African Phytosanitary Journal

Volume 2, Issue 1

November, 2020

EDITORIAL MANAGEMENT

Contact us:

Kenya Plant Health Inspectorate Service (KEPHIS)

P.O.Box 49592-00100

Nairobi, Kenya

Email: africanphytosanitaryjournal@kephis.org , director@kephis.org

Phone: +254206618000

Mobile: +254709891000

Foreword

The African Phytosanitary Journal is a peer refereed academic journal which is one of its kind. It provides a platform to inform the stakeholders involved in trade in agricultural products. This journal provides a clear understanding of phytosanitary issues in Africa and beyond. Subsequently, it provides a platform for the stakeholders to stay informed of the current and emerging phytosanitary issues. Research Institutes, National Plant Protection Organizations (NPPOs), Universities and industry constantly conduct research and come up with innovations and discoveries that affect plant health in one way or another yet the findings are not disseminated to the intended consumers in time.

With increased globalization, there is imminent increase in trade involving plant and animal products. It is prudent that this trade be conducted with materials free of contaminants. The regulatory protocols developed to actualize this are science based and are in line with sanitary and phytosanitary requirements. The stakeholders involved in this line of trade need to be updated regularly on the requirements inorder to facilitate their activities. Cognizant of this fact, the African Phytosanitary Journal provides a platform for research scientists, academia and industry to share their experiences and innovations. This is an open access journal with a wide scope in sanitary and phytosanitary issues. This issue has focused on pest management, pest surveillance, pest risk analysis, emerging technologies, pest identification and analysis.

The mission of this journal is to foster a deeper understanding of phytosanitary issues in Africa and provide a basis for their management. I would like to take this opportunity to thank the team that worked tirelessly on this journal and made the release of this issue possible. Lots of gratitude to the editorial board, the reviewers and the authors for their time and energy spent towards the production of this journal.

Dr. Isaac Macharia

Editor in Chief

Scope of the journal

This journal has been developed to bridge the knowledge and information gap in the following thematic areas:

- Pest surveillance
- Pest reporting
- Phytosanitary measures
- Pest Risk Analysis
- Pest identification and analysis
- Food safety
- Quarantine and Biosecurity
- Phytosanitary policy and regulation
- Phytosanitary treatment
- Emerging technologies
- Biological agents
- Pest Management
- Agricultural Chemistry
- Emerging phytosanitary issues
- Biosafety
- Phytosanitary issues on trade
- Other relevant phytosanitary issues

Table of Contents

EDITORIAL MANAGEMENT i

Foreword iii

Scope of the journal iv

Table of Contents v

Status of Maize Lethal Necrosis Disease in Zambia 1

The Effect of Seed Source and Post-harvest Practices on Quality of Soybean (*Glycine max*) Seeds in Busia County 17

Abundance and Diversity of Frugivorous Fruit Flies in Kandara, Murang'a County, Kenya 41

Next generation sequencing as a tool in modern pest risk analysis: a case study of groundnuts (*Arachis hypogaea*) as a potential host of new viruses in western Kenya 51

Maize Head Smut: Pathogenesis, Epidemiology, and Management Options 63

New scale insect country records for Kenya (Hemiptera: Coccomorpha) from old samples in insect collections 72

Pest Incursions Pose a Serious Threat To Food Security and the Kenyan Economy 105

Status of Maize Lethal Necrosis Disease in Zambia

Chomba, M. D*., Msiska, K. K., Abass M. S., Mudenda, M., Mukuwa, P.S.C.
Zambia Agriculture Research Institute, Plant Quarantine and Phytosanitary Service
P/B, 7, Chilanga, Zambia.
*Corresponding Author: Doreen Malekano Chomba, Email: dchomba71@gmail.com

Abstract

Maize is a staple food in Zambia and contributes immensely to food security for smallholder farmers. Disease outbreaks such as Maize Lethal Necrosis Disease (MLND) can be a key constraint to maize production. This disease is caused by synergistic co-infection with *Maize Chlorotic Mottle Virus* (MCMV) and any virus from the family Potyviridae, particularly, *Sugarcane Mosaic Virus* (SCMV), *Maize Dwarf Mosaic Virus* (MDMV) or *Wheat Streak Mosaic Virus* (WSMV). In 2011, an outbreak of MLND affecting almost all of the currently grown commercial varieties posed a challenge to maize production in Kenya and it has since been reported in DR Congo, Ethiopia, Kenya, Rwanda, Tanzania, and Uganda causing yield losses of up to 100%. Despite MLND having been reported in some neighboring countries, there is no information on the status of the disease in Zambia. Additionally, there is a lot of grain and seed trade between Zambia and other countries among which MLND has been reported. The aim of this study was to establish: (a) the status of MCMV; (b) agricultural practices used by farmers and (c) insect vectors associated with MLND. A survey was conducted in nine (9) provinces of Zambia during 2014/2015 and 2015/ 2016 cropping seasons. Farmers' maize fields were sampled at every five to ten-kilometer interval and tested using rapid diagnostic kits capable of detecting MCMV. Four hundred and nineteen samples collected all tested negative for MCMV. Zambian Agricultural Research Institute (ZARI), with all stakeholders in the maize value chain should continue implementing measures aimed at preventing the introduction of MLND in Zambia.

Key words: Survey, MLND, Losses, food security

INTRODUCTION

Maize (*Zea mays* L.) is one of the principal cereal crops in Sub-Saharan Africa (SSA) and is largely produced by smallholder farmers over 35 million hectares with an estimated production quantity of over 70 million metric tons of grain (Boddupalli *et al*., 2020). The crop is critical to food security in SSA; Eastern and Southern Africa uses 85% of the maize produced as food, while Africa as a whole uses 95% as food (Shiferaw *et al*., 2011). Maize is equally a very important food crop in Zambia and according to (Chapoto *et al*., 2010) the average consumption of maize grain in Zambia has been estimated at 133 kg per year making it the most popular food crop. It is cultivated in all the provinces and its production is dominated by small scale farmers who constitute an important and invaluable component of the Zambian economy (Chiona *et al*., 2014). According to the analysis of this study regarding the trend of maize production based on the crop forecasting survey estimates made available by the national Central Statistical Office (CSO) for the period 2011 to 2015, the country produced a cumulative total of 14.37 million MT. Zambia's small to medium holder farmers accounted for 89% of the total production over this period.

In terms of trade, Zambia has been a hub of seed and grain exports to her neighboring countries. A total of 120,000 MT of seed maize was exported within the Southern African Development Community (SADC) region and COMESA member countries (ACTESA, 2015), implying that maize has a significant contribution to the Zambia's Gross Domestic Product (GDP) and thereby to the National economy.

Although maize is widely grown, it is faced with several biotic constraints such as weeds, pathogens and insect pests thereby affecting its productivity (Oerke, 2006). In September 2011, a high incidence of a new maize disease called Maize Lethal Necrosis Disease (MLND) was reported at lower elevations (1,900 m a.s.l) in the Longisa division of Bomet County, Southern Rift Valley in Kenya. Since then, the disease has spread to many countries of East Africa rapidly due to insufficient knowledge on how the disease should be managed (Mahuku *et al*., 2015). MLND is caused by the synergistic co-infection of maize with *Maize Chlorotic*

Mottle Virus (MCMV) of the genus *Machlomovirus* and any of the *Potyviruses* such as: *Maize Dwarf Mosaic Virus* (MDMV), *Sugarcane Mosaic Virus* (SCMV), and *Wheat Streak Mosaic Virus* (WSMV) (Achon *et al*., 2017). In Eastern Africa, MLND was found to have resulted from co-infection of maize with MCMV and SCMV, although MCMV alone appeared to cause significant crop losses. SCMV has been known to be distributed widely in Africa since the 1970s (Mahuku *et al*., 2015). Therefore, this makes the detection of MCMV important as it is the only single virus needed together with SCMV to cause MLND (Mahuku *et al*., 2015). The disease is naturally known to affect all varieties of maize resulting in chlorotic mottling of the leaves, severe stunting and necrosis. This subsequently hinders the physiological processes of the plant such as photosynthesis, chlorophyll formation as well as denaturing enzymes necessary for the crop to produce. This further leads to low maize yields or plant death (Wangai *et al*., 2012). MLND is an economically devastating disease in maize growing areas of the world and is currently becoming an emerging threat in Africa and Asia (Achon *et al*., 2017). In Kenya, field losses for all commercial maize varieties were estimated at 30 to 100% depending on the stage of disease onset and severity (Mahuku *et al*., 2015). In 2012, MLND affected 77,000 ha in Kenya, translating into an estimated yield loss of 126 million MT valued at U.S.$52 million (Wangai *et al*., 2012a; Mahuku *et al*., 2015). Further annual losses due to MLN were estimated at 0.5 million MT/year (22%) or $187M in Kenya (De Groote *et al*., 2016). The transmission of MCMV occurs through insect vectors, mechanically, and also via seed at very low rates of about 0.04% (Jensen *et al*., 1991). This poses a challenge to detect this virus to prevent its introduction, infection and transmission (Liu *et al*., 2015). Infected soil has also been shown to transmit the viruses that cause MLN (Mahuku *et al*., 2015). Weeds such as Bermudagrass *(Cynodon dactylon)* Napier grass (*Pennisetum purpureum)* have been known to be hosts of viruses that causes MLND. It is further reported that insects such as thrips, beetles and aphids carry the viruses

from one plant to another in the field (Mahuku *et al.*, 2015).

Despite MLND being reported in some of Zambia's neighboring countries such as DR Congo and Tanzania, so far, no study has been carried out to establish its status in Zambia. In the light of this, a survey was conducted from 2015 to 2016 in order to investigate and establish if there was any occurrence of MLND. The objective of this study was to determine: (1) the presence or absence of Maize Chlorotic Mottle Virus (MCMV) in Zambia; (2) some of the agro cultural practices conducted by farmers which may predispose them to the attack of MLN in the study area in an event of an outbreak; (3) the presence or absence of insect vectors and weeds known to be hosts of MLND causing viruses.

MATERIALS AND METHODS

Study Location

The surveys were conducted from 2014/2015 to 2015/2016 cropping seasons and covered the following provinces: Copperbelt, Muchinga Northern, Lusaka, Luapula, North-Western, Southern, Central and Eastern. Areas along the border between Zambia, Tanzania and DR Congo covering the Copperbelt, Muchinga and Northern Provinces were targeted in 2014/2015. The survey was later extended to include other provinces covering Lusaka, Luapula, North-Western, Southern, Central and Eastern Provinces in the 2015/2016 cropping season (Fig. 1).

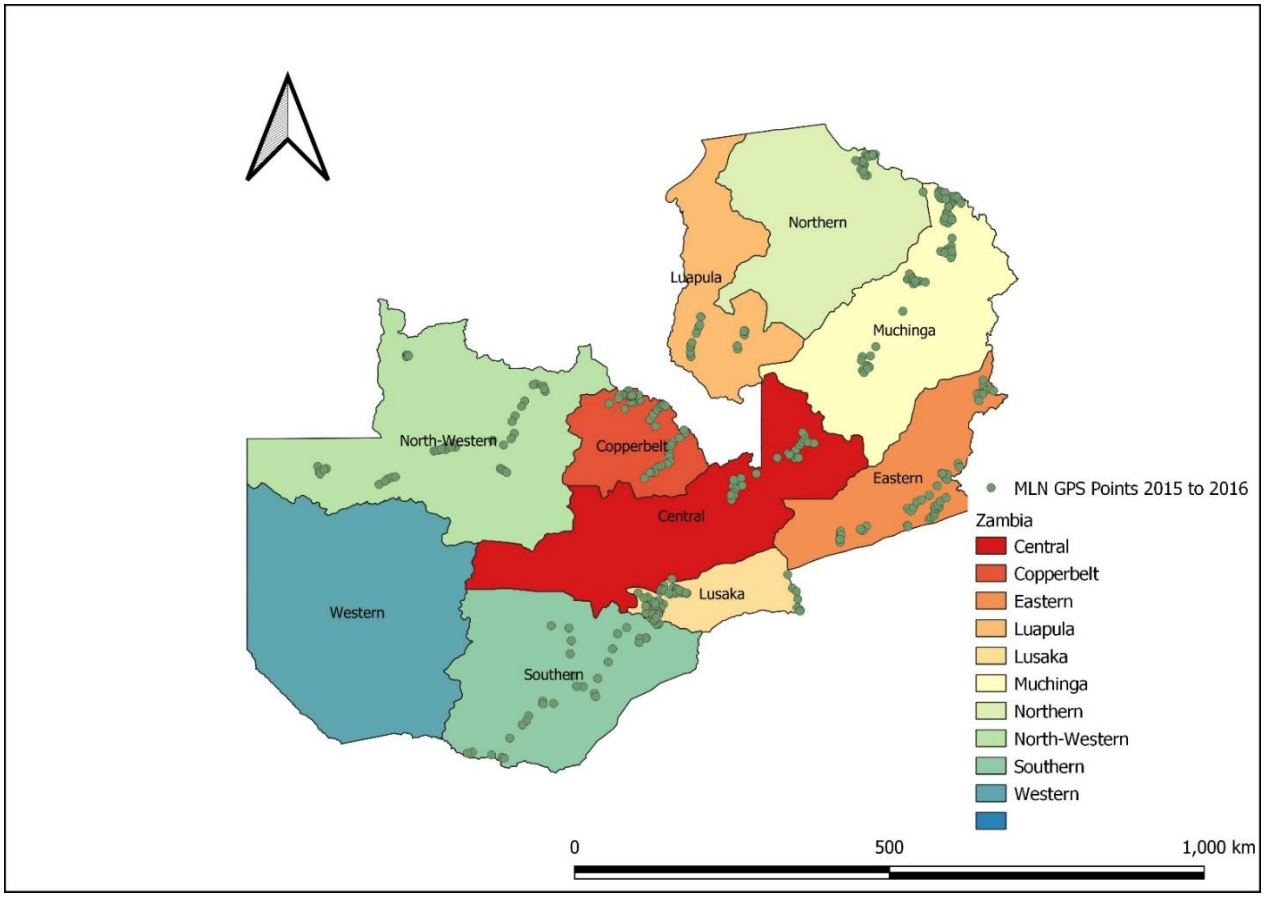

Figure 1: Map of Zambia showing areas surveyed.

2.2 Data collection using questionnaires

The questionnaires used in the survey were designed by the International Maize and Wheat Improvement Center (CIMMYT) and adopted to be utilized during the survey. The information collected included: name and sex of farmer, location, type and source of seed, planting date, field size, variety, stage of crop, crop rotation history, frequency of visits from extension agents, pesticides use, type of insects, type of diseases, symptoms and weeds. The global positioning system (GPS) latitude, longitude and elevation points were also recorded to help with the production of maps.

The agro cultural practices were collected by means of questionnaires. Farmers were asked to whether they practiced crop rotation, planted local varieties; and whether they used recycled seed among other questions. The farmer's responses were recorded on questionnaires. The information on agro cultural practices was collected because, in countries were MLN has

been reported, research has shown that crop rotation, use of certified seed are among interventions being used for disease management. Therefore, such information would be used by policy makers and extension staff to intensify the awareness on good agricultural practices that help to prevent the disease. Similarly, insects and weeds such as thrips, aphids and beetles among others have been reported to be vectors that aid the transmission of MLN causing viruses. Consequently, farmers would be advised on the control methods for such pests.

2.3 Field leaf sampling

Maize fields were inspected and sampled for detection of MCMV between January and March of each cropping season. The plants sampled were of varying growth stages ranging from flowering to the dough stages. Samples were picked at an interval of every five to ten (5-10) km distances between maize fields depending on the availability of maize fields in a particular area.

2.4 Sample collection

The survey team followed the X pattern (Fig. 2) to sample the field crops in order to maximize coverage and to have a thorough examination of the field (Muliokela, 1995). A total of six plants were randomly selected and inspected for the identification of MLN virus symptoms along each path within the X pattern (Suresh and Mezzalama, 2016).

A total of six flag leaf samples per field were cut out using scissors previously disinfected with bleach targeting both the symptomatic and non-symptomatic plants for general virus related symptoms. Depending on the size of the field the sampling was done as follows: for field of 5 ha one X pattern, 5 to 20 ha two X pattern and over 20 ha six or more. According to the CIMMYT MCMV detection protocol, it is recommended to test using flag leaves because these leaves are more succulent than the rest of the leaves, and more importantly the immunostrips are sensitive enough to detect the virus during the surveillance. Tissue paper towels were used to hold the leaves when cutting in order to avoid contamination during sampling. Each of the six individual wrapped leaves were placed in individual paper bags with uniquely identified barcodes placed on each sample. In order to avoid

contamination, the scissors were disinfected in between fields.

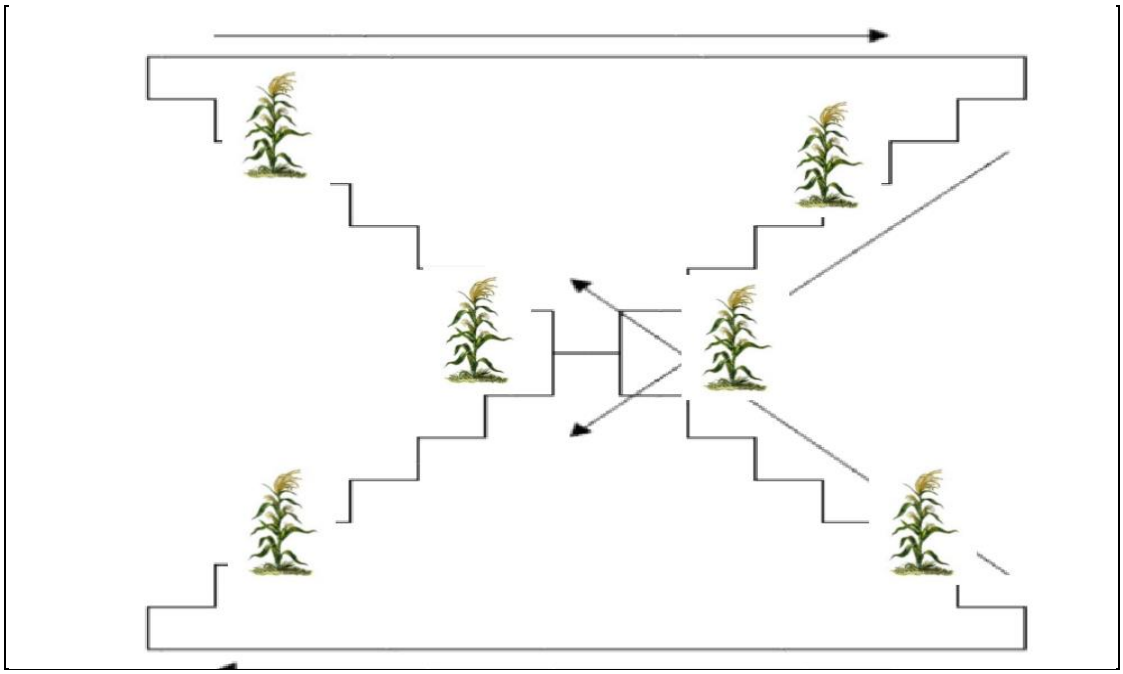

Figure 2: Schematic diagram on how sampling of plants was done.

Diagnostic procedure: Testing the leaves using the Immunostrips

With clean gloves, coin size leaf pieces were cut off from each of the six leaf samples collected and placed in a small size zip lock bags containing 4 mls of extraction buffer before crashing the leaves. One drop of extracted plant sap using a pipette was collected and placed in another small clean vial with three drops of extraction buffer added and mixed thoroughly. Thereafter, MCMV immunostrips was inserted into a test bag on its end marked sample and left for 10 to 15 minutes before reading the results.

Vector Pest Survey

General vector pest surveillance was conducted in order to check for presence or absence of all vectors associated with MLND such as aphids, thrips and stem borers without determining the numbers occurring per host. Pests found were noted and samples submitted to the entomology laboratory for identification.

Surveillance of Maize Aphid

The upper leaves were examined from each plant and a total of 50 plants were

sampled per field. Both winged and wingless aphids were collected and preserved in 70% ethyl alcohol and later submitted to the Entomology Laboratory for identification.

Surveillance of Maize Thrips

Observations for the presence of thrips was done along the path of the X pattern on all the internal plant parts, as well as in sheaths, under cob husks, on silk, between kernels, and in tassels, including individual spikelet. Plants were cut down, packed into sealed plastic bags and transported to the laboratory, where they were inspected under a microscope. All the collected thrips were preserved in 70% alcohol and later submitted to the Entomology Laboratory for identification.

Surveillance of Maize Stem Borer

The field was scanned for the presence of the stem borers. All the infested plants were dissected and the larvae found where collected. The collected larvae were preserved in 70% alcohol and later submitted to the Entomology Laboratory for identification.

Surveillance of weeds

The field was checked for the presence of weeds occurring using the weed identification book. Weed samples were collected and matched in line with the descriptors outlined in the weed pocket book and identified accordingly. This was done in order to check for the presence of weeds reported as hosts for MLN causing viruses

Results

Table 1 below indicates results for a total number of 419 samples obtained from the surveyed provinces of Zambia. Sampling sites per province ranged from 31 to 76. Muchinga Province had the highest number of samples with 76 while southern province had the lowest. The provinces such as Lusaka, Muchinga and Copperbelt had higher samples tested because they relatively have high interactions with countries outside Zambia. Muchinga and Copperbelt share borders with Tanzania and the Democratic Republic Congo, respectively while Lusaka is the hub for all grain and seed import and export activities. All samples collected and tested for MCMV were negative.

Table 1: Table of results for rapid diagnostic tests during surveillance for 2014 to 2016.

SN	Province	No of fields surveyed	Bulk AgriStrip Result MCMV (+/-)
1	Eastern	51	Negative
2	Central	33	Negative
3	Southern	31	Negative
4	Lusaka	73	Negative
5	Muchinga	76	Negative
6	Copperbelt	69	Negative
8	North -western	41	Negative
9	Northern & Luapula	45	Negative
Total		**419**	

All samples showed only one red line, which implied no presence of MLND in all the samples tested.

Information obtained from the questionnaires.

Results obtained from the questionnaires regarding farmers not implementing the agro-cultural practices are shown in figure 3 below. Two hundred and twelve farmers were planting local varieties while 164 farmers were not practicing crop rotation with 85 using recycled seed.

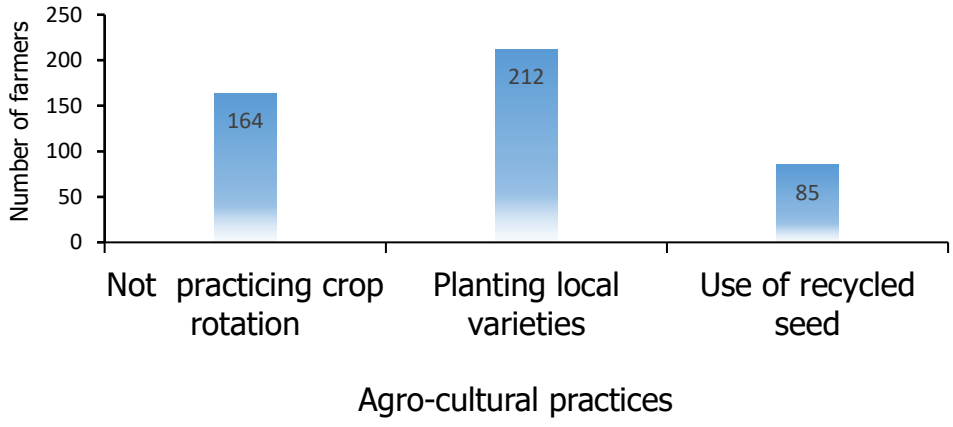

Figure 3: Number of farmers not implementing some agro cultural practices

Observed pests in farmer's fields

The insect pests that are known to be associated with MLND were observed in the farmer's maize fields. Those observed were as follows: Aphids, thrips, stalk borers and beetles, and the weeds that may act as reservoir for the virus: Bermudagrass *(Cynodon dactylon)* and Napier grass (*Pennisetum purpureum).* In several cases maize plants in the fields had more than one type insect occurring as shown in Figure 4. The most common insect type present on plants were stalk borers followed by the combination of stalk borers and aphids. However, many farmers' fields (200 out of the 419) surveyed fields had no insects observed on plants. Generally, aphids and beetles occurred on plants which were from vegetative to tasseling stages while stalk borers and thrips were mostly observed on physiologically matured ears and stems.

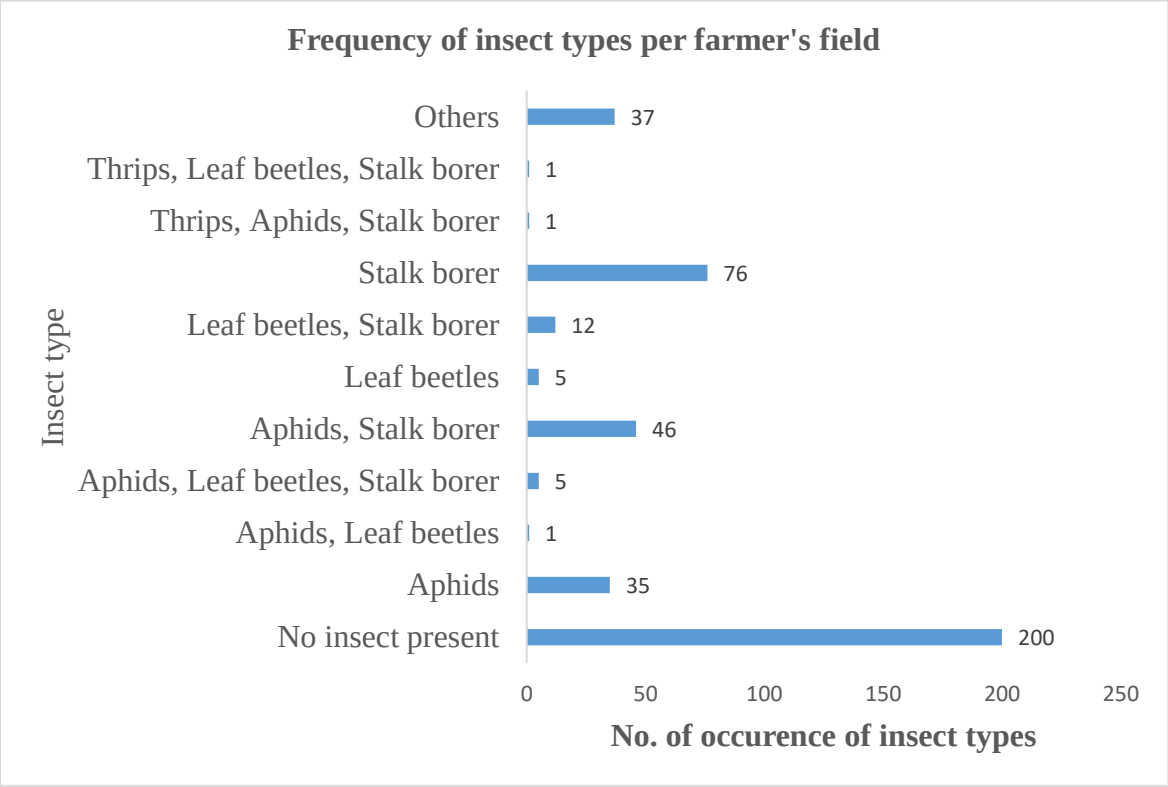

Figure 4: Frequency of insect types on maize plants in farmers' fields

Discussion

The survey findings revealed that the *Maize Chlorotic Mottle Virus* (MCMV) was absent. This implied that all the surveyed areas in Zambia were free from Maize Lethal Necrosis (MLN). It is important for Zambia to maintain this status if it is to continue as the hub of seed and grain exports to the neighboring countries. According to data on trade by ACTESA (2015) and CSO/MAL (2015) Crop Forecast Survey respectively, Zambia has been leading in the export of seed and it has been performing well with regards to seed and grain production.

Since the first report in Kenya, in 2011, the MLN has spread tremendously fast to other parts of Africa as well. The detection of MCMV and MLN in DRC and Tanzania the closest neighbors and trading partners for Zambia is of Concern. According to Lukanda (2014) MLND was detected in Kivu province in DRC in 2013 and the following year (2014), it was detected on maize affecting both the local and hybrid varieties in two provinces of Tanzania (Bini and Lubero) in and also in the northern part in Arusha. The fact that in the same period of 2014, MLN was detected in two countries confirms its fast geographical spread. This confirms the expressed concern in the report made by Isabirye and Rwomushana (2016) that MLN has the potential to spread and devastate maize production in Africa at a very fast rate. The predictive model on MLN spread shows that countries with the semi-arid and sub humid tropical type of climate in Central and Eastern Africa such as Ethiopia, Tanzania, and DRC were at risk of losing 662, 924 Km^2, 625, 690 km^2 and 615,940 Km^2 potential land of maize production respectively. From this information, Tanzania and DRC being Zambia's immediate neighboring countries implies, that the risk of MLN introduction to Zambia is very high. This state of affairs demands for strengthening surveillance and putting in place other measures like creating awareness to prevent further spread (Lukanda, 2014).

The practice of growing maize on a yearly basis predisposes farmers to incidences of crop diseases including

MLN. Planting the same crop on the same piece of land encourages the buildup of diseases and insect pests. Crop rotation, soil tillage, fertilization, liming and irrigation are among the agronomic practices that play an important role in preventing or reducing the risk of diseases (Heitefuss, 1989). Additionally, Mahuku *et al*. (2015) claims that some research conducted in USA and Kenya showed that interventions such as the insect vector control, crop rotation, and crop diversification are among the agronomic practices that play an important role in preventing or reducing the risk of MLND. Further, in Kenya, effective monitoring, rigorous implementation of maize-free periods and rotation with non-cereal crops have helped in minimizing MLND incidence.

Furthermore, despite the availability of many certified varieties on the market, the study revealed that 50% of farmers still planted local varieties and a further 20% used recycled seed as presented in Table 2. This type of seed in most cases results in poor yields and is highly susceptible to diseases. Mahuku *et al*. (2015) suggests that the use of resistant hybrids and cultivars, in combination with improved agronomic practices is likely to be the best solution in the long run.

Weeds such as Bermudagrass *(Cynodon dactylon)* Napier grass (*Pennisetum purpureum)* were detected in the surveyed fields as outlined in section 3.2. Mahuku *et al*. (2015) claims that these weeds are among the hosts of MLN causing viruses. Bockelman (1982) recommends that uncontrolled weeds that serve as hosts to the viruses causing MLN could act as reservoirs for the virus infection to the crops. For this reason, farmers should keep their fields weed free.

Similarly, some pests known to be vectors for MLN causing viruses such as stalkborers, aphids, thrips, and beetles were observed in the field during the survey. These pests are of concern even though seed transmission for both MCMV as well as SCMV as reported by (Wangai *et al*., 2012) is known to take place at very low rates. However, the presence of these vectors if not

controlled can spread the disease very fast resulting in epidemics (Mahuku *et al.*, 2015).

Conclusion

Findings from this study clearly indicate the absence of MLND in all the 419 fields of the provinces surveyed, which suggests that MLND is not present in Zambia. The revelation of the study that some of the farmers were inclined to certain agro-cultural practices that could encourage the spread and buildup of diseases in the fields needs redress as it might increase the risk for introduction of MLND.

Recommendation

There is need to strengthen extension services to enable farmers adopt good agronomic practices that help to prevent the spread of MLND such as crop rotation, crop diversification and controlling weeds and insects. The Government needs to continue to be proactive in conducting awareness and training on MLND. Further, the NPPO needs to put in more stringent phytosanitary measures to prevent MLND introduction and spread into Zambia by: continuing with conducting detection surveys; revision of maize phytosanitary import conditions; development and review of Standard Operating Procedures (SOPs); stakeholder consultations and sensitizations on strategies to prevent MLN and development of emergency response plan.

Acknowledgements

The surveillance was made possible through the financial support provided by the Agricultural Productivity Program for Southern Africa (APPSA) and the International Maize and Wheat Improvement Center (CIMMYT). NPPO staff, ZARI management and Department of Agriculture extension staff who were involved in the surveillance are also appreciated. We further thank the Farmers for their corporation during the survey.

References

Achon, M. A., Serrano, L., Clemente-Orta, G. & Sossai, S. (2017). First Report of Maize chlorotic mottle virus on a Perennial Host, Sorghum halepense, and Maize in Spain. *Plant Disease*. 101(2), pp. 393-393.

ACTESA, (2015). Eastern and Southern Africa Agricultural Trade Information Portal: Cross Border Trade Volumes Lusaka: ACTESA.

Bockelman, D. L., Claflin, L. E. & Uyemoto, J. K. (1982). Host range and seed transmission studies of Maize chlorotic mottle virus in grasses and corn. *Plant Disease*. 66, pp. 216– 218.

Boddupalli, P., Suresh, L. M., Mwatuni, F., Beyene, Y., Makumbi, D., Gowda, M., ... & Molnar, T. (2020). Maize lethal necrosis (MLN): Efforts toward containing the spread and impact of a devastating transboundary disease in sub-Saharan Africa. *Virus Research*, 197943.

CSO/MAL. (2015). Crop Forecast Survey. Lusaka, Zambia: CSO/MAL.

Chiona, S., Kalinda, T. & Tembo, G. (2014). Stochastic frontier analysis of the technical efficiency of smallholder maize farmers in Central Province, Zambia. *Journal of Agricultural Science*. 6(10), pp.108-118.

Chapoto, A., Govereh, J., Haggblade, S. & Jayne, T.S. (2010). Staple food prices in Zambia (No. 1093-2016-87954).

De Groote, H., Oloo, F., Tongruksawattana, S. & Das, B. (2016). Community-survey based assessment of the geographic distribution and impact of maize lethal necrosis (MLN) disease in Kenya. *Crop Protection*. 82, pp.30-35.

Heitefuss R. (1989). Crop and Plant Protection: The Practical Foundations. *Chichester, UK: Ellis Horwood Limited.*

Isabirye, B. E & Rwomushana, I. (2016). Current and future

potential distribution of maize chlorotic mottle virus and risk of maize lethal necrosis disease in Africa. *Journal of Crop Protection*. 5(2), pp. 215-228.

Jensen, S.G., Wysong, D.S., Ball, E.M., & Higley, P.M. (1991). Seed transmission of maize chlorotic mottle virus. *Plant Dissease*. 75, pp. 497–498.

Liu, Z., Xia, X., Yang, C. & Wang, L. (2015). Visual detection of Maize chlorotic mottle virus using unmodified gold nanoparticles. *RSC Advances*. 5(122), pp.100891-100897.

Lukanda, M., Owati, A., Ogunsanya, P., Valimunzigha, K., Katsongo, K., Ndemere, H. & Kumar, P.L. (2014). First report of Maize chlorotic mottle virus infecting maize in the Democratic Republic of the Congo. *Plant Disease*. 98(10), pp.1448-1448.

Mahuku, G., Lockhart, B.E., Wanjala, B., Jones, M.W., Kimunye, J.N., Stewart, L.R., Cassone, B.J., Sevgan, S., Nyasani, J.O., Kusia, E. & Kumar, P.L., (2015). Maize Lethal Necrosis (MLN), an Emerging Threat to Maize-Based Food Security in Sub-Saharan Africa. *Phytopathology*. 105(7), pp.956-965.

Muliokela, W.S. (1995). Zambia Seed Book Technology HandBook, Ministry of Agriculture, Food and Fisheries.

Oerke, E.C. (2006). Crop losses to pests. *The Journal of Agricultural Science*. 144(1), pp. 31-43.

Suresh, L.M. & Mezzalama, M. (2016). 'Sampling protocol for MLN and Introduction to Immunostrips for MLN pathogen detection' Workshop on Survey and Sampling for MLN Pathogens; March 3-4, 2016; CIMMYT, Harare.

Shiferaw B., Prasanna B. M., Hellin J. and Banziger M. (2011). Crops that feed the World 6. Past successes and furture challenges to the role played by maize in global food security.*Food Science.* 3, pp. 307 - 327.

Wangai, A.W., Redinbaugh, M.G., Kinyua, Z.M., Miano, D.W., Leley, P.K., Kasina, M., Mahuku, G., Scheets, K. & Jeffers, D. (2012). First report of maize chlorotic mottle virus and maize lethal necrosis in Kenya. *Plant Disease*. 96 (10), pp. 1582-1586.

The Effect of Seed Source and Post-harvest Practices on Quality of Soybean (*Glycine max*) Seeds in Busia County

Ochran, M. K.,[1] Narla, R. D.,[1] Muthomi J. W[1]. and Ochuodho J. O.[2]

[1]University of Nairobi P.O. Box 30197, GPO. Nairobi, Kenya

[2]University of Eldoret P. O. Box 1125-30100, Eldoret, Kenya

Corresponding Author: ochran.mutai@gmail.com

Abstract

Majority of farmers growing legumes use and recycle seeds from informal sources for the next crop. The quality of such seeds is unknown and usually leads to accumulation of pest, diseases and reduced yields. This study was carried out to determine production practices and quality of soybean seeds obtained from informal sources in Busia County. A survey was conducted to collect information on source of soybean seeds, production and post-harvest handling practices. Seed samples were collected from farmers, local market and agro-dealers. The seeds were evaluated for purity, seed coat damage, germination, vigour and fungal infection. Majority (48%) of farmers in Busia County used farm saved seeds, 29% and 23% used seeds from community based organizations and local markets, respectively. Most of the farmers (92%) used inappropriate threshing techniques like beating with sticks and about 80% of the farmers did not treat seed either before storage or during planting. Majority (68%) stored soybean for three months only. Seeds from informal sources had low purity, higher seed coat damage and infection as compared to certified seeds. The physical purity of seeds from the informal sources did not meet the recommended standard of 98% however their germination was comparable to 75% germination standard. Farmers therefore, should be advised to adopt use of certified seeds and appropriate handling techniques.

Keywords: Soybean, seed source, seed quality, seed production practices

Introduction

In Africa, seed that is used is supplied by formal and informal seed sector. According to Adetumbi *et al*. (2011) 100% of the seed dealers handle maize, 82% for cowpea and 82% vegetables compared to 27% for soybean seeds indicating a limited supply of soybean

seeds compared to other grain crops and vegetables. The informal sector provides the bulk of seed planted by farmers in developing countries (Bishaw, 2004). It operates at the farm level and depends on local knowledge of plant or seed selection and management practices. It does not involve seed certification procedures and although the role of the informal sector is recognized, few attempts have been made to assess the status of seed quality. Seed from the formal sector must meet specific quality standards prescribed by the national regulations and involve certification agency which establishes technical, administrative and regulatory frameworks to produce quality seed that meets specified minimum standards for marketing. Apart from good crop management practices to maintain varietal purity, laboratory tests are conducted to assess critical seed quality attributes. Seeds carry genetic potential of plants and influence the productivity of other agricultural inputs. Availability of, access to and use of quality seeds are determinant of the efficiency and productivity of other technologies in increasing crop productivity.

Soybean (*Glycine max*) is an important multipurpose crop utilized for food, livestock feed, industrial raw material and bio-energy (Myaka *et al*., 2005). It is also the world leading source of oil and protein (Fedaku *et al*., 2009) with 20% oil content, 40% protein and 35% of carbohydrates and is cholesterol free with low levels of saturated fatty acids. The biomass from soybean is an important source of animal feed, green manure and can also be used as mulch (Chianu *et al*., 2009). In Kenya soybean is being promoted as a cultural source of protein, cooking oil, income to farmers and for soil fertility improvement (Misiko *et al*., 2008). Sanginga *et al.* (2003) estimated that soybean can fix 44-103 kg/ha of nitrogen reducing the need for expensive nitrogen fertilizers. It adds nitrogen to the soil enriching infertile soils and stimulating crops productions in rotation especially with cereals (Ojiem, 2006).

Western region is the leading soybean production area in Kenya accounting for

80% of the total national soybean production with the main production areas being Butere/Mumias, Bungoma, Busia, Teso, Kakamega, Mt. Elgon, Lugari and Vihiga (Chianu *et al.*, 2008). However, production is still below their maximum potential due to some challenges facing the farmers. Lack of adequate and quality seed supply by the formal system and lack of knowledge on production and post-harvest practices of soybeans also hindered farmers from accessing improved quality seeds (Oshone *et al.*, 2014). This has led to about 70% of the farmers recycling seeds from informal sources and because seeds from such sources are of poor quality they result in poor yields. A baseline survey conducted by Odendo *et al.* (2008) revealed that communities in this region had interest in growing soybeans but had no access to improved varieties, good quality seeds and resorted to using seeds obtained from informal sources to raise crops in the following season. This study therefore aimed at determining the effect of seed source, production and post-harvest handling on quality of soybean seeds in Busia County.

Materials and Methods

A purposeful survey was conducted using a semi-structured questionnaire to collect information on source of soybean seeds, production and post-harvest handling practices. Seed samples were collected from farmers, local market and agro-dealers in Low Midland Zones I, II and IV. The seeds were evaluated for purity, seed coat damage, moisture content, germination, vigour and infection in the laboratory.

Seed quality tests

Determination of physical purity of soybean seeds

Analytical purity was conducted in accordance to ISTA 2015 guidelines. The different components comprising the sample were grouped into, pure seed, inert matter, other crop seeds and weed seed. Percentage of each component was calculated as a fraction of the initial weight as indicated below:

$$\text{Component (\%)} = \frac{\text{Weight of each component fraction X 100}}{\text{Initial weight of the sample}}$$

Determination of moisture content and seed coat damage

Moisture content was determined using a moisture meter by filling the meter cup with soybean seeds and recording the readings. The test was repeated four times. Seed coat damage was detected by sodium hypochlorite test as per Van Utrecht *et al*. (2000). Four replicates of 100 seeds were soaked in 1% sodium hypochlorite solution for 10 minutes. Seeds were considered to be damaged when the seed coat appeared wrinkled, swollen or with loose coats. Damaged seeds were counted and the percentage estimate of seed coat damage of a sample calculated using the formula:

$$\textbf{Damage}(\%) = \frac{(\text{No. of swollen seeds after test}) \text{ x}100}{\text{Total number of seeds used}}$$

Determination of germination and seedling vigour of soybean

Germination test was done according to ISTA 2015 guidelines on paper towel to determine the percentage of viable seeds in a sample. Seeds were surface sterilized in 2% sodium hypochlorite solution for five minutes to kill epiphytes followed by three changes of sterile distilled water. Four replications of 100 seeds each in transparent plastic boxes lined with absorbent towel. The boxes were arranged in a completely randomized design (CRD) in the laboratory under room temperature conditions. The seed were routinely misted with sterile distilled water. Seeds were considered germinated when 2mm of the radicals protruded and germination percentage calculated as shown below (Chirchir *et al.,* 2016).

$$\textbf{Germination}\ (\%) = \frac{(\text{Number of germinated seeds}) \text{ x}100}{\text{Total number of seeds}}$$

Germination rate index was also calculated as shown below:

$$\textbf{GRI} = \frac{\text{No. germ. seeds 1st}}{\text{Days of 1st count}} + \frac{\text{Germinated final count}}{\text{Days of final count}}$$

Seedling vigor was determined after 15 days by randomly selecting 10 seedlings from each replicate and measuring the root and shoot lengths using a ruler in centimeters. These were then used to calculate the seedling vigor index (SVI) using the formula described by Aliloo and Darabinejad, (2013).

$$\textbf{SVI} = \frac{\text{Germination \% x Seedling length}}{100}$$

Determination of fungal infection in soybean seeds using agar plate method

Soybean seeds were surface sterilized in 2% sodium hypochlorite for three minutes, followed by rinsing in three changes of sterile distilled water and blot dried on sterile paper towel. Four replications of 10 seeds per petri dish were plated on Petri dish containing potato dextrose agar media and incubated for 3-7 days at 25^0c in alternating dark and light conditions (Alemu, 2014). Sodium chloride was added to the media to inhibit germination and streptomycin sulphate to inhibit bacterial growth. The number of seeds infected and the individual pathogen types were recorded and results expressed as a percentage. Fungi growing on potato dextrose agar plates were identified both visually and under the stereomicroscope by observing colony characters and morphology of sporulating fungi (Shovan *et al*., 2008).

Data analysis

Survey data was analyzed using IBM Statistical Package for Social Science (SPSS) version 20. Laboratory tests data were subjected to analysis of variance (ANOVA) using GENSTAT 15^{th} edition. Means were separated using Fischer's protected Least Significant Difference (LSD) at 5% level of significance (Steel and Torrie, 1960).

Results

Source of soybean seeds and post-harvest practices used by farmers in Busia County

Most (48%) of the farmers used farm saved seeds, 29% and 23% were using seeds obtained from community based organizations and local markets respectively, across the three zones. Over (60%) of farmers in LM II used own saved seeds as compared to other zones while around 45 % sourced from community based organizations within their locality (Table 1).

Table 1: Percentage of farmers who obtained soybean seeds from different sources in three agro-ecological zones in Busia County.

n=60	Agro-Ecological Zones			
Source of seed	LM I (%)	LM II (%)	LM IV (%)	Mean
Farm saved	40±8.3	65±8.3	40±8.3	48.3±7.6
Local market	30±4.4	25±4.4	15±4.4	23.3±7.6
Community based organizations	30±10.1	10±10.1	45±10.1	28.3±7.6

N=Sample size, LM I= Lower Midland Zone I, LM II= Lower Midland Zone II, LM IV = Lower Midland Zone IV

About 92% of farmers threshed soybean crop by beating with sticks, 5% used their hands to remove seeds from the pods while 3% put soybean in a sack and beat them with sticks. All the farmers in LM I threshed soybean using sticks. Threshing inside the sack with sticks attracted only 5% of the farmers in LM II and IV. Hand threshing was practiced by 15% of the farmers in LM IV (Table 2).

Table 2: Percent farmers using different techniques to thresh soybean in three agro-ecological zones in Busia County.

n=60	**Agro-Ecological Zones**			
Threshing method	**LM I (%)**	**LM II (%)**	**LM IV (%)**	**Mean**
Removing seeds from the pods by hand	0±5.0	0±5.0	15±5.0	5.0±29.2
Beating with stick on the floor	100±6.0	95±6.0	80±6.0	91.7±29.2
Beating pods in a sack with sticks	0±1.7	5±1.7	5±1.7	3.3±29.2

N=Sample size, LM I= Lower Midland Zone I, LM II= Lower Midland Zone II, LM IV = Lower Midland Zone IV

About 67% of the farmers in Busia County did not treat soybean seeds. Around 60% of the farmers in LM I adopted seed treatment technology practice. In LM II and IV more than 70% of farmers did not treat the seeds either before storage or planting (Figure 1).

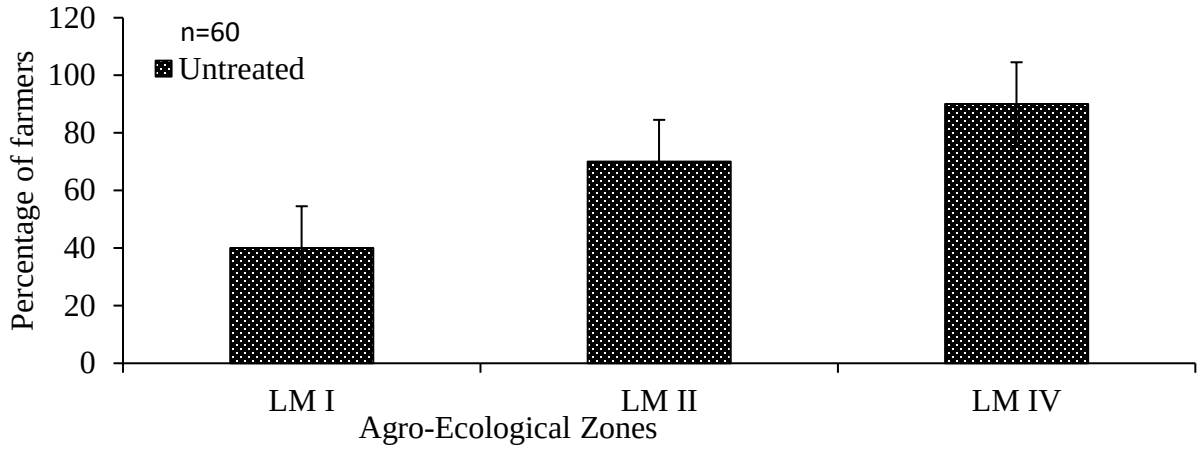

Figure 1: Percentage of farmers who did not treat soybean seeds before storage or planting in three zones Busia County.

Farmers stored soybean seeds up to a period of twelve months. However, majority (68%) reported to have stored seeds for three months. Only 5% as the smallest group stored their seeds for one year. Around 8% stored for one month, 7% stored for two months and about 12% of farmers stored for six months which was equivalent to two growing season. Farmers stored seeds for only two to six months period in LM II. While in zone IV seeds were stored for twelve months (Table 3)

Table 3: Percentage of farmers who reported the duration of storage of soybean seeds in three agro-ecological zones in Busia County.

n=60	Agro-Ecological Zones			
Duration of storage	LM I (%)	LM II (%)	LM IV (%)	Mean
Up to 1 month	10±4.4	0±4.4	15±4.4	8.3±12.1
1 to 2 months	5±1.7	10±1.7	5±1.7	6.7±12.1
2 to 3 months	75±9.3	80±9.3	50±9.3	68.3±12.1
3 to 6 months	10±1.7	10±1.7	15±1.7	11.7±12.1
Over 12 months	0±5.0	0±5.0	15±5.0	5.0±12.1

N=Sample size, LM I= Lower Midland Zone I, LM II= Lower Midland Zone II, LM IV = Lower Midland Zone IV

Quality status of soybean seeds used in Busia County

The analytical purity components of the seed samples from different sources differed significantly. Seeds obtained from agro-dealers had the highest percentage of pure seed followed by own saved seeds and lastly by seeds from the local market. However, all the seeds from the three sources did not meet the recommended physical purity standard of 98% (Table 4). The analysis indicated that seeds from the informal sources had damaged seed coats of 88% as compared to seed from agro-dealer (82%).

Table 4: Analytical purity of soybean seed samples from various sources in different agro-ecological zones in Busia County.

	Components		
Source	**Pure seed**	**Other crop seed**	**Inert matter**
Agro-dealer	97.2a	0.2b	0.1c
Farm-saved	92.5bc	0.3a	1.0a
Market	91.1c	0.4a	0.6b
Mean	93.6	0.3	0.6
LSD($P \leq 0.05$)	2.1	0.1	0.3
CV	5.0	118.8	120.8

LSD=least significance difference, CV=coefficient of variation.

Moisture content of seeds from the three sources did not differ significantly. Comparable results were recorded on damage caused by insects in seeds collected from the informal outlets and high compared to seed from formal source (Table 5).

Table 5: Percent moisture content, seed coat damage and insect damage of soybean seed samples collected from different sources in the three agro-ecological zones in Busia County.

Source	**Moisture content**	**Seed coat damage**	**Insect damage**
Agro-dealer	9.1a	81.8b	0.5b
Farm-saved	9.1a	88.4a	1.6a
Market	9.2a	87.7a	1.6a
Mean	9.1	86.0	1.2
LSD($P \leq 0.05$)	0.3	3.0	0.3
CV	7.5	8.0	83.2

LSD=least significance difference, CV=coefficient of variation

Germination percentage of soybean seeds from different sources differed significantly. Germination percentage of the seeds from the three sources met the minimum germination standard recommended for soybean seeds of 75%. Agro-dealer seeds recorded the highest germination percentage of 90% followed by local market (76%) and farm saved seeds (75%) respectively. High germination rate, number of normal seedlings and seedling vigour index was also observed on seeds from this source (Table 6).

Table 6: Percent germination and seedling vigour of soybean seed samples from different sources in three agro-ecological zones in Busia County.

	Germination		Seedlings		
Source	**Germination%**	**Germination rate**	**Normal**	**Seedling Length**	**Seedling vigour index**
Agro-dealer	90.0a	30.6a	82.5a	8.4a	7.6a
Farm-saved	75.2b	23.3b	69.0b	5.5c	4.8c
Market	75.9b	23.3b	66.5b	6.8b	5.7b
Mean	80.4	25.7	72.7	6.9	6.0
LSD($P \leq 0.05$)	7.7	3.4	7.6	1.4	1.4
CV	21.8	30.1	23.8	45.7	52.1

LSD=least significance difference, CV=coefficient of variation.

Infection of seeds collected from different sources and zones differed significantly. High number of seed infection was observed on seeds obtained from farmers and local markets. Infection by *Cercospora kikuchii and Penicillium spp.* were observed on seeds collected from local market. *Aspergillus falvus and Aspergillus niger* were the most prevalent in seed obtained from the farmers. Seeds collected from the agro-dealer outlets had the least incidence of fungi observed (Table 7).

Table 7: Percentage of fungi in soybean seed samples from various sources in different agro-ecological zones in Busia County.

	Fungi				
Source	**Infected**	***C. kikuchii***	***A. flavus***	***A. niger***	***Penicillium spp.***
Agro-dealer	10.0b	0.0b	0.0c	0.0b	0.0b
Farm-saved	28.6a	1.1ab	7.8a	5.2a	2.3a
Market	23.2a	2.2a	3.8b	2.0b	2.8a
Mean	20.6	1.1	3.9	2.4	1.7
LSD($P \leq 0.05$)	9.6	2.2	4.7	4.7	2.8

LSD=least significance difference, C-Cercospora, A-Aspergillus, spp.-species.

Discussion

Most of the farmers in Busia County utilized soybean seeds from the informal sources with majority of them using farm saved seeds usually obtained from the previous season. Most of the farmers have opted to save seeds to replant in the following season due to financial constraints, inadequate seeds of good quality and high cost of certified seeds. Informal seed sources readily availed the seed to farmers every season since most of them save up for the next planting. Findings reported by Oshone *et al*. (2014) similarly indicated that majority of the farmers in Ethiopia obtained common bean seed from informal sources a channel which contributed more than 95% of the common bean seed supply in Ethiopia. Similarly, Chianu *et al*. (2009) conducted a survey on soybeans in Kenya and revealed that most farmers used saved seeds or seeds sourced from the open air markets which at times were of mixed varieties to raise crops the following season due to their availability.

The formal seed source supplied inadequate seed and this was further confirmed with similar findings by Anon (2001) which revealed that the formal sector supplied only 4% of seeds sown by farmers and the remaining 96% was supplied by the informal sources in most African countries. Similarly, Adetumbi *et al.* (2011) reported that 100% of the seed industries which is a representation of formal sector handled maize, 82% for cowpea and 82% vegetables compared to 27% for soybean seeds. A baseline survey conducted by Odendo *et al*. (2008) revealed that most farmers in Western region of Kenya had interest in growing soybean but had a challenge in accessing improved varieties and good quality seed during planting. In addition, lack of adequate and quality seed supply by the formal system hindered farmers from accessing improved quality seeds (Oshone *et al*., 2014). The few seed industries in Kenya do not produce adequate seeds of soybean to meet the demand and most of them have given priority to cereals or high value crops for them to make profits (Lowaars *et al*., 2012).

Majority of the farmers interviewed reported to thresh soybean with sticks and as a result farmers experienced a challenge of reduced germination and vigor in soybeans. The results were in agreement with the findings by Surve *et al*. (2015) who reported that hand threshing practices usually used by few farmers of soybean recorded minimal seed coat damage and high germination compared to other techniques practiced by farmers such as stick beating and mechanical threshing. Similarly, Jha *et al*. (1995) found that hand threshing resulted in higher germination and less deterioration of seed than the other techniques. Hand threshing of soybean significantly increased seed yield compared with stick threshing and mechanical threshing (El-Abady *et al*., 2012). Threshing techniques like machine and stick beating produces more breaks, cracks, bruises and abrasions which results in reduced germination and vigor and increase in abnormal seedlings (Reddy *et al*., 1995).

Survey findings indicated that majority of the farmers do not treat seed during planting or before storage. Untreated seeds act as vehicles of transmitting pathogens which cause diseases in soybean seeds. These pathogens affect germination and seedling vigor and as a result lower emergence and productivity (Sinclair, 1991). Treatment provide additional assurance to crop establishment at reduced cost and allows germination of infected seeds by controlling pathogens and protecting seed from fungi (Araujo *et al*., 2005)

The survey revealed that most of the respondents stored their seeds for a maximum period of three months and the second best storage period being six months. Between 0 and 2 months was the period between harvest and the next season of planting hence few farmers reported this storage period. The period also could be after-ripening session of soybean for it to mature fully thus minimal deterioration. The observed reduction in percentage germination and vigor over time could be linked to depletion of reserved food for the embryo. This is in line with the findings of Iqbal *et al*. (2002) and

Demirkaya *et al*. 2010) that reduction of viability and vigor could be attributed to a reduction in enzyme activity within the seed. In addition, the reduction in seed quality with time could be as a result of membrane degradation (Singh and Dadlani, 2003), reduction in enzyme activity or changes in chemical composition of the cell (Verma *et al*., 2003). Similarly, Younesi and Azadi (2013) reported that an increase in duration of storage caused a decrease in the enzyme activity of sorghum seeds.

Other studies have shown a gradual decrease in germination and vigor of soybean cultivars with increasing period of storage up to six months (El-Abady *et al*., 2012) and extending storage period intensified deterioration hence low productivity of soybean (Adoba *et al*., 2016). Belesevic *et al.* (2010) found that storage conditions and durations affected germination but adversely affected seed vigor. According to Arif *et al.* (2006) seed viability decreased gradually from 64.5% to 39.2% as duration of storage increased from 2 months to 12 months. Similarly, Sadia *et al*. (2016) found that germination and seed vigor of cowpeas declined with increase in storage duration irrespective of genotypes or storage materials. In addition, more decline in germination of soybean stored under conventional condition due to variable temperature and humidity was also reported by Balesevic-Tubic *et al.* (2010). If the seeds are not dried properly high moisture content reduces seed viability by promoting fungal growth (Pradhan and Badola, 2012) which further results in reduction of germination. For oil crops such as soybeans and sunflower, increased content of fatty acid and auto oxidation of lipids during storage are the main causes of rapid deterioration of oil crops seeds (Balasevic-Tubic *et al*., 2005).

Analytical purity of seeds from different sources varied significantly but did not meet the minimum recommended purity standard of 98%. Seeds collected from agro-dealers were more pure as compared to seed from local market and farmers. Similar observations were made by Rahman *et al*. (2017) that okra seeds obtained from seed companies

were more pure followed by those from government organizations and then the farmer's seeds. Bishaw *et al*. (2012) working on barley also observed that seeds obtained from the formal sector had the highest analytical purity as compared to those collected from the farmers and local markets. Fujisaka *et al.* (1993) found that rice seeds samples that were obtained from farmers who used manual harvesting and threshing had higher analytical purity compared with those that were machine harvested. The use of non-cemented floors during threshing in the rural setting resulted in accumulation of foreign materials in farm saved seeds. Sarker *et al*. (2015) on quality of okra seeds from different sources and locations cited lack of careful attention during cleaning operations contributed to high percentage of reduced purity in farm saved seeds.

The present findings indicated that seeds obtained from the informal sources recorded higher percentage of damaged seeds compared to seeds from the formal sector. The extensive damage of seeds from the informal sector is due to poor handling and post-harvest techniques which in turn reduced seed quality. Similar findings were reported by Reddy *et al*. (1995) that seed damage occurred during threshing in soybean and resulted in shorter storage life of seeds. Costa *et al*. (2005) studies on zoning soybean crop found out that the spoilage caused by physical injury and moisture content were the main factors that contributed to reduced quality of soybean seeds. Similarly, mechanical damage was considered the most common reason for poor quality in most legumes especially when threshed at unsuitable seed moisture content (Greven *et al*., 2001). Pacheco *et al*. (2015) pointed out that physiological quality, vigor and performance of soybean seeds was highly influenced by seed coat injury. Soybean seeds obtained from informal sources had more physical injuries because the farmers had poor knowledge on how to handle them soybean seeds which have a sensitive seed coat. Minimal injuries on seeds obtained from the formal sources may be attributed to improved and planned processing technology and knowledge

of handling soybean (Araujo *et al*., 2008). Cracks on the seeds may also become entry points of microorganisms and become susceptible to insect attack which reduces the storage potential and the general quality of the seeds (Marcos Filho, 2005).

Percentage germination in seeds from the agro-dealer a formal sector was higher as compared to those from the informal sources. The high quality of seeds from the formal sector may be due to the fact that they have equipment and knowledge of handling seeds. According to Adetumbi *et al.* (2010), the formal sector is well equipped with sophisticated equipments and skills of handling seeds to ensure that the quality of seed is maintained all through. One major quality control mechanism that has been instituted in the formal seed sector is regular seed inspection at different levels from seed field to distribution channels. This has ensured that the quality of the seed is maintained from the field all the way to the hands of the final consumer, the farmer. Bishaw *et al*. (2012) also reported that percentage germination of certified seed from the formal sector was higher compared with seed obtained from other farmers, local markets and own saved seeds. Al-Faqeeh (1997) in his studies also found that certified seed had significantly higher germination in Lentils compared with seeds from other sources. Germination percentage of seeds from farmers was comparable to the minimum standard. This can be attributed to the fact that most farmers do selection of quality seeds for planting though using informal procedures which may vary from farmer to farmer. Similar results were also reported in Ethiopia where almost all samples collected from the farmersreached the minimum germination standards for wheat (Ensermu *et al*., 1998) and sorghum (Mekbib, 2008).

Seeds obtained from the local market and own saved were more infected as compared those from the agro-dealers. This is due to lack of standard procedures and regulations govern production of pathogen free seeds.

Cercospora kikuchii, *Penicillium spp.*, *Aspergillus niger and* flavus (Bhale *et al*., 2004) were recorded as the most common fungal pathogens that infected soybean in Busia County. Most pathogens that are seed-borne are difficult to detect by most farmers and may assume that seeds are healthy while they are not in reality. Our findings relate with those of Bhale *et al*. (2004); Wu *et al*. (1964) who isolated *Fusarium oxysporum*, *Aspergillus niger* and *Aspergillus flavus, Cercospora kikuchii, Penicillium sp., Phoma medicaginis, Macrophomina phaseolina Alternaria alternate* as seed-borne pathogens which are most predominant fungi in pre and post harvested soybean seeds. Presence of these fungal pathogens reduces physiological potential of soybean seeds (Galli *et al*., 2007). Singh and Agrawal (1986) reported 30% loss in seed viability and germination because of purple stain caused by *Cercospora kikuchii,* while, *Aspergillus flavus*, *Aspergillus niger*, *Fusarium spp* were responsible for seed rots and post emergence decays (Koning *et al*., 1995).

Seeds from the formal source registered low infection as compared to seed from the informal sector due to unhygienic storage conditions at the farmer's level (Utoba *et al*., 2011). Variation in fungal incidence was attributed to variations in climatic conditions during the crop cycle especially the prevailing humidity which favor the growth of the pathogens (Naqvi *et al*., 2013). The formal sector also practices seed treatment using fungicide, insecticides or combination of different chemicals which protect the seed from infections. Seed dressing protects the growing seedling for a specific period helping it escape infection (Ellis *et al*., 2011). This technique has shown significant improvement in field emergence, seed yield and reduced mycoflora association (Anuja *et al*., 2000).

Conclusion

Majority of farmers from the different agro-ecological zones in Busia obtained seeds for the following season from informal seed sources. These seeds were found to be of poor quality having

low germination and vigor, high infection incidence and physical quality that is below the ISTA recommended standard of 98%. Most of the farmers also did not treat soybean seeds and had limited knowledge on post-harvest handling and sensitivity of soybeans.

Recommendations

More training on appropriate post-harvest technologies is recommended in order to ensure high quality seeds. The use of fungicides, insecticides or combination of different chemicals which protect the seed from infections are also encouraged. Farmers should use seeds from the formal sector which are certified and of known good quality. In addition, extensive training should be done on the appropriate post-harvest handling techniques and sensitivity of soybean seeds.

Acknowledgement

My appreciation first goes to Almighty God and Nancy, Oliver, Titus and Bilha for their assistance.

References

Adetumbi, J. A., Jaime, A., Da Silva, T., & Alamu, O. (2011). Cowpea (*Vigna unguiculata* L. Walp) seed germination indices and yield as affected by length of storage. *Seed Science and Biotechnology.* 5 (1), pp. 11-14.

Odoba, A., Odiaka, N. I., Gbanguba, A. U., & Bashiru, M. (2016). Germination characteristics of twenty varieties of soybean (Glycine max (L.) Merr) stored for seven months. *Scientia Agriculturae*. 13 (3), pp. 151-155.

Alemu,K. (2014). Seed borne fungal pathogen associated with soybean (*Glycine max* L.) and their management in Jimma, Southwestern Ethiopia. *Journal of Biology, Agriculture and Healthcare*. 4 (25), pp. 14-19.

Al-Faqeeh, T. M. (1997). A survey of lentil seed quality in Jordan. University of Jordan, Amman, Jordan (Doctoral dissertation, Msc Thesis, 97p)

Aliloo, A.A. and Darabinejad, S, (2013). Evaluation of different techniques for breaking seed dormancy of *Heliotropium europaeum L.*

(Boraginanaceae). *Journal of Biological and Environmental Sciences.* 7(20), pp. 87-91.

Anna Aladjadjiyan (2010). Effect of microwave irradiation on seeds of lentils (*Lens culinaris*, med.) *Romanian Journal of Biophysics*. 20(3), pp. 213–221.

Anuja, Gupta, K. R., Aneja and Gupta, A. (2000). Field efficacy of seed dressing chemicals on seedling emergence. Seed yield and seed weight in soybean. *Seed Research.* 28(1), pp. 54-58.

Araujo, F. F., Henning, A. A. & Hungria, M. (2005). Phytohormones and antibiotics produced by *Bacillus xubtillis* and their effects on seed pathogenic fungi and on soybean root development. *World Journal of Microbiology and Biotechnology*. 21(8), pp. 1639-1645.

Araujo, R. F., Araujo, E. F., Vieira, R. F., Sofiatti, V., Zonta, J. B. and Souza, L. T. (2008). Physiological and Sanitary Quality of Mung beans subjected to post-harvest mechanical processing. *Revista Brasileira de Armazenamento*. 33(1), pp. 43-51.

Arif M., Jan M. T., Marwat, K. B., & Khan M. A. (2008). Seed priming improves emergence and yield of soybean. *Pakistan Journal of Bot*any. 40 (3), pp. 1169-1177.

Azadi, M. S., & Younesi, E. (2013). The effects of storage on germination characteristics and enzyme activity of sorghum seeds. *Journal of Stress Physiology & Biochemistry*. 9(4) pp. 289-298.

Balešević-Tubić, S., Malenčić, Đ., Tatić, M., & Miladinović, J., (2005). Influence of aging process on biochemical changes in sunflower seed. *Helia.* 28 (42), pp. 107-114.

Balesevic-Tubic, S., Tactic, M., Dordevic, V., Nikolic, Z., & Dukic, V. (2010). Seed viability of oil crops depending on storage conditions. *Helia*. 33 (52), pp. 153-160.

Bhale, M. S. (2004). Mycoflora associated with soybean seeds, detection and their management.

Journal Mycopathological Research. 42(1): 27-29.

Bishaw, Z, Struik, P.C. & Van Gastel, A.J.G., (2012). Farmers' Seed Sources and Seed Quality: Physical and Physiological Quality. *Journal of Crop Improvement*. 26(5), pp. 655-692.

Chianu, J. N., Mairura, F., Ekise, I., & Chianu, J. N. (2008). Farm input marketing in western Kenya: Challenges and opportunities. *African Journal of Agricultural Research*. 3 (3), pp. 167-173.

Chianu, J. N., Ohiokpehai, O., Vanlauwe, B., Adesina, A., De Groote, H., & Sanginga, N. (2009). Promoting a versatile but yet minor crop: Soybean in the farming systems of Kenya. *Journal of Sustainable Development in Africa.* 10 (4), pp. 324-344.

Chirchir, G. J., Mwangi, M., Nyamongo, D. O. & Gweyi-Onyango, J. P. (2016). Soybean farm-saved seed viability and vigour as influenced by agro-ecological conditions of Meru South Sub-county, Kenya. *Journal of Applied Bioscience.* 101(1), pp. 9634-9642.

Costa, N. P., Mesquita, C. M., Franca-Neto, J. B., Maurina, A. C., Krzyzanowski, F. C., Oliveira, M. C. N. & Henning, A. A. (2005). Validation of Ecological Zoning of the State of Parana for seed production cultivars of soybean. *Revista Brasileira De Sementes*. 27(1), pp. 37-44.

Demirkaya, M., Dietz, K. J., & Sivritepe, H. O. (2010). Changes in antioxidant enzymes during ageing of onion seeds. *Notulae Botanicae Horti Agrobotanici Cluj-Napoca*. *38* (1), pp. 49.

Edmeades, G., Fischer, R. A., & Byerlee, D. (2010). Can we feed the world in 2050: In *Proceedings of the New Zealand grassland association.* 72 (1), pp. 35-42.

El-Abady MI, El-Emam AAM, Seadh SE, Yousof F.I. (2012). Soybean Seed Quality as Affected by Cultivars, Threshing Methods and Storage Periods. *Research Journal of Seed Science*. 3, pp. 1-11.

Ellis, M. A., Broaders, K. D., Paul, P. A. & Darrance, A. E. (2011). Infection of soybean seed by Fusarium graminearum and effect of seed treatment on disease under controlled conditions. *Plant Disease*. 95(4), pp. 401-407.

Ensermu, R., Mwangi, W., Verkuijl, H., Hassena M. & Alemayehu, Z. (1998). Farmers'wheat seed source and seed management in Chilalo auraja, Ethiopia. Addis Ababa, Ethiopia: International Maize and Wheat Improvement Center (CIMMYT) pp. 1-36.

Fekadu, A., Wooderson, S. C., Markopoulo, K., Donaldson, C., Papadopoulos, A., & Cleare, A. J. (2009). What happens to patients with treatment-resistant depression? A systematic review of medium to long term outcome studies. *Journal of affective disorders*. 116 (1-2), pp. 4-11.

Forsberg, G., Kristensen, L., Eibel, P., Titone, P., & Haiti, W. (2003). Sensitivity of cereal seeds to short duration treatment with hot, humid air. *Journal of Plant Disease and Protection*. 110 (1): 1-16.

Fujisaka, S., Moody, K., & Ingram, K. (1993). A descriptive study of farming practices for dry seeded rain fed lowland rice in India, Indonesia, and Myanmar. *Agriculture, Ecosystems and Environment*. 45 (1-2), pp. 115-128.

Galli, J.A., Panizzi, R.C. & Vieira, R.D. (2007). Effect of *Colletotrichum dermatium* var. *truncate* and *Phomopsis sojae* in sanitary and physiological quality of soybean seeds. *Summa Phytopathologica.* 33(1), pp. 40–46.

Greven, M.M., McKenzie, B.A., Hampton, J.G., Hill, M.J. & Hill, G.D. (2001). Some factors affecting seed quality during the mechanical threshing of dwarf French bean (*Phaseolus vulgaris* L.). *Agronomy New Zealand*. 31, pp. 121-126.

International Seed Testing Association, **(**2015). International rules for seed testing. Basserdorf, Switzerland:

International Seed Testing Association.

Iqbal, N. A., Basra, S. H. M. A., & Rehman, K. (2002). Evaluation of vigour and oil quality in cottonseed during accelerated aging. *International Journal of Agricultural Biology.* 4 (3), pp. 318-322.

Jha, R. K., Saini, S. L. & Ram, C. (1996). Effect of threshing methods on yield and mechanical damage in soybean. *Crop Research*. 11, pp. 248-250

Koning, G., Hamman B., Eicker, A., Vener, H. A. & Devan (1995). First report of *Fusarium equisetti* on South African soybean cultivars. *Plant Disease*, 79 (7), pp. 754.

Loowars, N. P. & De Boef, W. S. (2012). Intergrated seed sector development in Africa: A conceptual framework for creating coherence between practices, programs and policies. Journal of crop improvement. 26(1), pp. 39-59.

Mekbib, F. (2008). Farmers'seed system of sorghum (*Sorghum bicolar* L. Moench) in the center of diversity: II. Seed quality, storage, protection and security. Journal of New Seeds, 9(3), pp. 191-211.

Marcos Filho, J. (2015). Seed vigor testing: an overview of the past, present and future perspective. *Scientia Agricola*. 72(4), pp. 363-374.

Misiko, M., Tittonell, P., Ramisch, J. J., Richards, P., & Giller, K. E. (2008). Integrating new soybean varieties for soil fertility management in smallholder systems through participatory research: Lessons from western Kenya. *Agricultural Systems*. 97 (1-2), 1-12.

Myaka, F. A. (2005). Soybean research in the eastern zone Tanzania. In *Proceedings of the First National Soybean Stakeholders Workshop.* pp. 10-11.

Naqvi, S. D. Y., Shiden, T., Merhawi, W. & Mehret, S. (2013). Identification of seed borne fungi on farmer saved sorghum (*Sorghum bicolar*

L.), pearl millet (*Pennisetum glaucum* L.) and groundnut (*Arachis hypogaea* L.) *seeds Agricultural Science Research Journal*. 3(4), pp. 107-114.

Odendo, M., & De Groote, H. (2008). Linking farmers to markets: The case of grain marketing information in Western Kenya. *African Association of Agricultural Economists Conference procceding, 2007,* pp.85-89.

Ojiem, J. O., De Ridder, N., Vanlauwe, B., & Giller, K. E. (2006). Socio-ecological niche: a conceptual framework for integration of legumes in smallholder farming systems. *International Journal of Agricultural Sustainability*. 4 (1), pp. 79-93.

Oshone, K., Gebeyehu, S., & Tesfaye, K. (2014). Assessment of common bean (*Phaseolus vulgaris* l.) Seed quality produced under different cropping systems by smallholder farmers in eastern Ethiopia. *African Journal of Food, Agriculture, Nutrition and Development*. 14 (1), pp. 8566-8584.

Pacheco, F. P., Nóbrega, L. H. P., LIMA, G. P. D., Santorum, M., Boller, W., & Formighieri, L. (2015). Physiological Quality of Soybean Seeds under Mechanical Injuries Caused by Combines. *Revista Caatinga*. 28 (4), pp. 190-201.

Pradhan, B. K., & Badola, H. K. (2012). Effect of storage conditions and storage periods on seed germination in eleven populations of *Swertia chirayita*: a critically endangered medicinal herb in Himalaya. *The Scientific World Journal*. 1, pp. 1-9.

Rahman, M. H., Sattar, M. A., Salim, M. M. R., Quddus, M. A., & Ali, M. M. (2017). Study on Quality of Okra (*Abelmoschus esculentus* L.) Seed Collected from Different Sources and Locations of Bangladesh. *American Journal of Plant Biology*. 2 (4), pp. 129-135.

Reddy, M. V. B., Kushalappa, A. C., Raghavan, G. S. V., & Stevenson, M. M. P. (1995). Eradication of

seedborne *Diaporthe phaseolorum* in soybean by microwave treatment. *Journal of Microwave Power and Electromagnetic Energy*. 30 (4), pp. 199–204.

Sanginga, N., Dashiell, K. E., Diels, J., Vanlauwe, B., Lyasse, O., Carsky, R. J.,... & Singh, B. B. (2003). Sustainable resource management coupled to resilient germplasm to provide new intensive cereal–grain–legume–livestock systems in the dry savanna. *Agriculture, Ecosystems & Environment*. 100 (2-3), pp. 305-314.

Shovan L. R., Bhuiyan A., Suitana N., Begum J. A. & Pervez Z. (2008). Prevalence of fungi associated with soybean seeds and pathogencity tests of the major seed borne pathogen. *International Journal of Sustainable Crop Production.* 3(4), pp. 24-33.

Sinclair, J.B. (1991). Latent infection of soybean plants and seeds by fungi. *Plant Disease*. 75 (3), pp. 220-224.

Singh, D. P. & Agarwal, V. K., (1986). Purple stain of soybean and seed viability. *Seed Research.* 14 (1), pp. 126.

Singh, K. K., & Dadlani, M. (2003). Effect of packaging on vigour and viability of soybean [*Glycine max* (L.) *Merrill*] seed during ambient storage. *Seed Research-New Delhi.* 31 (1), pp. 27-32.

Steel, R.G. & Torrie, J. (1960). Principles and Procedures of Statistics. Mcgraw-Hill Book Company, Inc.; New York; Toronto; London.

Surve, U. S.,Patil, P. C. & Gagre, K. C. (2015). Response of Soybean Varieties to different Threshing Methods and Storage Periods. *International Journal of Tropical Agriculture*. 33(2), pp. 1237-1240.

Utoba, E. B., Ogbod, E. N. & Nwogbaga, A. C. (2011). Seedborne mycoflora associated with rice and their influence on growth at Abakaliki, Southeast Agro-Ecology, Nigeria. Libyan *Agricultural Research Centre Journal International*, 2(2), pp. 79-84.

Van Utrecht, D., Bern, J. C. & Rukunudin, I. H. (2000). Soybean Mechanical Damage Detection. *Applied Engineering in Agriculture*. 16 (2), pp. 137-141.

Verma, S. S., Tomer, R. P. S., & Urmil, V. (2003). Loss of viability and vigour in Indian mustard seeds stored under ambient conditions. *Seed Research.* 31(1), pp. 98-101.

Abundance and Diversity of Frugivorous Fruit Flies in Kandara, Murang'a County, Kenya

Muriuki C.[1,2], Guantai M.[3], Samita N.[1], Mulwa J.[4], Nyamai M.[4] and Kasina M[2]

[1]School of Agriculture and Enterprise Development, Kenyatta University, P.O Box 43844 - 00100, Nairobi.
[2]National Sericulture Research Centre, Industrial Crops Research Institute, Kenya Agricultural and Livestock Research Organization, P.O. Box 7816 - 01000 Thika.

[3]Kenya Plant Health Inspectorate Service, P.O Box 49592 - 00100, Nairobi.

[4]National Agricultural Research Labs, Food Crops Research Institute, Kenya Agricultural and Livestock Research Organization, P.O. Box 14733 - 00800 Nairobi.

Corresponding Author: mguantai@kephis.org

Abstract

Fruits and vegetables are important source of livelihood to farmers and major horticulture sub sector with high contribution to agricultural GDP in Kenya. This study was conducted to determine diversity and abundance of frugivorous fruit flies in Kandara sub county, Murang'a County in 2018, at a place where first area of low pest population was created in Kenya for *Bactrocera dorsalis*. Three sets of pheromone traps (Methyl-Eugenol, Cuelure and Trimedlure) were set in six trap stations within farmers' orchards in four agro-ecological zones (LH1 (Lower Highland Zone), UM1 (Upper Mid-land Zone), UM2, and UM3). The trap catch data was collected fortnightly and data analyzed. Six fruit flies species namely; *Bactrocera dorsalis*, *Ceratitis cosyra*, *Ceratitis capitata, Zeugodacus cucurbitae, Dacus bivittatus* and *Perilampsis sp* were identified. *Bactrocera dorsalis* population was significantly (P<.001) different across the four agro-ecologies with lowest densities at LH1 and highest at UM3. Likewise, *C. capitata* recorded significant (P=0.042) difference densities across the agro-ecological zones, but no significant (P=0.386) difference was recorded for *C. cosyra* across the agro-ecological zones. Further, there was significant (P=0.012) difference in the number of *Perilampsis sp* across the agro-ecologies with the highest number recorded in UM1. Both *Z. cucurbitae* (P=0.061) and *D. bivittatus* (P=0.056) had low abundance across the agro-ecologies. The peak infestation period differed across the various fruit fly species, whereby *B. dorsalis* peaked in May, *C. capitata* in February and *C. cosyra*

in January. The study shows that abundance for the fruit flies is probably related to their preferred hostplant and the weather patterns. We recommend continuous monitoring and intensifying trapping activities during peak periods in order to control the pest and protect fruits from damage. Farmers should be trained on the use of pheromone traps to reduce over-reliance on pesticides.

Key words: Agro-ecologies, *Bactrocera dorsalis, Ceratitis* sp, fruit fly density, Pheromone,

Introduction

Fruits are an important source of livelihood for farmers and they further contribute immensely to the agricultural GDP for the country. They also improve diet by providing nutrients and essential vitamins (Thomas, 2008). Majority of households living in Kandara is composed of farmers who grow mangoes, avocados, and guavas as commercial fruits which unfortunately are among the main host plants for fruit flies. Therefore, the productivity and quality of these fruit crops is highly affected by fruit flies (Tephritidae) which cause damage directly by puncturing the fruits to lay eggs, the hatched maggots feed on the fruit creating galleries that serve as entry points for pathogens, fruit decay occurs and then falls to the ground, which contribute to high farm losses. Exported fruit have been intercepted due to presence of fruit flies by the importing countries (Bissdorf & Weber, 2005; Follett & Neven, 2006). For example, since 2015 to date, 19 interceptions have been received from EU due to Tephritidae flies in Mango, Capsicum, Eryngium and Cucurbits (EC, 2020).

Pest management practices by farmers in Kandara include use of pesticides that has resulted in their over-reliance control, increased cost of production and economic losses due to rejection of fruits as a result of maximum residue levels, increased pollution, health problems, (USDA-APHIS, 2008).

Poor farmer knowledge on fruit fly development in relation to host development, possible practical pest management options have resulted into

over-reliance on pesticides. Earlier studies indicate that lack of training and technical support to fruit farmers has contributed immensely to low adoption of Integrated Pest Management (IPM) technologies in developing countries (Parsa *et al*., 2014). The use of insect development and reproductive behaviour such as their activity density trends in the farmland is a major step towards their successful management. Earlier studies indicated that the activity density and distribution of Tephritid fruit flies is affected by biotic factors such as temperature and humidity (Vayssières *et al.,* 2008). For example, studies carried out in Thailand, indicated that the developmental time for the immature stages of *Bactrocera carambolae* and *Bactrocera papayae* increased with decrease in temperature (Danjuma *et al.,* 2014) whereas the optimum temperature for fruitflies development has been reported to lie between 20° and 30°C for *B. dorsalis* (Rwomushana *et al.,* 2008) and between 26 and 30°C for *B. cucurbitae,*. *The* purpose of this study was to determine the diversity and activity density trends of fruit flies across the agro ecological zones in Kandara from January to May 2018.

MATERIALS AND METHODS

Study site

The study was carried out in Kandara sub-County in Murang'a County in 2018. Kandara Sub County covers an area of about 236 km^2 and is located at latitude 0° 53'59.99"N and longitude 37° 00'0.00"E and an altitude of between 1520-1880 m above sea level. Kandara is composed of four agro ecological zones; Lower Highland Zone (LH1) - Tea and dairy zone, First Upper Mid-land Zone (UM1) - Coffee and tea zone, Second Upper Mid-land Zone (UM2) - Main coffee zone, Third Upper Mid-land Zone (UM3) - Marginal coffee zone (Fig 1). The average annual rainfall of the area studied ranges between 1,400 to 2,000mm and the annual mean temperature is between 18°c to 21°c (Jaetzold *et al.,* 2006).

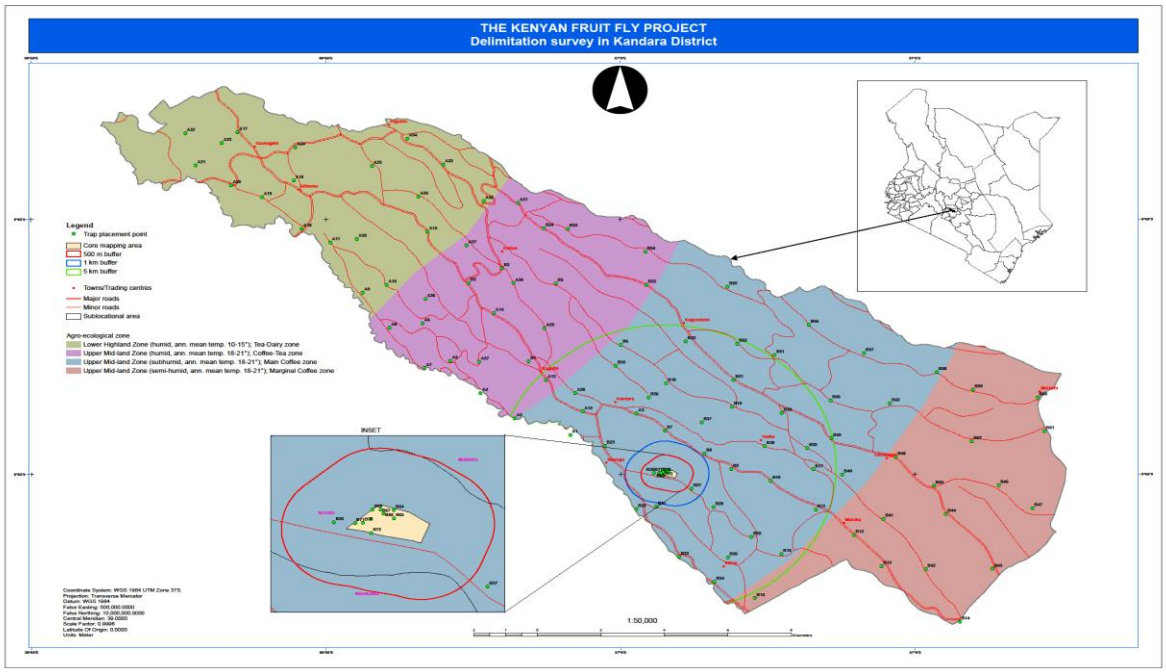

Figure 1. Map of the study sites

Data collection

Three sets of pheromone traps (Methyl-Eugenol, Cuelure and Trimedlure) were set in six trap stations within mango, avocado and guava farms in the four agro-ecological zones (LH1 (Lower Highland Zone), UM1 (Upper Mid-land Zone), UM2 (second Upper Midland Zone) and UM3 (third Upper Midland Zone) in the sub-county in 2018. Methyl –Eugenol was used to attract *Bactrocera dorsalis*, TrimediLure was used to attract *Ceratitis cosyra* and *Ceratitis capitata,* While Cuelure was used to attract both *Bactrocera cucurbitae* and *Dacus bivittatus.* Trap catch data was collected on a fortnight basis and servicing of the monitoring traps done every 6 weeks. Samples of collected trap catches were put in diffrent vials and taken to the laboratory at the Kenya Agricultural and Livestock Research Organization (KALRO) Sericulture, for further identification and counting. The ANOVA of the trap catch data was done using Genstat 17th edition. Significant means were separated using Fishers Protected

Least Significance Difference Test (LSD).

Results

Six species of fruit flies were identified across the four agro-ecological zones. These were *Bactrocera dorsalis Ceratitis cosyra*, *Ceratitis capitata, Bactrocera cucurbitae, Dacus bivittatus* and *Perilampsis sp.* (Table 1). A significant difference in the number *B. dorsalis* (P<0.001), *C. capitata* (P=0.042) and *Perilampsis* sp. (P=0.012) was recorded across the agro-ecological zones. However, there was no significant difference in the number of *C. cosyra* (P=0.386), *D. bivittatus* (P=0.056) and *B. cucurbitae* (P=0.061) across the agro ecologies. *Perilampsis sp* recorded the least activity density across all the agro ecologies, possibly due to low sensitivity of the lures towards this fruit fly. Generally, UM3 had the highest number of fruit flies (45.28%), followed by UM1 (24.77%), UM2 (22.40%) and the least was LH1 (7.55%). However, a significant number of *D. bivittatus* was recorded in LH1 (Table 1).

Table 1: Fruit fly densities across the agro-ecological zones.

Agro Ecology	***B. dorsalis***	***C. cosyra***	***C capitata***	***B. cucurbitae***	***D. bivittatus***	***Perilampsis sp***
LH1	3.64^{b}	17.7^{a}	27.21^{a}	4.267^{a}	7.752^{a}	0.2^{b}
UM1	65.69^{b}	41.25^{ab}	80.38^{b}	5.286^{a}	4.848^{b}	1.7905^{a}
UM2	106.9^{b}	25.54^{ab}	36.87^{c}	6.202^{a}	4.865^{b}	0.7212^{ab}
UM3	231.68^{a}	45.3^{b}	72.42^{b}	11.048^{b}	3.333^{b}	0.4762^{ab}
P value	<.001	0.386	0.042	0.061	0.056	0.012
s.e.	32.44	12.88	15.62	1.91	1.16	0.36

Abundance of *C. capitata* increased gradually from January peaking in February after which a gradual drop was recorded. In contrast, *C. cosyra* decreased gradually from January and almost flattened in April. A gradual increase in *B. dorsalis* was recorded from January to March and thereafter the population increased exponentially till the end of May (Fig 2).

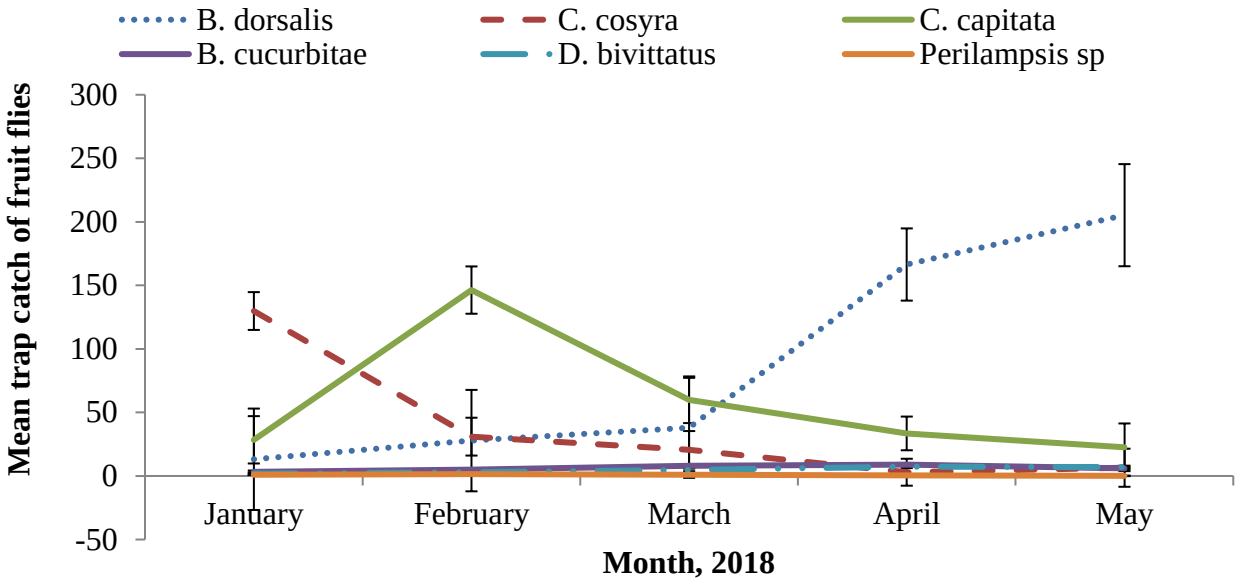

Figure 2: Fruit fly trap catch trend (Jan-May, 2018).

Discussion

Activity density of fruit flies differed across the four agro ecological zones probably due to variation in climatic conditions across the zones. The LH1, which is at a higher altitude and records lower temperatures explains why few fruit flies were recorded in this region. Further, fruit diversity is low in this zone. Temperature levels increase towards UM3 and this is likewise for fruit crop diversity and intensity of production. Previous studies on oriental fly (*B. dorsalis*) distribution in Kenya in the same area indicated that LH1 and UM1 had significantly lower pest population compared to UM2 due to cool weather LH1 (Kasina *et al.,* 2019). Earlier studies by Vayssières et al. (2008) indicated that Tephritid distribution and abundance depend on several abiotic factors (e.g., temperature, relative humidity, rainfall) and biotic factors (e.g., host plants, natural enemies). Low temperature was found to increase developmental time of immature stages of *Bactrocera carambolae* and *Bactrocera papaya* (Danjuma *et al.,* 2014) with the optimum temperature found to range

between 25 and 30°C for *B. invadens* (Rwomushana *et al.,* 2008).

The difference in the level of infestation of fruit fly species across the months (January to May 2018) is associated to the availability of suitable fruits for egg laying and multiplication. In Kandara, mangoes matured between February and April while avocadoes matured from March. This ensured sufficient food supply for multiplication of the fruit flies, especially *B. dorsalis* throughout the study period. *B. dorsalis* is known to attack at least 46 host plants, including many commercially grown fruit crops such as mango, oranges, guava, cucurbit, papaya and avocado, as well as many other species indigenous to Africa (José *et al.,* 2013). Earlier studies on preferred hosts in Zimbabwe indicate that availability of cultivated and wild fruit varieties throughout the year results in increased population of fruit flies making it difficult to manage them (Musasa *et al.,* 2019). A previous review on status of data from Afrotropical countries indicated that host availability and ecological niches affected the occurrence and impact of *Z. cucurbitae* (De meyer *et al.,* 2015). Other fruit fly species are more host specific explaining why their numbers may have remained relatively low in absence of their hosts. Our results shows that abundance for the fruit flies is probably related to their preferred hostplant and the weather patterns.

Recommendations

There is need to carry out continuous monitoring of fruit flies in Kandara throughout the growing season for their timely management to reduce fruit damage and meet phytosanitary requirements. There is need to train farmers on how to use pheromone traps with specific lures to reduce the pest population. A year round fruit fly management in the farmland is the only assurance for long term reduction and control of the diverse fruit fly pest in the locality.

Acknowledgement

This study was carried out with financial support from Horticulture Research Funds (HRF) to KALRO for fruit fly programme. We thank KALRO for facilitation and farmers for allowing access to farms. We also thank International Atomic Energy Agency through the project RAF 5074 on fruit flies for provision of lures and traps.

References

Bissdorf, J. & Weber, C. (2005). Field guide to non-chemical pest management in mango production. Pesticide Action Network (PAN), Hamburg, Germany.

Business Daily, (April 11, 2017). https://www.businessdailyafrica.com.

Clarke, A.R., Armstrong, K.F., Carmichael, A.E., Milne, J.R., Raghu, S., Roderick, G.K. & Yeates, D.K. (2005). Invasive phytophagous pests arising through a recent tropical evolutionary radiation: The *Bactrocera dorsalis* complex of fruit flies. *Annual Review of Entomology.* 50, pp. 293-319.

Danjuma, S., Thaochan, N., Permkam, S. & Satasook, C. (2014). Effect of temperature on the development and survival of immature stages of the carambola fruit fly, *Bactrocera carambolae*, and the Asian papaya fruit fly, *Bactrocera papayae*, reared on guava diet *Journal of Insect Science*, 14 (1), pp. 126.

De Meyer, M., Mohamed, S. & White, I.M. (2012). Invasive fruit fly pests in Africa. http://www.africamuseum.be/fruitfly/AfroAsia.htm. Date of access: 18th October 2017.

European Commission (2020). Interception of harmful organism in imported plants and other objects. European Union Notification System for Plant Health Interceptions-EUROPHYT; Monthly interceptions 2020.

Ekesi, S. & Billah, M. (2007). A field guide to the management of economically important Tephritid fruit flies in Africa. International

Centre of Insect Physiology and Ecology (ICIPE). Nairobi, Kenya: Science Press.

Ekesi, S., Mohamed, S. & Hanna, R. (2010). Rid fruits and vegetables in Africa of notorious fruit flies. CGIAR SP-IPM Technical Innovation Brief No. 4: 1-2, SP-IPM Secretariat, International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria.

Ekesi, S., Rwomushana, I., Gordon,I. & Ogol, C. K. (2008). Host plants and host plant preference studies for Bactrocera invadens (Diptera: Tephritidae) in Kenya, a new invasive fruit fly species in Africa. *Annals of the Entomologica Society of America*, 101(2), pp. 331-340.

Follett, P.A. & Neven, L.G. (2006). Current trends in quarantine entomology. *Annual Review of Entomology* 51, pp. 359–385.

José, L., Cugala, D. & Santos, L. (2013). Assessment of invasive fruit fly fruit infestation and damage in Cabo Delgado Province, Northern Mozambique. *African Crop Science Journal*. 21, pp. 21–28.

Jaetzold, R., Schmidt H., Hornetz B. & Shisanya C. (2006). Farm Management Handbook of Kenya. Natural Conditions and Farm Management. Second Edition. MoA/GTZ, Nairobi, Kenya.

Kasina M., Momanyi G., Mulwa J., Karanja T., Kithusi G. K., Matolo N. & Maingi P. (2019). Oriental Fruit fly distribution within Kandara sub-County, Murang'a, Kenya. *African Phytosanitary Journal.* 01(01), pp. 1-10.

Marc De Meyer, Hélène Delatte, Maulid Mwatawala, Serge Quilici, Jean-François Vayssières and Massimiliano Virgilio (2015). A review of the current knowledge on *Zeugodacus cucurbitae* (Coquillett) (Diptera, Tephritidae) in Africa, with a list of species included in Zeugodacus. *ZooKeys*. 540, pp. 539–557.

Musasa, S.T., Mashingaidze, A.B., & Musundire, R. (2019). Fruit fly identification, population dynamics and fruit damage during fruiting seasons of sweet oranges in Rusitu

Valley, Zimbabwe. *Science Reports.* 9, pp. 13578.

Parsa, S., Morse, S., Bonifacio, A., Chancellor, T.C., Condori, B., Crespo-Pérez, V. & Dangles, O. (2014). Obstacles to integrated pest management adoption in developing countries. Proceedings of the *National Academy of Sciences*. 111(10), pp. 3889–94.

Rwomushana, I., Ekesi, S., Ogol, C.K.P.O. & Gordon, I (2008). Effect of temperature on development and survival of immature stages of *Bactrocera invadens* (Diptera: Tephritidae). *Journal of Applied Entomology.* 132, pp. 832-839

Tan, K. H., Tokushima, I., Ono, H. & Nishida, R. (2010). Comparison of phenylpropanoid volatiles in male rectal pheromone gland after methyl eugenol consumption, and molecular phylogenetic relationship of four global pest fruit fly species: *Bactrocera invadens*, *B. dorsalis, B. correcta* and *B. zonata*. *Journal of Chemical Ecology.* 21, pp. 25-33.

Thomas, S.C.Li. (2008). Vegetables and Fruits: Nutritional and Therapeutic Values. CRO Press, 2008.

United States Department of Agriculture, Animal and Plant Health Inspection Service (USDAAPHIS), (2008). Federal import quarantine order for host materials of *Bactrocera invadens* (Diptera: Tephritidae), invasive fruit fly species.

Vayssières, J.F., Carel, Y., Coubes, M. & Duyck, P.F. (2008). Development of immature stages and comparative demography of two cucurbit-attacking fruit flies in Reunion Island: *Bactrocera cucurbitae* and *Dacus ciliates* (Diptera: Tephritidae). *Environmental Entomology.* 37, pp. 307-314.

Next generation sequencing as a tool in modern pest risk analysis: a case study of groundnuts (*Arachis hypogaea*) as a potential host of new viruses in western Kenya

Mukoye B[1*]., Mangeni B. C[2]., Sue J[3]., Mabele A. S[2]. and Were H. K[2].

[1] Kenya Plant Health Inspectorate Service (KEPHIS), Nairobi, Kenya.

[2] Department of Biological Sciences, Masinde Muliro University of Science and Technology (MMUST), Kakamega, Kenya.

[3] Cell and Molecular Biology Sciences, The James Hutton Institute (JHI), Dundee, UK.

*Corresponding Author: btemukoye@gmail.com

Abstract

Groundnut (*Arachis hypogaea,* L.*)* is grown in diverse environments throughout the semi-arid and sub-tropical regions of the world. Poor yields of 500-800kg/ha are attributed to poor agronomic practices, pests and diseases. The major disease reported in Kenya is Groundnut rosette disease (GRD). But recent observations in the field showed that the crop has varied and severe symptoms in addition to those caused by GRD. This required deeper analysis to establish the causal agents. Groundnut samples with virus-like symptoms were collected from western Kenya in 2016. Total RNA was extracted using All Prep RNA Mini Kit. Five mRNA libraries were prepared using the Illumina TrueSeq stranded mRNA library Prep Kit and pooled for multiplexed sequencing using an Illumina HiSeq 2500 to generate paired end reads (FastQ Sanger). The reads were analysed in the Galaxy project platform (customized). Quality reads were first mapped onto plant genome Refseq and unmapped reads isolated and mapped onto virus Refseq using Bowtie 2 (v2.2.3). Groundnut rosette virus satellite RNA, Groundnut rosette virus, Groundnut rosette assistor virus, Ethiopian tobacco bushy top virus, Cowpea polerovirus 2, Chickpea chlorotic stunt virus, Melon aphid-borne yellow virus, Phasey bean mild yellow virus, Beet mild yellowing virus, White clover mottle virus and Cotton leafroll dwarf virus were identified in four libraries. Other viruses (with less than 100 reads) including Bean common mosaic virus, Bean common mosaic necrosis virus, Cowpea chlorotic mottle virus RNA 3, Broad bean mottle virus RNA 3, Passion fruit woodiness virus among others were also mapped. Some of the viruses common in western Kenya were confirmed by PCR. The presence

of at least three viruses in groundnuts in Western Kenya highlights the importance of starting a germplasm clean-up program of the plant material used as seed in this crop.

Key words: Groundnuts, NGS, RefSeq, Viruses.

Introduction

Virus infection is prevalent across many types of plants, and is of specific importance in crops cultivated for food, where they cause significant yield and quality losses. Groundnuts (*Arachis hypogaea* L.), belongs to the family *Fabaceae,* and is the only domesticated species in the genus (Usman *et al*., 2013). Groundnut production is an enterprise of economic and nutritional value for farmers in east Africa (Okello *et al*., 2010). Resource poor smallholder farmers grow nearly 75 - 80% of the world's groundnuts in developing countries obtaining yields of 500-800kg/ha, as opposed to the potential yield of >2.5t/ha (Kayondo *et al.,* 2014). In western Kenya, an average of 600 – 700 kg/ha is achieved which is less than 30-50% of the potential yield. The low yields are mainly attributed to poor quality seeds, drought, poor agronomic practices, numerous pests and diseases caused by numerous pathogenic viruses, fungi, bacteria and nematodes (Mabele *et al*., 2020; Mutegi *et al*., 2010;). About 31 viruses have been reported to naturally infect groundnuts around the world (Kumar *et al.,* 2007). These viruses belong to various genera including *Potyvirus*, *Tospovirus, Cucumovirus, Pecluvirus, Soymovirus Umbravirus*, *Begomovirus, Bromovirus, Carlavirus, Ilarvirus, Luteovirus, Potexvirus, Rhabdovirus* and *Tymovirus.* Nineteen of these viruses were first isolated from groundnuts, while the rest from other hosts, but they commonly occur on groundnuts. The most economically important viruses of groundnuts are: *Groundnut rosette virus* (GRV), *Cucumber Mosaic Virus* (CMV), *Peanut mottle virus* (PeMoV), *Groundnut bud necrosis virus* (GBNV), *Indian peanut clump virus* (IPCV), *Groundnut rosette assistor virus* (GRAV), *Peanut stripe virus* (PStV), *Peanut clump virus* (PCV), *Tomato spotted wilt virus* (TSWV), *Tobacco streak virus* (TSV) (Okello *et*

al., 2014) and *Cowpea mild mottle virus* (CPMMV) (Mukoye *et al*., 2015). The observations made on groundnuts in western Kenya showed severe and highly variable virus-like symptoms which could be due to multiple infection of any of the groundnut viruses (Mukoye *et al*., 2020).

Proper diagnosis of plant viruses is the first step to the development of their management strategies in addition to preventing their introduction and spread. New viruses are identified on a regular basis and more are yet to be uncovered in some hosts or in other geographical regions where they have not been reported. Therefore, there is need for a robust tool to identify new viruses that have not been identified in new geographical areas, and in new hosts or new recombinants. Next generation sequencing (NGS) technologies are fast becoming a popular method to obtain whole plant virus genomes in a relatively short period of time (Boonham *et al*., 2014). NGS sequences complete genomes of plant viruses and still obtains excellent results due to its ability to use total RNA and DNA extractions (Adams *et al*., 2009). This study utilized NGS to establish viruses that could be causing the observed varied symptoms in groundnuts.

Materials and methods

Groundnut leaf samples showing virus-like symptoms of green mosaic, leaf distortion, downward curling, mottling, chlorotic areas, necrotic spots, local lesions, stunting or a combination of these were collected in RNA*later*® RNA Stabilization Solution and kept at 4^{o}C until further analysis. The leaves were collected in fields from Bungoma, Busia, Homabay, Kakamega, Siaya and Vihiga Counties through systematic sampling during the 2016-2017 short rains and long rains seasons.

Total RNA was extracted using All Prep RNA Mini Kit in the pooled samples. Five mRNA libraries were prepared using the Illumina TrueSeq stranded mRNA library Prep Kit and pooled for multiplexed sequencing using an Illumina HiSeq 2500 to generate paired end reads (FastQ Sanger). The reads were analysed in the Galaxy project

platform (customized). Quality control of the raw reads was conducted using Trimmomatic (Bolger et al., 2014) with parameters: LEADING: 20 TRAILING: 20 SLIDINGWINDOW: 4:20 and a minimum read length of 50. To remove host reads, trimmed reads were mapped to the concatenated genome sequences of two diploid ancestors of A. *hypogaea,* A. *duranensis* (Genbank GCA_000817695.2) and A. *ipaensis* (Genbank GCA_000816755.2) (Bertioli *et al*., 2016) and the chloroplast genome of A. *hypogaea* (Genbank KX257487.1) (Prabhudas *et al*., 2016) (as there is currently no sequenced genome for A. *hypogaea*). Mapping was conducted using Bowtie2 (Langmead, 2013) (score-min value"L,0,-0.2"). The un-mapped reads, designated as non-host reads were then assembled into contigs using Trinity (Grabherr *et al*., 2011) with a minimum contig length of 200bps. The contigs were mapped to the concatenated host ancestor genome using Bowtie2 (Langmead, 2013), and the unmapped contigs designated as non-host contigs. The non-host contigs (≥300 bp) were then aligned against a dataset of 691 K proteins from known virus genomes [extracted from GenBank (Benson *et al*., 2013) release 225] by selecting entries classified as 'virus' (VRL partition) and 'complete genome' using the NCBI's Assembly database (Kitts *et al*., 2016). The alignment was conducted using BlastX® (Altschul *et al*., 1990), and aligned contigs (e-values<10−6) denoted as virus sequences. The BlastX® alignments for virus-derived contigs were then checked manually to confirm virus sequence identification.

Verification of some of the common viruses detected by NGS (with less than 100 reads) was done by RT-PCR according to Naidu *et al.,* (1998a). This was done on some of the groundnut samples returned after sequencing (1, 2, 3, 4 and 5). The target viruses were Cowpea aphid-borne mosaic virus (CABMV), Bean common mosaic virus (BCMV), Bean common mosaic necrosis virus (BCMNV), Cowpea mild mottle virus (CPMMV) and Cucumber mosaic virus (CMV).

Total RNA was extracted using RNeasy Plant Mini Kit (Qiagen) following the

manufacturer's instructions. For samples 1, 2, 3 and 4, the leaf tissue was homogenized in liquid nitrogen while for sample 5, (which was split into 3 sets –A, B and C based on the fact that each leaf was from a different plant) the tissue was homogenized in lysis buffer provided in the kit. Coat protein primers for BCMV, BCMNV and CABMV were chosen using Primer3 software with reference to known accessions, namely; BCMNV NL-3 (accession Z17203.21) Pathogroup V1, BCMV NL-2 (accession L19472.1) Pathogroup V (Mangeni *et al*., 2014) and CABMV (accession X82873) Zimbabwe isolate (Mlotswa *et al*., 2002).

Results

Raw reads obtained ranged from 3.2 – 7.2 million. After trimming, the yield ranged from 2.8 – 6.3 million of which between 50 – 70% mapped to host genome. About 0.2 – 11% of the non-host reads mapped to the virus genome (VRL) (Table 1).

Table 1: Reads and contig counts information for each library RNA-seq dataset.

Library	E5	E7	E8	E9
Raw Reads	3,329,984	3,238,295	7,263,305	4,316,937
Reads after trimming	2,957,536	2,888,001	6,361,618	3,830,698
Reads after host mapping	996,404	1,472,648	3,019,197	2,139,196
Reads mapped to host	69.9%	54.5%	57.5%	50.2%
Reads mapped to Gb-VRL-cg (%)	0.88%	5.6%	11.6%	0.23%
Reads un-mapped to Gb-VRL-cg	989,041	1,407,511	2,713,214	2,136,764
Contigs assembled	15,183	79,111	24,346	10,658
Contigs mapping to host	210	756	1146	202
Contigs mapped to Gb-VRL-cg	1197	1769	1707	805
Unmapped contigs >= 1000bps	115	522	436	225

Groundnut rosette virus (GRV), its associated satellite RNA (sat-RNA) and Groundnut rosettes assistor virus (GRAV) were the common viruses detected in most of the libraries. Other viruses detected include Ethiopian tobacco bushy top virus, cowpea

polerovirus 2, chickpea chlorotic stunt virus, Melon aphid-borne yellow virus, Phasey bean mild yellow virus, Beet mild yellowing virus, White clover mottle virus, Cotton leafroll dwarf virus (Table 2).

Table 2: Viruses identified in the 4 libraries using the bioinformatics workflow. Viruses are only reported if they have ≥ 100 reads or ≥ 5 contigs mapped and 20% coverage.

Genbank Code	TaxID	Reads	%Cov	Contigs	%Cov	Virus name
E5						
AF195825.1	33761	1622	99.7	.	.	Groundnut rosette assistor virus clone N15GCP coat protein gene, complete cds
AF202870.1	127441	131	46.7	.	.	Satellite RNA of Groundnut Rosette Virus clone N310S, complete sequence
KY364847.1	1913125	590	38.6	10	50.6	Cowpea polerovirus 2 isolate BE179, complete genome
Z69910.1	47740	.	.	5	65.7	Groundnut rosette virus complete genome, strain MC1
KT962999.1	1756832	.	.	10	39.4	Phasey bean mild yellows virus isolate NSWCP15, complete genome
E7						
AF195825.1	33761	2025	100	9	100	Groundnut rosette assistor virus clone N15GCP coat protein gene, complete cds
AF202870.1	127441	1803	55	19	90.5	Satellite RNA of Groundnut Rosette Virus clone N310S, complete sequence
Z69910.1	47740	.	.	10	65.8	Groundnut rosette virus complete genome, strain MC1
KJ918748.1	1538549	.	.	8	49.3	Ethiopian tobacco bushy top virus isolate 18-2, complete genome
AY956384.1	328430	.	.	9	45.2	Chickpea chlorotic stunt virus isolate Et-fb-am1, complete genome
KY364847.1	1913125	.	.	5	43.9	Cowpea polerovirus 2 isolate BE179, complete genome
EU000534.1	471717	.	.	7	35.6	Melon aphid-borne yellows virus, complete genome
X83110.1	156690	.	.	5	32.2	Beet mild yellowing virus genomic RNA
LC192169.1	1913024	.	.	9	31.8	White clover mottle virus genomic RNA, complete genome, strain: CD
GU167940.1	312295	.	.	5	31.1	Cotton leafroll dwarf virus isolate ARG, complete sequence
KY364846.1	1913124	.	.	10	26.4	Cowpea polerovirus 1 isolate BE167, complete genome
E8						
AF195825.1	33761	1294	100	.	.	Groundnut rosette assistor virus clone N15GCP coat protein gene, complete cds
AF202870.1	127441	17408	56	26	62.4	Satellite RNA of Groundnut Rosette Virus clone N310S, complete sequence
Z69910.1	47740	.	.	11	43.1	Groundnut rosette virus complete genome, strain MC1
KJ918748.1	1538549	.	.	9	26.2	Ethiopian tobacco bushy top virus isolate 18-2, complete genome
E9						
KT456288.1	1188793	3219	93.7	.	.	Phaseolus vulgaris endornavirus 2 isolate PvEV-2_Brazil polyprotein gene, complete cds
AF202870.1	127441	.	.	5	94.3	Satellite RNA of Groundnut Rosette Virus clone N310S, complete sequence

The fifth library (E6) revealed some of the common viruses in western Kenya but with less than 100 reads. These were Bean common mosaic virus, Bean common mosaic necrosis virus, Broad bean mottle virus RNA 3, Passion fruit woodiness virus and Cowpea aphid-borne mosaic virus.

Table 3: Library E6 matched viruses with <100 reads.

RefSeq	Reads	Genome match annotation*
ref|NC_002738.1|	23	Groundnut rosette virus satellite RNA, complete genome
ref|NC_030236.1|	7	Impatiens flower break potyvirus isolate Asan, complete genome
ref|NC_003397.1|	5	**Bean common mosaic virus**, complete genome
ref|NC_003603.1|	2	Groundnut rosette virus complete genome, strain MC1
ref|NC_004047.1|	2	**Bean common mosaic necrosis virus**, complete genome
ref|NC_014790.2|	2	**Passion fruit woodiness virus**, complete genome
ref|NC_004013.1|	1	**Cowpea aphid-borne mosaic virus**, complete genome

*Bolded refers to common viruses in western Kenya.

RT-PCR verification

Samples 1, 2 and 4 had BCMNV, BCMV and CABMV. Leaf sample 3 was free of these viruses (Figure 1). In sample 5, portions A and C were negative for CABMV, BCMV and BCMNV. Portion B was positive for all these viruses (Figure 2).

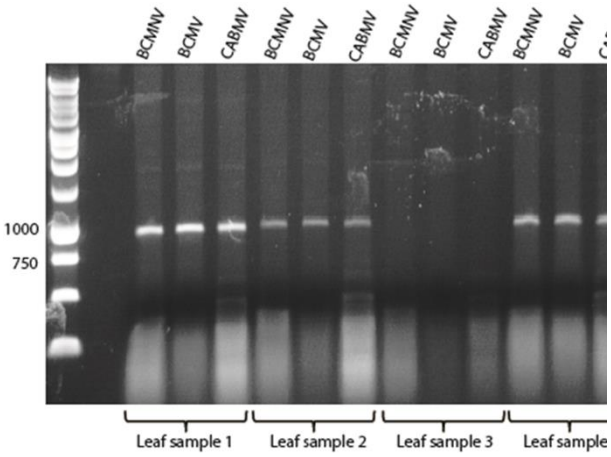

Figure 1: Gel electrophoresis view of RT-PCR results for samples 1-4.

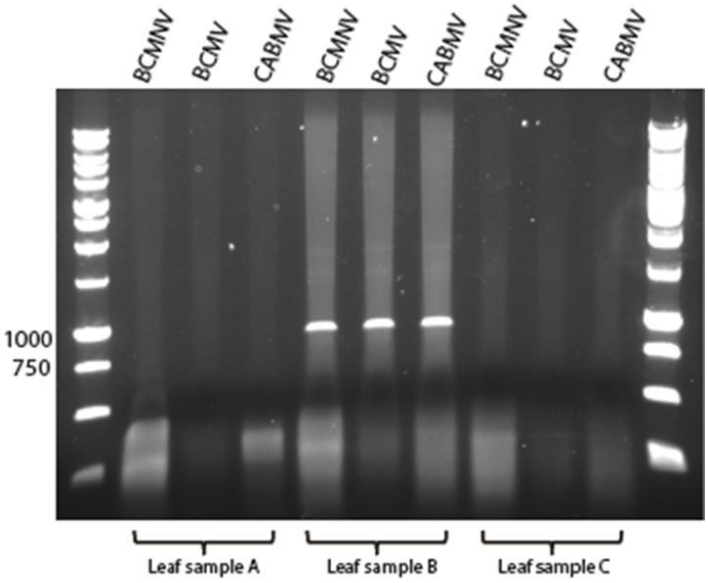

Figure 2: A gel electrophoresis view of RT-PCR results for sample 5 – A, B and C.

Discussion

Next generation sequencing (NGS) offers a great opportunity in diagnosis of plant viruses especially in the identification of new viruses. Detection of viral RNA and DNA genomes in infected plant material by NGS (Kreuze *et al.*, 2009) is possible through the extraction and sequencing of total RNA and DNA (Eichmeier *et al.*, 2016). NGS has the ability to sequence whole genomes of known and unknown viruses and the ability to detect multiple viruses from a mixed infection, thus providing a very sensitive diagnostic method for the rapid and routine detection of viruses. NGS being non-specific, can be used to detect all known and unknown viruses present in a host irrespective of their pathogenicity. In this study, common groundnut viruses namely: GRV, GRAV and sat-RNA were detected in almost all the libraries. In addition, several other new viruses were detected some of which have never been reported in groundnuts before. This confirms that NGS can be utilized in detection of known and unknown plant viruses.

The challenge to be addressed is proper analysis, interpretation and utilization of the huge data generated using NGS technology. Platforms to handle some of these challenges have been developed and still under constant improvement. The Galaxy platform is one of the effective one with proper tools in manipulation of NGS data. However, it needs a deeper understanding of the parameters involved in each tool contained.

The use of PCR and other serological virus detection methods are key in verification of the identified viruses using the NGS platform. This is key specifically when the technology is utilized by National Plant Protection Organizations (NPPOs) in virus diagnostics. The challenge will be in the detection of new/novel viruses whose quarantine status has not yet been established and therefore, this will require proper pest risk analysis (PRA) to be conducted. The NGS technology is a modern tool that is able to detect new viruses in plants, and therefore useful in enhancing phytosanitary operations in trade. Its use in determining the

groundnut virome revealed that the crop is a host of many viruses.

Recommendation

Utilization of new technologies like the new generation sequencing in pest diagnosis is recommended since it has the potential of eliminating ambiguity. A proper use of analysis of huge data generated and verification of the detected plant viruses. Virus containment in areas of detection is encouraged.

Acknowledgement

This work was funded by The royal society of UK. The sequencing was done at FERA Science (UK) and analysis done at James Hutton Institute (JHI).

References

Adams, I. P., Glover, R. H., Monger, W. A., Mumford, R. & Jackeviciene, E. (2009). Next generation sequencing and metagenomic analysis: a universal diagnostic tool in plant virology. *Molecular Plant Pathology*. 10, pp. 537–545.

Altschul, S. F., Gish, W., Miller, W., Myers, E. W. & Lipmon, D. J. (1990). Basic local alignment search tool. *Journal of Molecular Biology.* 215, pp. 403–410.

Boonham, N., Kreuze, J., Winter, S., van der Vlugt, R., Bergervoet, J., Tomlinson, J., & Mumford, R. (2014). Methods in virus diagnostics: from ELISA to next generation sequencing. *Virus research*. 186, pp. 20-31.

Benson, D. A., Cavanaugh, M., Clark, K., Karsch-Mizrachi, I., Lipman, D.J., Ostell, J. & Sayers, E.W. (2013). GenBank. *Nucleic Acids Research*. 41, pp. 36–42.

Bertioli, D. J., Cannon, S. B., Froenicke, L., Huang, G., Farmer, A. D., Cannon, E. K.,& Ren, L. (2016). The genome sequences of Arachis duranensis and Arachis ipaensis, the diploid ancestors of cultivated peanut. *Nature genetics*, 48(4), pp. 438-446.

Eichmeier, A., Kominkora, M., Kominek, P. & Baranek, M. (2016). Comprehensive virus detection

using next generation sequencing in grapevine vascular tissues of plants obtained from the wine region of Bohemia and Moravia (Czech Republic). *PloS ONE* 11(12):e0167966.doi:10.1371/journal.pone0167966.

Bolger, A. M., Lohse, M. & Usadel, B., (2014). Trimmomatic: a flexible trimmer for Illumina sequence data. *Bioinformatics*. 1–7.

Grabherr, M. G., Haas, B. J., Yassour, M., Levin, J. Z., Thompson, D. A., Amit, I., Adiconis, X., Fan, L., Raychowdhury, R., Zeng, Q., Chen, Z., Mauceli, E., Hacohen, N., Gnirke, A., Rhind, N., di Palma, F., Birren, B.W., Nusbaum, C., Lindblad-Toh, K., Friedman, N. & Regev, A., (2011). Full-length transcriptome assembly from RNA-Seq data without a reference genome. *Nature Biotechnology*. 29, pp. 644–652.

Kayondo, S. I., Rubaihayo, P. R., Ntare, B. R., Gibson, P. I., Edema, R., Ozimati, A. & Okello, D. K. (2014). Genetics of resistance to groundnut rosette virus disease*: African Crop Science Journal.* 22, (1), pp. 21-29.

Kitts, P. A., Church, D.M., Thibaud-Nissen, F., Choi, J., Hem, V., Sapojnikov, V., Smith, R.G., Tatusova, T., Xiang, C., Zherikov, A., DiCuccio, M., Murphy, T.D., Pruitt, K.D., Kimchi, A., (2016). Assembly: a resource for assembled genomes at NCBI. *Nucleic Acids Research*. 44, D73–D80.

Kumar, P. L. & Waliyar, F., (Ed). (2007). Diagnosis and detection of viruses infecting groundnuts. ICRISAT mandate crops: Methods Manual. Patancheru 502 324, Andhra Pradesh, India*: International Crops Research Institute for the Semi-Arid Tropics.* Pp 133.

Kreuze, J. F., Perez, A., Untiveros, M., Quispe, D., Fuentes, S. & Barker, I. (2009). Complete viral genome sequence and discovery of novel viruses by deep sequencing of small RNAs: *A generic method for diagnosis, discovery and*

sequencing of viruses. *Virology* 388, pp. 1-7.

Langmead, (2013). Fast gapped-read alignment with Bowtie 2. *Nature Methods*. 9, pp. 357–359.

Mabele, A. S., Were, H. K., Ndong'a, M. F. O. & Mukoye, B. (2020). Occurrence and genetic diversity of groundnut rosette assistor virus in western Kenya. *Elsevier Crop Protection.* 139, pp. 1-7.

Mangeni BC, MM Abang, H Awale, CN Omuse, R Leitch, W Arinaitwe, B Mukoye, JD Kelly & HK Were. (2014) Distribution and pathogenic characterization of bean common mosaic virus (BCMV) and bean common mosaic necrosis virus (BCMNV) in western Kenya. *Journal of Agri-Food and Applied Sciences*. 2: pp. 308-316.

Mlotshwa, S., Verver, J., Sithole-Niang, I., Van Kampen, T., Van Kammen, A. & Wellink, J. (2002). The genomic sequence of cowpea aphid-borne mosaic virus and its similarities with other potyviruses. *Archives of Virology*. 147, pp. 1043-52.

Mukoye, B., Were, H. K. & Ndong'a, M. F. O. (2020). Distribution and characterization of groundnut rosette associated viruses in western Kenya. *PhD thesis submitted to Masinde Muliro University of Science and Technology (MMUST)*, Kenya.

Mukoye, B., Mangeni, B.C., Leitich, R.K., Wosula, D. W., Omayio, D.O., Nyamwamu P.A., Arinaitwe, W., Winter, S., Abang, M.M. & Were, H.K. (2015). First report and biological characterization of cowpea mild mottle virus (CPMMV) infecting groundnuts in Western Kenya. *Journal of Agri-Food and Applied Sciences*. 3, pp. 1-5.

Mutegi, C. K. (2010). The extend of aflatoxin and aspergillus section flavi,penicillium spp.. and Rhizopus spp. contamination of peanuts from households in Western Kenya and the causative factors of contamination. *PhD dissertation,*

University of Kwazulu-Natal, Pietermaritzburg . South Africa.

Naidu, R. A., Robinson, D. J. & Kimmins, F. M. (1998a). Detection of each of the causal agents of grountnut rosette disease in plants and vector aphids by RT-PCR. *Journal of Virology Methods.* 76, pp. 9-18.

Okello, D. K., Birima, M. & Deom, C. M. (2010). Overview of groundnuts research in Uganda: Past, present and future. *African Journal of Biotechnology.* 9 (39): pp. 6448-6459.

Okello, D. V., Akello, L. B., Tukamuhabwa, P., Odongo, T. L., Ochwo-Ssemakula, M., Adriko, J. & Deom, C. M. (2014). Groundnut rosette disease symptom types, distribution and management of the disease in Uganda. *African Journal of Plant Science.* 8 (3): pp. 153-163.

Prabhudas, S. K., Prayaga, S., Madasamy, P., & Natarajan, P. (2016). Shallow whole genome sequencing for the assembly of complete chloroplast genome sequence of *Arachis hypogaea 1 Front. Plant Science*. 7, pp. 7–9.

Usman, A., Danquah, E. Y., Ofori, K. & Offei, S. K. (2013). Genetic analysis of resistance to rosette disease of groundnut (*Arachis hypogaea* L.). *A Thesis Submitted to the University of Ghana, Legon*. ISSN:10293978.

Maize Head Smut: Pathogenesis, Epidemiology, and Management Options

Chemeltorit, P.[1*] and Suresh, L.M[2].

[1]Kenya Plant Health Inspectorate Service (KEPHIS)

[2]International Maize and Wheat Improvement Center (CIMMYT), P.O. Box 1041 Village Market-00621, Nairobi.

[*]Corresponding Author: pchemeltorit@kephis.org

Abstract

Maize (*Zea mays*) head smut caused by *Sphacelotheca reiliana*, a basidiomycete with worldwide distribution, can cause devastating crop losses that pose a food security threat. While Kenya has experienced high incidences of the disease in the recent years, the seed certification regulation has a zero tolerance on *S. reiliana*. The spores of *S. reiliana* remain viable in soil for many years and serve as the main inoculum source as they germinate when the conditions are favourable and infect the host in the early stages of growth after germination. After penetration, the fungus grows systemically as the plant matures eventually transforming part or all of the inflorescence (ears and tassels) tissues into smut galls. The symptoms develop because the inflorescences have increased levels of reactive oxygen species, auxin, and misregulation of floral regulatory transcription factors. The most practical control strategy for maize head smut encompasses the use of resistant/tolerant cultivars, fungicide treatment (of seed or drenching of rows immediately after seeding), and field hygiene/ sanitation. Crop rotation may help when host crops are not planted for between 2-3 years or even more. Resistance genes, including ZmWAK, found in the major quantitative trait locus qHSR1/qHS2.09 regulate resistance of maize to head smut. The objective of this review paper is to provide an understanding of the head smut disease pathogenesis, epidemiology, and effective management options.

Key words: *Sphacelotheca reiliana*, basidiomycete, seed certification, smut galls, resistance gene

Introduction

Being the most important cereal crop in sub-Saharan Africa, Maize (*Zea mays*) is Kenya's staple crop with annual production steadily increasing over the last ten years, despite the area under production not changing significantly (FAOSTAT, 2018). Pest and diseases remain the main challenge to maize production and yield. Maize is affected by two types of smut diseases: head smut caused by *Sphacelotheca reiliana* (J. G. Kühn) G. P. Clinton [syn. *Sporisorium reilianum*], and common smut caused by *Ustilago maydis* (DC.) Corda. The head smut pathogen, *S. reiliana*, is a destructive soil borne basidiomycete of the ustilaginaceae family and has a worldwide distribution. *S. reiliana* is biotrophic (Martinez *et al*., 2002; Mohan *et al*., 2013) with systemic infection occurring in the very early stages of growth while the plant is in the seedling phase (Kosiada, 2011). *S. reiliana* has two formae speciales which are host specific, one infecting maize only while the other infecting sorghum (Martinez *et al*., 1999; Poloni & Schirawski, 2016). Distinct mechanisms in maize and sorghum determine specificity of host (Poloni & Schirawski, 2016; Zuther *et al*., 2012). *S. reiliana* reproduces sexually. One study (Schirawski *et al*., 2005) described two mating type loci of *S. reiliana* as triallelic 'a' and multiallelic 'b'. Each of the 'a' loci is composed of two pheromone genes where only one mating partner specifically recognizes each pheromone.

In severe cases, maize head smut can cause up to 80% yield loss (Frederiksen, 1977). The severity depends on the incidence since no viable kernels are produced by maize plants that are infected (Jackson-Ziems, 2014). This is why maize head smut needs to be urgently and effectively managed.

The Kenyan Seed and Plant Varieties Act (Cap 326) lists *S. reiliana* as a significant pathogen in seed production systems and places a zero tolerance on it during the final inspection of a maize seed crop that is due for seed certification. In August 2016, four counties of Kenya (Nandi, Elgeyo

Marakwet, Trans Nzoia, and Uasin Gishu) reported high incidences of head smut in maize. According to a surveillance (report unpublished) carried out by the Nakuru County Early Warning and Rapid Response Team, led by KEPHIS, in September 2019, Nakuru County reported increased cases of the disease, especially in Njoro sub-county. Most of the affected counties have the favourable climatic conditions (average soil temperatures of 28 °C and moderate to low soil moisture) for seedling infection. Figure 1 represents the situation of infection in one of the visited farmers' fields during the surveillance.

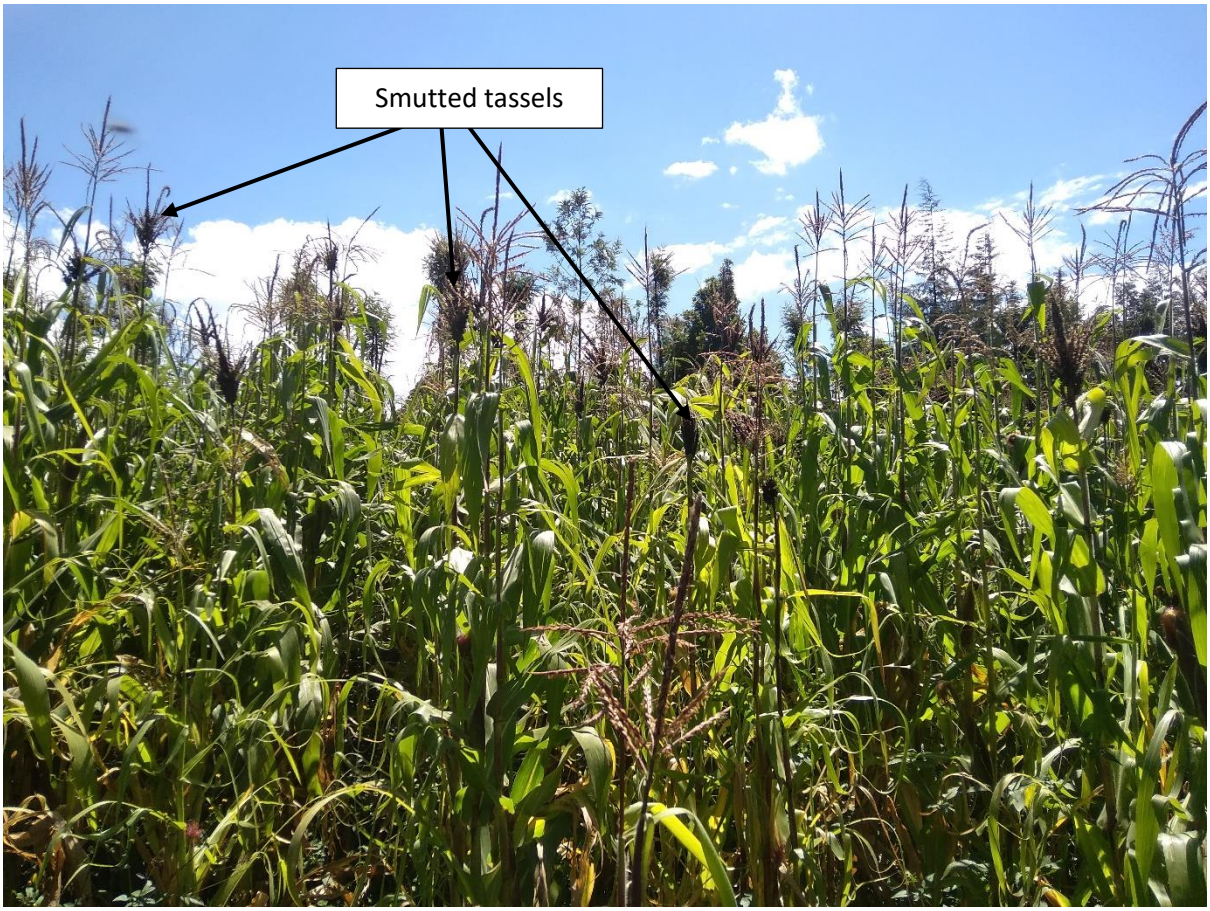

Figure 1: - High incidences of head smut in a maize field in Njoro, Nakuru County (September 10, 2019). All the infected plants are unproductive.

One of the findings of the surveillance was that in the previous cropping seasons, most farmers had fed infected maize debris to cows and the resulting farmyard manure were used in the following season. Such practices may have contributed to the increased disease epidemics in the following

seasons. With maize being Kenya's staple food crop, outbreaks of head smut can spell a food security crisis hence the need to understand the pathogen and disease dynamics. The objective of this review paper is, therefore, to provide a concise outline of head smut pathogenesis, epidemiology, and management options.

Disease cycle and symptoms

Spores from smutted inflorescence are easily dispersed by wind and rain and can remain viable in soil for up to four years (Mohan *et al.*, 2013) hence serving as the main source of inoculum. The germination of the *S. reiliana* spores in soil tend to be high in acidic soils. Under favourable conditions (acidic soils, moderate to low soil moisture, and warm temperatures of between 23-30°C), the spores germinate into infective hyphae which penetrate the roots of seedlings before they reach the six-leaf growth stage (Jackson-Ziems, 2014; Martinez *et al.*, 2002; Mohan *et al.*, 2013). The initial contact between the infective hyphae and the maize root entails the formation of a fungal sheath around the root tissue (Martinez *et al.*, 2000) which facilitates penetration whereas the initial infection of seedling is promoted by delayed rains (Jackson-Ziems, 2014). Following an infection of a maize seedling, the fungus passes through the host cell wall by lysis and mechanical pressure (Martinez *et al.*, 1999). The infective hyphae mainly grow intracellularly and systemically advances through the plant tissues, eventually transforming part or all of the inflorescence tissues into smut galls. Head smut then portrays on maize ears and tassels as galls, which later mature and sporulate to restart another disease cycle.

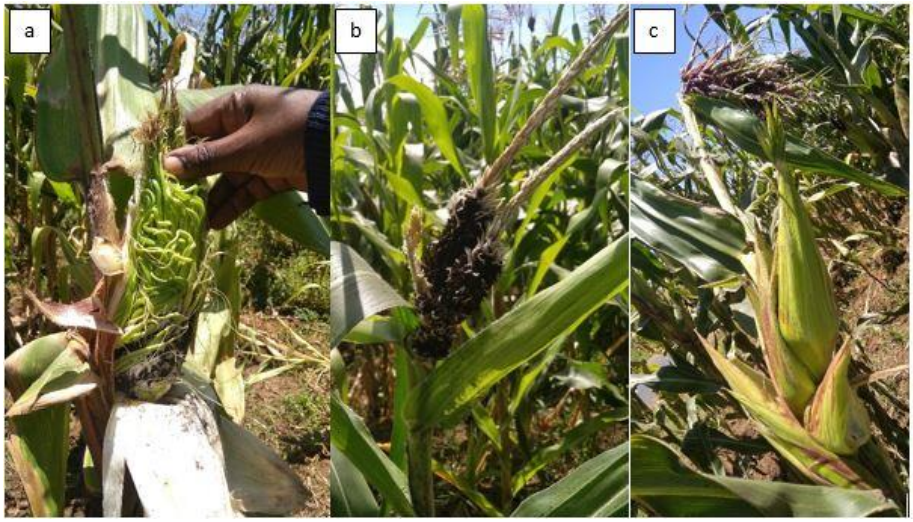

Figure 2. Symptoms of head smut disease on maize; (a) sori with teliospores and finger or wire-like proliferations on maize ear, (b) black mass of fungal teliospores on tassel, (c) malformation of the tassel and ear lacking silks.

Infected ears have rounded or pear-shaped smuts and lack silks whereas infected tassels turn into leafy structures with smutted spikelets (Jackson-Ziems, 2014; Mohan *et al*., 2013). In the early stages of ear development, these galls have a thin membrane that ruptures to expose dry, powdery, dark brown to black masses of teliospores (Mohan *et al*., 2013). The presence of fine, thread-like strands within the galls are remnants of the infected and damaged vascular tissue of the plant (Mohan *et al*., 2013). The symptoms develop because maize inflorescences (ears and tassels) that are infected by *S. reiliana* have increased levels of ROS (reactive oxygen species) and auxin, and misregulation of floral regulatory transcription factors (Ghareeb *et al*., 2011). It is important to note that even though infected, the vegetative tissues of maize do not show any symptoms and may appear healthy (Martinez *et al*., 1999) and thus mycelia in crop residue can serve as inoculum source in the following crop season (Anderson *et al*., 2016).

Disease Management

The increase in maize head smut occurrence and incidences over the years can be attributed to the continuous mono-cropping, use of susceptible varieties, misuse of seed coating agents and change in weather patterns (Li *et al*., 2015). These factors have contributed to the availability of favourable conditions (acidic soils, moderate to low soil moisture, and warm temperatures of between 23-30°C) for infection and disease proliferation. It is important to point out

that there are no curative measures for maize head smut and thus the only available options are preventive as described in the following paragraphs.

The use of resistant varieties - The resistance of maize to *S. reiliana* has received some remarkable attention through studies in the recent years and has been found to be quantitative and mostly additive (Anderson *et al*., 2016). Two quantitative trait loci, qHSR1/qHS2.09/q2.09HR (Konlasuk *et al*., 2015; Li *et al*., 2015; Weng *et al*., 2012) and q5.03HR (Li *et al.*, 2015), have been described as responsible for head smut resistance and is useful in marker assisted resistance for breeding programs. The resistance is conferred by the *ZmWAK* gene which is found in the qHSR1 locus (Konlasuk *et al*., 2015; Zhao *et al*., 2012; Zuo *et al*., 2015). The genome-wide association study (GWAS) by Wang *et al*., (2012) illustrated that resistance to head smut entails complex molecular interactions. In Kenya, two maize varieties, KH500-21A and PAN3M05, have been listed as resistant to head smut whereas WH699 is recorded as tolerant to smut, although the causative agent of the referred smut is not specified. Farmers must use certified seed in order to utilize the variety resistance. Whenever there are high head smut disease incidences in Kenya, farmers always blame the seed they used. This could be true because the cultivar they planted is susceptible to *S. reiliana* but may not have a direct connection with the quality of the seed, as long as the seed is certified.

Fungicide treatment – This is achieved by either seed dressing or by soil drenching, before or after seeding. Because *S. reiliana* is biotrophic in behaviour, systemic fungicide treatment will be effective if their mode of action will inhibit mycelial growth and/or sporogenesis, at least until floral induction occurs (Martinez *et al*., 1999). Some of the active ingredients that are effective in reducing *S. reiliana* infection, even at low application rates, are tebuconazole, fludioxonil, sedaxane (Anderson *et al*., 2015), propiconazole, and fiutriafol combined with imazalil sulphate (Wright *et al*., 2006). Most of these molecules belong to the azole group to which resistance development by fungi is rare and whose mode of action is inhibition of the synthesis of ergosterol hence loss of cell membrane integrity.

Cultural methods – This is best achieved by deep ploughing and rotation with non-host crops to reduce *S. reiliana* inoculum in soil and consequently lower disease incidences. For effective application, the rotation cycle should be at least 2-3 years (Mohan *et al*., 2013) without maize, sorghum, and any other grasses that serve as alternative sources of inoculum. Additionally, rogueing of infected plants and ensuring field

sanitation to rid the field of crop residue will reduce inoculum load and thus a good management strategy.

Conclusion

The maize head smut disease has a worldwide distribution. Disease incidences occur sporadically especially in high altitude areas and is associated with soils with nitrogen deficiency (Mohan *et al*., 2013), which means that it is difficult to predict its occurrence during a cropping season. Whereas head smut disease incidence (percentage of infected plants in a crop field) can reach 80%, disease severity in terms of yield loss is 100% because infected plants are not productive. Infections are favoured by acidic soils, moderate to low soil moisture, and warm temperatures. The most practical management strategy for maize head smut, therefore, integrates the use of resistant/tolerant cultivars, seed treatment and/or drenching of sown rows and observing field hygiene/sanitation. It is important to note that while cultural methods such as crop rotation are useful, they cannot limit disease incidence because spores remain viable for long periods.

Recommendation

Even though maize head smut can cause massive crop losses, there are practical options for its management. Phytosanitary challenges associated with maize head smut can be reduced by ensuring a clear understanding of the disease pathogenesis (biology, infection mechanisms, and symptoms) and epidemiology (spread in time and space).

References

Anderson, S. J., Simmons, H. E., French-Monar, R. D., & Munkvold, G. P. (2016). Susceptibility of maize inbreds and incidence of symptomless infection by the head smut pathogen, Sphacelotheca reiliana. *Plant Health Progress*. *17* (1), pp. 1–5.

Anderson, S. J., Simmons, H. E., & Munkvold, G. P. (2015). Real-time PCR assay for detection of Sphacelotheca reiliana infection in Maize (*Zea mays*) seedlings and evaluation of seed treatment efficacy. *Plant Disease*. 99 (12), pp. 1847–1852.

Frederiksen, R. A. (1977). Head smuts of corn and sorghum. *Proceedings of Corn Sorghum Resesearch Conf*erence. 32, pp. 89–104.

Ghareeb, H., Becker, A., Iven, T., Feussner, I., & Schirawski, J. (2011). Sporisorium reilianum infection changes inflorescence and branching architectures of maize. *Plant Physiology*. 156 (4), pp. 2037–2052.

Jackson-Ziems, T. A. (2014). Smut Diseases of Corn. *NebGuide*.

http://extensionpublications.unl.edu/assets/pdf/g2223.pdf

Konlasuk, S., Xing, Y., Zhang, N., Zuo, W., Zhang, B., Tan, G., & Xu, M. (2015). ZmWAK, a quantitative resistance gene to head smut in maize, improves yield performance by reducing the endophytic pathogen Sporisorium reiliana. *Molecular Breeding*. *35* (8), pp. 174.

Kosiada, T. (2011). In vitro influence of selected fungicides on Sphacelotheca reiliana and Ustilago maydis. *Journal of Plant Protection Research*. *51* (4), pp. 342–348.

Li, Y., Wu, X., Jaqueth, J., Zhang, D., Cui, D., Li, C., Hu, G., Dong, H., Song, Y., Shi, Y., Wang, T., Li, B., & Li, Y. (2015). The Identification of Two Head Smut Resistance-Related QTL in Maize by the Joint Approach of Linkage Mapping and Association Analysis. *Plos One*. 10 (12), e0145549.

Martinez, C., Jauneau, A., Roux, C., Savy, C., & Dargent, R. (2000). Early infection of maize roots by Sporisorium reilianum f. sp. zeae. *Protoplasma*. 213(1-2), pp. 83–92.

Martinez, C., Roux, C., & Dargent, R. (1999). Biotrophic development of Sporisorium reilianum f. sp. zeae in vegetative shoot apex of maize. *Phytopathology*. 89 (3), pp. 247–253.

Martinez, C., Roux, C., Jauneau, A., & Dargent, R. (2002). The Biological Cycle of Sporisorium reilianum f.sp. zeae: An Overview Using Microscopy. *Mycologia*. 94, pp. 505–514.

Mohan, S. K., Hamm, P. B., Clough, G. H., & Toit, L. J. (2013). *Corn Smuts*.

Poloni, A., & Schirawski, J. (2016). Host specificity in Sporisorium reilianum is determined by distinct mechanisms in maize and sorghum. *Molecular Plant Pathology*. 17 (5), pp. 741–754.

Schirawski, J., Heinze, B., Wagenknecht, M., & Kahmann, R. (2005). Mating type loci of Sporisorium reilianum: Novel pattern with three a and multiple b specificities. *Eukaryotic Cell*. 4 (8), pp. 1317–1327.

Wang, M., Yan, J., Zhao, J., Song, W., Zhang, X., Xiao, Y., & Zheng, Y. (2012). Genome-wide association study (GWAS) of resistance to head smut in maize. *Plant Science*. 196, pp. 125–131.

Weng, J., Liu, X., Wang, Z., Wang, J., Zhang, L., Hao, Z., Xie, C., Li, M., Zhang, D., Bai, L., Liu, C., Zhang, S., & Li, X. (2012). Molecular mapping of the major resistance quantitative trait locus qHS2.09

with simple sequence repeat and single nucleotide polymorphism markers in Maize. *Phytopathology*. 102 (7), pp. 692–699.

Wright, P. J., Fullerton, R. A., & Koolaard, J. P. (2006). Fungicide control of head smut (Sporisorium reilianum) of sweetcorn (Zea mays). *New Zealand Journal of Crop and Horticultural Science*. 34 (1), pp. 23–26.

Zhao, X., Tan, G., Xing, Y., Wei, L., Chao, Q., Zuo, W., Lübberstedt, T., & Xu, M. (2012). Marker-assisted introgression of qHSR1 to improve maize resistance to head smut. *Molecular Breeding*, 30(2), pp. 1077–1088.

Zuo, W., Chao, Q., Zhang, N., Ye, J., Tan, G., Li, B., Xing, Y., Zhang, B., Liu, H., Fengler, K. A., Zhao, J., Zhao, X., Chen, Y., Lai, J., Yan, J., & Xu, M. (2015). A maize wall-associated kinase confers quantitative resistance to head smut. *Nature Genetics*. 47 (2), pp. 151–157.

Zuther, K., Kahnt, J., Utermark, J., Imkampe, J., Uhse, S., & Schirawski, J. (2012). Host specificity of *Sporisorium reilianum* is tightly linked to generation of the phytoalexin luteolinidin by sorghum bicolor. *Molecular Plant-Microbe Interactions*, 25 (9), pp. 1230–1237.

New scale insect country records for Kenya (Hemiptera: Coccomorpha) from old samples in insect collections

Watson G. W. [1*], Ouvrard D.[2], Kasina M.[3], Achieng J.C.[4], Githae M.M.[5], Mulwa J.[3], Kinuthia W.[4], Macharia I.[6], Heya H.M[6], Polaszek A.[1]

[1]Department of Life Sciences, the Natural History Museum, Cromwell Road, London SW7 5BD, U.K.

[2]Anses, Laboratoire de la Santé des Végétaux, 755 avenue du campus Agropolis CS 30016, FR-34988 Montferrier-sur-Lez Cedex, France.

[3]Kenya Agriculture and Livestock Research Organisation, P.O. Box 57811-00200, Nairobi, Kenya.

[4]National Museums of Kenya, P.O. Box 40658-00100, Nairobi, Kenya.

[5]School of Biological Sciences, College of Biological and Physical Sciences, University of Nairobi, P.O. Box 30197-00100, Nairobi, Kenya.

[6]Kenya Plant Health Inspectorate Service, P.O. Box 49592-00100, Nairobi, Kenya.

*Corresponding author: Email: gillian.watson@nhm.ac.uk

Abstract

Scale insects (*Hemiptera: Sternorrhyncha: Coccomorpha*) are some of the least understood insects, particularly in agriculture, even though they can cause high crop losses. Due to their small size and cryptic habits they are rarely noticed at the onset of an infestation. In Kenya, efforts have been initiated to understand these pests better. Scale insects from Kenya, found in samples between 13 and 107 years old, were studied in the insect collections of the Natural History Museum, London, U.K. and the Kenya Agriculture and Livestock Research Organisation, Nairobi, Kenya. The study identified 51 new country records of scale insects including one new continental record for Africa, *Ferrisia dasylirii* (Cockerell) (*Pseudococcidae*). Of the new records, 35 species (68.6%) are native to Africa and 16 (31.4%) have been introduced from elsewhere. Six of the 51 species (11.8%) are pests in Kenya today. Amongst the introduced species, at least one (*Aonidiella comperei* McKenzie) could cause plant quarantine issues in trade, and four (25.0%) are pests, more than four times the frequency of pests amongst the African species (5.7%). The remaining 45 species have been present in Kenya for at least 13 years and many have not been collected again since the original samples, suggesting that either they have not survived or they

are rare because they are under good natural control. Most of the introduced species listed (75.0%) have not caused economic problems in Kenya to date, so it is thought unlikely that they will do so in the future.

Key words: Scale insects, introduced species, native species

Introduction

Specimens in old insect collections provide evidence of the species that were established in a country or region at a specific date. Review of historic insect collections can reveal new country records and early distribution records of species that have never been recorded in the literature: for example, study of the Wirjati collection in Indonesia (containing material collected 1916-1960) revealed three new Indonesian country records of mealybug species (Sartiami *et al.*, 2016), and study of Takahashi slides from Malaysia (containing material collected 1943-1944) revealed the earliest known Malaysian records of two mealybug species (Sartiami *et al.*, 2017). Such baseline information provides clarification of which non-native species have been accidentally introduced in more recent times (Sartiami *et al.*, 2016, 2017).

As part of the Darwin Initiative-funded project 25-032: "Agriculture and biodiversity: addressing scale insect threats in Kenya", old scale insect slide mounts in two important insect collections were studied. The collection at the Kenya Agriculture and Livestock Research Organisation (KALRO) National Agricultural Research Laboratory (KALRO-NARL) at Kabete, Nairobi, Kenya contains samples mostly collected between 1920 and 1970. Giovani De Lotto worked at this laboratory between 1950 and 1963 (Ben-Dov & Russo, 1991) and greatly enlarged the scale insect slide collection; the 3,597 slides were re-curated and databased during this work. The collection at the Natural History Museum, London, U.K. (NHMUK) contains samples mostly collected between 1910 and 2000, including the type material of many species. Study of Kenyan material in these collections has revealed multiple

unpublished first country records for Kenya and one new continental record for Africa. Here we document these new records, and provide data on recent collections of some of the species as evidence that they are still present in the country.

Materials and methods

Old samples of scale insects on slide-mounts in the KALRO-NARL and NHMUK collections were studied using a Zeiss Axiophot compound light microscope with phase contrast illumination and magnifications of 25x-800x. Species identities were checked using the most recently published identification keys available (listed in Table 1), together with unpublished keys being developed by the first author (GW) as part of the Darwin Initiative project. Specimens were also compared to type material when it was available. To assess which of the species identified had never been recorded from Kenya in the literature, a list compiled from the old collections was compared with a list searched online from the ScaleNet database (García Morales *et al*., 2016), which is based on the literature, particularly De Lotto's publications.

For each species, collection data are given for the historic samples first, followed by data from recently collected material where available. Species likely to have been introduced from outside continental Africa are marked with an asterisk (*****); those known to be native to Africa are marked with a dagger (**†**). New host-plant records are indicated by **[N]**. Collectors' names are provided where known. Recent samples from the present Darwin Initiative project are represented by slides that will be deposited in both the KALRO-NARL and NHMUK collections. Some recent material will be deposited also in the collections at the National Museums of Kenya, Nairobi, Kenya (NMK); University of Nairobi, Nairobi, Kenya (UoN); and Kenya Plant Health Inspectorate Service, Muguga, Kenya (KEPHIS).

Table 1. References used for identification of scale insect slide mounts from Kenya in the collections at KALRO and NHMUK. The references are in chronological order so that it is evident which is the most up-to-date.

Scale insect family	Identification aid resources
Aclerdidae	Howell & Williams (1976), **Ben-Dov (1977), Gill** (1993), Hodgson & Millar (2002), Miller et al. (2014)
Asterolecaniidae	Russell (1941), **Giliomee** & **Munting (1968),** Howell & Williams (1976), **Kosztarab** & **Kozár** (1988), Williams & Watson (1990), Gill (1993), Kosztarab (1996), **Stumpf & Lambdin (2001, 2006),** Giliomee & Kozár (2008), Miller et al. (2014)
Cerococcidae	Howell & Williams (1976), **Lambdin** & **Kosztarab** (1977), Williams & Watson (1990), Gill (1993), Kosztarab (1996), Miller et al. (2014), Hodgson & Williams (2016)
Coccidae	Hall (1925), **Laing** (1929), De Lotto (1954a, 1955, 1956a, 1956b, 1957a, 1957b, 1958a, 1958b, 1959a, 1960, 1961b, 1962, 1963, 1964b, 1965, 1966a, 1966b, 1966c, 1967b, 1969a, 1970, 1971, 1974a, 1975b, 1978, 1979), Hodgson (1967a, 1994), Howell & Williams (1976), Nakahara & Gill (1985), **Kosztarab** & **Kozár (**1988), Williams & Watson (1990), Gill (1988, 1993), **Kosztarab** (1996), Hodgson & Henderson (2000), Miller et al. (2014), Łagowska & Hodgson (2019)
Conchaspididae	Mamet (1954), Hodgson (1967b), Howell & Williams (1976), **Ben-Dov (1981),** Williams & Watson (1990), **Gill** (1993), Miller et al. (2014)
Dactylopiidae	**Karny** (1972), De Lotto (1974b), Howell & Williams (1976), Williams & Watson (1990), Gill (1993), Miller et al. (2014)
Diaspididae	Ferris (1937, 1938, 1941, 1942), Hall (1946), B**alachowsky** (1956, 1958), Mamet (1958), Williams (1963), Howell & Williams (1976), Tang (1986), **Kosztarab** & **Kozár** (1988), **Ben-Dov** (1988), Williams & Watson (1988a), Danzig (1993), **Gill** (1993, 1997), Kosztarab (1996), Watson (2002), **Miller** & **Davidson (2005),** Miller et al. (2014), Schneider et al. (2019)
Eriococcidae	Howell & Williams (1976), **Kosztarab** & **Kozár** (1988), Williams & Watson (1990), Gill (1993), Kosztarab (1996), **Kozár** et al. (2013), Miller et al. (2014)
Halimococcidae	Stickney (1934), Williams & Watson (1990), Miller et al. (2014)
Kermesidae	Howell & Williams (1976), **Bullington & Kosztarab (1985), Gill** (1993), Miller et al. (2014)
Kerriidae	Howell & Williams (1976), **Varshney (1984, 1990),** Williams & Watson (1990), Gill (1993), Kondo et al. (2011), Miller et al. (2014)

Kuwaniidae	Morrison (1928), De Lotto (1959b), **Gill** (1993), Hodgson & Foldi (2006), Miller et al. (2014)
Lecanodiaspididae	De Lotto (1955), Hodgson (1973), Howell & Williams (1976), Williams & Watson (1990), Gill (1993), Miller et al. (2014)

Table 1: Continued from previus page

Scale insect family	**Identification aid resources**
Margarodidae	Morrison (1928), de Klerk (1982a, 1982b, 1983), Foldi (2005a), Vahedi & Hodgson (2007)
Matsucoccidae	Boratynsky (1952a, 1952b), Gill (1993), Foldi (2005b), Miller et al. (2014)
Micrococcidae	Miller & Williams (1995), Miller et al. (2014)
Monophlebidae	Morrison (1928), De Lotto (1959b), Gill (1993), Kosztarab (1996), **Kosztarab** & **Kozár** (1988), **Unruh & Gullan (2008),** Foldi (2010), Miller et al. (2014)
Ortheziidae	Morrison (1925, 1952), Howell & Williams (1976), **Kosztarab** & **Kozár** (1988), Williams & Watson (1990), Gill (1993), Kosztarab (1996), **Kozár & Konczné Benedicty** (2000, 2001), **Kozár et al.** (2002), Miller & Koz**ár** (2002), **Kozár** (2004), Kondo et al. (2013), Miller et al. (2014)
Phoenicococcidae	Howell & Williams (1976), **Gill** (1993), Miller et al. (2014)
Pseudococcidae	James (1935), Ferris (1950, 1953), **De Lotto** (1954b, 1955, 1957c, 1958c, 1961a, 1964a, 1964c, 1967a, 1969a, 1969b, 1974c, 1975a), Ezzat & McConnell (1956), Williams (1958a, 1958b, 1961, 1970, 1986, 1996, 1998, 2001, 2004), Ezzat (1960, 1962), **Balachowsky & Matile-Ferrero** (1966), McKenzie (1967), **Matile-Ferrero** (1970), Howell & Williams (1976), **Cox** (1987, 1989), **Kosztarab** & **Kozár** (1988), Williams & Watson (1988b), Watson & Cox (1990), **Williams** & **Granara de Willink** (1992), Kosztarab (1996), Williams & Matile-Ferrero (1999, 2005), **Granara de Willink & Szumik (**2007), **Schneider** & **LaPolla (2011),** Miller & Giliomee (2011), Kaydan & Gullan (2012), Miller et al. (2014)
Putoidae	Gill (1993), Miller et al. (2014)
Rhizoecidae	Hambleton (1976), Kosztarab (1996), **Kozár & Konczné Benedicty** (2007), Miller et al. (2014)
Stictococcidae	Richard (1976), Williams et al. (2010), Miller et al. (2014)

Results

The scale insect species below were found in the KALRO-NARL, NHMUK and NMK collections. According to the ScaleNet database (García Morales *et al*., 2016) they have not been recorded from Kenya in the literature before.

Family Coccidae (13 species)

†A wax scale, *Ceroplastes ficus* Newstead: Kenya, Nairobi, National Museums of Kenya, Biodiversity Centre, on shrubs and trees, coll. G.W. Watson, 5.x.1996 (NHMUK).

†A wax scale, *Ceroplastes quadrilineatus* (Newstead): Kenya, Nairobi, on *Ficus* sp., coll. W.J. Hall, 26.iii.1949 (NHMUK).

†A wax scale, *Ceroplastes ?sinoiae* (Hall): Kenya, on *Coffea* sp., coll. H.C. James, [probably in the 1930s] (NHMUK).

†A soft scale, *Ceroplastodes zavattarii* Belio: Kenya: Yatta Plateau, Katangi, on ?Malvaceae, 6.viii.1977, coll. J.H. Martin (NHMUK); Malindi, on *Hoslundia opposita*[N], 20.v.1988, coll. J.H. Martin (NHMUK).

†A soft scale, *Coccus* sp. near ***cajani* (Newstead):** Kenya: Kiambu Co., Kikuyu, on *Acacia mearnsii*, v.1974 (NHMUK); Nairobi County, Waithaka, on *Cajanus* sp., coll. Alice, 19.vi.1977 (NHMUK).

†A soft scale, *Coccus milanjianus* Hodgson: Kenya, Kikuyu, on *Elaeodendron (=Cassine) buchananii*[N], v.1974 (NHMUK).

†A soft scale, *Inglisia theobromae* Newstead: Kenya: Limuru, on *Pelargonium*[N] sp., 16.i.1963, coll. G. De Lotto (NHMUK); Kikuyu, on *Acacia mearnsii*[N], v.1964 (NHMUK); Kikuyu, on *Abutilon* [N] sp., v. 1974 (NHMUK); Nairobi, N.A.L., on cotton, 4.vii.1977, coll. P. Nderi (NHMUK).

†A soft scale, *Lagosinia vayssierei* (Castel-Branco): Kenya, Kisumu, on *Grewia*[N] sp., 23.xi.1953 (NHMUK).

†A soft scale, *Pulvinaria merwei* Joubert: Kenya, Kiambu Co., Ruiru, on *Ipomoea batatas*, 20.viii.1957 (NHMUK).

***Cottony citrus scale, *Pulvinaria polygonata* (Cockerell):** Kenya: Mombasa, on *Mangifera indica*, 10.viii.1958 (NHMUK); Malindi, on *Mangifera indica*, 25.iii.1961, coll. J.F. Graham (NHMUK). **Recent:** Kilifi Co., Mtwapa, KALRO orchard, S 3º 56' 12", E 39º 44' 32", 10 m alt, on *Mangifera indica*, 10.vii.2019, coll. Extension officers (NHMUK, KALRO-NARL); Kwale Co., Lunga Lunga, S 4º 33', E 39º 52', 52 m alt., on *Citrus sinensis*, 28.viii.2019, Michael Githae (UoN). The species originated in southern Asia and is known to be a pest of citrus. In the coastal counties of Kenya it is a pest on citrus, causing serious honeydew and sooty mould fouling of leaves and fruits, impacting fruit quality.

***Urbicola soft scale, *Pulvinaria urbicola* (Cockerell):** Kenya: Kwale, on roots of *Capsicum* sp., 11.vi.1956 (NHMUK); Mombasa, on roots of *Solanum tuberosum*[N], 8.xi.1956 (NHMUK); Mombasa, on *Capsicum* sp., 10.vii.1957 (NHMUK). The species is of unknown origin but probably is not native to Africa; it is polyphagous and can cause significant defoliation of woody hosts. *Pulvinaria urbicola* has a history of damaging native forests on small islands in the Pacific Ocean, particularly if ants are present to attend it (Smith et al., 2004; Peck *et al*., 2014; Neumann *et al*., 2014, 2016).

***Iceplant scale, *Pulvinariella mesembryanthemi* (Vallot):** Kenya, Nairobi, on *Mesembryanthemum* sp., 6.ix.1951 (NHMUK). The insect is native to South Africa; in California in the absence of its natural enemies, it can kill large areas of highway ice plant (Aizoaceae: *Carpobrotus edulis* (L.) N.E. Br.) ground cover beside roads (Gill, 1993). No such damage has been recorded in Kenya in the literature but ice plant is not widely used in amenity plantings there, possibly because the scale makes them unsightly.

†Giant soft scale, *Pulvinarisca inopheron* (Laing): Kenya: Chogoria, on *Cajanus indicus*[N],

4.x.1937, coll. A.R.M. (NHMUK); Nairobi, on *Croton manostachys*, 16.ix.1951 (NHMUK); Nairobi, on *Chaetacme aristata*[N], 29.x.1953 (NHMUK); Nairobi, on *Salvia* sp., 7.vi.1954 (NHMUK). **Recent:** Kenya, Central Province, Muguga, on small farm, on *Caliandra carobensis*[N], 15.vii.2018, coll. G. Opondo (NHMUK, KALRO-NARL). In Kenya, this species forms very heavy infestations on *Calliandra* grown for animal fodder. Each adult female may be up to two cm long and produces a conspicuous large white ovisac.

Family Diaspididae (23species)

†**An armoured scale, *Africaspis communis* (Hall):** Kenya, Nairobi, on fig, 26.iii.1949, coll. W.J. Hall; Eldoma Ravine, on stems of shrub, 25.ii.1970, coll. E.S. Brown (NHMUK).

***False yellow scale, *Aonidiella comperei* McKenzie:** Kenya: Pemba Island, host not noted, coll. Anderson, pre-1962. **Recent:** Kilifi County, Mtwapa, KALRO orchard, S 3° 56' 12", E 39° 44' 32", 10 m alt., on *Citrus* sp. leaf undersides, coll. Extension Officers, 10.vii.2019, 2 samples; Mtwapa, S 3.93717°, E 39.7424, 169 m alt., on *Citrus sinensis*, coll. M.M. Githae, 12.xii.2019 and 13.xii.2019; Malindi, S 3.27643°, E 40.01251°, 139 m alt., on *C. sinensis*, coll. M.M. Githae, 12.x.2019; Malindi, S 3.27442°, E 40.04494°, 166 m alt., on *C. sinensis*, coll. M.M. Githae, 12.x.2019; Kwale County: Matunga, S 4.27996°, E 39.56794°, 68 m alt., on *C. sinensis*, coll. M.M. Githae, 25.viii.2019; Ukunda, S 4.28601°, E 39.5284°, 63 m alt., on *C. sinensis*, coll. M.M. Githae, 14.xii.2019; Njego, S 4.65341°, E 39.1998°, 53 m alt., on *C. sinensis*, coll. M.M. Githae, 16.xii.2019; Botela, S 4.5809°, E 39.10918°, 45 m alt., on *C. limon*, coll. M.M. Githae, 16.xii.2019. Worldwide, this species has been recorded on host-plants in 12 families including species of *Citrus*, *Annona* and other fruit trees, *Cocos nucifera*, *Carica*

papaya, Musa sp. and *Vitis vinifera* and may have a wider host range (Williams & Watson, 1988; Watson, 2002; García Morales *et al*., 2020). *Aonidiella comperei* secretes a circular, flat, yellow-brown scale cover that is often closely attached to the insect beneath. The anterior part of the adult female expands with maturity to become kidney-shaped, forming postero-lateral lobes that lie alongside the smaller abdomen, and becomes hard and brown (like *A. aurantiae* (Maskell)). Mounted on a microscope slide, the adult female *A. comperei* has the pygidial venter with one small group of perivulvar pores on either side of the vulva, and lacks prevulvar scleroses and apophyses (whereas *A. aurantiae* lacks perivulvar pores but has paired prevulvar scleroses and apophyses). *Aonidiella comperei* probably originated from tropical Asia, but has been spread to other continents through the movement of infested live plant material. Balachowsky (1958) recorded it previously from Tanzania and remarked that it has a preference for citrus. In Kenya it was found on citrus and may have the potential to become a citrus pest. Its presence on exported fruit could cause plant quarantine issues in trade.

***Aglaonema scale, *Aspidiotus ?excisus* Green:** Kenya, Siaya County, on *Lantana camara*, coll. Prof. Odihambo, 6.iv.1990. These specimens differ from typical *A. excisus* by having median lobes with basal scleroses. **Recent:** Mombasa Co., Likoni, S 4.0948°, E 39.64874°, 142 ft alt., on *Capsicum frutescens* leaf, coll. W. Kinuthia, J. Achieng, 20.ii.2020. CHECK for scleroses Aglaonema scale has been intercepted from Africa (Mozambique) at plant quarantine in South Korea (Suh, 2016). The species is considered to be a pest of ornamental plants (Davidson & Miller, 1990).

†Fried egg scale, *Aspidiotus ?ruandensis* Balachowsky: Kenya, Kericho County: Kericho,

on *Camellia sinensis*[N], 2.iii.1967; Nakuru, on *Camellia sinensis*, coll. G.W. Oloo, 30.x.1972; Kericho, on *Ca. sinensis*, coll. V. Sudoi, iv.1986; Kiambu County: Kiambu, on *Coffea arabica*[N], coll. J.W. Waikwe, 10.vii.1978; Kiambu, Makana Estate, on *Co. arabica*, coll. R.H. Markham, 18.iv.1983; Ruiru, on *Co. arabica* leaves, ?1991; Ruiru, Ruara Estate, on *Co. arabica*, coll. G.W. Watson, viii.1993; Ruiru, Ruara Estate, 1500 m asl,, on *Co. arabica*, coll. T.J. Crowe, 29.iv.1994; Kwale County: Diani forest, on *Diospyros squarosa*[N], 17.x.1983 (NHMUK). This is a native African species; there is some uncertainty about its identity because when *A. ruandensis* was described from Rwanda (Balachowsky, 1955), the scale cover was described as light grey-brown with a yellowish cast, and with dark exuviae; whereas specimens in Kenya have white scale covers with yellow exuviae. However, the morphology supports the material representing a single species with a variable number of submarginal prepygidial macroducts. Molecular analysis of material representing these two scale-cover colours would resolve whether there is more than one species involved. Fried egg scale may have been introduced to Kenya from further west; it has been present in the country since at least 1967. It occurs on shade trees in Kenyan beverage crop plantations; in dry conditions it spreads onto the foliage of coffee bushes, sometimes becoming a pest (T.J. Crowe, pers. comm. 1994). The sample data above and in García Morales et al. (2016) indicate that it is relatively polyphagous on tree foliage, including fruit trees.

Cactus scale, *Diaspis echinocacti (Bouché): Kenya, Nairobi, on *Diospyros abyssinica*[N], coll. G. De Lotto, 28.iii.1956 (KALRO-NARL).

†Mango scale, *Duplachionaspis natalensis* (Maskell): Kenya, Machakos, on *Panicum coloratum*[N], 9.v.1950 (KALRO-NARL).

†**An armoured scale, *Hemiberlesia mammillaris* (Lindinger):** Kenya, Magadi, on *Aloe* sp., 29.vii.1956 (KALRO-NARL).

† **An armoured scale, *Hulaspis dombeyae* (Hall):** Kenya, Kikuyu, on Dombeya goetzenii, v.1974 (NHMUK).

†**An armoured scale, *Lindingaspis musae* (Laing):** Kenya, Ruiru, on *Syzygium cordatum*[N], 31.x.1953 (KALRO-NARL).

†**An armoured scale, *Morganella conspicua* (Brain):** Kenya, Kajiado, on *Commiphora*[N] sp., 10.vi.1956 (KALRO-NARL).

†**An armoured scale, *Morganella spinigera* (Lindinger):** Kenya, Nairobi, on *Gelonium procerum*[N], 18.iv.1953 (KALRO-NARL).

†**Reed scale, *Odonaspis phragmitis* Hall:** Kenya, Ruiru, on roots of *Paspalum scrobiculatum*[N], 7.ii.1956 (KALRO-NARL).

Bermuda grass scale, *Odonaspis ruthae Kotinsky: Kenya, Nairobi, on roots of *Rhychelytrum repens*[N], coll. G. De Lotto, 20.iv.1954 (KALRO-NARL). This is the most polyphagous species in *Odonaspis*, and has been recorded damaging lawn grass in Egypt and Israel, and forage and turf grasses in the southern U.S.A. and Chile (Watson, 2002).

***Paragrass scale, *Odonaspis sacchariculis* (Zehntner)**: Kenya, on roots of lemon grass, coll. F.S. Notley, 28.v.1935; British East Africa, Kenya, on roots of lemon grass (*Andropogon* sp.), no date; Ramisi, on sugarcane, 8.xi.1971, coll. G.W. Oloo (NHMUK). The species can be a pest of sugarcane in India.

***Parlatoria date scale, *Parlatoria blanchardi* (Targioni Tozzetti):** Kenya, Turkana, on date palm, coll. Smead, 8.xi.1971 (NHMUK). The scale is not native to Africa but probably originated in the Middle East; it is a well-known pest of date palms. There are no recent records, probably due to lack of sampling in northern Kenya.

***Boxwood scale, *Pinnaspis buxi* (Bouché):** Kenya, Nairobi, Scott Agricultural Laboratory, on *Bauhinia purpurea*, 26.ii.1951

(NHMUK); Nairobi, on *Musa ensete*[N], 25.ix.1951 (KALRO-NARL).

†**An armoured scale, *Pseudaonidia baikeae* Newstead:** Kenya: Nairobi, on *Chaetachme aristata*[N], 24.vi.1951 (KALRO-NARL); Thika, on *Rawstonia usambaruensis*[N], 24.viii.1952 (KALRO-NARL).

†**An armoured scale, *Pseudotargionia glandulosa* (Newstead):** Kenya, Magadi, on *Acacia senegal*, coll. R.W. Le Pelley, 6.viii.1951 (NHMUK); Magadi, on *Acacia* sp., coll. G. De Lotto, 29.vii.1956 (KALRO-NARL).

†**An armoured scale, *Rolaspis polypora* Munting:** Kenya, Sultan Hamud, 19.viii.1956 (KALRO-NARL).

†**An armoured scale, *Rolaspis syrinx* Williams:** Kenya, Naivasha, on *Acokanthera schimperi*[N], 1.i.1953 (NHMUK).

***Lychee bark scale, *Rutherfordia major* (Cockerell):** Kenya, Nairobi, on *Ehretia sylvatica*[N], 4.xi.1951, coll. G. de Lotto (KALRO-NARL). **Recent:** Kenya, Nairobi, National Museums of Kenya, on unknown plant, 7.x.2019, coll. J. Achieng (NMK). Ebeling (1959) recorded this species as a pest of lychee (*Litchi chinensis*, Sapindaceae) in Florida. There is no record of it causing damage in Kenya.

†**An armoured scale, *Sclopetaspis ?malawica* Munting:** Kenya, Kikuyu, on 'Rwegethia' [=*Zehneria scabra*], v.1974 (NHMUK).

†**An armoured scale, *Umbaspis spatulata* (Hall):** Kenya, Nairobi, on *Tulia simplicifolia*[N], 10.iv.1953 (KALRO-NARL).

Family Eriococcidae (1 species)

†**A felt scale, *Acanthococcus ?etbaicus* (De Lotto):** Kenya, N 01º 07', E 35º 51', on *Acacia nilotica,* 1.i.1987 (NHMUK).

Family Kerriidae (1 species)

†**A lac insect, *Tachardina ?brachystegiae* (Hall):** Kenya, Buchuma N.W. of Mombasa, on twigs of *Acacia nilotica*, 16.v.1986 (NHMUK).

Family Lecanodiaspididae (1 species)

†A false pit scale, *Lecanodiaspis mimosae* (Maskell): Kenya, Naivasha, on *Acacia xanthaphloea*[N], 9.viii.1970, coll. H. Schmutterer (NHMUK).

Family Monophlebidae (2 species)

†Spiny monophlebid, *Aspidoproctus ?tricornis* (Newstead): Kenya, Marigat, Loruk, on *Acacia nilotica* ssp. *subalata*, 6.vi.1990, coll. J. Marohasy (NHMUK).

†A monophlebid, *Pseudaspidoproctus fulleri* (Cockerell): Kenya, Chiromo, on grass, 2.ii.1971, coll. H. Schmutterer (NHMUK).

Family Pseudococcidae (10 species)

†Acacia mealybug, *Acaciacoccus hockingi* Williams & Matile-Ferrero: Kenya, Lake Naivasha, lakeside, on whistling thorn *Acacia*, 27.v.1988, coll. J.H. Martin (NHMUK).

†A mealybug, Delottococcus phylicus (De Lotto): Kenya, Naromoru, on Asteraceae (=Compositae), 26.viii.1977, coll. J.H. Martin (NHMUK).

†Podocarpus mealybug, Eastia jouberti De Lotto: Kenya, Nyeri Province, on *Podocarpus ?gracilea*, 27.ix.1982 (NHMUK).

***A mealybug, Ferrisia dasylirii (Cockerell):** Kenya: Kiambu, on coffee, iv.1926, coll. T.W. Kirkpatrick (NHMUK); Central Province, Mitungu, 1,500 m alt., on *Tephrosia*[N] sp., viii.2007, coll. Dudutech 030907F (NHMUK). **New continental record.** The mealybug is of South American origin. It is polyphagous and was only identified recently because Kaydan & Gullan's (2012) revision of the genus provided an identification key. Like *F. virgata* (Cockerell), also present in Kenya, heavy infestations can cause honeydew and sooty mould fouling of foliage.

***A mealybug, *Ferrisia malvastra* (McDaniel):** Kenya, Namanga, on *Abutilon mauritiense*[N], 21.i.1961, coll. G. de Lotto (KALRO-NARL). The species is of

South American origin. Like the related species *F. virgata* (Cockerell), also present in Kenya, heavy infestations can cause honeydew and sooty mould fouling of foliage.

†**A mealybug, *Heliococcus* sp.** near ***osborni* Sanders:** Kenya, Mt Kenya, on grass, coll. H. Schmutterer, 14.ii.1971 (NHMUK).

***Hall's mealybug, *Planococcus halli* Ezzat & McConnell:** Kenya, Nanyuki, on *Pistacia*[N] sp., 30.ix.1977, coll. O. Barton (NHMUK). The origin of this polyphagous species is unknown; hosts include yams, groundnuts, cassava, pigeon pea, sugarcane, coffee and citrus. The mealybug is often intercepted at plant quarantine inspection in the U.S.A. on yam tubers from Nigeria (Cox, 1989).

***Passionvine mealybug, *Planococcus minor* (Maskell):** Kenya: Kikuyu, on *Jacaranda mimosifolia*[N], v.1974 (NHMUK); Malindi, Msabaha Ag. Res. Station, on potato in storage, 5.iii.1987, coll. B.L. Parker (NHMUK); Nairobi Arboretum, on buds of *Callistemon*[N] sp. with ants, 31.viii.1988, coll. J.H. Martin (NHMUK); Nairobi Arboretum, on *Clausena anisata*[N], 2.v.1988, coll. J.H. Martin (NHMUK). **Recent:** Kilifi Co., Mtwapa, KALRO orchard, S 3º 56' 11", E 39º 44' 28", 14 m alt, on *Psidium guajava*, 10.vii.2019, coll. Extension officers (NHMUK, KALRO-NARL). *Planococcus minor* is possibly of Pacific origin and is highly polyphagous, attacking many economically important plants; it is sometimes ant attended. The species is a fairly common pest on crops including citrus in the coastal counties of Kenya, where it occurs much more frequently than *P. citri* (Risso).

***Obscure mealybug, *Pseudococcus viburni* (Signoret):** NHMUK: Kiambu Co., on *Datura*[N] sp., coll. R.H. Le Pelley, 20.x.1929 (NHMUK). Of unknown origin, this species is highly polyphagous and there are many literature records of it being a pest on tree, field and glasshouse crops (García Morales

et al. 2016). No recent samples have been seen from Kenya.

†**Short-legged mealybug, *Vryburgia brevicruris* (McKenzie):** Kenya, on roots of *Bidens pilosa*[N], 5.iv.1930, coll. H.C. James (NHMUK).

Discussion

The 51 new country records of scale insect species for Kenya recorded above are based on samples between 13 and 107 years old. While 35 (68.6%) of these species are native to Africa, 16 (31.4%) have been introduced accidentally from outside the continent (Table 2). Most of the introduced species had been recorded previously from some other part of Africa but there is one new continental record: Ferrisia dasylirii (Pseudococcidae).

Table 2. A breakdown of taxonomic, geographic origin and economic data for the new Kenya species records found in old samples.

Families (in size order)	No. genera	No. spp.	No. spp. of African origin	No. spp. from outside Africa	Pests of African origin	Introduced pest species	No. species with pest potential
Diaspididae	18	23	15	8	1	2	3
Pseudococcidae	8	10	5	5	0	1	3
Coccidae	8	13	10	3	1	1	1
Monophlebidae	2	2	2	0	0	0	0
Eriococcidae	1	1	1	0	0	0	0
Lecanodiaspididae	1	1	1	0	0	0	0
Kerriidae	1	1	1	0	0	0	0
Totals	**39**	**51**	**35**	**16**	**2**	**4**	**7**

Six of the 51 species (11.8%) are pests in Kenya today (Tables 2 and 3). Amongst the 16 introduced species, at least one (*Aonidiella comperei*) could cause plant quarantine issues in plant produce trade, and four (25.0%) are pests (*Aonidiella comperei*, *Odonaspis ruthae*, *Planococcus minor* and *Pulvinaria polygonata*). *This is* more than four times more than the frequency of pests amongst the

African species (5.7%). Only two native African species recorded from Kenya for the first time are pests: *Pulvinarisca inopheron* (Coccidae) and *Aspidiotus ?ruandensis* (Diaspididae). *The difference in pest frequency between these two groups is* probably because the introduced species lack specialist natural enemies from their areas of origin. This may make them suitable for classical biological control, since many scale insects have host-specific parasitoids (Hymenoptera: Chacidoidea) in their native ranges.

Table 3. Newly recorded species in Kenya in this work, of economic importance or with pest potential: * = introduced, †=originating from Africa.

Family	Pest species	Potential pests
Coccidae	**Pulvinaria polygonata*	**Pulvinaria urbicola*
	†Pulvinarisca inopheron	
Diaspididae	**Aonidiella comperei*	**Odonaspis saccharicaulis*
	†Aspidiotus ?ruandensis	**Parlatoria blanchardi*
	**Odonaspis ruthae*	**Rutherfordia major*
Pseudococcidae	**Planococcus minor*	**Ferrisia dasylirii*

The remaining 45 non-pest species (12 (26.7%) of them introduced) have been present in Kenya for at least 13 years. Many have not been collected again since the original samples, suggesting that either they have not survived or they are rare because they are under good natural control.

Recommendations

In this small sample, introduced species were found to be almost three times more likely to become agricultural pests than native African species, probably due to the absence of specialist natural enemies from their areas of origin. Once identified, such introduced pests may be suitable for classical biological control using specialist natural enemies from their areas of origin. There is need for continued monitoring, awareness creation on the biology, spread and management of the pest in the surveyed counties.

Acknowledgements

The authors would like to thank the Director General, KALRO, Nairobi, Kenya, and Prof. Kenneth Norris, Head of Life Sciences, the Natural History Museum, London, U.K. for permission to study the collections at their institutions. The study was made possible by funding provided by the United Kingdom's Darwin Initiative scheme, project 25-032, supported by Department for Environment Food & Rural Affairs (Defra), Department for International Development (DFID) and the Foreign and Commonwealth Office (FCO).

References

Balachowsky, A.S. (1955). Contributions à l'étude de la faune entomologique du Ruanda Urundi (mission P. Basilewsky 1935). Pt I. Homoptera Coccoidea. Sur un *Aspidiotus* Bouché (Diaspididae-Aspidiotini) nouveau du Ruanda. *Annales du Musée royal du Congo Belge*, *36*, pp. 390-393.

Balachowsky**,** A.S. (1956). Les cochenilles du continent africain noir. Vol. 1. Aspidiotini (1ère Partie). *Annales du Musée royal du Congo belge*, *N.S.*, *3*, pp. 1-142.

Balachowsky, A.S. (1958). Les cochenilles du continent africain noir. Vol. 2. Aspidiotini (2me Partie), Odonaspidini et Parlatorini. *Annales du Musée royal du Congo belge*, *N.S.*, *4*, pp. 145-356.

Balachowsky, A.S., & Matile-Ferrero, D. (1966). Remarques sur les Pseudococcini, (Coccoidea-Coccidae) afro-tropicaux avec description d'un *Planococcus* Ferris nouveau vivant sur cacoyer en République Centrafricaine. *Cahiers de la Maboké, 4*, 68-72

Ben-Dov, Y. (1977). New species of *Aclerda* Signoret (Homoptera: Aclerdidae) from southern Africa. *Journal of Natural History, 11*, pp. 371-376.

Ben-Dov, Y. (1981). A catalogue of the Conchaspididae (Insecta, Homoptera, Coccoidea) of the world. *Annales de la Société entomologique de France*, *17*, 143-156.

Ben-Dov, Y. (1988). A taxonomic analysis of the armored scale tribe

Odonaspidini of the world (Homoptera: Coccoidea: Diaspididae). *United States Department of Agriculture Technical Bulletin, No. 1723*, pp. 1-142.

Ben-Dov, Y., & Russo, A. (1993). Giovanni De Lotto (1912-1990). *Bolletino del Laboratorio di Entomologia Agraria 'Filippo Silvestri' di Portici*, *48* (1991), pp. 245-251.

Boratynski, K.L. (1952a). Observations on *Matsucoccus pini* (Green) and a species of *Matsucoccus* Ckll. from Russia (Homoptera, Coccoidea, Margarodidae). *Annals and Magazine of Natural History*, *5*, pp. 507-508.

Boratyński, K.L. (1952b). *Matsucoccus pini* (Green, 1925) (Homoptera, Coccoidea: Margarodidae). Bionomics and external anatomy with reference to the variability of some taxonomic characters. *Transactions of the Royal Entomological Society of London, 103*, pp. 285-326.

Bullington, S.W., & Kosztarab, M.P. (1985). Studies on the morphology and systematics of scale insects - No. 12.I. Revision of the family Kermesidae (Homoptera) in the Nearctic region based on adult and third instar females. *Bulletin of the Virginia Polytechnic Institute and State University Agricultural Experiment Station*, *85*(11), 1-118

Cox, J.M. (1987). Pseudococcidae (Insecta: Hemiptera). Fauna of New Zealand. Duval, C. T. (series ed.), 11. DSIR Science Information Publishing Centre Wellington, New Zealand pp. 229.

Cox, J.M. (1989). The mealybug genus *Planococcus* (Homoptera: Pseudococcidae). *Bulletin of the British Museum (Natural History*). *Entomology, 58* (1), pp. 1-78.

Danzig, E.M. (1993). *[Fauna of Russia and neighbouring countries. Rhynchota, Volume X: suborder scale insects (Coccinea): families Phoenicococcidae and Diaspididae.]* St. Petersburg, Russia: Nauka Publishing House

Davidson, J.A., & Miller, D.R. (1990). 3.9.8 Ornamental Plants. In Rosen, D. (Ed.). *Armored Scale*

*Insects, Their Biology, Natural Enemies and Control (*pp. 603-632). *World Crop Pests, Vol. 4B.* Amsterdam, the Netherlands: Elsevier.

De Lotto, G. (1954a). Three apparently new coccids (Homopt.: Coccidae) from Eritrea. *Journal of the Entomological Society of southern Africa*, *17*, pp. 213-218.

De Lotto, G. (1954b). Three apparently new mealy bugs from Kenya. *Proceedings of the Royal Entomological Society of London*, *23*, pp. 110-114.

De Lotto, G. (1955). Three new coccids (Hemipt.: Coccoidea) attacking coffee in East Africa. *Bulletin of Entomological Research*, *46*, pp. 267-273.

De Lotto, G. (1956a). A new *Ceroplastodes* (Hom.: Coccoidea) from Kenya. *Journal of the Entomological Society of southern Africa*, *19*, pp. 310-312.

De Lotto, G. (1956b). The identity of some East African species of *Saissetia* (Homoptera, Coccidae). *Bulletin of Entomological Research*, *47*, pp. 239-249.

De Lotto, G. (1957a). Notes on some African species of *Saissetia* (Homoptera: Coccoidea: Coccidae). *Journal of the Entomological Society of southern Africa*, *20*, pp. 170-182.

De Lotto, G. (1957b). On some Ethiopian species of the genus *Coccus* (Homoptera: Coccoidea: Coccidae). *Journal of the Entomological Society of southern Africa*, *20*, pp. 295-314.

De Lotto, G. (1957c). The Pseudococcidae (Hom.: Coccoidea) described by H.C. James from East Africa. *Bulletin of the British Museum (Natural History). Entomology, Supplement*, *5*, pp. 185-232.

De Lotto, G. (1958a). A new species of *Saissetia* from South Africa (Homoptera: Coccoidea: Coccidae). *Journal of the Entomological Society of southern Africa*, *21*, pp. 66-68.

De Lotto, G. (1958b). New soft scales (Homoptera: Coccoidea: Coccidae) from Africa. *Proceedings of the Royal Entomological Society of London (B), 27*, pp.165-172.

De Lotto, G. (1958c). The Pseudococcidae (Hom.: Coccoidea) described by C.K. Brain from South Africa. *Bulletin of the British Museum (Natural History). Entomology, Supplement*, *7*, pp. 79-120.

De Lotto, G. (1959a). Further notes on Ethiopian species of the genus *Coccus* (Homoptera: Coccoidea: Coccidae). *Journal of the Entomological Society of southern Africa*, *22*, pp.150-173.

De Lotto, G. (1959b). Two new Margarodidae (Homopt.: Coccoidea) from Kenya. *Journal of the Entomological Society of Southern Africa, 22*, pp. 385-389.

De Lotto, G. (1960). The green scales of coffee in Africa south of the Sahara (Homoptera, Coccidae). *Bulletin of Entomological Research*, *51*, pp. 389-403.

De Lotto, G. (1961a). New Pseudococcidae (Homoptera: Coccoidea) from Africa. *Bulletin of the British Museum (Natural History). Entomology*, *10*, pp. 211-238.

De Lotto, G. (1961b). Two new *Ceroplastes* species from Africa (Homopt.: Coccidae). *Journal of the Entomological Society of southern Africa*, *24*, pp. 318-321.

De Lotto, G. (1962). A new *Coccus* from South Africa (Homoptera: Coccidae). *Journal of the Entomological Society of southern Africa, 25*, pp. 263-265.

De Lotto, G. (1963). New species and a new genus of hard scales from East Africa (Homoptera: Coccidae). *Proceedings of the Royal Entomological Society of London (B)*, *32*, pp.191-200.

De Lotto, G. (1964a). A new species of *Mombasinia* (Hom.: Pseudococcidae) from South Africa. *Proceedings of the Linnean Society of London*, *175*, pp. 135-136.

De Lotto, G. (1964b). A new species of *Pulvinaria* (Homopt.: Coccidae) attacking sugar cane in South Africa. *South African Journal of Agricultural Science*, *7*, pp. 863-866.

De Lotto, G. (1964c). Observations on African mealy bugs (Hemiptera: Coccoidea). *Bulletin of the British*

Museum (Natural History). Entomology, *14*, pp. 343-397.

De Lotto, G. (1965). On some Coccidae (Homoptera), chiefly from Africa. *Bulletin of the British Museum (Natural History). Entomology*, *16*, pp. 175-239.

De Lotto, G. (1966a). A new genus and four new species of Coccidae (Homoptera) from South Africa. *Proceedings of the Linnean Society of London*, *177*, pp. 143-149.

De Lotto, G. (1966b). Another new species of *Pulvinaria* (Hom.: Coccidae) from sugar cane. *South African Journal of Agricultural Science*, *9*, pp. 467-472.

De Lotto, G. (1966c). Descriptions of three new species of *Coccus* (Homoptera: Coccidae) from South Africa. *Proceedings of the Royal Entomological Society of London (B), 35*, pp. 41-46.

De Lotto, G. (1967a). The mealybugs of South Africa (Homoptera: Pseudococcidae), I. *Entomology Memoirs, Department of Agricultural Technical Services, Republic of South Africa. Pretoria*, *12*, pp. 1-28.

De Lotto, G. (1967b). The soft scales (Homoptera: Coccidae) of South Africa. I. *South African Journal of Agricultural Sciences*, *10*, 781-810

De Lotto, G. (1969a). On a few old and new soft scales and mealybugs (Homoptera: Coccoidea). *Journal of the Entomological Society of southern Africa*, *32*, pp. 413-422.

De Lotto, G. (1969b). The mealybugs of South Africa (Homoptera: Pseudococcidae), II. *Entomology Memoirs, Department of Agricultural Technical Services, Republic of South Africa. Pretoria, 20*, pp.1-30.

De Lotto, G. (1970). The soft scales (Homoptera: Coccidae) of South Africa, II. *Journal of the Entomological Society of southern Africa*, *33*, pp. 143-156.

De Lotto, G. (1971). On some genera and species of wax scales (Homoptera: Coccidae). *Journal of Natural History, 5*, pp. 133-153.

De Lotto, G. (1974a). New species of *Filippia* Targioni-Tozzetti, 1868 (Homoptera: Coccoidea: Coccidae) from South Africa. *Journal of the*

Entomological Society of Southern Africa, *37*, pp. 207-214.

De Lotto, G. (1974b). On the status and identity of the cochineal insects (Homoptera: Coccoidea: Dactylopiidae). *Journal of the Entomological Society of Southern Africa*, *37*, pp.167-193.

De Lotto, G. (1974c). On two genera of mealybugs (Homoptera: Coccoidea: Pseudococcidae). *Journal of the Entomological Society of Southern Africa*, *37*, pp. 109-115.

De Lotto, G. (1975a). New species of *Paracoccus* Ezzat & McConnell, 1956. (Homoptera: Coccoidea: Pseudococcidae) from southern Africa. *Journal of the Entomological Society of Southern Africa*, *38*, pp. 65-73.

De Lotto, G. (1975b). Two new genera of soft scales from Africa (Homoptera: Coccoidea: Coccidae). *Journal of the Entomological Society of southern Africa*, *38*, pp. 61-63.

De Lotto, G. (1978). The soft scales (Homoptera: Coccidae) of South Africa, III. *Journal of the Entomological Society of southern Africa, 41*, pp.135-147.

De Lotto, G. (1979). The soft scales (Homoptera: Coccidae) of South Africa, IV. *Journal of the Entomological Society of Southern Africa*, *42*, pp. 245-256.

Ebeling, W. (1959). *Subtropical fruit pests.* Los Angeles, U.S.A.: Division of Agricultural Sciences, University of California.

Ezzat, Y.M., & McConnell, H.S. (1956). A classification of the mealybug tribe Planococcini (Pseudococcidae: Homoptera). *Bulletin of the Maryland Agriculture Experiment Station*, *A-e84*, pp. 1-108.

Ezzat, Y.M. (1960). *Heliococcus osborni* (Sanders) redescribed as a new record from Egypt. *Bulletin de la Société entomologique d'Egypte, 44*, pp. 33-36.

Ezzat, Y.M. (1962). The genus *Trionymus* Berg in Egypt. U.A.R. *Bulletin de la Société entomologique d'Egypte, 46*, pp. 67-86.

Ferris, G.F. (1937). *Atlas of the scale insects of North America. Series*

1. Palo Alto, California, U.S.A.: Stanford University Press

Ferris, G.F. (1938). *Atlas of the scale insects of North America. Series 2.* Palo Alto, California, U.S.A.: Stanford University Press

Ferris, G.F. (1941). *Atlas of the scale insects of North America. Series 3.* Palo Alto, California, U.S.A.: Stanford University Press.

Ferris, G.F. (1942). *Atlas of the scale insects of North America. Series 4.* Palo Alto, California, U.S.A.: Stanford University Press.

Ferris, G.F. (1950). *Atlas of the Scale Insects of North America. (ser. 5) [v. 5]. The Pseudococcidae (Part I).* Palo Alto, California, U.S.A.: Stanford University Press

Ferris, G.F. (1953). *Atlas of the Scale Insects of North America, v. 6, The Pseudococcidae (Part II).* Palo Alto, California, U.S.A.: Stanford University Press.

Foldi, I. (2005a). Ground pearls: a generic revision of the Margarodidae *sensu stricto*. (Hemiptera: Sternorrhyncha: Coccoidea). *Annales de la Société entomologique de France*, *41*, pp. 81-125.

Foldi, I. (2005b). The Matsucoccidae in the Mediterranean basin with a world list of species (Hemiptera: Sternorrhyncha: Coccoidea). *Annales de la Société entomologique de France*, *40* (2), pp. 145-168.

Foldi, I. (2010). Trois espèces nouvelles du genre *Crypticerya* Cockerell de la region méditerranéenne (Hemiptera, Coccoidea, Monophlebidae). *Bulletin de la Société entomologique de France*, *115* (3), pp. 289-304.

García Morales, M., Denno, B.D., Miller, D.R., Miller, G.L., Ben-Dov, Y., & Hardy, N.B. (2016). *ScaleNet: a literature-based model of scale insect biology and systematics.* Database.

Giliomee, J.H., & Munting, J. (1968). A new species of *Asterolecanium* Targ. (Homoptera: Coccoidea: Asterolecaniidae) from South Africa. *Journal of the Entomological Society of Southern Africa, 31*, pp. 221-229.

Giliomee, J.H., & Kozár, F. (2008). Review of the family Asterolecaniidae (Hemiptera: Coccoidea) in South Africa, with the description of a new species. *African Entomology*, *16*(2), pp. 276-286.

Gill, R.J. (1988). *The Scale Insects of California: Part 1. The Soft Scales (Homoptera: Coccoidea: Coccidae).* Sacramento, California, U.S.A.: California Department of Food & Agriculture.

Gill, R.J. (1993). *The Scale Insects of California: Part 2. The Minor Families (Homoptera: Coccoidea).* Sacramento, California, U.S.A.: California Department of Food & Agriculture.

Gill, R.J. (1997). *The Scale Insects of California: Part 3. The Armored Scales (Homoptera: Diaspididae).* Sacramento, California, U.S.A.: California Department of Food & Agriculture.

Granara de Willink, M.C., & Szumik, C. (2007). Phenacoccinae de Centro y Sudamérica (Hemiptera: Coccoidea: Pseudococcidae): Sistemática y Filogenia. *Revista de la Sociedad Entomológica Argentina*, *66*(1-2), pp. 29-129.

Hall, W.J. (1925). Notes on Egyptian Coccidae with descriptions of new species. *Bulletin, Ministry of Agriculture, Egypt, Technical and Scientific Service*, *64*, pp. 1-31.

Hall, W.J. (1946). On the Ethiopian Diaspidini (Coccoidea). *Transactions of the Royal Entomological Society of London*, *97*, pp. 497-592.

Hambleton, E.J. (1976). A revision of the new world mealybugs of the genus *Rhizoecus* (Homoptera: Pseudococcidae). *United States Department of Agriculture Technical Bulletin*, *1522*, pp. 1-88.

Hodgson, C.J. (1967a). A revision of the species of *Inglisia* Maskell (Homoptera: Coccoidea) recorded from the Ethiopian region. *Arnoldia (Rhodesia)*, *3*, pp. 1-11.

Hodgson, C.J. (1967b). A species of *Conchaspis* (Homoptera: Conchaspididae) from central Africa. *Arnoldia (Rhodesia), 3*, pp. 1-3.

Hodgson, C.J. (1973). A revision of the *Lecanodiaspis* Targioni-Tozzetti (Homoptera: Coccoidea) of the

Ethiopian Region. *Bulletin of the British Museum (Natural History). Entomology*, *27*, pp. 413-452.

Hodgson, C.J. (1994). *The scale insect family Coccidae: an identification manual to genera.* Wallingford, U.K.: CAB International.

Hodgson, C.J., & Henderson, R.C. (2000). *Coccidae (Insecta: Hemiptera: Coccoidea). Fauna of New Zealand, 41*, pp. 1-259.

Hodgson, C.J., & Millar, I.M. (2002). A new subfamily, two new genera and three new species of Aclerdidae (Hemiptera: Coccoidea) from southern Africa, with a phylogenetic analysis of relationships. *Systematic Entomology*, *27*(4), pp. 469-517.

Hodgson, C.J., & Foldi, I. (2006). A review of the Margarodidae *sensu* Morrison (Hemiptera: Coccoidea) and some related taxa based on the morphology of adult males. *Zootaxa, 1263*, pp. 1-250.

Hodgson, C.J., & Williams, D.J. (2016). A revision of the family Cerococcidae Balachowsky (Hemiptera: Sternorrhyncha, Coccomorpha) with particular reference to species from the Afrotropical, western Palaearctic and western Oriental Regions, with the revival of *Antecerococcus* Green and description of a new genus and fifteen new species, and with ten new synonomies. *Zootaxa, 4091* (1), pp. 1-175.

Howell, J.O., & Williams, M.L. (1976). An annotated key to the families of scale insects (Homoptera: Coccoidea) of America, North of Mexico, based on characteristics of the adult female. Annals of the *Entomological Society of America*, *69* (2), pp. 181-189.

James, H.C. (1935). New hypogeic mealybugs (Coccidae) from East Africa. *Bulletin of Entomological Research*, *26*, pp. 379-390.

Karny, M. (1972). Comparative studies on three *Dactylopius* species (Homoptera: Dactylopiidae) attacking introduced opuntias in South Africa. *Entomology Memoirs, Department of Agricultural Technical Services, Republic of South Africa. Pretoria*, *26*, pp. 1-19.

Kaydan, M.B., & Gullan, P.J. (2012). A taxonomic revision of the mealybug

genus *Ferrisia* Fullaway (Hemiptera: Pseudococcidae), with descriptions of eight new species and a new genus. *Zootaxa, 3543*, pp. 1-65.

Klerk, C.A. de, Ben-Dov, Y., & Giliomee, J.H. (1982a). Redescriptions of four vine infesting species of *Margarodes* Guilding (Homoptera: Coccoidea: Margarodidae) from South Africa. *Phytophylactica*, *14*, pp. 61-76.

Klerk, C.A. de, Ben-Dov, Y., & Giliomee, J.H. (1982b). Redescriptions of three *Margarodes* Guilding species (Homoptera: Coccoidea: Margarodidae) found on grasses in South Africa. *Phytophylactica*, *14*, pp. 77-83.

Klerk, C.A. de, Ben-Dov, Y., & Giliomee, J.H. (1983). General morphology of South African species of *Margarodes* (Homoptera, Coccoidea, Margarodidae) with keys to nymphs and adult females. *Phytophylactica*, *15*, pp. 133-144.

Kondo, T., Gullan, P.J., & Pemberton, R.W. (2011). A new species of *Paratachardina* Balachowsky (Hemiptera: Coccoidea: Kerriidae) related to the lobate lac scale, *P. pseudolobata* Kondo & Gullan. *Journal of Asia-Pacific Entomology, 14*, pp. 141-146.

Kondo, T., Peronti, A.L., Kozár, F., & Szita, É. (2013). The citrus orthezia, *Praelongorthezia praelonga* (Douglas) (Hemiptera: Ortheziidae), a potential invasive species. In Pena, J. (Ed.). *Potential Invasive Pests of Agricultural Crops (*pp. 301-319*).* Wallingford, U.K.: CAB International.

Kosztarab, M., & Kozár, F. (1988). *Scale Insects of Central Europe*. Budapest, Hungary: Akademiai Kiado.

Kosztarab, M. (1996). *Scale insects of Northeastern North America. Identification, biology, and distribution.* Martinsburg, Virginia, U.S.A.: Virginia Museum of Natural History.

Kozár, F., & Konczné Benedicty, Z. (2000). Revision of *Newsteadia* of the Australian and Pacific regions, with description of eleven new species (Homoptera: Coccoidea, Ortheziidae). *Acta Zoologica*

Academiae Scientiarum Hungaricae, 46 (3), pp. 197-229.

Kozár, F., & Konczné Benedicty, Z. (2001). *Ortheziola* of Asia with the descriptions of three new species, and world distribution of the genus (Homoptera: Coccoidea, Ortheziidae). *Acta Zoologica Academiae Scientiarum Hungaricae, 47*(1), pp. 15-25.

Kozár, F., Foldi, I., & Konczné Benedicty, Z. (2002). Description of six new species of *Newsteadia* from Madagascar, Nepal and Peru (Hemiptera, Coccoidea, Ortheziidae). *Nouvelle Revue d'Entomologie, 19* (2), pp. 179-192.

Kozár, F. (2004). *Ortheziidae of the World.* Budapest, Hungary: Plant Protection Institute, Hungarian Academy of Sciences

Kozár, F., & Konczné Benedicty, Z. (2007). *Rhizoecinae of the world.* Budapest, Hungary: Plant Protection Institute, Hungarian Academy of Sciences.

Kozár, F., Kaydan, M.B., Konczné Benedicty, Z., & Szita, É. (2013). *Acanthococcidae and Related Families of the Palaearctic Region.* Budapest, Hungary: Hungarian Academy of Sciences.

Łagowska, B., & Hodgson, C.J. (2019). On some new and old soft scale insects (Hemiptera: Coccomorpha: Coccidae) from Africa, with description of a new *Coccus* species and introduction of a new genus. *Zootaxa*, *4612*(3), pp. 373-386.

Laing, F. (1929). Descriptions of new, and some notes on old species of Coccidae. *Annals and Magazine of Natural History*, *4*, pp. 465-501.

Lambdin, P.L., & Kosztarab, M. (1977). Morphology and systematics of the adult females of the genus *Cerococcus* (Homoptera: Coccoidea: Cerococcidae). *Bulletin of the Virginia Polytechnic Institute and State University Research Division, Blacksburg, 128*, pp. 1-252.

Mamet, R.J. (1954). A monograph of the Conchaspididae Green (Hemiptera: Coccoidea). *Transactions of the Royal Entomological Society of London, 105*, pp. 189-239.

Mamet, R.J. (1958). The *Selenaspidus* complex (Homoptera, Coccoidea). *Annales du Musée Royal du Congo Belge. N.S.*, *4*, pp. 359-429.

Matile-Ferrero, D. (1970). Hemiptera (Homoptera): Coccoidea. *South African Animal Life*, *14*, pp. 171-185.

McKenzie, H.L. (1967). *Mealybugs of California with taxonomy, biology, and control of North American species (Homoptera: Coccoidea: Pseudococcidae).* Berkeley, California, U.S.A.: University of California Press.

Miller, D.R., & Williams, D.J. 1995. Systematic revision of the family Micrococcidae (Homoptera: Coccoidea), with a discussion of its relationships, and a description of a gynandromorph. *Bollettino del Laboratorio di Entomologia Agraria 'Filippo Silvestri'. Portici*, *50* (1993), pp. 199-247.

Miller, D.R., & Kozár, F. (2002). Systematic analysis of Afrotropical *Newsteadia* (Hemiptera, Coccoidea: Ortheziidae) with descriptions of nine new species. *Acta Phytopathologica et Entomologica Hungarica, 37*(103), pp. 201-250.

Miller, D.R., & Davidson, J.A. (2005). *Armored Scale Insect Pests of Trees and Shrubs.* Ithaca, New York, U.S.A.: Cornell University Press.

Miller, D.R., & Giliomee, J.H. (2011). Systematic revision of the mealybug genus *Delottococcus* Cox & Ben-Dov (Hemiptera: Pseudococcidae). *African Entomology*, *19*(3), pp. 614-640.

Miller, D., Rung, A., Parikh, G., Venable, G., Redford, A.J., Evans, G.A., & Gill, R.J. (2014). *Scale Insects, Edition 2*. USDA APHIS Identification Technology Program (ITP), Fort Collins, Colorado, U.S.A.

Morrison, H. (1925). Classification of scale insects of the subfamily Ortheziinae. *Journal of Agricultural Research*, *30*, pp. 97-154.

Morrison, H. (1928). A classification of the higher groups and genera of the coccid family Margarodidae. *United States Department of Agriculture Technical Bulletin,* 52, pp. 1-239.

Morrison, H. (1952). Classification of the Ortheziidae. Supplement to "Classification of scale insects of the subfamily Ortheziinae". *United States Department of Agriculture Technical Bulletin, 1052*, pp. 1-80.

Nakahara, S., & Gill, R.J. (1985). Revision of *Philephedra*, including a review of *Lichtensia* in North America and description of a new genus *Metapulvinaria* (Homoptera: Coccidae). *Entomography*, *3*, pp. 1-42.

Neumann, G., O'Dowd, D.J, Gullan, P.J., & Green, P.T. (2014). First record of *Pulvinaria urbicola* Cockerell (Hemiptera: Coccidae), a potentially damaging scale insect, on Christmas Island, Indian Ocean. *Journal of Asia-Pacific Entomology*, *17*, pp. 27-30.

Neumann, G., O'Dowd, D.J, Gullan, P.J., & Green, P.T. (2016). Diversity, endemism and origins of scale insects on a tropical oceanic island: implications for management of an invasive ant. *Journal of Asia-Pacific Entomology*, *19*, pp. 159-166.

Peck, R., Banko, P., Pendleton, F., Schmaedick, M., & Ernsberger, E. (2014). *Arthropods of Rose Atoll with special reference to ants and Pulvinaria urbicola scales (Hemiptera: Coccidae) on Pisonia grandis trees.* Technical Report HCSU-057. Hilo, Hawai'i: Hawai'i Cooperative Studies Unit, University of Hawai'i.

Richard, C. (1976). Révision du groupe des *Stictococcus*, et création de taxa nouveaux (Homoptera, Coccoidea). *Annales de la Société entomologique de France*, *12*, pp. 653-669.

Russell, L.M. (1941). A classification of the scale insect genus *Asterolecanium*. *United States Department of Agriculture, Miscellaneous Publications, 424*, pp. 1-319.

Sartiami, D., Watson, G.W., Mohamad Roff, M.N., & Idris, A.B. (2016). New Indonesian country and species records of mealybugs (Hemiptera: Pseudococcidae) in Wirjati's historic collection. *REDIA*, *XCIX*, pp. 155-161.

Sartiami, D., Watson, G.W., Mohamad Roff, M.N., & Idris, A.B. (2017). A taxonomic update of Takahashi's

historic collection of mealybugs (Hemiptera: Pseudococcidae) from Malaysia and Singapore. *Serangga, 22* (2), pp. 91-114.

Schneider, S.A., & LaPolla, J.S. (2011). Systematics of the mealybug tribe Xenococcini (Hemiptera: Coccoidea: Pseudococcidae), with a discussion of trophobiotic associations with *Acropyga* Roger ants. *Systematic Entomology*, *36*, pp. 57-82.

Schneider, S.A., Fizdale, M., & Normark, B.B. (2019). Key to commonly intercepted armored scales: Aspidiotini. An interactive key to species of Diaspididae: tribe Aspidiotini.

Smith, D., Papacek, D., Hallam, M., & Smith, J. (2004). Biological control of *Pulvinaria rubicola* (Cockerell) (Homoptera: Coccidae) in a *Pisonia grandis* forest on North East Herald Cay in the coral sea. *General and Applied Entomology*, *33*, pp. 61-68.

Stickney, F.S. (1934). The external anatomy of the red date scale *Phoenicococcus marlatti* Cockerell, and its allies. *United States Department of Agriculture Technical Bulletin*, *404*, pp. 1-162.

Stumpf, C.F., & Lambdin, P.L. (2001). Taxonomic status of *Bambusaspis miliaris, B. robusta*, and *B. pseudomiliaris* (Hemiptera: Coccoidea: Asterolecaniidae). *Insecta Mundi, 13* (3/4), pp. 205-210.

Stumpf, C.F., & Lambdin, P.L. (2006). *Pit scales (Sternorrhyncha: Coccoidea) of North and South America.* Knoxville, Tennessee, U.S.A.: Tennessee Agricultural Experiment Station, Institute of Agriculture, University of Tennessee.

Suh, S.-J. (2016). Armoured scale insects (Hemiptera: Diaspididae) intercepted at the ports of entry in the Republic of Korea over the last 20 years. *Bulletin OEPP/EPPO Bulletin*, *46* (2), 313-331.

Tang, F.T. (1986). *[The scale insects of horticulture and forest of China. Volume III.]* Taigu, Shanxi, China: Shanxi Agricultural University Press.

Unruh, C.M., & Gullan, P.J. (2008). Identification guide to species in the scale insect tribe Iceryini

(Coccoidea: Monophlebidae). *Zootaxa*, *1803*, 1-106.

Varshney, R.K. (1984). A review of the family Tachardiidae (Kerriidae) in the Orient (Homoptera: Coccoidea). *Oriental Insects. New Delhi*, *18*, pp. 361-384.

Varshney, R.K. (1990). A synoptic updated catalogue of lac insects (Homoptera: Coccoidea: Tachardiidae). Pp. 71-74. In Koteja, J. (Ed.). *Proceedings of the Sixth International Symposium of Scale Insect Studies, Part II. Cracow, Poland: August 6-12, 1990.* Cracow, Poland: Agricultural University Press.

Vahedi, H.A., & Hodgson, C.J. (2007). Some species of the hypogeal scale insect *Porphyrophora* Brandt (Hemiptera: Sternorrhyncha: Coccoidea: Margarodidae) from Europe, the Middle East and North Africa. *Systematics and Biodiversity, 5*(1), pp. 23-122.

Watson, G.W., & Cox, J.M. (1990). Identity of the African coffee root mealybug, with descriptions of two new species of *Planococcus* (Homoptera: Pseudococcidae). *Bulletin of Entomological Research*, *80*, pp. 99-105.

Watson, G.W. (2002). *Arthropods of economic importance: Diaspididae of the world. An illustrated identification guide and information source.* University of Amsterdam, Netherlands: Expert Center for Taxonomic Identification (ETI) CD-ROM (with illustrations).

Williams, D.J. (1958a). Mealybugs (Pseudococcidae: Homoptera) described by W.M. Maskell, R. Newstead, T.D.A. Cockerell and E.E. Green from the Ethiopian region. *Bulletin of the British Museum (Natural History). Entomology*, *6*, 205-236

Williams, D.J. (1958b). The mealybugs (Pseudococcidae-Homoptera) described by W.J. Hall, F. Laing and A. H. Strickland from the Ethiopian region. *Bulletin of the British Museum (Natural History). Entomology*, *7*, pp. 1-37.

Williams, D.J. (1961). Notes on the genus *Heterococcus* Ferris (Coccoidea, Homoptera) with a description of a new species injurious to guinea-corn *(Sorghum*

vulgare) in Nigeria. *Bulletin of Entomological Research*, *51*, pp. 671-675.

Williams, D.J. (1963). Synoptic revisions of I. *Lindingaspis* and II. *Andaspis* with two new allied genera (Hemiptera: Coccoidea). *Bulletin of the British Museum (Natural History). Entomology, 15*, pp. 1-31.

Williams, D.J. (1970). The mealybugs (Homoptera, Coccoidea, Pseudococcidae) of sugar-cane, rice and sorghum. *Bulletin of Entomological Research*, *60*, pp. 109-188.

Williams, D.J. (1986). The identity and distribution of the genus *Maconellicoccus* Ezzat (Hemiptera: Pseudococcidae) in Africa. *Bulletin of Entomological Research*, *76*, pp. 351-357.

Williams, D.J., & Watson, G.W. (1988a). *The Scale Insects of the Tropical South Pacific Region. Pt. 1: The Armoured Scales (Diaspididae).* Wallingford, U.K.: CAB International.

Williams, D.J., & Watson, G.W. (1988b). *The Scale Insects of the Tropical South Pacific Region. Pt. 2: The Mealybugs (Pseudococcidae).* Wallingford, U.K.: CAB International

Williams, D.J., & Watson, G.W. (1990). *The Scale Insects of the Tropical South Pacific Region. Pt. 3: The Soft Scales (Coccidae) and Other Families.* Wallingford, U.K.: CAB International.

Williams, D.J., & Granara de Willink, M.C. (1992). *Mealybugs of Central and South America.* Wallingford, U.K.: CAB International.

Williams, D.J. (1996). A brief account of the hibiscus mealybug *Maconellicoccus hirsutus* (Hemiptera: Pseudococcidae), a pest of agriculture and horticulture, with descriptions of two related species from southern Asia. *Bulletin of Entomological Research*, *86*, pp. 617-628.

Williams, D.J. (1998). Mealybugs of the genera *Eumyrmococcus* Silvestri and *Xenococcus* Silvestri associated with the ant genus *Acropyga* Roger and a review of the subfamily Rhizoecinae (Hemiptera, Coccoidea, Pseudococcidae).

Bulletin of the Natural History Museum. Entomology Series, *67*, pp. 1-64.

Williams, D.J., & Matile-Ferrero, D. (1999). A new species of the mealybug genus *Cataenococcus* Ferris from Ethiopia on *Ensete ventricosum*, a plant infected by a virus (Hemiptera: Pseudococcidae: Musaceae). *Revue Française d'Entomologie*, *21*(4), pp. 145-149.

Williams, D.J. (2001). African species of the mealybug genus *Antonina* Signoret (Hemiptera: Coccoidea: Pseudococcidae). *Journal of Natural History*, *35*, pp. 833-848.

Williams, D.J. (2004). *Mealybugs of southern Asia.* The Natural History Museum, London, U.K. Kuala Lumpur, Malaysia: Southdene SDN. BHD.

Williams, D.J., & Matile-Ferrero, D. (2005). Mealybugs from Zanzibar and Pemba islands with a discussion of a potential invasive species (Hemiptera, Pseudococcidae). *Revue Française d'Entomologie*, *27*(4), pp. 145-152.

Williams, D.J., Matile-Ferrero, D., & Miller, D.R. (2010). A study of some species of the genus *Stictococcus* Cockerell (Hemiptera: Sternorrhyncha: Coccoidea: Stictococcidae), and a discussion on *Stictococcus vayssierei* Richard, a species injurious to cassava in Equatorial Africa with ... from Nigeria. *Zootaxa*, 2527, pp. 1-27.

Pest Incursions Pose a Serious Threat To Food Security and the Kenyan Economy

Macharia I.[1], Koome F.[1], Kosiom T.[1], Munguti F.,[1] Mukoye B.[1], Kimani E[1], Kimenju J.W.[2]
[1] Kenya Plant Health Inspectorate Service (KEPHIS)-P.O BOX 49592, Nairobi

[2] University of Nairobi Faculty of Agriculture, Department of Crop Production and Plant Protection, P.O. Box 29053, Nairobi.

Corresponding Author: macharia.isaac@kephis.org

Abstract

Although Kenya has a well-developed phytosanitary system to regulate introduction of plant and plant products, several pest incursions have been reported in the last two decades. The incursions have culminated in devastating impact on agriculture, biodiversity and the entire Kenyan economy. The objective of this review is to consolidate information on the pests involved, their distribution, estimate the economic losses associated with them and management measures in place. A total of 11 major pests and diseases namely Asian citrus psyllid (ACP), *Bactrocera dorsalis*, Banana xanthomonas wilt (BXW), Cassava brown streak disease (CBSD), Cassava mosaic disease (CMD), Fall army worm (FAW), Maize lethal necrosis disease (MLND), Papaya mealybug (PMB), *Parthenium hysterophorus,* potato cyst nematode (PCN) and tomato leaf miners (*Tuta absoluta*) have been reported in the last two decades. Some of the pests are persistent, invasive, vicious and fast spreading. For instance, the FAW has now spread to nearly all maize growing areas in Kenya in one year after the pest was first reported in 2017. The incursion pests are a major threat to food security, expensive to control and are a barrier to international market access. Integrated measures including improvement of diagnostic potential, increased pest and disease surveillance, improvement in rapid response and pest containment are needed in view of the dangers posed by incursion pests to the entire Kenyan economy whose mainstay is agriculture.

Key words: *Bactrocera dorsalis,* maize lethal necrosis disease, *Parthenium hysterophorus, Tuta absoluta*

Introduction

Agriculture is the backbone of the economies of many African countries contributing over 30% of the GDP. Production has been significantly affected by pest and diseases some of which have been introduced through trade. International movement of plant and plant produce has always been regulated due to the risk of introduction of pests and diseases. As a result of several serious pests having been introduced in different countries in the late 1800s, it was clear that there is need to undertake action to prevent further introductions. For instance, the late blight in Ireland which left over 2 million death due to starvation, the coffee leaf rust in Srinlanka introduced from Africa seriously affected coffee production, In Africa, the outbreak of coconut yellow lethal necrosis in Madagascar, and fusarium wilt of banana caused by strain TR4 in Mozambique are some of the examples of pests which have been reported to cause serious economic damage to the agricultural sector and the environment. Pest incursions has led to development of international phytosanitary measures which are currently being used to prevent introduction of quarantine pests or limit the entry of regulated non-quarantine pests while promoting international trade.

Kenya, like any other African countries has not been spared from the effect of introduction of new harmful pests and diseases. Inadequate phytosanitary capacity in many African countries has been cited as important factors which could be contributing to introduction and spread of new pests and diseases. Additionally, lack of capacity in diagnostics has sometimes delayed responses to emerging and endemic pathogens. Diagnostic tools might be available for some diseases and pests, but may not be applied in time to be effective. The widespread lack of equipment, supplies, reference materials and opportunities for training hamper the

ability of African scientists to provide these basic services and, further, to document the presence of dangerous pathogens and pests within their borders. Globalization, climate change, porous borders, financial constraints, and lack of awareness among farmers, importers, exporters and research scientists have been shown to be a challenge in preventing introduction and spread of harmful pests.

Although Kenya has developed a stringent phytosanitary system which regulates movement of plant and plant products, there has been several pest incursions which have negatively impacted on crop production, biodiversity, and human development. Some of the previous incursions reported before 1998 include Cassava mosaic disease, *Prostephanus truncates* (Larger grain borer-LGB), *Fusarium oxysporum f. sp. cubense* (Panama disease), *Salviniamolesta* (Salvinia) and *Eichhorniacrassipes* (Water hyacinth). These pests are still serious pests in Kenya with enormous resources being channeled to their management .The LGB which is native to Central and South America was introduced in Africa in early 1970s. The pest has spread into Kenya and other African countries through movement of infested grain. Water hyacinth infested Kenyan waterways in mid 1980s. It quickly spread and attained an estimated peak of 17,230 ha coverage on the Kenyan side of the Lake Victoria by 1998.

Foreign pests that have been reported on crops in Kenya since 1998

In the last two decades, Kenya has encountered several major pest incursions which include Asian citrus psyllid (ACP), *Bactrocera dorsalis*, Banana xanthomonas wilt (BXW), Cassava brown streak disease (CBSD), Cassava mosaic disease (CMD), Fall army worm (FAW), Maize lethal necrosis disease (MLND), Papaya mealybug (PMB), *Parthenium hysterophorus,* potato cyst nematode (PCN) and tomato leaf miners (*Tuta absoluta*), among others which have significantly affected food security, the environment and international trade (Table 1). A recent example is the invasive fall army worm which has been extensively damaging on

maize and other crops in Sub-Saharan Africa.

The fall army worm (FAW) *Spodoptera frugiperda*, a native to the tropical regions of the western hemisphere from the United States to Argentina, has caused heavy losses to cereal farmers in Africa since its introduction in the West African region in 2016. FAW was reported for the first time in Kenya in Trans Nzoia County in March 2017 in an offseason irrigated maize crop after which it spread first to all the maize production areas (KARLO, 2017). The pest has continued to cause serious losses in maize production and is threating horticultural export. In view of the importance of this pest, the government has instituted a multi-institutional technical team which has develop strategies for management of the pest.

Maize lethal necrosis is a serious viral disease affecting maize in Kenya. The disease is caused by co-infection of Maize Chlorotic Mottle Virus (MCMV) and Sugar Cane Mosaic Virus (SCMV) or with other cereal potyviridae viruses like the Wheat Streak Mosaic Virus (WSMV) or Maize Dwarf Mosaic Virus (MDMV).The disease was first reported in Bomet County in Kenya in 2011 and is currently spread across several maize production areas (Wangai *et al*., 2012). Estimated maize yield loss due to MLND varies from region to region, maize variety and season of the year. In Kenya, up to 100% yield losses have been reported in areas where the disease was very severe (Wangai *et al*., 2012). MLND causing viruses are transmitted by several vectors including thrips (*Frankliniella Williamsi*), and cereal leaf beetles (*Oulemame lanopus*).The disease is also seed transmitted. Several measures have been put in place to mitigate the negative effect caused by the viruses which include up-scaled seed certification system, use of systemic pesticide and breading for resistance.

Table 1. Foreign pests that have been reported on crops in Kenya since 1998.

Name of pest or disease	Year first reported	Status	Current Distribution	Yield loss Potential	References
Spodoptera frugiperda (Fall army worm)	2017	Widespread	All maize growing areas in Kenya	73%	CABI, 2018
Diaphorina citri (Asian citrus psyllid).	2016	Restricted	Coast Kenya	100% by greening disease	Rwomushana *et al*., 2017
Paracoccus marginatus (Papaya mealybug)	2016	Regulated	Coast Kenya	100%	Macharia *et al*., 2017
Globodera rostochiensis (Potato cyst nematode)	2015	Regulated	Potato production areas	80%	Mwangi *et al*., 2015
Tuta absoluta (Tomato leaf miner)	2014	Widespread	All tomato producing areas in Kenya	100%	Duressa, 2018
Maize lethal necrosis	2011	Regulated	Maize production areas	90%	Wangai *et al*., 2012
Parthenium hysterophorus (Parthenium weed)	2010	Noxious weed	Most open farming lands	High	Guyana, P., & Paraguay, S. 2014
Cassava brown streak disease	2006	Restricted	Coastal and Western Kenya	70%	Were *et al*., 2016
Xanthomonas campestris pv. Musacearum (Banana xanthomonas wilt)	2006	Restricted	Western Kenya	100%	Kwach *et al*., 2013
*Bactrocera (dorsalis) invades(*Mango fruit fly)	2003	Invasive	All host crops producing areas in Kenya	70%	Luc *et al* .,2003; Ekesi *et al*., 2011

Bactrocera dorsalis (formerly *Bactrocera invades*) is an invasive fruit fly species of Asian origin which was first reported in Kenya in 2003 (Lux *et al*., 2003). The pest has been reported to cause yield losses of up to 70% in mangoes (Ekesi e*t al*., 2011). Apart from the huge loss, introduction of *B. dorsalis* significantly affected international market for horticultural produce in Kenya. The most notable example is loss of the European Union (EU) market for mangoes and South Africa market for avocados coupled with inability to access other market such as USA, Australia among others. Use of pheromone traps and post-harvest treatments have been used in the management of the pest (Ekesi *et al.,* 2011)

Banana, a major fruit crop, has been threatened by Xanthomonus Wilt (BXW) caused by a bacterium *Xanthomonas vasicola.pv. musacearum (Xvm)*, formerly known as *Xanthomonas campestris,* which has been shown to cause up to 100% yield loss (Kwach *et al*., 2013). The disease was introduced from Uganda and has been shown to affect all banana cultivars. Xanthomonus Wilt (BXW) is best managed by use of clean planting materials and removing of male flower buds, and sterilization of tools. The disease has been reported in banana growing areas in Western Kenya and Nyanza (Kwach *et al*., 2013).

Tomato (*Lycopersicon esculentum* Mill) is one of the most important vegetable crops whose production has been threatened by *Tuta absoluta* (tomato leaf miner) which was introduced in Kenya in 2014. The pest has been reported to cause yield loss upto100% (Duressa, 2018). Chemical control and use of traps are the main management measures that have been used since the pest was reported.

Cassava Mosaic Disease (CMD) was reported in the 1980 as one of the most important viral disease affecting cassava in Kenya. Despite the effort through breeding for diseases resistance, introduction of cassava brown streak disease (CBSD) in 2006 rendered the effort made futile and breeders were forced to start all over again as all CMD resistant varieties were susceptible to CBSD. CBSD and CMD causing viruses are transmitted by *Bemisia tabaci* and

have the potential of causing up to 100% yield losses. In Kenya, among the CMD causal viruses, EACMV-Ug has recently been reported to be the most prevalent followed by EACMV and ACMV contrary to previous reports where ACMV was the most prevalent in Kenya (Were *et al*., 2016). It is not yet clear what factors have contributed to this change. Breeding for resistance is the most reliable means of control.

Other pest that have been introduced in Kenya include: *P. marginatus,* native of Central America and was first observed on the Africa in Ghana in 2010 from where it spread to other African countries. The pest was reported in Kenya in 2016 (Macharia *et al*., 2017). The pest is highly polyphagous, with hosts recorded from 84 plant species causing yield loss of up to 100%; *Diaphorina citri,* is native in Asia, and was first reported in Kenya in 2016. It causes up to 80% yield loss in citrus (Khan *et al.,* 2014); Potato cyst Nematode (PCN) is a serious pest of potato that was first reported in Kenya in 2014 in most potato production areas (Mwangi *et al*., 2015). Although the pest is native of in Europe, its introduction is still not clear. PCN has been reported to cause yield losses of up to 80%.

Prevention and management strategies of pest incursions in Kenya

Although introduction of pests still remains a challenge in Kenya and other African countries, Kenyan government through KEPHIS and other institutions has invested heavily on technology and policy to support phytosanitary regulation in the effort to prevent pest introduction and spread in the country. Regulating importation of plant and plant product plays an important role in minimizing introduction of harmful pest. Other measures instituted to reduce introduction of pest and diseases includes pest risk analysis which provide risks associated with importation and possible mitigation measures. Pest surveillance to establish occurrence and distribution of pests, pest identification, containment of materials likely to introduce pests in quarantine facilities among others.

KEPHIS Plant Quarantine and Biosecurity Station where most of the plant health facilities are located, is responsible for handling imported high risk plant materials. The imported material are grown under containment and monitored for pests and diseases for a period of time based on the pest before they are released to the importer. This reduces the chances of introduction and spread of harmful pests and diseases in the country. Importers are also allowed to establish quarantine facilities in their farms under strict regulation, monitoring and control from KEPHIS. All materials under quarantine are monitored and tested for quarantine pests before they are released. At the boarder points and point of entry and exit inspection, sampling grading of all imported consignment is undertaken.

In the last two decades several pests were identified in imported materials and appropriate measures undertaken to prevent their introduction. Among the pests detected were coconut case caterpillar *Mahasena corbetti*, *Curtobacterium flaccumfaciens* pv. *flaccumfaciens*, *Pectobacterium spp*, *Dickeya spp*, *Alternaria padwickii*, PVY^{NTN} and several others fungal, bacterial and viral diseases.

Conclusion

Various pest of economic importance have gained entry into the country and are posing as threats to food security and the general well-being of the people. The pests come in the form of plant pathogens, arthropod pests and invasive plant species.

Recommendation

There is need to conduct surveillance in all parts of the country for early detection which is important for management of pests. It is also important to strengthen phytosanitary measures and create awareness on pest management strategies for the county to attain the required food security. A surveillance database for the pests to be updated for ease of retrieval and build up on information.

Acknowledgment

The authors are grateful to the management of The Kenya Plant Health Inspectorate Service and The University of Nairobi for supporting the collaboration that led to this paper.

References

Duressa T. F. (2018). Newly Emerging Insect Pests and Diseases as a Challenge for Growth and Development of Ethiopia: The Case of Western Oromiya*. Journal of the Science of Food and Agriculture* 9, pp. 201.

Ekesi S., De Meyer M., Mohamed S.A., Virgilio M., & Borgemeister C. (2016). Taxonomy, ecology, and management of native and exotic fruit fly species in Africa. *Annual Review of Entomology*. 61, pp. 219–238.

Guyana, P. & Paraguay, S. (2014). Parthenium hysterophorus L. Asteraceae–Parthenium weed. *Bulletin OEPP/EPPO Bulletin*, 44 (3), pp. 474-478.

Khan A. A., Afzal M., Qureshi J.A., Khan A. M. & Raza A. M. (2014). Botanicals, selective insecticides, and predators to control *Diaphorina citri* (Hemiptera: Liviidae) in citrus orchards. *Insect Science.* 21, pp. 717–726.

Kwach J.K., Onyango M.A., Muthomi J.W. & Nderitu J.H. (2013). Status and spread of banana Xanthomonas wilt in Kenya. *African Crop Science Conference Proceedings*. 11, pp. 297-308 .

Lux S, Copeland RS, White IM, Manrakhan A, & Billah MK. (2003). A new invasive fruit fly species from the *Bactrocera dorsalis* (Hendel) group detected in East Africa. *International Journal of Tropical Insect Science* 23, pp. 355–361.

Macharia I., Kimani E., Koome F., Kosiom T., Heya H., Otipa M., & Oronje M. (2017). First Report and Distribution of the Papaya Mealybug, *Paracoccus marginatus*, in Kenya *Journal of Agricultural and Urban Entomology* 33, pp. 142-150.

Mwangi J.M., Kariuki G.M., Waceke J.W., Grundler F.M. (2015). First report of *Globodera rostochiensis* infesting potatoes in Kenya. *New Disease Reports* 31, pp. 18.

Rwomushana I., Khamis F.M., Grout T.G., Mohamed S.A., Sétamou M., Borgemeister C., Heya H.M., Tanga C.M., Nderitu P.W., Seguni Z.S., Materu C.L., & Ekesi S. (2017). Detection of Diaphorina citri Kuwayama (Hemiptera: Liviidae) in Kenya and potential implication for the spread of Huanglongbing disease in East Africa. Biological Invasions.

Smith J.J., Waage J., Woodhall J.W., Bishop J.S., Spence N.J. (2008). The challenge of providing pest diagnostic services for Africa. Eurropean Journal of Plant Pathology. 121, pp. 365-375.

Wangai AW, Redinbaugh MG, Kinyua ZM, Miano DW, Leley PK, Kasina M, Mahuku G, Scheets K, Jeffers D. 2012. First report of Maize chlorotic mottle virus and maize lethal necrosis in Kenya. *Plant Disease*, 96, pp. 1582-1583.

Were M.N., Mukoye B., Osogo A.K., Mangeni B.C., Nyamwamu P. A., Ogemah V. K., Muoma J. V., Winter S., Were H. K. (2016). Occurrence and Distribution of Begomoviruses Infecting Cassava in Western Kenya, *Plant*. 4, pp. 108-113.