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Screening Of a Selected Tomato Varieties for Response to *Ralstoniasolanacearum*INMASENO, Western Kenya

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Abstract:
Tomato (Solanumlycopersicum) is an important horticultural vegetable crop among small and large scale farmers in Kenya, grown for its nutritional and commercial purposes. However, gains from its production is limited by losses resulting from incidences of Bacterial wilt (Ralstoniasolanacearum) which has proved difficult to control and drastically decrease tomato yield and quality by up to 80%. The losses vary widely according to host, cultivar, climate, soil type, cropping practices and pathogen strain. The objectives of this study were to investigate the response of selected tomato varieties to Ralstoniasolanacearum at different stages of growth and their tolerance to bacterial wilt under greenhouse conditions. In addition, the study was aimed at investigating the agronomic performance of selected tomato varieties in Maseno, Western part of Kenya. The study was conducted between January to July 2013 and June to December 2020. The tomato varieties; 'Heirloom Tall vine' from Johnny's seed company, Maine, USA; 'Legend', 'Golden Jubilee', and 'Goliath Pear Hybrid' from Horticultural Products and services Div., WI, USA; and two commercially grown varieties 'Money maker' and 'Cal J', were grown in dystricnitisol sterilized soil medium in a 23 x 16 cm plastic pots; artificially inoculated at seedling stage (4 leaves), start of flowering and at 50% fruiting stage. A control experiment (non- inoculation at seedling, flowering and fruiting stage for each variety) was also included in the study. Disease incidence was scored on a scale of 0 (no symptoms) to 5 (death of the whole plant). The experiment comprised of three factors; six tomato varieties, two treatments (Inoculation and non-inoculation) and three stages of growth (seedling, flowering and fruiting stages) arranged in a 6 × 2 × 3 factorial in a Randomized Complete Block Design with three replications. Data was collected on days to flowering, days to fruiting, maturity period, plant height (cm), total yield, number of wilted plants, number of dead plants and response to BW. Data collected was subjected to Analysis of variance (ANOVA) using SAS Statistical package and effects declared Significant at 5% level. Means were separated using Duncan's Multiple Range Test at 0.05 Significance level. Linear correlation was done to compare the relationship between disease incidences at different stages of growth. The results obtained demonstrated variation between inoculated and non-inoculated varieties. Inoculated varieties took more number of days to flower, fruit, mature, and also produced less number of fruits with low weight (g) than in non-inoculated varieties due to the effects of BW. Goliath pear Hybrid took the shortest time to flower (42 days), fruit (59.17days) and mature (73.33 days) but produced the least number of fruits (146.67). Cal J and Money maker produced the highest number of fruits (209.67 and 191.17 respectively) but were susceptible to the pathogen. The results also showed that Heirloom Tall vine and Goliath pear Hybrid were tolerant to the disease with the lowest mean (2.00) of disease incidence. Fruiting stage had the highest mean (3.67) of disease incidence. There was a high positive correlation in disease incidence at different stages of growth. Goliath pear Hybrid was found to contain desirable characteristics such as; early maturing, large fruit size and was also tolerant to Bacterial wilt, hence a good candidate for Kenyan farmers.
Key Word**: *Ralstoniasolanacearum; Screening; Stage of growth; Tomato; Bacterial wilt; Pathogen.***

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I. Introduction

Tomato (*Lycopersiconesculentum*Mill) is one of the most popular vegetables in the world [13]. It is the world's largest vegetable crop after potato and sweet potato but it tops the list of canned vegetables [12]. It is a very versatile plant and it could either be grown for fresh market or for processing in which mechanical processes are involved [2]. Tomato and its products are rich in antioxidants and considered to be a good source of vitamins C, E and carotenoids, particularly lycopene and β-carotene and other phenolic compounds that protect the body against diseases [14]; [8]. Its fruits are used in salads or cooked as a vegetable, processed into tomato paste, sauce and puree. Like any other vegetable, it is a delicate crop that needs costly optimal management and application of inputs in order to obtain high yields. Among the most important is the cultural management of the notorious bacterial wilt disease. Bacterial wilt disease, caused by *Ralstoniasolanacearum*[18] conventionally classified as races and biovars, Bacterial wilt of tomato is caused by either race 1 or race 3 of *R. solanacearum*with a wide host range and rarely caused by race 2 [7]. The pathogen has caused significant yield and economic loss in tomato production in Kenya. Yield losses caused by bacterial wilt are estimated at 50-100% in tomato production areas in Kenya [11]. In Kenya the pathogen has been reported at both low and high altitudes [10]. Management is difficult due to high variability of the pathogen, limited possibility for chemical management, high capacity of the pathogen to survive in diverse environments and its extremely wide host range [6], hence the need to come up with a proper control method in order to minimize the prevalence pathogen.

According to [3], the pathogen is chiefly soil-borne and enters tomato plants through the roots, colonize vascular tissues, multiply, clog and cause wilting and eventually death of the plant [15]. In the early stages of the disease, the vascular system in the stem of a plant appears yellow or light brown in longitudinal or transverse section and as the disease progresses the system becomes dark brown. When the plant wilts completely, the cortex and the pith also become brown and as the disease progresses, the plant becomes permanently wilted with the entire root system showing brown rot [9]. The recommended control measures include; soil treatment with chemicals, crop rotation, use of tolerant varieties and field hygiene [4].The most effective and economic method of control of the disease is breeding and selection of varieties that are resistant to *R.Solanacearum*.

II. Material And Methods

2.1. Study Site

The study was carried out under greenhouse conditions at Maseno University Research farm which lies along Kisumu – Busiahighway in Western, Kenya. Its geographical coordinates are 0° 10' 0" South, 34° 36' 0" East and the altitude is 1,503 metrers above sea level. Maseno receives both short and long rains averaging 1750mm per annum with mean temperature of 28.7^0C. The study was carried out between January to July 2013 and June to December 2019.

2.2. Plant Materials

Four selected tomato varieties; Heirloom Tall vine from Johnny's seed company USA,Legend, Golden Jubilee, and Goliath Pear Hybrid from Horticultural Products and services USA and two varieties; Cal J and Money maker from Kenya were planted in sterilized soil medium in 23 by 16 cm polythene bags and in plastic buckets.

2.3. Preparation of the soil medium

Soil medium obtained from the field adjacent to the study site in Maseno was heat- sterilized for 30 minutes by placing enough amounts onto the steaming metallic plate, lightly wetted, and covered with a metallic lid. This was heated on a jiko fueled using wood shavings and, turned severely until the soil reaches an average of 100oC. The sterilized soil was then cooled, prepared to a fine tilth, then filled in the 23 x 16 cm plastic pots to three quarter full and labeled for each variety totaling to 324 bags. DAP fertilizer was applied at the rate of 150 kg/ha. One seed of each variety was then drilled 1 cm deep and then lightly covered with fine soil.The soil medium was maintained according to the agronomic requirements for raising tomato seedlings.

Plate 1: soil sterilization by heating using a murrajiko

2.4. Inoculation of Tomato plants

Inoculum from tomato plants showing bacterial wilt symptoms collected from different localities in Kisumu, Siaya, Vihiga and Kakamega was prepared in the microbiology laboratory in Maseno University. The isolates of R. Solanacearum were inoculated on tomato plants under greenhouse conditions. Root inoculation as per Thomas et al. [16] was administered at seedling stage (4 leaves), start of flowering and 50% fruiting stage from the date of sowing. A sterilized scalpel was used to cause sufficient damage to the secondary roots by punching each plant at the base of stem above the upper secondary roots about 1 cm away from the base to ensure that the plant is infected through the roots. Two millimeters of the standard bacterial suspension inoculum prepared in autoclaved distilled water according to [16] was then be poured over the wounded roots to augment natural infestation in every pot for 162 pots. The other half of the 162 plants were left untreated (control experiment). After inoculation, all pots were covered by polythene bags for 24 hours to maintain high humidity [1] for both inoculated and non-inoculated.

2.5. Experimental Design

The experiment was arranged in a 6 ×2 ×3 factorial laid out in a randomized complete block design (RCBD) with three replications. There were two treatments; Inoculation and Non – inoculation (control experiment) with three stages of growth; seedling stage (4 leaves), start of flowering and at 50% fruiting stage.

2.4 Data collection

Data on days to flowering, days to fruiting, days to maturity, plant height (cm), number of fruits, weight of fruits (g), number of wilted plants at different stages of growth, number of dead plants at different stages of growth and disease incidence at different stages of growth was collected.

BW scores were taken once in a week, at seedling stage (4 leaves), start of flowering and at 50% fruiting stage on a scale of 0 – 5, where; 0 = no symptom of bacterial wilt, 1 = one leaf partially wilted, 2 = one to two leaves wilted, 3 = all except 2 – 3 lower leaves wilted, 4 = all leaves wilted, 5= death of the whole plant. The number of wilted plants for each variety was then recorded and graded on a 0 – 5 scale with a modified rating scale given below to show resistance level in each variety [21]

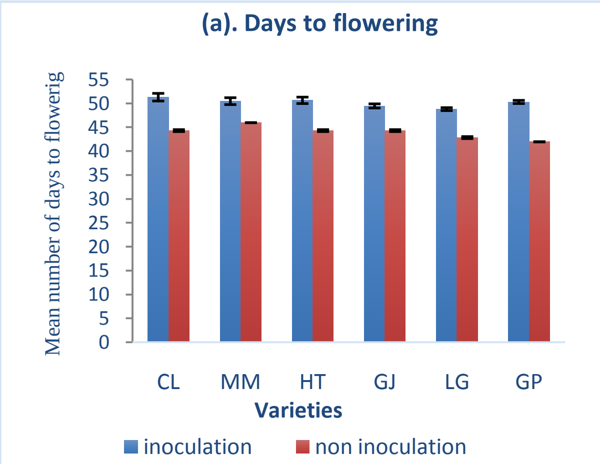
0 - Highly resistant (HR): Plants did not show any wilt symptoms
1 - Resistant (R): 1 – 20 % disease incidence
2 - Moderately resistant (MR): 21 - 40 % disease incidence
3 - Moderately susceptible (MS): 41 – 60 % disease incidence
4 - Susceptible (S): 61 – 80 % disease incidence
5 - Highly susceptible (HS): more than 80 % disease incidence

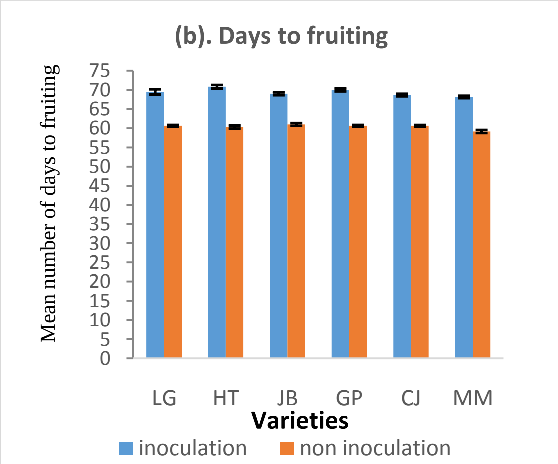
2.6. Statistical analysis

Statistical analysis of data was conducted using SAS 9.1 package.The count data on disease incidence on plants was log-transformed and aregression analysis performed using PROC REG in SAS. Data on growth parameters and disease incidence at the three stages of growth were analyzed using analysis of variance (ANOVA). Means that were considered significantly different ($P \leq 0.05$) were separated using Duncan's Multiple Range Test LSD.

III. Result

3.0 Growth parameters





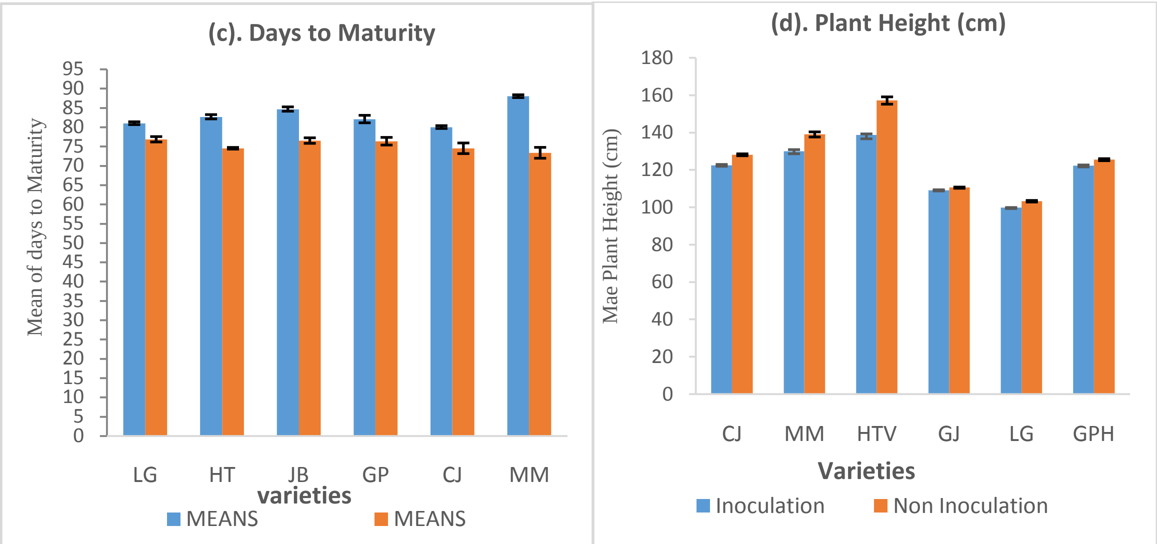


Fig1: Variation in growth parameters in all tomato varieties evaluated in two treatments. Error bars represent standard error of means calculated from six varieties per replicate. CJ = Cal J, MM= Money maker, HTV= Heirloom Tall vine, GJ = Golden Jubilee, LG =Legend, GPH= Goliath Pear Hybrid.

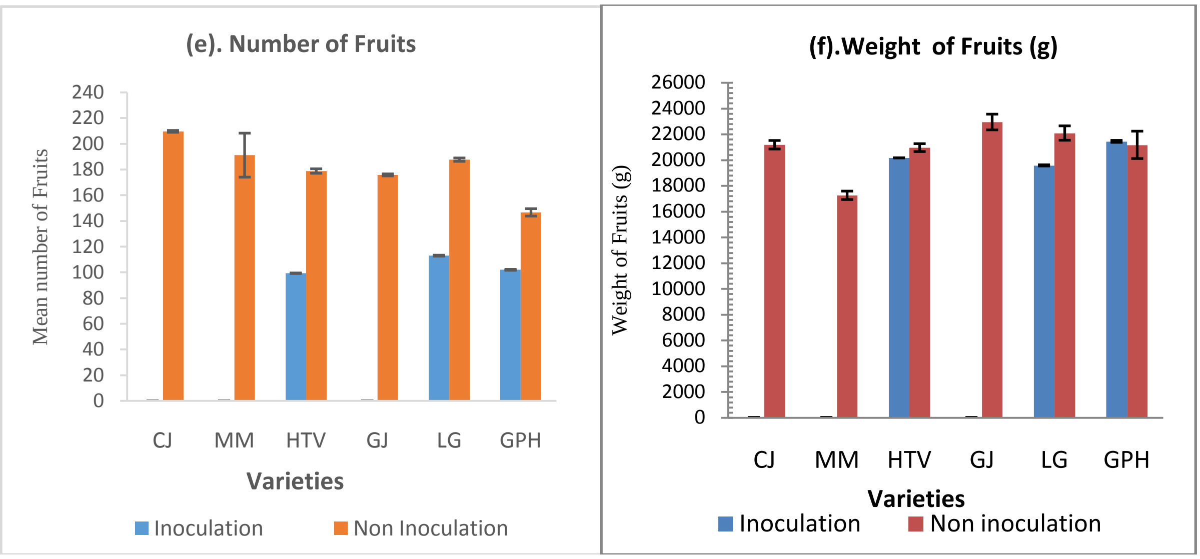


Fig1: Continued

3.1. Number of days to Flowering.

Table .1. Mean number of days to Flowering

VARIETY	INOCULATION	NON-INOCULATIN
Cal J	51.33^{a} ± 0.80	44.33^{b} ± 0.21
Money Maker	50.50^{ab} ± 0.72	46.00^{a} ± 0.00
Heirloom Tall vine	50.67^{ab} ± 0.67	44.33^{b} ± 0.21
Golden Jubilee	49.50^{bc} ± 0.43	43.33^{c} ± 0.21
Legend	48.83^{c} ± 0.31	42.67^{d} ± 0.21
Goliath pear Hybrid	50.33^{ab} ± 0.33	42.00^{e} ± 0.00
Mean	**50.19**	**43.78**
$LSD_{5\%}$	**1.297**	**0.00**
CV (%)	**2.00**	**0.00**

The mean number of days to flowering were determined at P≥0.05 significant level using DMRT. Means followed by the same letters in the same column are not significantly different (P≥0.05) according to DMRT.

In Table 1 and Fig 1(a). in Inoculation, Legend was significantly (p< 0.05) the first to flower (48.33 days) followed by Golden Jubilee (49.50 days), Goliath pear Hybrid (50.33 days), Money maker (50.50 days) and Heirloom Tall vine (50.67 days) respectively [Fig 1(a)]. Cal J was significantly (p< 0.05) the last to flower taking an average of 51.33 days. Golden Jubilee was significantly (p<0.05) different to Legend in means of number days to flowering. Heirloom Tall vine and Cal J were not significantly (p>0.05) different to Goliath Pear Hybrid in mean of number of days to flowering.A highly significant (p< 0.05) difference was observed in both experiments in means of number of days to flowering.

In non-Inoculation, Goliath Pear Hybrid was significantly the (p< 0.05) the first variety to flower (42.00 days) followed by Legend (42.67 days), Golden Jubilee (43.33 days) and, Cal J and Heirloom Tall vine which both took 44.33 days. Money maker was significantly (p< 0.05) the last to flower, which took an average of 46 days. Treatment differences were highly significant (p>0.001) in mean of number of days to flowering.

3.2. Number of days to Fruiting

Table 2. Mean number of days to Fruiting

VARIETY	INOCULATION	NON-INOCULATIN
Cal J	$69.50^{bc} \pm 0.67$	$60.67^{ab} \pm 0.21$
Money Maker	$70.83^{a} \pm 0.47$	$60.33^{b} \pm 0.42$
Heirloom Tall vine	$69.00^{cd} \pm 0.37$	$61.00^{a} \pm 0.37$
Golden Jubilee	$70.00^{b} \pm 0.37$	$60.67^{ab} \pm 0.21$
Legend	$68.67^{dc} \pm 0.33$	$60.50^{ab} \pm 0.22$
Goliath pear Hybrid	$68.17^{e} \pm 0.31$	$59.17^{c} \pm 0.40$
Mean	**69.36**	**60.39**
$LSD_{5\%}$	**0.614**	**0.598**
CV (%)	**0.68**	**0.77**

The mean number of days to fruiting were determined at P≥0.05 significant level using DMRT. Means followed by the same letters in the same column are not significantly different (P≥0.05) according to DMRT.

In Table 2 and Fig 1 (b). In Inoculation; Goliath Pear hybrid was significantly (p< 0.05) the first variety to fruit, which took an average of 68.17 days. Legend was the second variety to flower (68.67 days) followed by Heirloom Tall vine (69.00 days), Cal J (69.50 days) and Golden Jubilee (70.00 days) respectively. Money maker was significantly (p< 0.05) the last variety to fruit taking an average of 70.83 days. All the varieties were significantly (p< 0.05) different from each other in means of number of days to fruiting. A highly significant (p< 0.001) difference in means of number of days to fruiting was observed between experiment 1 and experiment 2.
In Non-Inoculation, Goliath pear Hybrid was also significantly (p< 0.05) the first variety to fruit taking an average 59.17 days followed by Money maker (60.33 days), Legend (60.50 days), Cal J, and Golden Jubilee both taking an average of 60.67 days to fruit. Money maker was significantly (p< 0.05) the last variety to fruit taking an average of 61.00 days. There was no significant (p> 0.001) difference in means of number of days to fruiting in experiment 1 and experiment 2. Treatment differences were highly significant (p>0.001) in means of number days to fruiting.

3.3. Number of days to Maturity

Table 3. Mean number of days to Maturity

VARIETY	INOCULATION	NON-INOCULATIN
Cal J	$81.00^{e} \pm 0.37$	$76.83^{a} \pm 0.70$
Money Maker	$82.67^{c} \pm 0.56$	$74.50^{b} \pm 0.22$
Heirloom Tall vine	$84.67^{b} \pm 0.56$	$76.50^{a} \pm 0.72$
Golden Jubilee	$82.07^{c} \pm 0.97$	$76.33^{a} \pm 0.99$
Legend	$80.00^{f} \pm 0.37$	$74.50^{b} \pm 1.38$
Goliath pear Hybrid	$88.00^{a} \pm 0.37$	$73.33^{c} \pm 1.41$
Mean	**83.06**	**75.33**

$LSD_{5\%}$	**0.00**	**0.678**
CV (%)	**0.00**	**0.69**

The mean number of days to fruiting were determined at P≥0.05 significant level using DMRT. Means followed by
the same letters in the same column are not significantly different (P≥0.05) according to DMRT. NB: CJ – Cal J, MM – Money maker, HTV – Heirloom Tall vine, GJ – Golden Jubilee, LG – Legend, GPH – Goliath Pear Hybrid.

In Table 3 and Fig 1(c). In inoculation; Legend was significantly (p< 0.05) the first variety to mature (80.00 days) followed by Cal J (81.00 days), Golden Jubilee (82.07 days), Money maker (82.67 days) and Heirloom Tall vine (84.67) respectively. Goliath pear Hybrid was significantly (p< 0.05) the last variety to mature which took an average of 88.00 days. Money maker was not significantly (p> 0.05) different to Golden Jubilee in means of number of days to maturity while the other four varieties were significantly (p< 0.05) different to each other in means of number of days to maturity. There was no significant (p> 0.001) difference in experiment 1 and experiment 2 in means of number of days to maturity.

In non-inoculation, Goliath pear Hybrid was significantly (p< 0.05) the first variety to mature which took an average of 73.33 days followed by Money maker and Legend which both took an average of 74.50 days, Golden Jubilee (76.33 days), Heirloom Tall vine (76.50 days) and Cal J (76.83 days) respectively. Cal J was not significantly (p> 0.05) different to Heirloom tall vine and Golden Jubilee while Money maker was not significantly (p> 0.05) different to Legend in means of number of days to maturity. Goliath pear Hybrid was significantly (p< 0.05) different to the other varieties in mean of number of days to maturity.
There was a significant (p< 0.001) difference in mean of number of number of days to maturity in experiment 1 and experiment 2. Maturity period differed significantly (p<0.001) in mean of number of days in treatments.

3.4. Plant Height

Table.4. Mean of Plant Height (cm)

VARIETY	INOCULATION	NON-INOCULATIN
Cal J	$122.53^{c} \pm 0.48$	$128.17^{c} \pm 0.51$
Money Maker	$130.07^{b} \pm 0.87$	$139.05^{b} \pm 1.37$
Heirloom Tall vine	$138.68^{a} \pm 0.67$	$157.20^{a} \pm 1.96$
Golden Jubilee	$109.12^{d} \pm 0.35$	$110.58^{d} \pm 0.33$
Legend	$99.83^{e} \pm 0.14$	103.32e ± 0.43
Goliath pear Hybrid	$122.22^{c} \pm 0.51$	$125.55^{c} \pm 0.55$
Mean	**120.41**	**127.31**
$LSD_{5\%}$	**1.202**	**3.076**
CV (%)	**0.78**	**1.88**

The mean number of plants height were determined at P≥0.05 significant level using DMRT. Means followed by the same letters in the same column are not significantly different (P≥0.05) according to DMRT.

In Table 4 and Fig 1(d). In inoculation; Heirloom Tall vine had significantly (p< 0.05) the longest height of 138.68 cm followed by Moneymaker (130.07cm), Cal J (122.53 cm), Goliath pear Hybrid (122.2 cm) and Golden Jubilee (109.12 cm) respectively. Legend had significantly (p< 0.05) the shortest plant height with an average height of 99.83 cm. Cal J was not significantly (p> 0.05) different to Goliath pear Hybrid in mean of plant height. Money maker, Heirloom Tall vine, Golden Jubilee and Legend were significantly (p< 0.05) different to each other in means of plant height in cm. There was no significant (p> 0.05) difference in experiment 1 and experiment 2 in means of plant height (cm). There was no significant (p> 0.05) difference in means of plant height in both experiment 1 and experiment 2.

In non-inoculation, Heirloom Tall vine was also significantly (p< 0.05) the tallest plant with an average height of 157.20 cm followed by Money maker (139.05cm), Cal J (128.17 cm), Goliath pear Hybrid (125.55 cm) and Golden Jubilee (110.58 cm) respectively. Legend was significantly (p< 0.05) the shortest plant with an average height of 103.32 cm. There was no significant (p> 0.001) difference in height (cm) of plants in both experiment 1 and experiment 2. There was a highly significant (p<0.001) variation in mean of plant height in treatments.

3.5. Number of Fruits

Table 5. Mean Number of Fruits

VARIETY	INOCULATION	NON-INOCULATIN
Cal J	$0.00^{d} \pm 0.00$	$209.67^{a} \pm 0.72$
Money Maker	$0.00^{d} \pm 0.00$	$191.17^{a} \pm 17.11$
Heirloom Tall vine	$99.33^{c} \pm 0.21$	$178.83^{b} \pm 1.74$
Golden Jubilee	$0.00^{d} \pm 0.00$	$175.83^{b} \pm 0.79$
Legend	$113.00^{a} \pm 0.37$	$187.67^{b} \pm 1.33$
Goliath pear Hybrid	$102.00^{b} \pm 0.37$	$146.67^{c} \pm 2.87$
Mean	**104.33**	**181.64**
$LSD_{5\%}$	**0.428**	**21.601**
CV (%)	**0.311**	**9.24**

The mean number of fruits were determined at P≥0.05 significant level using DMRT. Means followed by the same letters in the same column are not significantly different (P≥0.05) according to DMRT.

In Table 5 and Fig 1(d). In inoculation; Legend had significantly (p< 0.05) the highest mean (113.00) of number of fruits followed by Goliath pear Hybrid (102.00), while Heirloom Tall vine had significantly (p< 0.05) the lowest mean of number of fruits. Cal J, Money maker and Golden Jubilee had a mean (0.00) of number of fruits because they all succumbed to the effects of bacterial wilt hence no fruits were harvested. Legend, Goliath pear Hybrid and Heirloom Tall vine were significantly (p< 0.05) different in means of number of fruits. There was no significant (p> 0.001) difference in experiment 1 and experiment 2 in means of number of fruits.

In non-inoculation, Cal J had significantly (p< 0.05) the highest mean number of fruits which was 209.67 followed by Money maker (191.17), Legend (187.67), Heirloom Tall vine (178.83) and Golden Jubilee (175.83) respectively. Goliath pear Hybrid had significantly (p< 0.05) the lowest mean (146.67) of number of fruits. Cal J was not significantly (p> 0.05) different to Money maker in mean of number of fruits while Heirloom Tall vine, Golden Jubilee and Legend were not significantly (p> 0.05) different in means of number of fruits produced. Goliath pear Hybrid was significantly (p< 0.05) different to the other varieties in means of number of fruits produced. There was no significant (p> 0.001) difference in means of number of fruits in experiment 1 and experiment 2.

3.6. Weight of Fruits

Table 6. Mean Weight of Fruits (grams)

VARIETY	INOCULATION	NON-INOCULATIN
Cal J	$0.00^{d} \pm 0.00$	$21170.63^{b} \pm 330.70$
Money Maker	$0.00^{d} \pm 0.00$	$17247.10^{c} \pm 327.38$
Heirloom Tall vine	$20158.00\ ^{b} \pm 1.97$	$20948.35^{b} \pm 305.85$
Golden Jubilee	$0.00^{d} \pm 0.00$	$22394.35^{a} \pm 608.06$
Legend	$19569.00^{c} \pm 45.57$	$22077.80^{a} \pm 558.85$
Goliath pear Hybrid	$21433.00^{a} \pm 76.49$	$21160.55^{b} \pm 1060.88$
Mean	**20386.67**	**20833.11**
$LSD_{5\%}$	**0.00**	**670.62**
CV (%)	**0.00**	**2.50**

The mean number of days to fruiting were determined at P≥0.05 significant level using DMRT. Means followed by the same letters in the same column are not significantly different (P≥0.05) according to DMRT.

Table 6 and Fig 1(e). shows in inoculation; Goliath Pear Hybrid had significantly (p< 0.05) the highest mean (21,433.00g) of weight of fruits followed by Heirloom Tall vine (20,158.00g) and Legend (19,569.00g) respectively. Cal J, Money maker and Golden Jubilee had a mean (0.00) weight of fruits because they all succumbed to the effects of bacterial wilt hence no fruits were harvested. All the varieties were significantly (p<

0.05) different in means of weight of fruits. There was no significant (p> 0.001) difference in mean of weight of fruits in experiment 1 and experiment 2.

In non-inoculation, Golden Jubilee had significantly (p< 0.05) the highest mean (21,160.55g) weight of fruits followed by Legend (22,077.80g), Cal J (21,170.63), Goliath pear Hybrid (21,160.55g) and Heirloom Tall vine (20,948.35g) respectively. Money maker had significantly (p< 0.05) the lowest mean (17,247.10g) weight of fruits. Cal J, Heirloom Tall vine and Goliath pear Hybrid were not significantly (p> 0.05) different in means of weight of fruits. Golden Jubilee was not significantly (p> 0.05) different to Legend in mean of weight of fruits. Money maker was significantly (p< 0.05) different to the other varieties in mean of weight of fruits.

There was a significant (p< 0.001) difference in mean of weight of fruits in experiment 1 and experiment 2.

3.7. Disease incidences at different stages of growth

Table 7. Means of Disease Incidence at different stages of growth.

Stage of growth	**Seedling**			**Flowering**			**Fruiting**		
VARIETY	**EXP1**	**EXP 2**	**MEAN**	**EXP1**	**EXP 2**	**MEAN**	**EXP1**	**EXP 2**	**MEAN**
CJ	2.00^{a} ±0.00	3.00^{a} ±0.00	2.50^{a} ±0.12	4.00^{a} ±0.00	4.00^{a} ±0.00	4.00^{a} ±0.00	5.00^{a} ±0.00	5.00^{a} ±0.00	5.00^{a} ±0.00
MM	2.00^{a} ±0.00	2.00^{b} ±0.00	2.00^{b} ± 0.00	4.00^{a} ±0.00	4.00^{a} ±0.00	4.00^{a} ±0.00	5.00^{a} ±0.00	5.00^{a} ±0.00	5.00^{a} ±0.00
HTV	0.00^{c} ±0.00	0.00^{d} ±0.00	0.00^{e} ±0.00	2.00^{c} ±0.00	2.00^{c} ±0.00	2.00^{d} ±0.00	2.00^{c} ±0.00	2.00^{c} ±0.00	2.00^{c} ±0.00
GJ	0.00^{c} ±0.00	1.00^{c} ±0.00	0.50^{d} ±0.12	3.00^{b} ±0.00	3.00^{b} ±0.00	3.00^{c} ± 0.00	4.00^{b} ±0.00	4.00^{b} ±0.00	4.00^{b} ±0.00
LG	1.00^{b} ±0.00	1.00^{c} ±0.00	1.00^{c} ±0.00	3.00^{a} ±0.00	4.00^{a} ±0.00	3.50^{b} ± 0.12	4.00^{b} ±0.00	4.00^{b} ±0.00	4.00^{b} ±0.00
GPH	0.00^{c} ±0.00	0.00^{d} ±0.00	0.00^{e} ±0.00	2.00^{c} ±0.00	2.00^{c} ±0.00	2.00^{d} ± 0.00	2.00^{c} ±0.00	2.00^{c} ±0.00	2.00^{c} ±0.00
Mean	**0.83**	**1.17**	**1.00**	**3.00**	**3.17**	**3.08**	**3.67**	**3.67**	**3.67**
LSD$_{5\%}$	**0.00**	**0.00**	**0.00**	**0.00**	**0.00**	**0.00**	**0.00**	**0.00**	**0.00**
CV (%)	**0.00**	**0.00**	**0.00**	**0.00**	**0.00**	**0.00**	**0.00**	**0.00**	**0.00**

The mean disease incidence were determined at P≥0.05 significant level using DMRT. Means followed by the same letters in the same column are not significantly different (P≥0.05) according to DMRT. NB: CJ – Cal J, MM – Money maker, HTV – Heirloom Tall vine, GJ – Golden Jubilee, LG – Legend, GPH – Goliath Pear Hybrid.

3.7.1. Disease Incidence at Seedling stage

In Table 7. At seedling stage, in experiment 1; Cal J and Money maker had significantly (p<0.05) the highest mean (2.00) of disease incidence followed by Legend (1.00). Heirloom Tall vine, Golden Jubilee and Goliath pear Hybrid had significantly (p<0.05) the lowest mean (0.00). Cal J and Money maker were not significantly (p>0.05) different in mean of disease incidence. Heirloom Tall vine, Golden Jubilee and Goliath pear Hybrid were not significantly (p>0.05) different in mean of disease incidence. Legend was significantly (p<0.05) different to the other varieties.

In experiment 2; Cal J, had significantly the highest mean (3.00) of disease incidence followed by Money maker (2.00), Golden Jubilee and Legend which both had a mean of 1.00 of disease incidence. Heirloom Tall vine and Goliath pear Hybrid had significantly (p<0.05) the lowest mean (0.00) of disease incidence. Heirloom Tall vine and Goliath pear Hybrid were not significantly (p>0.05) different in mean of disease incidence. Golden Jubilee was not significantly (p>0.05) different to Legend in mean of disease incidence while Cal J was significantly (p<0.05) different to Money maker in mean of disease incidence.

There was no significant (p>0.001) difference in means of disease incidence in both experiment 1 and experiment 2 except in Cal J and Golden Jubilee.

Combined experiments showed, Cal J had significantly (p<0.05) the highest mean (2.50) of disease incidence followed by Money maker (2.00), Legend (1.00) and Golden Jubilee (0.50) respectively. Heirloom Tall vine and Goliath Pear Hybrid both had significantly (p<0.05) the lowest mean (0.00) of disease incidence. Cal J, Money maker, Golden Jubilee and Legend were significantly (p<0.05) different in means of disease incidence while Heirloom Tall vine was not significantly (p>0.05) different to Goliath pear Hybrid in means of disease incidence.

Linear Correlation between Mean of Disease Incidence at Seedling stage and Mean of Disease Incidence at Flowering stage

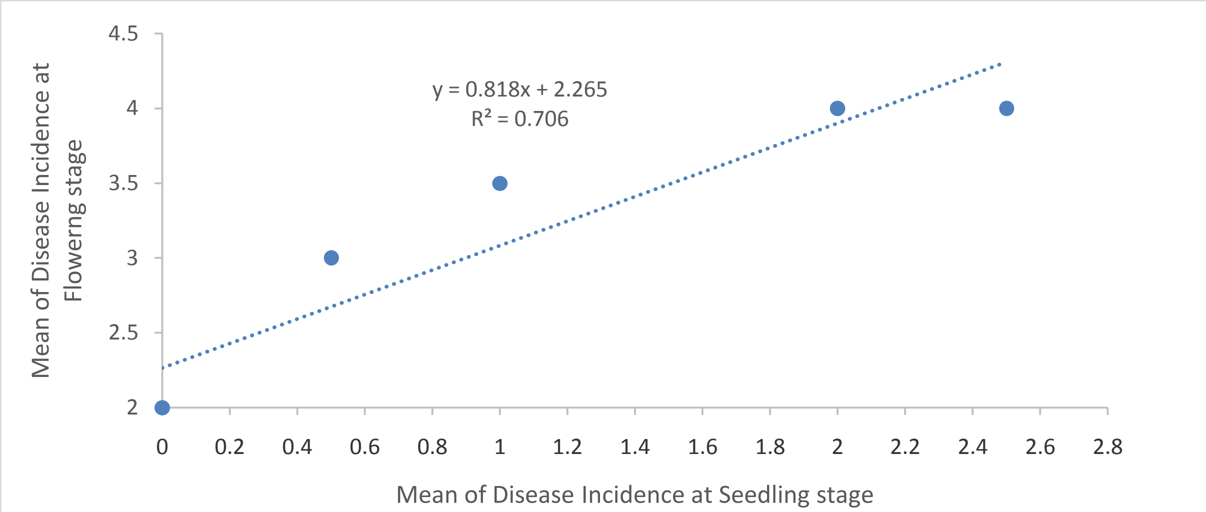


Fig 2: Linear Correlation between Mean of Disease Incidence at Seedling stage and Mean of Disease Incidence at Flowering stage.

There was a strong (R = 0.84, $R^2 = 0.7056$) and highly significant ($p< 0.001$) positive correlation between the mean of disease incidence at seedling stage and mean of disease incidence at flowering as shown in figure 2.

3.7.2. Disease Incidence at Flowering stage

In Table 7. At flowering stage, in experiment 1; Cal J and Money maker had significantly ($p<0.05$) the highest mean (4.00) of disease incidence followed by Golden Jubilee and Legend with a mean (3.00) of disease incidence. Heirloom Tall vine and Goliath pear Hybrid had significantly ($p<0.05$) the lowest mean (2.00) of disease incidence. Cal J, Money maker and Legend were not significantly ($p>0.05$) different in means of disease incidence. Heirloom Tall vine was also not significantly ($p>0.05$) different to Goliath pear Hybrid in means of disease incidence while Golden Jubilee was significantly ($p<0.05$) different to the other varieties in mean of disease incidence.

In experiment 2, Cal J, Money maker and Legend had significantly ($p<0.05$) the highest mean (4.00) of disease incidence followed by Golden Jubilee (3.00). Heirloom Tall vine and Goliath pear Hybrid had significantly ($p<0.05$) the lowest mean (2.00) of disease incidence. Cal J, Money maker and Legend were not significantly ($p>0.05$) different in means of disease incidence. Heirloom Tall vine was not significantly ($p<0.05$) different to Goliath pear Hybrid in mean of disease incidence. Golden Jubilee was significantly ($p<0.05$) different to the other varieties in mean of disease incidence.

In combined experiment, Cal J and Money maker had significantly ($p<0.05$) the highest mean (4.00) of disease incidence followed by Legend (3.50) and Golden Jubilee (3.00) respectively. Heirloom Tall vine and Goliath pear Hybrid had significantly ($p<0.05$) the lowest mean (2.00) of disease incidence. Cal J and Money maker were not significantly ($p>0.05$) different in mean of disease incidence. Heirloom Tall vine and Goliath pear Hybrid were also not significantly ($p>0.05$) different in mean of disease incidence, while Golden Jubilee was significantly ($p<0.05$) different to Legend in mean of disease incidence. There was no significant ($p>0.001$) difference in means of disease incidence in both experiments except in Legend.

Linear Correlation between Mean of Disease Incidence at Seedling stage and Mean of Disease Incidence at Fruiting stage

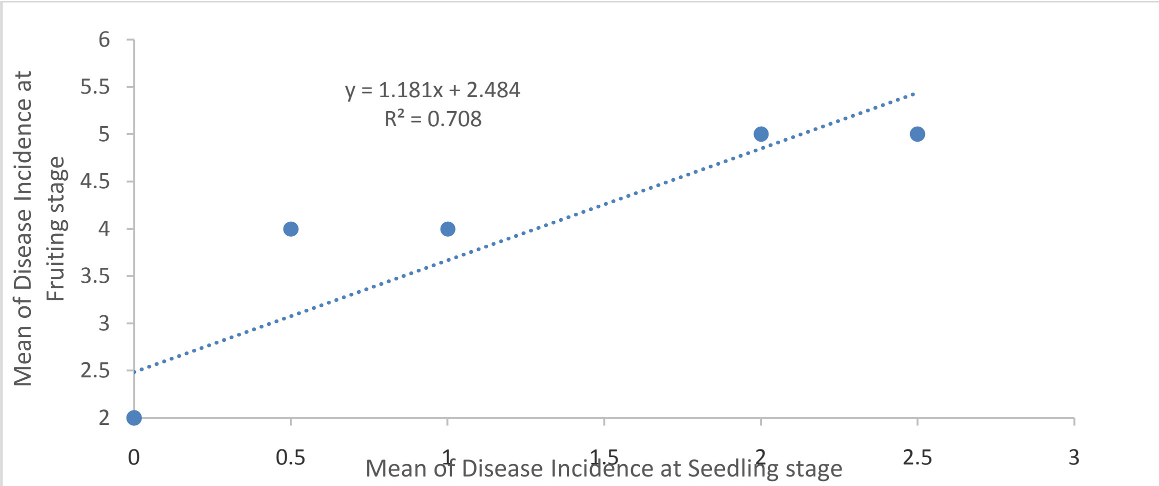


Fig 3: Linear Correlation between Mean of Disease Incidence at Seedling stage and Mean of Disease Incidence at Fruiting stage

There was a strong (R = 0.8414, $R^2 = 0.7081$) and highly significant ($p < 0.001$) positive correlation between the mean of disease incidence at seedling stage and mean of disease incidence at fruiting stage as shown in figure 3.

3.7. 3. Disease Incidence at Fruiting stage

In Table 7. At fruiting; in both experiment 1 and experiment 2, Cal J and Money maker had significantly ($p<0.05$) the highest mean (5.00) of disease incidence followed by Golden Jubilee and Legend which had a mean of 4.00 each. Heirloom Tall vine and Goliath pear Hybrid had significantly ($p<0.05$) the lowest mean (2.00) of disease incidence. In both experiments; Cal J and Money maker were not significantly ($p>0.05$) different in mean of disease incidence. Heirloom Tall vine was not significantly ($p>0.05$) different to Goliath pear Hybrid while Golden Jubilee and Legend were also not significantly ($p>0.05$) different in means of disease incidence. There was no significant ($p>0.001$) difference in experiment 1 and experiment 2 in means of disease incidence.

In combined experiment; Cal J and Money maker had significantly the highest mean (5.00) of disease incidence followed by Golden Jubilee and Legend which had a mean of 4.00 each. Heirloom Tall vine and Goliath pear Hybrid had significantly ($p<0.05$) the lowest mean (2.00) of disease incidence. In both experiments; Cal J and Money maker were not significantly ($p>0.05$) different mean of disease incidence. Heirloom Tall vine was not significantly ($p>0.05$) different to Goliath pear Hybrid while Golden Jubilee and Legend were also not significantly ($p>0.05$) different in means of disease incidence.

Linear Correlation between Mean of Disease Incidence at Flowering stage and Mean of Disease Incidence at Fruiting stage

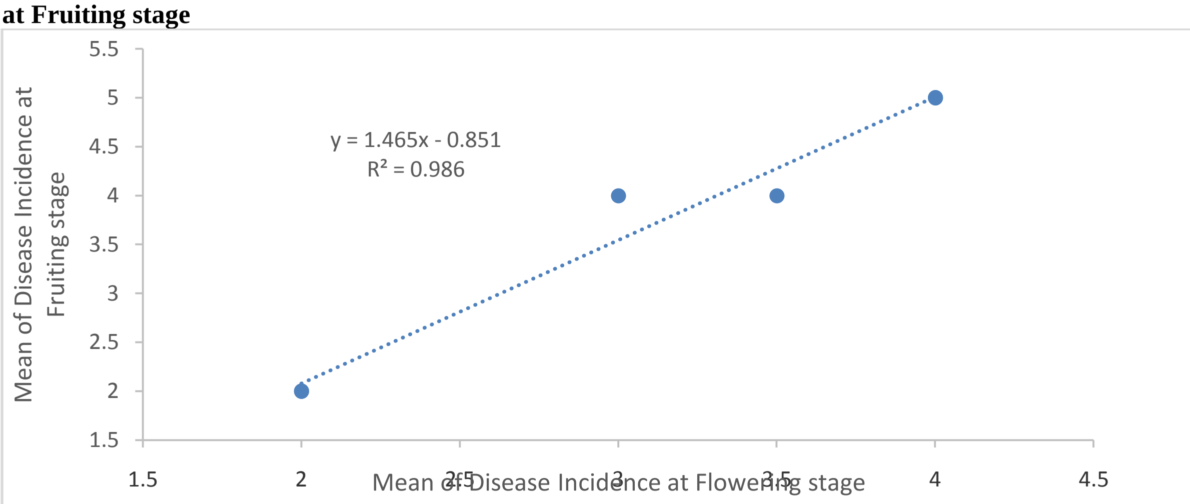


Fig 4: Linear Correlation between Mean of Disease Incidence at Flowering stage and Mean of Disease Incidence at Fruiting stage

Figure 4. Showed that there was a strong (R = 0.9929, R^2 = 0.9858) and highly significant (p< 0.001) positive correlation between the mean of disease incidence at flowering stage and mean of disease incidence at fruiting stage.

Inoculated Tomato varieties at fruiting.

Plate 2: Cal J

Plate 3: Money maker

Plate 4: Heirloom Tall vine

Plate 5: Golden Jubilee

Plate 6: Legend

Plate7: Goliath pear hybrid

IV. Discussion

4.1. Days to flowering

Highly significant variation in number of days to flowering in varieties were observed in inoculated and non-inoculated varieties, inoculated varieties took more number of days to flower than non-inoculated varieties, this was attributed to the effects of bacterial wilt. Bacterial wilt caused stunted growth in inoculated tomato varieties hence prolonged number of days to fruiting. Similar results were reported by [20].

4.2. Days to fruiting

Days to fruiting differed among the varieties in both inoculation and non-inoculation with inoculated varieties taking slightly longer time to fruit due to the effects of Bacterial wilt. The bacteria produce polysaccharides which clog the xylem preventing water flow throughout the plant [19] which caused water stress. This confirms why water is an essential compound in fruit formation; water stress caused delayed vegetative growth which in turn delayed fruiting.

4.3. Days to Maturity

The fruits were considered mature when the colour turned from green to red for Cal J, Money maker, Heirloom Tall vine, Legend and Goliath pear Hybrid; and yellow for Golden Jubilee. Maturity period differed among the varieties in both treatments. Inoculated varieties took more number of days to mature than non-inoculated varieties. This was a result of Bacterial wilt disease. This was caused by water stress in the inoculated tomato plants which led to delayed maturity period

4.4. Plant Height (cm)

Cumulative height of both the inoculated and non-inoculated plants was determined in centimeters.Highly significant variation in height of the varieties was observed among the varieties in both inoculated and non-inoculated. The study showed a significant reduction in plant height in the inoculated tomato varieties due to the effects of Bacterial wilt which caused stunted growth in the tomato plants. Similar results were reported by[20].

4.5. Fruit Number

The fruits were harvested twice per week and the number of fruits per plant recorded. There was a highly significant difference between the number of fruits produced in inoculated varieties and non-inoculated varieties. The study showed production of less number of fruits in the inoculated tomato varieties than in non-inoculated tomato varieties. Similar results were reported by [20], confirming that *Ralstoniasolanacearum* has great influence on fruit production in tomato varieties. Once inside the plant, *R. solanacearum* replicates and colonizes vascular tissue. Inoculated tomatoes; Cal J, Money maker and Golden Jubilee varieties succumbed to the effects of bacterial wilt before harvesting.

4.6. Fruit weight

The fruits were harvested twice a week for five harvestings and the yield recorded in grams for each variety. Fruit weight differed a mong varieties in both inoculation and non-inoculation. The study indicated that inoculated varieties produced fruits with low weight compared to non-inoculated varieties, this was as a result of the effects bacterial wilt disease. Bacterial wilt produced polysaccharides which clogged the xylem preventing water flow throughout the plant [19]. This caused water stress in the plants which led to production of fruits with low weight. This confirms why water is an essential compound in fruit formation; water stress causes formation of fruits with low weight [6].

4.7. Disease Incidence at seedling stage

Seedling stage had the lowest mean of disease incidences among the varieties. [17]The younger the seedling, the less was the susceptibility to the pathogen. The plants exhibited minimal disease incidence confirming that they were tolerant to the pathogen. In the early stages of infection, the wilting was only present on one side of the plant and occurred only in the afternoon when the temperatures were high. The young plants recovered overnight and appeared to be fine the next day. This was also reported by [2]. Younger plants exhibit less percent colonization and lower bacteria colony density [6]. This explains why all the tomato varieties evaluated at seedling stage (4 leaves) were tolerant to the pathogen. Physiology of resistant plants is also different from that of susceptible plants. According to research completed by [6] resistant plants still get infected by *R. solanacearum* however, they are capable of eliciting a defense response to contain the bacteria. This plant defense response contained the bacteria, which prevented higher colonization levels in the young plants.

4.8. Disease Incidence at flowering stage

With the advancement in age of the plants, there was significant increase in the disease incidence, the overall disease incidence at this stage was high compared to seedling stage. The results indicated that flowering stage was a crucial factor governing the susceptibility of tomato to the pathogen. As bacteria began to colonize more of the xylem at this stage, the bacteria synthesized proteins that inhibited the plant defense response. The bacteria then produced polysaccharides which clogged the xylem preventing water flow throughout the plant [19], resulting in death of the susceptible plants.

4.9. Disease Incidence at fruiting stage

The results indicated that the disease incidence at fruiting stage was relatively high in the susceptible varieties. This confirmed that the growth stage played a key role in determining the vulnerability of tomato to the pathogen. Similar results were reported by [17]. Cal J and Money maker exhibited the highest disease incidence causing high mortality rate thus the two varieties were highly susceptible to the pathogen. This was as a result of the degradation of xylem vessels and the destruction of surrounding tissues [20] preventing water up take. Heirloom Tall vine and Goliath pear Hybrid were tolerant to the pathogen. The two were tolerant to the pathogen at fruiting stage and this might be due to the activity of phenylalanine ammonia lyase (PAL) and polyphenol oxidase (PPO). Increased PAL is linked to the pathway that produces compounds such as phenols which are related to the plant defense response. PPO is related to the oxidation of phenols to make quinines which are also related to the plant defense response to pathogens [18].

V. Conclusion

Heirloom Tall vine and Goliath pear Hybrid showed tolerance to *R. Solanacearum* suggesting that the two tomato varieties are good candidates for Kenyan farmers. However there is need for further research to evaluate their performance in different ecological regions.

From the study, the results indicated that with advancement in age of the tomato plant, there was an increase in vulnerability to the *R. Solanacearum*. This proves that stage of growth is a determinant of the vulnarability to the pathogen. The three stages of growth evaluated showed different levels of tolerance to the pathogen. Seedling stage was tolerant to the pathogen while fruiting stage was the most susceptible stage to the pathogen.

Inoculated varieties took more days to flower, more days to fruit, more days to mature, low plant height (cm), less number of fruits and low weight of fruits (g) as compared to non-inoculated varieties, this was attributed to the effects of bacterial wilt. Bacterial wilt caused stunted growth and reduced vegetative growth in tomato plants that eventually led to reduced yields

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