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(RESEARCH ARTICLE)

Comparison of severity of *Phytophthora colocasiae* (taro leaf blight) disease on *in-vivo* and *in-vitro* Pacific-Caribbean and Kenyan taro (*Colocasiae esculenta*) grown in Kakamega county (Kenya)

Otieno Carren Adhiambo [1,*], Adema Valerie Palapala [2] and Opande George Timothy [3]

[1] *School of Science, department of biological sciences Rongo University , Rongo, Kenya.*
[2] *School of Science, USIU University, Nairobi, Kenya.*
[3] *School of Biological and physical sciences. Maseno University, Maseno, Kenya.*

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Abstract

Taro (*Colocasiae esculenta)* is an important tuber crop of Kakamega - Kenya. It is nutritionally very rich. Taro leaf blight (TLB) is the most devastating pathogen for its production worldwide and current management strategies are not effective in its control. Studies on TLB disease severity has been done worldwide. However, determining and comparing the disease severity between regions of the world has not been adequately done. Before this study, TLB disease severity on Pacific and Kenyan taro grown *in-vivo* in Kakamega - Kenya was unknown. Furthermore, knowledge on the differential effect of weather on taro accessions obtained from different regions of the world was scanty. TLB disease severity was assessed on Pacific-Caribbean taro accessions obtained through tissue culture from Pacific- Caribbean. Kenyan taro was obtained from various counties in Kenya. The research aimed at examining and comparing TLB disease severity on Pacific and Kenyan taro. The study was conducted at MMUST University of Science and Technology farm and Maseno university laboratory and greenhouse. All experiments were arranged in a CRD and replicated five times. Data obtained were subjected to ANOVA and the means separated with Least Significant Difference. The results showed that mean disease severity among the Pacific and Kenyan taro varied from 33.2% - 53.5% respectively. TLB disease severity in both *in-vivo* and *in-vitro* studies presented higher percentage on Kenyan than Pacific- Caribbean taro. The *in-vivo* severity range for Kenyan and Pacific taro were; 9.8-28.5% and 4.9-14.8% respectively. Likewise, Kenyan taro maintained higher percentage severity *in-vitro*, ranging from 20-44% whereas Pacific taro ranged from 9.4-30%. Kenyan Siaya accession KNY/SYA/51 recorded the highest disease severity *in-vivo*. The lowest disease severity *in-vivo* was obtained from the Pacific taro CE/IND/06. The study suggested that region of origin of taro, varietal difference and weather would influence TLB severity. This study indicated the need for breeding for resistance to taro leaf blight.

Keywords: *Phytophthora colocasiae*; *Colocasiae esculenta*; Severity; Accession; Region

1. Introduction

Taro is an increasingly important crop used as a vegetable, staple food and medicinal applications. It is a high-value crop and has immediate economic benefits for producers [1]. Additionally, it is an important source of essential nutrients providing long term nutritional benefits for consumers. The attack of *P. colocasiae* on the leaves reduces significantly the number of functional leaves which leads to yield reduction [2]. Globally, *P. colocasiae* causes corms to rot both in the field and in the storage and this has led to heavy storage losses [3]. The phenomenon of disease severity, in regard to taro leaf blight disease of taro have not been completely understood in Kenya and the comparision with the Pacific-Caribbean taro was unknown.

* Corresponding author
E-mail address: joycarotieno@gmail.com

According to Miyasaka et al., [4], taro cultivars differed significantly in severity of TLB and in severity of corm rots. It has been revealed that while some plants become severely diseased with continuous night time sporulation, others immediately adjacent may have little or no disease [5]. Chiejina and Ugwuja [6] reported that disease severity appeared to increase with increase in disease incidence across different locations. Environment has been recognized as one of the major factors that influence the process of an epidemic, having the capacity to induce or retard it [7]. The epidemiological parameters such as rainfall, temperature and relative humidity and their contribution to taro leaf blight disease severity has scantily been researched. Rainfall, humidity and temperature are the key factors controlling the taro leaf blight disease cycle and epidemiology. Favorable temperatures and regular periods of leaf wetness, particularly in the humid tropics promote TLB epidemics by favouring pathogen dispersal, infection, and disease development [8]. Outbreaks of the disease in new areas distant from known centers of infection probably result from the introduction of infected planting material.

The study of relationship between disease progressions with weather parameters is of paramount importance for effective disease management [9]. Fullerton and Tyson [10] revealed that apart from other environmental factors, moisture, sunshine and wind also influence fungal disease severity and epidemics generally flourish when night temperatures are in the range of 17–20 °C. The cool temperatures stimulate the release of infective zoospores, promoting multiple infections [9].

Taro leaves have waxy hydrophobic leaf cuticles, which assist the wash-off of sporangia and zoospores from the leaves into the soil, or their splash onto other leaves and petioles, particularly the lower older ones. However, in the absence of regular rainfall, conditions favourable to re-infection occur on most nights ensuring regular cycling and survival on infected plants thus making it endemic [8].

According to [9], TLB pathogen grows rapidly in areas with high humidity and heavy rainfall. Fullerton and Tyson [10] reported that moisture levels were positively correlated with the development of number and size of leaf blight of maize lesions in both susceptible and resistant varieties of maize. Similarly, Ayogu et al., [11] and Cabi [12] noted a prevalence of Northern leaf blight in highlands and wetter areas of the Kenya and Uganda. Temperature has been reported to govern the rate of reproduction of fungi and the physiological conditions of the host. Temperature highly affects the growth and aggressiveness of pathogens and expression of disease symptoms in the plants [13].

It is well known that temperature influences pathogen development as well as expression of host resistance. The effect of temperature on aggressiveness component had been established for many pathogen species and presents an optimum for spore germination, lesion development and sporulation. However, the response to temperature differed among individual plants. Spore production rate of two leaf rust isolates (*P. triticina*) were found to be identical at 2-18 °C but different at 10- 30 °C [14]. Growth was found to be faster between 27-30 °C [15].

Other factors involved in plant disease spread include; susceptible host, virulent pathogen, frequency of each element over time and duration and frequency of favourable environment. The host factors that affect epidemic development include; levels of genetic resistance or susceptibility of the host. The pathogen factor that affect epidemic include; levels of virulence, quantity of inoculum near host, type of reproduction of the pathogen, ecology of the pathogen and mode of spread of the pathogen. According to Benzohra et al., [16] the process of epidemic is influenced by environment [17]. Research has shown that absence of certain important nutrients such as calcium and phosphorus increase taro leaf blight disease infection. This needs to be investigated further [18]. Human activity also plays a role in TLB disease epidemics and they include; site selection and preparation, selection of propagative materials, cultural practices, disease control measures and introduction of new pathogens.

Before this study, severity of *taro leaf blight (Phytophthora colocasiae) disease on in-vivo and in-vitro Pacific-Caribbean and Kenyan taro (Colocasiae esculenta) grown* in Kakamega county of Kenya was unknown**,** therefore experiments were conducted to determine this unknown. Two field studies mainly; (1) to determine Taro leaf blight disease severity on Pacific-Caribbean, (2) to determine disease severity for the Kenyan and sampled Pacific accessions and (3) a greenhouse study was initiated.

2. Material and methods

2.1. Study area

The studies were established at two locations: Masinde Muliro University of Science and Technology in Kakamega county and Maseno University in Kisumu county. Kakamega town is located within the upper highland agro-ecological zone. Its climate is classified as tropical with a great deal of rainfall even in the driest month. It belongs to group Af

(Tropical rainforest) by Koppen- Geiger system of climatic classification [19]. The average temperature is 20.4 °C. The variation in temperature throughout the year is 2.0 °C. The lowest average temperature usually occurs in July when it is approximately 19.3 °C. The annual rainfall is approximately 1971 mm. The difference in precipitation between the driest and the wettest months is 212 mm (Kakamega –data.org).

MMUST University lies between longitudes of $34^0 32' 0''$E - $34^0 57' 0''$W and latitudes of $0^0 07' 30''$N - $0^0 10' 15''$S of the equator at an altitude of about 2000 m above sea level [19]. The trials were conducted from January 2013 to November 2013, and September 2013 to April 2014 at MMUST university garden and Kakamega Mlimani estate garden, respectively in the two respective cropping seasons. Maseno University lies within Latitude: 0° 00' 60.00" N and Longitude: 34° 35' 59.99" E and 1503 metres above sea level. The fields used for taro cultivation were non-flooded and rainfall provided all the water for plant growth. Harvesting occurred ten months after planting for the first experiment, and for the second and third it was after seven months. A completely randomized design was used in the two fields.

For the laboratory and greenhouse experiments, a total of four *Phytophthora colocasiae* isolates were obtained from University of Eldoret laboratory as earlier prepared through the process described by Whehan [20]. The isolates had earlier been collected from blight infected taro primarily from Kitale taro growing regions; Mois bridge, Maili saba, Bikeke and Kiminini in the year 2014.

2.2. Determination of taro leaf blight disease severity on Pacific-Caribbean in study-1

The field study – 1 was conducted on Pacific-Caribbean taro accessions. Three hundred Pacific-Caribbean taro plant tissue culture materials earlier imported from the Pacific community (Hawaii, Papua New Guinea, Samoa, Japan, Indonesia, Malaysia and Thailand) through the Secretariat of the Pacific Community (SPC) based in Suva, Fiji Islands in conformity to KEPHIS (Kenya Plant Health Inspectotate Service) requirements were used since they already had improved TLB resistance.

2.3. Culture multiplication of imported taro accessions

Three hundred imported taro plants obtained from 25 different accessions were used in field study-1. They included the following; (BL/HW/08, BL/HW/26, BL/HW/37, BL/PNG/10, BL/SM/111, BL/SM/116, BL/SM/120, BL/SM/128, BL/SM/132, BL/SM/143, BL/SM/149, BL/SM/151, BL/SM/152, BL/SM/158, BL/SM/43, BL/SM/80, BL/SM/92, CA/JP/03, CE/IND/01, CE/IND/06, CE/MAL/12, CE/MAL/14, CE/THA/07, CE/THA/09, CE/THA/24) replicated 12 times each. This experiment was established within Masinde Muliro University garden. Land not previously cultivated was used. The experimental area measuring 3500 m^2 (70m by 50m) was cleared using a machete, hand ploughed and harrowed twice using jembes and hoes before planting. Soil was made into raised beds in preparation for planting. Three hundred taro suckers were planted in 60 cm deep holes and each sucker firmly placed using hands according to the methods of [20]. The spacing was 0.5 m between plants and 1.0 m between rows. Watering was done in the morning and evening for one month approximately one liter per plant using a sprinkler. The plants were arranged in a completely randomized design (CRD). The 25 accessions with 12 plants per accession were replicated for 8 months.

2.4. Determination of disease severity for the Kenyan and sampled Pacific accessions in study-2

The same procedure as in field study- 1 above was repeated for field study-2 where both the Pacific -Caribbean and Kenyan taro accessions were used. Local taro accessions were collected from farmer's plots in seven regions in Kenya**;** Central Kenya in Kayole, Kisumu Dunga beach along Lake Victoria, Siaya along Dominion farm, Kakamega-Milimani, Mumias near sugar company, Kitale- Mailsaba, Busia, Bumala area, Eldoret, Langas area. Some Pacific-Caribbean taro accessions from the first experiment were also sampled, considering the least and the most susceptible accessions. A total of twenty-six taro plant materials were obtained. They included; KNY/KIS/81, KNY/BSA/41, KNY/ELD/75, BL/HW/8 CE/JP/3, BL/SM/120, KMM/MM1/75, KNY/KIS/20, KNY/CTR/33, BL/HW/26, BL/HW/80, KMM/MM2/76, KNY/KIS/21, KNY/KTL/61, CE/IND/1, BL/SM/28, KNY/SYA/50, KNY/KIS/22, KNY/SYA/51, CE/THA/7, CE/IND/6, BL/SM/48, KNY/KAK/16, CE/THA/24, CE/MAL/14, BL/SM/111. This experiment was established within Milimani estate garden. An experimental area measuring 2,240 m^2 (70 m by 50 m) not previously cultivated with taro [1].

2.5. Determination of disease severity for the Kenyan and sampled Pacific-Caribbean accessions under greenhouse conditions in study -3

The greenhouse experiment was performed in the laboratory and greenhouse respectively. General laboratory techniques were followed for preparation of media, sterilization, isolation and maintenance of fungal cultures with slight modifications where it seemed necessary. Petri dishes were placed in sterilization tins and sterilized in hot air oven at 160 °C for 90 minutes. Potato Dextrose Agar (PDA) media and water used in the study were sterilized at a temperature of 121.6 °C for 20 minutes in an autoclave as described by Asraku [21].

2.5.1. Sterilization and plating of medium

Work surfaces were sterilized by ethyl alcohol and sodium hypochlorite. Scalpel blades and inoculation loops were sterilized over flame. Plating of medium was done by melting the sterilized medium and distributing in 9 cm diameter petri plates. This was done aseptically at the rate of 20 ml per plate in the laminar flow-hood chamber and allowed to solidify. The cultures were incubated for 4 days maintaining them at room temperature in dark place in the laboratory according to the methods of Wambua [22]. They were then sub-cultured onto water agar till pure cultures were obtained. The remaining isolates were then stored at room temperature in 2 ml tubes containing 3-4 plugs of mycelium, 3- and 1-ml water. The isolates were left for 14 days.

2.5.2. Pathogenic nature of isolates

The pathogenic nature of the isolates was determined by proving Koch's postulates through pathogenicity test according to the methods of Singh et al., [23] where disease free taro leaves were placed on sterilized filter paper soaked with distilled water and placed in petri dishes. The plates were inoculated with 2 ml of sporangial suspension containing *Phytophthora colocasiae* which had earlier been sub-cultured in Maseno laboratory. The leaves were then covered with plastic bags and left for two days at room temperature. After two days, the inoculated sites showed water soaking lesions at the beginning but later turned brown according to the observations of Brooks [24].

2.5.3. Soil sterilization

Black soil from Maseno Botanical garden was sifted to remove stones, plastic materials and plant debris. The soil was steam sterilized in a barrel at 100 °C for two hours. The sterilized soil was left in the barrel overnight to cool before use according to the methods of Whehan [20]. The taro plants from the previous experiment two of Pacific-Caribbean and Kenyan taro were sampled considering the least and the most susceptible accessions as obtained from the previous result. They included; KNY/SYA/51, KNY/KAK/16, CA/JP/O3, CE/THA/07, KNY/BSA/41, BL/HW/26, BL/SM/80, KNY/SYA/50, KNY/KTL/61, BL/SM/92. KNY/MU/75, KNY/CNT/33, BL/HW/08, CE/THA/24, KNY/KSM/81. KNY/SYA/50.

2.5.4. Plucking and packaging of corm and cormels

The corm and cormels were carefully plucked from soil and packed in polythene bags then labeled according to place of origin. They were placed in a cool dry place for greenhouse evaluation. Plastic buckets were filled with the sterilized top soil and the samples placed at 1 m x 1 m using a complete randomized design with three replications. The crops were watered in the morning every two days using clean water and administered at the base of the crop. The tubers were covered with the soil and firmed down according to the methods of Shreshta [26].

2.5.5. Inoculation of pathogen

Two *Phytophthora colocasiae* pathogen treatments namely 21R1 and 3R1 isolates previously obtained from University of Eldoret laboratory were used for greenhouse inoculation. Distilled water was used on the leaves as control. The inoculation was done by using two most virulent isolates of *Phytophthora colocasiae* (showing very fast growth) in the culture medium. Mycelia mat from the culture were harvested using sterile scalpel into an electric blender. After blending for five minutes, 200 ml of sterile distilled water was added into 500 ml conical flask and filtered using double layer muslin cloth.

2.5.6. Soil inoculation

Soil inoculation was done by pouring 20ml of inoculums suspension at the base of the stem of each plant according to the methods of Shreshta [26]. This was done three months after planting. Control seedlings were treated with the same quantity of sterile distilled water. Both the inoculated and the control seedlings were covered with polythene bags to increase humidity around the plants according to the methods of Shreshta [26]. After 24 hours, polythene bags were removed for 20 minutes and the plants watered. Four days after inoculation, the polythene bags were finally removed. The plants were arranged in a completely randomized design. There were 16 accession with 3 plants per accession per treatment. There were two pathogen inoculation treatments and one control. The greenhouse experiment result was collected for 5 months. Similar procedure as described in the previous experiment for obtaining disease incidence was used.

2.5.7. Scoring for TLB disease severity

Collection of meteorological data from Kakamega weather station was performed on a monthly basis. Relative humidity recorded in the morning and afternoon, minimum, average and maximum monthly temperature, and average rainfall

prevailing at the observation sites were collected from Kakamega meteorological station for the interpretation of results. The weather changes were scored against the different accessions of taro used. Taro leaf blight disease symptoms were carefully observed to confirm the disease. Total area of leaves, total area of leaves infected and the disease severity were recorded at monthly intervals from the appearance of the first symptom (at 3 months) till the crop was harvested.

Areas of leaves were measured by using non-destructive methods [27-28] using the formula W_P x L_{PA} where; W_P=Leaf width passing the petiole attaching point, L_{PA}=Length of the petiole attaching point to the apex of leaf

Areas of leaves infected by the disease were assessed using the maximum length and breadth of the affected leaf area. Disease severity ratings per accession per experiment were undertaken using a subjective score scale of 1-9 adopted from Manza et al., [29]. However, records in this study were made as the percentage leaf area infected. The score was repeated monthly for eight months in the first experiment, five months for the second experiment and five months for the greenhouse experiment. The start of scoring took consideration the beginning of disease development i.e. first appearance of TLB symptoms on taro leaves.

2.6. Analysis

The results obtained on the different taro accessions were analyzed using analysis of variance (ANOVA) and the least significant difference (LSD) used to separate the means at 5%. Correlation analysis was used to compare mean differences as described by Chan et al., [30]

3. Results

3.1. Taro leaf blight disease severity of Pacific- Caribbean taro for study-1

Table 1 Percentage of TLB disease severity on Pacific- Caribbean taro

Month	Apr	May	Jun	July	Aug	Sept	Oct	Nov	Pooled
Age	3 mths	4 mths	5 mth	6 mths	7 mths	8 mths	9 mths	10 mths	mean
Pacific taro	Mean TLB disease severity								
BL/HW/08	6.4	5.2	3.7	5.5	6.8	17.8	12.0	14.9	9.0
BL/HW/26	5.8	3.4	7.3	6.8	13.7	14.6	12.5	10.0	9.3
BL/HW/37	6.6	3.2	11.7	8.2	34.2	30.3	26.7	12.4	16.6
BL/PNG/10	7.0	14.1	9.8	6.8	14.9	14.9	17.6	15.5	12.6
BL/SM/111	3.8	9.0	10.4	8.9	16.1	13.4	13.1	6.7	10.2
BL/SM/116	20.9	5.8	7.5	5.8	22.4	26.9	17.0	15.8	15.3
BL/SM/120	8.8	6.2	13.3	11.8	17.5	10.7	14.6	11.3	11.8
BL/SM/128	15.3	8.5	7.3	10.7	14.6	24.2	20.0	13.7	14.3
BL/SM/132	9.2	3.2	16.0	14.6	29.0	29.0	27.2	10.8	17.4
BL/SM/143	4.1	4.7	6.4	4.5	14.5	14.7	15.7	12.0	9.6
BL/SM/149	11.8	7.9	5.4	5.8	19.1	17.4	16.8	14.5	12.4
BL/SM/151	1.8	1.8	3.8	3.8	27.8	23.9	19.5	15.8	12.3
BL/SM/152	6.8	3.3	12.9	14.7	26.9	32.3	30.3	10.2	17.2
BL/SM/158	3.1	4.0	8.9	8.5	20.2	25.1	25.7	18.4	14.2
BL/SM/80	7.3	9.1	23.3	20.1	40.9	36.2	40.3	28.3	25.7
BL/SM/92	10.3	4.1	10.4	3.5	13.8	15.9	15.9	5.0	9.9
CA/JP/03	3.1d	7.3	8.8	9.6	26.0	32.3	25.6	14.5	15.9
CE/IND/01	22.3	30.3	31.1	12.1	22.8	16.3	21.7	15.8	21.5
CE/IND/06	35.8	42.5	48.2	34.0	45.8	50.0	56.3	39.2	44.0
CE/MAL/12	8.8	11.3	23.7	19.5	33.8	35.9	33.8	23.7	23.8
CE/MAL/14	5.0	3.8	6.0	5.2	26.5	35.3	38.6	24.7	18.1
CE/THA/07	26.1	22.2	21.9	22.8	29.5	29.5	29.5	29.5	26.4
CE/THA/09	11.4	11.4	26.3	24.5	26.0	29.9	27.8	22.4	22.5
CE/THA/24	8.8	14.8	27.3	27.8	33.4	35.5	33.7	18.3	25.0
Mean	10.2	9.7	14.3	12.0	23.6	25.1	24.3	16.6	17.0
CV	89.7	87.4	74.4	94.9	68.2	66	69.3	66.9	78.6
LSD	2.539	2.696	3.40	2.951	4.270	4.269	4.186	3.283	

The result of disease severity of Pacific-Caribbean taro accessions was as shown on table 1. A non-uniform trend of percentage disease severity was experienced in this study. The significantly ($p<0.05$) highest disease severity of 28.9% was recorded with Thailand taro accession CE/THA/07 and significantly the least ($p< 0.05$) disease severity of 10.7% from Hawaiian taro accession BL/HW/26. The highest month in disease severity was September with a severity of 25.1% and significantly ($p<0.05$) lowest month in disease severity was May with 9.7% severity as shown on table 1 Accessions BL/HW/08, BL/HW/26 and BL/HW/37, had similar reaction to taro leaf blight in that their percentage severity was higher at the beginning of the study, decreased and finally increased as shown on table 1.1 Similar finding was obtained with accession CE/THA/24 (25% severity) and CE/THA/O7 (26.4% severity) which were also statistically the same ($p>0.05$) in terms of TLB disease severity. This similarity in behavior could be due to their genetic relatedness.

The low severity could be due to accession's low susceptibility to TLB disease. Simongo et al., [31] in his study, linked low disease severity of a cultivar to the cultivar being less prone to diseases. The low TLB disease severity was linked to clean planting materials and good agricultural practices [20]. It could also be as a result of external factors like temperature, rainfall and relative humidity being unfavorable to the pathogen. This fact was supported by the report of Lin et al., Adomako [28] that weather conditions such as high rainfall, temperature and humidity influence infection rates in all plants, including those with a degree of genetic resistance.

3.2. Taro leaf blight disease severity of Pacific- Caribbean taro for study-2

The results on TLB disease severity on Pacific- Caribbean taro under field experiment two were as shown on table 2. The highest disease severity of 13.5% was observed on accession CE/IND/01 whereas the lowest percentage severity of 4.9% was realized on accession CE/IND/06. All the Pacific- Caribbean taro accessions in the study two showed a steady increase in taro leaf blight severity from February to April except for Samoa accession BL/SM/111 which decreased between March (31.7%) and April (17.7) (Table 2). Disease severity for December and January were statistically insignificant and therefore were not recorded on the table below.

Table 2 Percentage of TLB disease severity on Pacific -Caribbean taro under study-2

	Feb (5mths)	Mar (6mths)	April (7mths)	Pooled mean
Pacific taro	**Mean percentage disease severity**			
BL/HW/26	1.4±3.8	13.6±18.4	44.3±20.9	11.9±20.9
BL/HW/8	0±0	2.5±5	31.3±31.5	6.8±17.9
BL/HW/80	0±0	23.3±23.1	41.7±38.2	13±24.3
BL/SM/111	10±0	31.7±37.5	17.7±28	11.9±21.6
BL/SM/120	18.8±23.9	21.3±36.1	33.8±19.7	14.8±23.2
BL/SM/28	1±1.7	6.7±5.8	25±43.3	6.5±19.3
BL/SM/48	3.3±5.8	23.3±23.1	17.7±28	8.9±17.2
CE/IND/01	2.5±5	21.3±21.7	43.8±23.9	13.5±21.8
CE/IND/06	0±0	2.5±5	21.3±21.7	4.9±12.3
CE/JP/03	12.5±25	8.8±11.8	26.3±32.5	9.8±19.5
CE/MAL/14	2.5±5	15±12.2	30±30.8	9.5±17.9
CE/THA/24	5±5.8	3.8±4.3	30±23.1	7.8±15.1
CE/THA/07	0±0	2.5±5	68.8±12.5	14.3±28.5
Mean	4.4±5.85	13.56±16.1	33.2±27.2	10.36±20.1
LSD $p<0.05$	3.68	6.53	9.42	
C.V	245.4	143.7	78.01	193.63

BL/SM/111 showed progressive increase in disease severity then reduced finally in April. April recorded the highest disease severity of 33.2%. This could be due to high rainfall amount which was more favorable for the fungus in April

just as reported by [27] that rainfall increased taro leaf blight severity. The increase in disease severity could be due to increase in inoculums and increasing age of plant. This finding corroborates that of Simongo et al., [31] that severity of *Colocasiae* blight depended on the availability of inoculum, time of disease appearance and stage of growth of plant. Furthermore, the severity of blight disease increased with age of plant. However, a contrary finding reported a decrease in TLB lesion diameter with increasing plant age. This could be due to high resistance to taro leaf blight disease exhibited by some taro accessions [29].

3.3. Taro leaf blight disease severity of Kenyan taro for study-2

The result of this study was as shown on table 3. Taro leaf blight disease severity started right from the third month although it was statistically insignificant. April recorded significantly ($p<0.05$) highest severity of 53.5% while significantly ($p<0.05$) lowest disease severity of 1.1% was recorded in January as shown on table 1.3. All the accessions increased in disease severity from January to April except Busia accession KNY/BSA/41 and Central Kenya accession KNY/CTR/33 that reduced between March and April as 58.3-46.7% and 49-47% respectively (Table 3).

Table 3 Percentage of TLB disease severity on Kenyan taro

Month and age	Jan (4 mths)	Feb (5 mths)	Mar (6 mths)	April (7 mths)	
Kenyan taro		**Mean TLB disease severity**			**Pooled mean**
KMM/MM1/75	1.2±1.6	8.8±9.5	24±24.1	52±27	17.3±24.9
KMM/MM2/76	1.2±1.6	1.8±1.6	16±19.5	66±28	17.1±29.2
KNY/BSA/41	2±1.7	33.3±14	58.3±14.4	46.7±38	28.3±29
KNY/CTR/33	0±0	10.6±13	49±35.6	47±23	21.3±29
KNY/ELD/75	0±0	1.5±1.7	10±0	37.5±14	9.8±15.8
KNY/KAK/16	0±0	0.6±1.3	20.6±26.9	35±22	11.2±20.4
KNY/KIS/20	1.5±1.7	5±5.8	46.3±26.9	50±20	20.6±26.9
KNY/KIS/21	0.8±1.5	13.3±25	31.3±23.9	56.3±13	20.3±26.2
KNY/KIS/22	3±0	17.5±8.7	30±23.1	56.3±13	21.4±23.7
KNY/KIS/81	1.5±1.7	1.5±1.7	7.5±5	75±0	17.1±29.9
KNY/KTL/61	0±0	2.6±4.3	8±4.5	55±21	13.1±23.3
KNY/SYA/50	1.5±1.7	3.3±4.7	27.5±26.3	56.3±24	17.7±26.5
KNY/SYA/51	1.5±1.7	25±20	52.5±30.7	62.5±14	28.5±30.5
MEAN	1.1±1	9.6±8.6	29.3±	53.5±20	
L.S.D $p<0.05$	0.5	4.61	9.09	7.57	
CV	139	150	91.6	40.4	141

This could imply that there were inherent properties to reduce disease severity in these accessions. The highest disease severity of 30.5% was recorded in Siaya Accession KNY/SYA/51 and the lowest severity of 15.85% in Eldoret accession KNY/ELD/75. The study indicated that disease severity levels for the accessions evaluated differed ($p<0.05$) significantly. Different taro accessions exhibit different levels of disease resistance. Location from which the accessions were obtained could also play a role in determining the level of disease susceptibility.

3.4. Comparison of taro leaf blight disease severity between Kenyan and Pacific -Caribbean taro (Dec 2013-April 2014).

The result comparing Kenyan and Pacific-Caribbean taro accessions in terms of disease severity under field conditions were as shown on figure 1 below. Percentage taro leaf blight disease severity on Kenyan and Pacific - Caribbean taro were similar during the first month of the experiment after which, the severity remained higher in Kenyan accessions than Pacific-Caribbean taro. This was a clear indication of higher susceptibility to taro leaf blight of the Kenyan taro

accessions than the Pacific- Caribbean (Figure 1). The highest disease severity for both the Pacific -Caribbean and the Kenyan took place in April with error bars showing significantly higher infection in Kenyan taro (53.5%) than the Pacific-Caribbean ones (33.2%) during the month of April. December and January however, showed minimal disease severity with both Pacific-Caribbean and Kenyan taro (Figure 1).

The study showed that disease severity remained low at initial stage but increased progressively for both Kenya and Pacific- Caribbean. This present finding suggests that during the first stages of TLB infection, taro uses non-specific mechanisms to eliminate pathogens by increasing phenolic levels which protects cells from oxidation and accelerate recovery during inflammation. Harplapur, (2005) in his research on epidemiology and management of *turcicium* leaf blight of maize caused by *Exserohilum turcicium* reported a similar result that more susceptibility of wheat plants to *Helminthosporium sativum* mostly occurred at a later age after flowering. There was higher increase in disease severity in Kenyan taro than the Pacific- Caribbean (Figure 1). The low severity value suggests that a number of Pacific -Caribbean accessions investigated were resistant to taro leaf blight.

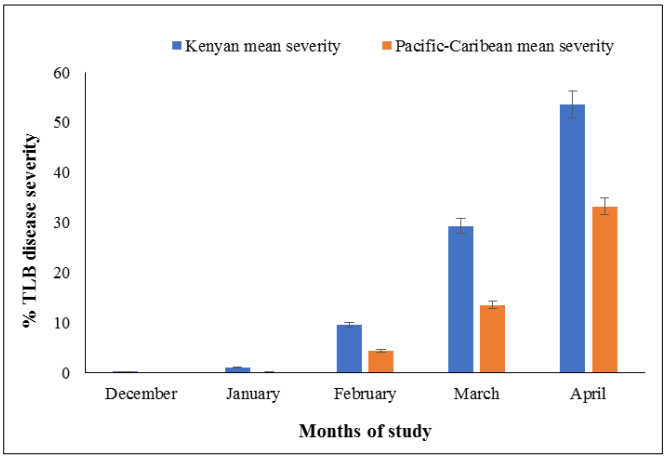

Figure 1 Comparison of taro leaf blight disease severity between Kenyan and Pacific -Caribbean taro under field study 2 of Dec 2013-April 2014

3.5. Taro leaf blight disease severity of Pacific- Caribbean taro greenhouse study of September 2015 -January 2016

Table 4 Percentage TLB disease severity on Pacific-Caribbean accessions of taro under greenhouse study

Month and age	Sept (3 mth)	Oct (4 mth)	Nov (5 mth)	Dec (6 mth)	Jan (7 mth)	
Pacific taro	**Mean TLB disease severity**					**Pooled mean**
BL/HW/08	11.78±21.71	14±20.8	20.56±19.9	26.11±27	27.78±26	20±23.2
BL/HW/26	3.22±4.09	3.56±3.91	6.67±5	11.67±10.9	21.67±23	9.4±13.2
BL/SM/80	12.56±12.2	17.78±16.8	30.56±24.3	30.56±24.3	36.11±28	26±22.8
BL/SM/92	5.11±4.78	10±9.68	21.67±22.7	23.33±22.2	11.67±11	14±16.6
CA/JP/03	2.44±3.21	5.89±4.96	11.67±10.9	22.22±19.5	33.33±25	15±18.5
CE/IND/01	9.33±11.82	20.89±23.2	25±21.6	30.56±24.3	33.33±25	24±22.4
CE/THA/07	16±20.76	14.78±20.4	25.56±31.96	27.78±26.35	41.67±33	25±27.6
CE/THA/24	12±16.35	20.56±19.9	36.11±28.26	41.67±33.07	41.67±33	30±28.5
MEAN	9.06±11.9	13.4±14.96	22.23±21	26.74±23.46	30.9±26	
LSD	6.14	7.5	10.17	10.82	11.91	
C.V	151.66	124.87	102.3	90.5	86.21	111.68

The result on taro leaf blight disease severity on Pacific- Caribbean greenhouse taro was as indicated on table 4 below. The *Colocasiae* blight disease severity steadily increased from third month to seventh month. Samoa accession BL/SM/92 increased in severity from September to December then decreased from December to January. This reflected a unique characteristic of the taro accession. The mean disease severity per month showed that September was significantly ($p<0.05$) the least in disease severity with a severity of 9.06% while January was significantly ($p<0.05$) the highest with a severity of 30.9% (Table 4). Thailand accession CE/THA/24 scored significantly ($p<0.05$) the highest mean severity of 30% while the Hawaiian accession BL/HW/26 recorded significantly ($p<0.05$) the least disease severity of 9.4%.

The result revealed that different locations from which taro was obtained influenced TLB disease severity. Even the taro accessions from Pacific -Caribbean portrayed differences in disease severity. This could be attributed to genetic and environmental differences as described by [4].

3.6. Taro leaf blight disease severity of Kenyan taro in the greenhouse (September 2015 -January 2016)

Results of greenhouse experiment on Kenyan taro are shown on table 5. Accession KNY/BSA/41 scored significantly ($p<0.05$) the highest blight disease severity of 44%. The significantly ($p<0.05$) lowest severity of 20% was recorded with accession KNY/KAK/16 (Table 5). All the accessions showed a steady increase in disease severity from the third to seventh month (Table 5). The study showed that as the plants increased in age, severity of TLB also increased [32, 33].

Table 5 Percentage of TLB disease severity on Kenyan accessions of taro under greenhouse

Month and age	**Sept (3 mths)**	**Oct (4 mths)**	**Nov (5 mths)**	**Dec (6 mths)**	**Jan (7 mths)**	
Kenyan taro	**Mean TLB disease severity**					**Pooled mean**
KNY/BSA/41	34.03±26.8	40.28±30.6	45.28±34.3	48.1±35.6	50.14±36	44±33.1
KNY/CNT/33	17.6±17.2	27.5±23.9	36.32±28.5	41.4±31.8	45.56±34	34±29.3
KNY/KAK/16	4.36±9.21	11.19±13.9	21.78±23.3	30.7±27.8	33.89±29	20±24.5
KNY/KIS/81	7.97±10.6	19.03±18.5	31.33±25.5	42.8±32.6	48.78±36	30±30
KNY/KTL/61	18.33±26.2	23.33±25.5	25±25	36.1±28.3	44.44±35	29±28.6
KNY/MU/75	9.28±13.9	13.28±13.6	24.33±21.7	35.4±30.1	41.39±34	25±26.8
KNY/SYA/50	8.44±9.86	12.56±11.8	21.94±20.5	33.9±30.4	45.83±35	25±26.9
KNY/SYA/51	9.56±12.7	21.11±23.9	36.28±28.5	41.7±32.1	45.28±35	31±30.1
MEAN	13.7±15.8	21.04± 20.2	30.28±25.9	38.8±31.1	44±34	
LSD $p<0.05$	8.38	10.34	12.35	14.08	15.12	
CV	130.07	103.8	86.8	79.68	76.15	97

Severity was lowest at 13.75% during the month of September when the plants were three months old, it was however highest in the last month of study with a severity mean of 44% (Table 1.5). The result showed increase in TLB disease severity with increase in age [31] that late growing periods of taro revealed higher TLB disease levels than early periods.

3.7. Comparison between taro leaf blight disease severity on Kenyan and Pacific- Caribbean in a greenhouse study

Comparison of disease severity by region was given on figure 2. Both categories of taro accessions showed steady increase in TLB disease severity from the third month to the seventh month. The error bars showed that Kenyan accessions were significantly higher in disease severity than the Pacific- Caribbean taro particularly in January. The highest disease severity for the Kenyan accession was 44% in January while the highest for Pacific -Caribbean was 30% also in January (Figure 2). The lowest disease severity for Kenya was 13.7% while the lowest severity for Pacific - Caribbean was 9.06%.

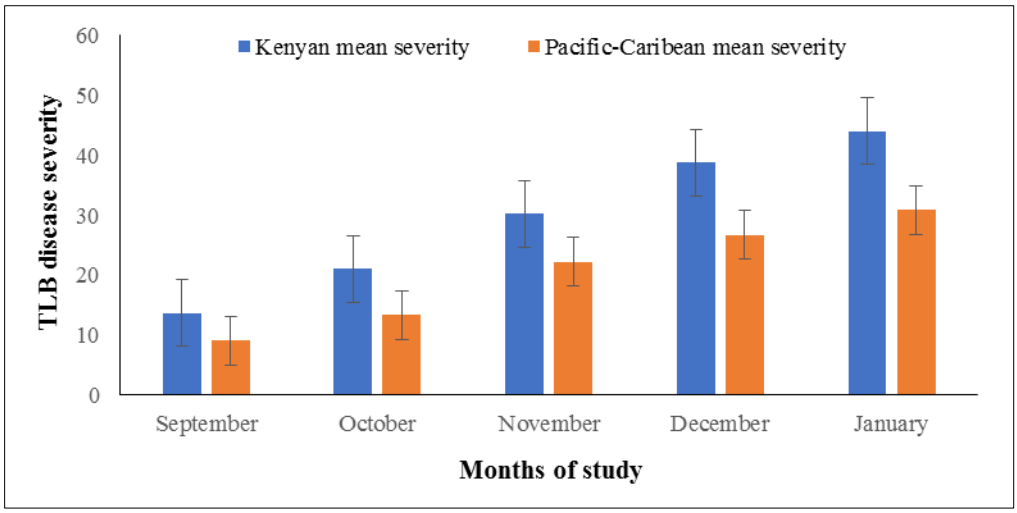

Figure 2 Comparison of mean percentage TLB disease severity on Kenyan and Pacific-Caribbean taro under greenhouse study

This could be a proof that Pacific -Caribbean taro had earlier been screened for taro leaf blight disease and were more tolerant to TLB disease. This was consistent that improved Samoan accession BL/SM/132 from Pacific-Caribbean neither showed tissue collapse even after TLB pathogen inoculation nor symptom of taro leaf blight disease compared to other taro accessions which showed high severity rates [30]. Adomako et al., [27] attributed the differences in disease severity to genetic differences among taro plants. There was a differential degree of response against taro leaf blight disease among different taro accessions [18].

The increase in disease development in Pacific-Caribbean taro was relatively low as compared to Kenyan. These results alluded to more physiological adaptation of *Phytophthora colocasiae* on Kenyan than Pacific-Caribbean improved accessions. In the TLB-Kenya pathosystem, there is evidence of physiological adaptation of *P. Colocasiae*. This finding was also in concurrence with the report of CABI [12] that the role of pathogen variability and adaptation cannot be precluded and may in fact account for the observations.

4. Conclusions and recommendations

Region of origin has been found to have effect on TLB disease severity. The two categories of taro accessions (Kenyan and Pacific-Caribbean) differed in their disease severity levels. Kenyan categories were generally more severely infected by taro leaf blight than the Pacific -Caribbean categories. More accessions from different parts of the continent should be investigated for TLB disease severity to enable more comparison and to arrive at the most resistant accession. The months of the year varied in disease severity. This has possibly been linked to the effect of varied weather conditions which have profound effect on the prevalence of the disease. When the temperatures are high and relative humidity is favorably high, disease severity increase. The experiment revealed that the disease spreads very fast particularly when conditions are favorable and can be very destructive. The study of severity of taro leaf blight in response to various weather conditions should be extended to other taro producing regions of Kenya. The period of study needs to be extended to capture more years with varied weather conditions.

Compliance with ethical standards

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Disclosure of conflict of interest

The authors proclaim that there is no conflict of interests with respect to publication of this paper.

References

[1] Otieno CA, Palapala VA and Opande GT. (2018). Evaluation of taro leaf blight disease (*Phytophthora colocasiae*) incidence on Kenyan and Pacific Island taro (*Colocasiae esculenta*) accessions in Kakamega (Western Kenya). International Journal of Biosciences, 12(6), 355-361.

[2] Tsopmbeng GR, Lienou JA and Fontem DA. (2014). Influence of altitudes on Sporangia size and aggressiveness of Phytophthora colocasiae isolates in Cameroon. International journal of sciences: Basic and applied research (IJSBAR), 13(1), 333-341.

[3] Mbong GA, Fokunang CN, Manju EB, Njukeng AP, Tembe-Fokunang EA and Itanna R. (2015). Mycelia growth and sporulation of *Phytopthora colocasiae* isolates under selected conditions. American Journal of Agriculture, 8(4), 193- 201.

[4] Miyasaka SC, Culloch MC and Nelson SC (2012). Taro germplasm evaluated for resistance to taro leaf blight 22(6).

[5] Adinde JO, Anieke US, Nwankwo OG, Agu CJ, Aniakor AC, Nwagboso AA and Eze CO (2016). Incidence and severity of taro leaf blight in Iwollo, South Eastern Nigeria. International journal of current research in Biosciences and plant Biology, 3(10), 163-168.

[6] Chiejina NV and Ugwuja FN. (2013) Incidence of *Phytophthora* Leaf-Blight Disease of Cocoyam in Nsukka Area of South-Eastern Nigeria. Journal of Botanical Research, 4(1), 21-24.

[7] Ayogu CJ, Ike CU, Ogbonna OI and Nnaemeka Gk. (2015). Agricultural Extension Roles towards adapting to the effects of taro leaf blight (TLB) disease in Nsukka Agricultural Zone, Enugu State. Journal of Biology, Agriculture and Health care, 5(12), 2015.

[8] Shakywar RC, Pathak SP, Pathak M, Tomar KS and Singh H. (2013). Developmental behavior of leaf blight of taro caused by *Phytophthora colocasiae*. Society for plant research, 26(1), 167-170.

[9] Adipala E, Lipps PE and Madden LV. (1993). Use of disease assessment methods in predicting yield loss due to northern leaf blight of maize. African crop science journal, 1(2), 159-173.

[10] Fullerton R and Tyson J. (2003). The biology of *Phytophthora colocasiae* and implication for its management and control. In Proceedings of the Third Taro Symposium, Nadi, Fiji Islands, Secretariat of the Pacific Community: Noumea, New Caledonia, 107– 111.

[11] Ayogu CJ, Ike CU, Ogbonna OI, Nnaemeka Gk. (2015). Agricultural Extension Roles towards adapting to the effects of taro leaf blight (TLB) disease in Nsukka Agricultural Zone, Enugu State. Journal of Biology, Agriculture and Health care, 5(12), 2015.

[12] Cabi. 2016. *Phytophthora colocasiae* (taro leaf blight) accessed at http://www.cabi.org/isc/datasheet/40955 on 13/11/2017.

[13] Rai B, Kumar S and Kumar B. (2002). Effect of environmental parameters on the development of turcicum leaf blight of maize. Annals of Biology, 18, 153-155.

[14] Adipala E, Lipps PE and Madden LV. (1993) Use of disease assessment methods in predicting yield loss due to northern leaf blight of maize. African crop science journal, 1(2), 159-173.

[15] Ramathani I. (2009). Characterization of turcicum blight epidemics and pathogen population in the *Exserohilum turcicum*-Sorghum pathosystem in Uganda. *MSc Thesis, Makerere University Kampala, Uganda.*

[16] Benzohra IE, Bendahmane BS, Benkada MY and Labdi M. (2018). Temperature effects on cultural and morphological aspects of *Ascochyta fabae* Sand, agent of Ascochyta Blight on Faba Bean (*Vicia faba* L. Subsp. Major). Advances in Biological Research, 12(2), 58-63.

[17] Tsopmbeng GR, Lienou JA and Fontem DA (2014). Influence of altitudes on Sporangia size and aggressiveness of Phytophthora colocasiae isolates in Cameroon. International journal of sciences: Basic and applied research (IJSBAR), 333-341.

[18] Scot N, Brooks F and Teves, G. (2011). Taro Leaf Blight in Hawaii 'I. Plant Disease, 71, 1 - 14.

[19] Agrios GN. (2005). Plant Pathology (5th ed). Academic Press, New Yolk.

[20] Whehan HG. (1992). The effect of crop yield potential on disease yield loss relationship in barley. A thesis submitted in fulfillment of the requirements for the degree of Doctor of Philosophy in crop science at Lincoln university-New Zealand.

[21] Asraku JS (2010). Identification of the major foliar fungal disease of *Colocasiae esculenta* (L.) Schott and its management in the Kumasi Metropolis. A thesis submitted to the school of research and graduate studies, Kwame Nkrumah in partial fulfilment of the requirement for the award of the degree of Masters of Science in plant pathology, KNUST, 1-68.

[22] Wambua J. (2004). Mushroom cultivation in Kenya. Mushroom community supporting group. Mushroom growers hand book chapter 10.

[23] Singh D, Jackson D, Hunter D, Fullerton R, Lebot V, Tailor M, Josef T, Okpul T and Tyson J. (2012). Taro Leaf Blight-A threat to food security. Open access Agriculture 2, 182-203.

[24] Brooks FE. (2011). Methods of measuring taro leaf blight severity and its effects on yield. www 2.ctahr.hawaii.edu/adap/scc-/and Grant/ Dr brooks accessed 3/9/2012.

[25] Nath VS, Hedge VM Muthulekshmi LJ, Misra R S, Veena SS, Jaj M, Suresh K, Unnikrashnan and Darveekaran SS. (2014). Rapid and sensitive detection of *Phytophthora colocasiae* responsible for the taro leaf blight using conventional and real –time PCR assay.

[26] Shrestha SK. (2012). Investigation of *Phytophthora species*: *Phytophthora colocasiae* on taro and *Phytophthora* recovered from streams in Eastern Tennessee. A thesis presented for the master of science degree- University of Tennessee, 4, 1-57.

[27] Adomako J, Kwoseh CK, Moses E and Larbi-Koranteng S. (2016). Prevalence of *Phytophthora* Leaf Blight of taro (*Colocasiae esculenta* (L.) Schott) in the semi deciduous forest zone of Ghana. *AJEA,* 11(4), 1-7.

[28] Lin MJ and Ko WH. (2008). Occurrence of isolates of *Phytophthora colocasiae* in Taiwan with homothallic behavior and its significance. Mycologia, 100, 727-734.

[29] Manza WS, Zarafi AB and Alabi O. (2008). Incidence of leaf blight caused by *Fusarium pallidoroseum* on varied age of caster (*Ricinus communis*) inoculated using different methods. African journal of general Agriculture 4(2) 1-5.

[30] Chan LF, Lu CT, Lu HY and Lai CH. (1993). A simple method for estimating leaf area in wetland taro (*Colocasiae esculenta* (L.) Schott). Journal of Agricultural Research, 42, 162–172.

[31] Simongo DK, Gonsales IC and Mesangkei. (2016). North Philippine root crop research and training center, Benguet state University, La Trinidad, Benguet 2601. International Journal of advancement in research and technology, 5 (6).

[32] Omege TE, Ugwuoke KI, Adinde JO, Ogwulumba SI and Unigwe LO. (2016). Effect of cropping season on the control of taro leaf blight (*Phytophthora colocasiae*) of cocoyam (*Colocasiae esculenta* C.) in Nsukka, South Eastern Nigeria. International journal of advanced biological research, 6(1), 30-39.

[33] Sarkar, Adhikary NK and Tarafdar J. (2017). Field management of taro leaf blight using promising germplasm. International Journal of Current Microbiology and Applied Sciences, 6(12), 1399-1407.

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