

Genetic Variation and Foliar Yield Index in Amaranth (*Amaranthus* spp)

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ABSTRACT

Foliar yield are important traits in amaranth. Thirty-eight genotypes of amaranth were evaluated to determine genetic variation and selection indices for foliar yield in the crop. Field experiment was carried out at the Federal University of Agriculture, Abeokuta during the late growing season of 2010 and 2011. Randomized complete block design with three replicates was used. Significant ($p < 0.05$) difference was observed among the genotypes for the traits evaluated. Specific leaf area, fresh leaf, stem and root weights and leaf dry matter had high phenotypic and genotypic coefficient of variations, heritability estimates and genetic advance. Leaf dry matter had significant genotypic correlation coefficients with specific leaf area (-0.70), fresh leaf weight (0.59), fresh root weight (0.60) and biomass weight (0.39). Biomass weight (10.72) and leaf area (0.56) had positive direct effect on leaf dry matter was revealed for. Co-heritability between leaf dry matter and other traits varied from 0.94 (harvest index) to 1.29 (leaf area) which revealed simultaneous inheritance between the traits. Biomass weight and specific leaf area were considered as important selection index for foliage yield in amaranth.

Key words: Heritability, Path analysis, Specific leaf area, Trait association, Vegetable amaranths

INTRODUCTION

Amaranth is an important leafy vegetable in Africa (Adebooye *et al.*, 2003; Maundu *et al.*, 2009), and the most consumed vegetable in Southwest Nigeria. It is the most adapted vegetable crop in Africa due to its ability to withstand drought, heat, pests and diseases (Kadereit *et al.*, 2003). About 70 species of amaranth had been documented (Brenner *et al.*, 2000), of which 17 species are with edible leaves (Grubben and Denton, 2004). Common species cultivated in Africa include *A. blitum*, *A. caudatus*, *A. cruentus*, *A. dubius*, *A. hypochondriacus*, *A. spinosus*, *A. thunbergii*, *A. tricolor* and *A. viridis* (Enoch *et al.*, 2014). Tender leaves, petioles and stems of amaranth species usually grown as vegetables (*A. cruentus*, *A. dubius* and *A. tricolor*) are consumed raw or cooked and they have better taste than the grain types (Alegbejo, 2013).

Amaranth is a potential crop that can reduce under-nutrition and used to treat constipation, anaemia and kidney complaints in pregnant women and children (Smith and Eyzaguirre, 2007; Onyango, 2010). Extract from the root boiled with honey are used as laxative, tape worm expellant and wound dressing (Schippers, 2002). The leaves are rich in lysine, methionine and cysteine (Segura-Nieto *et al.*, 1994). Also, the leaves contain high level of vitamin A, vitamin K, vitamin B₆, vitamin C, riboflavin and folate, and dietary minerals including calcium, iron, magnesium, phosphorus, potassium, zinc, copper and manganese (Priya *et al.*, 2007). Amaranth has high nutritional quality because

of its high dry matter content compared to other important vegetables, (Aletor *et al.*, 2002; Iheanacho and Udebuani, 2009; Schönfeldt and Pretorius, 2011). However, it contains anti-nutritional properties like hydrocyanic and oxalic acids which are easily removed by boiling (Grubben and Denton, 2004).

In spite of the importance of this crop as a vegetable, less emphasizes has been placed on genetic improvement of its foliar yield. Therefore, there is need to explore breeding potentials in this crop. This study was carried out to determine the extent of genetic variability and selection indices for foliar yield in amaranth.

MATERIALS AND METHODS

Thirty-eight accessions of amaranth maintained at the National Centre for Genetic Resources (NACGRAB), Nigeria were used. Field experiment was carried out at the Federal University of Agriculture, Abeokuta, Nigeria (between Lat 7°10'N and 7°58'N, Long 3°20'E and 4°37'E, Alt 159 masl) at the late growing seasons of 2010 and 2011. The genotypes were assessed for foliar yield and related traits in a randomized complete block design with three replicates. The plots consisted of three rows, 1.80 m long, and were separated at 0.30 m. Seedlings raised in the nursery using top soils were transplanted at 2 weeks after

planting with plant spacing of 0.30 m. Inter plot spacing was 0.50 m. The plants were thinned to one plant per hill at 2 weeks after transplanting (WAT) to maintain a total of 15 plants per plot. At 5 WAT when the plants are still tender, the following traits were evaluated on 10 randomly selected plants per plot: plant height (cm); stem width at 0.20 m from soil level; number of leaves; length and width of 3rd leaf from top of the plant (cm), petiole length (cm), fresh weights of leaf (including petiole), stem and root weight (g); leaf/stem ratio estimated as the ratio of fresh weights of leaf to stem; leaf area was estimated with regression equation $Y = 0.93 + 0.63X$ (cm²) where X is the product of leaf length and width (Adetimirin, 2007); specific leaf area was estimated as ratio of leaf area to leaf dry matter (cm²/g); biomass weight was computed as the total fresh weight of leaf and stem (g); harvest index as ratio of biomass weight to whole plant weight; leaf dry matter was estimated from 20 g fresh leaf oven dried at 50°C for 12 hrs.

Statistical analysis

Analysis of variance was performed across years for each character evaluated. Genotype and year were considered as fixed factors in a linear model (SAS, Version 9.1.1, SAS, Inc., Cary, NC), $Y_{ijk} = \mu + b(y)_{jk} + g_i + y_j + (g \times y)_{ij} + e_{ijk}$ where Y_{ijk} = overall observation, μ = overall mean, $b(l)_{jk}$ = block within year effect, g_i = fixed genotype effect, y_j = fixed year effect, $(g \times y)_{ij}$ = genotype \times year effect and e_{ijk} = random error. The mean squares were used to estimate phenotypic (σ_p^2) and genotypic (σ_g^2) variances and the variances were used to calculate coefficients of variation ($\sqrt{\sigma_p^2 \text{ or } \sigma_g^2} / \bar{X} \times 100$), broad-sense heritability (σ_g^2 / σ_p^2) and genetic advance at 5% selection differential ($\sigma_p \times \sigma_g^2 / \sigma_p^2 \times k$) where k is selection differential and \bar{X} is grand mean (Johnson *et al.*, 1955; Miller, 1974).

Correlation coefficients ($Cov_{xy} / \sqrt{\sigma_x^2 \times \sigma_y^2}$) were evaluated between the characters where σ_x^2 and σ_y^2 are phenotypic and genotypic variances of characters x and y and Cov_{xy} is covariance between the characters (Falconer, 1989). Genotypic correlation coefficients were partitioned into direct and indirect effects of the traits on leaf dry matter (Dewey and Lu, 1959). Also, co-heritability was estimated between characters ($GCov_{xy} / PCov_{xy}$) where $GCov_{xy}$ and $PCov_{xy}$ are genotypic and phenotypic covariance between characters, respectively (Janssens, 1979).

RESULTS AND DISCUSSION

Biomass and leaf dry matter yields are important traits in amaranths selected for genetic improvement by breeders and genetic variability of these traits and related traits are important to breeding program of the crop. Significant variation among the amaranth genotypes for all traits except number of leaves revealed that selection is possible for genetic improvement of vegetative characters in amaranth (Table 1). Year effect was not significant for most of the traits. However, genotype \times year effect was significant for stem width, petiole length, leaf length and biomass. This suggested that the genotypes had different performance for these traits across the years, indicating the importance of evaluating the genotypes across several environments to develop stable and specific genotypes. Among the traits, genotypic coefficient of variation (GCV) was high for fresh stem (62.98), root (62.29) and leaf (43.73) weights, specific leaf area (53.40), biomass weight (50.63) and leaf dry matter (38.86) as presented in Table 2. This indicated higher extent for which selection can be done for these traits. High heritability estimates and genetic advance above 50% of the genotype mean were also observed for these traits. Heritability revealed possibility for direct selection of traits based on phenotype and is paramount to genetic advance (Nyquist 1991). High heritability estimates observed for the traits revealed that the expression of the characters are conditioned more by genetic than environmental factors. Furthermore, genetic advance for these traits suggested influence of additive gene on the characters. Therefore, genetic gain is expected for genetic improvement of the traits through selection. Phenotypic correlation coefficients were similar in direction (-/+) and slightly higher in magnitude than genotypic coefficients for character association in amaranth (Table 3). Other studies have also reported this in amaranth (Shukla and Singh, 2002; Shukla *et al.*, 2010; Chattopadhyay *et al.*, 2013). Specific leaf area, fresh leaf weight, fresh root weight, and biomass weight had significant phenotypic and genotypic coefficients with leaf dry matter. However, the association was negative between specific leaf area and leaf dry matter. Specific leaf area had positive genotypic coefficients with leaf length, leaf area and harvest index; and negative with fresh root weight. Biomass weight had positive genotypic coefficient with plant height, petiole length and fresh leaf, stem and root weights. Association observed between the traits indicated pleiotropic nature of genes (Kebede *et al.*, 2001; Nausherwan *et al.*, 2008). Also, since phenotypic correlation coefficient is a function of genotypic and environmental correlation coefficients (Falconer, 1989), the inter-character association was reliable.

Table 1: Mean squares of vegetative characters evaluated in 38 genotypes of amaranths across two years

Source of variation	Block/year (df = 4)	Genotype (G)(df = 37)	Year (Y) (df = 1)	G x Y (df = 37)	Error (df = 148)	CV
Plant height (cm)	43.33	159.34**	71.67	48.8	40.98	0.3
Stem width (cm)	51.23**	7.09**	610.22**	6.98**	2.6	0.36
Number of leaves	335.74**	22.71	864.75**	19.64	18.25	0.63
Petiole length (cm)	20.37**	18.04**	7.88	4.98*	3.03	0.18
Leaf length (cm)	5.65	55.78**	159.69**	14.77**	8.07	0.27
Leaf width (cm)	6.59**	5.85**	35.35**	1.99	1.61	0.15
Leaf area (cm ²)	745.29	1829.89**	10579.67**	533.23	376.09	0.33
Specific leaf area (cm ² /g)	20.74	247.97**	129.14	48.02	44.69	0.62
Fresh leaf weight (g)	4.65	81.97**	3.47	7.98	5.86	0.3
Fresh stem weight (g)	32.96	596.55**	30.46	26.24	18.55	0.33
Leaf/Stem	0.01	0.20**	0.01	0.02	0.02	0.24
Fresh root weight (g)	1.88	47.91**	0.52	2.43	2.03	0.32
Biomass weight (g)	117.44	1409.20**	0.02	111.19	65.6	0.36
Harvest index	0.01	0.03**	0.01	0.01**	0	0
Leaf dry matter (g)	2.52	50.40**	1.68	4.34	3.45	0.26

* significant at $p \leq 0.01$, **significant at $p \leq 0.05$, Degree of freedom (df), Coefficient of variation (cv)

Table 2: Range, mean, phenotypic and genotypic coefficients of variability, heritability (broad-sense) and genetic advance of vegetative characters in amaranths

Character	Range	Mean	PCV (%)	GCV (%)	Heritability (%)	GA (%)
Plant height (cm)	11.98 - 34.54	23.16	22.25	18.53	69.37	31.8
Stem width (cm)	2.16 - 6.89	4.49	24.21	3.02	1.55	0.77
Number of leaves	2.89 - 11.85	6.78	28.69	10.55	13.52	7.99
Petiole length (cm)	6.31 - 12.58	9.92	17.48	14.87	72.39	26.07
Leaf length (cm)	7.14 - 18.32	10.7	28.5	24.43	73.52	43.16
Leaf width (cm)	6.89 - 11.61	8.43	11.71	9.51	65.98	15.92
Leaf area (cm ²)	36.76 - 93.56	58.1	30.06	25.3	70.86	43.88
Specific leaf area (cm ² /g)	4.36 - 29.22	10.81	59.47	53.4	80.63	98.78
Fresh leaf weight (g)	2.43 - 18.30	8.03	46.03	43.73	90.26	85.59
Fresh stem weight (g)	6.90 - 46.64	15.48	64.41	62.98	95.6	126.86
Leaf/Stem	0.21 - 0.95	0.58	31.56	29.86	89.55	58.22
Fresh root weight (g)	1.45 - 11.06	4.42	63.93	62.29	94.93	125.02
Biomass weight (g)	12.22 - 70.02	29.05	52.76	50.63	92.11	100.1
Harvest index	0.63 - 0.93	0.81	8.13	6.76	69.23	11.59
Leaf dry matter (g)	3.11 - 14.11	7.13	40.65	38.86	91.39	76.53

Phenotypic coefficient of variation (PCV); genotypic coefficient of variation (GCV); genetic advance (GA)

Correlation may not give precise information about causal effect on dependent variable because it only measures mutual association between two traits. It is important therefore to distinguish traits used as selection index from correlated traits that may not have direct association with the dependent trait. Furthermore, co-heritability will improve efficiency of selection. It will reveal traits that are jointly inherited (Srivastava and Jain, 1994; Singh and Narayanan, 1997). Causal effects of other traits on leaf dry matter revealed high and positive direct effects of biomass weight and leaf area (Table 4). Direct values of the two traits were higher than their correlation coefficients with

leaf dry matter. This was due to their decreasing effect through fresh leaf and stem weights. Fresh leaf and root weights had negative direct effect on leaf dry matter, their high and positive correlation coefficients with leaf dry matter were through biomass weight. Consistently, specific leaf area had negative effect on leaf dry weight. It had increasing negative effect on leaf dry matter through leaf length and biomass weight. It was reported in a range of crop that low specific leaf area maintained high water use efficiency and was related to higher chlorophyll per unit leaf area (Songsri *et al.*, 2009). They reported that thicker leaves have low specific leaf area compared with thinner

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Table 3: Phenotypic (above diagonal) and genotypic (below diagonal) correlation coefficients between vegetative characters in 38 genotypes of amaranth

Character	Plant height (cm)	Stem girth (cm)	Number of leaves	Petiole length (cm)	Leaf length (cm)	Leaf width (cm)	Leaf area (cm ²)	Specific leaf area (cm ² /g)	Fresh leaf weight (g)	Stem leaf weight (g)	Leaf / Stem	Fresh root weight (g)	Biomass weight (g)	Harvest index	Leaf dry matter (g)
Plant height (cm)		0.12	-0.1	0.48**	0.40**	0.01	0.43**	0.11	0.28	0.31*	-0.07	0.24	0.33*	0.1	0.26
Stem width (cm)	0.16		-0.21	-0.14	-0.19	0.39*	-0.06	-0.05	0.12	0.09	-0.17	0.07	0.11	0.05	0.17
Number of leaves	-0.12	-0.27		-0.12	0.07	0	0.06	0.09	-0.02	-0.06	0.11	-0.04	-0.06	-0.05	-0.11
Petiole length (cm)	0.54**	-0.15	-0.15		0.35*	0.22	0.45**	0.1	0.44**	0.42**	-0.04	0.54**	0.52**	-0.08	0.23
Leaf length (cm)	0.36*	-0.22	0.16	0.41**		-0.17	0.93**	0.65**	0.21	0.11	-0.02	0.06	0.13	0.33*	-0.06
Leaf width (cm)	-0.09	0.49**	-0.07	0.26	-0.27		0.2	0.03	0.35*	0.15	0.24	0.25	0.26	0.02	0.27
Leaf area (cm ²)	0.37*	-0.05	0.14	0.55**	0.93**	0.09		0.64**	0.36*	0.19	0.05	0.2	0.26	0.31*	0.07
Specific leaf area (cm ² /g)	-0.01	-0.03	0.22	0.12	0.62**	-0.04	0.58**		-0.11	0.01	-0.11	-0.29	-0.1	0.46**	-0.66**
Fresh leaf weight (g)	0.3	0.12	-0.04	0.48**	0.23	0.41**	0.42**	-0.17		0.55**	0.37*	0.73**	0.79**	0.05	0.52**
Fresh stem weight (g)	0.34*	0.09	-0.1	0.44**	0.12	0.17	0.23	0	0.54**		-0.46**	0.37*	0.93**	0.38*	0.14
Leaf/Stem	-0.1	-0.18	0.16	-0.02	-0.03	0.31*	0.06	-0.16	0.37*	-0.47**		0.24	-0.19	-0.32*	0.23
Fresh root weight (g)	0.27	0.08	-0.04	0.56**	0.07	0.28	0.23	-0.35*	0.75**	0.37*	0.26		0.64**	-0.49**	0.57**
Biomass weight (g)	0.37*	0.12	-0.09	0.54**	0.14	0.29	0.3	-0.13	0.79**	0.93**	-0.2	0.65**		0.18	0.36*
Harvest index	0.11	0.05	-0.1	-0.1	0.38*	0.03	0.37*	0.54**	0.02	0.38*	-0.35*	-	0.17		-0.29
Leaf dry matter (g)	0.3	0.17	-0.16	0.24	-0.05	0.29	0.1	-0.70**	0.59**	0.16	0.27	0.60**	0.39*	-0.29	

Table 4: Causal effects of vegetative characters on leaf dry matter in 38 genotypes of amaranth

Character	Direct effect	Indirect effect													
		Plant height	Stem girth	No leaves	Petiole length	Leaf length	Leaf width	Leaf area	Specific leaf area	Fresh leaf weight	Fresh stem weight	Leaf/Stem ratio	Fresh root weight	Biomass weight	Harvest index
Plant height	0.07		0.02	-0.02	0.09	-0.13	0.02	0.21	0.01	-0.85	-2.89	0.01	-0.26	3.91	0.12
Stem girth	0.11	0.01		-0.04	-0.03	0.08	-0.11	-0.03	0.03	-0.35	-0.76	0.01	-0.08	1.28	0.05
Number of leaves	0.15	-0.01	-0.03		-0.03	-0.06	0.02	0.08	-0.19	0.12	0.83	-0.01	0.04	-0.97	-0.11
Petiole length	0.17	0.04	-0.02	-0.02		-0.15	-0.06	0.31	-0.1	-1.35	-3.74	0	-0.54	5.81	-0.11
Leaf length	-0.36	0.03	-0.02	0.02	0.07		0.06	0.52	-0.52	-0.65	-1	0	-0.06	1.46	0.41
Leaf width	-0.23	-0.01	0.05	-0.01	0.04	0.1		0.05	0.03	-1.16	-1.43	-0.02	-0.27	3.1	0.03
Leaf area	0.56	0.03	-0.01	0.02	0.09	-0.34	-0.02		-0.49	-1.18	-1.92	0	-0.22	3.18	0.4
Specific leaf area	-0.84	0	0	0.03	0.02	-0.22	0.01	0.33		0.46	-0.01	0.01	0.34	-1.4	0.59
Fresh leaf weight	-2.8	0.02	0.01	-0.01	0.08	-0.08	-0.1	0.24	0.14		-4.65	-0.02	-0.73	8.46	0.02
Fresh stem weight	-8.53	0.02	0.01	-0.01	0.07	-0.04	-0.04	0.13	0	-1.52		0.03	-0.36	9.99	0.41
Leaf/Stem ratio	-0.06	-0.01	-0.02	0.02	0	0.01	-0.07	0.03	0.14	-1.03	4		-0.25	-2.12	-0.38
Fresh root weight	-0.97	0.02	0.01	-0.01	0.1	-0.02	-0.06	0.13	0.29	-2.11	-3.17	-0.01		6.95	-0.55
Biomass weight	10.72	0.03	0.01	-0.01	0.09	-0.05	-0.07	0.17	0.11	-2.21	-7.96	0.01	-0.63		0.18
Harvest index	1.09	0.01	0.01	-0.02	-0.02	-0.14	-0.01	0.21	-0.46	-0.05	-3.23	0.02	0.49	1.8	

Table 5: Co-heritability between vegetative characters of 38 genotypes of amaranth

Character	Stem girth	Number of leaves	Petiole length	Leaf length	Leaf width	Leaf area	Specific leaf area	Fresh leaf weight	Stem leaf weight	Leaf/Stem ratio	Fresh root weight	Biomass weight	Harvest index	Leaf dry matter
Plant height	1.17	0.89	1.01	0.75	-7.21	0.67	-0.07	0.95	1	1.25	1.01	0.99	0.96	1.05
Stem girth		0.99	1	0.99	1.06	0.7	0.5	0.96	0.97	0.97	1.04	0.98	0.87	0.95
Number of leaves			1	1.71	15.58	1.69	1.61	1.38	1.19	1.11	0.79	1.17	1.4	1.13
Petiole length				1.03	1.01	1.02	1.03	1.01	0.99	0.42	1.01	1	1.09	0.99
Leaf length					1.33	0.79	0.75	0.95	0.99	1.52	0.97	0.97	1	0.67
Leaf width						0.35	-0.94	1.01	0.96	1.08	0.98	0.98	1.12	0.95
Leaf area							0.69	0.98	0.99	0.97	0.97	0.98	1.01	1.29
Specific leaf area								1.23	0.14	1.17	1.04	1.14	1	0.91
Fresh leaf weight									0.95	0.91	0.98	0.94	0.3	1.06
Fresh stem weight										0.95	0.98	0.97	0.94	1.07
Leaf/Stem ratio											0.99	0.96	0.98	1.07
Fresh root weight												0.98	0.97	1.01
Biomass weight													0.88	1.04
Harvest index														0.94

leaves. Also, higher chlorophyll per unit leaf area will have greater photosynthetic capacity (Akkasaeng *et al.*, 2003; Arunyanark *et al.*, 2008) and hence higher dry matter content. In general, most of the traits had increasing effect on leaf dry matter through biomass weight and reducing effect via fresh leaf and stem weights. Eighty-seven percent (87%) of the cause-effect relationship of the traits on dry matter yield was explained by the path analysis.

All the traits had positive co-heritability with leaf dry matter which revealed simultaneous inheritance between leaf dry matter and the traits (Table 5). The highest co-heritability value was recorded for leaf area (1.29) followed by number of leaves (1.13). High and positive co-heritability was also recorded among number of leaves, specific leaf area and biomass weight. Negative co-heritability observed among specific leaf area, plant height and leaf width indicated that these traits are not jointly inherited.

CONCLUSION

Direct and indirect selection of low specific leaf area (thicker leaves) and higher biomass weight can be considered for genetic improvement of leaf dry matter in amaranth. Also, genetic gain is expected for joint improvement of biomass weight through higher number of leaves, and low specific leaf area through wider leaf width.

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