# Diallel analysis of sweetpotato for beta-carotene content and yield components

Martin Chiona
Presented at the Sweetpotato Breeders'
Annual Meeting
Mukono, Uganda, June 22-25, 2010

#### Introduction

- VAD is one of the major nutritional deficiencies worldwide
- VAD is a leading cause of early childhood death and a major risk factor for pregnant women in Africa
- Vitamin A is essential for the normal development of a person and deficiency can
  - lead to night-blindness (estimated to afflict 3 million sub-Saharan children under the age of five)
  - result in an increased susceptibility to a variety of other diseases due to a weakened immune system
- In low-income, impoverished populations, 82% of the dietary vitamin A is derived from plant sources (van den Berg et al., 2000).

#### Introduction Cont...

- VAD is a problem of public health significance in Zambia
  - Subclinical VAD is significant, with a prevalence rate of 13-17% among children ranging from 6 months to 12 years of age.
- Improved vitamin A intake through increased consumption of OFSP will make a significant contribution to improved health.
- The enrichment of  $\beta$ -carotene in the local sweetpotato varieties is an attractive option to improving vitamin A intake.
- There is wide genetic variability for vitamin A occurring naturally in sweetpotato
- This means conventional breeding techniques can be employed to incorporate β-carotene into local sweetpotato varieties.

#### Introduction Cont...

- Diallel crosses have been widely used in genetic research to investigate the inheritance of important traits among a set of genotypes.
- Diallel mating designs were devised, specifically, to investigate the combining ability of the parental lines for the purpose of identifying superior parents for use in hybrid development programmes.
- A diallel cross is a set of  $p^2$  possible single crosses and selves between p homozygous or heterozygous parents;
- It provides a powerful method for investigating the relative genetic properties of these parents.
- It is possible to partition genetic variation into components due to general combining ability (GCA) and specific combining ability (SCA) and
- to thus obtain a relative estimate of the inheritance of a given trait.

#### Introduction Cont...

- Analysis of diallel data is usually conducted according to the methods of Griffing (1956) which partition the total variation of diallel data into GCA of the parents and SCA of the crosses (Yan and Hunt, 2002).
- The variances of GCA and SCA are related to the type of gene action involved.
- Variance for GCA includes additive portion while that of SCA includes non additive portion of total variance arising largely from dominance and epistatic deviations
- Diallel analysis has also found application for analyzing β-carotene content in sweetpotato.

# Objectives

- The present research examined the basic nature of the quantitative inheritance of selected traits in sweetpotato carried out by means of diallel analysis with a view to
  - estimating the components of genetic variance (GCA and SCA), and
  - to determine the type of gene action controlling β-carotene, root dry mass (RDM), harvest index (HI) and root fresh yield (RFY) in F<sub>1</sub> progenies

# Methodology

- Hand crosses were carried out in a 5 x 5 full diallel
  - from 2006 to 2008
  - at Mansa Research Station (11° 14.396' S and 028° 57.226' E)
- The parents consisted of
  - two introductions from CIP and
  - three advanced breeding lines

			Root dry mass	Source
No.	Genotype	Flesh colour	(%)	
1	Excel	deep orange	29	CIP
2	L4-138/3	white	30	Local
3	W-119	orange	25	CIP
4	Unknown 2/1	cream	32	Local
5	L3-199084/1	orange	21	Local

- Botanical seed obtained
- Germinated in a sand and vermiculite mix in polystyrene
   24 x 8 cell seedling trays.
- Seedlings were transplanted to 0.5 L plastic pots.
- Once the plants were about 5 cm tall, they were transplanted to the field in the wetland
  - to allow for further growth in a non competitive environment.
- The F<sub>1</sub> progeny were randomly selected from the wetland site on the basis of producing adequate good quality vegetative cuttings for the field trial.
- This was the only pre-trial selection that was imposed on the progeny to be evaluated.



















Raising planting materials in the wetland (dambo)

- On adequate planting material basis for a given  $F_1$  cross, 20 progeny were randomly chosen to represent each cross.
- The selected F<sub>1</sub> progeny along with their parental lines were planted in the same field trial.
- The trial was planted in the rainy season in November
  - cultural practices and weed control were performed according to standard field practices.
  - No external inputs such as fertilizers were applied.
  - Hills were planted for a final plant density of about 40 000 plants ha<sup>-1</sup>.
  - The trial was laid out as a 5 x 5 triple lattice
  - The experimental plot was a single row for each F<sub>1</sub> cross, 6 m long and 1 m apart,
  - with plant to plant distance of 30 cm.
  - Each single row plot comprised 20 progeny of a cross.



- All data were recorded on individual plant basis and then averaged across the 20 progeny of each F<sub>1</sub> cross.
- The quantitative characters were evaluated as follows:
  - RFY expressed as harvested fresh roots in tonnes per hectare;
  - HI expressed as a ratio of RFY to total biomass (mass of roots and vines);
  - β-carotene content expressed as mg 100 g<sup>-1</sup>; and
  - RDM expressed as a percentage of root dry mass divided by the root fresh mass.

# Statistical Analysis

- General analyses of variance were performed for all hybrids and parents using GENSTAT for β-carotene content, RDM, HI, and RFY.
- Pseudofactors for analysing with the triple lattice design were generated within the GENSTAT programme.
- The block corrected means of full-sib families for each of three replications were used in the diallel analyses.
- To test the null hypothesis of no genotypic differences among treatments (parents and crosses) a one way analysis of variance was performed.

# Statistical Analysis Cont...

- Treatment sum of squares were broken into three components, parents (P), crosses (C), and P vs. C.
- GCA and SCA variance components were computed according to the fixed-effects model I, method 1 (F<sub>1</sub>s; parents and reciprocals included) described by Griffing (1956)
- used the DIALLEL-SAS05 program developed by Zhang et al. (2005).

# Results

Source	df	Mean Squares			
		β-carotene	Root dry	Harvest	Root fresh
		content	mass (%)	index	yield (t ha <sup>-1</sup> )
		(mg 100g <sup>-1</sup> )			
Rep	2	0.57 <sup>NS</sup>	6.99 <sup>NS</sup>	0.004 <sup>NS</sup>	000.28 <sup>NS</sup>
Treatments	24	42.34***	38.28***	0.065***	198.16***
Blocks within reps	12	11.20***	22.64	0.032	079.68
Intra-block error	36	0.12***	4.60***	0.003***	001.22***
Total	74				

Parents/ Crosses <sup>a</sup>	β-carotene content (mg 100 g <sup>-1</sup> )	Root dry mass (%)	Harvest index	Root fresh yield (t ha <sup>-1</sup> )
1 x 2	13.69	28.27	0.55	1.12
1 x 3	0.18	24.74	0.84	8.21
1 x 4	0.06	31.49	0.92	14.95
4 x 5	0.13	30.83	0.73	26.49
Parent 4	0.07	32.51	0.78	10.92
Parent 2	0.19	35.54	0.63	11.35
Parent 3	4.89	24.43	0.81	25.44
Parent 5	3.51	36.12	0.69	18.93
Parent 1	6.65	25.78	0.76	4.84
Mean	2.51	29.91	0.71	10.93
s.e	0.34	2.14	0.05	1.11
CV (%)	13.7	7.2	7.6	10.1
LSD <sub>(0.05)</sub>	0.65	4.05	0.10	2.09

Parents/ Crosses <sup>a</sup>	content	Root dry mass (%)	Harvest index	Root fresh yield (t ha <sup>-1</sup> )
	(mg 100 g <sup>-1</sup> )			
Reciprocals (be	low the diagonal	)		
3 x 1	0.06	27.18	0.87	16.67
4 x 1	0.07	28.75	0.86	15.53
5 x 1	0.16	37.89	0.88	11.76
3 x 2	13.72	29.41	0.74	7.59
4 x 2	0.08	31.44	0.91	8.84
4 x 3	0.24	33.66	0.83	32.79
Parent 4	0.07	32.51	0.78	10.92
Parent 2	0.19	35.54	0.63	11.35
Parent 3	4.89	24.43	0.81	25.44
Parent 5	3.51	36.12	0.69	18.93
Parent 1	6.65	25.78	0.76	4.84

Source	df	Mean Squares			
		β-carotene	Root dry	Harvest	Root fresh
		content	mass (%)	index	yield (t ha <sup>-1</sup> )
		(mg 100g <sup>-1</sup> )			
Rep	2	0.57***	7.00 <sup>NS</sup>	0.01 <sup>NS</sup>	000.11 <sup>NS*</sup>
Parent	4	25.62***	101.24***	0.25***	269.38***
Parent x Cross	1	9.48***	19.74 <sup>NS</sup>	0.01 <sup>NS</sup>	251.33***
Crosses	9	56.41***	22.96***	0.21***	225.03***
GCA	4	71.36***	42.32***	0.27***	235.77***
SCA	5	44.45***	7.47 <sup>NS*</sup>	0.17***	217.87***
Reciprocal	10	43.20***	8.17 <sup>NS*</sup>	0.23***	225.03***
Error	48	00.104**	5.46 **	0.003	1.50***
Total	74				

# GCA

Parent <sup>a</sup>	β-carotene content (mg 100g <sup>-1</sup> )	Root dry mass (%)	Harvest index	Root fresh yield (t ha <sup>-1</sup> )
1	1.650**	-2.612**	0.0623**	-5.082**
2	-0.023 <sup>NS</sup>	2.981**	0.0129 <sup>NS</sup>	1.301**
3	0.354**	-2.807**	-0.0848**	2.533**
4	-1.044**	1.837**	0.0126 <sup>NS</sup>	-0.152 <sup>NS</sup>
5	-0.937**	0.602 <sup>NS</sup>	-0.0031 <sup>NS</sup>	1.400**

Crosses <sup>a</sup>	β-carotene content (mg 100 g <sup>-1</sup> )	Root dry mass (%)	Harvest index	Root fresh yield (t ha <sup>-1</sup> )
1 x 2	5.655**	-0.745 <sup>NS</sup>	-0.141**	-4.754**
1 x 3	-2.105**	1.144 <sup>NS</sup>	0.114**	-3.320**
1 x 4	-3.018**	1.257 <sup>NS</sup>	0.194**	9.939**
1 x 5	-2.416**	-0.378 <sup>NS</sup>	-0.193**	-10.399**
2 x 3	-2.722**	0.563 <sup>NS</sup>	0.153**	6.797**
2 x 4	-1.373**	1.849*	-0.070**	-4.517**
2 x 5	3.184**	-1.343 <sup>NS</sup>	0.097*	6.414**
3 x 4	5.088**	-0.690 <sup>NS</sup>	-0.084**	-7.629**
3 x 5	-3.364**	-2.904 <sup>NS</sup>	0.547**	-14.979**
4 x 5	-0.113 <sup>NS</sup>	0.571 <sup>NS</sup>	-0.292**	-4.111**

Crosses <sup>a</sup>	β-carotene content	Root dry mass (%)	Harvest index	Root fresh yield (t ha <sup>-1</sup> )
	(mg 100 g <sup>-1</sup> )			
Reciprocals (E	Below the diago	onal)		
2 x 1	3.917**	-1.437 <sup>NS</sup>	-0.0002 <sup>NS</sup>	-0.722 <sup>NS</sup>
3 x 1	-2.303**	-0.112 <sup>NS</sup>	0.127**	3.611**
4 x 1	0.005 <sup>NS</sup>	1.465 <sup>NS</sup>	0.027 <sup>NS</sup>	-0.370 <sup>NS</sup>
5 x 1	0.003 <sup>NS</sup>	0.653 <sup>NS</sup>	0.000 <sup>NS</sup>	0.000 <sup>NS</sup>
3 x 2	0.000 <sup>NS</sup>	0.000 <sup>NS</sup>	-0.025 <sup>NS</sup>	-5.111**
4 x 2	-0.040 <sup>NS</sup>	-1.763 <sup>NS</sup>	-0.316**	-4.889**
5 x 2	2.257**	-1.972*	-0.117**	-15.042**
4 x 3	-6.802**	-0.317 <sup>NS</sup>	0.332**	-2.434**
5 x 3	-0.005 <sup>NS</sup>	0.728 <sup>NS</sup>	0.160**	7.889**
5 x 4	0.005 <sup>NS</sup>	-1.160 <sup>NS</sup>	-0.344**	0.780 <sup>NS</sup>



#### Conclusions

- Differences among the genotypes were highly significant for all the characters providing an opportunity for selection
- GCA mean squares were highly significant (p=0.01) for all characters studied.
- SCA and Reciprocal effects were significant (p=0.01) for all characters studied except RDM.
- Non-additive type of gene action was dominant over additive effect for β-carotene, HI and RFY because SCA variance was greater than GCA variance.
- In case of RDM, additive gene action was more important in controlling the trait; though, both additive and non-additive gene actions were involved in the expression of the traits.

#### Conclusions

- The ratios of GCA to SCA variances were small (0.1-0.35:1) except for RDM (7.56:1).
- The two high  $\beta$ -carotene parents used in this study exhibited high GCA, indicating that additive gene effects were predominant in the inheritance of  $\beta$ -carotene.
  - However, β-carotene parents with high GCA did not result in progeny with the highest SCA for most of the crosses.
  - The high β-carotene progenies were obtained from a cross of a parent with high GCA plus a parent with low GCA (1 x 2 and 3 x 4) and a parent with low GCA plus a parent with low GCA (2 x 5).
  - The reason for this is unclear, however sweetpotato is a highly heterozygous hexaploid, and it is possible that on crossing parents selected for β-carotene content more favourable combinations of genes are broken down than reassembled in the progeny.

#### Conclusions

- In crosses between parents with low GCAs or between a parent with high GCA and a parent with low GCA which produced progeny with high SCA effects, it is possible that dominance gene effects resulting from non-allelic interactions from the parent with low GCA increase the expression of the trait in the progeny.
- Therefore, parents must be selected on the basis of their SCA. Conversely, high RDM parents that exhibited high GCA produced only one cross with significant high RDM progenies.
- Reciprocals were not significant for RDM but they were significant for β-carotene content. These results suggest that improvement for β-carotene content and RDM may not be achieved simultaneously.

- From the above results, GCA alone was not sufficient for selecting parents for a hybrid programme.
- Parents with low GCA may produce progeny with high SCA effects for β-carotene, HI, and RFY.
- Hence,  $\beta$ -carotene can be transferred into improved progeny using parents with either high or low levels of  $\beta$ -carotene.
- Conversely, high RDM can be transferred into improved progeny using parents with high RDM.





# Thank you