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Original Article

Floral Maturation Indices of African Yam Bean (*Sphenostylis stenocarpa* Hochst Ex. A. Rich) Harms (Fabaceae)

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Abstract

The present investigation aimed at understanding the floral morphology and maturation indices of African yam bean (AYB), knowledge of which is very relevant for the crop's improvement programs. Ten accessions of AYB were grown in rows of ten plants, in three replications. Thirty anthesized flowers were collected from each accession for the study of the floral parts. The progressive development of flowers from buds was additionally investigated. AYB flower is large and complete. The calyx was fused and the corolla has five petals (one standard, two wings and two keels). AYB accessions exhibited racemes and indeterminate form of inflorescence. The mean standard petal length and width were 2.94 cm and 3.59 cm respectively. The androecium which surrounded the carpel consists of ten filaments (nine fused, one free). The gynoecium comprised flattened stigma with hairy edge. The mean length of style and ovary was 1.60 cm and 1.00 cm respectively. A distinct purple line (along the enclosing end of the standard petals) became visible at flower bud stage, when bud length reached 2.2 cm - 2.7 cm. Anthesis occurred within 24 hours beyond this stage and petals dropped a day after anthesis. Certification of stigmatic receptivity and pollen fertility may be dependent on the above information. The present study further provides basis for the understanding of pollination mechanism and breeding system within the species.

Keywords: androecium; anthesis; floral morphology; flower bud; gynoecium

Introduction

An overview of the societal importance of legumes (Fabaceae) provides ample justification for significant investment in the botanical family (Gepts *et al*., 2005). African yam bean (*Sphenostylis stenocarpa* Hochst ex. A. Rich) Harms is an underutilized species in the family with high nutritional profile. Its nativity is West and Central Africa. The Avatime (Ghana), the Bandudus (Democratic Republic of Congo), Ekiti (Nigeria) etc. are some of the ethnic groups which place significant cultural values for the crop (Potter and Doyle, 1992). Many lives were sustained during the Nigerian civil war as the crop provided the major protein source in the Igbo (Nigeria) refugee camps between 1964 to 1967 (Nwokolo, 1996). The production of the crop is remote and limited to few cultural niche where it is grown in mixed and as a companion crop with some primary crops especially yam (*Dioscorea* spp.). It is grown as a secondary crop to provide meal for the farming family during off season.

An old documentation by Yampolsky and Yampolsky (1922) revealed that plant species exhibits various sexual forms. From the 120,000 plant species catalogued by then, 86% were bisexual (72% hermaphrodite, 7% monoecius and 7% gynomonoecious, andromonoecious or androgynomonoecious), 4% were unisexual (dioecious) and 10% were both unisexual and bisexual (gynodioecious, androdioecious or trioecious). Whichever of these forms a plant species exhibits, flowering initiates the sexual reproductive cycle in angiosperms (Elliot *et al*., 1982).

The taxonomy of angiosperms has traditionally been dependent on flower characteristics such as morphology, e.g. petal color (Endress, 2010). The name, *Sphenostylis* was evolved by Harms (1899) to replace *Dolichos* in the description of a small group of legume with wedge shaped cuneate style. "*Sphen*" in *Sphenostylis* is a Greek word which describes the dorsiventrally flattened stigma and wedge-shaped style (Allen and Allen, 1981; Potter and Doyle, 1994). Owing to the remark of Gepts *et al*. (2005) that each legume has unique features of botanical relevance, the re-description of the genus *Sphenostylis* was meant to uphold the distinction of the small group within the Fabaceae family.

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Members of the genus *Sphenostylis* are closely related to and were formally grouped with *Dolichos* and *Vigna;* hence, most of them bore *Dolichus* and *Vigna* synonyms (Harms, 1899; 1911). Gillett (1966) considered them to be closely related to *Lablab*. However, *Nesphostylis* is the nearest sister to *Sphenostylis* because of the cuneate style and dorsiventrally flattened stigma (Milne-Redhead and Polhill, 1971; Potter and Doyle, 1994). The presence of aril on the seeds, appendages on standard petals, apical dilation on the stamens, a tooth at the base of the vexillary stamen, large persistent bracteolate and hair below the terminal stigma (Milne-Redhead and Polhill, 1971) differentiated *Nesphostylis* from *Sphenostylis*. Diversity of floral forms exists at intra-generic levels and the understanding of the peculiarity of the floral system of each species is necessary for the species' genetic improvement.

Insufficient understanding of species' floral biology has been repeatedly cited as a shortcoming in the recovery plan of endangered species (Tear *et al*., 1995; Clark *et al*., 2002), low productivity (Ganapathy *et al*., 2008; Eradasappa and Mohana, 2016) and poor initiation of applied research capable of leading to improvement of most underutilized species. Young *et al.* (2007) further remarked that basic biological knowledge of a species can help to identify factors that limit their long-term persistence. The unavailability of the knowledge of species floral biology has in varied proportion contributed to continual underutilization of the less privileged crop species.

Conventional crop improvement dwells on the good knowledge of the floral morphology of a crop because effective advancement on population and trait of interest would be unachievable without the flowers. Information on the flowering and the breeding system of AYB has not been well documented. Since this critical information is not available for the crop, the present study therefore, offered to provide hints to identifying characteristic(s) linked to AYB flower maturation. The import of the information is essential for breeding and improvement of the crop. It will further serve as a basic guide for morphological characterization and pollination mechanism within the species.

Materials and Methods

The present experiment was carried out on ten selected accessions from the 79 accessions of African yam bean collected from the Genetic Resources Centre of the International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria, whose morphological diversity was earlier reported by Adewale *et al.* (2012). Each was planted in a row of ten plants in three blocks at spacing of 1 m x 1 m in a field at IITA, Ibadan (7.5 °N, 3.9 °E). Thirty anthesized flowers were collected from each accession across the three replications and measurements on the floral parts were recorded. Data were collected on: length of standard petal, breadth of standard petal, length: breadth ratio of standard petals, standard filament length, ovary length and style length. The mean and standard error of the six parameters were computed for the ten accessions following the procedure in Gomez and Gomez (1984).

The trend of growth and development of the flowers from visible and handy flower bud stage (of about 10 mm length) were further studied on a single flower basis following the protocol proposed by Dafni (1992). The study involved six African yam bean accessions (TSs10, TSs39, TSs58, TSs98, TSs118 and TSs125). Four developing flower buds (whose petals have not opened and bud lengths were about 10 mm long) were tagged on three plants on a row in each of the three replications. Progressive development of the tagged flower buds was daily monitored and length (BL) measured until anthesis. Other data recorded included: days to anthesis (DTA) and days to flower drop (DTP). Changes in flower buds size and colour were carefully observed daily.

Variability among the accessions for the parameters was studied by analysis of variance (ANOVA) in SAS using PROC GLM. Means were separated using the Duncan Multiple Range Test.

Results

The calyx of AYB flower is fused, however, the components of the corolla, androecium and gynoecium are presented in Fig. 1. The mean and the standard error of some floral parts of the 10 AYB accessions are presented in Table 1. Mean length and width of the standard petals of the ten accessions were 2.94 cm and 3.59 cm respectively; TSs10 had the least (2.67 cm and 3.21 cm), while TSs125 had the highest (3.18 cm and 3.87 cm) values respectively (Table 1). The width of the standard petal was longer than the length in all the studied accessions. The ratio of the standard petal length to the width ranged between 0.80 (TSs111 and TSs118) and 0.85 (TSs23). The mean length of the only free filament (e.g. the vexilary), ovary and style were: 2.55 cm, 1.60 cm and 1.00 cm respectively (Table 1).

The studied AYB accessions exhibited racemose and indeterminate form of inflorescence. The flower buds grew and developed progressively along its length and width (Fig. 2). Bud length of AYB flowers became visible and handy at a length of 1.00 cm (Fig. 2A). Daily monitoring for growth in length of the developing flower buds was done at 24 hours interval. For the six genotypes considered for the current study, flower bud lengths did not differ significantly from day one to four (Table 2). However, bud length at the fifth day (BL5), days to anthesis and days to petal drop differed significantly ($P \leq 0.05$) among the six accessions (Table 2). Mean length of the flower buds increased linearly on daily basis.

The six accessions recorded mean progressive flower bud length increase of 0.26 cm, 0.32 cm, 0.37 cm and 0.43 cm in the four intervals between day one to five (Table 3). Mean daily addition to bud length was about 0.35 cm. Days to anthesis and petal drop was attained earlier in TSs58 compared to the other accessions (Table 3). A distinct purple lateral line ran along the enclosing end of the standard petals of well-developed flower buds of AYB (Fig. 3). Anthesis was most imminent at this stage. Length of buds at this stage ranged between 2.20 cm - 2.50 cm. This range coincided with bud length measurements of the fifth day (BL5, Table 3), a day to flower anthesis.

Table 1. The measurements of the floral parts of anthesized flowers of some accessions of African yam bean

Parameters	Accessions										Grand Mean
	TSs10	TSs23	TSs39	TSs40	TSs58	TSs96	TSs98	TSs111	TSs118	TSs125	
Standard petal length (cm)	2.67±0.042	2.98±0.113	3.01±0.075	3.07±0.061	2.81±0.048	3.06±0.092	2.85±0.071	2.9±0.078	2.86±0.061	3.18±0.101	2.94± 0.0742
Standard petal width (cm)	3.21±0.067	3.51±0.154	3.69±0.079	3.7±0.123	3.45±0.067	3.79±0.066	3.45±0.082	3.65±0.105	3.58±0.051	3.87±0.084	3.59± 0.0878
Standard petal L:W ratio	0.83±0.012	0.85±0.019	0.82±0.010	0.83±0.013	0.81±0.013	0.81±0.018	0.83±0.013	0.80±0.015	0.80±0.018	0.82±0.012	0.82± 0.0143
Standard Filament L (cm)	2.60±0.038	2.95±0.117	2.40±0.105	2.57±0.036	2.48±0.056	2.37±0.177	2.4±0.128	2.56±0.086	2.49±0.026	2.70±0.073	2.55± 0.0842
Ovary length (cm)	1.72±0.029	1.66±0.042	1.64±0.034	1.6±0.053	1.52±0.036	1.4±0.046	1.56±0.049	1.58±0.059	1.64±0.048	1.65±0.043	1.60± 0.0439
Style length (cm)	0.94±0.069	1.08±0.049	1.06±0.034	1.07±0.029	0.91±0.042	1.01±0.064	1.04±0.042	1.03±0.041	0.67±0.078	1.20±0.037	1.001± 0.049

L = Length, W = Width; n = 30

Table 2. Analysis of variance summary of six floral morphological traits

Source of Variation	Degree of freedom	Mean squares						
		BL1	BL2	BL3	BL4	BL5	DTA (days)	DTP (days)
Accessions	5	0.00011	0.0032	0.0211	0.0429	0.0381**	0.3886**	0.2635*
Error	215	0.00021	0.0145	0.0291	0.0204	0.0017	0.1137	0.0961
Mean		1.0012	1.260	1.578	1.953	2.381	5.862	6.888
Range		1.0-1.2	1.0-1.6	1.2-2.0	1.4-2.4	2.2-2.7	5.0-6.0	6.0-7.0
CV (%)		1.46	9.55	10.81	7.32	4.54	5.75	4.50

Note: BL1 – BL5 = Measurement of bud lengths for days 1 to 5 in centimeters, DTA = Days to Anthesis, DTP = Days to Petal drop (after anthesis), CV = Coefficient of variation

Table 3. Mean of floral bud lengths of different ages

Accessions	BL1	BL2	BL3	BL4	BL5	DTA	DTP
TSs118	1.004a	1.265a	1.581a	1.991a	2.412a	5.925a	6.944a
TSs10	1.002a	1.256a	1.565a	1.933a	2.384a	5.869a	6.891a
TSs125	1.000a	1.241a	1.531a	1.869b	2.355ab	5.931a	6.931a
TSs39	1.000a	1.268a	1.598a	1.977a	2.398a	5.844a	6.889a
TSs58	1.000a	1.264a	1.597a	1.942ab	2.328c	5.667b	6.722b
TSs98	1.000a	1.265a	1.586a	1.933a	2.386a	5.956a	6.956a
Grand Mean	1.001	1.259	1.575	1.947	2.378	5.865	6.889

Note: *Means with the same letter are not significantly different and mean comparison is along each column
BL1 – BL5 = Measurement of bud lengths for days 1 to 5 in centimeters, DTA = Days to Anthesis, DTP = Days to Petal drop (after anthesis).

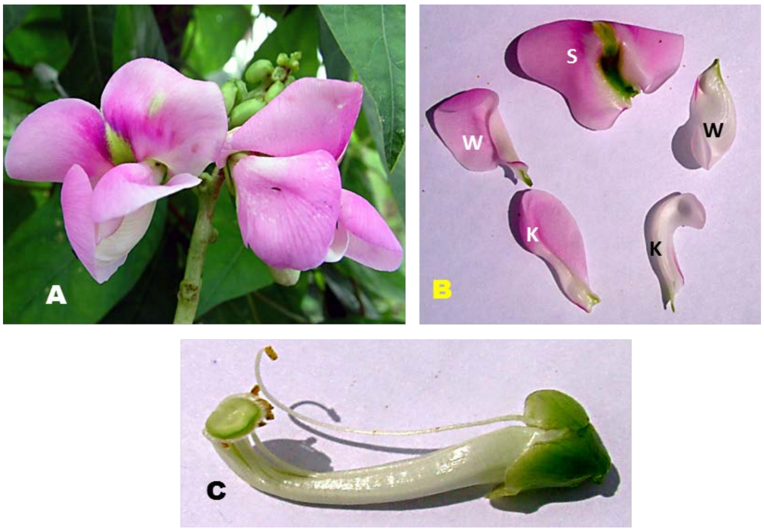

Fig. 1. Morphological parts of the African yam bean flower A - Anthesized flower; B - Corolla components of African yam bean (S - Standard/banner, W - Wings (2) and K - Keels (2)); C - Diadelphous stamens (nine filaments fussed from base and one free, each bearing one anther) and "*sphen*" (Greek) e.g. dorsiventrally flattened ovary and wedge or cuneate style

Fig. 2. Progressive length measurements of the flower buds of African yam bean A - Flower bud length of about 1.0 cm; B - Flower bud length of about 1.5 cm; C - Flower bud length of about 2.0 cm; D - Flower bud length of about 2.5 cm

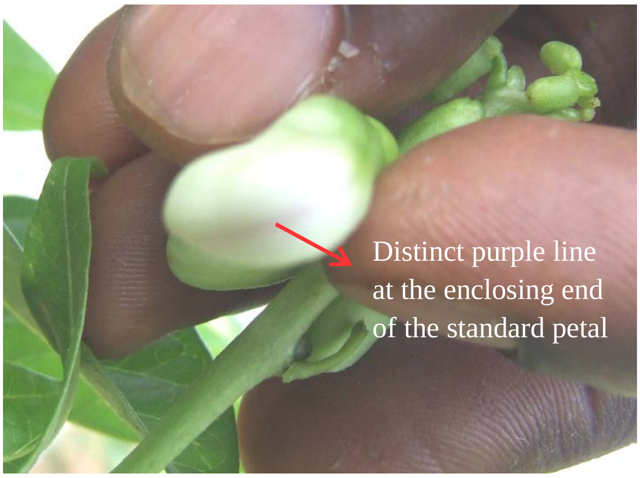

Fig. 3. A qualitative indicator of flower bud maturation in African yam bean

Discussion

The basic understanding of the structure, sexuality and phenology of the flower is a pre-requisite for understanding the life cycle as well as the necessary background for any pollination and reproductive biology studies (Driscoll, 1990; Dafni, 1992). Therefore, the familiarization of a plant breeder with the floral morphology and pollination mechanism is key to propounding genetic improvement for the crop of interest. African yam bean is a legume in the sub-family of Papiloinoideae. Milne-Redhead and Polhill (1971) had much earlier remarked that the inflorescence of African yam bean was raceme. The development of flower buds according to Pareek *et al.* (2007) occurs in stages. From the study, it was observed that the apical meristem initiated bracts in acropetal succession and a floral bud developed on the axil of each bract. The petal forms of African yam bean flower conformed to that of most Papilionoideae (Tucker, 1987). Each anthesized flower had three petal forms: a single standard petal (or vexillum), two wing petals and two keel petals. The vexillum petal enlarges greatly and encloses the rest of the flower, but remains bilaterally symmetrical.

African yam bean flower is large, complete and hermaphroditic, having the four basic floral parts (calyx, corolla, stamen and pistil). The length and the width of the standard petal, length of the vexillary filament, ovary and style varied significantly among the accessions. The standard petal width was consistently longer than the standard petal length. The stamen had nine fused filaments and one standard whose mean length was 2.55 cm. The description of the diadelphous Papilionoid filaments by Tucker (2003) accurately matches the staminate features of African yam bean, in which nine filaments were fused and one was free. Moreover, the shape of the anther on the ten filaments was tetra locular, fitting into the description of the Papilionoideae by Milne-Redhead and Polhill (1971). The incomplete fusion of the stamen produced a hole or fenestrae leading to the nectary base. The gap seems to facilitate the entry of bee's proboscis into the nectary gland.

Identifiable developmental changes on flowers are inferences to precede anthesis or anther dehiscence. Frankel and Galum (1977) documented some of such indicators to include: change of colour at the tip of the closed corolla (as observed in Flax, Tobacco and Tomato), increase in the size of the flower buds, protrusion of petals from the calyx etc. This study identified two floral maturity indicators signifying the termination of the flower bud stage for the onset of anthesis in African yam bean.

Firstly, the daily data generated on flower bud length measurements gave a meaningful inference (Frankel and Galum, 1977; Dafni, 1992). When flower bud length reached about 2.4 cm (± 1cm), there was an indication that the bud stage was about to be terminated for anthesis. Secondly, the notice of a purple stripe along the joint of the standard petal of the flower bud was another indicator. These two developmental inferences provided signal for the nearness of anthesis in African yam bean. A strong correlation was observed between the two; therefore, anthesis in African yam bean is imminent when flower bud length reaches about 2.4 cm (± 1cm) and a purple stripe along the joint of the standard petal becomes visible.

The present study summarily identified indicators of floral maturity as basic information on which further investigation such as accurate timing of stigma receptivity, timing of pollen viability, etc. could stem from. The study envisages further research in this line for a thorough understanding of the possible pollination mechanism and breeding systems of the crop.

Conclusions

African yam bean is a legume with large, perfect and complete dialdelphous flower. From the flower bud stage at which the length equals 1.0 cm, metric measurement of the length of the flower bud linearly increased daily and reached 2.2 to 2.5 cm at the fifth day. A conspicuous purple line along the enclosing end of the standard petal appears when bud length reaches about 2.4 cm length. The two coinciding features agree with progressive timing development and thus (from our study) became the indicators of floral maturation of *Sphenostylis stenocarpa*.

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