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Research Application Summary

Characterization of tomato accessions for breeding research in Kenya

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Abstract

Understanding the phenotypic traits of germplasm through cultivation and assessing the morphological and agronomic characters facilitates selection of suitable parents for a breeding programme. Although tomato is the leading vegetable crop in Kenya, little work has been done to develop improved tomato varieties with farmer preferred and market demanded traits. The objective of this study was to characterize new sources of resistance to tomato bacterial wilt and other traits, farmer selections and commercial varieties to identify potential parental lines for university of Nairobi breeding program. Study genotypes were evaluated for various morphological and agronomic traits including reaction to bacterial wilt. The experiments were conducted at Kabete Field Station, University of Nairobi, Mwea Research Station of the Kenya Agricultural and Livestock Research Organization (KALRO), and in a bacterial wilt infested sick plot in a farmer's field at Kanyei location, Kirinyaga County in 2017 and 2018. Study materials were 10 genotypes including accessions with genes for resistance to bacterial wilt, farmers' selections and commercial varieties. Trials were laid out in a randomized complete block design with three replicates. Data were recorded on days to flowering, days to maturity, fruits per truss, fruits diameter, fruit length, fruits per plant, number of locules per fruit, average weight of fruit and resistance to bacterial wilt. Bacterial wilt incidence and severity were scored on a 0 to 5 scale (0 = no disease symptoms, 5 = entire plant dead) for two seasons. There were significant differences ($P<0.05$) for days to flowering, days to maturity, fruit diameter, fruit length, average fruit weight, number of fruits per plant, number of trusses per plant and the yield per hectare. High resistance to *Ralstonia solanacearum* was recorded on parent lines AVTO1424, AVTO1429, and AVTO1314. Moderate resistance to bacterial wilt was recorded on parent lines Danny select and Valoria select, and UC82 recorded moderate susceptibility, Cal J VF and Riogrande were susceptible while tomato Roma VF (location severity index of 4.75 on sick plot) was highly susceptible. The recorded growth parameters were : mean days to flowering: 34.50 - 40.83 days, days to maturity: 77 - 91.17 days, fruit diameter: 71.16 - 41.17 cm, fruit length: 59.83 - 46.54 cm, average fruit weight: 115-35.20 gm, number of fruits per truss: 2.83 - 4.74, number of trusses per plant: 20.10 - 30.0, yield per hectare: 59260 - 9547 kg per hectare, growth type: determinate and indeterminate, number of locules: 11.10 - 2.17 and plant stem colour: green and purple. There was high genetic variation among the test genotypes for the traits measured. This provides variability that can be exploited in a tomato breeding programme.

Key words: Host resistance, Kenya, phenotypic traits, *Ralstonia solanacearum*, tomatoes

Résumé

Comprendre les traits phénotypiques du matériel génétique par la culture et évaluer les caractères morphologiques et agronomiques facilite la sélection des parents appropriés pour un programme de sélection. Bien que la tomate soit la principale culture légumière au Kenya, peu de travail a été fait pour développer des variétés de tomates améliorées avec les caractéristiques préférées des agriculteurs et demandées par le marché. L'objectif de cette étude était de caractériser de nouvelles sources de résistance au flétrissement bactérien de la tomate et à d'autres caractères, des sélections d'agriculteurs et des variétés commerciales pour identifier les lignées parentales potentielles pour le programme de sélection de l'université de Nairobi. Les génotypes de l'étude ont été évalués pour divers caractères morphologiques et agronomiques, y compris la réaction au flétrissement bactérien. Les expériences ont été menées à la Station au terrain de Kabete, l'Université de Nairobi, la Station de recherche de Mwea de l'Organisation de Recherche Agricole et d'Elevage du Kenya (KALRO), et dans une parcelle malade infestée de flétrissement bactérien dans un champ d'un agriculteur à Kanyei, dans le comté de Kirinyaga en 2017 et 2018. Le matériel d'étude était composé de 10 génotypes, y compris des accessions avec des gènes de résistance au flétrissement bactérien, des sélections d'agriculteurs et des variétés commerciales. Les essais ont été présentés dans un dispositif de bloc complet randomisé avec trois répétitions. Les données ont été enregistrées sur les jours de floraison précoce, les jours de maturité précoce, les fruits par grappe, le diamètre des fruits, la longueur des fruits, les fruits par plante, le nombre de locules par fruit, le poids moyen des fruits et la résistance au flétrissement bactérien. L'incidence et la sévérité du flétrissement bactérien ont été notées sur une échelle de 0 à 5 (0 = aucun symptôme de maladie, 5 = plante entière morte) pendant deux saisons. Il y avait des différences significatives ($P < 0,05$) pour les jours de floraison précoce, les jours de maturité précoce, le diamètre des fruits, la longueur des fruits, le poids moyen des fruits, le nombre de fruits par plante, le nombre de grappes par plante et le rendement par hectare. Une résistance élevée à Ralstonia solanacearum a été enregistrée sur les lignées parentales AVTO1424, AVTO1429 et AVTO1314. Une résistance modérée au flétrissement bactérien a été enregistrée sur les lignées parentales Danny Select et Valoria Select, et UC82 a enregistré une susceptibilité modérée, Cal J VF et Riogrande étaient susceptibles tandis que la tomate Roma VF (indice de sévérité de la localisation de 4,75 sur la parcelle malade) était très susceptible. Les paramètres de croissance enregistrés étaient: jours moyens de floraison précoce: 34,50 - 40,83 jours, jours de maturité précoce: 77 - 91,17 jours, diamètre du fruit: 71,16 - 41,17 cm, longueur du fruit: 59,83 - 46,54 cm, poids moyen du fruit: 115-35,20 g, nombre de fruits par grappe: 2,83 - 4,74, nombre de grappes par plante: 20,10 - 30,0, rendement par hectare: 59260 - 9547 kg par hectare, type de croissance: déterminé et indéterminé, nombre de locules: 11,10 - 2,17 et couleur de la tige de la plante: vert et violet. Il y avait une forte variation génétique entre les génotypes testés pour les caractères mesurés. Ceci fournit une variabilité qui peut être exploitée dans un programme de sélection de tomates.

Mots clés: Résistance de l'hôte, Kenya, caractères phénotypiques, *Ralstonia solanacearum*, tomates

Introduction

Tomato (*Solanum lsycopersicum* L) has been rated as the second most important vegetable crop in Kenya after potato, and is commercially important to many small-scale farmers, processing industry and consumers in Kenya. However, the increasing demand for tomatoes has not matched with the production because of low yield estimated at 18.7 t ha^{-1} compared to 56 t ha^{-1} in China and 56 t ha^{-1} in Morocco (FAO, 2016). Tomato is an introduced crop in Kenya but a key crop with promising potential in horticultural expansion and development in Kenya. It has been recorded to contribute 14% of the total vegetable produced and subsequently 6.72% of entire horticultural crops. The area under tomato production in Kenya increased to 24,531 ha in 2016 from 21,874 ha in 2012. Even with increase in area under production, the total production has declined from 444,862 tonnes in 2012 to 410,033 tonnes in 2016. Fruit yield has declined from 20 t ha^{-1} in 2012 to 18.7 t ha^{-1} in 2016 (FAO, 2016).This decline in productivity is due to various biotic stresses mainly diseases and insect pests, and abiotic stresses such as high temperatures, high humidity, excessive rainfall in some regions, low light intensity and low nutrients in the soil. Most of the seed merchants import tomato seeds from abroad making seed expensive, and with reduced accessibility to small-scale growers. Some of the imported varieties are susceptible to biotic and abiotic factors. Tomatoes have multiple uses including in fresh markets and processing. Preferred traits differ with markets and end-users. The main goal of breeders is to develop varieties that have traits which are demanded by target users and markets. The tomato varieties vary in terms of yield, agronomic and commercially desirable traits and this makes it necessary to understand the morphological and agronomic traits before undertaking any genetic improvement for important agronomic traits (Huang *et al.,* 2012).

Small-scale farmers continue to intensify tomato cultivation to make up for reduced yields through off-season cultivation. This results in diseases and pests build-up, further complicating the production system. Farmers have continued to over rely on own saved seeds and other informal sources because the imported seeds are expensive and therefore are not accessible to the majority of the produces. As a result, many continue to rely on out-dated low yielding, disease susceptible varieties such as Roma VF, Marglobe, Moneymaker, Riogrande and uncertified farm selected seeds (Fufa *et al.,* 2009). Breeding programmes in Kenya have focused on improvement of cereals, root crops, beverage crops and pulses. There is a paucity of information on tomato breeding programme initiated by public or private company in Kenya. The only well documeted tomato breeding programme in the East African region is the Asian Vegetable Research and Development Centre (AVRDC) programme centred in Arusha, Tanzania (Fufa *et al.,* 2009).

Phenotypic characterization of morphological and agronomic traits including leaf, floral, growth habit, crop yields and yield components provide valuable information for crop improvement programmes because it then becomes possible to analyse the genetic variability amongst the accessions and identify potential parents for hybridization and selection (Valls, 2007). Determination of variability in the local tomato germplasm is critical especially for the utilization, maintenance as well as the acquisition of germplasm resources for use in breeding programmes (Mwirigi *et al.,* 2009). The objective of this study was to characterize ten tomato accessions to facilitate breeding for improved agronomic traits and resistance to bacterial wilt disease casual agent (*Ralstonia solanacearum*).

Materials and Methods

Experimental sites. The field experiment was organized into three sites between 2017 and 2018 at Kabete Field station, University of Nairobi and at Mwea Research Station of Kenya Agricultural and Livestock Research Organization (KALRO) in Kirinyaga county and a bacterial wilt infested farmer's field (Kanyei location, Kanyeki-ini sub-location in Kirinyaga County). Kabete field station experiences a bimodal rainfall (1059 mm per year) and temperature range of 12.3°C to 22.5°C. Kabete site is also characterized by humic nitisols that are deep and well-drained with a pH of about 5.0 to 5.4. The Mwea research station site and farmer's site are both located in Kirinyaga county and experience a bimodal rainfall regime (850 mm) and a temperature range of 15.6°C to 28.6°C and with vertisol soils with a pH of about 5.1. The plant materials were evaluated in a randomized complete block design with three replicates for the three sites.

Plant material and method. Ten tomato genotypes were used in this study. They included three elite breeding accessions from the World Vegetable Centre (AVRDC) in Taiwan namely AVT01424, AVT01429 and AVT01314, four commercial cultivars sourced from Continental Seeds Company Limited (Riogrande, Roma VF, Cal J VF and UC 82), and three selections (Eden, Danny and Valoria) from farmers in Kirinyaga County. The commercial varieties are purelines with good marketable traits but lacking key agronomic traits and resistance to biotic stresses. These selections require further genetic improvement for increased yields, earliness and resistance to major biotic stresses especially bacterial wilt. The accessions from AVRDC Taiwan are donor parents for resistance to major biotic challenges but lacking some traits to allow direct commercialization without further improvements (Fufa *et al.,* 2009).

The ten lines were sown in germination trays and transplanted after 28 days to the field following normal agronomic practices to ensure healthy crop. Diammonium phosphate (DAP) at a rate of 12 g plant^{-1} and 12 g plant^{-1} of N:P:K (17% nitrogen: 17% phosphorus: 17% potassium) was applied during transplanting. Manual cultivation was carried out to keep plots weed free. Metalaxyl-M and Propineb 700 g/kg at the rate of 50 g / 20 litres water were used to control early and late blight per fortnight alternatively. Imidaclopride+Betacyfluthrine100+45 g/L at rate of 0.2L/Ha and Thiamethoxam at the rate of 8gm / 20 litres water were used to control aphids, whiteflies and leaf miners during the crop growth cycle. Supplemental irrigation was provided with drip system as needed.

Sick plot development. The *Ralstonia solanacearum* sick plot was developed in a farmer's field with history of bacterial wilt in Kanyei location, Kanyeki-ini sub location in Kirinyaga County by incorporating chopped debris of diseased plants collected from different farms in the region to build up the inoculum load of the pathogen. Susceptible check variety Roma VF was used to test the pathogen load and establish the location severity index (Jitendra et al., 2004). The dead plant parts after the disease infection were chopped and incorporated back to the soil to further prepare the sick plot for germplasm evaluation. Random samples were taken from the diseased plants for laboratory confirmation of *R. solanacearum*. Adequate infestation of the sick plot was ensured by continuous recycle of *R. solanacearum* plant debris throughout the research period. The sick plot was considered

ideal for *Ralstonia solanacearum* evaluation after attaining location severity index of over 76 % with the susceptible check varieties. The bacterial wilt disease incidence score of 0 - 5 scale (0=no disease symptoms, 1=one leaf wilted, 2=two leaves wilted, 3=three leaves wilted, 4=entire plant wilted except the tip, 5=entire plant dead) was used to rank the germplasm resistance reaction and further classified into percent severity (Jitendra *et al.,* 2004). The location severity index (LSI) was calculated for the field with bacterial wilt using the formula;

$$LSI = \frac{\Sigma \text{ Score x number of genotypes}}{\text{Total number of genotypes observed}} \times 100$$

Data collection and analysis. Morphological and agronomic data were collected at various growing stages for ten quantitative traits following the International Plant Genetic Resources Institute (IPGRI) tomato descriptor for plant height, days to 50% flowering, days to maturity, fruit diameter, fruit length, average fruit weight, number of locules, number of truss per plant and yield per plant. Qualitative traits like presence of green shoulders, stem colour and growth type were recorded. Quantitative trait data were subjected to analysis of variance (ANOVA) using Genstat software (15th edition). Least significant difference (LSD) test was used to compare and separate means of the genotypes at 5% significance P-value thresholds.

Results

There was a significant accession effect for qualitative and quantitative traits and bacterial wilt among the ten accessions.

Days to 50% flowering: The ten accessions showed three categories of maturity, i.e., early maturity in which 50% flowering occurred between 32 - 36 days, medium maturity at 37 - 45 days and late maturity where flowering occured 46 - 50 days (Table 1). Some of the local commercial varieties such as UC82 flowered very early and were comparable to some elite breeding lines. Riogrande and farmer selection of Valoria flowered late. Duration to 50% flowering is important to breeders in managing the flowering harmonization when planning hybridization programmes.

Days to maturity: The ten accessions showed significant variation for 50% days to first harvest. The mean duration to the first harvest varied from 77 days for Danny FS to 91.17 days for Valoria FS for the two growing seasons. Earliness is an important factor especially in rainfed agriculture where the rainfall is not reliable, and crop must take shorter time to be in production.

Presence of green shoulders: Only one genotype (AVTO 1429) showed green shoulders on fruits. Green shoulders refer to darker chlorophyll pigmentation on the upper part of the tomato fruit during the early mature growing phase of fruit but later the fruits turn colour to red. There were no green shoulders on the other nine genotypes.

Stem colour at seedling stage: Three genotypes from AVRDC (AVTO 1424, AVTO 1429, AVTO 1314) had a green stem while other genotypes had purple stem.

Growth habit: One genotype was indeterminate (AVTO 1429), seven genotypes were determinate (UC82, Danny FS, Roma VF, Riogrande, Valoria FS, Eden FS, Roma VF), and two genotypes were

semi-determinate (AVTO 1424, AVTO 1314).

Number of fruits per truss: The mean value for the number of fruits ranged between 2.83 fruits per truss for AVTO1314 to 4.74 fruits for Danny FS.

Number of trusses per plant: The number of fruits per plant varied greatly among the parental accessions. The mean trusses per plant ranged between 20.10 for AVTO1314 to 30.0 (Roma VF).

Fruit diameter: Fruit diameter ranged between 71.16 cm (AVTO 1429) to 41.17 cm (UC82).

Fruit length and number of fruits per plant: The mean fruit length among the parental accessions varied significantly from 59.83 cm (Roma VF) to 46.54 cm (UC 82). The number of fruits per plant varied from 114.70 (AVTO 1314) to 288.70 (Danny FS) (Table 1).

Number of locules per fruit and fruit weight: Number of locules per fruit ranged from 2.17 (Riogrande) to 11.10 (AVTO 1429). Fruit weight varied from 35.20 g (UC 82) to 115g (AVTO 1429). The fruit weight represents a significant market trait of concern useful during fruit grading.

Yield per hectare: Fruit yield varied from 9547 kg ha^{-1} (Roma VF) to 59, 260 kg ha^{-1} (AVTO 1424). Yield is an important trait to any breeder when evaluating genotypes to be included in a hybridization programme for genetic improvement.

Bacterial wilt resistance. The reactions of the ten genotypes to *Rastonia solanacearum* were as shown in Table 2. Three elite genotypes from AVRDC (AVTO 1424, AVTO 1429, and AVTO 1314) recorded highest levels of resistance to bacterial wilt in the infected field. Two local farmer selections (Danny FS and Valoria FS) were moderately resistance. The commercial variety UC82 was moderately susceptible to the bacterial wilt. Two genotypes (Cal JVF, Riogrande) were susceptible. Roma VF was highly susceptible with a severity index of 4.75. The results indicated variability that could be exploited by breeders in developing lines with resistance to bacterial wilt.

Table 1. Mean performance of parental genotypes for growth and yield traits over two seasons

Genotypes	Days to 50% flowering	Days to maturity	No. of fruits per truss	No. of trusses per plant	Fruit diameter (cm)	Fruit length (cm)	No. of fruits per plant	No. of locules per fruit	Average fruit wt. (g)	Yield ($kgha^{-1}$)
AVT01424	36.5	84.2	3.5	22.9	46.9	51.9	247.3	2.6	41.7	59260.0
AVT01429	36.5	81.3	3.4	26.0	71.2	51.9	166.7	11.1	115.0	16187.0
AVT01314	36.5	85.7	2.8	20.1	56.3	49.4	114.7	5.3	59.6	29815.0
Danny FS	34.5	77.0	4.7	23.9	48.2	49.5	288.7	2.5	52.0	53612.0
Valoria FS	40.3	91.2	3.8	23.3	51.6	55.5	246.7	2.4	66.0	48241.0
UC 82	34.0	74.8	4.5	23.7	41.2	46.5	266.3	2.6	35.2	44167.0
Cal J VF	41.2	84.7	3.4	20.9	43.7	49.9	235.3	2.3	51.7	12560.0
Roma VF	39.3	85.8	4.1	30.0	42.7	59.8	236.3	2.7	40.7	9547.0
Eden FS	37.0	83.7	3.5	21.9	45.0	53.7	182.3	3.3	74.1	12480.0
Riogrande	40.8	88.0	3.5	20.6	48.4	58.4	263.7	2.2	65.9	56575.0
Mean	37.7	83.6	3.7	23.3	49.5	52.6	224.8	3.7	60.2	34244.0
$LSD_{0.05}$	0.01	0.01	0.01	0.01	4.3	3.8	34.4	1.4	26.8	4497.0
CV (%) 2.0	1.1	7.8	5.9	13.3	11.1	8.9	55.2	67.7	7.70	

CV: coefficient of variation; LSD: Least significant difference at 5% P-value threshold

Table 2. Disease scores of tomato accessions in a bacterial wilt sick plot at9 Kanyei location, Kanyeki-ini sub-location, Kirinyaga County, 2016 -2017

Bacterial wilt incidence score	Bacterial wilt severity	Resistance reaction	Genotypes
0	0	Highly resistant	AVTO 1424, AVTO 1429, AVTO 1314
1	1-10	Resistant	None
2	11.25	Moderately resistant	Danny FS, and Valoria FS
3	26-50	Moderately susceptible	UC82
4	51-75	Susceptible	CAL JVF, Riogrande
5	76-100	Highly susceptible	Roma
Location severity Index		4.20	

Discussion

Tomato characterization through morphological traits has largely been used in understanding the genetic diversities among the test genotypes. This study shows that there is considerable genetic variation for traits in terms of morphological, phonological and reproductive traits measured among the ten tomato genotypes. The results also showed clear differences for resistance to *Rastonia solanacearum* which could be utilized for breeding for different traits (Dagnoko *et al.,* 2013). There was variability in the days to flowering that necessitates breeders to consider during sowing seeds and transplanting for hybridization purposes. Days to maturity which refers to time taken from transplanting to onset of harvesting varied among the genotypes and this make it possible to design breeding objective for earliness. The growth habit among the genotypes varied greatly indicating opportunities for developing new lines with determinate, semi-determinate and indeterminate growth habits.

The results displayed variations in the fruit traits ranging from fruit diameter and length and number of fruits per truss. The market puts lots of interest in the fruit characters. The variability observed is valuable to breeders in developing potential varieties with improved marketable fruit traits to meet diverse preferences (Ogwu *et al.,* 2017). Significant variation exists in these genotypes for fruit weight, fruit yield, and yield components presenting a great opportunity to improve the yield potential of some local commercial varieties like Roma VF, Cal JVF and local selection such as Eden FS through hybridization and selection. The genotypes showed wide variation for resistance to bacterial wilt. Resistance varied from high levels in introduced accessions to high susceptibility among the local genotypes. This present great opportunity to breeders for improving bacterial wilt resistance levels of the local susceptible commercial genotypes. The moderately resistant genotypes can be improved for disease resistance through additive gene effects (Monma *et al.,* 1997).

Conclusion

The objective of this study was to characterize local and recently introduced tomato germplasm for morphological and horticultural traits and reaction to infection by bacterial wilt. Results showed that there is adequate variation for all traits studied. The lines can be used for hybridization and

selection programme to develop new varieties which meet farmer, consumer and market demanded traits in Kenya, The differences in the morphological traits and in the reaction to wilt provide valuable information that can be exploited by breeders in selecting parental materials to use in a hybridization scheme or breeding for hybrids. The varying days to flowering pointed out the need to have harmonized flowering when designing a hybridization scheme.

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