World Agroforestry Centre TRANSFORMING LIVES AND LANDSCAPES	STANDARD OPERATING PROCEDURE		No of pages 5
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	Title: Soil Sample Reception and Processing at ICRAF Soil-Plant Spectral Diagnostic Laboratory		

I. SCOPE AND APPLICATION

This standard operating procedure is designed for use by ICRAF Soil-Plant Spectral Diagnostic Laboratory. The procedure covers sample reception and logging, soil sieving and weighing, soil sub-sampling, soil sample storage, archiving and disposal.

All samples received from the field and AfSIS sites through regional laboratories (sentinel sites, agronomic trials, legacy samples) will be processed using this standard operating procedure. This SOP applies to samples that are processed and sieved through 2 mm sieve prior to submission to the laboratory. This SOP describes the process of receiving, logging and processing of samples before making them available for various analyses in the laboratory.

II. RELATED SOPS

i. A separate guide is available for data management and submission

ii.

III. PRINCIPLE

Soil is an incredibly complex matrix. The properties of soil vary greatly from one point to the other under the influence of a wide range of factors including parent material, climate, slope and management. From the laboratory's perspective, soil presents a series of problems in terms of the homogenisation and pre-treatment of the sample, all of which can impact significantly on the final analytical result. There are guideline documents and standards detailing different sample preparations, but they are not definitive. Depending on the type and purpose of the analyses that are to be carried out on the samples, each laboratory selects and adopts its own sample processing procedures. Some of the analyses carried out in the ICRAF Soil-Plant Spectral Diagnostic Laboratory use a very small sample, often <1 g, and it is very difficult to achieve reproducible results unless strict standards are adhered to in the preparation of the samples and a good representative sample is obtained for analyses.

IV. EQUIPMENT

a. Drying trays
b. Oven
c. Metal boxes
d. Balance

e. Sieve 2mm
f. Wooden rolling pin

V. MATERIALS

a. Plastic sheet
b. Markers
c. Brown paper bags size 5
d. Plastic zip-lock bags
e. Particulate respirator (eg., N95 of 3M make)
f. Nitrile gloves

VI. PROCEDURES

a. Soil sample reception and logging

- Logging of samples involves transferring the researcher information on the samples material to the laboratory record sheet (Log-in form).
- Authorised samples are received at the reception area and are logged on priority basis i.e. First come first served. All authorized samples must be accompanied by electronic copy of the logging information.
- Lay out the samples received in order of labeling and check against field sheets. Samples collected within the AfSIS project have field descriptors in electronic format already entered in the AfSIS database.
- Make detailed notes in a laboratory record book of any labeling discrepancies or problems due to damaged sample bags or lost samples. In case of AfSIS samples enter them in the comments field of the logging database.
- To the extent possible, resolve the problems noticed at this stage. This is particularly important for samples that are missing or incomplete login information.
- Assign the Sample Serial Number (SSN) starting from 000,001 up to 999,999. Write the assigned SSN number on the sample bags using a permanent marker. Including the thousand separator (comma) helps in avoiding recording errors. In case of AfSIS samples, the number should be prefixed with the laboratory location code assigned by AfSIS (sel for Selien; sot for Sotuba; chi for Chitedze; iia for IIAM Maputo).
- Logging in of samples also involves inputting the requested analysis that the researcher needs to do on the samples.
- Samples collected within the AfSIS project have field descriptors in electronic format already entered in the AfSIS database.
- For foreign samples a phytosanitary certificate issued by the country of origin certifying that the samples has been inspected and certified free from any harmful diseases or pathogens is filed. See Appendix 3 for the shipping procedure to be followed.
- Next copy the field descriptors on the sample bag plus the assigned serial number to a sample logging sheet (Appendix 1). For AfSIS sentinel site samples, the field descriptors should already be in the database. For other samples, ancillary data on the recording sheet will need to be entered into the database.

b. Drying

- Soil samples are dried for storage and to make grinding and homogenisation easier.
- Samples should be thoroughly dry until a constant weight is obtained
- Spread the soil out as a thin layer into shallow trays or plastic or paper sheets. It is important to ensure that no material from a sample is lost or discarded, as weights of soil fractions are to be recorded on processing.
- Break up clods as far as possible to aid drying. Take care to avoid crushing gravel sized particles.
- Great care should be taken at all stages to ensure sample labels remain with the samples.
- Exercise care to avoid contamination from dust, plaster or other potential contaminants during drying, as soils are subjected to trace element analysis.
- Air dry the samples in shade. Drying can also be done in a large room, custom-made solar dryer, or a forced-air oven at 40° C.
- Drying time will depend on the condition of the samples and ambient conditions, but the samples should be thoroughly dry (i.e. constant weight).

c. Weighing and sieving

- Weigh the whole dried soil sample to 0.1 g using a calibrated top-pan balance and record the weight. It is important that no material is lost from the sample during sample processing.
- Mix the dry sample thoroughly while still on the drying tray.
- Spread the sample on a plastic sheet.
- Using a wooden rolling pin, crush the sample to pass through a 2 mm sieve. While crushing, remove any plant materials and any possible pieces of gravel (making sure they are gravel and not soil aggregates) and place in a separate pile (the coarse fraction).
- Pass the crushed sample through the 2 mm sieve by shake the sieve gently to allow the soil to pass through. If a large amount of soil needs to be sieved, it is easier to do it in small batches rather than all at one time.
- Place whatever remains on the sieve back onto the plastic sheet and crush again gently. Then pass again through the 2 mm sieve. Make sure that all soil materials are crushed, but do not attempt to crush gravel and rocks.
- Throw anything that now remains on the sieve into the coarse fraction pile.
- While crushing, remove any plant materials (e.g. roots) and any possible pieces of gravel (making sure they are gravel and not soil aggregates) and place in a separate pile (the coarse fraction).
- The whole sample should be processed and no material should be discarded. You will remain with two fractions:
  - The coarse fraction (>2 mm), which cannot pass through the sieve.
  - The soil fines (<2 mm), which have passed through the sieve.
- Weigh the coarse fraction and record the weight to 0.1 g and record the data in “Sample Logging Sheet”. The logging sheet is given as Appendix 1 to this SOP.
- Enter the weight of the coarse fractions into the AfSIS database. The weight of the fine fraction is calculated in the database.
- Clean off the bench with a damp cloth to remove soil dust, so as to prevent contamination from one sample to another.

d. Sub-sampling of fine fractions

- Soil samples received will be divided into two sets viz., full set and reference subset. See appendix 4 and Appendix 5 for details. Complete analysis including wet chemical analysis will be conducted on the reference subset while only MIR and particle size analysis will be carried out on the full set. The sample requirement for full set is 20 g and that for reference subset is 350 g.
- Sub sampling of large samples to obtain a uniform sub sample is done in lab after crushing and sieving using coning and quartering described in Appendix 2. This method ensures a random and unbiased process to ensure representation.
- Through coning and quartering method, collect 20 g sub sample.
- Sub divide the 20 g sub sample into 15 g (for particle size analysis using LDPSA) and place it in a ziplock polythene bag labeled with the sample serial number (SSN).
- Transfer the remaining 5 g sample (for MIR spectra) into a Ziploc polythene bag labeled with the sample serial number (SSN).
- Store the remaining sample until MIR spectra on the full set is conducted and samples that constitute the reference subset are identified. Note: In case of AfSIS samples, the full set samples are identified before the MIR spectral analysis.
- For the reference subset, collect 350 g sample into a strong size 5 brown paper bag labeled with the sample serial number (SSN). If necessary use the coning and quartering method described in Appendix 2.
- From this create the following sub-samples. All the sub samples should be placed in a ploythene Ziploc bag of appropriate size labeled with the sample serial number (SSN)
    a. A 10 g sub sample for fine grinding and for MIR spectra, XRF, CNS and 13C/15N and 210Pb/137Cs analyses
    b. A 30 g (20 g if sample is insufficient) sub sample for wet chemical analysis at crop nutrition laboratory
    c. A 10 g sub sample for microbial profiling
    d. A 200 g sub sample for soil physical analysis including bulk density, Atterberg and plastic limits, shrinkage and swelling
    e. A 35 g sub sample for soil moisture characteristic curve
    f. Remaining 65 g sub sample for XRD and MPA NIR
- When sub sampling for Microbial profiling it is important to decontaminate the surrounding and the tools being used with 70 % ethanol before start and in between handling of samples.
- Store the sub samples at appropriate designated places for analysis. Note: Foreign soils should be stored separately in the foreign soil store.
- Store the excess soil fines in a labeled bag for possible use in future.

e. Sub-sampling of coarse fractions

- The coarse fraction of the samples selected above for reference analyses (i.e. the coarse fraction of the 350 g samples) is to be sub-sampled to collect a 20 g sub sample. In case of AfSIS samples this was done before shipping to the ICRAF Soil-Plant Spectral Diagnostic Lab
- Crush the coarse fraction as much as possible to homogenize the sample before sub-sampling

- Use coning and quartering (Appendix 2) or a riffle box to obtain a 20 g subsample of the coarse fraction
- Place the subsample in a zip-lock polythene bag labeled with the SSN, site, cluster, plot, depth_std code, and “coarse”. Example: sel000,002. Kiso1.1.TOP.Coarse”.
- Place the remainder of the coarse fraction into a labeled paper bag and store at regional laboratory for possible future analysis.

VII. CALCULATIONS

All the calculations are automated in the database management system

VIII. QUALITY ASSURANCE/QUALITYCONTROL

Not applicable

IX. DATA VALIDATION

Not applicable

X. HEALTH AND SAFETY

- Wear nitrile gloves to reduce the incidence of skin contact with potentially contaminated soil and to reduce the risk of cross-contamination
- Wear respirator that covers the mouth and nose to filter out harmful dust particles. Inhaling such particles irritates the nostrils and sinuses and can lead to lung diseases.
- Refer to the site-specific Health and Safety Plan for other safety concerns and applicable personal protective equipment

XI. REFERENCES

XII. APPENDIX

Appendix 1: Sample Logging Sheet

SSN	Sampling date	Cluster	Plot	Depth_std code	Depth_ top	Depth_ bottom	Total air dried soil weight	Coarse fragments

Appendix 2: Coning and quartering procedure

Coning and quartering procedure

1. Place the sample on a large cleaned surface or heavy-duty plastic sheeting.

2. Thoroughly mix the soil sample and spread the sample into a conical pile.

3. Further mix the soil by circumventing the cone symmetrically, repeatedly taking a spatula-full of soil from the base and transferring the soil to the apex of the cone.

4. Ensure the spatula is large enough to reach to centre of the cone. Circumvent the cone twice.

5. Flatten the cone to a height of about 1 cm.

6. Use a flat spatula or ruler, divide the pile into quarters along two lines intersecting 90° to each other.

7. Select one pair of opposite quarters as the sample to be retained.

8. If the sample is still too large then repeat the procedure from the beginning.

Appendix 3: Sample shipping procedures

In case AfSIS and those projects following AfSIS protocols, the following sub-samples should be shipped to the ICRAF Soil-Plant Spectral Diagnostic Lab, Nairobi from each site:

- A 20 g sub-sample of fine fractions for all soil samples.
- 32 samples of 350g soil fines for 0-20 and 20-50 depths from Plot One (or whichever plot has both top and subsoil samples)
- The corresponding 20 g sub-sample of coarse fraction of the 32 samples of soil fines. If there are no coarse fragments, that is fine.

1. Communicate and inform ICRAF Soil-Plant Spectral Diagnostic Lab prior to shipping to ensure all legal requirements are met and to ensure the safety of the samples.
2. In advance of shipment, send the details of your samples to the ICRAF Soil-Plant Spectral Diagnostic Lab at ICRAF Headquarters, Nairobi to: Keith Shepherd (k.shepherd@cgiar.org) copied to Elvis Weullow (e.weullow@cgiar.org). The information required is
    a. A description of the material (e.g. air-dried 2 mm-sieved soil samples)
    b. Number of soil samples for shipping
    c. Total weight of the soil in the batch
    d. Name, institutional address and fax number of the scientist shipping the samples.
3. Obtain a phytosanitary certificate from your country's plant inspectorate authorities or, if this is not possible, a letter from the relevant government authority indicating that the soils are specifically meant for research purposes only and have no commercial value.
4. Send the phytosanitary certificate or letter to the ICRAF Soil-Plant Spectral Diagnostic Laboratory.

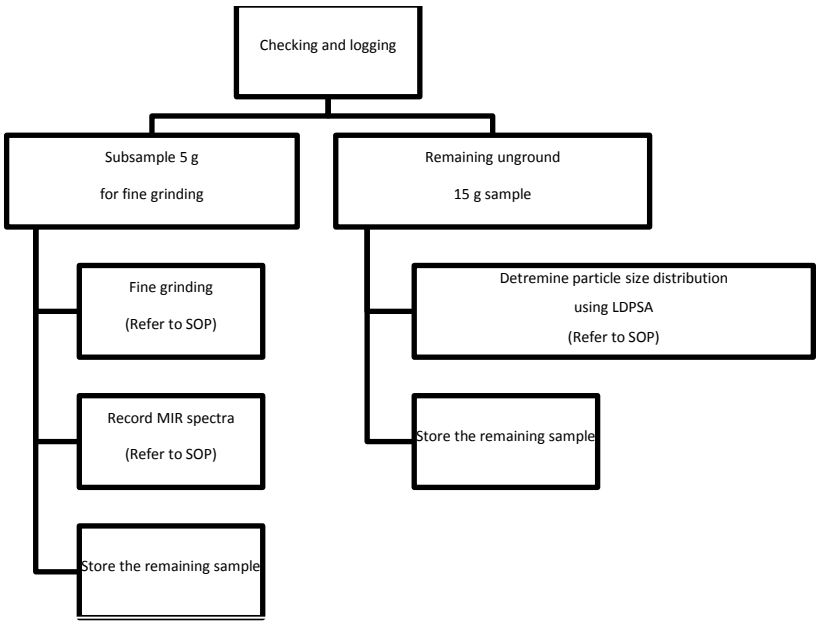
5. Based on the above documentation, the ICRAF Soil-Plant Spectral Diagnostic Laboratory will obtain the import permit from the Kenya Plant Health Inspectorate Service (KEPHIS) and a scanned copy of the permit[1] will be mailed to you.
6. The samples should be shipped together with a copy of the KEPHIS permit and a copy of the phytosanitary certificate or government letter from the source country. Failure to do so may result in the samples being destroyed by KEPHIS!
7. The soil samples to be shipped should be carefully double-packed into strong polythene bags that cannot be easily ripped or damaged in transit, and packed into strong shipping cartons.
8. Also have the shipping agent repack the consignment. Secure packing is critical because damaged packets will be destroyed by KEPHIS and our agreement with KEPHIS may be revoked.
9. Make sure that the final packing will stand the rough handling at airports.
10. The shipping address is:

    Dr. Keith Shepherd
    Att: Elvis Weullow
    World Agroforestry Centre (ICRAF).
    P. O. Box 30677-00100
    Nairobi, KENYA
    Tel: +254 20 7224000
    Fax: +254 20 7224001

11. Immediately after shipping, fax or email the shipping details (e.g. airway bill number) to Samuel Gaturu (s.gaturu@cgiar.org), Elvis Weullow (e.weullow@cgiar.org), and Mercy Nyambura (m.nyambura@cgiar.org), copied to Dr Keith Shepherd (k.shepherd@cgiar.org). This will allow us to alert the shipping agent's Nairobi office about the arrival of shipment and prepare for their clearance on arrival.
12. ICRAF Soil-Plant Spectral Diagnostic Laboratory will arrange for clearance of the shipment and inspection of the soils by KEPHIS.
13. Upon clearance by KEPHIS, ICRAF will arrange for collection and transport of samples to ICRAF Soil-Plant Spectral Diagnostic Laboratory.
14. At the laboratory, samples will be logged as foreign soils, stored in dedicated foreign soil store and handle them as per the specified procedures for handling foreign soils laid down in their agreement with KEPHIS.
15. Note that the ICRAF Soil-Plant Spectral Diagnostic Laboratory charges US$100 to cover all the expenses involved in sample clearance protocols, including KEPHIS fee, visits to the KEPHIS office, and clearance when the samples arrive. The shipping and permit costs are the responsibility of the sender.

Appendix 4: Flow chart for full set samples

[1] KEPHIS also issue a quarantine (Q) label that the ICRAF Soil Lab will retain for clearance purposes.



Appendix 5: Flow chart for reference subset samples