

**IMPACT OF HYBRIDIZATION ON OKRA (*Abelmoschus esculentus*) NUTRIENT
ENRICHMENT****ABSTRACT**

In this study, hybridization was carried out on two varieties of okra namely: Clemson spineless and dwarf long green to raise F_1 so as to assess the expression of heterosis or improved vigour. Studies have shown that significant differences exist in the morphological, Phytochemical and nutrient characteristics among okra varieties. The aim of plant breeders ~~try~~ is to substitute the undesirable qualities of ~~in~~ plants with desirable ones so that it ~~will~~ result in higher yield of crops of improved quality. ~~and~~

~~this is achieved through the process of hybridization. In this study, hybridization was carried out on two varieties of okra namely: Clemson spineless and dwarf long green to raise F_1 so as to assess the expression of heterosis or improved vigour and~~

The impact of hybridization on okra nutrient enrichment as shown in vitamin compositions (vitamin A, B₁, B₂, B₃ and C (fruits part) of the parental plants compared with the F_1 hybrid. ~~This was done using standard methods.~~ Results were analyzed using one way analysis of variance (ANOVA). Results showed that the parental and the F_1 plants contained varied quantities of all the investigated vitamins. Higher contents of Vitamins A, B₂ and C were recorded for the F_1 hybrid when compared with the parental plants. So, there was an expression of heterosis indicating that hybridization could greatly enriched vitamin nutrients in okra.

Moreover, F_1 generation combined the characters of Clemson Spineless and Dwarf long green varieties and also had additional improved genetic attributes. This can be seen in most nutrients being higher in the F_1 hybrid and also the reduced spines in the fruit of the F_1 hybrid as opposed to the prominent spines in dwarf long green parent.

Keywords: hybridization, okra varieties, Clemson spineless, dwarf long green, heterosis, vitamin composition, nutrient enrichment

30 INTRODUCTION

31 *Abelmoschus esculentus* L.(Moench), Okra, is an important vegetable in the family Malvaceae. It
32 ~~is a green fruit which is strictly used as a vegetable and~~ is valued for its edible green ~~seed~~ pods
33 containing round, white seeds (Jesus *et al*, 2008) which is commonly used vegetable of India. It is
an economically important vegetable crop cultivated in tropical, subtropical and warm temperate
regions around the world (Vaughan and Geissler, 2009). The fruits are said to be shaped like a lady's
fingers - one of its common names in British English. The flowers and upright plants give okra an
ornamental value (Duzyaman, 1997). Okra provides dietary fibres and distinct seed protein balanced in
both lysine and tryptophan - amino acids (unlike the proteins of cereals and pulses) thus, referred
as “a perfect villager’s vegetable (Camciuc *et al.*, 1998). Okra is an annual herb, 3 to 6 feet tall with a
hibiscus-like flower, a deep branched taproot and succulent stem with scattered, stiff hairs. The pods of
Abelmoschus esculentus have a unique flavour and texture and release slimy mucilage on cooking, which
can be

42 used to thicken sauces and add smoothness to soups. Its mature fruit and stems contain crude fibre,
43 which is used in the paper industry. (Mabberley, 2008).

44 In the Eastern part of Nigeria, precisely Anambra, there are five varieties which are the most
45 common. They are; Green emerald, Dwarf long green, the local long pod variety, perkins spineless
46 and Clemson spineless. The two varieties which are the focus of this study are the Clemson spineless
47 and the dwarf long green. The Clemson spineless is long, narrow and do not possess spines; while
48 the dwarf long green variety is shorter, greater in diameter and possess spines. The Dwarf long green
49 variety has a darker shade of green colour than the Clemson spineless (Van, 2005).

50 Vitamin is an organic compound and a vital nutrient that an organism requires in limited
51 amounts. The body can only manufacture three vitamins D, K and Biotin a B vitamin. The rest must
52 be obtained from dietary sources. Studies have shown that significant differences exist in the
53 morphological, phytochemical, nutrient and anatomical characteristics among okra varieties (Van,
54 2005). Furthermore, the modern scientific method of crop improvement was necessitated by the
55 present demands of modern man for good quality ~~food~~ crops; ~~crops-with~~ having good taste,
~~crops-that-mature~~ early maturity, ~~crops-that-contain-most-of-the-essential-food~~ more nutrients and
~~crops~~ that can meet the immediate needs of man in terms of food and raw materials (Duzyaman, 2010;
Ilodibia *et al.*, 2014 and 2015). ~~Plant breeders try to substitute the undesirable qualities in plants with~~
~~desirable ones so that it will result in higher yield of crops of improved quality and this is achieved~~
~~through the process of crossbreeding or hybridization.~~ Hence, this research aimed at hybridizing two
varieties of

61 okra Clemson spineless and dwarf long green to produce a hybrid that might combine the qualities of

62 the two varieties with ~~the removal~~ substitution of their ~~bad~~ undesirable traits to meet the
63 popular demands of the society.

64 Such a hybrid of the two varieties is likely to be of better quality and demand than either of the two
65 varieties. Accordingly, the problem and focus of the researcher is to hybridize the two common
66 varieties found in Nigeria and compare the vitamin contents of the hybrid and the parental plants
67 (Clemson spineless and the dwarf long green). It will cover the first filial generation in related
68 qualities.

69 MATERIALS AND METHODS

70 Collection, identification of plant materials and study area

71 The seeds of the two varieties of *Abelmoschus esculentus* (Clemson spineless and the dwarf long
72 green) used in this work were obtained between March – April 2016 and authenticated at the
73 Agricultural development project (ADP), Kwata, Anambra state, Nigeria. The experiments were
74 carried out at the Botany laboratory, Nnamdi Azikiwe University, Awka, Anambra state and Emery
75 Biotechnology Laboratory, Ahia-Eke, situated in Umuahia, Abia State. The design of the study was
76 pure experimentation of randomized complete block design.

77 Materials and experimental procedure

78 ~~Seeds of two varieties of okra (Clemson spineless and dwarf long green (Fig. 1), twenty (20) black~~
79 ~~polythene bags, weighing balance, a pair of forceps, aluminium foil, animal manure and loam soil.~~

80 After the seeds were obtained, a viability test was carried out on them. This was done by soaking a
81 handful of each variety in a beaker containing distilled water for approximately 3 minutes. The seeds
82 that floated on the surface of the soil were discarded while those that sank to the bottom of the
83 beaker were sown in a transparent glass jar containing loamy soil. After a few days, germination
84 occurred proving the seeds to be viable. A total of twenty black polythene bags were obtained,
85 perforated and filled with 5kg of loamy soil each. Ten of the polythene bags were labeled ‘Clemson
86 spineless’ while the other ten were labeled ‘Dwarf long green’ which are the two varieties to be
87 studied. The seeds of each variety were broadcast accordingly on the soil surface (five seeds per
88 bag). At different stages, the seedlings were pruned to two seedlings per bag when they were 12cm
89 high. Flowering of the dwarf long green variety started at eight weeks while that of the Clemson
90 spineless variety started at nine weeks. It was at this stage that the crossing or hybridization was

done. Artificial crossing was adopted because okra is a self-pollinating crop. This involved the removal of anthers with a pair of forceps from the dwarf long green variety thus using it as a female parent before it dehisces and covering it with a study bag (foil) to avoid natural crossing by insects. This was followed by the collection and transference of ripe pollen grains from the Clemson spineless (male parent) to the stigma of the emasculated plant (dwarf long green). This pollination process was followed by fertilization and subsequent production of fruits (the F₁ hybrid). This procedure is as outlined by Ilodibia *et al.* (2014).

Final experimental detail of hybrid seeds and layout is missing.

Vitamin nutrient analysis

Dried fruits of two varieties of okra (Clemson spineless, Dwarf long green) and the F₁ hybrid were ground into fine (100-mesh screen) powder with a dry mill and then examined for the following vitamins (A, B₁, B₂, B₃ and C) using the standard methods described by AOAC (2005).

STATISTICAL ANALYSIS

Data obtained was statistically analyzed using one way analysis of variance (ANOVA). The Duncan's multiple range test was used to test the difference among treatments at 0.05% level. Results were presented in Mean± Standard Error.

RESULTS AND DISCUSSION

Results of the study were shown in Table 1 and Figures 1-4.

Results showed that the parental and the F₁ plants contained varied quantities of all the investigated vitamins except B₃ which was absent in F₁ (Table 1). This conforms to the report of Velu *et al.* (2011) and Ilodibia *et al.* (2015), that several hundred wheat accessions showed four to five fold variability for grain Fe and Zn concentrations and the two species of pepper (*Capsicum frutescens* and *C. annuum*) and F₁ hybrid all contained different nutrient compositions respectively. The fruit of the dwarf long green variety contained higher quantities of Vitamins B₁ and B₃ when compared with the fruit of the Clemson spineless variety and the F₁ hybrid.

Higher contents of Vitamins A, B₂ and C were recorded for the F₁ fruit when compared with the fruits of parental plants. Vitamin A: 8.75 ± 0.00, 9.40 ± 0.00 and 9.75 ± 0.00 for clemson spineless, dwarf long green and F₁ hybrid respectively (Table 1, Fig. 1). Vitamin B: 0.45±0.00, 0.35±0.00 and 0.50±00 for clemson spineless, dwarf long green and F₁ hybrid respectively (Table 1). Vitamin C:

18.80±0.14, 17.53±0.11 and 19.20±0.10 for clemson spineless, dwarf long green and F₁ hybrid respectively (Table 1). This is in line with the report of Allard (1960) that the progeny of hybridization will combine many of the qualities of both parents thus producing a crop which is superior to some extent to either of the parents. Again, Nwakile (1994) reported that when the male pollen of a plant is manually transferred, into the female stigma of another plant which may be of a different variety, species or even genera, a hybrid with improved characters is produced. Also, with that of Ilodibia *et al.* (2014 and 2015), who reported that F₁ generation combined the characters of both parents and other improved attributes.

Furthermore, the F₁ generation combined the characters of both the Clemson spineless and Dwarf long green varieties, although the characters of the Dwarf long green dominated that of the Clemson spineless variety. This can be seen in the reduced number of spines possessed by the F₁ hybrid as opposed to the Clemson spineless which has no spines. This agrees with what Mendel (1866) pointed out that when two plants with pair of contracting characters are hybridized, one of the characters would often appear in the offspring while the other remain masked. The character that appeared in the offspring he called dominant character while the character that did not appear he called recessive character. Vitamin A provides healthy skin, good eye sight, a powerful antioxidant that protects against cancer and heart disease and enhances the immune system. Vitamin B1 provides healthy heart and nervous system, optimizes metabolism and brain formation. Vitamin B2 helps in the formation of antibodies and red blood cells, facilitates carbohydrate, fat and protein metabolism, aids against stress and fatigue. Vitamin C is an anti-stress vitamin, protects the cells from toxic wastes and is the most powerful and effective of all the antioxidants, reduces infections, allergies, asthma and common cold.

154 **Table 1: Mean Percent Vitamin compositions of the fruit of Clemson spineless, Dwarf long**
 155 **green and F₁hybrid (%).**

Parameters	Clemson spineless (F)	Dwarf long green (F)	F ₁ Fruit	p-value
Vit. A (UI/100g)	8.75±0.00 ^a	9.40±0.00 ^b	9.75±0.00 ^c	0.01
Vit. B ₁ (mg/100g)	0.45±0.00 ^b	0.35±0.00 ^a	0.50±0.00 ^c	0.05
Vit. B ₃ (mg/100g)	0.88±0.00 ^b	1.04±0.00 ^c	0.10±0.00 ^a	0.00
Vit. C (mg/100g)	18.80±0.14 ^b	17.53±0.11 ^a	19.20±0.10 ^c	0.00

156 Results are in Mean ±Std.

157 The same letter in a column is not significantly different by Duncan's multiple range test at (p<0.05)

158 F: fruit

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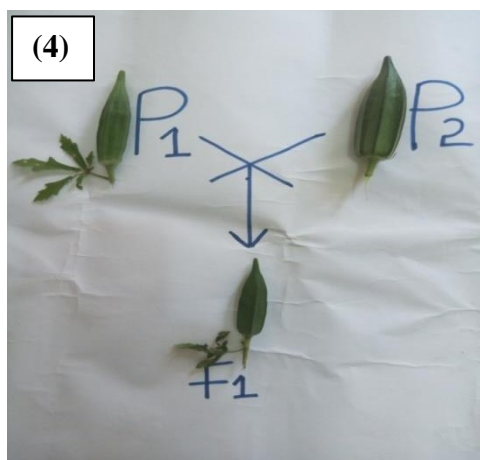
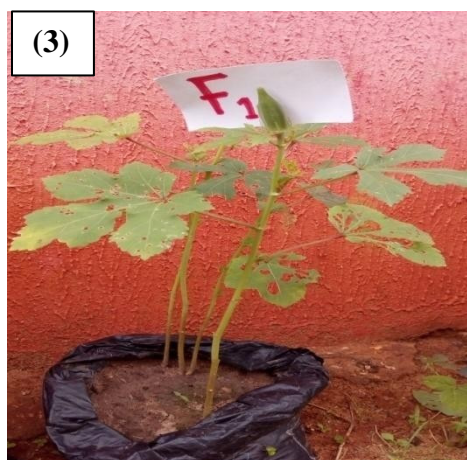
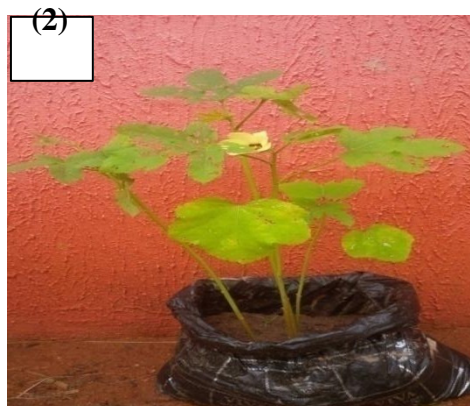
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Fig. (1) The parental variety P_1 (Clemson spineless) (2) Parental variety P_2 (Dwarf long green) (3) F_1 hybrid and (4) Fruits of parents compared with the fruit of the F_1 hybrid.

Conflict of Interests: Authors have declared that no competing interests exist.

182 **CONCLUSION**

183 Results of this study showed that the F₁ generation combined the characters of Clemson Spineless
 184 and Dwarf long green varieties and also had additional improved genetic attributes. This can be seen
 185 in most nutrients being higher in the F₁ hybrid and also the reduced spines in the fruit of the F₁
 186 hybrid as opposed to the prominent spines in dwarf long green parent. Thus hybridization could
 187 greatly enriched vitamin nutrients in okra.

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