Laboratory Standard Operating Procedure

NIRS Acquisition on Fresh Matooke Fingers using the Benchtop NIRS FOSS DS2500 and Relating Spectra to Finger Dry Matter Content by Oven Method

High-Throughput Phenotyping Protocols (HTPP), WP3

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Ephraim NUWAMANYA, National Crops Resources Research Institute (NaCRRI), Kampala, Uganda

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Ethics: The activities, which led to the production of this document, were assessed and approved by the CIRAD Ethics Committee (H2020 ethics self-assessment procedure). When relevant, samples were prepared according to good hygiene and manufacturing practices. When external participants were involved in an activity, they were priorly informed about the objective of the activity and explained that their participation was entirely voluntary, that they could stop the interview at any point and that their responses would be anonymous and securely stored by the research team for research purposes. Written consent (signature) was systematically sought from sensory panelists and from consumers participating in activities.

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WP3: High-Throughput Phenotyping Protocols (HTPP)

SOP: NIRS Acquisition on Fresh Matooke Fingers using the Benchtop NIRS FOSS DS2500 and Relating Spectra to Finger Dry Matter Content by Oven Method

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Written by: Ephraim NUWAMANYA

For information on this SOP please contact:

- Ephraim NUWAMANYA (nuwamanyaephraim@gmail.com)
- Brigitte UWIMANA (B.Uwimana@cgiar.org)

This document has been reviewed by:

Karima MEGHAR (CIRAD)	26/10/2020
Fabrice DAVRIEUX (CIRAD)	12/11/2020
Emmanuel ALAMU (IITA)	13/11/2020

Final validation by:

Fabrice DAVRIEUX (CIRAD)	17/11/2020

CONTENTS

Table of Contents

ABSTRACT

Determination of dry matter is important in selection of banana with acceptable qualities for consumer utilization. DM determination in banana is quickly and reliably determined using NIRS technology. The Standard Operating Procedures (SOPs) detailed allows for the acquisition of mashed banana fingers. The procedures describe the scanning and spectral acquisition of banana using the small sample cup of the FOSS DS2500 NIRS equipment. Banana samples are harvested using approved harvesting tools and labelled after which preparation commences by peeling and grating using a grater. This is followed loading the grates into a sample cup and scanning of the grated material filled in the sample cup. The procedure is repeated by loading fresh material from the main sample into the sample cup producing two subsets of scans from one particular accession. Spectral data produced from these scans is downloaded and further processed for use in calibration development and finger/cluster chemical composition determination. Reference data generation is carried out by approved reference methods in repeatability analyses carried out at NaCRRI and NARL. Critical points of consideration include the development of a sample flow and spectra acquisition matrix coupled to correct labelling since sample numbers involved are usually many.

Key words: Banana, spectra, procedure, Reference data, repeatability

1 SCOPE AND APPLICATION

This SOP applies to the spectral acquisition of fresh Matooke fingers using the benchtop NIRS FOSS DS2500 and relating spectra to finger dry matter content by oven method. The SOP will help to ensure the quality spectral data collection from a representative sample of Fresh Matooke fingers for appropriate phenotyping of the Matooke/banana fingers.

2 REFERENCES

Part of this protocol has been informed by work from the following publications

1. Sánchez, T., Ceballos, H., Dufour, D., Ortiz, D., Morante, N., Calle, F., ... & Davrieux, F. (2014). Prediction of carotenoids, cyanide and dry matter contents in fresh cassava root using NIRS and Hunter color techniques. Food chemistry, 151, 444-451.
2. Ikeogu, U.N., Davrieux, F., Dufour, D., Ceballos, H., Egesi, C.N., Jannink, J.L. (2017). Rapid analyses of dry matter content and carotenoids in fresh cassava roots using a portable visible and near infrared spectrometer (Vis/NIRS). PLoSONE 12(12), e0188918. https://doi.org/10.1371/journal.pone.0188918
3. Alamu, E. O., Nuwamanya, E., Cornet, D., Meghar, K., Adesokan, M., Tran, T., … Davrieux, F. (2020). Near-infrared spectroscopy applications for high-throughput phenotyping for cassava and yam: A review. International Journal of Food Science and Technology, 1–11. https://doi.org/10.1111/ijfs.14773

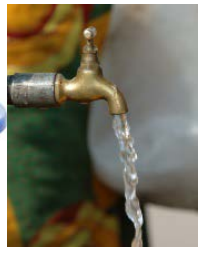
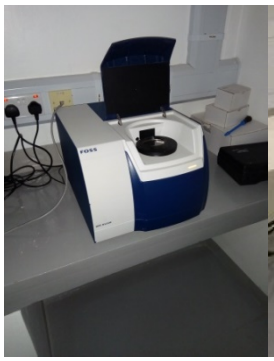
3 DEFINITIONS

In this SOP, phenotyping refers to a quantitative description of the banana finger anatomical/physical properties in relation to biochemical properties of the finger in question.

4 PRINCIPLE

In this SOP, the relationship between the spectra acquired from fresh banana (matooke) finger discs and the dry matter content is investigated. Developing the right metric for dry matter is important in defining measurable parameters related to biochemical properties that define Matooke yield and Matooke quality or tokeness.

5 APPARATUS

Process	Equipment/pictorial presentation of the process
Harvesting equipment	 
Labelling equipment such as markers or bar code generator and bar code reader	 
Washing equipment and water source	  
Sample cutting or chopping and grating equipment	   
Benchtop NIRS equipment for spectra generation	 

6 PROCEDURE

Sample preparation

I. Harvest 1(one) mature bunch from the plot. Bunches are of different sizes and shapes but should have fingers that have length and diameter measurements allowing for peeling to be undertaken on the sample.

II. After harvest, label the samples, move them to the lab and select the cluster of interest. From each cluster, remove the two outer fingers and thereafter select at least four fingers in a zigzag fashion (Figure 1). Determine the number of fingers based on the maximum number of fingers that would give more than 100g of peeled fresh matooke. The zigzag fashion of selecting fingers allows you to select both fingers in the upper and lower row on the cluster hence ensuring the randomisation of the sample.

III. Wash the selected fingers with water in a bucket to remove debris. Dry the samples by wiping with a paper towel.

IV. For each of the selected fingers in step II above, immediately peel and cut off at least 2 cm (depends on the length of the finger) from both ends. Remove sap by wiping with the wipes provided where necessary. Note that peeling should be done to avoid blackening of the sample due to oxidation.

V. Aggregate the peeled fingers and give the same label matching the accession name using a barcode.

VI. For each of the aggregated samples slice the 0.3cm sections using the slicing portion of the grate to produce thin slices. Immediately after slicing, the sample is placed in the small sample cup and scanned using a benchtop NIRS equipment

VII. The sample is scanned twice by picking from the same sample two times to produce two average spectra representing one sample (see Figure 1).

VIII. Pick a sample from the remaining pool sample and use it for dry matter determination.

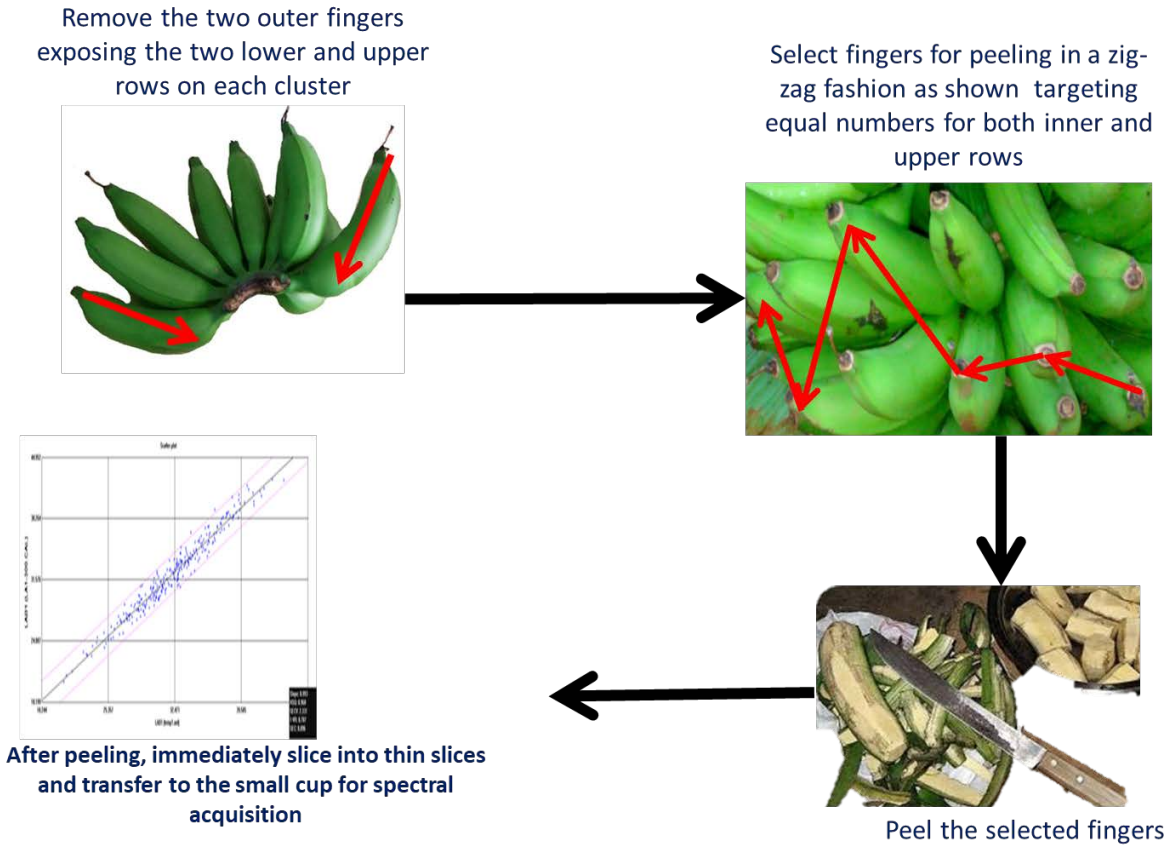


Figure 1: Description of sample preparation, scanning and spectra acquisition process

7 EXPRESSION OF RESULTS

The dry matter content (%) and of each of the peeled Fresh Matooke fingers portions are determined and used in calibration.

8 CRITICAL POINTS OR NOTE ON THE PROCEDURE

I. Be sure to maintain the same sample id (accession name) for all root sections derived from the same root.

II. Be sure to read the manual supplied with the NIRS FOSS DS2500 before use in acquiring spectra

III. Be sure to use the minimal time between sample preparation (peeling and slicing) and sample spectra generation as scanned samples rapidly lose moisture.

IV. Maintain sample homogeneity and state by only preparing the sample when you are ready for spectral acquisition

V. Take care to position the slices in the cell to cover the reading surface as much as possible.

VI. Use an agreeable digital or manual platform to enter reference information. Where bar code labels are used, scan the bar code with the tablet to ensure you are entering data to the right accession

VII. Have a well-documented and reviewed sample flow prior to the start of exercise

Dry samples from dry matter measurements conducted at a temperature between 50°C and 60°C can be used to make flour which is later assessed for other properties.

Institute: Cirad – UMR QualiSud

Address: C/O Cathy Méjean, TA-B95/15 - 73 rue Jean-François Breton - 34398 Montpellier Cedex 5 - France

Tel: +33 4 67 61 44 31

Email: rtbfoodspmu@cirad.fr

Website: https://rtbfoods.cirad.fr/