

Managing within varietal variation in the context of a genebank: the options

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Meeting on Cassava Germplasm: Collection Process and Action
Plan for South, East and Central Africa
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Constraints: Cassava Germplasm Collection

7 million germplasm in 1750 gene banks; 1-2 million are distinct (FAO, 1996)

Mismatch (TME and TMe) and other nomenclature issues

Presence of duplicates

Mismatch between field bank and in vitro bank

Maintenance cost and implication on distribution

Low genetic diversity

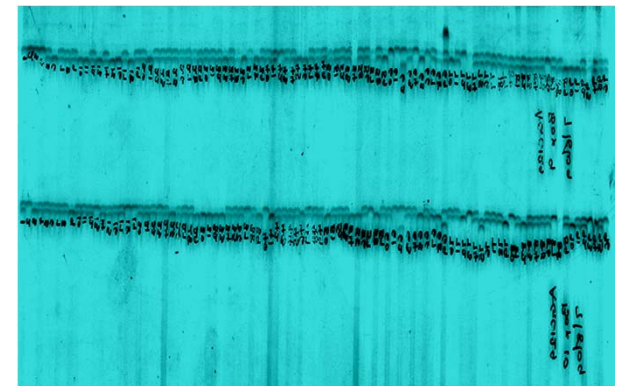
GCDT Funds: Identification of genetically unique accessions in cassava/yam using SSR markers

Question: Differentiate two accessions as Unique

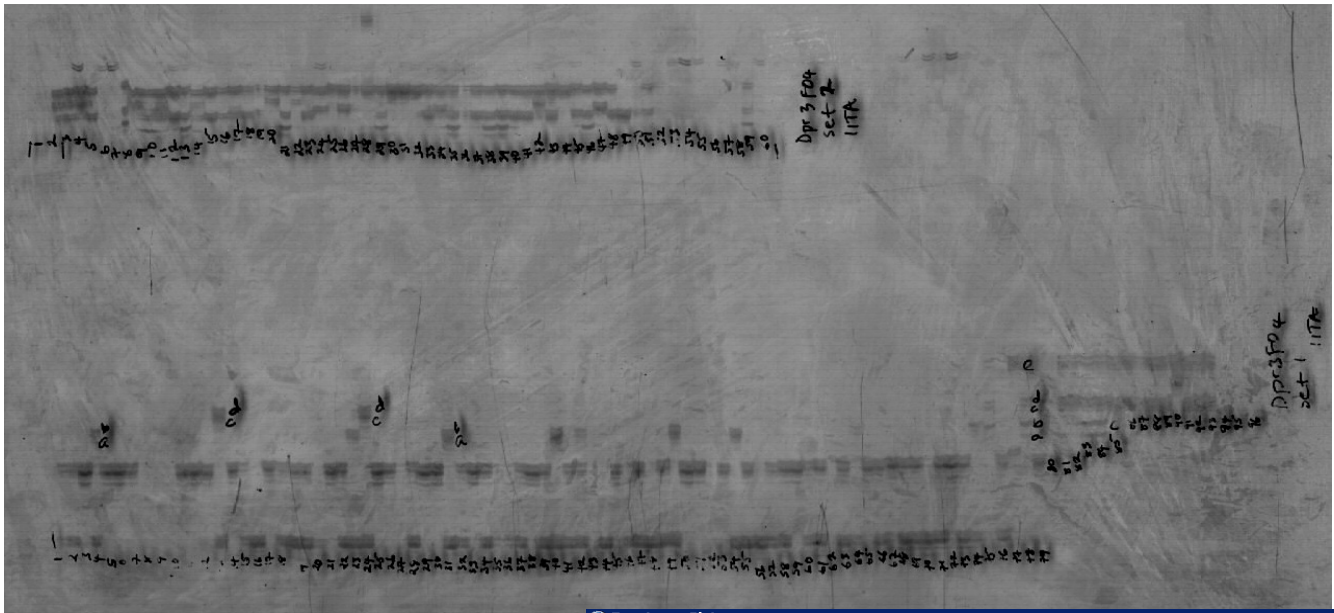
- **Number of accessions studied: Cassava: 2418 and Yam: 1280 *D. alata* (815 IITA and 465 NARS); 878 *D. rotundata* (626 IITA and 252 NARS); 70 *D. cayenensis* (59 IITA and 11 NARS) ; 57 *D. spp.* (mainly landraces)**
- **Number of SSRs used: Cassava: 25; Yam: 18 genomic and 25 EST-SSRs**
- **System used: PAGE and Capillary Electrophoresis (ABI3130)**
- **Data scored as presence (1) or absence (0) for each allele per marker per accession (PAGE)**
- **Genetic distances (GD) were calculated among pairs of accessions following Modified Roger's distance method**
- **Two accessions were defined as "Absolute Duplicates", if GD = 0 between them**
- **Two accessions were defined as "Potential Duplicates", if GD = 0 and 0.15**



**High-throughput DNA Extraction
(192 – samples per day)**



High-throughput PAGE gels



PAGE and ABI analysis



Identification of Duplicates: Cassava

893 (36.9%) duplicates identified

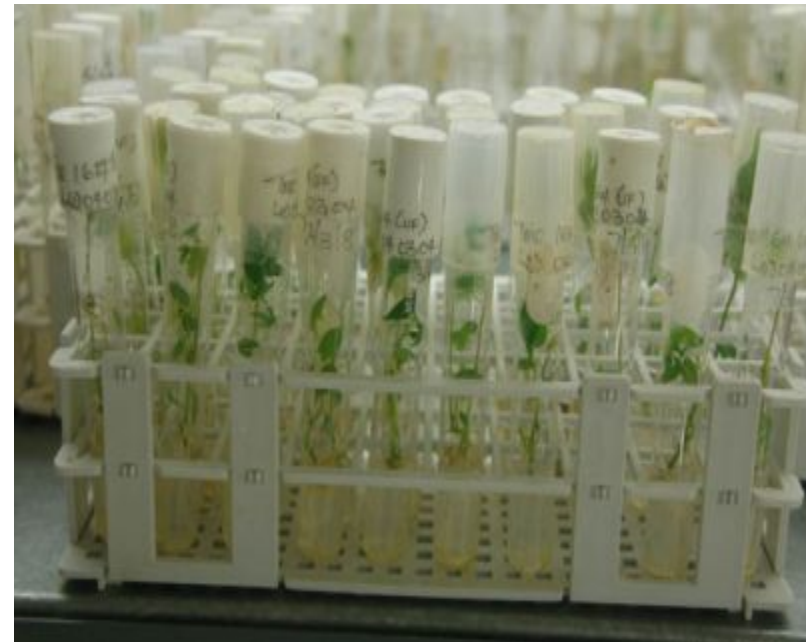
Further fingerprinting with additional 15 SSR markers using capillary electrophoresis (multiplexed) (ABI3130)

Data scored in base pairs based on actual peaks observed (GeneScan and GeneMapper)

The average number of alleles per locus was 4.7, which ranged from 8 alleles (AT47) to 3 alleles (TCT140, TTC59 and AT101)

Further Fingerprinting with SNPs or GBS, conduct field trials for morphological evaluation, conservation in long-term storage

Somaclonal variation study in Yams under *in vitro* regeneration



Background

- More than 3000 accessions of 8 yam species conserved under Genetic Resources Center of IITA.
- GRC is now conserving around 1200 meristem derived accessions of yams *in vitro*.
- New explants and sub culture from old culture are both used for regular regeneration.
- The extent of somaclonal variation and its genotypic and phenotypic effect in yam is not yet studied.

Objectives

- To evaluate the extent of somaclonal variation of meristem derived *in vitro* clones of *D. rotundata* and *D. alata* accessions with their corresponding field materials.
- To analyse the effect of somaclonal variation on phenotypes.

Methods

- Different accessions constituting of *D. rotundata* (58) and *D. alata* (26) being regenerated under *in vitro* regeneration will be assessed.
- The materials was selected based on introduction date *in vitro*. Hence, includes and grouped as old introduction, middle and recent introduction.
- *in vitro* materials acclimatized and will son be transferred to screen house. Similarly the accessions under field regeneration will be planted under similar condition but from tuber.
- Morphological descriptors and molecular tools will be used for assessment of possible variation.

Yam field genebank management (mismatch identification)

Challenges

- Individual variation within accessions
- This is either due to mis-labelling and material mix up during planting
- or storage along the years
- For some accessions the mix was from the beginning during germplasm
- collection and inclusion to genebank
- Misplacement and loss of labels in the field
- Implications for reliability of further regeneration techniques

Activities performed to validate accessions

- Assessment of morphological variation among individual plants within accessions.
- Identification of distinct morphological characters as a reference for each accession.
- Maintenance of uniform individuals within accessions in yam field genebank collection.

Achievements

- Significant percentage (~20%) of the total collection was found misidentified.
- Mislabeling/misidentification between field and *in vitro* was identified among species and within species.
- Few descriptors were found with the most discriminant power among 53 different yam descriptors used in the study.
- Improved maintenance quality of accessions and reliability of further regeneration techniques (*in vitro* and cryopreservation).
- Introduction of bar coding of field bank