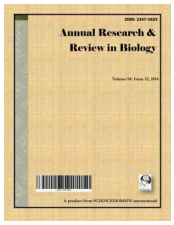

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Assessing the Incidences of Late Blight Disease on Irish Potato Varieties in Kisii County, Kenya

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Authors' contributions

This work was carried out in collaboration between all the authors. Author MOM designed the study wrote the protocol, gathered the initial data and performed preliminary data analysis. Authors JN and JKM being supervisors, managed the literature searches, anchored the field study, interpreted the data and produced the initial draft. All authors read and approved the final manuscript.

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ABSTRACT

Background: Late Blight Disease, caused by the fungal pathogen, *Phytophthora infestans,* is a major constraint of Irish potato production in Kenya. The disease can destroy a crop, causing up to 100% yield loss. Small scale holder farmers in Kisii County continuously grow Irish potato that is susceptible to *P. infestans* which require a number of fungicides sprays. The study was formulated out of the realization that Irish potato plays a major role in food security and contributes to poverty alleviation. Also, the commonly used protectant fungicides for the crop are expensive, hazardous and are not effective against *Phytophthora infestans*.
Aim: To assess the incidences of late blight disease on Irish potato varieties in Kisii County, Kenya.
Study Design: The Irish potato farms were selected randomly following a line transect. Twenty nine stops at every ten kilometres were made, three potato farms around every stop were inspected for the presence of late blight infected Irish potato varieties. A total of eighty seven farms were inspected for the presence of late blight. Five late blight infected leaves from each farm were collected for examination in the laboratory.

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Methodology: A random sampling method was used to collect late blight infected Irish potato plants from various farms along the transect routes. *P. infestans* were isolated, cultured on a potato dextrose agar and identified by microscopic techniques at National Agricultural Research Laboratories and Kenya Agricultural research and Livestock Organisation Kisii. An inoculum prepared was standardized to $1x10^7$ sporangia/ml concentration using haemocytometer. Pathogenicity test was done on health Irish potato plants to confirm the pathogenicity nature of the *Phytophthora infestans*. The data collected was subjected to analysis of variance using GENSTAT directive version 12.0 and chi-square.
Place and Duration of the Study: Sampling was carried out in the month of April, May and June 2015 during the long rain season in Kisii County, Kenya.
Results: Late blight disease was found to be rampant in all the 87 farms inspected in the County. Late blight effect differed significantly at $P<.001$ among the Irish potato varieties assessed. There was no significant different among the farms inspected and this indicated that all farms visited were infected with late blight the same way. Along the transect routes six Irish potato varieties were identified notably Mang'ere (55.6%), Nyayo (17.2%), Tigoni (3.4%), Asante (3.4%), Shangi (10.3%) and Purple gold (10.3%).
Conclusion: Late blight disease remains a serious threat to Irish potato production causing significant yield and economic losses to farmers. The diversity of *Phytophthora infestans* in the region calls for the most sustainable approach for reducing late blight problem significantly and this would enable the farmers to get good yields from their Irish potato crop.

Keywords: Assessment; incidences; Phytophthora infestans; varieties; Solanum tuberosum; Kisii.

1. INTRODUCTION

The Late Blight Disease (LB) is caused by a fungus, *Phytophthora infestans*. Irish potato blight is one of the most devastating plant diseases. The disease destroyed Irish potato crop and led to mass starvation. In the great Irish famine of 1845 to 1847, up to one million people died and similar number of people emigrated to the rest of Europe and United States of America. It was not until 1861 that Antony de Barry, who was considered the father of plant pathology, that the question as the cause of blight was finally settled [1]. Irish potato was introduced to East Africa by British farmers in the 1880s. It was first introduced in Kenya by Irish missionaries, for whom this was the staple food hence the name "Irish potato" [2]. In Kenya Irish potato had been grown for more than half a century before blight was first observed and reported in 1941 [3]. In Kenya Irish potato (*Solanum tuberosum* L*)* would easily be grown, providing more nutritious food faster on less land than any other food crop. Potato plays a major role in food security and contributes to poverty alleviation through income generation and employment creation. It is mainly grown by smallholder farmers as a cash crop and therefore plays an important role in food security [4,5]. The major constraint of Irish potato production include late blight disease [5]. Limited availability of good quality potato seeds and low yielding varieties are also additional production constraints experienced by Irish potato farmers in the County. Irish potato yields have been low due to attack by late blight disease, caused by *Phytophthora infestans* [3,4,6]. The magnitude of yield loss is high due to susceptibility of Irish potato varieties to *Phytophthora infestans* [7]. The diversity of *Phytophthora infestans* in the region has been aggravated by many alternative hosts to the pathogen and small scale holdings of land, which has led to impracticable crop rotations. Irish potato (*Solanum tuberosum* L) belongs to genus *Solanum* and *Solanacea* family. Late blight disease attacks all solanaceous plants [8]. The numerous plant species attacked by late blight disease include not only important economic hosts such as tomatoes (*Lycopersicon esculentum*), peppers (*Capsicum spp*), black night shade (*Solanum nigrum*) but also a number of weeds such as Sodom apple (*Solanum incanum*). The weather conditions prevailing in the study area favours *P. infestans* spores' development and germination on wet leaves. During humid weather, whole plants may be attacked and killed in a short time. Late blight disease of Irish potato is one of the diseases that can destroy a crop, causing up to 100% yield loss [7,9]. Late blight disease of Irish potato is a serious production constraints in major growing regions of Kenya [7]. Except for the two varieties released in 1998 (Tigoni and Asante), most of the others including nearly all the farmer's types are susceptible to late blight disease. Lack of quality potato seeds which are resistant to *Phytophthora infestans*

among growers is also a major problem adversely affecting the expansion of Irish potato production in Kenya [3,4,10]. The study was formulated out of realization that Irish potato plays a major role in food security and contributes to poverty alleviation. The commonly used protectant fungicides against late blight to control the disease, are expensive, environmentally unfriendly, pose serious health hazards for small holders who grow most of the crops and are not effective against *Phytophthora infestans* [3]. As a basis of the information technology gaps about incidences of Late blight disease, there is need to assess the occurrence of Late blight disease and their effect on Irish potato varieties grown in the County.

2. MATERIALS AND METHODS

2.1 Research Site

This research was conducted in a randomly selected farms following a line transect. The County is located at 0°41' 0 S and 34°46' 0"E. It exhibits a highland equatorial climate with a bimodal rainfall pattern with average annual rainfall of 1,500 mm, with long rains between February and June, and short rains between September and November. The Months of January and July are relatively dry. The area has Maximum day time temperature range between 21 and 30℃ and minimum temperature range between 15 and 20℃. The altitude is between 1500 m-1800 m above sea level and above. The area provides favourable environmental conditions for late blight development and progress on Irish potato varieties [11].

2.2 Sample Size and Sampling

The farms were selected using random sampling method following a line transect. According to the terrain, nine transect routes that included Keroka-Nyangusu; Nyangusu-Kisii; Keroka-Kisii; Mogonga-Nyamarambe; Masimba-Nyamasibi; Kiamokama-Keumbu; Kisii-Suneka; Nyamache-Nyabisabo and Masimba- Mogweko- Keroka transects were used. During sampling, 29 stop overs were made and three Irish potato farms around every stop were inspected for the presence of infected Irish potato varieties by *Phytophthora infestans*. At each farm, collections of infected Irish potato leaves samples were made and occurrence of late blight disease was scored. Five late blight disease infected Irish potato leaves from each farm were randomly sampled and a total of 435 infected leaves were collected from the study area. To assess the incidences of Late blight disease on Irish potato varieties in Kisii County a random sampling method was used to collect late blight diseased plants from the various farms [12]. The researcher and extension officers from the Ministry of Agriculture travelled using a vehicle along a given transect route, and made stops at every 10 km and scored late blight incidences. The incidences was established by calculating the percentage of blighted plants per farm and was subjected to ANOVA test. The prevalence of late blight was calculated as the number of farms affected by the disease expressed as percentage of all farms visited in the County and was subjected to ANOVA and Chi-square tests. Late blight disease infected Irish potato leaves were identified and collected randomly based on the symptoms, marked and grouped according to the varieties [13]. They were then placed in a cool moist box and transported to the laboratory for microscopic examination [14]. Late blight infected Irish potato leaves samples from each variety were randomly picked and used to isolate *Phytophthora infestans* [15]. Irish potato varieties which were growing in the study area were also collected and identified along the transect routes in each farm inspected and records were made.

2.3 Laboratory Experiment

Sixty petri-dishes were sterilised in a potable autoclave for fifteen minutes at a temperature of 121℃ and at 1atmospheric pressure and cooled to 55-60℃. The petri-dishes were then dried with the lids half-off in the lamina flow chamber and were grouped according to the number of Irish potato varieties from where the infected leaves were obtained. Thirty nine grams (39 gms) and 31.6 gms of potato dextrose agar was added to one litre and 810 ml sterile distilled water respectively. It was autoclaved at 121℃ for 15 minutes. The potato dextrose agar solution was cooled to 50-60℃ before pouring into the petri-dishes [16]. This yielded sixty petri-dishes. Thirty millimetres of the sterile potato dextrose agar media was poured into each sterile petri-dish. It was then placed in a lamina air flow chamber and was left to solidify for two hours. The late blight infected Irish potato leaves from the field were washed under running tap water. They were surface sterilised by dipping in 70% ethyl alcohol for 30 seconds and were then gently blot dried on paper towel for quick removal of alcohol [16]. Small fresh and recently late blight infected leaf piece from the edge of actively growing

lesion were cut using a sterilised scalpel and were placed in each petri-dish containing solidified potato dextrose agar. They were then incubated at 18℃ for fourteen days in a germination chamber. The petri-dishes were examined regularly for the growth of mycelium and sporangia and other morphological characteristics [16]. The mycelium and sporangia were isolated, mounted on a microscope slide and were observed [14]. The identified *Phytophthora infestans* spores were isolated and transferred to a fresh sterile potato dextrose agar and was incubated at 18℃ for fourteen days and a pure culture was prepared. The petri-dishes were examined regularly for the growth of the sporangia [15]. The sporulating petri-dish was flooded with 10 ml distilled sterile water and *P. infestans* spores were harvested [16] and a total of six hundred millimetres of the sporangia suspensions were obtained from sixty petri-dishes. Serial dilutions were made and standardized using a haemocytometer until a concentration of the inoculum of $1x10^7$ sporangia/ml in a fresh pure culture was obtained [16]. Pathogenicity test was done by spraying the prepared inoculum into healthy Irish potato test plants using hand pump sprays. The Irish potato plants were observed regularly for the symptoms after inoculation [13]. The *P. infestans* were isolated from the inoculated potato test plants, cultured and were microscopically identified [14].

2.4 Data Analysis

Late blight infected leaves were collected randomly following a line transect from various farms. The data collected was subjected to analysis of variance (ANOVA) using GENSTAT directive version 12.0 and Chi-square.

3. RESULTS AND DISCUSSION

Late blight was found to be rampant and differed significantly among the Irish potato varieties in the region (Tables 1 and 3). Late blight infected Irish potato farms indicated that the diversity of *Phytophthora infestans* were mainly aggravated by high humidity and wet months during the study which provided favourable conditions for the growth of *P. infestans* spores. (Figs. 1, 3a, 3b and 3c). These results are in conformity with [11] who reported about the rainfall patterns in the region and [7] who reported about weather conditions favourable for *P. infestans* spores development and germination on wet leaves. Petri-dishes were dried with the lids half-off in the lamina air flow chamber and this discouraged any free water or condensation on the lids as water encourages the growth and spread of bacterial contaminants. Surface sterilized late blight infected leaves were blot dried, and this prevented bacterial growth which suppressed the growth of *Phytophthora infestans. Phytophthora infestans* were incubated at a temperature of 18℃ and in a dark germination chamber which provided favourable conditions for its growth and also slowed down bacterial growth. *P. infestans* produced sporangia quickly on the surface of potato dextrose agar leaving the bacterial contaminants behind. The sporangia of *P. infestans* were observed at the tips of the sporangiophores and they appeared lemon shaped under microscopic examination. The sporulating *P. infestans* in the Petri-dishes were identified based on branching of the sporangiophores on which the sporangia were borne, the characteristics of the mycelium and the spores produced on the potato dextrose agar assisted in identification. This results are in conformity with that of [14,15,16] who reported about the morphological characteristics of sporulated *P. infestans.*

Pathogenicity test which was done on healthy Irish potato test plants proved Koch's postulates which showed that *Phytophthora infestans* cause brown/black lesions on plant leaves and stems and this confirmed presence of the pathogen in the inoculum prepared (Figs. 3b and 3c). The symptoms on leaves was small at first and appeared water soaked, but soon expanded rapidly and became necrotic (Fig. 1). These results are in agreement with the findings of [13] who reported about late blight symptoms. The result in Table 1 indicated that late blight infected samples were significantly different at $P<.001$ among the varieties (Table 1, Figs. 1, 3a, 3b and 3c). Late blight infected Irish potato samples were significantly different $F_{(5, 48)} = 8.77$, $p<.001$ along the transect routes (Table 1). Mang'ere and Nyayo were identified as susceptible varieties. In this study, humid conditions aggravated sporulation which resulted in a visible white growth at the leading edge of lesions on lower surface of leaves. It was a white growth that distinguished late blight from other foliar diseases. This result was in conformity with [16] who reported about late blight white growth. *Phytophthora infestans* were found destructive on susceptible Irish potato varieties grown (Figs. 3b and 3c). This result is in agreement with the findings of [7] who reported about susceptible Irish potato varieties.

Fig. 1. Shangi Irish potato variety severely infected with late blight disease in one of the farmer's plots

Source: Researcher

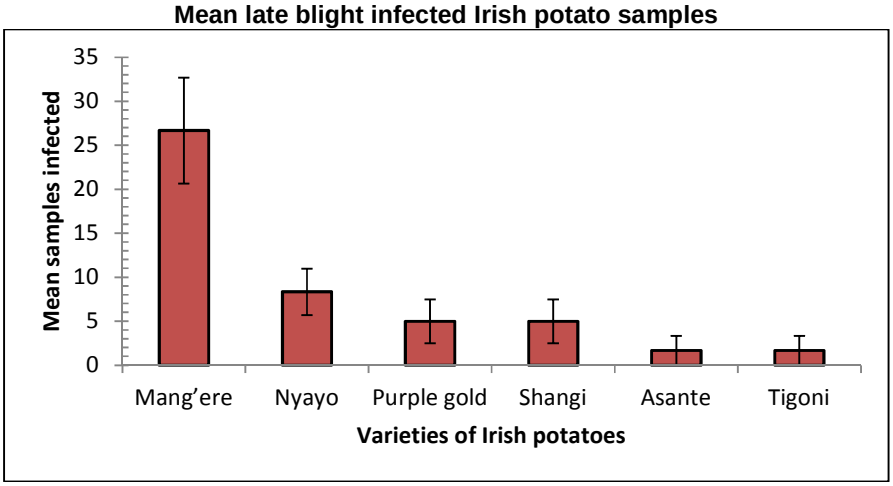

Fig. 2. Bars representing standard error of means of LB infected Irish potato varieties

Table 1. ANOVA test for late blight infected Irish potato samples

S. no	Source of variation	d.f.	s.s.	m.s.	F	p-value
1.	Variety	5	4020.83	804.7	8.77	<.001
2.	Residue	48	4400	91.67		
	Total	53	8420.83			

Significant at p<.001

Table 2. Prevalence of Irish potato late blight disease in Kisii County

S. no	Transect route	Observed N	Expected N	Residual
1	Keroka-Kisii	9	9.7	-.7
2	Kiamokama-Keumbu	9	9.7	-.7
3	Kisii-Suneka	3	9.7	-6.7
4	Masimba-Mogweko-Keroka	9	9.7	-.7
5	Masimba-Nyamasibi	9	9.7	-.7
6	Mogonga-Nyamarambe	15	9.7	5.3
7	Nyamache-Nyabisabo	9	9.7	-.7
8	Nyangusu-Keroka	9	9.7	-.7
9	Nyangusu-Kisii	15	9.7	5.3
Total	9	87	87.3	-38.1

a **b** **c**

Figs. 3a, b and c: Show different late blight reactions to various Irish potato varieties
Source: Researcher

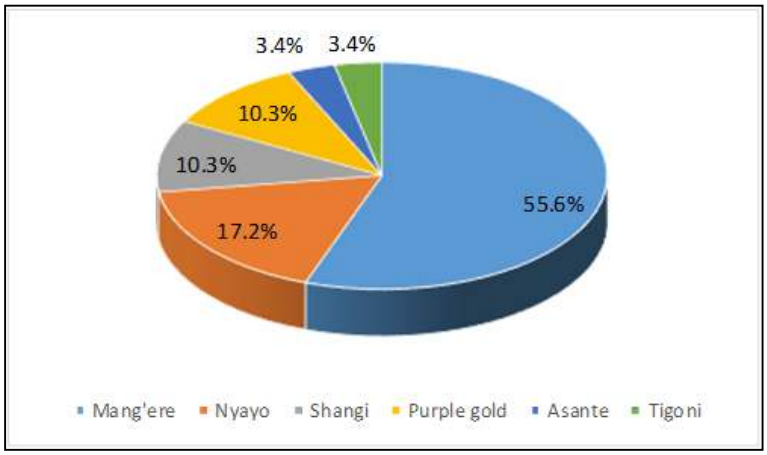

Fig. 4. Percentage distribution of Irish potato varieties in the County
Source: Researcher

There was no significance difference among the late blight infected farms (Tables 2 and 3). The results in Table 3 indicated that all the farms visited were infected with late blight disease the same way (Tables 2 and 3). The Irish potato identified had different reactions to *Phytophthora infestan* (Figs. 1 (Shangi), 3a (Purple gold), 3b (Mang'ere) and 3c (Nyayo)). These results are in agreement with the findings of [3] who reported that late blight is a major constraint of Irish potatoes and [7] who reported that the disease can destroy a crop causing up to100% yield loss (Figs. 3b and 3c).

Table 3. Chi-square test for the prevalence of late blight

S. no	Test statistics	Transect
1	Chi-square value	10.759
2	d.f.	8
3	p-value	0.216*

Irish potato varieties grown for instance Mang'ere (Fig. 3b) and Nyayo (Fig. 3c) were susceptible to late blight disease in all the farms visited. This findings are in conformity with [7] who reported about lack of quality Irish potato seeds.The result indicated that six Irish potato varieties were identified in the County along the transect routes: Mang'ere (55.6%), Nyayo (17.2%), Shangi (10.3%), Purple gold (10.3%), Asante (3.4%) and Tigoni (3.4%) (Fig. 3). The local Irish potato varieties were grown as intercrop in some farms, others as pure stand and growing naturally.

4. CONCLUSION

On the basis of the present study late blight disease remains a serious threat to Irish potato production causing significant yield and economic losses to farmers. This study confirmed that late blight disease was rampant in the region and the environmental conditions

available were favourable for the pathogen development and progress on Irish potato varieties. Late blight infected Irish potato were significantly different among the varieties. All the farms visited were infected with late blight the same way. The diversity of *Phytophthora infestans* in the region calls for the most sustainable approach for reducing late blight problem significantly and this would enable the farmers to get good yields from their Irish potato crop. These results along with similar information and other Irish potato production constraints can be used as a basis for research on the most sustainable management for reducing late blight disease problem significantly.

5. RECOMMENDATION

Further research is recommended to determine the effect of host tolerance on foliar late blight severity, diseases development and progress on selected Irish potato varieties. This recommendation comes from the findings that late blight is rampant and infected Irish potato varieties differed significantly in the County. This was mainly contributed by susceptible Irish potato varieties, high humidity and wet conditions which were favourable for *Phytophthora infestans* survival. Therefore the protectant fungicides available are not effective in the control of late blight and this calls for Irish potatoes which are tolerant to the pathogen.

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COMPETING INTEREST

Authors have declared that no competing interests exist.

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