Scientia Horticulturae 213 (2016) 287–293

Contents lists available at ScienceDirect

Scientia Horticulturae

journal homepage: www.elsevier.com/locate/scihorti

Genetic variation for vigour and yield of cocoa (*Theobroma cacao* L.) clones in Ghana

Atta Ofori*, Francis K. Padi, Frank O. Ansah, Abraham Akpertey, Gilbert J. Anim-Kwapong

Cocoa Research Institute of Ghana, PO Box 8, Akim Tafo, Ghana

ARTICLE INFO

Article history:
Received 5 August 2016
Received in revised form 1 November 2016
Accepted 4 November 2016
Available online 10 November 2016

Keywords:
Bean weight
Germplasm
Heritability
Pod value
Yield efficiency

ABSTRACT

The lack of agronomic information on germplasm clones has been suggested as one of the reasons for their poor utilization and management. Several cocoa genotypes from different genetic origin have been introduced into Ghana to broaden the crops genetic base. The present study investigates genetic variation and associations of vigour (estimated as stem diameter increments) and yield and its component traits (bean weight, number of beans per pod, pod value and yield efficiency) in 116 cocoa clones introduced into Ghana over different time periods. The clones were transplanted in June 2010, after eight-months of grafting, following a randomised complete block design with four replications consisting of four trees per clone per block. Stem diameter increments (SDI) in both juvenile and productive stage and yield component traits were evaluated between July 2010 and June 2015. Clone effects were highly significant ($P < 0.01$) for all the traits except SDI in the productive stage. Pod value and bean yield varied from 14 to 57 and 183 to 952 kg/ha, respectively. Heritability was generally low for all the traits and the highest observed (0.27 ± 0.06) was for bean weight. A positive genetic correlation ($r = 0.47$, $P \leq 0.001$) was observed between SDI in the juvenile stage and bean yield. Some of the best performing clones T65/238, ICS 40, T16/613, SGU 50 and T63/961 combined high yields with high bean weight and high yield efficiency. Results from the study indicate that there is considerable genetic variation for yield in the available germplasm clones, and yield increase could be achieved by developing hybrids from some of the best clones.

1. Introduction

Cocoa (*Theobroma cacao* L.) is a humid tropical tree crop grown in areas of high annual rainfall. Even though South America is the primary centre of diversity (Cheesman, 1944; Motamayor et al., 2002), cocoa cultivation has spread to most tropical regions and notably among them is the Caribbean, Southeast Asia and West Africa (Allen and Lass, 1983). It is cultivated for its fruit, from which the seed is used for the production of chocolates and confectionaries. Currently, about 70% of the world's cocoa production comes from West Africa, with Côte d'Ivoire and Ghana being the major producing countries (ICCO, 2013).

The *T. cacao* species, has traditionally been grouped into Trinitario, Criollo and Forastero, based on morpho-geographic classification (Cheesman, 1944), until recently when Motamayor et al. (2008) proposed 10 genetic groups. The ten groups are Marañon, Guiana, Contanama, Curaray, Nanay, Iquitos, Nacional, Purus, Criollo and Amelonado. Across years, a large number of genetically diverse accessions have been collected in the primary center of diversity and introduced into the various cocoa-growing countries through intermediate quarantine and genebank centers.

In Ghana, the first introductions occurred before 1938 and consisted mainly of the Amelonado and Trinitario genetic groups. They dominated plantations until the beginning of formal research when A. F. Posnette introduced the Trinidad clones (referred to as T) in 1943/44 (Lockwood and Gyamfi, 1979). Between 1946 and 1971, several clones belonging to the Criollo, Trinitario, Nanay, Iquitos, Nacional, Parinari (a sub-group of the Marañon) and Scavina (a sub-group of the Contanama) genetic groups were also introduced (Lockwood and Gyamfi, 1979). Later introductions occurred between 1972 and 2004 and largely comprised the Curaray, Purus and Guiana populations (Abdul-Karima et al., 2006). After 2004, clones from several genetic groups available from the International Cocoa Quarantine Centre, Reading were introduced annually and currently a little over 1000 different clones are available in the CRIG germplasm (Padi et al., 2015).

The evaluation of the cocoa clones in Ghana is very important for their efficient utilization and management. Currently, very few

* Corresponding author.
E-mail address: ofori77@yahoo.com (A. Ofori).

http://dx.doi.org/10.1016/j.scienta.2016.11.003
0304-4238/

of the clones have been evaluated. Moreover, those that have been evaluated were carried out at different periods. The first evaluation conducted in 1947 consisted of 20 clones mainly of the Trinitario, Criollo and Amelonado genetic groups for yield (Glendinning, 1964). The next clone trial conducted in 1956 evaluated 14 Upper Amazon cocoa clones for establishment and yield (Lockwood, 1971). A more recent evaluation, the International and Local clone trials were established in 2000 to evaluate 25 International clones and 25 local clones for yield, diseases and pests (Adomako et al., 2007). A comprehensive evaluation of the germplasm clones for yield, diseases and pests is important to the cocoa breeding program for an effective comparison and determination of clones for future breeding efforts.

Different breeding strategies have been adopted to evaluate and select high yielding cocoa clones. Cilas et al. (2010) used bean weight, number of beans per pod and number of ovules per ovary to determine yield production in cocoa. Average weight, length and breadth of cocoa beans were used to select some high yielding clones in the Guiana population (Lachenaud and Oliver, 2005). The relationship between juvenile tree growth and early yields has been used for selecting high yielding cocoa genotypes in later years (Glendinning, 1960; Padi et al., 2012; Ofori et al., 2015). Yield efficiency, the ratio of cumulative yield to stem increments has currently gained interest and used as indicator for selecting high yielding cocoa cultivars (Adomako et al., 1999; Daymond et al., 2002; Padi et al., 2012, 2016). Cocoa clones that partition smaller proportions of assimilates to vegetative growth are preferred for establishment because they facilitate crop management operations such as pruning, application of foliar pesticides and fertilizers, and harvesting. For the effective utilization and selection of cocoa clones as parents in breeding programs, knowledge of genetic variation of key agronomic traits is important. Information on genetic parameters such as variance, heritability as well as correlation among agronomic traits in cocoa clones is limited. Such knowledge is useful for cocoa breeders to identify potential parents for breeding improved cultivars with broad genetic base. The aim of this study is to analyze the genetic variation for stem diameter increments (SDI) and yield component traits as well as the relationship between these traits in a set of 116 cocoa clones. The specific objectives were, (1) to evaluate the performance of cocoa clones for SDI in both juvenile and productive stage, yield and its component traits, (2) to estimate heritability for SDI in both juvenile and productive stage, yield and its component traits, and (3) to assess the phenotypic and genotypic correlations between SDI in the juvenile and yield and its component traits in cocoa clones in Ghana.

2. Materials and methods

2.1. Plant materials

A total of 116 genotypes were selected for this study. These comprised of clones from different genetic origins (Table 1) and selected on the basis of differences in their introduction period and breeding history. These include clones introduced before the beginning of formal research in 1938 which are mostly the local Amelonado and the local Trinitario introduced by Tetteh Quarshie and Governor Griffiths, respectively (Lockwood and Gyamfi, 1979). These were the dominant planting materials in Ghana until 1944 when Posnette brought in the Trinidad introductions clones (referred to as T). These composed of different combinations of open and hand pollinated pods from the upper Amazon, lower Amazon and Trinitario genetic group (Lockwood and Gyamfi, 1979). An agreement with the British Overseas Development Administration to finance a multi-disciplinary team of research workers at CRIG in 1969 (Legg, 1981), led to the introduction of true to type clones of Nanay (Na), Iquitos (IMC), Parinari (PA) a sub-group of the Marañon and Scavina (SCA) a sub-group of the Contanama from the International Cocoa Quarantine Centre, Reading between 1970s to 1990s (Abdul-Karima et al., 2006). Clones introduced after this period largely comprised of the Guyana (GU) and Purus genetic group (Abdul-Karima et al., 2006). Ten seed garden clones selected based on combining ability tests of introduced clones in crosses with local selections for vigor, yield and resistance to diseases (Glendinning, 1966) were included.

Table 1
Genetic type of 116 cocoa clones use for the study.

Genetic type	Clone
Amelonado	A 196, CAS 3, MA 12, P 30, TF 6, TF20
Nacional	MO 20*, T56/118 (MO 14 open-pollinated)
Iquito	IMC 6, T16/613, T16/618 (IMC 24 open pollinated), T17/358, T17/1856 (IMC 53 open pollinated), IMC 60*, IMC 61, IMC 67, IMC 83, AMEZ3/2
Guiana	GU 168/H, GU 212/H, GU 225/V
Local selection	ALPHAB 36, L6/428, N8/112, N33/326
Nanay	NA 33, T11/94 (NA 33 open-pollinated), T14/233, T14/295 (NA 43 open-pollinated), T13/472 (NA 60 open-pollinated), NA 124, NA 227, NA 427, Pound 7
Purus	RB 41, RB 45, RB49
Parinari	PA7*, PA 13, PA 16, PA 20, T53/46 (PA 37 open-pollinated), PA 65, PA 70, T30/539 (PA 103 open-pollinated), PA 118, PA 140, PA150*
Hybrids	SGU 50 (Criollo x Matina hybrids) T60/887*, T60/885*, T60/1052, T60/1774 and T60/975 (PA 7 x NA 33), T61/1239 and T61/13/26 (NA 33 x NA 32), T62/205 (NA 33 x NA 34), T63/762, T63/961 T63/971* T63/967* and T63/882 (PA 35 x NA32), T65/236, T65/238 and T65/328 (PA 7 x IMC 47), T72/1768 (NA 3 x IMC 60), T73/1931 (NA 33 x IMC 60), T76/1224 and T76/1835 (PA 35 x NA 31), T79/1064 and T79/467* (NA 32 x PA 7), T81/1879 and T81/1880 (NA 31 x NA 32), T82/503 (NA 32 x PA 35), T85/874 and T85/799* (IMC 60 x NA 34), T87/98 and T87/368 (IMC 60 x NA 34), T88/2130 (ICS 85 self), T90/114 (IMC 76 x NA 32), T92/1614 (NA 32 x NA 31), T99/129 and T99/395 (ICS 56 x ICS 85)
Scavina	SCA 6, SCA 9, T12/61, T12/63 and T12/151 (SCA 12 open-pollinated)
Trinitario	A 45, A 72, A 164/43, A 164/45, D 26, D 70, T3/247 and T3/335 (local trinitario) ICS 16, ICS 25, ICS 39, ICS 40, ICS 43, ICS 95, E 9/195, E 75, K 5, K 9, O 2, R15, S 19, S 72, T24/229 (ICS 80 open-pollinated), T26/277 (ICS 45 open-pollinated), T39/659 (ICS 6 open-pollinated), T45/145 (ICS 70 open-pollinated), T57/308 and T57/368 (ICS 60 open-pollinated)

*Seed Garden parents included in the experiment.

2.2. Field evaluation and plant culture

Field experiment was conducted at the Cocoa Research Institute of Ghana (CRIG), Tafo (a humid rainforest belt, with latitude 06^0 13′N, 0^0 22′W). The experiment was set up on a previously cropped land without fallowing, with some of the overhead shade trees removed. Soil samples randomly taken from the experimental site and analysed before planting indicated 0.9 g/kg nitrogen, 8.6 g/kg carbon, 7.08 (μg/g) available phosphorus and 0.60 (meq/100 g) potassium. Based on the chemical components, the soil is much lower in fertility than that recommended for cocoa cultivation in Ghana (Ahenkorah et al., 1982). In generating experimental materials, scions from fan branches of the 116 selected genotypes were budded onto six-month old rootstocks generated from PA7/808 x PA 150. The clones were transplanted in July 2010, eight-months after grafting following a randomised complete block design with four replications consisting of four trees per clone per block. Within each block, the trees of all 116 clones were arranged following a single-tree randomisation procedure. The clones were planted at 3.0 m^2 spacing under both plantain as temporary shade and *Terminalia* spp. as permanent shade. The plantain was planted at 3.0 m^2

Table 2
Mean sum of squares for stem diameter increments (SDI) in the juvenile stage, stem diameter increments (SDI) in the reproductive stage, bean weight, number of beans per pod, pod value, yield and yield efficiency for 116 cocoa clones evaluated.

Source	df	SDI in the juvenile stage (cm)	SDI in the productive stage (cm)	Bean weight (g)	Number of beans per pod	Pod value	Average yield (kg/ha)	Yield efficiency (g/cm^2)
Block	3	1.46	1.11	0.05	155.43*	168.4*	5453.0	10.36
Clones	115	2.21*	0.40	0.017**	84.06**	92.85**	20512**	29.7**
Error	345	1.65	0.32	0.008	55.05	56.26	10992	18.46
CV (%)		21.2	16.99	7.90	22.88	23.76	21.78	24.35

*, ** Significant at 5% and 1% probability level, respectively; *CV*, coefficient of variation.

spacing and the permanent shade at 18.00 m^2 spacing. The shade crops and trees were planted three-months before the cocoa clones were transplanted.

In July of 2010 and 2011, each cocoa plant was fertilized with 70 g nitrogen (N) supplied as ammonium sulphate because of the low levels of fertility of the soil used for the experiment. At the start of flower production, application of fertilizers and agro pesticides, following recommended practices for cocoa production in Ghana were followed. Essentially, N:P:K (0:22:18) fertilizer was applied by broadcasting 350 kg/ha in May each year. Insect pests were controlled using imidacloprid (200 SL) as Confidor (Bayer Crop Science, Monheim, Germany) at the rate of 150 ml/ha from August to December each year. Black pod disease caused by *Phythophthora* spp. was controlled by applying Ridomil Gold (Syngenta; 60% copper oxide and 6% mefenoxam) at the rate of 3 × 4 g/l of water and applied to drench developing pods on trees. The rate of fungicide application per hectare was therefore dependent on the number of pods per tree at the time of fungicide application.

2.3. Data collection

Stem diameter of each plant was measured at 15 cm above the graft union with the aid of electronic callipers at 6-month intervals from September 2010 to June 2015. Yield data collection was carried out between July 2013 and June 2015. At harvest, the number of matured pods for each clone within blocks was combined and counted. A sample of 30 pods per clone per block was used to estimate the mean bean weight and number of seeds per pod after fermentation and drying to a moisture content of about 7%. Pod value (number of pods needed to produce 1 kg of dry beans) was estimated from the mean seed weight and number of seeds per pod. Total yield was determined from the total number of pods produced per clone per block divided by the pod value. Yield efficiency was estimated as the cumulative yield during the productive stage (averaged per tree) divided by the increase in stem cross sectional area over the same period.

2.4. Statistical analysis

Analysis of variance (ANOVA) based on best linear unbiased estimates (BLUE) using average trait values across years were performed with the following model:

$$Yij = \mu + bj + gi + eij.$$

Where, Y_{ij} is the BLUE of genotype i in block j; μ is the mean, b_j is the effect of block j (for j = number of blocks); g_i is the effect of clone i (for i = number of clone); and e_{ij} is the error. Test for normality of data bases on residual error plots a condition for ANOVA was carried out for all the traits. The REML (restricted maximum likelihood) method (Corbeil and Searle, 1976) was used to determine the genotypic variance and the error variance. All these statistical analyses were performed using GenStat statistical software version 11 (VSN International Ltd, Hemel Hempstead, UK). The phenotypic variance was then calculated as sum of the error and the genotypic variance. Broad sense heritabilities were calculated as the ratio of genetic variance (clone) and phenotypic variance as,

$$h^2_b = \sigma^2_G/\sigma^2_P$$

Where,

h^2_b, σ^2_g, and σ^2_P are broad sense heritability, genetic variance and phenotypic variance, respectively. Confidence intervals for heritabilities were estimated following (Singh et al., 1993) as,

$$[M_g/M_e - \alpha/2]/[M_g/M_e + (b-1)\alpha 2] \leq h^2 \leq [M_g/M_e - 1/\alpha 2]/M_g/M_e + [(b-1)1/\alpha 2]$$

Where,

M_g, M_e, b and $\alpha/2$ are genotypic mean square, error mean square, number of blocks and probability point of the F distribution, respectively. Genotypic and phenotypic coefficients of variation were estimated following (Burton, 1952) as,

$$GCV = \sqrt{\sigma^2}_g/\text{MandPCV} = \sqrt{\sigma^2}_p/M$$

Where,

GCV, PCV, σ^2_g σ^2_p and M are respectively genotypic coefficients of variation, phenotypic coefficients of variation, genotypic variance, phenotypic variance and trait mean. Genotypic and phenotypic correlation coefficients were estimated for all pairs of traits using multivariate model with SAS Proc Mixed procedure (Isik, 2009).

3. Results

3.1. Variation among clones for traits

Analysis of variance showed a highly significant ($p < 0.01$) variation among clones for all the traits except stem diameter increments (SDI) in the productive stage (Table 2). The effects of block were significant ($p < 0.05$) for only number of beans per pod and pod value. Stem diameter increments (SDI) in the juvenile stage varied from 4.12 cm (clone T12/61) to 7.83 cm (clone T57/308). Distribution of bean weight among the clones showed that 81% of it had weight above the minimum value (1.0 g) established by the chocolate industry. Number of beans per pod ranged from 17.0 in clone T39/651 to 57 in clone K5. Pod value, an indirect measurement of yield components had a mean of 32. Clones T16/613, T65/238, and T73/1931 had superior yielding ability than the best seed garden clone (T63/671–852 kg/ha) (Table 4). Regarding yield efficiency, the mean was 15.70 g/cm^2 and varied from 9.90 g/cm^2 to 38.86 g/cm^2 for clone ICS 16 and T65/238, respectively.

3.2. Genotypic and phenotypic coefficient of variation and heritability for traits

The phenotypic coefficient of variation (PCV) was higher than the genotypic coefficient of variation (PCV) for all the traits studied (Table 3). Yield efficiency had the highest genotypic coefficient of variation (GCV) of 10.6% and SDI in the productive stage had the lowest (3.89%) compared with the other traits. The phenotypic

Table 3
Estimates of mean, range and genetic variability components for stem diameter increments (SDI) in the juvenile stage, stem diameter increments (SDI) in the reproductive stage, bean weight, number of beans per pod, pod value, yield and yield efficiency for 116 cocoa clones evaluated.

Genetic parameters	SDI in the juvenile stage (cm)	SDI in the productive stage (cm)	Bean weight (g)	Number of beans per pod	Pod value	Average yield (kg/ha)	Yield efficiency (g/cm^2)
Mean	5.85	3.37	1.16	33.7	32	498	15.70
Range	4.12–7.70	2.03–4.68	0.79–1.72	14.07–57.0	15–59	183–952	4.78–38.86
Vg	0.15	0.017	0.003	7.14	8.98	2380.07	2.76
Vp	1.80	0.347	0.011	62.24	65.35	13372.3	20.25
GCV	6.62	3.86	4.72	8.09	9.31	9.80	10.6
PCV	15.2	17.3	9.04	23.5	25.3	23.0	28.9
h^2bs	$0.02 \leq 0.08 \leq 0.14$	$0.006 \leq 0.05 \leq 0.11$	$0.21 \leq 0.27 \leq 0.33$	$0.06 \leq 0.11 \leq 0.16$	$0.08 \leq 0.14 \leq 0.20$	$0.14 \leq 0.18 \leq 0.23$	$0.07 \leq 0.14 \leq 0.19$
Ve/Vg	11.0	19.0	2.65	7.70	6.26	4.61	6.60

Vg – genotypic variance, Vp – phenotypic variance, GCV – genotypic coefficient of variation, PCV – phenotypic coefficient of variation, h^2bs – broad sense heritability and its confidence intervals and Ve – environmental variance.

Table 4
The ten best and worst yielding clones with their corresponding bean weight, number of beans per pod, pod value, stem diameter increments (SDI) in the juvenile stage, stem diameter increments (SDI) in the reproductive stage and yield efficiency for 116 cocoa clones evaluated.

Clone	Average yield (kg/ha)	Bean weight (g)	Number of beans per pod	Pod value	SDI in the juvenile stage (cm)	SDI in the productive stage (cm)	Yield efficiency (g/cm^2)
Best ten clones							
T16/613	952	1.45	36.91	20	6.69	4.43	30.68
T65/238	938	1.38	43.00	19	5.70	3.49	38.86
T73/1931	852	1.35	42.50	21	5.87	4.04	30.07
T63/961*	850	1.33	35.14	24	5.89	3.92	31.94
ICS40	829	1.47	30.40	25	7.10	3.90	29.03
IMC83	820	1.20	41.00	20	7.77	4.39	24.39
T60/887*	813	1.42	47.67	18	5.74	3.90	29.17
SGU50	812	1.22	38.00	25	5.10	3.83	32.02
ICS39	800	1.38	33.33	24	6.99	3.96	29.94
PA150*	796	1.21	40.50	25	5.89	3.91	28.48
Worst ten clones							
SCA6	183	0.79	29.67	52	4.60	2.20	12.97
A196	211	0.80	25.50	51	4.65	2.31	13.38
SCA9	213	0.84	28.00	43	4.90	2.62	11.87
E75	218	1.05	19.67	56	5.60	2.95	10.68
NA427	253	1.08	25.75	42	6.12	3.72	10.08
ICS16	255	1.17	32.17	30	6.19	3.71	9.90
T53/46	267	0.98	36.00	33	6.10	3.67	10.78
T79/1150	267	0.99	19.00	53	5.75	4.00	9.94
P30	275	1.17	22.07	40	4.95	2.9	12.82
T39/651	277	1.16	17.60	57	6.20	3.54	11.31
SED ($P < 0.05$)	76.5	0.07	5.04	5.3	0.90	0.40	3.97

SED, Standard error of difference, * Seed garden parents included in the study.

coefficient of variation (PCV) followed the same trend with yield efficiency having the highest (28.9%) and bean weight the lowest (13.7%). The environment to genetic variance ratio indicates the importance of environmental effects in controlling traits. This was positive for all the traits and led generally to low heritability estimates, which ranged from $h^2 = 0.05 \pm 0.05$ for SDI in the productive stage to $h^2 = 0.27 \pm 0.06$ for bean weight.

3.3. Genotypic and phenotypic correlations among traits

The genotypic and phenotypic correlations between traits produced estimates that were similar in direction and magnitude (Table 5). Genotypic correlations (r_g) and phenotypic correlations (r_p) among bean yield component traits (bean weight, number of bean per pod and yield efficiency) were significantly positive. Generally, both genotypic and phenotypic correlations between SDI in juvenile stage and yield component traits were positive whereas that between SDI in the productive stage and yield component traits were negative. Pod value negatively correlated with all the other traits with yield recording the highest ($r_g = -0.89$, $P \leq 0.01$; $r_p = -0.64$, $P \leq 0.01$). Juvenile SDI negatively correlated with SDI in the productive stage ($r_g = -0.23$, $P \leq 0.01$; $r_p = -0.44$, $P \leq 0.01$). In comparison, the genotypic correlations were considerably higher than their corresponding phenotypic correlations in most cases for trait combinations.

4. Discussion

Information on the level of genetic variation and relationship among key agronomic traits of cocoa is very important in selecting parental clones for yield improvement in plant breeding programs. Till present, very few of the cocoa germplasm clones in Ghana have been evaluated for traits such as bean weight, number of bean per pod, pod value and yield efficiency (Glendinning, 1964; Lockwood, 1971; Adomako et al., 2007). In addition, the previous evaluations were carried out at different periods, making it difficult to compare clones because of differences in weather conditions (humidity, temperature and rainfall) across years. A comprehensive evaluation of clones introduced across years for key agronomic traits is relevant because it enables valid comparison to be made among clones. The experiment was established on a previously cropped land without fallowing, reflecting the lower level of fertility of soils used in the experiment compared to soils recommended for cocoa cultivation in Ghana (Ahenkorah et al., 1982).

Results from the present study indicated the presence of significant variation among clones for bean yield and its component traits and stem diameter increments. Even though the trial was

Table 5
Phenotypic (above diagonal) and genotypic (below diagonal) correlation coefficients for stem diameter increments (SDI) in the juvenile stage, stem diameter increments (SDI) in the reproductive stage, bean weight, number of beans per pod, pod value, yield and yield efficiency for 116 cocoa clones evaluated.

Traits	SDI in the juvenile stage (cm)	SDI in the productive stage (cm)	Bean weight (g)	Number of beans per pod	Pod value	Average yield (kg/ha)	Yield efficiency (g/cm^2)
SDI in the juvenile (cm)	–	−0.44**	0.02	0.12	−0.11	0.22*	0.28**
SDI in the productive stage (cm)	−0.23**	–	0.11	−0.05	−0.002	0.09	−0.39**
Bean weight (g)	0.04	0.39**	–	0.02	−0.36**	0.28**	0.17
Number of beans per pod	0.28**	−0.08	0.09	–	−0.72**	0.51**	0.49**
Pod value	−0.49**	−0.01	−0.49**	−0.87**	–	−0.64**	−0.61**
Average yield (kg/ha)	0.47**	0.15	0.42**	0.79**	−0.89**	–	0.70**
Yield efficiency (g/cm2)	0.56**	−0.09	0.18*	0.84**	−0.87**	0.85**	–

*, ** Significant at 5% and 1% probability level, respectively.

established on a previously cropped land without fallowing, the block effects were not significant for most traits. This points to a less heterogeneous environment and indicate that the experiment was not affected by cropping history or shade species planted, and the design selected for the experiment was suitable. Zhang et al. (2015) in comparing single plant randomization within plots and multiple plot randomization concluded that the former have the advantage of testing large number of clones simultaneously with good replication giving high statistical precision. In all, a total of 16 plants per clone were used for the study. This is adequate because clones are true to type and such a number is enough to determine their mean performance as indicated in an earlier experiment (Cilas et al., 2010).

Despite the non-significant block effect, heritability was generally low for the studied traits. This is supported by the relatively higher environmental variance, which were 2.6 for bean weight, 7.77 for number of beans per pod, 6.20 for pod value, 4.58 for bean yield, 11.00 for SDI in the juvenile stage and 6.61 for yield efficiency. Increased sampling of the experimental area (through increased number of tree per replications) may be required to increase the precision of the observed differences and minimize the error variance in the study. Bean weight had the highest heritability of 0.27 ± 0.06. This is lower than 0.51 reported by Cilas et al. (2010) and 0.78 by Pardo and Enriquez (1988). The heritability for number of beans per pod of 0.11 ± 0.05 was also lower than 0.29 observed by Cilas et al. (2010).

Nevertheless for bean yield, heritability value found in this study ($h^2 = 0.18 \pm 0.04$) is quite comparable with earlier report of 0.15 (Lockwood et al., 2007) and 0.11 ± 0.1 (Padi et al., 2016). Heritability of SDI in the juvenile stage of $h^2 = 0.08 \pm 0.06$ was also similar to 0.16 ± 0.19 (Padi et al., 2016). The genetic coefficients of variation (GCV) provides a measure for comparing genetic variability in quantitative traits, and together with heritability estimates determine the extent of heritable variation (Burton, 1952). In this study, bean weight with the highest heritability of 0.27 had only 4.72 for GCV as against 10.6 for yield efficiency which had heritability of 0.14. These inconsistencies have been reported (Nyadanu and Dikera, 2014) and the selection of parents for further breeding activities should rely mainly on genotypic performance.

Variations among the clones for the traits were quite high and this may be attributed to the larger number of clones included in the study. Stem diameter increments in the Juvenile stage was lowest in clone T12/61, an open pollinated clone of SCA 12 of the Contanama genetic group. Knight and Rogers, (1955) described SCA 12 as moderately tall with short weeping branches. Clone T57/308 which had the best SDI in the juvenile stage is an open pollinated tree of ICS 60 of the Trinitario genetic group. The high SDI in the juvenile stage of Clone T57/308 is consistent with what has been found in other studies (Toxopeus, 1969; Laurent et al., 1994; Phillips, 2003) that ICS genotypes are generally vigorous.

Number of seeds per pod is largely influence by the number of ovules per ovary, the fertility of the ovules and the tree reproductive nature (self-compatibility or self-incompatibility). These attributes may vary among genotypes and may have accounted for the large differences between clones for number of beans per pod, which agrees with previous reports that number of bean per pod is very variable (Cilas et al., 2010; Tan 1990). Bean weight on the other hand is highly heritable. The local Trinitario clone A 72 had the best bean weight (1.72 g), confirming Motamayor et al. (2002) observations that clones of Trinitario origin generally have large seeds. The worst bean weight of Clone SCA 6 (0.79 g) corresponds to findings of Cilas et al. (2010) who reported 0.71 g. SCA 6 is generally discriminated in most breeding applications because it is characterised by poor yield components even though it tolerates black pod diseases (Barreto et al., 2015). The bean yield of SCA 6 was the worst (183 kg/ha) as against (952 kg/ha) for clone T16/613, an open pollinated tree of IMC 24. The yield of T16/613 was similar to levels obtained in commercial plantations in Ghana which have average maximum yields of 450 kg/ha, 700 kg/ha and 1000 kg/ha, under low-input, medium-input or high-input systems, respectively (Gockowski et al., 2011).

Yield efficiency varied among the clones and two different types could be identified among the high yielding clones; high yielding clones with moderate SDI in the productive stage (e.g. T65/238) and high yielding clones with high SDI in the productive stage (e.g. T16/613). For instance, clone T16/613 invariably had 14 kg yields higher than T65/238, but the later had SDI in the productive stage of 3.49 cm and yield efficiency of 38 g/cm^2 whereas the former had SDI in the productive stage of 4.43 cm and yield efficiency of 30 g/cm^2. These differences enable the selection of high yielding varieties with suitable tree sizes.

Out of the best ten clones for SDI in the juvenile stage, bean yield and yield efficiency, seven (IMC 67, IMC 83, T73/1930, T65/238, PA 150, T16/613, T60/887 and T63/961) comes from either the Parinari or Iquitos genetic groups. This concur with previous combining ability studies for SDI in the juvenile stage, bean yield and yield efficiency (Padi et al., 2016) which observed favorable GCA effects for clones of the Parinari and Iquitos genetic group. Future utilization of the best clones could be done with-out further general combining ability studies.

The significant positive (genotypic and phenotypic) correlations observed between bean yield and the other yield component traits indicate that direct selection for any of those traits would favor bean yield. The correlation between bean weight and number of beans per pod was very weak ($r_g = 0.09$; $r_p = 0.02$) and agrees with Cilas et al. (2010) who reported a correlation of $r_g = 0.001$. This implies that both traits are independent, so it is possible to select materials with high bean weight and large number of beans per pod. The significant positive genotypic correlations observed between SDI in the juvenile stage and yield component traits especially

for bean yield ($r_g = 0.47$, $P \leq 0.001$) implies that SDI in the juvenile stage could be a better indicator for determining bean yield in later years as indicated in other studies (Glendinning, 1960; Padi et al., 2012; Ofori et al., 2015). With regards to SDI in the productive stage, the negative correlations with yield component traits could be attributed to assimilate partitioning during production. Competition for assimilate during the production stage favors pod development than stem growth in high yielding clones relative to clones with low bean yield which reflected in the large variation for yield efficiency. This is consistent with earlier reports (Daymond et al., 2002; Padi et al., 2016), who argues that yield efficiency which is an index that integrates yield with vegetative growth is much better indicator of productivity than yield itself.

The negative correlation between pod value and yield component traits with the highest occurring in bean yield ($r_g = -0.89$, $P \leq 0.001$), indicates that the lower the pod value the higher the bean yield. This concurs with (Tan, 1990) who reported a negative correlation of $r = -0.73$ between pod value and bean weight, ($r = -0.7$) between pod value and number of beans per pod and ($r = -0.33$) between pod value and bean yield. Generally, the clone with good pod value were among the best for bean yield, high number of beans per pod and seed weight, and comes from either the Parinari, Iquitos or Trinitario genetic groups.

In conclusion, this study has demonstrated the effectiveness of using a single-tree randomisation within block design for evaluating a large number of clones for key agronomic traits. With the large number of cocoa clones belonging to various genetic groups tested for their agronomic performance, the available information could be valuable for selection of parental clones for breeding. Results from the study showed that there is considerable genetic variation among the clones for yield component traits and stem diameter increment in the juvenile stage. In fact clones from the Parinari, Iquitos and Trinitario genetic groups (e.g. T16/613, T65/238, ICS 40, IMC 83 and SGU 50) represent the top ten percent and they could be the group of choice in future breeding programs. Previous evaluation studies on combining abilities, resistance to disease and pest, and other physiological traits have extensively covered these three populations (Adomako et al., 1999; Padi et al., 2016). They could be used directly as parental clones in hybridization programs without further tests on those traits. The moderately high genetic correlation ($r_g = 0.47$, $P \leq 0.01$) between SDI in the juvenile stage and bean yield indicate that, even though the study covers juvenile tree growth and two years yield component traits data, high yielding parents could be selected at this stage for use in other breeding activities.

Acknowledgments

The authors gratefully thank the field and technical staff of the Plant Breeding Division, CRIG, for their support and assistance, especially Mr. Robert Dorbgadzi for their support and assistance. This work is published with the kind permission of the Executive Director of Cocoa Research Institute of Ghana as manuscript number CRIG/03/2016/024/006.

References

Abdul-Karima, A., Adomako, B., Adu-Ampomah, Y., 2006. Cocoa introduction into Ghana. Ghana J. Agric. Sci. 39, 227–238.

Adomako, B., Allen, R.C., Adu-Ampomah, Y., 1999. Combining abilities for yield and vegetative traits of Upper Amazon cocoa selections in Ghana. Plant. Res. Dév. 6, 183–189.

Adomako, B., Padi, F.K., Opoku, S.Y., Assuah, M.K., Domfeh, O., Owusu-Ansah, F., 2007. International Clone Trial and Local Clone Selection Trial. Annual Report. Cocoa Research Institute of Ghana43–49, 2007/08.

Ahenkorah, Y., Halm, B.J., Appiah, M.R., Akrofi, G.S., 1982. Fertilizer use on cacao rehabilitation projects in Ghana. In: Proceeding of the 8th International Cocoa Research Conference, Cartagena, Colombia 1981, pp 165–170.

Allen, J.B., Lass, R.A., 1983. London cocoa trade amazon project. Cocoa Growers Bull. 34, 74.

Barreto, M.A., Santos, J.C.S., Corrêa, R.X., Luz, E.D.M.N., Marelli, J., Souza, A.P., 2015. Detection of genetic resistance to cocoa black pod disease caused by three Phytophthora species. Euphytica 206, 677–687.

Burton, G.W., 1952. Quantitative inheritance in grasses. Grassland Congress In: Proceedings of the 6th Int, 1, pp. 227–283.

Cheesman, E.E., 1944. Notes on the nomenclature, classification and possible relationships of cocoa populations. Trop. Agric. 21, 144–159.

Cilas, C., Machado, R., Motamayor, J.C., 2010. Relationship between several traits linked to sexual plant reproduction in *Theobroma cacao* L: number of ovules per ovary, number of seed per pod, and seed weight. Tree Genetic Genomes 6, 219–226.

Corbeil, R.R., Searle, S.R., 1976. Restricted maximum likelihood (REML) estimation of variance components in the mixed model. Technometrics 18, 31–38.

Daymond, A.J., Hadley, P., Machado, R.C.R., Ng, E., 2002. Genetic variability in partitioning to the yield component of cacao (*Theobroma cacao* L.). HortScience 37, 799–801.

Glendinning, D.R., 1960. The relationship between growth and yield in cocoa varieties. Euphytica 9, 351–355.

Glendinning, D.R., 1964. A study of clonal cocoa variety. Hort. Res. 4, 89–97.

Glendinning, D.R., 1966. Further developments in the breeding programme at the Cocoa Research Institute, Tafo. Ghana J. Sci. 6, 52–62.

Gockowski, J., Afari-Sefa, V., Dziwornu, A., Fredua-Agyeman, N., 2011. Economic cost-benefit analysis of fine flavor and certified sustainable cocoa production systems in Ghana. Q. J. Int. Agric. 50, 175–200.

International Cocoa Organization (ICCO), 2013. Quarterly Bulletin of Cocoa Statistics, vol. XXXIX. International Cocoa Organization, London, UK, No. 4, Cocoa year 2012/13.

Isik, F., 2009. Genetic Correlations and Correlated Response Quantitative Forest Genetics Lecture 9: Course Notes with Codes on SAS Proc Mixed Procedures., pp. 1–23.

Knight, R., Rogers, H.H., 1955. Recent introductions of West Africa *Theobroma Cacao* and Related Species: a review of the first ten years. Emp. J. Exp. Agric. 23, 113–125.

Lachenaud, P., Oliver, G., 2005. Variability and selection for morphological bean traits in wild cocoa trees (*Theobroma cacao* L.) from French Guiana. Genet. Resour. Crop Ev. 52, 225–231.

Laurent, V., Risterucci, A.M., Lanaud, C., 1994. Genetic diversity in cocoa revealed by cDNA probes. Theor. Appl. Genetic 88, 193–198.

Legg, J.T., 1981. The British cacao swollen shoot project in Ghana, 1969–1981. In: Proceedings of the 7th Int. Cacao Res. Conference, Douala, Cameroun 1979, pp. 399–406.

Lockwood, G., Gyamfi, M.M.O., 1979. The Cocoa Research Institute of Ghana Cocoa Germplasm Collection with Notes on Codes Used in the Breeding Programme at Tafo and Elsewhere Technical Bulletin No 10. Cocoa Research Institute of Ghana, New-Tafo, Akim, Ghana.

Lockwood, G., Owusu-Ansah, F., Adu-Ampomah, Y., 2007. Heritability of single plant yield and incidence of black pod yield in cocoa. Exp. Agric. 43, 455–462.

Lockwood, G., 1971. Early results from a trial of Upper Amazon cocoa clones in Ghana. Exp. Agric. 7, 321–327.

Motamayor, J.C., Risterucci, A.M., Lopez, P.A., Ortiz, C.F., Moreno, A., Lanaud, C., 2002. Cacao domestication I: the origin of the cacao cultivated by the Mayas. Heredity 89, 380–386.

Motamayor, J.C., Lachenaud, P., Mota, S.J.W., Loor, R., Kuhn, D.N., Brown, S., Schnell, R.J., 2008. Geographic and genetic population differentiation of the Amazonian chocolate tree (*Theobroma cacao* L.). PLoS One 3, 10.

Nyadanu, D., Dikera, E., 2014. Exploring variation: relationships and heritability of traits among selected accessions of sorghum (*Sorghum bicolor* L. Moench) in the Upper East Region of Ghana. J. Plant Breed. Genetics 2, 101–107.

Ofori, A., Padi, F.K., Acheampong, K., Lowor, S., 2015. Genetic variation and relationship of traits related to drought tolerance in cocoa (*Theobroma cacao* L.) under shade and no-shade conditions in Ghana. Euphytica 201, 411–421.

Padi, F.K., Opoku, S.Y., Adomako, B., Adu-Ampomah, Y., 2012. Effectiveness of juvenile tree growth rate as an index for selecting high yielding cocoa families. Sci. Hort. 139, 14–20.

Padi, F.K., Ofori, A., Takrama, J., Djan, E., Opoku, S.Y., Dadzie, A.M., Bhattacharjee, R., Motamayor, J.C., Zhang, D., 2015. The impact of SNP fingerprinting and parentage analysis on the effectiveness of variety recommendations in cacao. Tree Genetics Genomes 11, 44.

Padi, F.K., Ofori, A., Auther, A., 2016. Genetic variation and combining abilities for vigour and yield in a recurrent selection programme for cacao. J. Agric. Sci., 1–21, http://dx.doi.org/10.1017/S0021859616000459.

Pardo, J., Enriquez, G.A., 1988. Herencia de agunos components de la calidad industrial en almendras de cacao (*Theobroma cacao* L.). In: Pro 10th Int. Cocoa Res. Conf., Cocoa Producers Alliance, Santo Domingo, pp. 695–699.

Phillips, W., 2003. Origin, Biogeography, Genetic Diversity and Taxonomic Affinities of the Cacao Fungus Moniliophthora Rorei (cif.) Evan Et Al., as Determined Using Molecular, Phytopathological and Morpho-physiological Evidence. PhD. Dissertation. the University of Reading, UK (pp. 349.).

Singh, M., Ceccarelli, S., Hamblin, J., 1993. Estimation of heritability from varietal trials data. Theor. Appl. Genetic 86, 437–441.

Tan, G.Y., 1990. Combining ability analyses of yield and its components in cacao. J. Am. Soc. Hortic. Sci. 115, 509–512.

Toxopeus, H., 1969. Cacao (*Theobroma Cacao* L.) In: Outlines of Perennial Crop Breeding in the Tropics. Misc. Papers 4. Landbouwhogeschool Agric Univ., Wageningen. The Netherlands, pp. 81–109.

Zhang, J., Peter, G.F., Powell, G.L., White, T.L., Gezan, S.A., 2015. Comparison of breeding values estimated between single-tree and multiple-tree plots for a slash pine population. Tree Genetics Genomes 11, 48.