

STUDIES OF GENETIC CONTROL OF PRO-VITAMIN A MAIZE HYBRIDS IN GHANA

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Abstract: Vitamin A deficiency (VAD) is responsible for a number of disorders that range from impaired iron mobilization, growth retardation, and blindness to a depressed immune response, increased susceptibility to infectious disease and increased childhood mortality and morbidity affecting several children and pregnant women worldwide. Breeding for Pro-vitamin A (PVA) maize varieties requires the understanding of the genetic control of the PVA trait in order to inform breeders about appropriate breeding schemes to use in their breeding programmes. The objective of this research therefore was to unravel the genetic control of Pro-vitamin A trait in maize hybrids in Ghana. The parents for the crosses included Akposoe (Quality Protein Maize (QPM) variety 80-85 days maturity), Aburohemaa (QPM variety 90 days maturity), Honampa (normal orange PVA variety 110 days maturity) and ZM305 (normal orange PVA inbred line 85 days maturity). Field evaluations were conducted in 2014 major and minor seasons in eight environments. A three-parameter additive-dominance model was adequate to explain the genetic variation and its importance in the inheritance of the PVA trait in the two hybrids studied. Investigations regarding the gene action of the PVA trait revealed that both additive and dominance gene effects were present. However, the additive gene effect was higher than the dominance gene effect for the PVA trait indicating the predominant role of additive component of gene action in the inheritance of the PVA trait. The implications of these results are that, the PVA trait was highly heritable and development of PVA varieties through hybridization and early segregating generation selection may play a significant role in the improvement of the PVA trait. It was therefore recommended that, the use of population improvement, bi-parental mating, recurrent selection, self progeny selection and diallel selective mating system might help in the breeding for the PVA trait in maize.

Keywords: Vitamin A deficiency, Additive, Dominance, Gene effect, Genetic variation.

INTRODUCTION

Vitamin A deficiency (VAD) is responsible for a number of disorders that range from impaired iron mobilization, growth retardation, and blindness to a depressed immune response, increased susceptibility to infectious disease and increased childhood mortality and morbidity affecting several children and pregnant women worldwide (Sommer and Davidson, 2002; WHO, 2009). Breeding for biofortified crops including maize offers a long term

solution to this problem. However, breeding efforts would require the knowledge of the genetic control of the PVA trait in order to inform breeders of appropriate breeding schemes to use in their breeding programmes.

An approach to study the genetic effects could be accomplished by Generation Means Analysis (GMA) (Frank and Hallauer, 1997). GMA is a simple but useful technique for estimating genetic effects for a polygenic trait. It is very useful in estimating epistatic genetic effects such as additive×additive, dominance×dominance, and additive×dominance effects (Singh & Singh, 1992; Sher et al. 2012; Frank & Hallauer 1997). Since this analysis takes into account trait means and not variances, its sensitivity and accuracy might be of significant influence to maize breeding programmes (Hallauer & Miranda 1988).

In GMA, the A, B, C and D scaling tests are used to explain the adequacy of the various parameter models used. The significance of A and B scale tests indicates the presence of all types of non-allelic gene interactions. The significance of C scale suggests dominance×dominance [I] type of epistasis while that of D scale reveal additive×additive [i] gene interaction (Singh & Narayanan 1993). When the values of one or more of A, B, C and D scaling tests are not significantly different from zero in a cross, it implies that, complex factors such as maternal effects and non-allelic interactions may be absent in the genetic control of the trait being studied. Thus, the additive–dominance model would be enough to show the genetic variation in traits being studied (Kearsey & Pooni, 1996). On the other hand, if these parameters were significantly different from zero for a cross, it implied model was inadequate and further tests such as joint scaling tests may be required (Wannows et al. 2015; Fisher & Yates, 1963; Kearsey & Pooni, 1996). Joint scaling tests may normally begin first with a mean [m] model only. If this model is adequate in explaining the variation in the trait, then fitting of further genetic parameters may not be necessary. Otherwise, the simplest model next to fit would be [m] and additive [d] model or if these two models are not adequate then dominance [h] parameter would be added. It should be noted that, only significant parameters are retained at each step and additions made to arrive at a fit that would be adequate to explain the data considering few and simple parameter models (Kearsey & Pooni 1996).

When additive genetic effects are significantly detected, it may suggest that recurrent selection, intra-population improvement schemes, such as recurrent mass, half-sib, or selfed progeny selection and early generation selection may be useful (Miles et al. 1980). When dominance genetic effects are significantly observed, hybridization or heterosis breeding may

be recommended (Hasib et al. 2002). Considering, the phenomenon of heterosis, a better parent with more favourable alleles may require a little amount of dominance to bring about hybrid vigour in the F1s (Kearsey & Pooni 1996). The presence of duplicate type of epistasis may be evidenced from opposite sign of dominance [h] and dominance \times dominance [l]. This may cancel or weaken hybrid combination and hinder the progress made under selection and therefore, selection would have to be deferred until later generation of segregation where dominance effects might be dissipated (Hasib et al. 2002). When both additive and non-additive gene effects are significantly detected, the use of population improvement, biparental mating, recurrent selection and diallel selective mating system might help (Hasib et al. 2002; Azizi et al. 2006). However, when the dominance gene effect is higher than additive gene effect, indicating predominant role of dominant component of gene action in inheritance of traits, then the selection for such traits should be delayed to later generation when dominant effect is diminished (Azizi et al. 2006; Ishfaq 2011; Sofi et al. 2006; Iqbal et al. 2010; El-Badawy 2012; Shahrokhi et al. 2013; Wannows et al. 2015).

The negative or positive signs for additive effects and additive \times dominance effect depend on which parent is chosen as P1 (Cukadar-Olmedo & Miller 1997; Edwards et al. 1975; Shahrokhi et al 2011). Epistasis in the form of duplicate gene interactions may occur where opposite signs for the estimates of dominance [h] and dominance \times dominance [l] effects are observed. However, if dominance [h] and dominance \times dominance [l] effects have the same sign it implied complementary type of gene action and therefore heterosis breeding may be recommended (Shahrokhi et al 2011; Haq et al. 2013). Opposite sign between additive [d] and additive \times additive [i] gene effect may suggest the oppositional nature of interaction in these traits (Shahrokhi et al 2011; Haq et al. 2013). A preponderance of non-additive gene action for PVA carotenoids was reported by Halilu et al. (2016).

The objective of this research therefore was to unravel the genetic control of Pro-vitamin A trait among maize hybrids in Ghana.

MATERIALS AND METHODS

The parents for the crosses included Akposoe (Quality Protein Maize (QPM) variety 80-85 days maturity), Aburohema (QPM variety 90 days maturity), Honampa (normal orange PVA variety 110 days maturity) and ZM305 (normal orange PVA inbred line 85 days maturity). Study sites included Kwadaso, Fumesua (Forest zone), Ejura, Akumadan (Forest-transition zone), Pokuase (Coastal savanna), Nyankpala (Guinea savanna) in the major season

of 2014 and at Kwadaso and Pokuase in the minor season of the same year representing 8 environments.

Design, management of trials and data recording

The design used was a randomized complete block design with 3 replications. The row length was 5m and row spacing was 75 cm x 25 cm but number of rows and plants for data recording differed according to the type of generation as indicated by Iqbal et al. (2010). Thus, the samples for data could be representative enough as follows: Parents and F1s= 2 row plot, data on 10 random plants; BC1s and BC2s= 4 row plot, data on 20 random plants; F2s= 8 row plot, data on 30 random plants. Controlled pollinations were used in the stated samples for carotenoid analysis from 5 environments where the pollinations were done.

Determination of total pro-vitamin A content

Total PVA content in $\mu\text{g/g}$ of dry matter was calculated for each sample as the sum of β -carotene + 0.5 (β -cryptoxanthin) (Babu et al. 2013a). Since PVA values for the white materials were zero the PVA contents obtained were transformed as: PVA value obtained + 0.05 to enable analysis to be done.

Generation Mean Analysis

The Generation Mean Analysis of the six populations (P1, P2, F1, F2, BC1 and BC2) was performed using R software version 3.3.1. First, scaling tests A, B, and C were performed according to according to Mather & Jinks (1982). In order to estimate the net additive and net dominance effects, a joint scaling test was performed. A three-parameter weighted multiple linear regression model (additive-dominance) was fitted to obtain m , $[d]$, and $[h]$, representing mid-parental value, net additive, net dominance effects respectively. The weighted multiple linear regression model had the form:

$$wt_i y_i = wt_i (\beta_0 + \beta_1 x_{Ai} + \beta_2 x_{Di} + \varepsilon_i)$$

Where; wt_i = weights of the generation means (family size/variance)

y_i = response variable (mean of the i th generation/family)

B_0 = the intercept = the mid-parent value, m

B_1 = the net additive effect = $[d]$

B_2 = the net dominance effect = $[h]$

E = the random error

x_{Ai} and x_{Di} = the coefficients of the additive and dominance effects in the equations for predicting the expected generation means using an additive-dominance model (Table 1).

The observed generation means values were then compared to the expected means after the model fitting by performing a Chi-square goodness of fit test as described by Mather and Jinks (1982). A non-significant Chi-square test indicated the adequacy of the simple additive-dominance model.

Table 1. Expectations of the six families on an additive-dominance model

Generation	M	A	D
		<i>x_A variable</i>	<i>x_D variable</i>
P2	1	1	0
P1	1	-1	0
F1	1	0	1
F2	1	0	0.5
B1	1	0.5	0.5
B2	1	-0.5	0.5

RESULTS & DISCUSSIONS

Gene Effects

Table 2 shows mean, standard errors, coefficient of variation, variances, weights and additive-dominance model for pro-vitamin A contents of top cross hybrid (Akposoe x ZM305). There were significant differences between the generation means for the PVA contents. ZM305 donor parent had the highest content of 3.19 µg/g. Table 3 shows Mean, standard errors, coefficient of variation, variances, weights and additive-dominance for pro-vitamin A contents of varietal cross hybrid (Aburohemaa x Honampa). There were significant differences between the generation means for the PVA contents. Honampa donor parent had the highest content of 4.22µg/g.

Table 4 shows the results of estimates of gene effects for the PVA contents of the two hybrids studied. The three-parameter model was adequate to demonstrate the genetic variation and its importance in the inheritance of the PVA trait in the two hybrids studied. In the top cross hybrid (Akposoe x ZM305), Additive [d] and dominance [h] effects were prominent though both were not significant but $[d]=0.27^{ns}>[h]=-0.62^{ns}$. A similar trend was observed in the varietal cross hybrid (Aburohemaa x Honampa) however, additive [d] effects were significant with $[d]=1.86^{*}>[h]=-0.32^{ns}$. The additive [d] and dominance [h] gene effects observed in both crosses with additive values higher than dominance indicated that genetic inheritance of the PVA trait was highly heritable and was controlled by additive genetic effects (Wong et al.

1998; Egesel et al. 2003b; Menkir & Maziya-Dixon, 2004; Grüneberg et al. 2005; Pfeiffer & McClafferty 2007; Kandianis et al. 2013). The results were in contrast with Azizi et al. (2006) & Halilu et al. (2016) who observed in their PVA trial that, dominance genetic effects were higher than additive effects for the PVA trait. Since both additive and dominance gene effects were significantly detected in the present study, the use of hybridization, population improvement, bi-parental mating, recurrent selection and diallel selective mating system, self progeny selection and early generation selection might help in the breeding for the PVA trait (Miles et al. 1980; Hasib et al. 2002; Azizi 2006; Al-Tabbal & Al-Fraihat 2012).

Table 2. Mean, standard error, coefficient of variation, variances, weights and additive-dominance model for five basic generations of pro-vitamin A contents of top-cross hybrid.

Generation	\bar{X}	S_x^2	$S_{\bar{x}}^2$	Akposoe x ZM305			
				Weighted \bar{X}	m	[d]	[h]
P2	3.19	0.32	0.03	31.1	1	1.0	0.0
F1	2.30	0.61	0.03	32.8	1	0.0	1.0
F2	2.54	1.21	0.06	16.5	1	0.0	0.5
BC1	2.63	0.40	0.02	50.1	1	0.5	0.5
BC2	3.09	0.47	0.02	42.6	1	-0.5	0.5
SED	0.23**						
CV%	23.2						

SED=Standard Error of Difference of Means; S^2 =Variance; P2 (ZM305)=Parent; F1,F2 and BC1, BC2=First and second filial and backcross generations respectively; CV=Coefficient of variation; p=probability; **=Highly significant at $p<0.01$.

N.B Parent 1 (P1) was equated to zero due to zero PVA content.

Table 3. Mean, standard errors, coefficient of variation, variances, weights and additive-dominance model for five basic generations of pro-vitamin A contents of varietal cross hybrid.

Generation	\bar{X}	S_x^2	Aburohemaa x Honampa				
			$S_{\bar{x}}^2$	Weighted \bar{X}	m	[d]	[h]
P2	4.22	0.26	0.03	37.8	1	1.0	0.0
F1	2.01	0.36	0.02	56.2	1	0.0	1.0
F2	1.87	0.49	0.02	40.6	1	0.0	0.5
BC1	1.26	0.26	0.01	76.2	1	0.5	0.5
BC2	3.06	0.79	0.04	25.4	1	-0.5	0.5
SED	0.23**						
CV%	23.2						

SED=Standard Error of Difference of Means; S^2 =Variance; P2 (Honampa)=Parent; F1, F2 and BC1, BC2=First and second filial and backcross generations respectively; CV=Coefficient of variation; P=Probability; **=Highly significant at $p<0.01$
N.B Parent 1 (P1) was equated to zero due to zero PVA content.

Table 4. Estimates of gene effects for pro-vitamin A contents of two maize hybrids

Parameter	Estimate	Akposoe x ZM305			Aburohemaa x Honampa			
		Std. error	t-value	P ($> t $)	Estimate	Std. error	t-value	P ($> t $)
Mean	3.05	0.25	12.01	0.007**	2.29	0.19	11.85	0.007**
Additive [d]	0.27	0.23	1.15	0.371 ^{ns}	1.86	0.19	9.79	0.010*
Dominance [h]	-0.62	0.41	-1.51	0.270 ^{ns}	-0.32	0.31	-1.03	0.41 ^{ns}
Scaling Test								
A	P=0.09 ^{ns}			P=0.81 ^{ns}				
B	P=1.09 ^{ns}			P=1.06 ^{ns}				
C	P=0.03*			P=0.28 ^{ns}				
χ^2 (0.05,df=2)	3.78 ^{ns}			3.34 ^{ns}				

P=Probability; **,*=Highly significant at $p<0.01$ and $p<0.05$ respectively; ns=Non-significant.

CONCLUSIONS

It was concluded from the study that the PVA trait was highly heritable and both additive and dominance gene effects were important in the breeding of Pro-vitamin A maize. Thus, various breeding methods for pro-vitamin A maize trait were suggested.

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