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Preliminary study on variability and heritability estimates of micronutrient composition in the immature fruits of okra (*Abelmoschus esculentus*) genotypes in South Africa

Abe Shegro Gerrano^{1*}

Abstract: Okra is one of the important fruit vegetable crop but is an under-utilized and neglected plant species in South Africa. The immature fruits of okra is a good source of protein, minerals and vitamins. Despite its contribution to food, nutritional and health security, this crop is rarely cultivated and researched in South Africa. The study was carried out to assess the genetic variability and heritability estimates using micronutrient content such as iron, zinc and manganese in the immature green fruits of okra plant for future use in the okra breeding programme to increase nutritional value. The experiment was conducted at the Roodeplaat research farm of the Agricultural Research Council in a randomized complete block design replicated three times. The analysis of variance showed highly significant differences for all of three micronutrients recorded. This indicated that there were high level of genetic differences among the genotypes studied. Phenotypic and genetic coefficient of variations, broad-sense heritability (repeatability) and genetic advance were also estimated for iron, zinc and manganese, which could be exploited in selecting suitable and potential okra parents when breeding for high micronutrient contents.

ABOUT THE AUTHOR

Abe Shegro Gerrano is a researcher with the research focus on the assessment of genetic diversity in crop and crop wild relatives towards the genetic improvement of crops for especially yield, nutritional quality as well as stress breeding for drought tolerance in different environmental conditions towards resilience of climate change in South Africa. AS Gerrano is a plant breeder and Geneticist by training. He supervises and co-supervises students' research projects. Dr Gerrano has a huge experience of working together with the communities in conservation of natural resources; improvement of agricultural production and productivity; conduct farmers participatory research that helps the improvements of communities livelihood and technology. Dr Gerrano is working on the leafy, fruit and seed vegetables in South Africa as a breeder in the Agricultural Research Council of the South Africa.

PUBLIC INTEREST STATEMENT

A wide variety of indigenous fruit vegetable crops such as okra are grown and consumed in South Africa. Despite the contribution for food, nutritional and health security importance of this crop, it is recognized as an underutilized and neglected crop and little attention has been given for the improvement of the micronutrient content in the breeding programmes. This work was done on the quantification and heritability analysis of micronutrient contents in the fruits of different okra genotypes in South Africa for future breeding purposes. The current results from this study confirmed that these genotypes were rich sources of micronutrients such as iron, zinc and manganese in the green immature fruits and a great genetic variability was observed. Therefore, this paper provides a basic information on the genetic background for the use of indigenous fruit vegetable crops and their impact and contribution to food security, nutritional value, local and national economy of South Africa.

The phenotypic coefficient of variation ranged from 23.04% in Zn to 75.48% in Fe. Furthermore, the highest values for the genetic coefficient of variation was found in Fe. High heritability estimates were observed in all mineral elements Fe (99.86%), Mn (99.72%) and Zn (96.32%) evaluated indicating potential genetic gain as percentage of mean for Fe (155.38), Mn (69.20) and Zn (45.79), respectively when breeding for these micronutrient traits. The present study revealed the genetic potential of the genotypes studied and their importance for use in the quality-breeding programme aimed towards addressing malnutrition in South Africa.

Subjects: Agriculture & Environmental Sciences; Agronomy; Fruit & Vegetables; Nutrition

Keywords: fruit; genetic diversity; genetic advance; genetic variance; micronutrient

1. Introduction

South Africans depend on a few food crops for nutrition despite the availability of numerous nutritionally-rich indigenous leafy, fruit and seed vegetables that can broaden the food base. Okra is one of neglected crops. It is an annual fruit vegetable crop grown in tropical and sub-tropical regions as well as in warmer temperate regions of the Mediterranean region (Dhankhar & Mishra, 2009) of the world. It is one of the most important African indigenous fruit vegetable crops belonging to the family Malvaceae. The crop originated in Ethiopia (Sathish & Eswar, 2013) and was cultivated by the ancient Egyptians. It's cultivation spread throughout Middle East and North Africa (Lamont, 1999). In Ethiopia it has various names such as Kenkase (Berta), Andeha (Gumuz), Bamia (Oromica/Amharic) (Gemede, Ratta, Haki, Woldegiorgis, & Beyene, 2015). Gemede et al. (2015), Gemede, Haki, Beyene, Woldegiorgis, and Rakshit (2016), Habtamu and Negussie (2014) and Schippers (2000) reported that okra is a multipurpose crop due to its various uses of the pods, fresh leaves, buds, flowers, stems and seeds. They also reported that the immature fruits could be consumed as vegetables, in the form of salads, soups and stews, fresh or dried, fried or boiled. It contains mucilage in various parts of the plant, which is associated with other important substances including tannins (Woolfe, Chaplin, & Otchere, 1977). Okra contains proteins, carbohydrates and vitamins (Lamont, 1999) that plays a substantial role in human health (Saifullah & Rabbani, 2009) and nutritional security. Consumption of fresh young and green immature okra fruits is very important as fresh fruits, and it can be consumed in different forms (Ndunguru & Rajabu, 2004) such as boiled, fried or cooked. Okra seeds contain about 20% protein and 20% oil (Tindall, 1983). The seeds can be dried and the dried seeds are a nutritious material that can be used to prepare vegetable curds or roasted and ground to be used as coffee additive or substitute (Moekchantuk & Kumar, 2004). Moreover, it was reported that okra leaves can be used as animal feed. In similar fashion, the green leaf buds and flowers are also edible (Doijode, 2001). The stalk/stem of okra can be used for fences as well as firewood in the rural areas in Ethiopia.

Okra is a traditional crop, which requires relatively low agronomic input, but can contribute substantially to sustainable agricultural production and productivity in South Africa and beyond. This species is under-utilized and have the potential for contributing towards food security, with increased nutritional and health benefits for an alarmly fast growing population. It is a valuable source of nutrients (Yang & Keding, 2009) with important medicinal properties (Hilou, Nacoulma, & Guiguemde, 2006). Okra also has the potential to contribute a vital role in income generation and poverty alleviation. It was reported that micronutrient malnutrition, specially the deficiencies in iron, and zinc, affects more than three billion people in the world (Wang, Yin, Tanaka, Tanaka, & Tsujimoto, 2011). In South Africa, one out of five children is stunted and one out of ten children is underweight, due to food malnutrition, which leads to chronic nutritional deprivation (Department of Health, 2010). Therefore, this preliminary study will identify the genotypes with high micronutrient content. Improving the genetic potential of indigenous fruit vegetables like okra species is of paramount

importance for micronutrient concentration. Evaluation and characterization of okra germplasm is important to the breeders who desire sources of genes for novel nutritional traits such as iron, zinc and manganese. This fruit vegetable crop is essential for human nutrition since it is an excellent source of mineral elements and protein. Breeding for high nutritional quality in okra remained largely untapped and is at an infant stage in our breeding programmes. This fruit vegetable crop can thus play a significant role in addressing issues of food security and malnutrition in South Africa. To date, there are no reports of any improved cultivars developed in the country for high micronutrient contents, micronutrient screening and characterization in okra plants and this is the first report on this crop. Improvement of nutritional quality of different okra genotypes in South Africa requires information of the magnitude of genetic variation and genetic heritability present in okra, which will lead to development of new cultivar for the traits of interest to reduce malnutrition. Hence, the objective of the study was to assess the level of micronutrient variability, heritability estimates and select superior parental lines for future nutritional quality breeding programme in South Africa.

2. Material and methods

Forty-six okra genotypes were obtained from the AVRDC-World Vegetable Center, Taiwan and were planted at the ARC research farm at Roodeplaat (25° 59' S; 28° 35' E) in South Africa during the 2016/2017 cropping season. The research farm is situated at an altitude of 1,168 m above sea level. Roodeplaat has annual maximum average temperature ranging from 15.38 to 30.36°C and receives an average annual rainfall of 584.21 mm. The experimental site has loam clay type of soil. Each okra genotypes were planted, as two seeds in three rows of 4 m length spaced at 0.85 m between rows and 0.4 m apart between the plants and the seedlings were thinned into one when fully established at the field. A randomized complete block design with three replications were applied. Trial management such as plot preparation, and hand weeding were done when required and supplementary irrigation was employed when rainfall is not enough for the growth and development of the crop under research. Fertilizer was not applied to simulate low-input conditions for the production of okra for climate change.

The fresh and immature fruits from three randomly selected plants per plot from three replicates were harvested and analysed for selected micro-elements (iron, zinc and manganese) at the analytical laboratory of the ARC in Pretoria, South Africa. The samples were oven dried at 70°C for 24 h, ground into fine powder, and 0.5 g samples were used for analysis. Fe²⁺, Zn²⁺ and Mn²⁺ contents in the samples of immature green fruits were then determined using the Inductively Coupled Plasma Optical Emission Spectrometric method (Zasoski & Burau, 1997).

2.1. Data and analysis

The nutritional data were subjected to estimate genetic variability using Agronomix (2008) computer software. The means of okra genotypes were compared by the least significance difference (LSD) at 0.01 probability level for these mineral elements. Furthermore, genetic parameters such as genetic and phenotypic variances, genetic and phenotypic coefficient of variations, and expected genetic advance under selection were estimated using the functions suggested by Farshadfar, Romena, and Safari (2013), Uguru (1995) and Comstock and Robinson (1952) as follows:

Genetic variance (V_g) = $MS_g - Mse/r$, where MS_g = Mean squares of genotype; Mse = Mean squares of error; r = number of replications

Phenotypic variance (V_p) = $V_g + MSe$

Genotypic coefficient of variation (GCV) = $(\sqrt{V_g}/X)100$ and

Phenotypic coefficient of variation (PCV) = $(\sqrt{V_p}/X)100$, where X is the grand mean for the phenotypic traits; V_g = genotypic variance; V_p = phenotypic variance.

Broad sense heritability (h^2_{bs}) = $(V_g/V_p) \cdot 100$.

Genetic advance under selection (G_s) = $(k) (\sqrt{V_p}) (V_g/V_p)$, where the selection differential, k , is based on the mean phenotypic values of the selected genotypes and of the base population, $k = 2.063$.

Genetic advance as percentage mean is calculated as, $GA\% = GA/X \cdot 100$.

3. Results and discussion

3.1. Genetic variability

The ranges of mean values, mean squares, coefficient of variation, and genetic parameters as well as heritability estimates for the micro-mineral elements studied are presented in Table 1. Substantial variability was observed from the magnitudes of the ranges of means, PCV and GCV for the concentration of micronutrient elements (Fe, Zn, and Mn) evaluated. This difference is an indication of the inherent genetic variation among the okra genotypes. Furthermore, this could provide the genetic background information for the future genetic studies in okra quality breeding programme in South Africa. Screening and characterization of existing germplasm collection of the ARC in South Africa for nutritional variability such as most limiting factors in diets of resource poor farmers is a first step to include nutritional objectives in our breeding programme. This would also play a great role in biofortification to increase the micronutrient content in the okra breeding programme, and is currently receiving widespread attention for the opportunities it creates to tackle hidden hunger.

Highly significant differences were observed for the concentration of these selected micro-mineral elements (Table 1) in the immature and green fruits of okra genotypes evaluated and exhibited large value ranges. The existence of this wide genetic variability among the genotypes would help in selection of potential parental genotypes in the improvement programme of okra breeding programme in the ARC. Inuwa, Ajeigbe, Muhammad, and Mustapha (2012), Shegro, Shargie, van Biljon, and Labuschagne (2012), Gerrano, Adebola, Jansen van Rensburg, and Laurie (2015) and Gerrano, Jansen van Rensburg, and Adebola (2017) reported that the existence of high genetic variability is a pre requisite in a pre-breeding programme for effective and efficient selection of the traits of interest for improvement. The highest average mean value was recorded in Fe compared to Zn and Mn. The variation in the concentration of these mineral elements might be due to the variation in genetic

Table 1. Mean squares, genetic parameters and heritability estimates for the concentration of selected microelements in the immature and green fruits of okra

Genetic parameters	Fe	Zn	Mn
Range of mean values (mg/100 gm)	11.83–74.65	2.694.81	1.50–3.84
Grand mean of base population (X)	33.32	3.83	2.37
Genetic variance (V_g)	63,164.88	75.10	63.40
Phenotypic variance (V_p)	63,254.92	77.98	63.58
Phenotypic coefficient of variability (PCV)	75.48	23.04	33.64
Genetic coefficient variability (GCV)	75.42	22.62	33.60
Broad sense heritability (H _{bs})	99.86	96.32	99.72
Phenotypic standard deviation	251.33	8.83	7.97
Genetic advance under selection (G_s)	517.76	17.55	16.40
Genetic advance (GA) as percentage of mean	155.38	45.79	69.20
Mean squares for genotypes	63,194.89**	76.06**	63.46**
Mean squares for error	90.04	2.87	0.18
LSD (1%)	15.39	2.79	0.69
CV (%)	2.8	4.5	1.8

**Highly significant at 1% probability level.

factors as well as the soil-nutrient absorption capacity of the genotypes into the plant system (Shegro et al., 2012). Liu et al. (2003) similarly reported the significant differences among rice cultivars in the absorption and translocation of Fe, Mn, Zn, Cu, and Mg.

3.2. Genetic parameters

The heritability estimates provides a background genetic information on the inheritance of traits from parents to progenies as well as the amount of genetic progress that can be made in selection of a trait or traits of interest (Sabesan, Suresh, & Saravanan, 2009); while genetic advance under selection is helpful in detecting the actual genetic gain expected (Ogunniyan & Olakojo, 2014). Heritability estimates in broad sense, PCV, GCV, genetic and phenotypic variances, as well as genetic advance under selection are presented in Table 1. Understanding and knowledge of the magnitude of genetic variability among the okra genotypes for the traits evaluated is very important and a prerequisite for selection of good parental lines to make significantly improvement in okra breeding.

The values for the phenotypic variance was relatively large in magnitude than the corresponding values of genotypic variance for the mineral elements (Table 1) and the highest phenotypic and genotypic variances were observed in Fe. Similarly, the values for PCV was relatively greater than that of their respective GCV values. The PCV ranged between 23.04% for Zn and 75.48% for Fe. Similarly, the GCV varied from 22.62% for Zn and 75.42% for Fe. In general, the PCV values were relatively higher than the GCV values for all the traits, which might be due to the environmental factors. The highest PCV and GCV helps in the selection of the promising parental lines for breeding. However, the low PCV and GCV values observed in this experiment revealed that the breeders would look for the genetic sources of high variability for the traits of interest to develop new improved okra cultivar. This finding was similar to what Kowsalya and Raveendran (1996) found. Heritability in broad sense was estimated for the selected micronutrient elements in the green and immature fruits of okra genotypes. The estimated broad sense heritability varied from 96.32 to 99.86% and the highest was observed in Fe. In this study, highest heritability estimate was observed for all the traits under study indicating that these mineral elements are highly heritable among the genotypes evaluated to the progenies.

According to Bello, Abdulmalik, Afolabi, and Ige (2010), the traits that showed a high heritability estimate values would respond positively to a selection pressure in the breeding programme. Those micro-mineral traits, which showed high heritability and high genetic advance as percentage of mean, could be used in selection process since they are controlled by the additive gene action and less influenced by the environmental factors. A high heritability for these mineral traits indicates that these traits are less influenced by the environmental factors and would help selection based on the phenotype expression due to additive genetic value that could be worthwhile for genetic improvement; which are the limiting factors in the diets particularly for rural communities. Moreover, the genetic advance estimates under selection for these mineral elements indicated a great variation for the traits under study. All the mineral elements under evaluation showed high heritability estimates along with high genetic advance under selection as percent population mean, it would be then suitable and important to improve these microelements through selecting the potential okra genotypes as parental lines in the okra-breeding programme. Muhammad et al. (2015) also reported that for efficient selection, combination of high heritability with high genetic advance would provide a clear base line on the reliability of that specific trait in the selection of the genotypes. A combined consideration of GCV, broad sense heritability estimates and genetic advance is important as selection criteria and hence all the traits combined high heritability estimates with high (Fe) and moderate (Zn and Mn) GCV and genetic advance, which could be explained by additive gene action in the improvement programme.

4. Conclusion

The wide range of genetic diversity found in this preliminary study makes a strong case that future breeding efforts of okra can contribute to food, and nutritional security, health benefit and income diversification in the subsistence farming system that predominate in the different parts of the

world. Quantification and identification of micronutrient contents in immature green fruits of okra is important for improvement of the traits of interest. Based on the results of the present study, okra genotypes showed a considerable range of genetic differences for Fe, Zn and Mn studied which could be exploited for future use in nutritional quality breeding programme in South Africa.

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Competing Interests

The author declares no competing interest.

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