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Full Length Research Paper

Combining ability and heterosis for different agronomic traits in maize (*Zea mays* L.) under drought stress in the Sudan Savanna of Borno State, Nigeria

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Combining ability variances were estimated for grain yield and its related traits in maize (*Zea mays* L.). Nine maize varieties consisted of five IITA open pollinated varieties (OPVs) and four local varieties were crossed in line x tester mating design. During the 2007 cropping season to determine the general combining ability (GCA), specific combining ability (SCA) effects and heterosis. Parents and hybrids were evaluated in Damboa during the cropping seasons of 2008/2009. Significant level of genetic variability among the parental lines and their hybrids for days to tasseling, days to silking, anthesis silking interval (ASI), plant height, ear height, weight of cobs, dehusked cobs and grain yield, thus suggesting the possibility for genetic improvement. The relatively smaller proportion of GCA to SCA ratio indicated that the predominance of non-additive genetic effects with respect to all the traits except number of cobs per plant and number of cobs per plot. This suggests that high performing hybrids such as EVDT-99WSTRC0 x EX-DAMBOA WHITE, EVDT-99WSTRC0 x EX-BIU WHITE, EVDT-99WSTRQPMC0 x EX-DAMBOA YELLOW and TZECOMP₃DTC₁ x EX-BIU YELLOW may be used to develop potential varieties. Grain yield superiority of some hybrids over the higher parents was recorded suggesting the possibility of their commercial exploitation. The parents: EVDT-99WSTRC0, TZE-WDTSTRQPMC0, and EX-DAMBOA WHITE were identified as the best combiners in terms of GCA for days to tasseling, days to silking, number of cobs/plant, number of cobs/plot, dehusked cobs and grain yield.

Key words: Combining ability, heterosis, Nigeria maize, drought.

INTRODUCTION

Maize (*Zea mays* L.) is the most important cereal crop in Sub-Sahara Africa (SSA) and an important stable food for

more than 1.2 billion people in SSA and Latin America. Globally, maize is ranked the third most important cereal

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crop, after wheat and rice. It is one of the widely cultivated cereal crops due to its adaptation to a wide range of environments and considered as major staple food crop in Nigeria and receiving much attention in industrial development. Africa harvests 29 million hectares, with Nigeria, the largest producer harvesting 3% (FAO, 2009).

World maize production was estimated to be 950 million tonnes, for the 2012/2013 season, an increase of 9% from 2011/2012 (Brandt, 2013). However, according to IITA, (2011), Africa produces 6.5% and the largest African producer is Nigeria with nearly 8 million tonnes, followed by South Africa. Africa imports 28% of the required maize from countries outside the continent. Drought stress is one of the most important environmental stresses affecting agricultural productivity worldwide particularly in the study area (Sudan savanna). It is a major abiotic constraint to maize production. A lack adequate rainfall can lead to decrease in yield and trigger famines. It is the most devastating maize production constraints in Sudan Savanna of Nigeria. This is because rainfall in this region is unpredictable in terms of establishment (may start early or very late in the season), quantity (some times less than 600 mm/annum), and distribution (could be poorly distributed) (Izge and Dugje, 2011).

Combining ability and heterosis concepts had been successfully studied in this work for the production of high yielding and drought tolerant hybrids. Information about combining ability and heterotic patterns among maize gene pools and populations should assist research programmes in their hybrid development activities (Beck et al., 1991). The needs for breeding maize crop tolerance important for increasing adaptability under stress conditions. The choice for selection and breeding procedure to be used for genetic improvement of crop plants therefore largely depend on the magnitude of genetic variability and the nature of gene action governing the inheritance of desirable traits. It is eminent for plant breeders to be familiar with the potentials of local materials before embarking on population improvement (Aminu and Izge, 2013). To establish a sound basis for any breeding programme, aimed at achieving higher yield, breeders must have information on the nature of combining ability of parents, their behaviour and performance in hybrid combination (Chawla and Gupta, 1984). Such knowledge of combining ability is essential for selection of suitable parents for hybridization and identification of promising hybrids for the development of improved varieties for a diverse agro-ecology (Alabi et al., 1987). As such, drought tolerance breeding has been used as a tool in identifying traits that are most vital in selection in order to improve crop yield and other yield attributes (Hallauer and Miranda-Filho, 1988).

This study aimed to improve maize varieties depending on their great diversity which has yield potential and drought tolerance traits. Estimate of the general and specific combining ability effects and determine the high

parent heterosis existing among the traits.

MATERIALS AND METHODS

Five maize lines (EVDT-99WSTRC0, TZE-WDTSTRQPMC0, EVDT-99WSTRQPMC0, TZECOMP₃DTC₁, and BG9TZECOMP_{3x4}) were selected and classified as drought tolerant genotypes and open pollinated varieties (OPVs), developed in International Institute of Tropical Agriculture (IITA) in Ibadan, Nigeria from diverse sources of germplasm. The second set of parents consisted of four local cultivars (EX-BIU WHITE, EX-BIU YELLOW, EX-DAMBOA WHITE and EX-DAMBOA YELLOW) susceptible to drought predominantly growing by the farmers in the study areas. The materials were crossed in line x tester mating design at the Faculty of Agriculture, Teaching and Research Farm, University of Maiduguri, Nigeria, to generate for the initial breeding population (F₁ hybrids) during the rainy season of 2007 under manual irrigation to generate a total of 20 hybrids. The hybrids produced together with their parents were evaluated during the two rainy seasons of 2008 and 2009 in Damboa. Damboa is located in Sudan Savanna with an average annual rainfall of 500 – 1000 mm distributed within the rainy season period of 100 – 120 days the soil type is sandy loam. The treatments were laid out in a randomized complete block design (RCBD) replicated three times. Each plot size was 5 x 2.75 m, with four rows spaced of 75 x 40 cm intra-row spacing. The sowing was done in (15th August) in order to subject the genotype to moisture stress.

NPK (15:15:15) fertilizer at the rate of 333.3 kg/ha was applied 10 days after planting and urea was applied at the rate of 110 kg/ha four weeks after planting. Growth data were recorded on number of stands/plot, days to 50% tasseling, days to 50% silking, anthesis silking interval (ASI), plant height (cm) and ear height (cm). Harvest data recorded include; number of cobs/plant, number of cobs/plot (g), 100 seed weight (g) and grain yield (kg/ha). The combining ability analysis and the estimates of general combining ability (GCA) and specific combining ability (SCA) effects were done based on the procedures described by Kempthorne (1957) and Singh and Chaudhary (1985) using SPAR 2.0 Statistical Package for Agricultural Research. The significant differences among GCA effects and SCA effects were tested using the formula of Cox and Frey (1984). High parent heterosis was estimated according to Liang et al. (1972).

RESULTS AND DISCUSSION

Analysis of combining ability

Analysis of combining ability variance and variance components for twelve agronomic traits in line x tester design in maize are presented in Table 1. The results showed that there were statistical significant differences among the lines in their variances in plant height, ear height and number of cobs per plot. Similarly, the results for testers indicated that mean squares due to ear height, number of cobs per plant and dehusked cobs expressed significant differences. In another development, the results indicated that, the variance for line x tester interaction were highly significant for days to tasseling, days to silking, plant height and ear height among all the other traits. The results showed additive and non additive effects were both significant ($P < 0.05$) and responsible

Table 1. Analysis of general combining ability (GCA) and specific combining ability (SCA) variance for twelve agronomic traits combined years.

Source of variation	DF	NSP	DTT	DTS	ASI	PHT	EHT	NCPL	NCPT	WC	DC	HSW	GRY
Line	4	29.270	29.425	27.175	0.321	825.300*	959.26**	0.480	89.533*	511734.167	794620.00	15.978	138476.014
Tester	3	20.911	21.964	21.564	0.097	324.243	1125.373**	1.318*	32.62	718411.11	1134960.833**	4.402	688938.326
Line x Tester	12	102.960*	44.908*	43.536*	0.465	1082.056**	1082.056**	0.407	16.511	513315.278	558837.222	8.084	443865.929
Error variance component estimates	56	52.436	19.608	18.656	0.603	383.66	413.66	0.576	42.618	561726.332	590952.808	6.830	387721.082
Line		-3.070	-0.645	-0.682	-0.006	7.408	-5.117	0.003	0-537	-65.880	9824.282	0.329	-12724.579
Tester		2.735	-0.765	-0.732	-0.012	-10.775	1.444	0.034	0.262	6836.528	19204.120	-0.123	8169.080
σ^2_{gca}		0.415	-0.100	-0.101	-0.001	-0.199	-0.277	0.002	0.339	467.714	2051.845	0.016	-373.535
σ^2_{sca}		0.184	2.016	1.951	-0.050	16.121	74.426	0.037	4.183	2770.151	37701.468	0.428	6380.03
$\sigma^2_{gca/sca}$		2.26	-0.050	-0.052	0.02	0.012	-0.004	0.054	0.081	0.169	0.054	0.037	-0.059
Proportional contribution to total variation													
Line		8.27	16.229	15.62	17.93	27.41	19.00	17.84	54.75	19.75	23.92	36.71	6.97
Tester		4.43	9.12	9.30	4.07	8.08	16.72	36.75	14.96	20.80	25.62	7.58	26.01
Line x Tester		87.30	74.59	75.08	78.00	64.51	64.29	45.41	30.29	59.45	50.46	55.71	61.02
GCA (Line + Tester)		12.70	25.41	14.92	22.00	35.49	35.72	53.59	69.71	40.55	49.54	44.29	32.98
GCA/SCA		0.146	0.341	0.199	0.282	0.550	0.557	1.180	2.301	0.682	0.982	0.795	0.540

NSP = Number of stands per plot, ASI = Anthesis silking interval, NCPL = Number of cobs per plant, DC = Dehusked cobs, DTT = Days to 50% tasseling, PHT = Plant height, NCPT = Number of cobs per plot, HSW = 100seed weight, DTS = Days to 50% silking, EHT = Ear height, WC = Weight of cobs, GRY = Grain yield, * = Significant, ** = Highly significant.

for the genetic expression. These results are in agreement with that of Kadams et al. (1999), Izge et al. (2007), Premlathan and Kalamani (2010) and Aminu and Izge (2013). The fact that both additive and non-additive gene actions were important in genetic control of most traits studies means that there is the existence of tremendous amount of variability in the genetic materials evaluated, confirming the results of Bello and Olaoye (2009) and Aminu and Izge (2013). The results for the variance component estimates showed that dehusked cobs had the highest value among the lines. However, the variance component estimates for testers also expressed the highest value for weight of the cobs, dehusked

cobs and grain yield. The estimate of SCA variance was higher than the GCA variance. However, in few cases the estimates of GCA were higher than SCA indicating that additive gene effect was in controlled while, non-additive genetic effect was more important than the additive gene effect as most of the GCA/SCA ratios were less than unity. These results are in agreement with Rojas and Sprague (1952) and Gama et al. (1995) who worked on millet and maize respectively. These results showed that parental lines would be utilized in the development of maize hybrids.

The results for the proportional contribution of lines to total variation are higher than the testers in most of the traits. The lowest contribution to

total variations among the lines was given by grain yield. The results of the interaction between line x tester were higher for all the traits except number of cobs per plant and number of cobs per plot. The GCA/SCA ratio shows that, high values were obtained in respect to number cobs per plant, number of cobs per plot, dehusked cobs and 100-seed weight. However, the low and moderate values were obtained in the remaining traits. The lower proportion of GCA/SCA also indicated that additive x non additive and non additive interactions were not significant among hybrids. However, the importance of additive genetics effects was reported by Alamnie et al. (2006) and Aminu and Izge (2013) in respect of

Table 2. Estimate of general combining ability (GCA) effect for male and female parents for twelve agronomic traits combined years.

Line entries	STD	DTT	DTS	ASI	PHT	EHT	NCPL	NCPT	WC	DC	HSW	GRY
EVDT-99WSTRC0	0.650	-2.200	-1.783	-1.200*	77.612**	54.522**	0.217	26.833**	3139.683**	2807.017**	4.518**	2227.241**
TZE-WDTSTRQPMC0	4.067	-3.200*	-4.033*	2.817**	88.028**	38.822**	-0.042	29.333**	3539.683**	3181.183**	4.943**	2331.483**
EVDT-99WSTRQPMC0	1.733	-2.700	-2.450	-1.533**	1.528	-12.487	-1.308*	1.417	303.017	363.683	-0.998	159.665
TZE-COMP ₃ DTC ₁	-3.267	7.217**	6.550**	-1.117*	-79.497**	-31.770*	3.308**	-26.500*	-3853.733**	-3557.233*	-1.648*	-2345.509**
BG97TZECOMP ₃ x ₄	-3.183	0.883	1.717	-1.367*	-87.672**	-47.087**	-2.175**	-31.083**	-3128.650**	-2794.650**	-6.815**	-2374.880**
SE (±)	2.414	1.52	1.48	0.26	7.48	6.78	0.25	2.17	249.83	256.25	0.83	207.56
Male Entries												
EX-DAMBOA WHITE	1.117	0.133	0.050	1.617**	11.872	4.793	4.532**	7.767**	761.283*	612.450*	3.478*	312.992
EX-DAMBOA YELLOW	3.183	-1.867	0.517	-1.283*	33.645**	16.693**	1.662*	4.833*	668.017*	727.850**	-0.648	439.798*
EX-BIU WHITE	-1.750	1.200	-1.417	-1.550*	-29.762**	-8.167	-1.128*	-6.100*	-424.650*	-367.483	-1.495	-382.214*
EX-BIU YELLOW	-2.550	0.533	0.850	-1.350*	-15.755*	-13.320*	-1.742*	-6.500*	-1004.650**	-972.817**	-1.335	-370.577*
SE (±)	2.09	1.31	1.28	0.22	6.48	5.87	0.22	1.89	216.36	221.92	0.75	179.75

NSP = Number of stands per plot, ASI = Anthesis silking interval, NCPL = Number of cobs per plant, DC = Dehusked cobs, DTT = Days to 50% tasseling, PHT = Plant height, NCPT = Number of cobs per plot, HSW = 100seed weight, DTS = Days to 50% silking, EHT = Ear height, WC = Weight of cobs, GRY = Grain yield, * = Significant, ** = Highly significant.

grain weight in maize.

General combining ability effects

Estimates of general combining ability effects for twelve agronomic traits in maize are presented in Table 2. Among the parents, TZE-WDTSTRQPMC0 expressed positive significant GCA values effects for all the traits except for days to tasseling expressed negative GCA effects, while in case of other characters positive GCA effects are desirable. Similarly, EVDT-99WSTRC0 is the second highest general combiner with positive significant GCA effects for number of cobs per plot, weight of cobs, dehusked cobs, 100-seed weight and grain yield. Therefore, TZE-WDTSTRQPMC0 and EVDT-99WSTRC0 had exhibited highly significant GCA effects in desirable direction for almost all the

traits. These findings are in accordance with Bello and Olaoye (2009) and Aminu and Izge (2013).

The results for testers indicated that EX-DAMBOA WHITE and EX-DAMBOA YELLOW had the highest significant GCA effects. These were due to the adaptation of the testers which were originated from the study area. Maize breeders have therefore, devoted effort to developing superior genotypes for grain yield and adaptation to different stress factors (Olaoye et al., 2005). The results also indicated that, EX-BIU WHITE and EX-BIU YELLOW had negative significant GCA effects for almost all the traits.

Specific combining ability effects of hybrids

The estimates of specific combining ability for twelve agronomic traits in maize evaluated are presented in Table 3. Specific combining ability

effects are used to identify the best cross-combinations for hybrids production (Izge et al., 2007). These studies identified a number of desirable hybrids for some of the agronomic traits such as anthesis silking interval, plant height, ear heights, cobs per plant, cobs per plot, dehusked cobs and grain yield. The SCA effects were significant or highly significant in the twenty hybrids studied for the different agronomic traits. The study revealed that hybrids with high SCA effects involved at least one or two of the several higher general combiners as parent namely: EVDT-99WSTRC0, EVDT-99WSTRQPMC0, EX-BIU WHITE and EX-DAMBOA WHITE. Gama et al. (1995) reported similar result where a hybrid with high SCA effects involved one or both of the good general combiners as parents. Hybrid EVDT-99WSTRC0 x EX-DAMBOA WHITE expressed negative and significant SCA effects for days to tasseling and days to silking.

Table 3. Estimate of specific combining ability (SCA) effect for the hybrids for twelve agronomic traits combined years.

Entries	NSP	DTT	DTS	ASI	PHT	EHT	NCPL	NCPT	WC	DC	HSW	GRY
EVDT-99WSTRC0 x EX-DAMBOA WHITE	5.117	-6.617*	-7.983*	4.467*	-39.538*	-37.802*	3.428*	15.650*	2917.060**	2705.050**	-2.578	2220.038**
EX-DAMBOA YELLOW	-5.517	0.133	0.650	1.200	-15.645	5.568	1.437	-9.167*	-1059.683*	-1053.683*	1.082	-839.798*
EX-BIU WHITE	6.417	3.533	3.250	-3.267*	41.428*	37.258*	4.503*	28.583**	3210.317**	3156.317**	1.028	2415.656**
EX-BIU YELLOW	4.217	1.467	-0.350	0.600	13.755	6.112	0.850	13.167*	679.650	583.650	0.468	523.305
TZE-WDTSTRQPMC0 x EX-DAMBOA WHITE	-8.533*	-0.800	0.300	-4.350*	-6.955	0.798	-4.698*	-9.267*	-1052.950*	1179.117*	-4.870*	-739.659*
EX-DAMBOA YELLOW	-3.600	-1.867	4.33	2.117	-37.395*	-31.002*	1.762	-5.000	-459.683	-361.183	1.723	-156.162
EX-BIU WHITE	5.333	-0.133	0.833	1.383	16.678	-9.908	1.562	7.267*	699.650	867.483*	0.470	699.789
EX-BIU YELLOW	6.800	2.800	2.900	0.850	27.672*	40.112*	1.375	7.000*	812.983	672.817	2.677	196.032
EVDT-99WSTRMC0 x EX-DAMBOA WHITE	0.800	-5.300*	5.2317*	-3.600*	81.878**	30.040*	-2.232	-7.100*	-7.100	-871.617*	3.238*	-625.720
EX-DAMBOA YELLOW	2.067	-4.033	-4.350	1.200	50.772**	14.673	3.790*	13.100*	1132.983*	1341.650*	4.898*	1942.214**
EX-BIU WHITE	-10.333*	2.700	2.917	0.800	-81.255**	-31.800*	0.228	-18.483**	-2863.683**	-	-0.488	-
EX-BIU YELLOW	7.467	6.633*	6.650*	1.600	-51.395**	-12.913	-0.225	-25.750**	-3263.683**	2795.017**	-	2106.574**
TZE-COMP ₃ DTC ₁ x EX-DAMBOA WHITE	17.467**	-7.450*	-8.450*	4.317*	-22.997	23.257*	4.752*	-1.433	-1276.533*	-	7.648**	-
EX-DAMBOA YELLOW	3.733	0.050	0.650	-5.217*	-9.137	-5.577	-5.455*	-6.167	-912.933*	3066.350**	9.522**	2529.120**
EX-BIU WHITE	-7.000	-3.883	4.417	-5.283*	36.170*	7.150	-5.388*	-0.233	903.067*	-871.033*	-5.018*	-818.939*
EX-BIU YELLOW	4.200	-5.113*	-3.355*	4.817*	-4.037	-29.830*	3.908*	9.833*	1286.400*	-962.767*	-4.838*	-1045.048*
BG97TZECOMP _{03x4} x EX-DAMBOA WHITE	-4.617	0.783	0.383	-4.100	-12.388	-26.293*	-4.032*	2.150	165.383	529.233*	2.335	175.994
EX-DAMBOA YELLOW	3.317	5.717*	6.483*	0.700	11.405	27.473*	0.028	-8.250*	-778.017	1304.567	-5.312*	1687.994**
EX-BIU WHITE	5.583	-2.217	2.583	1.633	-13.022	-2.700	2.095	8.350*	127.983	216.717	-2.685	-35.720
EX-BIU YELLOW	-4.283	-4.283	3.517	1.767	14.005	-8.840	1.908	-2.250	484.650	-778.683	3.828*	-374.647
SE (i)	4.18	2.61	2.56	1.48	12.96	11.74	1.44	3.77	432.72	56.650	4.168*	288.577
										505.317	1.51	121.789
										443.83		359.50

NSP =Number of stands per plot, ASI=Anthesis silking interval, NCPL = Number of cobs per plant, DC = Dehusked cobs, DTT =Days to 50% tasseling, PHT = Plant height, NCPT = Number of cobs per plot, HSW= 100seed weight, DTS =Days to 50% silking, EHT = Ear height, WC = Weight of cobs, GRY = Grain yield, * = Significant, ** = Highly Significant.

Negativity of these traits is important, implying that these hybrids could mature earlier and could escape drought. Similar results were reported by Bello and Olaoye (2009) and Aminu and Izge (2013). With respect to ASI, TZECOMP₃DTC₁ x EX-DAMBOA WHITE had the highest positive and significant SCA effects. ASI is a trait used mostly in screening for tolerance to stresses. It is a measure of nicking (synchronization) of pollen shed with silking. This report is in accordance with

finding of Shanghai et al. (1983), Paul and Debenth (1999) and Bello and Olaoye (2009). Nine hybrids expressed significant SCA effect for plant height and ear height. However, four and five of them expressed negative and significant SCA effects with EVDT-99WSTRQPMC0 x EX-BIU WHITE had the highest negative and significant SCA effects. Negative plant height is desirable especially in drought prone and windy areas against drought lodging (Izge et al., 2007;

Aminu and Izge, 2013). For 100-seed weight, hybrid TZECOMP₃DTC₁ x EX-DAMBOA WHITE had the highest positive and significant SCA effects. The hybrids EVDT-99WSTRC0 x EX-DAMBOA WHITE, EVDT-99WSTRC0 x EX-BIU WHITE and TZECOMP₃DTC₁ x EX-BIU YELLOW exhibited positive and significant SCA effects for grain yield. These are good hybrids when breeding for drought stress and grain yield. These hybrids probably have potential as parents of

Table 4. Heterosis of the hybrids over higher parents for twelve agronomic traits combined years.

Entry	NSP	DTT	DTS	ASI	PHT	EHT	NCPL	NCPT	WC	DC	HSW	GRY
EVDT-99WSTRC0 x EX-DAMBOA WHITE	13.05	-4.86	-2.26	5.76	-2.82	-18.33	12.07	18.36	12.68	15.80	5.82	38.34
EX-DAMBOA YELLOW	14.72	-4.00	-3.41	6.26	5.05	14.88	14.29	6.78	-3.10	-4.72	-9.08	-16.24
EX-BIU WHITE	11.31	-6.30	-8.24	4.81	-5.48	-29.43	6.44	19.38	15.89	13.97	5.64	39.46
EX-BIU YELLOW	14.14	7.64	7.85	-10.29	9.18	13.42	3.97	5.88	3.79	7.46	-7.41	10.10
TZE-WDTSTRQPMC0 x EX-DAMBOA WHITE	8.65	-6.64	-4.24	-5.20	-11.73	-25.16	6.20	13.03	9.11	8.69	2.90	18.55
EX-DAMBOA YELLOW	6.34	-6.15	5.97	-4.76	8.38	12.68	3.10	3.17	13.69	9.34	1.93	11.62
EX-BIU WHITE	-13.26	-10.60	-8.78	3.06	-12.89	-32.87	7.36	12.50	4.22	4.58	14.86	11.17
EX-BIU YELLOW	-1.44	-2.17	-1.14	-5.26	-2.28	6.21	4.73	2.94	5.12	7.73	0.95	9.57
EVDT-99WSTRQPMC0 x EX-DAMBOA WHITE	3.66	-3.00	-1.12	2.26	6.78	-8.79	-3.73	1.52	5.60	0.76	2.75	-4.44
EX-DAMBOA YELLOW	13.96	-1.80	-2.23	5.81	-2.27	-18.86	17.86	21.31	19.17	18.86	7.88	40.76
EX-BIU WHITE	9.52	-14.33	-12.5	-14.29	-15.46	-32.90	5.58	12.50	10.60	-10.95	5.31	-31.96
EX-BIU YELLOW	-2.74	-10.51	-8.66	-4.76	-4.51	-33.38	13.57	27.94	12.09	23.07	4.01	21.12
TZE-COMP ₃ DTC ₁ x EX-DAMBOA WHITE	-9.27	-1.18	-1.09	-10.00	-1.99	-5.73	-9.54	-17.81	-32.23	-38.51	-18.88	-39.78
EX-DAMBOA YELLOW	-10.39	-5.60	-5.99	-4.76	-5.75	-23.59	1.30	-26.03	-14.59	-24.18	-5.79	-23.69
EX-BIU WHITE	-2.53	-3.15	-2.93	-4.76	-2.44	-25.23	4.23	-5.48	6.35	-3.52	-20.46	-1.16
EX-BIU YELLOW	13.93	-7.08	-5.18	4.76	-2.78	-23.94	16.78	23.70	21.55	15.12	7.27	41.72
BG97TZECOMP _{3x4} x EX-DAMBOA WHITE	6.42	7.60	8.47	-19.05	2.78	22.03	-16.60	-7.58	0.58	-5.59	-1.01	-0.33
EX-DAMBOA YELLOW	12.92	-2.44	-1.69	-11.31	8.74	2.47	-2.16	-6.78	5.19	0.27	-6.93	1.60
EX-BIU WHITE	3.43	-4.58	-4.79	4.26	1.39	2.61	4.37	13.44	6.15	4.25	4.64	12.05
EX-BIU YELLOW	s8.62	-6.40	-5.65	-5.26	-1.84	-33.09	14.68	13.88	8.80	10.07	0.19	21.92

NSP =Number of stands per plot, ASI=Anthesis silking interval, NCPL = Number of cobs per plant, DC = Dehusked cobs, DTT =Days to 50% tasseling, PHT = Plant height , NCPT = Number of cobs per plot, HSW= 100seed weight, DTS =Days to 50% silking, EHT = Ear height, WC = Weight of cobs, GRY = Grain yield.

hybrid varieties, as well as for inclusion in breeding programs, since they may contribute superior alleles in new populations for high grain yield and other abiotic stresses in maize production especially in Sudano-Sahelian zone. These results are in line with earlier independent studies of Perez-Velasquez et al. (1996), Kumar et al. (1998) and Bello and Olaoye (2009) who reported that maize grain yield and flowering traits were under the control of non-additive (SCA effects) type of gene action.

Heterosis

Estimates of heterosis for twelve agronomic traits in maize are presented in Table 4. The degree of heterosis varied from hybrid to hybrid and from traits to traits. This study showed that great potentials for increased maize yield exist because of the high level of heterosis observed. Both positive and negative heterotic values were recorded for all agronomic traits studied. However, the high positive higher parent heterosis

observed for stands count, number of cobs/plant, cobs/plot, weight of cobs and dehusked cobs directly indicated their importance for total grain yield increased. Aminu and Izge (2013) reported significant and positive level of heterosis for 1000 for these traits in maize. The negative heterosis recorded for traits like days to tasseling, days to silking, plant height and ear height are desirable in breeding for earliness and short stature hybrids that could resist lodging particularly in windy environment. Hybrids EVDT-99WSDTRC0 x EX-

DAMBOA WHITE, EVDT-99WSDTRC0 x EX-DAMBOA YELLOW and EVDT-99WSTRQPMC0 x EX-DAMBOA YELLOWH expressed positive and significant higher parent heterosis for ASI and could be recommended for environment with low and erratic rainfall, because it is one of the drought tolerant traits.

This study indicated tremendous level of higher parent heterosis in grain yield. EVDT-99WSTRC0 x EX-DAMBOA WHITE, EVDT-99WSTRC0 x EX-BIU WHITE, EVD-99WSTRQPMC0 x EX-DAMBOA YELLOW and TZE-COMP₃DTC₁ x EX-BIU YELLOW are among the hybrids that expressed the highest significant higher parent heterosis for grain yield. Low levels of heterosis were observed which could be attributed to narrow genetic base of the materials used in the development of some parents. It is noteworthy that these hybrids appeared to have genes that can be introgressed to exploit heterosis for earliness and high grain yield. These results are in line with earlier independent studies of Bello and Olaoye (2009), Kumar et al. (1988), Joshi et al. (1998) and Perez-Velasquez et al. (1996) who reported that maize grain yield and flowering traits were under the control of non-additive (SCA effect) type of gene action.

Conclusion

The study identified parents: EVDT-99WSTRC0, TZE-WDTSTRQPMC0 and EX-DAMBOA YELLOW as the best general combiners, while hybrids EVDT-99WSTRC0 x EX-DAMBOA WHITE, EVDT-99WSTRC0 x EX-DAMBOA YELLOW and EVDT-99WSTRQPMC0 x EX-DAMBOA YELLOW as the best among the 20 hybrids evaluated since they have the best level of high parent heterosis in ASI, number of cobs/plant, cobs/plot, weight of cobs and dehusked cobs and grain yield. The desirable heterotic levels in days to tasseling, days to silking, plant height and ear height are desirable in areas with marginal rainfalls and windy environment like the study area.

Conflict of Interest

The authors have not declared any conflict of interest.

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Full Length Research Paper

Evaluation of bean qualities of indigenous Arabica coffee genotypes across different environments

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Evaluation of bean qualities of 30 Arabica coffee genotypes were carried out at four different locations (South-western Ethiopia). The genotypes had high overall yield potential, during a preliminary evaluation carried out at Gera. The differences among genotypes for cherry weight (CW), bean weight, parchment length (PL), bean length (BL), floater beans and outturn percent at each location were highly significant ($p < 0.01$). Two genotypes: 8143 and 8213 exhibited exceptionally higher mean values for CW, hundred beans weight (HBW) and BLs. However, coffee genotypes with higher CW or HBW did not exhibit higher outturn compared to those genotypes with lower CW or HBW indicating the needs to apply intensive agronomic practices such as mulching to conserve moisture, pruning to adjust optimum fruit to leaf ratio and adequate fertilization to avoid nutrient shortage. Generally, genotypes exhibited higher CW, HBW, outturn, BL and parchment growth at Gera and Metu than Agaro and Jimma which had relatively favorable climate during the season. Irrespective of the prevailing environmental factors and its higher overall yield potential, genotype 8143 consistently exhibited higher CW, HBW and BL and lower percentage of floater coffee beans.

Key words: Arabica coffee, bean quality, environments, genotypes, indigenous.

INTRODUCTION

Arabica coffee is grown in about 80 tropical and sub-tropical countries. The majority of these countries supply the product to world market. Ethiopia is among these countries which heavily depend on coffee exports for foreign exchange earnings. About 40% of its export is coffee (Alemayehu et al., 2008; Nigussie et al., 2008). The involvement of such many countries in the production and trade increased competition for a sustainable market. Such conditions force the market to consider quality as major criterion to prioritize and ensure higher price for

desirable coffee beans.

The term quality refers to beans flavor in fragrance, aroma, sweetness, acidity, caffeine content or overall taste felt by consumer after drink as well as physical characteristics such as length, width, thickness or weights, shape or color of coffee beans and so on (Giomo et al., 2012; Agwanda et al., 2003; Fox et al., 2013). Basically, there are two economic species of coffee, Arabica and robusta, which are supplied in the world market (Vander Vossen, 1997) and Ethiopia

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produce solely Arabica. There is wide difference among varieties for qualities even within Arabica coffee types (Vander Graaf, 1981; Desse, 2008; Behailu et al., 2008).

Despite the role coffee plays in the national economy and in spite of the fact that Ethiopia is origin of Arabica coffee, an in-depth research study to improve its quality has not been yet undertaken, apart from the limited selection work done to develop desirable varieties with fine flavor. Results of these studies illustrated there were peculiar coffee types in Ethiopia which exhibited fine cup taste (Behailu et al., 2008; Desse, 2008). However, in addition to flavor, improving varieties for desirable bean attribute is important to ensure higher prices (Getachew, 1990). Coffee types with larger beans usually fetch higher prices than smaller ones even though the former does not necessarily produce desirable roast or liquor than the latter (Cavaco Bicho et al., 2010). Both genotype and environment affects beans physical as well as organoleptic properties such as caffeine contents (Agwanda et al., 2003; Fox et al., 2013; Yonas, 2005; Tesfaye et al., 2008; Alemseged and Tesfaye, 2012). Moisture amount received during bean growth is very critical to affect growths of coffee beans (Teskaye et al., 2008; Alemseged and Tesfaye, 2012). Fertility levels of the edaphic factors where coffee bushes grow also affects beans growth (Yonas, 2005). Other than the independent effects of genotypes across environments, there is interaction where genotypes exhibit differential performance across different environments (Mawardi and Hulupi, 1995; Yonas, 2005). But through selection, it was possible to breed coffee genotypes that exhibited minimum interaction for bean quality across wide environments (Yonas and Bayetta, 2008). Since Ethiopia has both wide genetic diversity of Arabica coffee and diverse environments for growing it, conducting adaptation tests across such environments is important to select those genotypes which exhibit consistently superior performance for bean quality traits.

Thus, the objective of this study was designed to assess the bean qualities of the different Arabica coffee genotypes across wide environments and select the superior ones for commercial use.

METHODOLOGY

Experimental sites

The trials were conducted at four different locations in South-western region of Ethiopia: Jimma, Agaro, Metu and Gera. The first three locations represent medium altitude, Gera represents high land and their description is given in Table 1.

Materials

The experimental plots consisted of 30 Arabica coffee genotypes planted out in randomized complete block design (RCBD) of three replications. They represent all the three types of canopy configuration in Arabica coffee: compact, intermediate or open. The

genotypes were selected for, cup quality and yield and their higher resistance to RCB, during a preliminary evaluation at Gera. Primarily, they were collected from farmers' field of different coffee growing parts of the country in South-western Ethiopia. Each plot consisted of 10 bushes in single row. The spacing between rows and bushes within rows were 2 × 2 m, respectively. The materials are presented in Table 2. They were grown in field which had shade of *Susbania susban*. However, the shades were very sparse and light penetration was high. The plots received uniform application of fertilizer and cultural practices throughout the period of data collection. All the coffee bushes were maintained in single stem pruning system. The coffee fruit were matured and harvested on 2 and 15 September at Agaro and Gera, and on 3 and 7 December at Jimma and Metu, respectively. During harvest all the red cherries from all the 10 bushes in a plot were harvested and thoroughly mixed. Ten (10) sample cherries were taken and weighed using Sartorius sensitive balance (with a precision level of four decimal places) immediately after harvest. The weight of 100-sample coffee beans was weight using sensitive balance at 11% standard moisture level. The outturn percent was determined by dividing the dried clean coffee at the stated moisture level to the corresponding sample fresh cherry weight (CW) from which the clean coffee was prepared and multiplied by 100. The mean bean length (BL) of the different coffee genotypes was determined by measuring the length of 10 sample coffee beans and then the sum was divided to the total numbers of beans. The mean parchment length (PL) was also determined in a similar fashion as for BL. The percent of floater beans was determined by taking 100 sample cherries, pulping it immediately after harvest and the pulped beans were soaked in distilled water. The pulped beans that settled were counted. The floating beans with endosperm filled incompletely were also counted, excluding the empty locules. Then the settled and floating beans (incompletely filled beans) were added. Later, the number of floating beans was divided to the sum of the total number of settled and floating beans and multiplied by 100.

Statistical analysis

First, analyses of variances of each trait were carried out at the specific environments using Agrobases software. Later combined analyses of variance for all traits were carried out after error variances at the different environments were confirmed to be homogeneous, to calculate environmental, genotypic and genotype by environment interaction effects. Since the error variances of the different traits at each location were homogenous, pooled error mean squares were used to calculate coefficient of variance (CV) and least significant differences (LSD) for the combined means.

RESULTS AND DISCUSSION

Bean qualities and outturn percent

The differences among coffee genotypes for ten cherries weight (TCW), hundred beans weight (HBW), BL, PL, outturn and floater beans percent (FBP) at the specific locations were highly significant ($p < 0.01$) (Tables 3, 4 and 5). This shows that the indigenous Arabica coffee types in Ethiopia exhibited genetic variability for traits related to determine bean qualities. It also showed that the possibility of these traits could be improved through selection. This enables the country to produce and supply high standard coffee beans to the world coffee market. However, the interactions of genotype by environment of

Table 1. Characteristics of test locations.

Location	Altitude (masl)	Latitude	Longitude	Temperature (°C)		Annual rainfall (mm)
				Minimum	Maximum	
Jimma	1753 m	7°36'5"	36°E	11.5	26.2	1531.8
Agaro	1600	7°9'	36.6E	NA	NA	NA
Gera	1940 m	7°7'	36°E	10.4	24.4	1878.9
Metu	1550	8°33"	36°E	12.5	28.6	1810.6

NA = Not available.

Table 2. Thirty Arabica coffee genotypes evaluated at four different locations.

Serial number	Genotype designation	Serial number	Genotype designation
1	74191	16	8011
2	75187-B	17	8017
3	7453	18	8019
4	74145	19	8021
5	75194	20	8112
6	7512	21	8133
7	7574	22	8136
8	7803-A	23	8143
9	7803-B	24	8144
10	7809-B	25	827
11	802	26	878
12	804	27	8211
13	808	28	8213
14	809	29	8219
15	8010	30	8223

the different traits were highly significant ($p < 0.01$) (Table 6). This may indicate the fact that a genotype which is superior in performance at one set of environments for one agronomic trait may not be superior at a different set and therefore its performance at the different environments must be inspected before it is recommended for commercial use.

Generally, there are four major stages of bean growth in Arabica coffee (Tesfaye et al., 2008). The first stage is pin head and it starts immediately after fertilization. During this stage, the fertilized flower undergoes internal cellular activities such as cell division and does not exhibit much change in size. The second is the berry expansion stage where exocarp and endocarp, that encloses the parchment and beans, respectively, are grown to their full genetic limit unless restricted by external environmental factors such as age of bearing bush, moisture availability, presence of pruning practices, crop load, plant population density, shade level and availability of nutrients in adequate amounts (Tesfaye et al., 2008). The third is bean filling stage during which the parchment is filled with photosynthetic assimilates. The fourth or the last is the maturity stage during which much change in the size of

the berry as well as bean is not noticed except internal processes that facilitate maturity. Each stage stays for nearly 2 months at medium altitude and may be longer at higher elevation areas where the climate is cooler.

From the genotypes evaluated: 8213, 7803A, and 8143 exhibited higher mean value for CWs (Table 3). The weight of coffee cherries across the distinct locations ranged from 14 to 19 g, the least and the highest being observed at Jimma and Metu, respectively. This was a very pronounced difference and showed that environment plays an important role in determining berry size apart from genetic factors. The moisture received from June to September at Jimma during 2009/10 was adequate, however; the restricted berry growth at the particular site could be attributed to shortage of moisture in May which was critical for berry expansion (Figure 1). On the other hand, the highest CW observed at Metu could be attributed to the optimum rainfall received throughout all stages of berry growth at the particular location as shown in Figure 1. Similar justification was stated by Tesfaye et al. (2008) and Tesfaye and Ismail (2008) that moisture amount received during fruit growths has a significant influence on bean quality.

Table 3. Ten cherries weight and hundred beans weight (g) of 30 Arabica coffee genotypes at four different locations during 2009/2010.

Genotype	Ten cherries weight in (g)					Hundred beans weight (g)				
	Jimma	Agaro	Gera	Metu	Combined mean	Jimma	Agaro	Gera	Metu	Combined mean
74191	12.60	12.89	15.38	18.35	14.80	14.49	11.14	17.51	15.89	14.76
75187B	15.82	17.95	17.91	21.13	18.20	18.52	15.21	16.12	18.01	16.97
7453	13.31	13.37	13.92	18.08	14.67	15.35	13.18	15.27	14.81	14.65
74145	15.67	13.69	16.05	16.48	15.47	14.68	10.30	15.81	16.36	14.29
75194	14.13	12.90	18.13	18.42	15.89	15.91	10.54	15.70	15.67	14.46
7512	14.40	14.68	17.35	17.87	16.08	15.35	10.90	16.34	17.57	15.04
7574	10.84	14.36	17.35	20.03	15.65	12.19	11.43	14.21	16.36	13.55
7803A	16.44	16.17	20.72	24.67	19.50	16.98	13.51	19.42	19.22	17.28
7803B	16.48	16.23	17.49	20.05	17.56	16.23	15.05	18.02	17.83	16.78
7809B	15.09	16.80	18.83	20.21	17.73	15.93	14.49	17.84	17.53	16.45
802	12.22	15.51	18.23	18.65	16.15	12.60	11.57	14.51	16.77	13.86
804	13.79	16.30	13.00	17.20	15.07	14.05	13.37	16.96	15.61	15.00
808	12.32	12.99	16.89	18.01	15.05	14.47	10.27	14.05	13.69	13.12
809	16.41	18.88	17.82	19.60	18.18	17.86	14.58	17.62	20.60	17.67
8010	12.80	13.54	14.38	15.93	14.16	13.96	13.26	16.79	12.20	14.05
8011	15.41	13.26	14.74	18.76	15.54	13.57	10.53	14.09	16.77	13.74
8017	13.37	13.86	15.41	18.20	15.21	14.32	9.77	14.73	16.91	13.93
8019	13.30	16.51	18.05	20.76	17.16	13.14	13.56	15.08	15.00	14.19
8021	13.04	12.67	15.70	19.66	15.27	13.86	10.65	14.24	16.28	13.76
8112	14.34	16.71	17.04	18.96	16.76	12.48	13.06	17.54	14.64	14.43
8133	11.21	14.25	14.47	20.16	15.02	12.91	11.75	15.68	15.63	13.99
8136	14.68	16.57	16.47	19.93	16.91	13.28	12.11	17.11	16.52	14.75
8143	16.63	18.94	22.97	22.33	20.22	17.81	14.99	18.86	17.49	17.29
8144	13.48	15.77	18.23	20.47	16.99	14.75	12.37	14.81	16.98	14.73
827	12.80	14.67	16.26	19.33	15.77	15.23	11.51	15.63	16.86	14.81
828	16.62	16.21	17.12	20.69	17.66	15.59	13.04	16.74	18.41	15.94
8211	20.76	19.92	21.54	21.11	20.83	21.82	16.10	18.70	19.89	19.13
8213	16.20	17.08	19.03	25.46	19.44	19.78	12.81	19.05	19.31	17.74
8219	13.62	17.07	18.92	20.27	17.47	16.83	13.26	16.39	17.93	16.10
8223	12.69	15.98	22.97	16.28	16.98	12.73	12.48	12.80	16.30	13.58
Mean	14.35	15.52	17.41	19.57	16.71	15.22	12.56	16.25	16.77	15.2
CV	4.89	5.64	4.41	4.78	4.94	4.11	3.84	3.55	3.65	3.8
LSD _{0.05}	0.71	0.88	0.78	0.95	1.38	0.63	0.49	0.58	0.61	0.97
LSD _{0.01}	0.96	1.19	1.05	1.28	1.87	0.85	0.66	0.79	0.83	1.31

LSD, Least significant differences.

The range among genotypes for HBW was 13.37 to 19.89 g. This is also a very pronounced difference. On average three genotypes: 7803A, 8143 and 8213 exhibited the three highest mean values for HBW as shown in the work. They also exhibited higher mean value for overall mean yields (Yonas and Bayetta, 2008). The range for HBW across the distinctive locations was 12.56 to 16.97 g the least and the highest being observed at Agaro and Metu, respectively. The highest mean values observed for HBW at Metu was attributed to the favorable environmental factor (climatic condition) mentioned earlier. On the other hand, the lowest mean

value of the genotypes for HBW at Agaro might be attributed to lack of adequate moisture in May which was critical for bean filling at the particular location as there was no rain in the same month at Jimma. Similar justification was reported by Tesfaye et al. (2013a, b) that the amount of moisture available during the critical period of fruit growth has significant influence on physical quality of coffee beans. The existence of genotypes combining large bean size along with high yield potential and fine cup taste is preferable as it fulfills both productivity and all aspects of qualities. Coffee beans with larger sizes usually achieve higher grading and fetch higher price

Table 4. Parchment and beans lengths of 30 Arabica coffee genotypes at four different locations during 2009/2010.

Genotype	Parchment length (cm)					Bean length (cm)				
	Jimma	Agaro	Gera	Metu	Combined mean	Jimma	Agaro	Gera	Metu	Combined mean
74191	1.095	1.064	1.159	1.191	1.127	0.816	0.782	1.000	0.898	0.874
75187B	1.241	1.127	1.215	1.207	1.198	1.013	0.834	0.947	0.976	0.943
7453	1.075	1.142	1.149	1.137	1.126	0.884	0.915	0.933	0.951	0.921
74145	1.133	1.033	1.097	1.122	1.096	0.872	0.771	0.909	0.886	0.860
75194	1.099	0.987	1.135	1.103	1.081	0.858	0.751	0.855	0.915	0.845
7512	1.067	1.036	1.140	1.180	1.106	0.869	0.743	0.932	0.924	0.867
7574	1.034	1.107	1.285	1.254	1.170	0.814	0.859	0.915	0.999	0.897
7803A	1.203	1.151	1.340	1.261	1.239	0.955	0.871	1.095	1.025	0.986
7803B	1.145	1.251	1.285	1.228	1.227	0.955	0.865	0.986	0.968	0.943
7809B	1.147	1.271	1.324	1.233	1.244	0.922	1.015	1.057	0.954	0.987
802	0.991	1.091	1.064	1.147	1.073	0.768	0.853	0.889	0.943	0.863
804	1.037	1.161	1.216	1.131	1.136	0.801	0.907	1.009	0.894	0.903
808	1.097	1.007	1.192	1.028	1.081	0.921	0.739	0.916	0.823	0.850
809	1.171	1.160	1.279	1.322	1.233	0.901	0.891	0.985	1.023	0.950
8010	1.063	1.113	1.214	1.131	1.130	0.796	0.877	0.989	0.963	0.906
8011	1.081	1.109	1.152	1.153	1.124	0.807	0.771	0.894	0.883	0.839
8017	1.142	1.099	1.115	1.145	1.125	0.895	0.835	0.949	0.875	0.889
8019	1.094	1.213	1.186	1.195	1.172	0.792	0.936	0.929	0.992	0.912
8021	1.087	1.106	1.163	1.193	1.137	0.870	0.839	0.889	0.948	0.887
8112	1.123	1.170	1.277	1.191	1.190	0.863	0.835	1.031	0.877	0.901
8133	0.941	1.027	1.129	1.113	1.053	0.831	0.851	0.940	0.890	0.878
8136	1.078	1.171	1.321	1.229	1.200	0.849	0.787	1.013	0.951	0.900
8143	1.159	1.268	1.401	1.102	1.232	0.982	0.980	1.123	1.039	1.031
8144	1.082	1.152	1.141	1.109	1.121	0.913	0.938	0.945	0.949	0.936
827	1.197	1.126	1.218	1.153	1.174	0.845	0.836	1.031	0.966	0.919
828	1.111	1.169	1.181	1.353	1.204	0.867	0.885	0.935	0.998	0.921
8211	1.269	1.289	1.264	1.261	1.271	0.964	0.949	1.060	0.992	0.991
8213	1.327	1.168	1.313	1.306	1.279	1.015	0.902	1.101	1.037	1.014
8219	1.195	1.189	1.283	1.210	1.219	0.949	0.953	1.004	0.899	0.951
8223	1.025	1.115	1.137	1.113	1.098	0.851	0.937	0.897	0.946	0.908
Mean	1.117	1.136	1.213	1.183	1.162	0.881	0.864	0.972	0.946	0.916
CV	4.442	4.296	4.102	3.258	2.45	4.395	2.843	4.07	3.927	3.45
LSD _{0.05}	0.050	0.05	0.050	0.039	0.048	0.04	0.025	0.04	0.038	0.053
LSD _{0.01}	0.068	0.067	0.068	0.053	0.064	0.053	0.034	0.054	0.051	0.072

LSD, Least significant differences.

than smaller ones. However, currently, high demand and premium prices are ensured for those coffee types which combine high bean and cup qualities.

The parchment that grows inside the exocarp determines the ultimate bean sizes: bean weight as well as length. It is in turn determined by the size of the exocarp. The overall range among genotypes and locations for PL was 1.053 to 1.279 cm and 1.117 to 1.213 cm, respectively (Table 4). The highest was observed at Gera followed by Metu and Agaro, respectively and the least was observed at Jimma. However, HBWs and BLs at the distinct locations were

not in the same order with their corresponding PLs noticed at the respective locations. The absence of correlation between PL and BL or HBW at the different locations could be attributed to absence of correspondence in the amount of moisture received during berry expansion and bean filling stages at the distinct locations as the latter two processes take place at different times and this illustrates that larger berry volume does not necessarily ensure larger bean size (Figure 1).

The range among genotypes for outturn was 13.22 to 15.73% (Table 5). The range across the distinct locations for outturn was 13.32 to 16.23% the least and the highest

Table 5. Outturn and floater bean percent of 30 Arabica coffee genotypes at four different locations during 2009/2010.

Genotype	Outturn (%)					Floater bean (%)				
	Jimma	Agaro	Gera	Metu	Combined mean	Jimma	Agaro	Gera	Metu	Combined mean
74191	11.51	15.77	18.54	17.11	15.73	11.44	11.18	0.65	1.02	6.07
75187B	10.14	16.39	16.10	14.13	14.19	9.02	12.96	3.47	11.05	9.13
7453	12.97	15.79	16.42	13.70	14.72	4.38	21.42	2.21	1.42	7.36
74145	12.56	13.92	16.68	16.12	14.82	2.51	23.92	1.87	0.23	7.13
75194	13.08	14.33	16.56	14.34	14.58	4.08	27.69	2.81	2.71	9.32
7512	14.06	14.31	17.10	17.19	15.66	5.55	18.17	1.92	0.90	6.64
7574	13.51	13.05	16.82	12.47	13.96	14.23	38.10	4.03	3.07	14.86
7803A	13.70	14.21	15.46	16.46	14.96	17.44	57.52	13.72	10.67	24.84
7803B	13.71	16.26	17.15	12.76	14.97	5.05	15.22	1.87	2.80	6.23
7809B	14.61	14.50	17.66	13.07	14.96	2.28	21.32	2.57	3.52	7.42
802	13.74	11.46	17.04	14.32	14.14	7.71	25.71	4.21	3.23	10.22
804	14.41	14.47	15.45	14.12	14.61	3.09	14.73	3.03	1.67	5.63
808	13.51	14.25	14.07	14.73	14.14	1.70	10.46	0.77	0.20	3.28
809	13.34	14.18	17.03	14.12	14.67	3.06	35.02	1.38	9.14	12.15
8010	16.55	12.43	17.34	16.07	15.60	1.70	12.26	2.34	0.67	4.24
8011	12.56	13.56	15.79	16.37	14.57	3.77	20.60	2.07	1.25	6.92
8017	15.16	12.18	15.39	14.39	14.28	2.03	21.19	1.44	3.89	7.14
8019	12.44	12.43	15.38	12.93	13.30	3.52	17.95	3.35	1.04	6.46
8021	14.21	13.27	14.66	14.93	14.27	1.96	40.54	1.46	0.71	11.17
8112	13.17	14.01	17.26	13.53	14.49	6.85	30.74	4.10	1.69	10.85
8133	13.53	12.97	13.70	13.42	13.41	3.52	27.81	6.73	4.18	10.56
8136	11.33	14.13	17.22	13.73	14.10	1.82	15.00	3.51	2.30	5.66
8143	12.27	12.77	14.97	12.88	13.22	4.45	9.77	2.93	2.21	4.84
8144	13.48	13.31	15.22	16.82	14.71	4.54	18.47	2.73	5.41	7.79
827	13.54	13.06	16.73	14.33	14.42	2.64	34.66	2.39	5.39	11.27
828	11.94	12.81	18.00	13.78	14.13	5.24	37.42	3.22	1.20	11.77
8211	11.45	14.63	16.55	12.98	13.90	2.51	41.13	2.97	1.40	12.00
8213	14.46	13.24	16.64	12.61	14.24	1.70	51.27	2.71	3.43	14.78
8219	14.33	14.69	14.83	13.27	14.28	2.66	29.59	3.51	2.80	9.64
8223	14.35	12.11	15.18	15.98	14.40	1.27	40.32	5.81	4.13	12.88
Mean	13.32	13.82	16.23	14.42	14.45	4.72	26.07	3.19	3.11	9.27
CV	5.52	3.9	3.59	4.15	4.29	9.42	4.57	14.53	13.02	7.62
LSD _{0.05}	0.74	0.54	0.59	0.61	1.01	0.45	1.2	0.47	0.41	1.18
LSD _{0.01}	1	0.74	0.8	0.82	1.36	0.61	1.62	0.63	0.55	1.6

LSD, Least significant differences.

being observed at Jimma and Gera, respectively. Genotypes 8010, 7512 and 74191 exhibited the three top outturn percent as shown in the work. However, these genotypes were not among the top for overall yield (Yonas and Bayetta, 2008), HBW or BL. On the other hand, genotypes: 8143, 8019 and 8133 which were top for overall yield, HBW or BL exhibited the three least values for outturn. This shows that genotypes with higher CW or HBW do not necessarily exhibit higher outturn.

The overall range among genotypes for FBP was 3.28 to 24.84% (Table 5). The range across the distinct locations was 3.11 to 26.07%, the highest and the lowest

being observed at Agaro and Metu, respectively. The floater bean percent of genotypes at Agaro was much higher and it ranged from 9.77 to 57.52%. Three genotypes: 808 (3.28), 8010 (4.24), and 8143 (4.84) however exhibited the three least floater percent. The higher floater beans noticed at Agaro compared to the other locations is attributed to the fact that high proportion of the parchments was not filled by an endosperm during the critical stage of grain filling for reason mentioned earlier. However, irrespective of the prevailing environmental conditions genotype 8143 consistently exhibited minimum floaters in addition to its relative

Table 6. Mean squares of combined analysis of variance for ten cherries weight, hundred beans weight, beans length, parchment length, outturn and floater beans %.

Parameter	Environments (E)	Genotypes (G)	GXE	Pooled error
Traits (DF)	3	29	87	232
Ten Cherries weight	469.370**L	35.485**L	6.725**	0.681
Hundred beans weight	316.176**L	28.724**L	4.552**	0.333
Bean length	0.239**L	0.030**L	0.007**	0.001
Parchment length	0.172**L	0.048**L	0.009**	0.000
Outturn	145.559**L	4.423**	5.141**	0.383
Floater percent	11335.387**L	213.910**L	111.614**	0.000

**L and ** significant against mean square of G x E and mean square of error at 0.01 probability level, respectively.

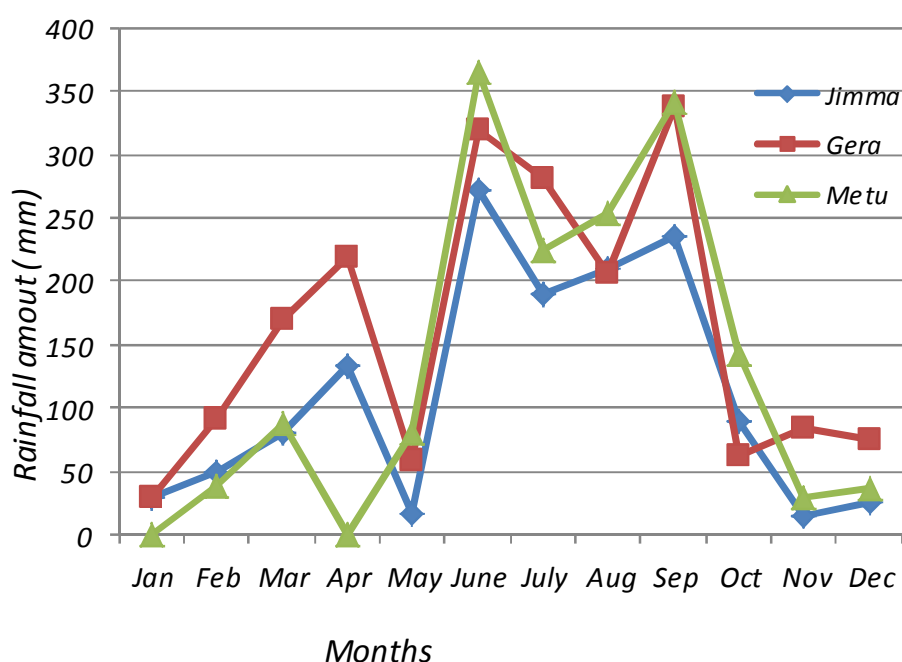


Figure 1. The rainfall amount and distributions received during the twelve months at Jimma, Gera and Metu in 2009/10

higher overall yield potential. On the other hand, genotype 8213, the highest yielder from among the genotypes (Yonas and Bayetta, 2008), exhibited relatively higher floater beans.

Generally, the genotypes exhibited higher values for TCW, HBW, outturn, BL and PL at Gera and Metu than Agaro and Jimma. This is attributed mainly to differences of climatic elements prevailed at the different locations during the bean growth periods. The endosperm development of beans at Gera and Agaro was traced back to have taken place in May and June as it usually occurs 2 months in advance before maturity (harvest) stage. During this month, the rainfall received at Jimma, which is adjacent to Agaro, was low (Figure 1). The same dry spell might have encountered at the latter as the

mean values observed for bean weights or BLs at this particular location were the least. The berry expansion and bean filling at Gera and Metu coincided from March to June and June to September, respectively as traced back from their harvest/maturity time. During these months the rainfall received at the latter was more optimal than the latter (Figure 1) and this might have favored the development of larger beans. The optimum moisture received during berry expansion and bean filling from June to September favored better bean growth at Jimma than Agaro however the outturn observed at the former was lower than the latter. The reason is attributed largely to high flower abortion resulted from the high temperature and little moisture received in May (Figure 1) which subsequently affected the outturn. The case at Agaro was

clear that part of the bean filling coincided in May and during this month the rain received at Jimma, which is adjacent to it, was little (17 mm) and similar conditions might be true at the former to affect bean growth adversely. Higher floater beans were noticed at Agaro than the other locations (Table 4). This was largely attributed to favorable environments (nutrients or/and moisture) that had favored luxurious berry expansion and parchment growth, while the shortage of rain immediately after the expansion in May might have induced incomplete bean filling ultimately resulting in higher percentage of floater beans. In conformity to the result of the present study, Cavaco Bicho et al. (2010) also stated that moisture amount available during fruit growths affect berry and bean growth. The author being impressed by the higher floater beans observed at Agaro at the first picking took another sample 45 days later from the same site. The results of the second evaluation however revealed that the floater percentage in this case was significantly reduced. The outturn percentage was also increased significantly compared to the first sample suggesting the fact that it is the moisture during critical stages which determines degree of bean filling. Nevertheless, the BL and HBW did not exhibit significant changes relative to the first picking as the same dry spell in May has rather considerably restricted berry and parchment expansions.

Conclusion

Both the chemical and physical attributes of coffee beans are important criteria which determine value of Arabica coffee beans in world market. The Ethiopian coffees are among the preferred coffee types in cup taste. But higher prices are paid to those coffees which fulfill both criteria. In this regard three genotypes: 7803A, 8143, and 8213 exhibited higher mean values for bean size in addition to having higher cup taste and overall yield potential and therefore could be recommended for commercial production.

Genotypes having higher bean sizes did not exhibit higher outturn. This may imply that the proportion of pulp percent could be higher than expected for those genotypes with higher CW. Genotypes also exhibited higher outturn at higher altitude areas which have adequate moisture and fair distribution than mid altitude areas with less moisture and erratic distribution. This illustrates the need to apply intensive agronomic practices such as irrigation, pruning, mulching and maintenance of shade at optimum level at the latter to alleviate the problem.

Genotypes exhibited higher FBP and lower outturn at Jimma and Agaro than Gera and Metu. On the other hand, higher outturn and lower floater beans were observed at latter than the former. This is attributed to the fact that the moisture received at the former was more

optimal than the latter during the critical time of bean growth. Therefore, coffee orchards which grow at moisture stress areas should be grown with moderate shades or adequate mulch to retain the available moisture in the soil for longer periods.

Conflict of Interests

The author(s) have not declared any conflict of interests.

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Full Length Research Paper

Crop nutrition studies on grain filling and chalkiness in rice

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Rice grain chalkiness can reduce crop quality resulting in lower grades and returns. It is as a consequence of loose starch and protein particles packing during grain filling and maturation, and may be due to nutritional, biochemical, physiological and environmental conditions particularly in varieties with high heritable chalkiness. This study set out to evaluate the response to varying levels of nitrogen and potassium [N:K] fertilizer combinations and its time of application [TOA] on the partitioning of assimilates and their effect on grain filling and chalkiness in a commercial rice production system. The results indicate that while the N level increased within a particular K level, the crop maintained a similar rate of photosynthesis and dry matter production. The K which is involved in active translocation manifested its effect later in the partitioning after 14WAS, and during grain filling. At the N:K ratio of 2:1, the % chalkiness was at an acceptable level (<2%), and the plant tissue analysis N:K was close to 1:1. Chalkiness is an agronomic problem that is best managed in susceptible varieties through adequate crop nutrition using the simple N:K ratio. The study confirmed that when N:K was applied at a 2:1 ratio, grain yield was the highest and chalkiness at its lowest

Key words: Grain filling, chalkiness, net assimilation rate, partitioning of assimilates, potassium fertilizer.

INTRODUCTION

Grain chalkiness in rice is a highly undesirable industrial quality trait in the marketing and consumption, and influences the commercial value. Chalky kernels range from 1 to 15% [U.S. No. 1 to 6, respectively] for long rice grains (USDA, 2005). These grains break more readily than translucent ones (Bridgemohan, 1997), thus reducing whole grain head rice yields (HRY). Currently, world markets do not accept any rice which has more than 2% chalky rice.

Chalkiness is characterized by opaque areas in various parts of the rice kernel which occurs when starch and protein particles become loosely packed or incompletely filled within the endosperm during grain maturation (Ebron, 2013). This may be influenced by a combination of factors including, physical, biochemical, physiological, water quality and availability (Chen et al., 2012), temperature (Shen et al., 1997), crop nutrition (Mingli and Yonggui, 2005; Sun et al., 2014), cultivar genetics

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(Yamakawa et al., 2007) and production environment and cropping systems (Carlos et al., 2008).

The molecular basis of this trait is poorly understood but the quantitative trait locus (QTL) is identified as *Chalk*. This encodes a vacuolar H⁺-translocating pyrophosphatase (V-PPase) with inorganic pyrophosphate (PP_i) hydrolysis and H⁺-translocation activity and has been shown to influence grain chalkiness, which also affects HRY (Yibo et al., 2014).

Ishimaru et al. (2009) reported that high-temperature stress during grain ripening causes induces loose packing of the amyloplasts. Shen et al., (1997) found that with high temperature treatment, peaks of materials synthesis and enzyme activity in endosperm were earlier than those of proper temperature treatment. This affected the sequence and pattern of changes of materials and enzymes in endosperm during normal grain filling, and further intensified chalkiness. Molecular mapping (Chandusingh et al., 2013) and magnetic resonance images (Ishimaru et al., 2009) of the early stage caryopses in the high-temperature condition confirmed loose packing of amyloplasts in the chalky part of the grain. Rice breeding programs usually discard chalkiness lines, but it can still manifest over time in some environments.

Zhou et al. (2009) reported that chalkiness is related to "source-sink" of rice, dynamics of grain filling, and biosynthesis and accumulation of starch in endosperm. It is a complicated quantitative trait, which is controlled by maternal, endosperm and cytoplasmic effects. They noted that there are some stable QTL for rice chalkiness on many rice chromosomes, which have an impact on starch synthesis, starch metabolism, and fruit development. However, the manipulative network and formative mechanisms of rice chalkiness remain unclear, and the decrease of rice chalkiness is a major aim in rice quality breeding.

Sun et al. (2014) showed that different water-nitrogen management patterns and P:K fertilizer combined application significantly affect rice quality, nutrient absorption and distribution and grain HRY. They found that amylose and protein contents under different water - nitrogen management patterns are higher than those of different P-K fertilizer combined applications, while the % chalkiness and gel consistencies are the opposite.

Motoki et al. (2013) confirmed that chalky grains are caused by high temperature stress at the ripening stage. Chikako et al. (2013) showed that deep-flood treatment enhanced N uptake, and consequently photosynthetic activity, resulting in the reduction of chalky grain formation. Shenggang et al. (2013) surmised that there is a role for plant growth regulators in grain yield, quality and antioxidant enzyme activities, but little is known about.

This study set out to evaluate the response to varying levels of nitrogen and potassium [N:K] fertilizer combinations and its time of application (TOA) on the

partitioning of assimilates and their effect on grain filling and chalkiness in a commercial rice production system.

MATERIALS AND METHODS

Two commercial field studies were conducted the Caroni Rice Project [CRP] and one pot study at the Centre Bioscience, Agriculture and Food Technology, Waterloo Research Campus, University of Trinidad and Tobago.

The soil type was the Frederick soil series loam with an average CEC of 4.8 meq/100 g; a pH (water soil solution) of 5.25; N at 0.313 % ; P of 7.5 to 11.9 mg.kg⁻¹ ; K at 124 to 160 mg.kg⁻¹, and a sand, silt, and clay content of 61.0, 14.5, and 24.5%, respectively, in the upper 0.5 m soil. The pot trials were established using the same soil type as that of the commercial fields. For this purpose, the soil was pulverized and sieved to remove all debris, and solarized to kill weed seeds.

The seed material used was var. Oryzica 1 which is the major commercial variety. This is a high yielding variety and is considered to have low percentage chalkiness. However, over the last six years, this has varied from 2.5 to 20% depending on the field conditions, time of the year, rainfall patterns, and the timeliness of the fertilizer application. The seeds were broadcasted by aerial application at seeding rate of 50 kg seed.ha⁻¹. The crop was cultivated under rain fed conditions of the wet season (June to October) and there was no need for supplemental irrigation.

Aerial application for the seed, fertilizer and pesticides were done using a Cessna AJ Truck A188B aircraft which is calibrated to deliver the appropriate rates needed for the study. All applications were done during the hours of 6.30 to 8.30 am when the wind speed was less than 10 knots h⁻¹ flying at 90 knots h⁻¹ at 8 to 10 m above ground level, and average daily temperatures between 22 to 25°C.

The crop was subjected to all the standard agronomic and estate management practices for commercial fields with respect to land preparation, sowing, water management and weed control. All treatments received a standard dressing of phosphorous (P₂O₅) at a rate of 58 kg.ha⁻¹ as Triple Super Phosphate (TSP) at the time of the first nitrogen application. The normal agronomic practices of insect control [cypermethrin 225 CE (1/ha) and fipronil (1/ha)] and diseases control [Difenoconazole] were conducted. Weed control was done using propanil at 3.0/ha⁻¹ at 2 weeks after sowing (WAS) and a mixture of Oxadiazon (1.25 ka. ai.ha⁻¹) and Bromoxyl + 2,4D ester (1.0 ai./ha⁻¹) at 7 WAS. The field crop was harvested by combine harvester and plot yield and HRY recorded.

Study 1

This field experiment was established at CRP on the entire field of Nos. 003 and 005 which were 12 ha each. The fertilizer treatments were Nitrogen at 2 rates (75 and 90 kg.ha⁻¹ (applied as split TOA at 4.5 and 9.0 weeks after sowing (WAS) and Potassium at 3 rates (0, 50, and 90 kg.ha⁻¹) at the similar TOA (Table 1). The trial was laid out as a split plot, with nitrogen as the main plots and potassium in the sub-plots, with time of application randomized within sub-plots and replicated three times. The experimental plots were 12 m in width by 500 m in length and were normally manually harvested, whilst the remainder of the field was mechanized.

Study 2

This field experiment was conducted the next year following study 1 at CRP in field 205, with experimental plots sizes of 10 m × 50 m. It was laid out according to split plot design with 4 main plots and 3

Table 1. Treatment combinations of the N:K fertilizer [kg.ha⁻¹].

Time of application [TOA] WAS	Potassium [K] rates kg.ha ⁻¹	Nitrogen [N] rates kg.ha ⁻¹
0	K ₀ (0)	N ₁ (75)
4.5	K ₁ (50) K ₂ (50)	N ₂ (90)
9.0	K ₃ (90) K ₄ (90)	

Table 2. Treatment combinations of the N:K fertilizer [kg.ha⁻¹].

Potassium [K] rates kg.ha ⁻¹ at TOA (6 and 9 WAS)	Nitrogen (N) rates kg.ha ⁻¹ at TOA (3, 6 and 9 WAS)
K ₁ (45)	N ₁ (30)
K ₂ (60)	N ₂ (45)
K ₃ (90)	N ₃ (60)
	N ₄ (90)

subplots, with 3 replicates. The N:K treatments were Main Plot: N (30, 45, 60 and 90 kg N.ha⁻¹) in split TOA (3,6,9 WAS) and Subplot: Muriate of Potash (45, 60, and 90 kg K₂O.ha⁻¹) which was applied only at 6 and 9 WAS in a split TOA (Table 2).

Study 3

The pot experiment was conducted under full sunlight conditions and established in plastic drums filled with 50 L of the solarized soil. The N:K fertilizer treatments were nitrogen at 4 rates [50, 75 100 and 125 kg.ha⁻¹] and potassium fixed at 45 kg.ha⁻¹ using similar TOA as Study 2. The pots were placed 30 cm × 30 cm apart and water level kept flooded above field capacity during growth and grain filling period. Leaf growth and plant dry matter production were monitored at 10 days intervals until early panicle emergence.

In all studies, leaf tissue and soil samples were analyzed to determine the levels of N, P and K. At the times of aerial application of fertilizer, containers (30 × 45 cm) were randomly distributed throughout the experiment field to monitor the aerial discharge and quantity of fertilizer reaching the crop. Grain yield for each treatment was computed from five 10m² quadrant. The data collected included tillering ability, filled spikelets, yield components and grain yield. The N was determined by means of the of a Coleman Junior 11A Linear Absorbance Spectrophotometer and the K by Flame Photometry using a Corning Eel Flame Photometer.

Plants were harvested for biomass measurements starting from 20 days after emergence (DAE) at intervals of 10 days until harvest. At each harvest, plants were separated into leaf laminae, petioles, tillers, and roots and dried in a forced-air oven at 65°C for 72 h (to constant weight) for dry matter determination. Best fit polynomial regression curves were used for growth analysis between two successive harvest periods [HP]. Relative growth rate (RGR), net assimilation rate (NAR), specific leaf area (SLA), leaf Area (LA), leaf area ratio (LAR), leaf weight fraction (LWF), shoot dry matter as described by Hunt et al. (2002). Kernel chalkiness determined by visual rating of the chalky proportion of the grain and is used to measure chalkiness based on the standard Evaluation System SES scale presented below: Select, segregate and weigh the chalky

grains (SES Scale 9). Determine the % chalky grain using the equation:

Scale	% area of chalkiness
1	less than 10
5	10 - 20
9	more than 20

$$\% \text{ Chalky grain} = \frac{\text{Weight of chalky grains}}{\text{Weight of milled rice}} \times 100$$

Air temperature, rainfall, and solar radiation during the experiments were calculated with an automated weather station (Davis Instruments, California) located within the station compound and at close proximity to commercial fields. Light interception was determined during the morning (10.00 to 11.00 am) by measuring the radiation above the canopy, and at soil level using an integrated quantum sensor (Model LJ – 185B Quantum Radiometer/Photometer, LICOR, Inc, Lincoln, NE 68504). Leaf area was measured using an area meter (Type AAM – 5 Hayashi Denko Co. Ltd. Japan). All data were analyses as multiple regression analysis using Minitab Statistical package.

RESULTS AND DISCUSSION

Study 1

The results (Table 1) indicated that the varying rates and time of application of the N:K combinations had no significant effect on final plant height at harvest (82.1 ± 0.823 cm), and tiller number (240.4 ± 7.66%). There appeared to be no trend on the productive particle (%). In all cases, regardless of the N:K combination, the % chalkiness was in excess of the acceptable industrial standard (Table 1). At the lower nitrogen N₁ rate (75 kgN.ha⁻¹), it was observed that by increasing the potassium

Table 3. The response of rice variety Oryzica 1 to varying rates and time of application of nitrogen and potassium fertilizer.

Treatment combination	Plant height	Tiller Nos. (Nos. on ⁻²) (cm)	% Productive panicle	Chalkiness (%)	Yield (t.ha ⁻¹)
N ₁ K ₀	84	280	86	11.8	2.6
N ₁ K ₁	83	208	95	13.0	2.6
N ₁ K ₂	79	247	93	15.4	3.1
N ₁ K ₃	84	256	91	10.2	3.7
N ₁ K ₄	81	226	94	10.6	2.4
N ₂ K ₀	84	254	90	15.6	2.8
N ₂ K ₁	84	251	88	13.6	2.7
N ₂ K ₂	85	205	87	17.0	2.4
N ₂ K ₃	78	256	93	15.0	2.3
N ₂ K ₄	79	221	89	9.0	2.1
Mean	82.1	240.4	90.6	13.1	2.6
S.E [±]	0.82	7.66	0.98	0.85	0.15

Table 4. The effect of various combinations of nitrogen and potassium fertilizer on the development and yield of rice variety Oryzica 1 1996.

Treatment combination	Tiller Nos. (Nos. on ⁻²) (cm)	% Productive panicle	Chalkiness (%)
N ₁ K ₁	212	100	3.3
N ₂ K ₁	216	100	5.3
N ₃ K ₁	304	96	5.0
N ₄ K ₁	278	96	3.0
N ₁ K ₂	275	100	4.0
N ₂ K ₂	333	100	5.3
N ₃ K ₂	414	99	3.7
N ₄ K ₂	293	100	4.0
N ₁ K ₃	348	100	2.0
N ₂ K ₃	440	100	4.3
N ₃ K ₃	491	96	3.3
N ₄ K ₃	436	100	6.7
Mean	336.7	98.9	4.13
S.E[±]	26.2	0.514	0.36

rate, the rice grain yield increased linearly (Equation 1).

$$Y_{\text{Grain yield}} = 2.05 + 0.38 \text{ N:K } R^2 = 0.88 \quad (1)$$

However, an opposite trend was observed at the higher N₂ rate (90 kg N. ha⁻¹), as the K₂O rate increased, the grain yield decreased linearly (Equation 2).

$$Y_{\text{Grain yield}} = 2.85 - 0.19 \text{ N:K } R^2 = 0.96 \quad (2)$$

It was difficult to partition between the time of application between 4.5 and 9.0 WAS, compared to rates. However, it appeared that N₁K₃ (75 kg N + 90 kg K₂O at 4.5 WAS) produced the highest yield and was above the normal

yield of 3.5 t. ha⁻¹ (Table 3).

Study 2

The key difference between experiments 2 and 1 was that the nitrogen fertilizer was applied at 4 rates in three split applications, and the K₂O at three rates, and split into two TOA. It is to note that the first N application was at 3 WAS and applied single with no K₂O₅ at this time. The results showed that increasing the N rates (Table 4 and 5) at 45 kg K₂O ha⁻¹ increased grain yield linearly (Equation 3)

$$Y_{\text{Grain yield}} = 1.9 + 0.4 \text{ N:K } R^2 = 0.83 \quad (3)$$

Table 5. The effect of various combinations of Nitrogen and Potassium fertilizer on the development and yield of rice variety Oryzica 1 1996.

Nitrogen kg ha ⁻¹	Rates of fertilizer		
	Potassium [K ₂ O] kg ha ⁻¹		
	45	60	90
	Rice grain yield (t.ha ⁻¹)		
30	2.5	3.4	2.5
45	2.5	3.2	3.0
60	2.9	2.9	3.2
90	3.7	2.4	3.0
Equations	3	4	

Table 6. Leaf tissue and soil analysis of the of the varying N:K treatment combinations at 9 WAS.

N*K fertilizer rates	NPK content (%)					
	Leaf tissue			Soil analysis		
	N	P	K	P	K	pH
N ₁ K ₁	3.27	0.30	2.31	10.00	129.33	5.20
N ₂ K ₁	3.02	0.31	2.31	10.67	140.00	5.18
N ₃ K ₁	3.17	0.28	2.19	9.67	134.67	5.25
N ₄ K ₁	2.93	0.31	2.15	8.67	143.33	5.32
N ₁ K ₂	3.15	0.31	2.25	10.00	129.33	5.15
N ₂ K ₂	2.72	0.29	2.21	8.33	128.00	5.17
N ₃ K ₂	3.22	0.29	2.20	9.33	177.33	5.33
N ₄ K ₂	3.45	0.27	2.15	10.00	120.00	5.22
N ₁ K ₃	2.83	0.29	2.05	9.33	117.33	5.17
N ₂ K ₃	2.87	0.25	2.19	9.00	125.33	5.23
N ₃ K ₃	3.23	0.31	2.49	10.67	144.00	5.20
N ₄ K ₃	2.90	0.30	2.13	9.00	152.00	5.18

However, the opposite occurred when the rate increased to 60 kg (K₂O ha⁻¹) and it decreased linearly (Equation 4).

$$Y_{\text{Grain yield}} = 3.8 - 0.33 N^*K \quad R^2 = 95.9\% \quad (4)$$

Grain yield peaked at the highest nitrogen rate (90 kg N₂ ha⁻¹) and lowest potassium (45 kg K₂O. ha⁻¹) producing 3.7 t. ha⁻¹ of quality rice grains with an acceptable chalkiness of 2.0% at the split fertilizer TOA at 6 and 9 WAS (Tables 4 and 5). This compared to the highest grain yield in Trial 1 which had a similar yield (3.7 t. ha⁻¹) at 70 kg N. ha⁻¹ plus 50 kg (K₂O) at a single application [4.5 WAS], but a chalkiness of 10.2%.

The difference in both trials attaining at high of 3.7 t. ha⁻¹ was that the treatment with the lowest chalkiness had both nitrogen and potassium as split applications. That is, a single application of nitrogen was made at 3 WAS, and the other 2 application of N coincided with the potassium application at later dates of 6 and 9 WAS, respectively. The results suggested that splitting the nitrogen

application at equal intervals TOA (3, 6, 9 WAS) , and using a lower rate in a split application (6 and 9 WAS) produced the highest yield, and the lowest chalkiness. Further, the crop was making better use of the Nitrogen prior to panicle initiation at 9 WAS. At that treatment (Table 6), the tissue analysis, indicated that the % NPK was adequate (Table 7), and the crop was making better uptake and use of the soil Phosphorous.

Study 3

This pot trial was established to validate the TOA of the N:K combinations in experiments 1 and 2, and determine the influence on the sink-source relationship, and the subsequent partitioning of assimilates and grain filling and chalkiness in rice.

The partitioning of assimilates analyzed through growth analysis revealed that the varying N:K combinations treatment had no effect over the various harvest period

Table 7. Levels of nutrient adequacy from tissue analysis for rice variety Oryzica 1 (Shand, 1997).

Nutrient	Levels of adequacy		
	Deficient	Critical	Adequate
N	<2.9	2.9	>3.0
P	<0.12	0.13 to 0.21	>0.22
K	<0.84	0.85 to 1.96	>1.97

$$Y_{LA} = -863 + 51.4 HP - 0.167HP^2 : R^2 = 0.69$$

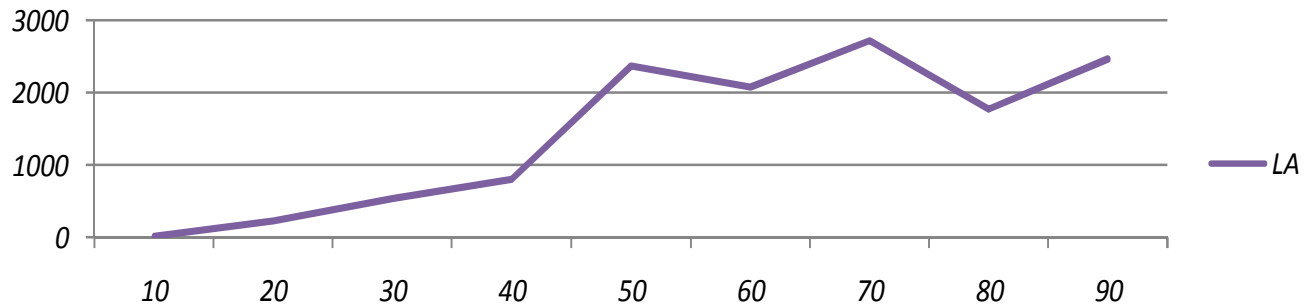


Figure 1. Leaf area (cm²) of rice (var. Orizica1) under varying levels and time of application of N:F fertilizer.

$$Y_{LAR} = 54.9 - 0.529 HP : R^2 = 0.64$$

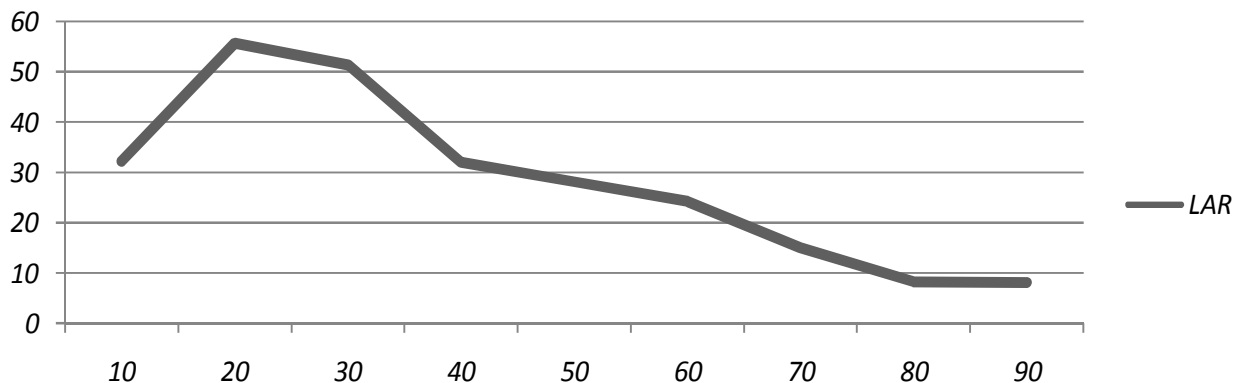


Figure 2. Leaf area ratio [LAR (g/cm²/day⁻¹)] of rice (var. Orizica1) under varying levels and time of application of N:F fertilizer.

[HP] until flowering at 95 DAS. It showed that the leaf area (Figure 1) and LAI (Figure 4) exhibited the usual quadratic response over HP. Similarly, the LAR (Figure 2) and LWF (Figure 3) declined linearly and were unaffected. The crop reached its maximum leaf area index [3.85] at 70 to 80 days after sowing. Both the RGR (Figure 5) and NAR (Figure 6) showed no treatment effect over the HP, and displayed quadratic responses.

Like the other two studies, the N*K combinations did not influence the number of panicles or the tiller number /stool, but the number of grains and grain yield increased linearly with increasing levels of nitrogen (Table 8). The acceptable level of chalkiness [2%] was attained at the 100 kg N : 45 kg K], and increased with lower or higher levels of N to the fixed K.

Whilst the N fertilizer level was increased at the fixed K

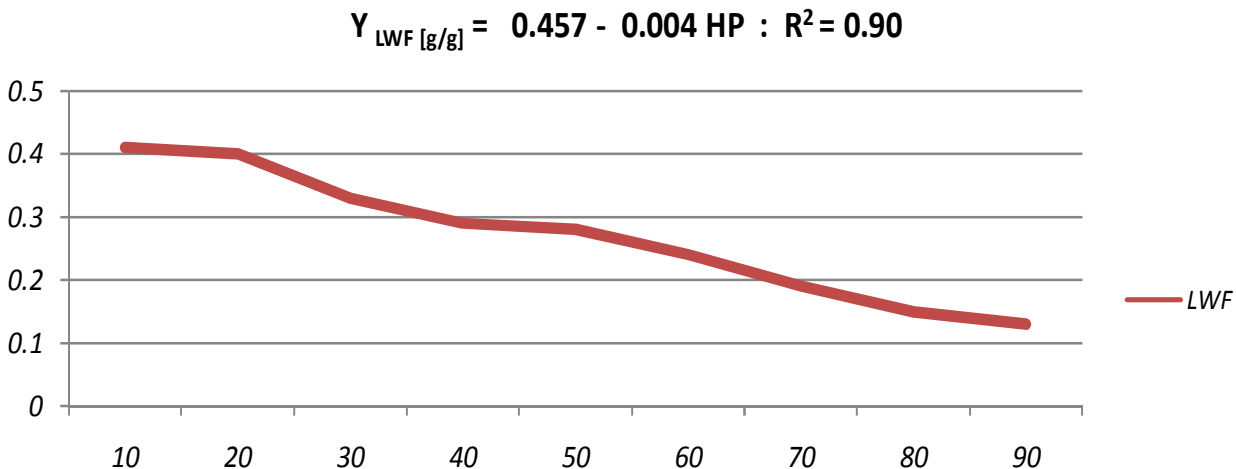


Figure 3. The Leaf Weight Fraction [LWF (g/g)] of rice (var. Orizica1) under varying levels and time of application of N:F fertilizer.

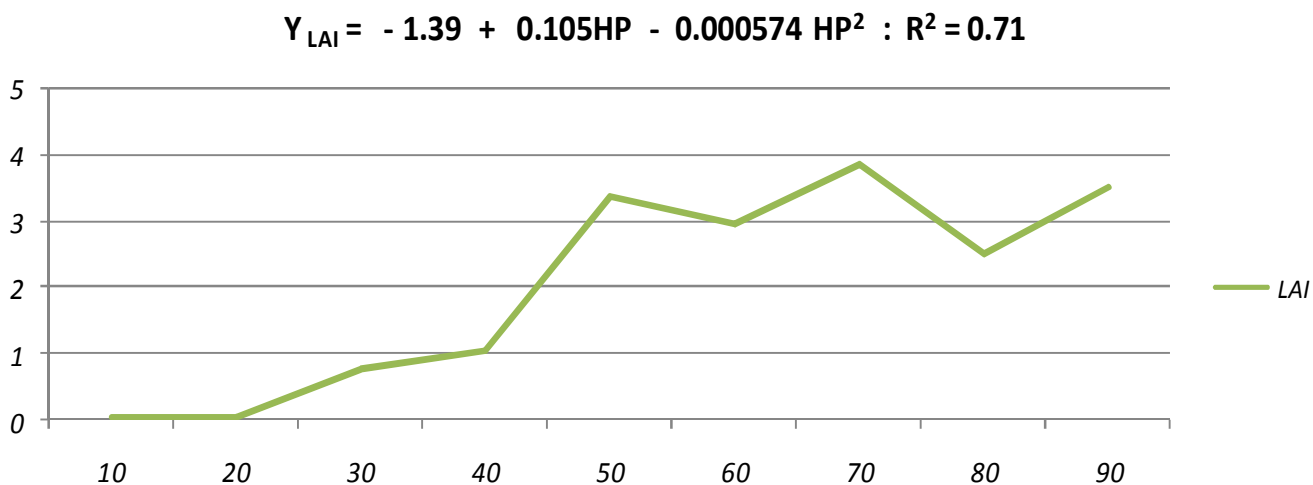


Figure 4. The leaf area index [LAI] of rice (var. Orizica1) under varying levels and time of application of N:F fertilizer.

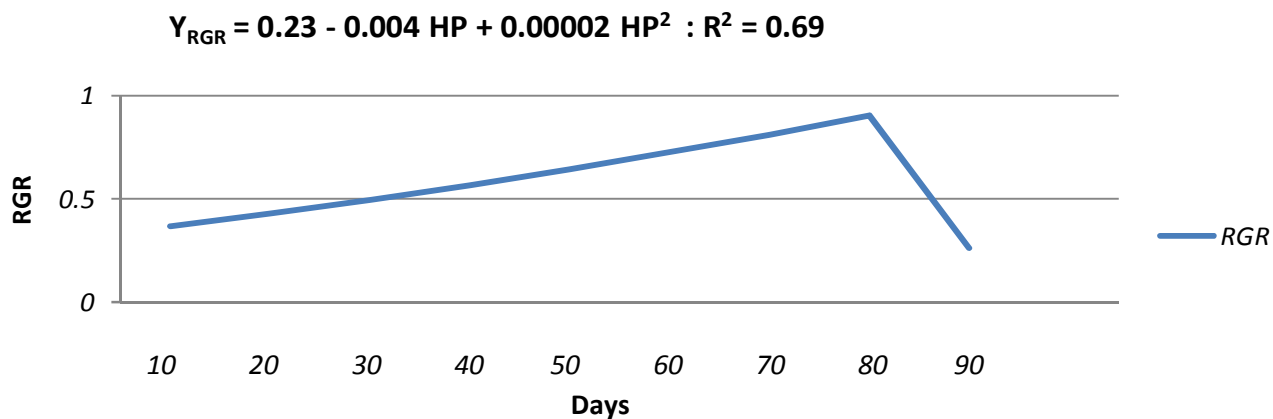


Figure 5. The Relative growth rate [RGR (g.m².day⁻¹)] of rice (var. Orizica1) under varying levels and time of application of N:F fertilizer.

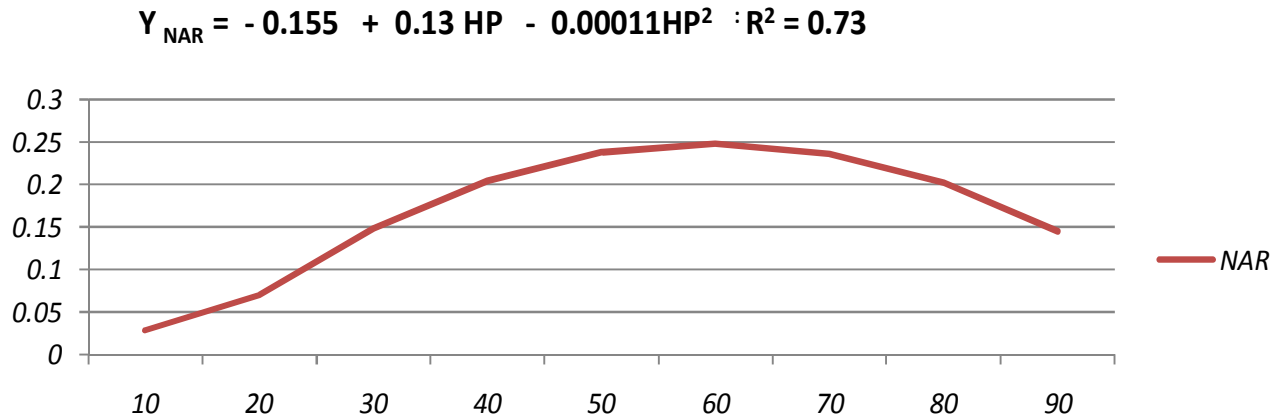


Figure 6. The net assimilation rate [NAR (g.m².day⁻¹)] of rice (var. Orizica1) under varying levels and time of application of N:F fertilizer.

Table 8. The effect of varying N:K treatment combinations on the reproductive capacity of rice.

Nitrogen kg ha ⁻¹	No. tillers / stool	Nos. panicle / stool	Chalkiness %	Number of grain/stool	Grain yield / stool
50	46	32	3	2872	78.1
75	43	37	4.7	2902	78.2
100	28	37	1.9	3056	83.1
125	42	36	6.1	3570	97.1
Mean (S.E)	42 (2.07)	34.1 (1.16)	-	3100 (185)	Y=37.5+6.12 N:K+21.0 HP; R ² = 88.6%

level, the crop maintained a similar rate of photosynthesis and dry matter production. The K which is involved in active translocation manifested its effect later in the partitioning of assimilates after 90 DAS, and during grain filling. N is significant in photosynthesis and production of assimilates, but K is the key element in determining the efficiency of the 'sink- source' relationship and the prevention of the loose packing of the starch and protein particles (Chikako et al., 2013).

At this period, the evidence showed that when the N:K ratio was 2:1, the % chalkiness was at an acceptable level (Bridgemohan, 1997). However, above the ratio, the number of grains appeared to increase (not significant), and the grain yield increased linearly (Table 6), but also the % chalkiness reached in excess of 6%. Mingli and Yonggui (2005) found that the grain yield in rice can be improved applying a urea solution during ripening period. However, while the amylose content was improved and gel consistency was extended, the chalkiness was increased.

Chalkiness is an agronomic problem that is best managed in susceptible varieties through adequate crop nutrition using the simple N:K ratio. The study confirmed that when N:K was applied at a 2:1 ratio, grain yield was the highest and chalkiness at its lowest, and that the plant

tissue analysis N: K was close to 1:1. Since most of the experiments were conducted in the wet season, it is suggested that the growing environment could have impacted in kernel chalkiness, which can be attributed to nighttime air temperature during grain filling. It is also accepted that there are some susceptible chalky cultivars, but the selected var. which is known to be of good quality has succumbed to variations in N:K nutrition in the wet season.

Conflict of Interest

The authors have not declared any conflict of interest.

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