Hormonal Response of Gibberellin (Ga3), Grafting and Seasonal Variations on Growth and Yield Parameters on Okra (Abelmoschus Esculentus)

Aminu, Y.
Biology Department,
College of Education and Preliminary Studies, Kano State, Nigeria

Maryam, M.G.
Chemistry Department,
College of Education and Preliminary Studies, Kano State, Nigeria

Kabiru, S.A.
Geography Department,
College of Education and Preliminary Studies, Kano State, Nigeria
E-mail: aminuyahayafagge@gmail.com
GSM: +2347035347997

Abstract
The effect of Gibberellin Hormonal Response and Grafting methods on the Growth and Yield of Okra was investigated, with the aim of inducing variability that could be exploited in the improvement of some quality traits in Okra plants. Gibberellin, grafting and a combination of grafting with Gibberellin) were applied on to the Okra seeds. The seeds of Okra were treated at four different concentrations of Gibberellin (0.1mM, 1.0 mM, 2.0 mM and 0.0 mM as control) after which some of the mutants were grafted using tongue grafting method at seedlings stage. The other set of seeds were not treated with the hormone but were grafted at seedlings stage. The results obtained revealed highly significant difference (P≤ 0.01) in the effects of Gibberellin on survival rates, number of fruits, and fruit weight. Similarly, highly significant differences (P≤0.01) were found between the treatments in Survival rate, except on the number of fruits and fruit weight, where no significant differences exist. More so, significant differences were found in the traits between the seasons except in fruit number. The result showed that Gibberellin and grafting improve important quality traits of Okra. The mutants and grafted Okra could be grown both during the rainy and dry seasons, while 0.1 mM concentration of Gibberellin and grafting improve some important quality traits of Okra that could be utilized for further improvement of Okra. However, the response of Okra to grafting was higher. Based on the findings, the study concluded that, grafting improves some important quality traits of Okra that are of high economic value and possible recommendations that could be made.

Keywords: Grafting, Hormone, Season, Variation, Interaction, Gibberellin, Okra.

1. Introduction
Okra (Abelmoschus esculentus (L.) Moench) known in many English speaking countries as lady's fingers, bhindi in India, krajaj kheaw in Thailand, okra plant, ocho, okoro, quigmombo, quingumbo, gombo, kopiarab, kacang beni and bhindi in South East Asia. However, in Middle East it is known asbamia, bamyayor bamiieh and gumbo in Southern (Ndunguru, et al.,2004). Portuguese and Angola, okra is known as quiabo, and asquimbombo in Cub, gombo commun, gombo, gumbo in France, mbamia and mbinda in Sweden,and in Japan as okura (Chauhan et al.,2001), (Lamont, et al.,1999). Taiwan is called qiu kui (Siemonsma, et al.,2000).Nigeria spoken in Igbo (Whoter, et al.,2000). It belongs to family malvaceae and genus Abelmoschus.

Okra is a multipurpose crop due to its various uses of the fresh leaves, buds, flowers, pods, stems and seeds (Mihretu et al., 2014). Okra immature fruits (green seed pods), which are consumed as vegetables, can be used in salads, soups and stews, fresh or dried, fried or boiled (Ndunguru & Rajabu,2004). It offers mucilaginous consistency after cooking. Often the extract obtained from the fruit is added to different recipes like soups, stews and sauces to increase the consistency. Okra mucilage has medicinal applications when used as plasma replaces men or blood volume expander. The mucilage of okra binds cholesterol and bile acid carrying toxins dumped into it by
the liver. The immature pods are also used in making pickle. The entire plant is edible and is used to have several food (Madison, 2008; Maramag, 2013).

Okra seeds are source of oil and protein. Okra seeds have been used on a small scale for oil production. It can be also used as no caffeinated substitute for coffee. Okra seeds may be roasted and ground to form a caffeine free substitute for coffee (Calisir, & Yildiz, 2005). Okra also has industrial applications and is used in confectionary (Adetuyi et al., 2011).

To promote the use of indigenous vegetables like Okra that have play significant role in mitigate food insecurity and alleviate malnutrition in the country. However, Okra has been considered a minor crop and no attention was paid to its improvement in the international research program in past (Sanjeet et al., 2010).

On the other hand, the demand for vegetable oils is rapidly increasing due to the growing human population and the expanding oil industry with health promoting oil components, the exploration of some underutilized and newer resources of vegetable oils is of much concern (Schalau, 2002). Okra, which is currently grown mainly as a vegetable crop, has potential for cultivation as an essential oilseed crop because okra seeds contain high amount of oil (20-40%) (Sorapong, 2012; MEF, 2013). Among PGRs, gibberellin play vital role in regulating developmental processes within plant bodies (Gou et al., 2010). Gibberellin helps in cell growth of stem, leaves and other aerial parts by causing cell elongation, and increase in intermodal length. A higher concentration of gibberellins increases plant growth (Bora and Sarma, 2006). The different concentrations of GA had significant effect on growth in mustard (Akter et al., 2007). Baydar (2000) reported that oil synthesis increases with increasing dose of GA in safflower. Gibberellins, are effective in increasing oil yield. Besides yield, the fiber strength and fineness is also improved by the application of gibberellin (Ayala-Silva et al., 2005). Mckenzie and Deyholos (2011) reported that treatment of GA causes stem elongation, expansion and proliferation and cell wall thickening in bast fiber of linseed. This study aimed to investigate and determine the effects of Gibberellin, Grafting and Seasonal Variations on Some Growth Parameters on Okra (Abelmoschus esculentus).

2. Materials and Method
The research was conducted in (September, 2013) at the Green House of the Botanical Garden of the Department of Biological Sciences, Ahmadu Bello University Zaria, Kaduna State. (Lat11°12’N, Long 7°37’E, Alt 550-700 m above sea level) (Anonymous, 2014).

2.1 Sources of the Seeds
Seeds of three varieties of cultivated tomato (Aji amarillo, Aji chili and Aji cristal) were collected from the Institute for Agricultural Research (I.A.R), Ahmadu Bello University Zaria, Nigeria.

2.2 Treatment and Experimental Design
The treatments used in the research are Grafting and mutation, three concentrations of Gibberellin (0.1mM, 1.0mM, 2.0Mm). These were laid out in a Completely Randomized Design (CRD) with three replications. The seeds of the three Chilli pepper varieties were soaked/treated with three different concentrations of Gibberellin (0.1mM, 1.0mM, 2.0Mm) for 4hours while 0.0mM as control.

The seeds used for root stocks were planted 2 days prior to that of the shoots. Tongue grafting approach was used. This is due to the relative advantage of the method over others such as its being used on larger plants, three times faster than other techniques, high success rate and is easy to handle.

After two weeks of planting, and a day prior to grafting, the plants used for grafting were watered fully to make them turgid. One-fourth of the plants used for rootstock were cut at slant early in the morning. The shoots were also cut in the same way. The two cut ends were placed in direct contact and use a small clip was used to hold the cut surfaces together. This was repeated in the 3rd and 4th week of planting, while the remainders were left as control as described in McVoy (2005) protocol.

2.3 Data Collection
Data were observed and collected on number of Survival rate, number of fruits/plant, and fruit weight.

2.4 Survival Rate (%)
The number of grafts that survive during the emergence of first flower were determined and their percentages taken and recorded. Leafless grafts were considered as dead.
2.5 Number of Fruits/Plant
The number of fruits produced per plant was determined through counting per treatment for each variety after twelve weeks of planting and recorded.

2.6 Dry Weights of the Fruits (g)
The weights of 100 fruits dried in the oven at 90 °C for 48 hours were determined in grams using a balance and recorded. (Mettier, PS15. Max-15000g).

2.7 Data Analysis
All the data collected were analyzed using Analysis of Variance, and the means were separated using Duncan’s Multiple Range Test, (DMRT).

3. Results
The results from the combined analysis of variance on the effects of mutation and grafting on some selected traits of Okra are presented in (Table 1). The results showed highly significant difference (P≤0.01) in the effect of concentrations of Gibberellin on all the selected traits of the Okra. Similarly, high significant difference (P≤0.01) was found among the varieties in terms of all the selected traits. However, no significant difference was found in the effect of the treatments on fruits number and fruit weight, except on survival rate where the effect is significant (P≤0.05). Highly significant difference (P≤0.01) was found among the seasons in terms of fruit weight, and significant difference (P≤0.05) was found among the seasons on survival rate, while no significant difference was found among the seasons in terms of fruits number.

However, no significant difference in the interactions of Gibberellin with variety and between Gibberellin with the treatments on all the selected traits of the Okra plant. Similarly, no significant difference was found in the interactions of Gibberellin with seasons in almost all the selected traits. However, highly significant difference (P≤0.01) was found in the interaction of varieties with treatments on fruit number and fruit weight, while no significant difference was found in the interaction of varieties with treatments on other remaining selected trait. More so, highly significant difference (P≤0.01) was found in the interaction of varieties with seasons on Survival rate and number of fruit, except on fruit weight, where no significant difference was found.

Furthermore, highly significant difference (P≤0.01) was found in the interaction of the treatments with seasons on the fruit weight except on fruit number; where the interaction is significant (P≤0.05) and on survival rates, where no significant difference was found in the interaction. However, no significant difference was found in the interaction of Gibberellin concentrations with varieties and treatments and interaction of Gibberellin with varieties and seasons on the selected traits of the Okra cultivars.

Table 1. Mean Squares for the Combined Effects of Grafting and Gibberellin Interactions on Some Growth Parameters on Okra in Two Different Seasons

| Sources of Variation | dF | Survival Rate (%) | Number of Fruits | Fruit Weight (g) |
|----------------------|----|-------------------|------------------|------------------|
| Replication          | 2  | 74.54*ns          | 11.87*           | 55.92**          |
| Concentration        | 3  | 14265**           | 188.59**         | 383**            |
| Variety              | 2  | 3803.53**         | 37.26**          | 564.34**         |
| Treatments           | 2  | 878.43*           | 5.26ns           | 21.68ns          |
| Seasons              | 1  | 874.67*ns         | 0.01ns           | 189.84**         |
| Conc. x Trt.         | 6  | 72.30**ns         | 1.36ns           | 6.23ns           |
| Conc. x Var.         | 6  | 44.73**ns         | 3.19**ns         | 14.17**ns        |
| Conc. x Seas.        | 3  | 35.35**ns         | 0.91ns           | 9.67**ns         |
| Var. x Trt.          | 4  | 164.47**ns        | 14.79**ns        | 70.64**ns        |
| Var. x Seas.         | 2  | 4860.2**ns        | 95.36**ns        | 12.12**ns        |
| Trt. x Seas.         | 2  | 138.49**ns        | 12.22x           | 45.54**ns        |
| Conc. x Var. x Trt. x Seas. | 18 | 153.02**ns | 2.82ns | 1.63ns |
| Error                | 142| 148.56            | 2.78             | 7.53             |

Keys: ns= No significant difference * = Significant difference (P≤0.05) **= Highly significant difference (P≤0.01)
The result of the analysis of variance ANOVA on the effects of grafting, and seasons on some growth parameters on the Okra was shown in (Table 2). below. It revealed that grafting is more efficient during the dry season in terms of all the selected traits and efficient on survival rate and number of fruit except on fruit weight where it is more efficient in rainy season.

Table 2. Response of Grafting and Seasons on Some Growth Parameters on Okra

| Treatment | Season | Survival rate (%) | Number of fruit | Fruit weight |
|-----------|--------|-------------------|-----------------|-------------|
| Grafting  | DRY    | 47.51<sup>a</sup> | 4.33<sup>a</sup> | 11.69<sup>a</sup> |
|           | RAINY  | 41.43<sup>b</sup> | 4.00<sup>b</sup> | 15.27<sup>a</sup> |

NB: Means within the columns with the same letter(s) are not significantly different using 5% DMRT.

The result of the analysis of variance ANOVA on the effects of mutation, and seasons on some growth parameters on the Okra was shown in (Table 3) below: Mutation was found to be more efficient during the dry season in terms survival rate, except on number of fruit and fruit weight where it was found to be efficient. More so, mutation was found to be more efficient during the rainy season on fruit number, fruit weight, except on survival rate, where it was found to be efficient. This was due to effect of concentration of Gibberellin on the selected trait of the Okra.

Table 3. Response of Mutation and Seasons on Some Growth Parameters on the Okra

| Treatment | Season | Survival rate (%) | Number of fruit | Fruit weight |
|-----------|--------|-------------------|-----------------|-------------|
| Mutation  | DRY    | 51.62<sup>a</sup> | 3.30<sup>b</sup> | 12.72<sup>b</sup> |
|           | RAINY  | 50.75<sup>b</sup> | 4.22<sup>a</sup> | 13.20<sup>a</sup> |

NB: Means within the columns with the same letter(s) are not significantly different using 5% DMRT.

The result of the analysis of variance ANOVA on the effects of mutation, and grafting on some growth parameters on the Okra plant was shown in (Table 4) below. The interactive effect of mutation and grafting was more pronounced during the dry season with regard to survival rate and number of fruits, while with respect to fruit weight, it was found to be higher during the rainy compared to dry season.

Table 4. Response of Mutation and grafting, and seasons on Some Growth Parameters on the Okra

| Treatment | Season | Survival rate (%) | Number of fruit | Fruit weight |
|-----------|--------|-------------------|-----------------|-------------|
| Mutation  | DRY    | 47.51<sup>a</sup> | 4.33<sup>a</sup> | 11.69<sup>a</sup> |
| X Grafting| RAINY  | 41.43<sup>b</sup> | 4.00<sup>b</sup> | 15.27<sup>a</sup> |

NB: Means within the columns with the same letter(s) are not significantly different using 5% DMRT.

4. Discussion

The differences observed in most of the quantitative and qualitative traits among the Gibberellin induced mutants of okra evaluated showed significant improvements in the selected traits. Although there were few traits with no significant differences in responses to the applied treatments; the ability of the mutants to germinate faster after one and two weeks of planting in respect to the controls showed that the mutagenic treatments induced increase enzymatic activities, which could be responsible for the early germination. This finding is in agreement with the findings of Mensah et al. (2007) who reported a decrease in germination with increase in the dose of chemical mutagens. In the present investigation, germination, survival percentage, plant heights and leaf number and area
decreased with increasing concentration of Gibberellin. This finding conformed to the earlier report by Ahloowalia and Maluszynski (2001) that, the viable mutants observed are mainly dependable measure of genetic effect in mutagen. The increased in the number of leaves, plant heights and number of fruits per plant due to Gibberellin treatments was also in conformity with the work of Adamu and Aliyu (2007) who reported increased in growth and yield parameters of tomato due to Gibberellin treatments. There were reductions in the germination and survival percentages with increasing concentrations for both chemicals in the C4 generation. Reductions in germination and survival percentages due to the effects of mutagens on various crop plants have earlier been documented by Mensah and Akomeah(1997)and Mensah et al. (2005).

More so, the improvement in the growth and yield components of Okra due to Gibberellin treatments stressed the effect of mutation on the growth and yield of plants. This is in agreement with the work of Adamu et al. (2002) who observed when groundnut was treated with gamma rays and Sheeba et al. (2005) who reported the effect of gamma rays and EMS were used to treat Sesanum indicum L. where seed germination, seedling survival, were reduced significantly with an increase in dosage levels of both mutagens. However, in contrast, Sasiet et al. (2005) showed that all plant mutant types registered lower yields compared to their parents in the study of the effects of diethyl sulphate and EMS on Okra (Abelmoschus esculentum (L.) var. MDU-1).

The increased in fruit quality (such as juice and fruit weight) and number due to induced mutagenesis by Gibberellin signifies the vital role played by the mutagen in improving the quality traits of Okra.

The distinct differences observed in most of the qualitative traits among the grafted Okra plants compared to the controls showed improvements in fruits quality induced by grafting. The improved fruit characteristics of Okra due to grafting is in agreement with the findings of Lee (1994) and Pogonyi et al. (2005), who independently reported improvements in fruits quality due to grafting. The increase in the number of fruits among the grafted tomatoes was consistent with the work of Ibrahim et al. (2001) who found that the total number of fruits per plant in non-grafted plants was statistically different from those of grafted plants. In a similar study by Khah et al. (2006) fruit weight of grafted plants was found to be higher than in non-grafted plants, and plants grafted onto Heman and Primavera produced more fruit than the non-grafted, both in the greenhouse and in the open field. In the present study, the number of fruits and fruit weights of non-grafted plants were significantly lower than the corresponding values for plants grafted onto both rootstock cultivars.

Higher fruit yield parameters found in this study could be due to the fact that grafting combines novel traits of the two grafted plants after grafting operation. This has also been reported by Tsouvaltiz et al. (2004).

The fruit characteristics of grafted plants were compared with those of non-grafted plants. The results showed that the fruit index (number of fruits and fruit weight) were significantly influenced by grafting. The results agree with those reported by Lee (1994) who concluded that fruit shapes are influenced by rootstocks. Pogonyi et al. (2005) reported that when Lemance F1 was grafted onto Beaufort rootstock, increased yield was caused mainly by higher average fruit weight. Ibrahim et al. (2014) also found that the total number of fruits in non-grafted plants was statistically different from the total for grafted plants.

In grafted combinations, the total fruit yield per plant increased significantly in comparison with that of the control plants. Ibrahim et al. (2001) observed similar results in grafted and non-grafted Chilli plants. These investigators suggested that the higher yield of fruit from grafted Chilli plants was most likely an effect of the vigorous root system of the rootstock. According to Lee (1994), the increased yield of grafted plants is also believed to be due to enhanced water and mineral uptake. Similarly, Osvald (2004) reported that Chilli grafting on suitable rootstocks has positive effects on cultivation performance, especially under greenhouse conditions.

5. Conclusion
The effect of Gibberellin and season interactions was found to be beneficial in improving certain qualitative traits of Okra plant. The effect of grafting and mutation was found to be beneficial in improving certain qualitative traits of tomato varieties. The use of grafting in crop improvement helps to understand the mechanism of grafting induction and to quantify the frequency as well as the pattern of changes in different selected plants by grafting. Okra is therefore recommended for processing industries. More so, Induced mutation using various concentrations of Gibberellin and grafting technique were employed singly and in combination on the three varieties of Okra with the aim of improving the growth and yield parameters of the plants in both the wet and dry seasons. Significant improvements were found among the grafted and mutant tomatoes in both the dry and wet seasons. It was concluded that, Gibberellin via mutation improves some important quality traits of Okra that are of high economic value and possible recommendations made.
6. Recommendation

Since Okra production in the study area has been shown to be very profitable and has huge potential for in some generation, it is recommended that farmers should go in Chilli pepper production to make more income and enhance their livelihoods.

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