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Full Length Research Paper

Grain yield and stability of selected early and medium duration cowpea in Ghana

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Changes in climate are a major driver for climate-smart crops with short duration on-field and adaptation to diverse growing conditions. This study evaluated the performance of nine early duration and 10 medium duration cowpea genotypes at six locations within the Guinea and Sudan savanna zones of Ghana. Genotypes for each maturity group were laid out in a Randomized Complete Block Design with three replications for each location. There were significant ($p<0.001$) genotype, environment and genotype x environment effects of the cowpea genotypes of both maturity groups for grain yield. Among the early duration cowpea tested, GGE biplot analysis revealed SARI-2-50-80, SARI-13-17-2, IT99K-1122, SARI-3-11-80, and IT07K-299-6, respectively, as having high yield and stable performance across the six test environments; and out-performed the check variety, Kirkhouse Benga. With the medium duration trials, IT86D-610, IT10K-837-1, and SARI-6-2-6 had high yields, which were comparable to the check, Padituya. IT10K-837-1 was the most stable and had a relatively shorter maturity period. Grain yield performance of early duration cowpea was discriminated by three mega environments while only two mega environments discriminated grain yield of medium duration cowpea. The selected genotypes could be used in hybridizations or released as cowpea varieties in the country.

Key words: Genotype x environment, maturity period, multi-location, biplot analysis, genetic variability.

INTRODUCTION

Cowpea (*Vigna unguiculata* (L.) Walp) is one of the most important cultivated grain legumes in sub-Saharan Africa, possibly because of its relatively wide adaptation to drought and ability to give appreciable yields on low-nutrient soils, where other crops would fail (Pule-Meulenberg et al., 2010). It has key importance in ensuring food and income security of smallholder farmers in sub-Saharan Africa (Langyintuo et al., 2003). It is widely cultivated in all ecologies and it is a constituent crop in most farming systems, grown either as intercrop or relay crop, particularly in the northern parts of Ghana (Quaye et al., 2011). At different places and times in Africa, the grain, the green pods, the dried leaves, and hay all command good market prices. The most important factor driving its demand is the high protein it offers which could be as high as 40 % in both fodder and grains

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(Dakora and Belane, 2019). Cowpea can derive a substantial amount of its nitrogen nutrition from symbiotic nitrogen fixation (Pule-Meulenberg et al., 2010) and N-fixation in cowpea has been reported to vary from 16.6-23.0 kg/ha in Northern Ghana (Naab et al., 2009).

The yield of cowpea in Ghana is, however, still the lowest in the world averaging 0.5 t/ha despite the numerous importance of this crop (Langyintuo et al., 2003; Ofosu-Budu et al., 2008). Yield is a quantitative trait that is influenced by the environment. Cowpea production is constrained by various field pests, disease infestation, amount of rain, drought, and photoperiod and this varies from one agroecology to another. This differential environmental condition resulting in differing yield responses of cowpea genotypes can be attributed to genotype-by-environment interaction (Odeseye et al., 2018). To identify high-yielding and well-adapted cowpea genotypes, multi-location evaluation of a large number of diverse improved cowpea genotypes must be carried out within the different agro-ecologies of the region. Multi-environment evaluation trials of crop cultivars are essential as genotype performance will be determined across different environments. Through this, the genotype by environment analysis can be conducted to help identify high yielding and stable genotypes for the test environments (Asio et al., 2009; Horn et al., 2018; Sousa et al., 2018). Since genotype-by-environment interaction (GEI) limits selection response there is a need to investigate its presence. Previous studies have dwelt on using conventional methods of analysis to determine the genotype x environment interaction and these methods provide little information on the patterns in the interaction (Kempton, 1984). A simple crop physiological model to study the yield basis and environmental effects of cowpea genotypes was reported in a multilocational study (Marfo and Waliyar, 1997). Recently AMMI and GG-Biplot analysis have been conducted on most crops to understand the genotype x environment effects. The GG-Biplot however, offers more advantages, as it is much easier to understand, identify high performing and stable genotypes, identify mega environments, and select discriminative and representative environments for the traits of interest (Dos Santos et al., 2016; Sousa et al., 2018). The GG-Biplot methodology has, however, not been much exploited in identifying high-yielding and stable cowpea genotypes in Ghana.

This study aims at evaluating cowpea genotypes across six locations in the Guinea and Sudan savanna agroecological zones of Ghana, identifying high-performing cowpea genotypes and making recommendations for its utilizations in either hybridizations or future release. Specifically, this study sought to: (i) Evaluate the genotype x environment interaction, (ii) Identify high-yielding and stable cowpea genotypes, and (iii) Identify mega-environments, as well as discriminative and representative environments for cowpea grain yield in Northern Ghana.

MATERIALS AND METHODS

Experimental materials and location of study

A total of 19 cowpea advanced breeding lines and two checks were used in this study. This includes nine early duration lines with Kirkhouse Benga as a check variety, and 10 medium duration genotypes with Padituya as check variety (Table 1). Evaluations were conducted in the 2018 cropping season in six environments namely, Nyankpala, Yendi, Damongo, Manga, Wa, and Tumu.

Experimental design and analysis

The experiment was laid out in a Randomized Complete Block Design with three replications. Two separate experiments were set up for each location, one for early duration genotypes and another for medium duration genotypes. Experimental design consists of 4 rows of 4 m length with spacing 20 cm within rows and 60 cm between rows. Crops were established under rainfed conditions with no irrigation. Rainfall pattern during the growing period is described in Table 2. Field pests were controlled using K-Optimal (Cyhalothrin 15 g/L + Acetamiprid 20; EC) at the rate of 500 ml per ha at vegetative, flowering, and podding stages. Weeds were manually controlled as, and when, necessary. Data on agronomic performance such as days to 90 % maturity, grain yield and 100 seed weight were taken for the various trials. Two middle rows were harvested (net plots) to estimate grain yield per plot and this was converted to t/ha

Data were subjected to analysis of variance (ANOVA) using the GENSTAT 12th edition (Payne, 2002). Means were separated using LSD at 5 %. The combined analysis was also conducted to test for the presence of genotype by environment interaction effect. This led to genotype x environment interaction analysis using the genotype-by-environment interaction (GGE) -biplot model (Yan et al., 2000, 2007) done using GENSTAT.

RESULTS

Days to 90% maturity, 100 seed weight, and grain yield

There were variations in days to 90 % maturity among early maturing genotypes (Table 3), and medium maturing genotypes (Table 4) performance within locations and across locations. Among the early maturing category, SARI-5-5-5, SARI-6-2-9, SARI-2-50-80, and SARI-1-3-90 attained 90 % maturity in less than 65 days after planting (DAP); while the check variety (Kirkhouse Benga) took 65 days (Table 3). All the nine medium maturing lines evaluated reached mean maturity of 67 DAP, which was much earlier than the check variety, Padituya which reached maturity at 69 DAP (Table 4).

Variations were also observed in 100 seed weight of cowpea genotypes. SARI-2-50-80 had the highest seed weight and the only genotype that outperformed the check variety in terms of 100 seed weight (Table 5). Apart from the check variety with an average 100 seed weight of 20.86 g, IT10K-837-1 had the highest 100 seed weight among the other medium maturing lines with a weight of 18.4 g (Table 6). There were significant differences ($P<0.05$) in the grain yield of early maturing

Table 1. List of genotypes used for the study.

Genotype	Source	Maturity group	Market class
IT99K-1122	IITA	Early (60-70 DAP)	Brown seeded
IT07K-298-15	IITA	Early (60-70 DAP)	White seeded
SARI-1-3-90	CSIR-SARI	Early (60-70 DAP)	Mottled
SARI-5-5-5	CSIR-SARI	Early (60-70 DAP)	White seeded
IT07K-299-6	IITA	Early (60-70 DAP)	Medium size, white seeded with black hilium
SARI-13-17-2	CSIR-SARI	Early (60-70 DAP)	White seeded
SARI-1-50-81	CSIR-SARI	Early (60-70 DAP)	Whites seeded
SARI-6-2-9	CSIR-SARI	Early (60-70 DAP)	White seeded with brown eye
SARI-3-11-80	CSIR-SARI	Early (60-70 DAP)	Large, white seeded with black eye
SARI-2-50-80	CSIR-SARI	Early (60-70 DAP)	Large, white seeded with black eye
Kirkhouse-Benga (check)	CSIR-SARI	Early (60-70 DAP)	Large, white seeded with black eye
IT06K-137-1	IITA	Medium (70-75 DAP)	White seeded
IT07K-298-15	IITA	Medium (70-75 DAP)	White seeded
IT07K-299-69	IITA	Medium (70-75 DAP)	White seeded
IT08K-126-19	IITA	Medium (70-75 DAP)	White seeded
IT09K-456	IITA	Medium (70-75 DAP)	White seeded
IT10K-837-1	IITA	Medium (70-75 DAP)	Large, white seeded with black eye
IT86D-610	IITA	Medium (70-75 DAP)	Brown seeded
IT98K-628	IITA	Medium (70-75 DAP)	White seeded
SARI-6-2-6	CSIR-SARI	Medium (70-75 DAP)	Mottled
Padituya (check)	CSIR-SARI	Medium (70-75 DAP)	Large, white seeded with black eye

Table 2. Description of trial location and rainfall pattern in 2018 cropping season.

Environment	Agroecology	Latitude	Longitude	Mean Precipitation (mm)		
				July	August	September
Nyankpala	Guinea savanna	09° 23' N	01° 00' W	150	190	220
Yendi	Guinea savanna	09° 30' N	00° 01' W	180	220	250
Damongo	Guinea savanna	09° 01' N	01° 36' W	160	220	250
Manga	Sudan savanna	11° 01' N	00° 16' W	180	230	170
Tumu	Guinea savanna	10° 54' N	01° 95' W	280	270	170
Wa	Guinea savanna	10° 04' N	02° 30' W	140	190	195

cowpea genotypes grown at each of the six locations (Table 7). Grain yield of SARI-2-50-80 (1.92 t/ha), SARI-13-17-2 (1.91 t/ha), IT99K-1122 (1.84 t/ha), SARI-3-11-80 (1.76 t/ha) and IT07K-299-6 (1.75 t/ha) were higher than the check, Kirkhouse Benga (1.69 t/ha). The highest grain yields were obtained for trials conducted at Damongo (2.09 t/ha) and the lowest was obtained for in trials conducted at Tumu (1.32 t/ha). There were significant ($p<0.05$) differences in grain yield of medium maturing lines (Table 8). Yields of IT86D-610 (2.06 t/ha), IT10K-837-1 (1.94 t/ha) and SARI-6-2-6 (1.82 t/ha) were comparable the Padituya (2.09 t/ha). There was no significant difference ($p<0.05$) between the yield of IT10K-837-1 and Padituya in five out of the six locations. Damongo had the highest grain yields (1.91 t/ha) and the lowest grain yields were obtained in Tumu (1.47 t/ha).

Genotype by environment effect and stability analysis for grain yield

Environment, Genotype, and Genotype x Environment were significant ($p<0.001$) and respectively explained 45.91, 25.74, and 13.47% of yield variance (Table 9). The environment was the most important source of variation. SARI-2-50-80, SARI-13-17-2, IT99K-1122, SARI-3-11-80, and IT07K-299-6 out-performed the check in terms of yield and had stable performance across the six test environments (Figure 1). Environment, Genotype, and Genotype x Environment were significant ($p<0.001$) and respectively explained 10.02, 56.67, and 30.11 % of yield variance (Table 10). The genotype was the most important source of variation. The grain yield of IT10K-837-1 was the most stable across the six environments

Table 3. Days to 90 % maturity of early maturing lines.

Genotype	Days to 90 % maturity						
	Damongo	Nyankpala	Yendi	Wa	Tumu	Manga	Mean
IT99K-1122	65.67^{bc}	64.33	62.67^{ab}	68.33^{e}	70.33^{cd}	60	65.22
IT07K-298-15	66^{bc}	64.67	64.33^{de}	61.67^{abc}	69^{cd}	59.67	64.22
SARI-1-3-90	67.67^{bc}	64	62.33^{a}	62.33^{abc}	66.33^{a}	59	63.61
SARI-5-5-5	61^{a}	64	63.33^{bc}	63^{bc}	71^{d}	60	63.72
IT07K-299-6	65.33^{bc}	65	65^{e}	63.33^{bc}	70.33^{cd}	59.33	64.72
SARI-13-17-2	64.33^{b}	64.33	66f	67.33^{de}	69.67^{cd}	59.33	65.17
SARI-1-50-81	68.67^{c}	64	65^{e}	64^{cd}	68.67^{bc}	59	64.89
SARI-6-2-9	66^{bc}	64.33	63.33^{bc}	59^{a}	67^{ab}	59	63.11
SARI-3-11-80	67.33^{bc}	65	66.67^{f}	62.33^{abc}	69^{cd}	59	64.88
SARI-2-50-80	65^{b}	65	63.67^{cd}	59.67^{ab}	69.33^{cd}	59	63.61
Kirkhouse-Benga	65.67^{bc}	64.67	63.67^{cd}	67.33^{de}	70.33^{cd}	59.33	65.17
Mean	65.7	64.485	64.182	63.48	69.18	59.33	
cv%	2.7	0.9	0.8	3.2	1.5	1.2	

CV: Coefficient of variation. Genotypes with different letters are significantly different at P<0.05.

Table 4. Days to 90 % maturity of medium maturing lines.

Treatment	Days to 90 % maturity						
	Damongo	Nyankpala	Yendi	Wa	Tumu	Manga	Mean
IT06K-137-1	67.33^{cd}	69^{cd}	64.67^{ab}	66.67^{ab}	68.67^{d}	64.67^{c}	66.84
IT07K-298-15	67^{c}	65^{a}	65.67^{bc}	65.33^{a}	70^{e}	63.67^{a}	66.11
IT07K-299-69	64.33^{a}	69.33^{cd}	65.67^{bcd}	68^{cd}	72.67^{g}	65.67^{d}	67.61
IT08K-126-19	68^{cde}	69.33^{cd}	65.33^{b}	66.33^{ab}	67.33^{bc}	66.67^{e}	67.17
IT09K-456	65.67^{b}	70^{d}	66.67^{c}	66.33^{ab}	66.33^{ab}	67.67^{f}	67.11
IT10K-837-1	64.67^{ab}	67.33^{b}	64^{a}	68.33^{cd}	68.33^{cd}	68^{fg}	66.78
IT86D-610	68.33^{de}	68.33^{bc}	64.67^{ab}	65.33^{a}	65.67^{a}	63.67^{ab}	66
IT98K-628	65.67^{b}	67.67^{b}	63.67^{a}	67.33^{bc}	69.33^{de}	64.67^{ac}	66.39
SARI-6-2-6	67.33^{cd}	67.33^{b}	66.67^{cd}	68.67^{cd}	69de	68.67^{g}	67.95
Padituya	69^{e}	67.67^{b}	69.33^{e}	69.33^{d}	71.33^{g}	70h	69.44
Mean	66.73	68.1	65.63	67.17	68.87	66.33	
cv%	0.9	0.9	0.9	1.1	1	0.8	

and its yield was not significantly different from the check, Padituya in 5 out of the 6 environments (Figure 3).

GGE biplot analysis shows the relative performance of cowpea genotypes for grain yield across six environments (Figures 1 and 3). The GGE biplot for early maturing lines shows a high proportion of total GGE variance (82.96 %) of which PC1 accounts for 72.69% and PC2 accounting for 10.27 % (Figure 1). The GGE biplot for medium maturing genotypes also shows a high proportion of total GGE (81.96) with PC1 accounting for 69.89% and PC2 accounting for 12.07% (Figure 3). The Average Environment Coordination (AEC) axis, as indicated by the two arrows pointing in the opposite direction of the Biplot origin separates the genotypes that are below the average from those that are above the average. Only four genotypes have below-average grain yield for early duration cowpea (SARI-6-2-9, SARI-1-3-90, SARI-1-50-81, SARI-5-5-5) and seven genotypes have above-average grain yield (SARI-2-50-80, SARI-13-17-2, IT99K-1122, SARI-3-11-80, IT07K-299-6, Kirkhouse Benga and IT07K-298-15) (Figure 1). In terms of stability and high grain yield, SARI-2-50-80 and SARI-13-17-2 were the most outstanding. Four genotypes had below-average grain yield (IT06K-137-1, IT98K-628, IT07K-298-15, and IT09K-456) and six genotypes had above-average grain yield (Padituya, IT86D-610, IT10K-837-1, SARI-6-2-6, IT07K-299-69 and IT08K-126-19) for the medium duration cowpea genotypes (Figure 3). IT10K-837-1 and IT86D-

Table 5. 100 seed weight of early maturing cowpea lines.

Genotype	100 seed weight (g)						Mean
	Damongo	Nyankpala	Yendi	Wa	Tumu	Manga	
IT99K-1122	12.00^{a}	13^{a}	10.67^{aa}	13.5^{a}	10.67^{a}	14.33^{ab}	12.36
IT07K-298-15	15.33^{bc}	12.67^{a}	14^{b}	17.5^{de}	14^{b}	14^{a}	14.58
SARI-1-3-90	15.33^{bc}	14.67^{b}	14.67^{bc}	15^{abc}	14.67^{bc}	14.67^{abc}	14.84
SARI-5-5-5	14.33^{b}	14.67^{b}	15^{bcd}	14.33^{ab}	15^{bcd}	15.67^{abcd}	14.83
IT07K-299-6	15.33^{bc}	14.67^{b}	15.33^{cde}	14.6^{abc}	15.33^{cde}	15.33^{abc}	15.10
SARI-13-17-2	17^{d}	17.33^{c}	16de	14.8^{abc}	16de	16.33^{bcde}	16.24
SARI-1-50-81	16.67^{d}	15.67^{b}	16.33^{ef}	15.67^{bcd}	16.33^{ef}	15.67^{abcd}	16.06
SARI-6-2-9	16^{cd}	15^{b}	16.33^{ef}	15.5^{bc}	16.33^{ef}	16.67^{cde}	15.97
SARI-3-11-80	18.67^{ef}	17.33^{cd}	18^{gh}	16.33^{cd}	18^{gh}	16.67^{cde}	17.5
SARI-2-50-80	19.67^{f}	18.67^{ce}	18.67^{h}	17.43^{de}	18.67^{h}	18.33^{e}	18.57
Kirkhouse-Benga (check)	18.33^{e}	18.67^{e}	17.33^{fg}	18.73^{e}	17.33^{fg}	17.67^{de}	18.01
Mean	15.24	15.67	15.67	15.76	15.67	15.94	
cv%	3.6	4.7	4.5	6.5	4.5	7.5	

Table 6. 100 seed weight of medium maturing lines.

Genotype	100 seed weight (g)						Mean
	Damongo	Nyankpala	Yendi	Wa	Tumu	Manga	
IT06K-137-1	18.33^{de}	18.33^{bc}	17.33^{c}	18.33^{d}	17.33^{d}	15.7^{b}	17.56
IT07K-298-15	15.33^{ab}	16.33^{ab}	15.67^{b}	17.47^{cd}	16.03^{c}	15^{ab}	15.97
IT07K-299-69	13.67^{a}	16^{ab}	15^{b}	15.63^{b}	15.83^{bc}	14.4^{a}	15.09
IT08K-126-19	17.33^{cd}	16.67^{ab}	15.67^{b}	17.5^{cd}	17.1^{d}	17^{c}	16.88
IT09K-456	16.33^{bc}	15^{a}	13.67^{a}	15.67^{b}	15.67^{bc}	14.33^{a}	15.11
IT10K-837-1	19.33^{e}f	18.33^{bc}	17.67^{c}	19.53^{e}	18.03^{e}	17.67^{c}	18.43
IT86D-610	14^{a}	17.33^{ab}	12.67^{a}	15.23^{ab}	15.23^{b}	14^{a}	14.74
IT98K-628	14.67^{ab}	15^{a}	15^{b}	14.33^{a}	14.13^{a}	15^{ab}	14.69
SARI-6-2-6	17.33^{cd}	17^{ab}	15.67^{b}	17.13^{c}	16.9^{d}	17^{c}	16.84
Padituya (check)	21f	20.33^{c}	21.67^{d}	21.87f	19.97f	20.33^{d}	20.86
Mean	16.73	17.03	16	17.27	16.623	16.04	
CV%	5.9	8.6	4	3.5	2.1	3.6	

610 were the most stable among the high-performing genotypes.

Best environments for genotypes

The results of the biplot who-wins-where showed three mega-environments for grain yield of early duration cowpea. This includes Nyankpala and Wa; Tumu and Yendi; and Damongo and Manga with the best average performers for these environments being IT99K-1122, SARI-13-17-2, and SARI-2-50-80, respectively (Figure 2). Two mega environments were identified for grain yield of medium duration cowpea (Figure 4). Wa, Nyankpala, Manga, and Damongo formed one mega environment with IT86D-610 being the best performing line. Yendi and Tumu were grouped into another mega environment with Padituya being the best performing variety.

Four environments, namely, Nyankpala, Yendi, Damongo, and Manga had relatively longer vectors and therefore discriminated the grain yield of early duration cowpea. Tumu has the least vector length, followed by Wa. Vector angles between Nyankpala and Wa, Tumu and Yendi, Damongo and Manga were less than 90º and are said to be correlated with each other (Figure 5a).

Five out of six environments well discriminated the grain yield of the medium duration cowpea (Figure 5b). Wa had the least vector length, all the other environments had longer vectors. Vectors angles between Nyankpala, Wa, Manga, and Damongo is less than 90º and can be said to be correlated together. Vector angle between Tumu and Yendi are also less than 90º and are also

Table 7. Grain yield of early maturing cowpea advanced breeding lines in six locations.

Genotype	Grain yield (t/ha)						
	Damongo	Nyankpala	Yendi	Wa	Tumu	Manga	Mean
IT99K-1122	2.23 ^{cd}	1.9 ^{e}	1.53 ^{bc}	1.9 ^{cde}	1.5 ^{bc}	2 ^{bc}	1.84
IT07K-298-15	2.23 ^{cd}	1.37 ^{bcd}	1.57 ^{bc}	1.23 ^{a}	1.33 ^{bc}	1.77 ^{b}	1.58
SARI-1-3-90	1.63 ^{b}	1.2 ^{bc}	1.47 ^{bc}	1.56 ^{b}	1.1 ^{ab}	1.53 ^{ab}	1.42
SARI-5-5-5	1.07 ^{a}	1 ^{ab}	0.53 ^{a}	1.2 ^{a}	0.7 ^{a}	1.03 ^{a}	0.92
IT07K-299-6	2.4 ^{d}	1.37 ^{bcd}	1.57 ^{bc}	2.01 ^{de}	1.47 ^{bc}	1.67 ^{b}	1.75
SARI-13-17-2	2.2 ^{cd}	1.57 ^{cde}	2.13 ^{d}	2.07 ^{e}	1.37 ^{bc}	2.1 ^{bc}	1.91
SARI-1-50-81	2.13 ^{cd}	0.73 ^{a}	1.13 ^{b}	1.55 ^{b}	1.13 ^{ab}	1.53 ^{ab}	1.37
SARI-6-2-9	2.23 ^{cd}	1.03 ^{ab}	1.33 ^{bc}	1.53 ^{b}	1.17 ^{ab}	1.47 ^{ab}	1.46
SARI-3-11-80	2.4 ^{d}	1.47 ^{bcde}	1.73 ^{cd}	1.81 ^{cd}	1.4 ^{bc}	1.73 ^{b}	1.76
SARI-2-50-80	2.43 ^{d}	1.73 ^{de}	1.67 ^{cd}	1.72 ^{bc}	1.47 ^{bc}	2.5 ^{c}	1.92
Kirkhouse-Benga (check)	2 ^{c}	1.33 ^{bcd}	1.7 ^{cd}	1.57 ^{b}	1.83 ^{c}	1.7 ^{b}	1.69
Mean	2.09	1.34	1.49	1.65	1.32	1.73	
cv%	7.4	18.3	17.8	7.2	19.9	19.2	

CV: Coefficient of variation. Genotypes with different letters are significantly different at P<0.05.

Table 8. Grain yield of medium maturing cowpea advanced breeding lines in six locations.

Cowpea genotype	Grain yield (t/ha)						Mean
	Damongo	Nyankpala	Yendi	Wa	Tumu	Manga	
IT09K-456	1.2^{a}	0.77^{a}	1.4^{b}	1.3^{a}	1.28^{c}	0.9^{a}	1.14
IT07K-298-15	1.4^{b}	1.84^{d}	1.13^{a}	1.44^{ab}	0.87^{a}	1.23^{b}	1.32
IT06K-137-1	1.63^{c}	1.36^{b}	1.38^{b}	1.83^{de}	1.15^{b}	1.45^{c}	1.47
IT98K-628	1.68^{c}	1.53^{c}	1.36^{b}	1.58^{bc}	0.85^{a}	1.45^{c}	1.41
IT08K-126-19	1.98^{d}	2.0^{de}	2.02^{cd}	1.26^{a}	1.77^{e}	1.18^{b}	1.7
IT10K-837-1	2.12^{e}	1.92^{d}	2.17^{d}	1.87^{de}	1.53^{d}	2.04^{e}	1.94
IT86D-610	2.27^{f}	2.14^{e}	2.07^{cd}	1.69^{cd}	1.92^{f}	2.27^{f}	2.06
SARI-6-2-6	2.27^{ef}	1.9^{d}	1.49^{b}	1.74^{cde}	1.7^{e}	1.85^{d}	1.82
IT07K-299-69	2.34^{f}	1.23^{b}	1.92^{c}	1.62^{bc}	1.47^{d}	1.95^{de}	1.75
Padituya (check)	2.20^{ef}	1.93^{d}	2.17^{d}	1.94^{e}	2.2g	2.07^{e}	2.09
Mean	1.91	1.66	1.71	1.628	1.47	1.64	
CV%	4.2	5.5	5.6	6.9	4.9	5.3	

CV: Coefficient of variation. Genotypes with different letters are significantly different at P<0.05.

highly correlated.

DISCUSSION

Field evaluation of advanced breeding materials across multiple environments is one way of identifying high performing genotypes. Parameters such as days to maturity, 100 seed weight, and grain yield are very important in selecting cowpea genotypes for release. In this study, early and medium duration cowpea genotypes were evaluated across six environments in Northern Ghana. The materials evaluated exhibited wide phenotypic variability in terms of maturity, 100 seed weight, and grain yield. These may be due to the inherent genetic variability and environmental differences. SARI-2-50-80, combines earliness with large seed size and high grain yield among the early maturing lines evaluated as it outperformed the check variety, Kirkhouse Benga. Some other genotypes were as early as SARI-2-50-80 but did not give comparable yields. This may be due to inherent genetic differences. In a similar study by Owusu et al. (2020), where the check variety was Bawutawuta, SARI-2-50-80 outperformed the check in terms of grain yield and seed size. These results indicate their utility in breeding programs.

All the medium maturing lines evaluated matured earlier than Padituya (check), but at the same time, they

Table 9. ANOVA results for grain yield of early maturing genotypes.

Source of variation	d.f.	s.s.	s.s. (%)	m.s.	v.r.	F pr.
Environment	5	27.05132	45.91128	5.41026	97.26	<0.001
Environment.Rep	12	2.09213	3.550746	0.17434	3.13	<0.001
Genotype	10	15.16534	25.73849	1.51653	27.26	<0.001
Genotype.Environment	50	7.93652	13.4698	0.15873	2.85	<0.001
Residual	120	6.67555		0.05563		
Total	197	58.92086				

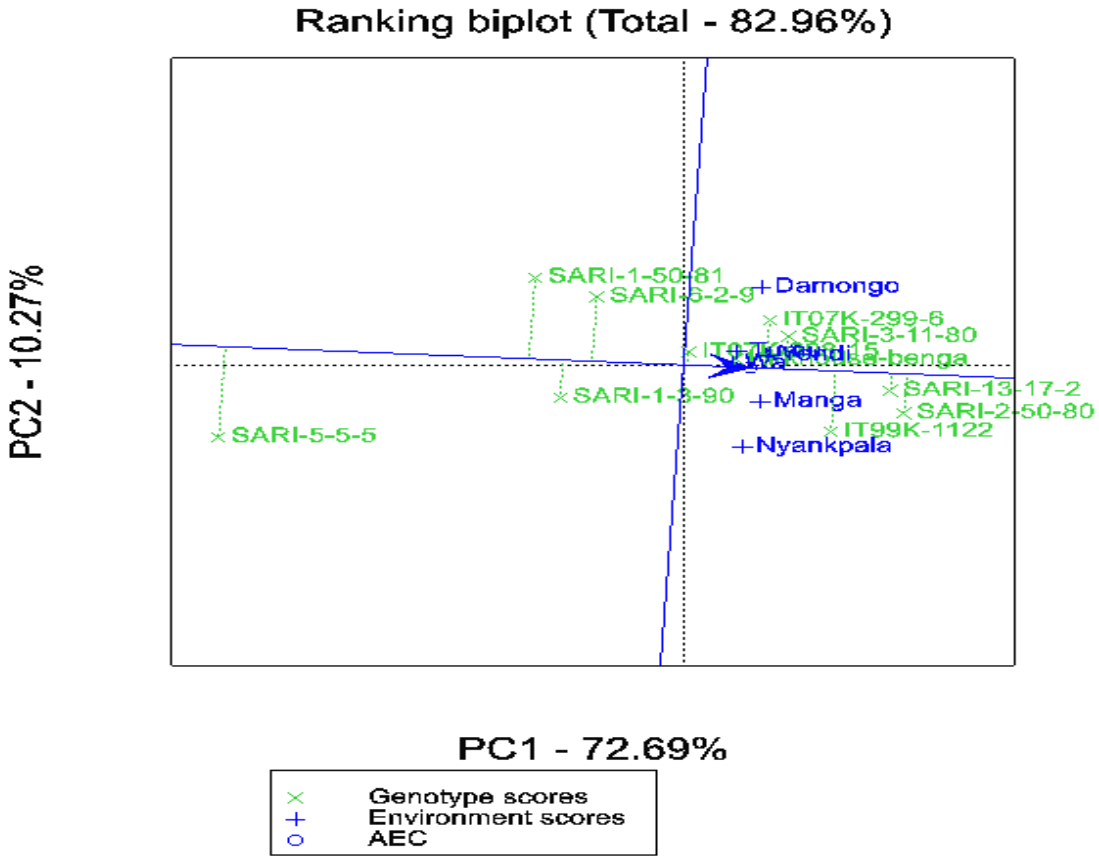

Figure 1. Grain yield and stability of early maturing cowpea genotypes across six environments.

had relatively lower seed weight and grain yield than the check. This could mean the longer the maturity period the larger the grain size and the higher the yield. Despite these, some medium maturing lines IT86D-610 (2.06 t/ha), IT10K-837-1 (1.94 t/ha), and SARI-6-2-6 (1.82 t/ha) had yields comparable to Padituya (2.09 t/ha), the check variety. Among these, IT10K-837-1 had the highest 100 seed weight (18.43 g), white seeded, and will be more preferred by consumers in Ghana. This line, therefore, combines earliness with large seed size and grain yield and will suit the changing climatic conditions than Padituya. High yields were for IT86D-610 and IT10K-837-1 were also reported in a study by Owusu et al. (2020).

The efficiency of genetic gain is reduced through selection by the presence of GxE interaction when genotypes are compared across a range of environments. When there are inconsistencies in the performance of a genotype from one environment to another environment, GxE is said to have occurred. However, if a genotype's performance is consistent across the environments it is said to be stable and therefore shows a general adaptation.

GGE-biplot is an effective tool in evaluating genotypes based on their means and stability (Yan, 2001).

Table 10. ANOVA results of grain yield for medium maturing genotypes.

Source of variation	d.f.	s.s.	s.s (%)	m.s.	v.r.	F pr.
Environment	5	3.000177	10.02452	0.600035	73.54	<0.001
Environment.Rep	12	0.074486	0.248881	0.006207	0.76	0.689
Genotype	9	16.95962	56.66733	1.884402	230.96	<0.001
Genotype.Environment	45	9.012928	30.11497	0.200287	24.55	<0.001
Residual	108	0.881183		0.008159		
Total	179	29.92839		0.167198		

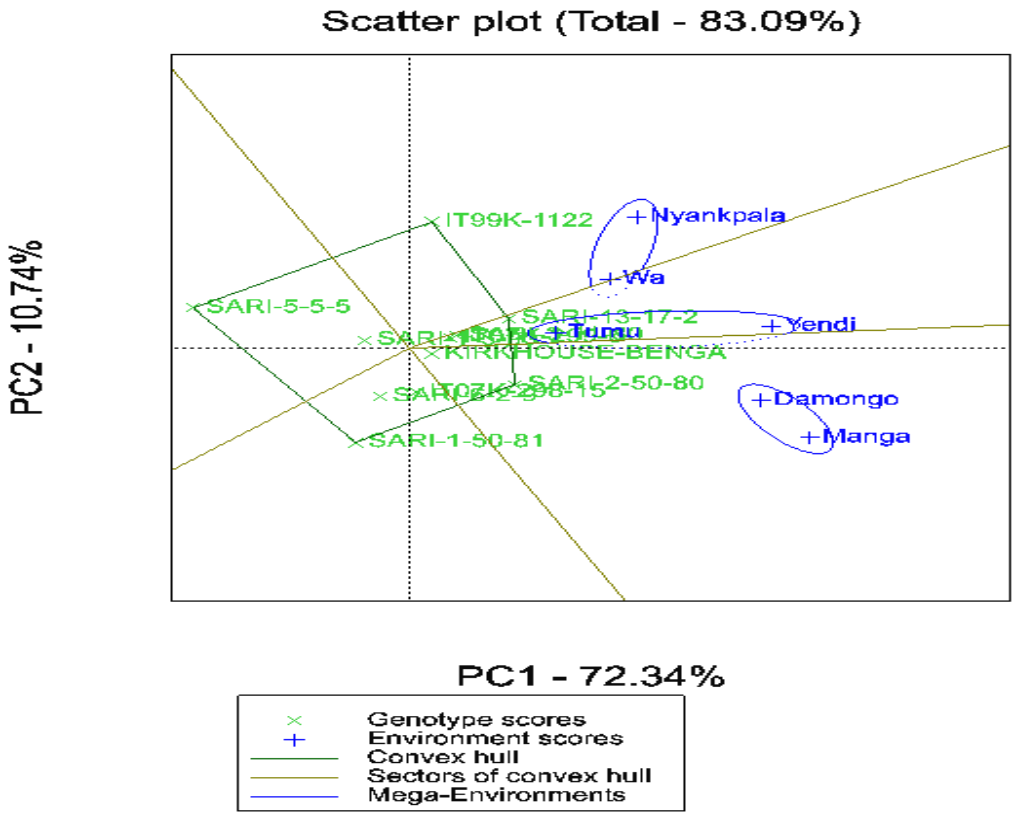

Figure 2. A which-won-where graph and mega environment grouping of early maturing cowpea genotypes.

A type is said to have lower stability if it is greatly projected on PC1 and higher stability if it is closer to the PC2 axis. A high and positive PC score means a higher average value; and a high negative PC score indicates a lower average value (Yan et al., 2000). Thus, SARI-2-50-80, SARI-13-17-2, IT99K-1122, SARI-3-11-80, and IT07K-299-6 have a high yield and stable performance across the six test environments and out-performed the check variety (Figure 1 and Table 7). Also, IT86D-610, IT10K-837-1, and SARI-6-2-6 had high yield and stable performance; but IT10K-837-1 was the most stable (Figure 3). The stability of IT86D-610, IT10-837-1 and SARI-2-50-80 is contrary to what was reported in a recent study in Ghana (Owusu et al., 2020). Desirable genotypes have been selected using this method (Sousa et al., 2018).

The GGE-Biplot method facilitates the selection of superior genotypes by grouping environments into mega-environments and determining which genotypes performed best in these mega-environments. Positively correlated environments in each sector of the polygons are grouped into mega-environments (Yan et al., 2000). This makes it possible to explore GxE, with greater accuracy in identifying mega-environments and selecting stable and adapted genotypes for the environments (Silva and Benin, 2012). The mega-environment

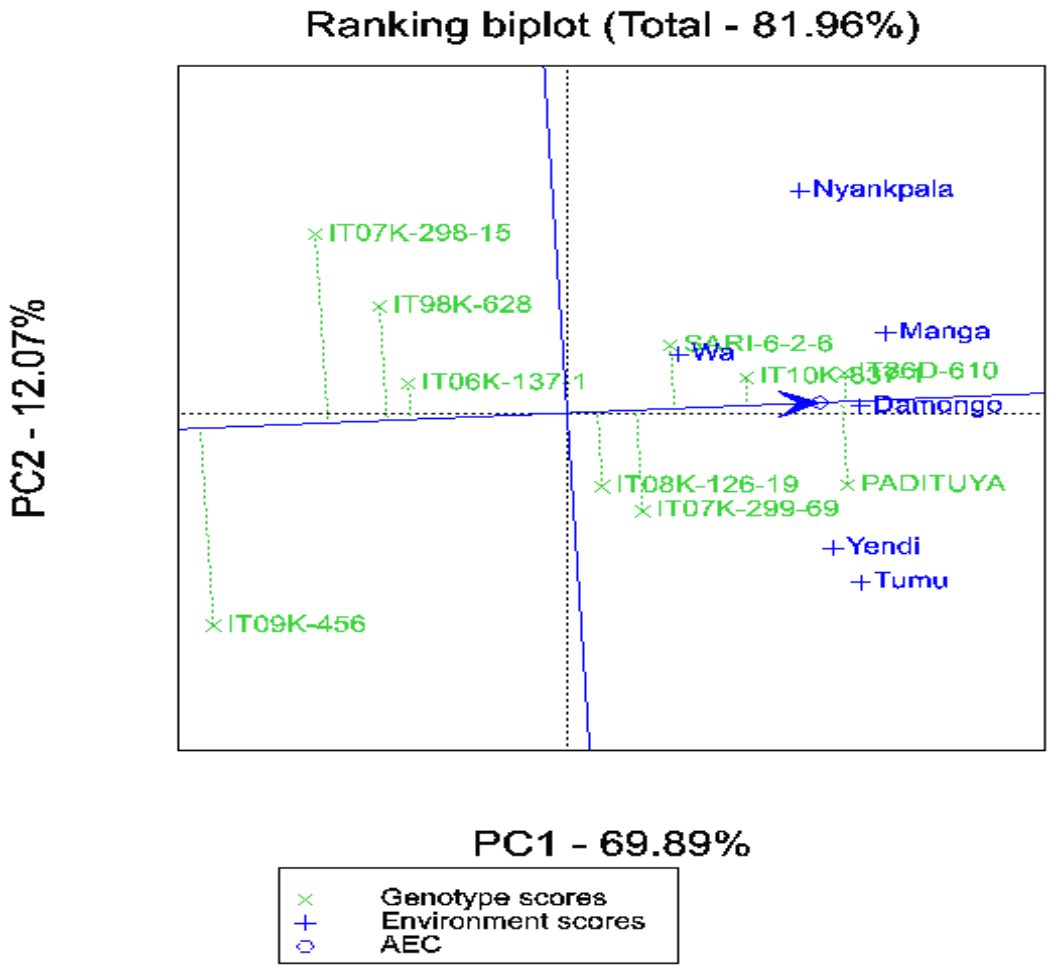

Figure 3. Grain yield and stability of medium maturing cowpea genotypes across six environments.

groupings were discriminative of the grain yield of cowpea. Environments within each mega- environment can be said to be correlated with each other as the angle between the vectors as shown in Figure 5 are less than 90°. The lower the angle between two vectors than 90º the better the correlation (Yan and Tinker, 2006). Similar occurrences of positive correlation of encompassing environments have been reported in other studies (Dos Santos et al., 2016; Sousa et al., 2018). The length of the vector can also be used to tell which of the environments are most discriminating of the target trait. Apart from Tumu which was the least discriminating environment for grain yield of early duration cowpea (Figure 5a) and Wa which was the least discriminating environment for medium duration cowpea (Figure 5b), the rest of the environments well discriminated the genotypes and can be used to select superior genotypes. Tumu being the least performing environment for the early maturing cowpea could be attributed to it receiving the highest mean rainfall. Wa being the least performing environment among the medium duration, genotypes could also be attributed to the region experiencing the least rainfall during the growing season of the crop (Table 2). There is, therefore, evidence that cowpea with different maturity groups differs in their response to water stress. Too much rain will have a more pronounced effect on early duration cowpea than medium duration cowpea and too little rainfall will have a negative impact on grain yield of medium duration cowpea than early duration cowpea.

Some genotypes originated from the vertices of the polygon but contain no clustered environment. Examples of such genotypes are SARI-5-5-5 and SARI-1-50-81 (Figure 2), IT07K-298-15, IT09K-456, IT98K-628, and IT06K-137-1 (Figure 4). These genotypes can be said to be unfavorable to the test environments due to their low grain yields and are therefore not recommended. This similar situation has been reported by other authors (Dos Santos et al., 2016; Karimizadeh et al., 2013). The identification of suitable genotypes and an ideal environment using the GGE-Biplot method could help future evaluation studies as genotypes will be tested in environments with greater GxE.

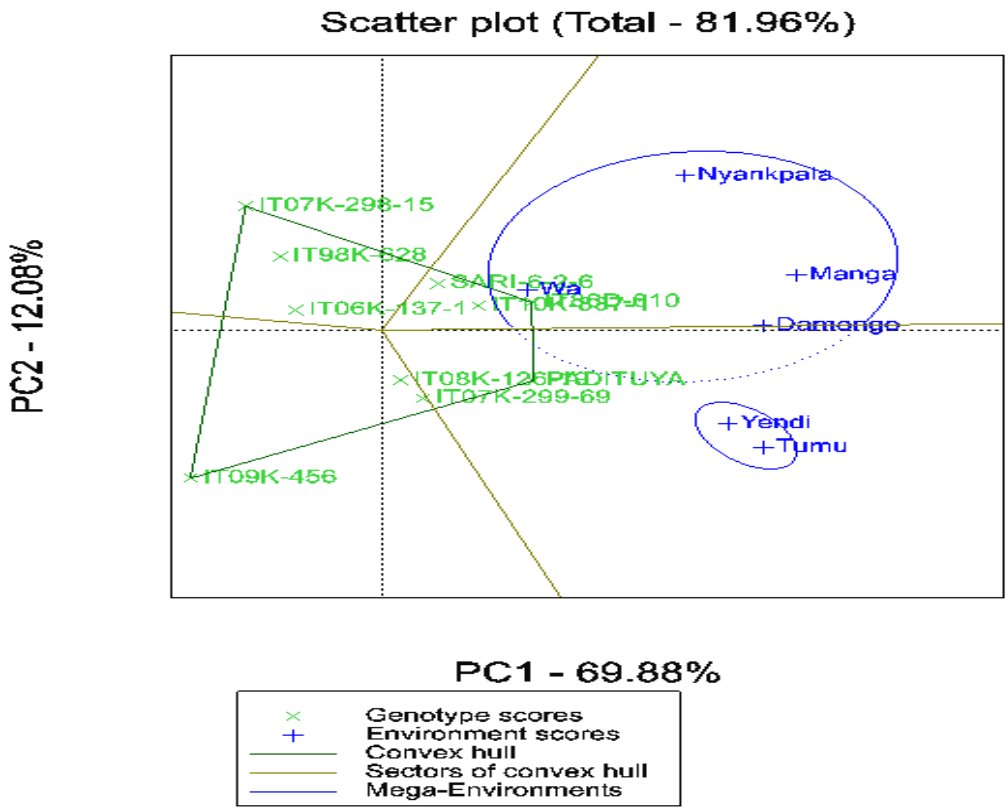

Figure 4. A which won graph and mega environments classification of medium maturing cowpea genotype.

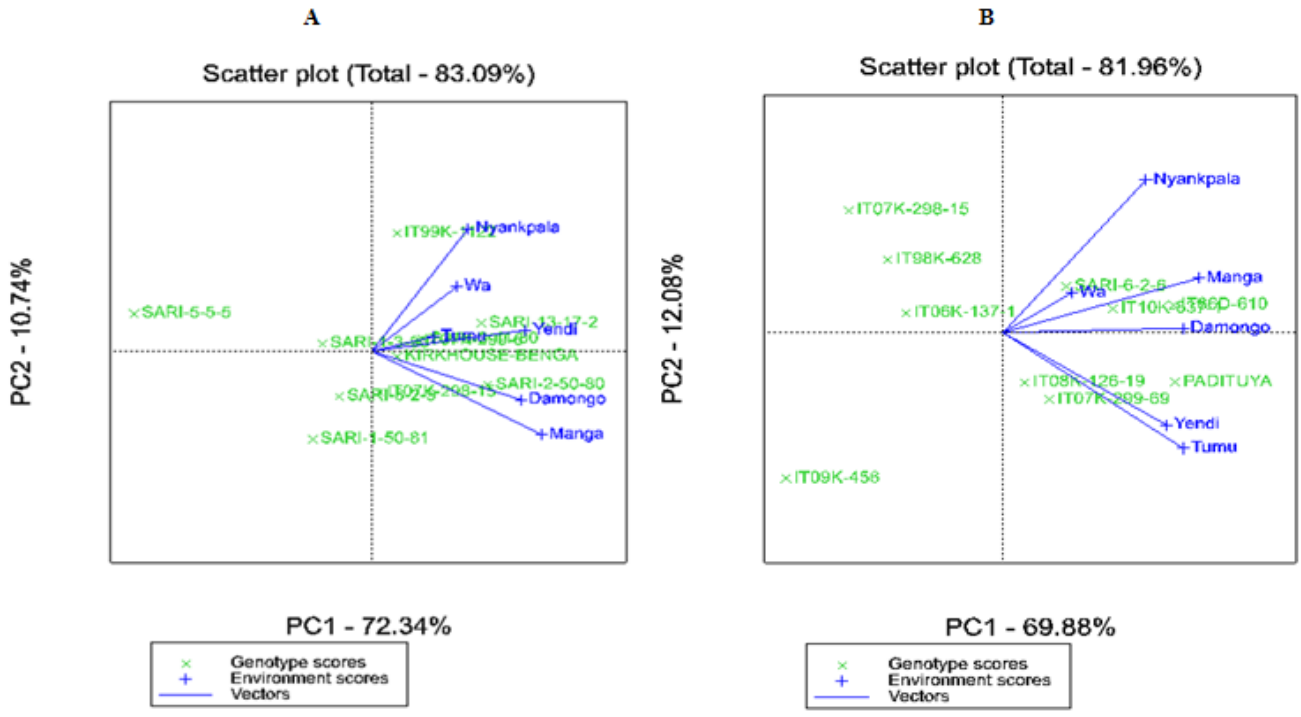

Figure 5. Discrimination and representativeness of grain yield of early duration cowpea genotypes (left) and medium duration cowpea genotypes (right).

Conclusion

Grain yield has consistently been high for Damongo and Yendi for the early maturing and medium maturity cowpea genotypes that were evaluated. For early maturing lines, Tumu was the least performing environment; while Wa was a poor performing environment for medium maturing lines. Notwithstanding that, high yielding and stable early maturing lines (such as SARI-2-50-80, SARI-13-17-2, IT99K-1122, SARI-3-11-80, and IT07K-299-6) outperformed the check (Kirkhouse Benga); and SARI-2-50-80 had the highest 100 seed weight. Medium maturing lines (such as SARI-6-2-6, IT86D-610, and IT10K-837-1) could also be selected as candidate lines, because they matured earlier than the check and have yields comparable to the check. In terms of 100 seed weight, however, only IT10K-837-1 came close to the check. These selected genotypes can further be evaluated on-farms and released as varieties in the future.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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The multi-level table and circular diagnostic chart as alternative taxonomic key formats for plant identification

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Correct identification of plants is a prerequisite to achieving desirable results in health care delivery, sustainable food production and housing, forest resources management and environmental protection. However, many of the paper-based/printable taxonomic key formats available to the taxonomist for this important responsibility are fraught with inadequacies some of which include fixed sequence of plant identification steps, non- or hardly-susceptible to computerisation, lack of provision for confirmation of suspected plant identity and indeterminable character states, and tedious construction and navigation procedures. This paper with the aim of making the practice of plant taxonomy more attractive, less laborious and dreaded, proposes two new key formats with highlights of their design/features, construction procedures and usage. These alternative key formats, with varying capacities to circumvent some of the enumerated challenges are multi-level table of identification and multi-layer circular diagnostic chart. The status of each of the proposed key formats is discussed with reference to the inadequacies observed in the dichotomous key format with which most taxonomists are familiar. Based on their structural features and functionality attributes, it is conclusive that the two alternative key formats constitute useful templates upon which reliable plant diagnostic tools can be based.

Key words: Automated plant identification, computerised key, diagnostic key, dichotomous key, multi-access key, plant identification, single-access key, taxonomic key.

INTRODUCTION

The basic terminology in systematics is 'taxon', a formal category of living things, or a 'taxonomic group' recognised by having certain characteristics in common which we take as evidence of genetic relationship, and possessed some degree of objective reality (Rickett, 1958). It is a group of one or more individuals, or of lower taxa judged sufficiently similar to each other to be treated together formally as a single evolutionary or informational unit at a particular level in the taxonomic hierarchy, and sufficiently different from other groups of the same rank to be treated separately from them (Radford et al., 1974). Going by these regular definitions, each taxon (except the highest), such as a species belongs to one and only one taxon of the next higher rank, such as a genus, implying each individual belongs to exactly one species (and has one name) in any particular taxonomic

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treatment of its group. Taxonomists frequently present organised written descriptions of the characteristics of similar taxa such as species or genera, etc., to facilitate identification or recognition of unknown organisms. These organised descriptions are referred to as diagnostic keys, which come in different formats and styles such as in Hopkins and Stanfield (1966), Lowe and Stanfield (1974), Payne et al. (1974), Payne and Preece (1980), Jones et al. (1998) and Javatpoint (2018), each with its merits and demerits.

Outside the understanding of the terminology from the systematics (strict) point of view, and for all practical purposes, it is useful to operationally define 'taxon' as one or more objects recognized by sharing certain characteristics, and representing a group or category, the members of which may or may not be related taxonomically. Such objects will include tangible items as plant specimens of one or more taxonomic groups (e.g. species, sub-genus, genus, etc.), plant organs (e.g. root, flower, seed, fruit, etc.), or intangibles and other forms of categorisation such as plant diseases, colours, sounds, odours and so on (Amante and Norton, 2003). So, in the context of this study, taxon is more widely viewed beyond its conventional usage as 'taxonomic group' or assemblage of plants or animals that are genetically related, or so related to the best of our knowledge (Rickett, 1958) to mean a unit of classification (taxonomic or otherwise) of objects (tangible or intangible), recognised by a set of features that distinguish (or diagnose) the group from such other groups. This position equally recognises both the relationship of inclusion between levels and of complementarity within levels as aptly described by Price (1967) regarding the objects classified into groups under groups. That is, given a large group of taxa (or objects), and based on the relationships among the objects, recursive classification or compartmentalisation of the group members into taxa of lower categories/ranks is possible until the entire group is resolved into the smallest manageable clusters of taxa, or each taxon as it were.

Correct identification of plants is important in health care delivery (Upton and Romm, 2010), sustainable food production (Amante and Norton, 2003) and housing, criminal justice (Bock and Norris, 2016), forest resources management and environmental protection. Medicinal plants misidentification and misrepresentation are two known root causes of herb adulteration or substitution, which in turn, is the basic cause of serious health problems to consumers of herbal medicinal products (Panter et al., 2014), and a motivation for bad publicity and legal burdens sometimes faced by the pharmaceutical industry (Dukes, 2006). For these reasons, there is a huge responsibility on the shoulders of plant taxonomists, who, unfortunately, often have to contend with a number of challenges including the intricate nature and complexity of plant life, and variability in their characteristics (Tilling, 1987), perceived tediousness of taxonomic practices along with obsolete tools for identification (Stagg and Donkin, 2013) and the attendant declining interest in botany (Drea, 2011), especially plant taxonomy (The Conservation, 2020). The number of botany specialists is reducing by the day. In the United States of America, the number of undergraduate degrees earned in botany is said to have decreased by 50% since the late 1980s (Bidwell, 2013), a trend that should never be taken lightly. The aforementioned challenges therefore formed the basis for conceptualising this study to make a contribution towards ameliorating the declining interest in plant taxonomy.

A taxonomic key is derived from a data matrix of a given number of 'objects', and it is usually possible to contrive a large number of different key formats for one set of objects such as plant species, but the keys will vary in their usability (Pankhurst, 1970). Dichotomous key is widely acknowledged as the most popular type of identification key (Sinh et al., 2017), and had been a clever means of organising taxonomic information before the age of computers (Godfray et al., 2007). The use of this key format is known to have contributed to increasing the quality and durability of knowledge of plant classification acquired in comparison to traditional teaching techniques (Andic et al., 2019) and an established method for teaching plant identification skills (Stagg and Donkin, 2013). However, a number of seemingly demoralising weaknesses are associated with dichotomous key format, including: being tedious to construct (Lobanov, 2003), having fixed point of entry and daunting path of navigation (Hagedorn et al., 2010), the problem of unanswerable couplet (Rambold and Martellos, 2010), being unusable for confirmation of suspected identity, and non-readily amenable to automation (Yin et al., 2016). So, invention of new key formats shall continue to be a welcome development in taxonomy. In providing a way out of the challenges enumerated earlier, the present study aimed at making the practice of plant taxonomy more attractive, less laborious and dreaded, and so, the objectives are to propose two new taxonomic key formats with highlights of their features, construction procedures and usage that should possibly make them desirable, either as alternative or complementary tools for plant identification.

MATERIALS AND METHODS

Adoption of heuristic approach to addressing the weaknesses in dichotomous key format

The first step taken in actualising the objectives of this study was to align with the thoughts of Pankhurst (1970) on the two complementary problems in taxonomy, which are still valid till date. Firstly, given a set of objects (e.g., plants), examine their characteristics in order to find a classification, that is, group the objects into subsets (or taxa), and assign names to the subsets; and secondly, given a classification and an object, identify that object. In other words, given a list of the characteristics of named subsets which are known to exist, and an additional object, decide

which subset the object belongs (that is, recognise it, or find its name). Accepting that the taxonomists' diagnostic key was an important tool in the process of identification, the format and styles of the frequently used single-access diagnostic keys along with the challenges associated with their features, construction and application were critically examined (Walter and Winterton, 2007). In an effort to address some of these inadequacies, consideration was also given to selection criteria for construction of efficient diagnostic keys (Payne, 1981, 1988). Information obtained from the steps highlighted earlier were integrated into a thought to develop two alternative key formats, namely: the multi-level table and multi-layer circular diagnostic chart, each with far reaching desirable qualities in terms of design/features, construction, navigation efficiency and possibility of automation.

Data procurement for purpose of illustration

Wood anatomical data on five medicinal herbs marketed as plant roots in Ogbomoso township, south western Nigeria were sourced for the purpose of illustration from the 2019 compilation of unpublished results at the medicinal plants research laboratory in the Department of Pure and Applied Biology, Ladoke Akintola University of Technology, Ogbomoso, Nigeria. The species are *Aristolochia ringens, Calliandra haematocephala, Parquetina nigrescens, Sarcocephalus latifolius* and *Zanthoxylum zanthoxyloides*. The data items were obtained in accordance with the standard procedures: tissue sectioning/maceration (Schoch et al., 2004), staining, dehydration (Ogunkunle and Oladele, 2014), mounting (Arx et al., 2016), and microscopic observations (de Parnia and Miller, 1991), while the terminology and descriptions of observed features followed those of the International Association of Wood Anatomists (IAWA Committee, 1989). Twenty-five characters, consisting of ten qualitative (Table 1) and fifteen quantitative features (Table 2), all of which were diagnostic of the species were compiled. Staining was done in 1% alcoholic safranin, mounting was carried out in Canada balsam and observations made using Olympus biological microscope CH20i Model with binocular facility. Quantitative characters were considered diagnostic of the species only if the means of the replicated values were statistically significant at $\alpha = 5$ following One-Way Analysis of Variance, and Duncan multiple range classification of the means (Landau and Everitt, 2004).

Conceptualisation of procedures for constructing and applying the two alternative key formats

For a given number of taxa with certain observable characters, the features as well as the procedures for constructing and navigating a multi-level table of identification on one hand, and the multi-layer circular diagnostic chart on the other hand were heuristically conceptualised as recursive or repetitive process of 'divide and conquer' algorithms (Hagedorn et al., 2010). Further to the achievement of the objectives of this study, the algorithm in each case was systematically executed, and is here, being proposed as a number of logical steps.

Design and statement of the features of multi-level table of identification

The multi-level table of identification was conceived as a diagnostic tool having features similar to the conventional table of results displaying characters and plant taxa in columns and rows. Unlike in a conventional table of results, the characters in the key are stated either as unit characters or combinations of two or more features observable in either a taxon or in clusters of taxa. Also unlike in the conventional table of results, the characters are arranged in tiers or levels: primary, secondary, tertiary, quaternary, quinary, senary, septenary, octonary, nonary denary, etc., representing first, second, third, …tenth level, respectively; indicating the levels of successive classification/compartmentalisation of the plant group using the characters. Characters of the first tier/level (that is, of the primary classification of the taxa) are listed in the first row on top of the table while the next row is used to display all the names of the taxa (that is, a cluster of plants) as defined by each character or characters in the first/primary level; characters of the second tier/level are listed in the third row of the table while the fourth row is used to display the clusters of taxa as defined by each character or characters in the second level, etc., thus rows of lists of characters alternate with rows of lists of taxa, and at the end of each successive level, the number of taxa in a cluster progressively reduces down the line.

Construction procedure of multi-level table of identification

The essential activities in building the multi-level table of identification consist of the following steps:

(1) constructing a conventional table of character comparison for the plant group under study, displaying characters in rows and the taxa in columns as in Tables 1 and 2;
(2) selecting one, or few characters as character combinations, which is/are considered as being of primary importance for classifying the group under study into few clusters of taxa that may be mutually or non-mutually exclusive, and stating those characters in the first row of the table; meanwhile, maximum number of clusters should be four to avoid the key being unwieldy;
(3) enumerating the taxa in each of the few clusters in the next row of the table as defined by the characters or character combinations in the previous row;
(4) considering the clusters of taxa obtained from primary level of classification, one at a time, selecting another character or character combination as being of secondary importance to further circumscribe the taxa in the primary cluster into smaller clusters (again, mutually-exclusive or not), and enumerating such taxa as a subset of that cluster in the next row;
(5) considering again, each cluster of taxa obtained from second tier of characters, selecting another, or few characters as being of tertiary importance to further circumscribe the taxa therein and enumerating such taxa as a subset of the secondary cluster in the next row;
(6) recursively selecting one or few characters (of quaternary, quinary, senary, *etc*., importance) for subsequent circumscription of the taxa as earlier described until every taxon in each cluster of taxa has been sorted/keyed out separately towards the bottom of their respective columns in the table;
(7) noting that if mutually exclusive clusters of taxa were possible and achievable throughout the recursive classifications of the group in steps '2' to '6', each taxon would be keyed out only once. However, if mutual exclusivity of the clusters was not achievable throughout the entire classifications, that is, if one or more non-mutually exclusive clusters were involved, at least one taxon would be keyed out more than once in the table.

Application of multi-level table of identification

In order to apply the multi- level table of identification, the following steps were formulated, adopted and are herein proposed:

(1) Evaluate the plant material based on the provisions of the first level of character combinations and decide which of the few clusters of taxa in the next row the plant belongs;

Table 1. Some wood anatomical descriptive features in the roots of five medicinal herbs marketed in Ogbomoso, Nigeria.

S/N	Diagnostic features	*ARRI*	*CAHA*	*PANI*	*SALA*	*ZAZA*
	Vessels					
1	Pore type	Diffuse-porous	Diffuse-porous	Ring-porous	Diffuse-porous	Diffuse-porous
2	Occurrence	Solitary***; Radial chains of 2***; Clusters of 3*	Solitary**; Radial chains of 2-7****	Solitary***; Radial chains of 2-3***	Solitary	Solitary*; Radial chains of 2-8****
3	End walls	Oblique	Oblique***; Truncate****	Oblique**; Truncate****	Oblique***; Truncate***	Oblique***; Truncate****
	Wood fibres					
4	Occurrence	Aggregates	Aggregates and diffuse; non-storied	Diffuse; non-storied	Aggregates; non-storied	Aggregates; non-storied
5	Frequency/relative abundance	high***	high***	Low**	Low**	Low**
	Wood parenchyma cells (WPC)					
6	Type of WPC in transverse section	Apotracheal (diffuse)	Apotracheal (diffuse-aggregate)	Apotracheal (diffuse-aggregate)	Apotracheal (diffuse)	Paratracheal (scanty**; vasicentric**; aliform**)
	Wood rays (WRY)					
7	Shape of WRY in Transverse Section	Square*; procumbent****	Square**; procumbent****	Square***; procumbent***	Square**; procumbent****	Procumbent
8	Width of WRY in Tangential Longitudinal Section	Uniseriate	Uniseriate****; biseriate**; multiseriate*	Uniseriate****; biseriate**; ultiseriate*	Uniseriate***; biseriate**; multiseriate**	Biseriate**; multiseriate****
9	Composition of WRY in Tangential Longitudinal Section	Homocellular	Homocellular****; heterocellular*	Homocellular****; heterocellular**	Heterocellular	Homocellular***; heterocellular***
10	General shape of WRY in Tangential Longitudinal Section	Linear	linear**; Mono-convex**; bi-convex****	Mono-convex**; linear****; dumb-bell**	Bi-convex****; dumb-bell*	Mono-convex**; bi-convex****

*ARRI= Aristolochia ringens; CAHA= Calliandra haematocephala; PANI= Parquetina nigrescens; SALA= Sarcocephalus latifolius; ZAZA=Zanthoxylum zanthoxyloides;*****(very frequent/usually observed/very high frequency , that is 60-99% occurrence); ***(frequent/averagely observed/high frequency, that is,40-59% occurrence); **(less frequent/sometimes observed/low frequency, that is, 10-39% occurrence); *(seldom frequent/rarely observed/very low frequency, that is, 1-9% occurrence).
Source: Extract from the 2019 unpublished data compiled at the medicinal plants research laboratory, Ladoke Akintola University of Technology, Ogbomoso, Nigeria.

(2) Once a cluster of taxa has been selected in the row as the likely group in which the plant belongs, the few other clusters of taxa and the columns in which they fall should no longer be considered in subsequent steps;

(3) Re-evaluate the plant on the basis of the provisions of the next level of character combinations to decide which of the clusters of taxa in the next row the plant belongs; and if a decision is difficult or impossible because the plant features do not match the stated character combinations in the row, refrain from making a selection, but proceed to evaluate the unknown plant material on the basis of the characters in the next level;

Table 2. Mean quantitative wood anatomical characteristics of some types of cells in the roots of five medicinal herbs marketed in Ogbomoso, Nigeria.

S/N	Diagnostic features	*ARRI*	*CAHA*	*PANI*	*SALA*	*ZAZA*
	Vessels (VS)					
1	Density/mm^2 in transverse section (TS)	37^{d}±1.20	74^{e}±2.73	10^{b}±0.72	5^{a}±0.22	31^{c}±1.06
2	% frequency of VS shapes in TS	Round(50); Oval(50)	Round(54); Oval(46)	Round(58); Oval(42)	Round(47); Oval(53)	Round(53); Oval(47)
3	VS diameter (µm)	101.97^{b}±5.54	63.81^{a}±1.85	231.94^{e}±11.77	197.03^{d}±10.01	146.86^{c}±3.65
4	VS lumen width (µm)	89.51^{b}±5.34	53.25^{a}±1.76	208.13^{e}±11.45	181.47^{d}±5.48	132.95^{c}±3.73
5	Length of VS member (µm)	194.25^{a} ±7.47	605.01^{c}±39.40	804.52^{d}±52.86	499.78^{b}±24.26	528.04^{bc}±11.84
	Wood fibres (FB)					
6	Density/mm^2	289^{c}±14.76	270^{c}±23.11	63^{a}±5.05	120^{b}±6.03	127^{b}±11.09
7	Diameter (µm)	20.82^{b}±1.07	14.59^{a}±0.81	34.05^{d}±0.92	31.57^{d}±1.15	25.43^{c}±1.09
8	Lumen width (µm)	11.95^{a}±0.99	9.64^{a}±0.67	26.03^{c}±0.95	25.17^{c}±0.89	20.14^{b}±1.09
9	FB length (µm)	514.72^{a}±17.45	856.06^{b}±40.63	604.50^{a}±14.38	1091.58^{c}±67.40	1059.02^{c}±31.31
	Wood parenchyma cells					
10	Density/mm^2 in transverse section	59^{b}±4.05	256^{d}±19.64	197^{c}±11.14	32^{a}±1.57	83^{b}±7.25
	WRY in TLS					
11	Density/mm^2 in TLS	19^{d}±0.48	11^{b}±0.43	20^{d} ± 0.70	13^{c}±0.37	6^{a} ± 0.27
12	Number of cells across WRY width in TLS	1^{a}±0.00 (1-cell)	2^{b}±0.09 (1-3 cells)	2^{b} ± 0.10 (1-3 cells)	2^{b}±0.14 (1-3 cells)	3^{c} ± 0.07 (2-3 cells)
13	WRY thickness in TLS (µm)	12.97^{a}±0.84	29.02^{a}±1.78	31.40^{a}±1.55	55.30^{a}±2.93	86.02^{a}±2.89
14	Number of cells in WRY height (TLS)	6^{a}±0.30	13^{b}±0.89	13^{b} ± 1.63	25^{c}±2.02	34^{d}±1.71
15	WRY height in TLS (µm)	187.39^{a}±6.21	308.57^{ab}±19.93	435.20^{b} ± 49.82	1022.50^{c}±73.98	882.35^{c}±42.87

ARRI= Aristolochia ringens; CAHA= Calliandra haematocephala; PANI= Parquetina nigrescens; SALA= Sarcocephalus latifolius; ZAZA=Zanthoxylum zanthoxyloides. TS= transverse section, TLS= tangential longitudinal section. The mean values of data in a row with the same superscripts are not significantly different (p>=0.05) while those with different superscripts are significantly different (p<0.05). The ranges of number of cells across ray width are shown in parentheses.
Source: Extract from the 2019 unpublished data compiled at the medicinal plants research laboratory, Ladoke Akintola University of Technology, Ogbomoso, Nigeria.

(4) Repeat step 3, with the assurance that the features of the plant being identified align with those in the stated character combinations until a single taxon is achievable, which is taken as the identity of the unknown plant.

Design and statement of the features of multi-layer circular diagnostic chart

The multi-layer circular diagnostic chart was conceived as consisting of two parts: the first part is a number of concentric circles, not drawn to scale, but partitioned by means of radial lines into a number of sectors, and the rings into compartments or 'cells'; each sector representing a taxon in the key as indicated at the circumference; and each compartment is either assigned a number 1 or 2 or 3, *etc*., or is left void/empty as the case may be. The second part of the key is a list of characters or character combinations, assigned numerical values 1, 2, 3, *etc.*, pertaining to the plant taxa in the circular diagram as appropriately indicated in the compartments.

Procedure for constructing a multi-layer circular diagnostic chart

The essential activities/steps in constructing the multi-layer circular diagnostic chart are as follows:

(1) A multi-level table of identification is first constructed as earlier described in steps '1' to '7';
(2) All the characters or character combinations in the table of identification obtained in step '1' above are serially numbered 1, 2, 3, *etc*., from top to bottom, column after column, from the left to the right;
(3) A circle is drawn (not to scale) with a number of concentric rings not less than the number of levels/tiers of characters in the table of identification;
(4) The circle is divided into sectors equal to the number of taxa in the key, thereby partitioning the concentric rings

into ‘cells’, boxes or compartments, and each sector is assigned a name of taxon at the circumference;
(5) The compartments in the circle are labeled using information in the appropriate table of identification by following a number of sub-steps:

(a) Consider the first taxon in the circular diagram, proceed to the first level of characters in the appropriate table, examine one column in the table after the other, and take note of every number (earlier assigned in step 2 above) attached to the character combinations 1, 2, 3, etc., that are contributors to the classification of the taxon being considered;
(b) Transfer those numbers attached to the relevant character combinations for the identification of the taxon into the compartments in the circle in reverse order, that is, the appropriate number at the first (topmost) level in the table is inserted in the last (innermost) compartment for the taxon; the next appropriate number at the next relevant level of characters down the table is inserted in the next upper compartment in the diagram and so on. Once the taxon is observed to have been keyed out in the table, subsequent transfer of numbers attached to character combinations for the taxon should be done in parentheses, indicating such characters to be of secondary importance, that is, although they are, or may be diagnostic of the taxon or a cluster of taxa, such characters need not be observable for taxa recognition to occur;
(c) Consider the next taxon in the diagram and repeat sub-steps ‘a’ and ‘b’;
(d) Remove any extra, entirely empty rings of compartments at the periphery of the circle, and the first part of the key would have been achieved;
(e) Compile, as an attachment to the circular chart, a list of all the characters/character combinations in the chronological order of numbering in the appropriate table, that is, 1, 2, 3, *etc*., thereby achieving the second part of the key.

Application of multi-layer circular diagnostic chart

In order to apply the multi-layer circular diagnostic chart for identification, the user enters the key at the centre and proceeds centrifugally (toward the circumference) by selecting the characters observable on/applicable to the unknown plant specimen at the successive rings of compartments. This process progressively narrows down on the choice of the possible identities (that is, sectors) of the unknown specimen until only one choice is achievable, which represents its identity.

Illustrative execution of the proposed procedures

The proposed steps for constructing and using the two new key formats developed from this study were executed using the wood anatomical data in Tables 1 and 2 to obtain two single-entry diagnostic keys usable for identifying five medicinal herbs sold as roots in Ogbomoso, Nigeria.

RESULTS

Tables 3 and 4 and Figures 1 and 2 are the results obtained following execution of the proposed procedures for constructing and using two alternative key formats. Construction of a multi-level table of identification (Tables 3 and 4) is a major step in making the multi-layer circular diagnostic chart (Figures 1 and 2), and both of these types of key are single-access devices. While Table 3 and Figure 1 were products of classifications of the five taxa into two mutually exclusive groups, Table 4 and Figure 2 are the results of classification of the taxa into three non-mutually exclusive groups.

DISCUSSION

Narrowing the lines of demarcation between the major components of taxonomy

In constructing a taxonomic key, an important suggestion by Radford et al. (1974) is to “identify all groups to be included in the key and prepare a description of each taxon”. This position has not changed till date. Morse (1971) explained that in preparing a key, one usually divides the initial group of taxa by a character couplet into two subgroups, each of which is independently divided into further subgroups, and so forth, until every taxon is distinguished from all others. Again, this procedure is valid till date, more so with a recent consenting publication (Hagedorn et al., 2010) regarding keys as ‘divide and conquer’ search algorithms that reduce the result set recursively until the remainder is small enough to be solved by direct comparison. These submissions point to the fact that although, identification is a separate activity in systematics, but in practice, it involves the other three major components of taxonomy namely classification, description and nomenclature (Radford et al., 1974). This scenario played out in the course of this study because the three activities were brought to bear in developing two new key formats for the purpose of identification.

Examining beliefs and opinions on identification keys

One implication of adopting the above-stated suggestion by Radford et al. (1974) is that the author of a key should or will not, ordinarily require same key to identify any of the taxa included in it. *Ab-initio,* he identified all the taxa and created the key. If one views this scenario on the surface, along with the general belief that the use of identification keys requires intensive training and experience, which only few individuals do have (Waldchen et al., 2018), one will agree with Lobanov (2003) that “keys are compiled by those who do not need them for those who cannot use them”. However, on a closer examination of the two pillars on which Lobanov’s conjecture reclines, one would tend to disagree with him. Firstly, a taxonomist is not expected to have worked on all plant groups, nor is he obliged to keep in mind separately the names and diagnostic features of those taxa included in all the keys he has authored. Therefore, as a specialist, he not only uses keys created by his

Table 3. Type I multi-level table of identification (with two mutually exclusive groups of taxa) for diagnosing some medicinal herbs sold as roots in Ogbomoso Nigeria based on their wood anatomical features.

Tier/level of character	Diagnostic character combinations and the plant taxa	
First (primary) level	Both uniseriate and biseriate rays are observable in the wood TLS	Either uniseriate or biseriate rays (but not both) are found in the wood TLS
Taxa	*Calliandra haematocephala, Parquetina nigrescens, Sarcocephalus latifolius*	*Arristolochia ringens, Zanthoxylum zanthoxyloides*
Second (secondary) level	Vessels (TS), occur in solitary units and in radial chains of 2 to 7 axial parenchyma (TS), abundant; 40 to 50% in composition relative to other wood tissues (that is, fibres, vessels, and rays	Rays (TLS), exclusively uniseriate and linear in shape; rays in TLS, relatively short; mean height, less than 200 µm and mean number of cells in height, 6
Taxa	*C. haematocephala, P. nigrescens*	*A. ringens*
Third (tertiary) level	Vessels (TS), occur only in solitary units; fibre, fairly long; mean length, above 1000µm; axial parenchyma (TS), relatively low, being less than 10% in composition relative to other wood tissues (that is, fibres, vessels, and rays)	Rays (TLS), biseriate and multiseriate, the general shape being mono-convex or bi-convex; rays in TLS, relatively tall; mean height, about 900 µm and mean number of cells in height, 34
Taxa	*S. latifolius*	*Z. zanthoxyloides*
Fourth (quaternary) level	Vessels (TS), relatively narrow; mean diameter, less than 65 µm, occurring in solitary units and in radial chains of 2 to 7; fibres (TS), more abundant; about 45% in composition relative to other wood tissues (that is, vessels, axial parenchyma and rays)	-
Taxa	*C. haematocephala*	-
Fifth (quinary) level	Vessels (TS), relatively wide; mean diameter, about 230 µm; occurring in solitary units and in radial chains of 2 to 3; fibres (TS), less abundant; about 13% in composition relative to other wood tissues (that is, vessels, axial parenchyma and rays)	-
Taxa	*Parquetina nigrescens*	-

TS, transverse section; *TLS*, tangential longitudinal section.

colleagues who are specialists in various other plant groups, those written by him are also potential tools for him to carry out identification of the taxa afterwards. Considering the second leg of the argument, it is to be understood that writers and users of keys do not necessarily occupy mutually exclusive positions, so the question of certain persons outside of a clique not being able to use identification keys ought not to be overstressed in the first place.

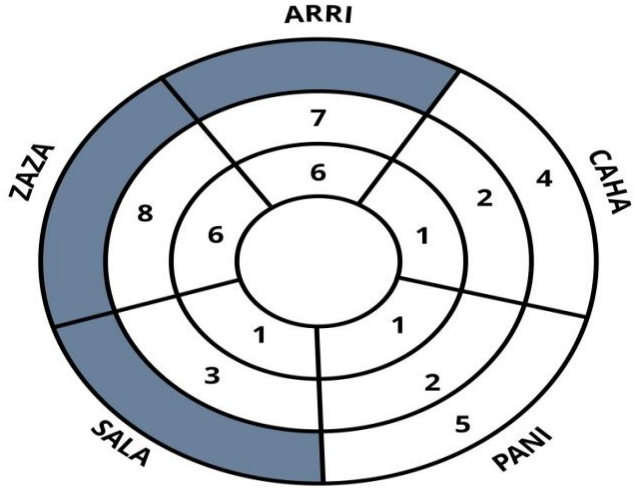
One valid deduction from Lobanov's hypothesis on identification keys is that there is 'plentiful harvest, but few workers to gather it in', and this position is explainable as follows: Identification is the basic prerequisite to understanding biodiversity and ecology (Randler, 2008), and is indispensable in many facets of human life. However, species identification is perceived by many practitioners as onerous task, being comparable with the learning of new words of a new language, while others believe that the act

Table 4. Type II multi-level table of identification (with three non-mutually exclusive groups of taxa) for diagnosing some medicinal herbs sold as roots in Ogbomoso Nigeria based on their wood anatomical features.

Tier/level of character	Diagnostic character combinations and the plant taxa		
First (primary) level	Linear shape type of rays, found in wood TLS, mean fibre length, less than 900 µm	Uniseriate, biseriate and multiseriate rays are all present in wood TLS	Homocellular rays are found in wood TLS
Taxa	*Arristolochia ringens, Calliandra haematocephala, Parquetina nigrescens*	*Calliandra haematocephala, Parquetina nigrescens, Sarcocephalus latifolius*	*Arristolochia ringens, Zanthoxylum zanthoxyloides*
Second (secondary) level	Uniseriate, biseriate and multiseriate rays, present; the cellular composition of rays, both homocellular and heterocellular (not either) are observable; mean density of wood parenchyma, greater than 100/mm^2	Vessels occur as solitary units and as radial chains of 2-7; wood parenchyma are apotracheal of diffuse aggregate type; both homocellular and heterocellular types of rays are found	Wood parenchyma, apotracheal of diffuse type; ray cells in TS, square (isodiametric) and radially procumbent types; rays in TLS, all uniseriate, homocellular in composition and linear in shape; mean vessel diameter, about 100µm; mean density of fibres, about 300/mm^2;and of rays, about 20/mm^2; mean fibre length, about 500 µm
Taxa	*C. haematocephala, P. nigrescens*	*C. haematocephala, P. nigrescens*	*A. ringens*
Third (tertiary) level	Shape of rays, all linear; entirely uniseriate, all homocellular, never heterocellular; mean density of wood parenchyma, about 60/mm^2; mean density of rays, about 20/mm^2	Vessels occur only as solitary units; wood parenchyma are apotracheal of diffuse type; rays are exclusively heterocellular in composition; density of axial parenchyma, about 30/mm^2	Wood parenchyma, paratracheal, of scanty, vasicentric and aliform types; ray cells in TS, all radially procumbent; rays in TLS, biseriate and multiseriate (2-3 cells thick), of homocellular and heterocellular types in composition, and mono-convex and bi-convex in shape; mean vessel diameter in TS, about 150 µm; density of fibres, about 130/mm^2 , and of rays, about 6/mm^2; mean fibre length, greater than 1000µm
Taxa	*A. ringens*	*S. latifolius*	*Z. zanthoxyloides*
Fourth (quaternary) level	Biconvex rays, found along with linear and mono-convex shapes; mean vessel diameter in TS, about 65µm; mean densities of fibres, wood parenchyma and rays, about 270, about 250 and about 10/mm^2 respectively	Dumb-bell shaped (that is, constricted) rays, present; ; mean vessel diameter in TS, about 280 µm; mean densities of fibres, wood parenchyma and rays, about 60, about 200 and about 20 mm^2 respectively	-
Taxa	*C. haematocephala*	*P. nigrescens*	-
Fifth (quinary) level	Dumb-bell shaped (i.e. constricted) rays, found along with linear and mono-convex shapes; mean vessel diameter in TS, about 240 µm; mean densities of fibres, wood parenchyma and rays, about 60, about 200 and about 20/mm^2 respectively	Biconvex-shaped rays, found; mean vessel diameter in TS, about 65 µm; mean densities of fibres, wood parenchyma and rays, about 270, about 260 and about 10 /mm^2 respectively; Mean fibre length greater than 850 µm	-
Taxa	*P. nigrescens*	*C. haematocephala*	-

TS, transverse section; *TLS*, tangential longitudinal section.

is much more difficult and complex (Randler, 2008). Arising from this perception, more and more students and researchers are showing reduced interest in taxonomy. True, there are many non-botany specialists who desire correct identification of plants but are constrained by lack



List of characters

1. Both uniseriate and biseriate rays are observable in the wood TLS
2. Vessels (TS), occur in solitary units and in radial chains of 2 to 7; axial parenchyma (TS), 40 to 50% by volume, relative to other wood tissues (that is, fibres, vessels, and rays)
3. Vessels (TS), occur only in solitary units; axial parenchyma (TS), less than 10% by volume, relative to other wood tissues (that is, fibres, vessels, and rays)
4. Vessels (TS), relatively narrow; mean diameter, less than 65 µm, occurring in solitary units and in radial chains of 2 to 7
5. Vessels (TS), relatively wide; mean diameter, about 230 µm; occurring in solitary units and in radial chains of 2 to 3
6. Either uniseriate or biseriate rays (but not both) are found in the wood TLS
7. Rays (TLS), exclusively uniseriate and linear in shape; relatively short; mean height, less than 200 µm and mean number of cells in height, 6
8. Rays (TLS), biseriate and multiseriate, the general shape being mono-convex or bi-convex; relatively tall; mean height, about 900 µm and mean number of cells in height, 34

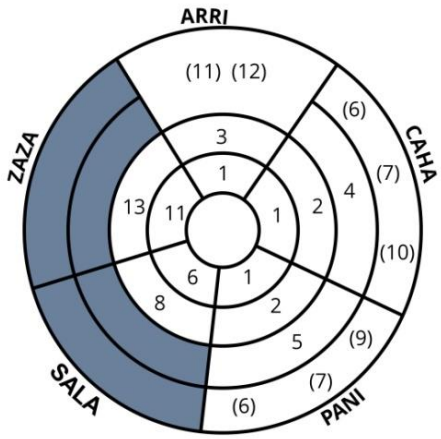
Figure 1. Type I multi- layer circular diagnostic chart for identifying five medicinal herbs sold as roots in Ogbomoso, Nigeria based on their wood anatomical features. *ARRI= Aristolochia ringens; CAHA= Calliandra haematocephala; PANI= Parquetina nigrescens; SALA= Sarcocephalus latifolius; ZAZA=Zanthoxylum zanthoxyloides*;*TS*, transverse section; *TLS*, tangential longitudinal section.

the technical know-how. The up-coming botany specialists who ought to be of assistance to the former category of people in this regard are not better-off either. Furthermore, the current rates of species lost to extinction (IUCN, 2017) necessitate concerted attempts to protect and conserve biodiversity. Species conservation, however, requires species identification skills, but there seems to be perpetual shrink in the number of those interested in this mission (Woodland, 2007).

Comparing the structural and functionality attributes of dichotomous key, multi-level table of identification and circular diagnostic chart

Although enumeration of the features of dichotomous keys is not included in the core objectives of this study, a brief highlight of the challenges associated with the construction and use of this important key format will pave the way for a hands-on approach to comparing the features of the newly proposed key formats with those of the dichotomous format, a single-access identification tool with which key users are most familiar (Sinh et al., 2017). In a dichotomous key, as well as in each of the two newly developed keys in this study, there is only one point of entry, so that there is a single path to be followed by the user. This is a property shared by all single-access key formats, and is accompanied by the problem of 'unanswerable couplet', that is, a user may get stuck and identification will be impossible if a choice cannot be decided at any point (Hagedorn et al., 2010). Also, of concern to users of single access keys are the issues of 'dead ends', and the 'momentary distractions' that can cause a user to forget his or her position in a key (Walter and Winterton, 2007). These situations can arise when a character cannot be observed or adequately scored (e.g. when the feature is in its developmental stage or is season-based, and hence not visible in the specimen) or because the options are not stated clearly enough in the key. The magnitude of the frustration that may set in due to these problems can be intolerable, especially to novice taxonomy students. While the dichotomous key format is notoriously prone to these challenges, such difficulties can be more tolerable with the application of the multi-level table and circular diagnostic chart being proposed since it is much easier to retrace one's steps in case a wrong choice has been made.

It is the belief in certain quarters that construction and use of dichotomous keys are daunting tasks for many students (Jacquemart et al., 2016), and that the format is difficult to automate, if at all amenable to conventional programming techniques (Yin et al., 2016). In contrast, both the construction and navigation procedures of the newly developed single access key formats, that is, multi-level table and circular diagnostic chart have clear-cut algorithms, which can be followed by key makers and users with relative ease. Additionally, these algorithms



List of Characters/Character Combinations

1. Linear shape type of rays, found in wood TLS, mean fibre length, less than 900 µm.
2. Uniseriate, biseriate and multiseriate rays, present; the cellular composition of rays, both homocellular and heterocellular (not either) are observable; mean density of wood parenchyma, greater than 100/mm^2.
3. Shape of rays, all linear; entirely uniseriate, all homocellular, never heterocellular; mean density of wood parenchyma, about 60/mm^2; mean density of rays, about 20/mm^2.
4. Biconvex rays, found along with linear and mono-convex shapes; mean vessel diameter in TS, about 65 µm; mean densities of fibres, wood parenchyma and rays, about 270, about 250 and about 10/mm^2 respectively.
5. Dumb-bell shaped (that is, constricted) rays, found along with linear and mono-convex shapes; mean vessel diameter in TS, about 240 µm; mean densities of fibres, wood parenchyma and rays, about 60, about 200 and about 20/mm^2 respectively.
6. Uniseriate, biseriate and multiseriate rays are all present in wood TLS.
7. Vessels occur as solitary units and as radial chains of 2-7; wood parenchyma are apotracheal of diffuse aggregate type; both homocellular and heterocellular types of rays are found
8. Vessels occur only as solitary units; wood parenchyma are apotracheal of diffuse type; rays are exclusively heterocellular in composition; density of axial parenchyma, about 30/mm^2.
9. Dumb-bell shaped (that is, constricted) rays, present; mean vessel diameter in TS, about 280 µm; mean densities of fibres, wood parenchyma and rays, about 60, about 200 and about 20 mm^2 respectively
10. Biconvex-shaped rays, found; mean vessel diameter in TS, about 65 µm; mean densities of fibres, wood parenchyma and rays, about 270, about 260 and about 10/mm^2 respectively; Mean fibre length greater than 850 µm.
11. Homocellular rays are found in wood TLS.
12. Wood parenchyma, apotracheal of diffuse type; ray cells in TS, square (isodiametric) and radially procumbent types; rays in TLS, all uniseriate, homocellular in composition and linear in shape; mean vessel diameter, about 100 µm; mean density of fibres, about 300/mm^2; and of rays, about 20/mm^2; mean fibre length, about 500 µm.
13. Wood parenchyma, paratracheal, of scanty, vasicentric and aliform types; ray cells in TS, all radially procumbent; rays in TLS, biseriate and multiseriate (2-3 cells thick), of homocellular and heterocellular types in composition, and mono-convex and bi-convex in shape; mean vessel diameter in TS, about 150 µm; density of fibres, about 130/mm^2, and of rays, about 6/mm^2; mean fibre length, greater than 1000 µm.

Figure 2. Type II multi-layer circular diagnostic chart for identifying five medicinal herbs sold as roots in Ogbomoso, Nigeria based on their wood anatomical features. *ARRI= Aristolochia ringens; CAHA= Calliandra haematocephala; PANI= Parquetina nigrescens; SALA= Sarcocephalus latifolius; ZAZA=Zanthoxylum zanthoxyloides*; Characters in parentheses are regarded as being of secondary importance i.e. although they are, or may be diagnostic of a taxon or a cluster of taxa, such characters need not be observable for taxa recognition to occur. *TS*, transverse section; *TLS*, tangential longitudinal section.

can be coded using the desired programming languages; so automation of these activities should not be an intractable problem.

While both the paper-based and computerised dichotomous keys (Tofilski, 2018) are not readily usable for confirmation of suspected identity of a plant, this exercise is manually practicable and electronically achievable using the two newly created single access key formats in this study. If for an unknown plant specimen, one of the taxa included in a key is suspected by a user as its identity, the procedure to confirm or otherwise is first locate the position of the suspected taxon in a key and then work on the key along the established route of identifying the taxon, paying particular attention to only those statements/questions regarding the suspected taxon name, and ensuring that all such (not most) statements are in agreement with the observable features of the specimen in the hand. For the purpose of illustration, if a user in applying the key in Table 3 suspects the identity of a plant to be *C. haematocephala*, confirmation is done by first locating the positions at which the suspected name has been successively keyed out in the column on the left and then the specimen is evaluated based on those characters, that is, the first, second and fourth level of characters where the taxon name occurs. Similarly, if the same taxon is suspected using the key in Figure 1, the specimen is evaluated based on characters 1, 2 and 4 only. So, if given a key, and the assurance that a suspected taxon is included in that key, plant identity confirmation can be explored as a means of assessing leaners' extent of familiarity with the vegetation around them. The learners will not only find the exercise pleasing and refreshing, but also inspiring, much like a game.

Arresting the declining interest in botanical knowledge

Stagg and Donkin (2013) believed that the demise of botanical interest was due to the way botany was taught, if it was taught at all. In order to curtail this undesirable development, appealing plant identification resources are needed, making botany relevant to people's lives is necessary, and correct use of new teaching aids is important (Tilling, 1987). Each of the two newly designed identification key formats in this study has provided answers to these calls. With the likes of multi-level table of identification (Tables 3 and 4) and circular diagnostic chart (Figures 1 and 2) in place, taxonomic key construction and use for identification or identity confirmation turn out to be favourite pastime for specialists and novices alike. In lieu of dichotomous keys, the circular diagnostic chart has additional merit of being adaptable (possibly with enhancements in form of multiple attractive colours) for use at the primary school level, or as braille, with or without sound effects, for use by visually challenged persons (Andic et al., 2019).

CONCLUSIONS AND RECOMMENDATIONS

In this study, two new taxonomic key formats have been designed, illustrated and proposed for use in plant taxonomy. They are namely: multi-level table of identification and multi-layer circular diagnostic chart, both of which are single access devices. Using these key formats, the trio activities of key construction, plant identification, and plant identity confirmation are made possible through robust algorithms. Since each of these algorithms is in conformity with the principal features of a good/executable computer algorithm (that is, being deterministic, general, finite, and with capacity to act on at least one input to produce at least one output), it is believed that these alternative key formats should be programmable. Going by their features and functionality attributes, the two new key formats proposed in this paper are recommended as useful templates upon which reliable plant diagnostic tools can be based. This paper has also contributed wood anatomy-based diagnostic keys usable for authenticating five medicinal herbs marketed as plant roots in Ogbomoso, Nigeria.

CONFLICT OF INTERESTS

The author has not declared any conflict of interests.

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