# <u>Applications of Molecular Markers in</u> <u>Sweetpotato</u>



**CIP Sweetpotato Molecular Markers** 

#### (I) **RFLPs Restriction Length Polymorphism**

non PCR (polymerase chain reaction), 1 loci per assay, 5g DNA, Codominant, 5 days per assay

#### (II) RAPDs Random Amplified Polymorphic DNA PCR, 10 loci per assay, 0.2g DNA, Dominant, 5 hours per assay, unreliable

#### (III) AFLPs Amplified Fragment Length Polymorphism PCR, 50 loci per assay, 0.2g DNA, Dominant, 1 day per assay,

#### (IV) SSRs Simple Sequence Repeats

PCR, 1 loci per assay, 0.2g DNA, Co-dominant, 5 hours per assay, large number of alleles per marker loci

#### (V) DarTs Diversity Array Technology

PCR, Applying bioarry chips to AFLPs, >100 loci per assay, 5ng DNA, Dominant, 1.5 to days per array

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**Application I - Germplasm** 

Identification of Duplicates Verifying Distinct Accessions Identifying Structures in Germplasm

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Fig. 2 Dendrogam of the UPGMA cluster analysis of 92 sweetpotato cultivars (85 cultivar of African origin) on the basis of Jaccard's SSR based genetic similarities

(origin of clones: orange = Africa, green lines = South American germplasm, light blue = Pacific, deep blue = North America, and red = China; origin of East Africa farmer varieties: UG = Ugandan, cultivars, KE = Kenya, TZ = Tanzania, ZB = Zambia;  $\Box$  = White fleshed clones,  $\Delta$  = Orange-fleshed clones)

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# Application II – Building Heterotic Genepools

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#### Fig. Cluster analysis of 575 accessions of sweetpotato based on 20 SSR markers

(Central and North America in black, South America in green, Pacific including New Zealand, Austria and Papua New Guinea in blue, Asia in yellow and Sub Saharan Africa in red).

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#### Fig. Cluster analysis of 575 accessions of sweetpotato based on 20 SSR markers

(farmer variety accessions in blue and accessions generated by breeding programs in green)

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## Marker Application for Heterosis and Genetic Distance

Heterosis increases with the Genetic Distance

& deceases if the Genetic distance is too large



Schematic representation of the relationship between mid-parent heterosis for yield and parental genetic distance based on unselected DNA markers (1) Crosses between related parents (2) intra-genepool crosses between unrelated lines (3) - (5) inter-genepool crosses (Lefort-Buson 1985, Melchinger 1999, modified)

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### Heterosis in Sweetpotato



**Fig. Distribution of mid-offspring – mid parent heterosis in 231 families** evaluated on basis of 30 clones per family and two plot replications for each clone across two locations.

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# **Application III – Marker Assisted Selection**

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# <u>Markers and Sweetpotato virus</u> <u>disease (SPVD)</u>





Aphids

← SPFMV+ SPCSV and / or Crinivirus other viruses





Whiteflies



In experiments: Yield reduction up to 90%

In practice: Yield reduction up to 75%

Apparently healthy SPFMV and / or other viruses <u>SPVD</u> <u>SPCSV + SPFMV and / or</u> <u>Other viruses</u>

### Marker development

- 1) More SSR markers
- Using unexploited set of EST available 48 new SSR loci were identified.
- already available 64 SSR marker loci for sweetpotato.
- 2) <u>Resistance groups formed from PJ parents & offspring</u> (- -) 0 in SPCSV and SPFMV ELISA scores\_\_\_\_

A) -- (5 clones)

- B) + (23 clones) inc. doubt full clones ↑SPFMV
- C) + (31 clones)
- D) - (763 clones)

individual DNA -> CIP (AFLP) & Australia (DArT)

- 3) Extreme groups formed from PJ parents & offspring
  - (-) 0 or very low in SPCSV ELISA scores
  - A) 10 clones
  - B) + 298 clones

bulked DNA – CIP (AFLP)

Marker Assisted Selection for A) SPVD

### **B)** Nematodes

GCP



#### Population PJ05.\_\_\_\_ x DLP3163 + parents for AFLP marker screening

number	SFMV	SPCSV
10	-	-
39	+	-
28	-	+
747	+	+

10 clones

298 clones out of 747 (Highest ELISA titers)

# Form the AFLP markers screened so far 4 markers have been selected

E33M49.187	Chi-Square	11.82	Prt. 0.0006
E35M36.293	Chi-Square	6.16	Prt. 0.0130
E35M36.267	Chi-Square	22.35	Prt. <.0001
E33M59.163	Chi-Square	11.04	Prt. 0.0009

For the combination 1<sup>st</sup> E33M49.187, 2<sup>nd</sup> E33M59.163, 3<sup>rd</sup> E35M36.293, 4<sup>th</sup> E35M36.267

	E33M49.187	E35M36.293	E35M36.267	E33M59.159
no band (O)	1	0	3	9
band (1)	9	5	2	1
missin values	0	5	5	0
no band (O)	186	155	17	94
band (1)	106	123	268	157
missin values	6	20	13	47

2 wrong classifications of10 resistant clones8 wrong classifications of298 susceptible clones

Note resistance group still too smal for higher power of significance tests

# **Application IV – The Sweetpotato Map**

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### Sweetpotato AFLP & SSR markers

#### The Sweetpotato Map

- Beauregard x Tanzania
- (SPK004 x Xushu18)
- Mapped: dry matter, starch, sucrose, ß-carotene

Work independently made at CIP and NCSU

All 90 chromosomes can be distinguished, not all chromosomes can be assigned to their homologous linkage group (15 homologous groups)

### Sweetpotato Map



CIP – Sweetpotato Map

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### Thank you for your Attention

CIP - Sweetpotato Molecular Markers