where : m = moisture content in %
a = weight of the empty container
b = weight of the container + sample before drying
c = weight of the container + sample after drying

Table 2: Time table for hydrometer readings

Start stopwatch
Time : 0:00’00" hr:min’sec"

	Sample 1	Sample 2	Sample 3	Sample 4
shake until:	00"	02’00"	08’00"	10’00"
1. reading	40"	02’40	08’40"	10’40"
2. reading	04’00"	06’00"	12’00"	14’00"
3. reading	4:00’00"	4:02’00"	4:08’00"	4:10’00"

	Sample 5	Sample 6	Sample 7	Sample 8
shake until	16’00"	18’00"	24’00"	26’00"
1. reading	16’40"	18’40"	24’40"	26’40"
2. reading	20’00"	22’00"	28’00"	30’00"
3. reading	4:16’00"	4:18’00"	4:24’00"	4:26’00"

	Sample 9	Sample 10	Sample 11	Sample 12
shake until	32’00"	34’00"	40’00"	42’00"
1. reading	32’40"	34’40"	40’40"	42’40"
2. reading	36’00"	38’00"	44’00"	46’00"
3. reading	4:32’00"	4:34’00"	4:40’00"	4:42’00"

	Sample 13	Sample 14	Sample 15	Sample 16
shake until	48’00"	50’00"	56’00"	58’00"
1. reading	48’40"	50’40"	56’40"	58’40"
2. reading	52’00"	54’00"	1:00’00"	1:02’00"
3. reading	4:48’00"	4:50’00"	4:56’00"	4:58’00"

	Sample 17	Sample 18	Sample 19	Sample 20
shake until	1:04’00"	1:06’00"	1:08’00"	1:10’00"
1. reading	1:04’40"	1:06’40"	1:08’40"	1:10’40"
2. reading	1:08’00"	1:10’00"	1:12’00"	1:14’00"
3. reading	5:04’00"	5:06’00"	5:08’00"	5:10’00"

	Sample 21	Sample 22	Sample 23	Sample 24
shake until	1:16’00"	1:18’00"	1:24’00"	1:26’00"
1. reading	1:16’40"	1:18’40"	1:24’40"	1:26’40"
2. reading	1:20’40"	1:22’00"	1:28’00"	1:30’00"
3. reading	5:16’00"	5:18’00"	5:24’00"	5:26’00"

USEFUL ADRESSES

Part of the equipment used in the laboratory is purchased from overseas. In case of restocking spare parts, breakdowns or replacement here some adresses:

Dispensers/diluters, multisample stirrer, superfloc, etc.:

Custom Laboratory Equipment Inc.
P.O. Box 757
205 E. Michigan Avenue

Orange City, Florida 32763
USA

Telephone: 904-775-2322
Telex: none

Contact: Dr. Alan B. Hunter

Superfloc:

American Cyanamid Company
P.O. Box 32787

Charlotte, North Carolina 28232
USA

Automatic extractor:

Centurion International Inc.
P.O. Box 82846
4555 North 48th Street

Lincoln, Nebraska 68501
USA

Telephone: 402-467-4491
Telex: 48-4377 CENTURION LCN

Contact: Debra Hilde

Barnstead water purification system, steam washer, ultrasound bath:

Curtin Matheson Scientific International
9999 Stuebner Airline

Houston, Texas 77038
USA

Telephone: 713-820-9898
Telex: 46-20313

Contact: Williams

Kjeltec distiller:

Tecator AB
P.O. Box 70

S-263 01 Hoeganaes
Sweden

Telephone: 042-42330
Telex: 72 695

Automatic titrator, pH-electrodes, conductivity cell:

Schott Geraete GmbH
P.O. Box 1130
Im Langgewann 5

D-6238 Hofheim a.Ts
Fed. Rep. Germany

Telephone: 06192-8081
Telex: 410471

or

Klever instruments
P.O. Box 483

Jukskei Park 2158
South Africa

Telephone: 011-704-3017/8
Telex:

Atomic Absorption Spectrometer:

Varian Techtron Pty. Ltd.

Springvale
Australia

Telephone:
Telex:

or

SMM Instruments
P.O. Box 38622

Booysens 2016
South Africa

Telephone: 434-1010
Telex: 4-85881

Balances:

Sartorius GmbH
P.O. Box 3243

D-3400 Goettingen
Fed. Rep. Germany

Telephone: 0551-308 289
Telex: 96 723

or

Zeiss West Germany
P.O. Box 1561

2000 Johannesburg
South Africa

Telephone: 402-2860
Telex: 4-88346

Contact: Frank Bender

Shakers, sieve shaker, test sieves:

Labotec Pty. Ltd.
P.O. Box 43161

Industria 2042
South Africa

Telephone: 673-4140
Telex: 4-51117

Contact: Mrs. Sharon Brett

Chemicals:

Saarchem Pty. Ltd.
P.O. Box 144

Muldersdrift 1747
South Africa

Telephone: 762-5456
Telex: 4-26089

Contact: Jeff Pye

General Laboratory ware, Chemicals, Special Requests:

Quadralab Pty. Ltd.
P.O. Box 519

Bergvlei 2012
South Africa

Telephone: 786-8042/5
Telex: 4-27884

Contact: Mr. Pat Horey

Printed by Agricultural Information Services
Ministry of Agriculture
P/Bag 003, Gaborone, Botswana
12/89