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Identification of major insect pest associated to taro leaf blight disease transmission

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ABSTRACT
Objective: This study aimed to identify potential entomopathogenic pest associated with taro damage and transmission of taro late blight disease.
Methodology and Results: Insect pest in taro infected fields were collected from infected crops and soil for identification using an insect identification guide. The percentage abundance of each insect phylum was calculated, and insect photographs taken. Pathogenicity Assessment was done to isolate spores of *P. colocasiae* from insect samples and the isolated spores used in inoculating taro cultivars growing in a screen house. The average lesion diameter *P.colocasiae* infected area was recorded for 14 days.
Conclusion and Application of Results: The Phylum Arthropoda had the highest abundance (100 %) in Bambui and Yaounde, while Phylum Annelida had the lowest abundance (7.69 %) in Ekona. No lesions developed on inoculated cultivars in the screen house, indicating that taro leaf blight is not vectored by insect pests. However, these insect pests created wounds on leaves for rapid growth and proliferation of the pathogen.
Key words: Identification, insect pest, Taro leaf blight disease, transmission.

INTRODUCTION
Taro (*Colocasia esculenta*) has become a major staple food and has gained popularity most countries in the tropics. However, taro production has declined due to severe pests and diseases attacks (Fokunang *et al.,* 2016). Taro leaf blight disease (TLBD) cause by *Phytophthora colocasiae* is characterized by large necrotic zonate spots on the leaves often expand coalesced and destroy large areas of leaves (Manju *et al.,* 2017). The margin of the lesion is marked by a white powdery band of sporangia and numerous droplets of orange or reddish exudates (Bandyopadhyay *et al.,* 2011). Taro leaf blight caused 90 % yield lost in Cameroon (Mbong *et al., 2013*) and 95% leaf yield in susceptible genotypes (Nelson *et al.,* 2011). Infection also led to a significant loss in planting materials and crop failure when infected corms were used and some farmers abandoned their farms and cultivated other

crops in taro growing regions of Cameroon (Guarion, 2010; Fontem and Mbong, 2011). Carmical *et al.(*2008) reported that pest common to taro destruction and disease transmission were *Apis gossy, Aleurodicus disperses*, *Hippotion celerio*, *Pterostichus melanarius, Phenacoccus saenopsis,*Tetrancy spp, plant leafhopper (Tarophagus spp), and *Spodoptera litura.* Insect pests are considered to be responsible for approximately 10–20% of yield losses in significant crops worldwide and about 50% of annual production in developing countries (Badii*et al.,* 2015). Taro beetles are important pests of taro which reduces corm yields and quality by creating wounds on taro during storage which permits the entering of pathogens (Waterhouse and Norris, 1987; Droby *et al.,* 1992). Normally, the high infestation of insect pest discourages farmers from cultivating desired crops, and some tend to abandon infested crop fields. (Tindo and Tamo 1999). In Cameroon, pest common in taro fields are intensive in taro-growing regions. These pests usually appear in the fields during the taro planting seasons, between July and November, when rainfall and high relative humidity levels are high and favour pest growth and reproduction. Taro farmers in the tropics experience substantial plant protection-related risks that threaten agricultural production safety and sustainable food supply (Ratnadass *et al.,* 2021). Many research efforts towards reducing taro yield losses have focused on identifying the disease's causal agent and pathogenicity and proposing suitable control measures(Fontem and Mbong, 201; Mbong *et al.,*2013;Fokung *et al.,* 2016; Manju *et al.,*2017; Manu *et al.,*2022). Despite these efforts, taro leaf blight disease remains a constraint during production, and limited data exists on insect pests associated with taro leaf blight disease. Therefore, study was aimed to identify potential entomopathogenic pests associated with taro damage and transmission potential of the pests. This knowledge should influence disease management recommendations of taro leaf blight disease. The result of this research would highly contribute to the development of integrated pest and disease management systems in taro production.

MATERIALS AND METHODS

Description of experimental sites: These studies were conducted at three field research sites: screen house and Phytopathology Laboratories at the Institute of Agricultural Research (IRAD), Bambui, in North West Region, (IRAD), Ekona, in South West Region and the International Institute of Tropical Agriculture (IITA) Nkolbisson, Yaoundé, Centre Region of Cameroon. These sites were chosen based on the climatic oscillation and rainfall regime of theses agro-ecological zones. The positions of the experimental sites were recorded using GPS mark Garmin etrex 20. IRAD, Bambui is situated at 32º, 0627′ N latitude, 0659′ E longitude and altitude 1262 m above sea level. IRAD Ekona is located at latitude 32° 0537′ N, longitude 0467′ E and altitude 411 m above sea level and IITA is situated at latitude 32º86′ N, longitude 270′ E and altitude 777 m above sea level. Three agro regions of Cameroon were used for the field studies which were Centre, South West and North West Regions.

Climate of the three agro-ecological regions: Bambui falls under the Western Highlands Plateau agro-ecological region of Cameroon. The climate is tropical, characterised by two seasons: a long rainy season which starts from mid-March and ends in mid-November and a dry season from mid-November to mid-March. Bambui has an annual rainfall of 2145 mm and an average temperature of 21.5^0 C. The soils of the area are ferralistic, vertisol and androsol which are easily eroded and cannot support dense vegetation (Gwanfobe *et al.,* 1983). Ekona falls under the Humid Forest with

monomodal rainfall agro-ecological regions with warm temperate climate. This region has two seasons as describe in the Western Highlands Plateau. Ekona has an average temperature of 23-25^0 C, average rainfall of 2815 mm and the soil consist of basalts because of volcanic activity (Gountie and Monoa, 2013). Nkolbisson, Yaoundé falls under the Humid Forest with bimodal rainfall agro-ecological regions: This zone has Guinea type of climate with four seasons which consist of two periods of rainfall, September to mid-November, March to early July and with two dry seasons, November to March and July to September. In Yaoundé, red ferrelitic and laterites soils are common and are caused by the decomposition of crystalline rock. The average temperature and rainfall are 25^0 C and 1000- 2000 mm respectfully (Gwanfobe *et al.,* 1983).

Pest identification: Pests were collected from taro cultivars showing young lesions of taro leaf blight at the growing sites (IITA Yaoundé, IRAD Bambui and IRAD Ekona) between July and November when environmental conditions (rainfall, relative humidity and temperature) were favourable for reproduction. Pest collection was done between 7.30 and 10.00 a. m daily when the pest were less active in the field due to high relative humidity. Pests were collected using hand picking and sweep net for those that were found on leaves and stems. Soil around infected taro plant was carried and infected corms were harvested. These pests were carried in plastic bags to the entomology laboratory for identification by a crop entomologist. The identification of pest associated with the taro fields was done with the assistance of an entomologist using the insect identification guide. Percentage abundance of pest phyla was calculated and inventory of pest and photographs taken.

Collection, isolation and identification of *P. colocasiae* from infected plant materials: Infected taro leaves with young lesions of taro blight were collected from the field at IITA, Yaoundé, IRAD Bambui and IRAD Ekona, each preserved in separate plastic bags and transported to the Phytopathology laboratory. These leaves were cut with a razor blade in to small fragments of 2 mm from the advancing edges of the disease and surface-sterilized in 5 % diluted solution of sodium hypochlorite for 30 seconds and rinsed in three successive changes of sterile distilled water for 3 minutes. The leaf fragments were dried on sterilized filter paper and four fragments placed on solidified cool culture medium in each Petri dish. These dishes were incubated at room temperature of 21-26 ^{0}C (Brunt *et al.,* 2001). Identification of the fungi was done under the microscope and confirmation made using the procedure describe by Nelson et al. (2011).

Preparation of spore suspensions of *P. colocasiae:* Pests were washed in sterile distilled water and separated into two parts. One part was placed inV6 juice agar medium while the other was surface sterilized in 5 % diluted solution of sodium hypochlorite for 30 seconds and rinsed in three successive changes of sterile distilled water for 3 minute. After drying on sterilized filter papers, the pests were crushed and mixed with 20 ml of sterile distilled water in a small mortar. The extracts were filtered through a sterile muslin cheesed cloth and supernatant placed on V6 juice agar medium containing antibiotics to inhibit bacterial growth. There were 10 plates of five insects per plate. The plates were incubated at ambient temperature of 24-26^0 C for 8 days (Brunt *et al.,* 2001). Plates were observed microscopically for fungi growth (Nelson *et al.,* 2021). The surface of the growing colonies in each Petri dish was flood with 5ml of sterile distilled water and the mycelia dislodged with a small brush. The suspension was centrifuged for 3 minutes, and the supernatant was filtered through a 2 layered sterile muslin cheesed cloth. A drop of Tween 80 per 100 ml inoculum was added as wetting agent.

Assessment of pathogenicity: Local cultivars under investigation were planted in plastic pots

filled with sterilized soil in a screen house. These plants were arranged in a complete randomized design with four replicate of four plants per replicate. The taro was inoculated 49 days after planting with mycelia suspension of *P. colocasiae* obtained from the pests. Inoculation was done by using a syringe to inject the spore suspension on three spots on the leaves. Control plants were treated with sterile distilled water. Observations were carried out and lesion diameter was measured using a ruler. Data for average lesion diameter was recorded for 14 days. Temperature and humidity were also recorded with the Hobo metre (Fokunang *et al.,* 2000).

Statistical analysis: All data collected from percentage occurrence of pest phyla and lesion diameter was subjected to analysis of Variance (ANOVA) using statistical software (J M P 8). Their treatment means were separated using student T-test (STT) and the Least Significant Difference (LSD) at statistical significance of 95 % confidence interval. Mean data were used to plot graphs for appropriate representation of the results.

RESULTS

Pests identified from soil samples, corm samples, sweep net and hand picking in the three experimental field sites: Results of pest collected from corms and soil surrounding the corms showed that *Phenacoccus saenopsis* were absent in Bambui, *Pterostichus melanarius, Tetropium fascum* and *Cordyceps sinensis* in Yaounde and *Gryllus rubens* in Ekona. Adult *Diosophila melanogaster*, *Deroceras reticulatum*, *Lasius niger*, *Glenurus gratus* , *Lumbriscus terrestris* and *Helix aspersa* were found only at Ekona and *Sitophilus zea* only in Bambui. *Lithobius ferticatus* were the common pest in all the three experimental sites (Tables 1, 2 and 3). Results of pest that were caught by hand picking and sweep nets in the three experimental field sites also showed that *Rairanda aurantia* , *Forticula auriculario*, *Helix aspersa* , *Zenocerus variegatus* , *Lasius niger* , *Aphis pomi* , beetles, *Diosophila melanogaster* and *Musca domestica* were common in the three sites. *Gryllus rubens* and *Caenurgina erechtea* were absent in Ekona and *Polyommatus icarus*, *Cordyceps sinensis* in Bambui. *Lithobius ferticatus* were observed only in Yaounde. *Aphis pomi*, *Helix aspersa*, *Forticula auriculario*, *Lasius niger*, *Diosophila melanogaster* and *Musca domestica* were generally more abundant and frequently encountered (Tables 4, 5 and 6). *Aphis pomi* were collected only from stems and leaves while *Phenacoccus saenopsis* and *Piomerus crassipes* were collected from corms and soils. Organisms that were common or found in all the sites were *Lithobius ferticatus*, *Rairanda aurantia*, *Forticula auriculario*, *Helix aspersa*, *Zenocerus variegatus*, *Lasius niger*, *Aphis pomi*, *Pterostichus melanarius, Diphucephala colaspidoides, Tetropium fascum,Diosophila melanogaster* and *Musca domestica*. *Lithobius ferticatus*, *Phenacoccus saenopsis*, *Lumbriscus terrestris* and *Glenurus gratus* were found only on corm or in soils.

Table1: Pest identified from soil and corm samples in the Bambui taro field

Scientific name	Common name	Phylum	Class	Order

Pterostichus melanarius *Diphucephala colaspidoides* *Tetropium fascum*	Black Beetles Green Beetles Brown Beetle	Arthropoda Arthropoda Arthropoda	Insecta Insecta Insecta	Coleoptera Coleoptera Coleoptera
Rairanda aurantia	Spiders	Arthropoda	Arachnida	Araneae
Sitophilus zea	Weevils	Arthropoda	Insecta	Coleopteran
Gryllus rubens	Crickets	Arthropoda	Insecta	Orthoptera
Cordyceps sinensis	Caterpillars	Arthropoda	Insecta	Lepidoptera
Lithobius ferticatus	Centipedes	Arthropoda	Chiplopoda	Craterostigmomorpha

Table2: Pests identified from soil and corm samples at the Yaounde taro field

Scientific name	**Common name**	**Phylum**	**Class**	**Order**
Phenacoccus saenopsis	Mealy bug	Arthropoda	Insecta	Homoptera
Lithobius ferticatus	Centipede	Arthropoda	Chilopoda	Craterostigmomorpha
Julius terrestris	Millipede	Arthropoda	Diplopoda	Spirostreptida
Lasius niger	Black Ants	Arthropoda	Insecta	Hymenoptera
Gryllus rubens	Crickets	Arthropoda	Insecta	Orthoptera
Rairanda aurantia	Spider	Arthropoda	Arachnida	Araneae

Table3: Pests identified from soil and corm samples in the Ekonataro field sites

Scientific name	**Common name**	**Phylum**	**Class**	**Order**
Helix aspersa	Snail	Mollusca	Gastropoda	Pulmonata
Lumbriscus terrestris	Earthworm	Annelida	Oligochaeta	Magadrili
Deroceras reticulatum	Slug	Mollusca	Gastropoda	Pulmonata
Lilioceris lillii *Tetropium fascum*	Red Beetles Brown Beetle	Arthropoda Arthropoda	Insecta Insecta	Coleoptera Coleoptera
Lygaeus kalmii	Red Bugs	Arthropoda	Insecta	Hemiptera
Lithobius ferticatus	Centipede	Arthropoda	Chiplopoda	Craterostigmomorpha
Cordyceps sinensis	Caterpillar	Arthropoda	Insecta	Lepidoptera
Diosophila melanogaster	Fruit fly	Arthropoda	Insecta	Diptera
Julius terrestris	Millipede	Arthropoda	Diplopoda	Spirostreptida
Lasius niger	Black Ants	Arthropoda	Insecta	Hymenoptera
Rairanda aurantia	Spider	Arthropoda	Arachnida	Araneae
Phenacoccus saenopsis	Mealy bugs	Arthropoda	Insecta	Homoptera
Glenurus gratus	Ant lion	Arthropoda	Insecta	Meuroptera

Table4: Pests identified from sweep net and hand picking inthe Bambui taro field sites

Scientific name	Common name	Phylum	Class	Order
Pterostichus melanarius	Black Beetles	Arthropoda	Insecta	Coleoptera
Diosophila melanogaster	Fruit flies	Arthropoda	Insecta	Diptera
Tetranychus urticae	Red spiders	Arthropoda	Insecta	Araneae
Musca domestica	House flies	Arthropoda	Insecta	Diptera
Helix aspersa	Snails	Mollusca	Gastropoda	Pulmonata
Deroceras reticulatum	Slugs	Mollusca	Gastropoda	Pulmonata
Gryllus rubens	Crickets	Arthropoda	Insecta	Orthoptera
Zenocerus variegatus	Grasshoppers	Arthropoda	Insecta	Orthoptera
Forticula auriculario	Ear wigs	Arthropoda	Insecta	Dermaptera
Lasius niger	Black ants	Arthropoda	Insecta	Hymenoptera
Aphis pomi	Aphids	Arthropoda	Insecta	Hymenoptera

Table5: Pests identified from sweep net and hand picking in the Yaounde taro field sites

Scientific name	Common name	Phylum	Class	Order
Rairanda aurantia	Spider	Arthropoda	Arachnida	Araneae
Forticula auriculario	Ear wig	Arthropoda	Insecta	Dermaptera
Helix aspersa	Snail	Mollusca	Gastropoda	Pulmonata
Lithobius ferticatus	Centipede	Arthropoda	Chilopoda	Craterostigmomorpha
Zenocerus variegatus	Grasshoppers	Arthropoda	Insecta	Orthopthera
Lasius niger	Black Ants	Arthropoda	Insecta	Hymenoptera
Aphis pomi	Aphids	Arthropoda	Insecta	Hemiptera
Polyommatus icarus	Butterflies	Arthropoda	Insecta	Lepidoptera
Cordyceps sinensis	Caterpillars	Arthropoda	Insecta	Lepidoptera
Tetropium fascum	Brown Beetles	Arthropoda	Insecta	Coleoptera
Diosophila melanogaster	Fruit Fly	Arthropoda	Insecta	Diptera
Gryllus rubens	Cricket	Arthropoda	Insecta	Orthoptera
Musca domestica	House flies	Arthropoda	Insecta	Diptera

Table 6: Pests identified from sweep net and hand picking in Ekona taro field sites

Scientific name	Common name	Phylum	Class	Order
Forticula auriculario	Ear wig	Arthropoda	Insecta	Dermaptera
Julius terrestris	Millipede	Arthropoda	Diplopoda	Polydesmida
Helix aspersa	Snail	Arthropoda	Gastropoda	Pulmonata
Zenocerus variegatus	Variegated grasshopper	Arthropoda	Insecta	Orthoptera
Rairanda aurantia	Spiders	Arthropoda	Arachnida	Araneae
Lasius niger	Black ants	Arthropoda	Insecta	Hymenoptera
Solenopsis geminata	Red ants	Arthropoda	Insecta	Hymenoptera
Pterostichus melanarius	Black beetles	Arthropoda	Insecta	Coleoptera
Lilioceris lilii	Red Beetles	Arthropoda	Insecta	Coleoptera
Piomerus crassipes	Assassin bugs	Arthropoda	Insecta	Hemiptera

Aphis pomis	Aphids	Arthropoda	Insecta	Homoptera
Cordyceps sinensis	Caterpillars	Arthropoda	Insecta	Lepidoptera
Diosophila melanogaster	Fruit flies	Arthropoda	Insecta	Doptera
Musca domestica	House fly	Arthropoda	Insecta	Doptera
Caenurgina erechtea	Moths	Arthropoda	Insecta	Lepidoptera
Polyommatus icarus	Butterfly	Arthropoda	Insecta	Lepidoptera

Percentage abundance of pest phyla identified from soils, corms, sweep net and hand picking in three experimental field sites: A study and classification of pests identified from soil and corms associated in the transmission of *P.colocasiae*in three experimental sites showed that arthropods were the most represented phyla with maximum mean occurrence of 100 % in Bambui and Yaounde and minimum mean occurrence of 7.69 % in the phylum Annelida in Ekona (fig1). Results of mean occurrence of pest phyla identified from sweep net and hand picking in the three experimental sites showed that highest mean percentage occurrence of 92.31 % was recorded in phylum Arthropoda at Ekona and minimum mean percentage occurrence of 7.69 % in phylum Mollusca at Ekona (Fig 2).

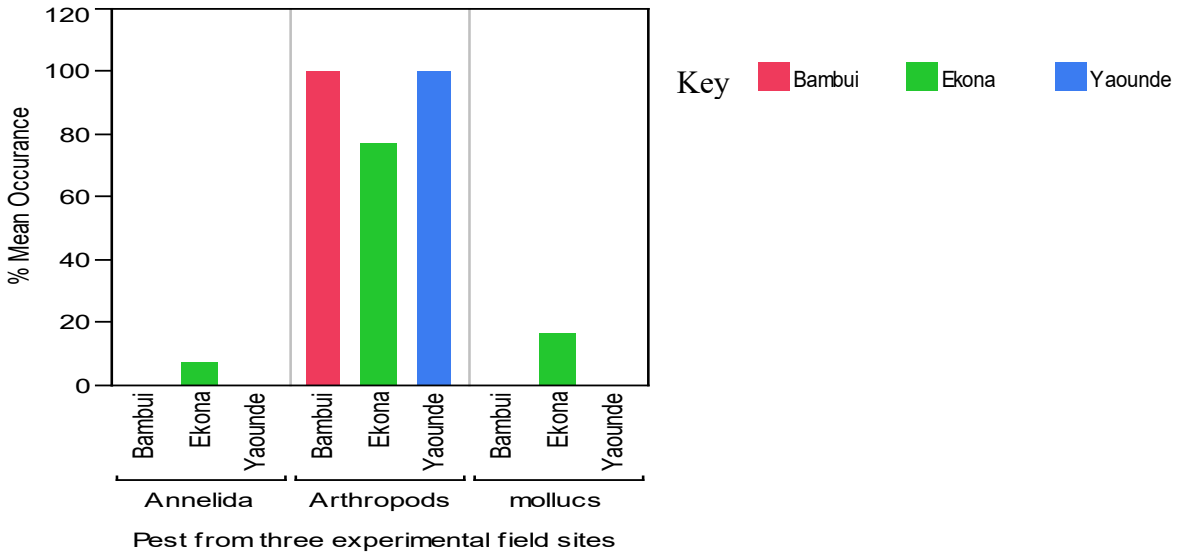


Fig.1: Means percentage abundance of pest phyla identified from soils and corms in three experimental field sites

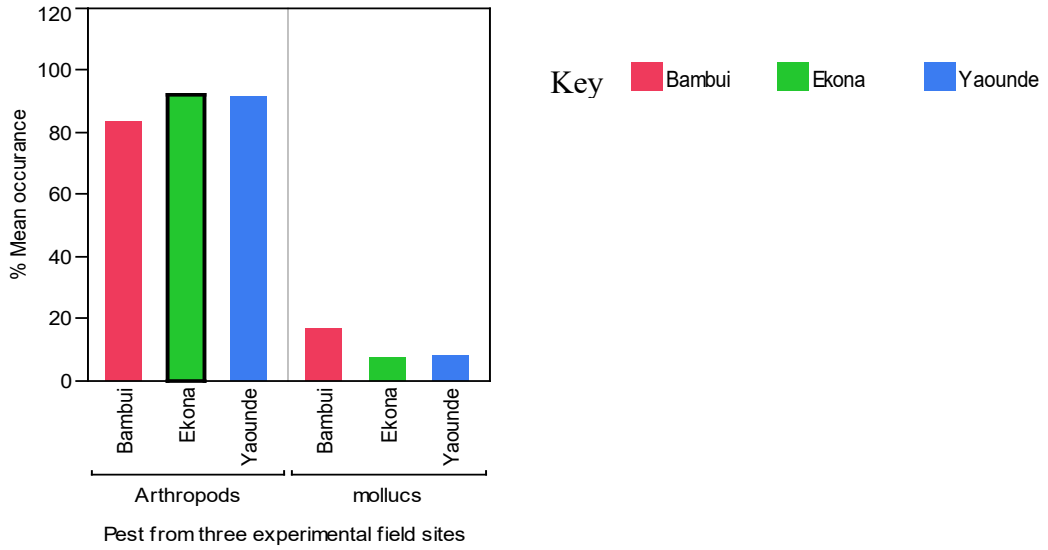
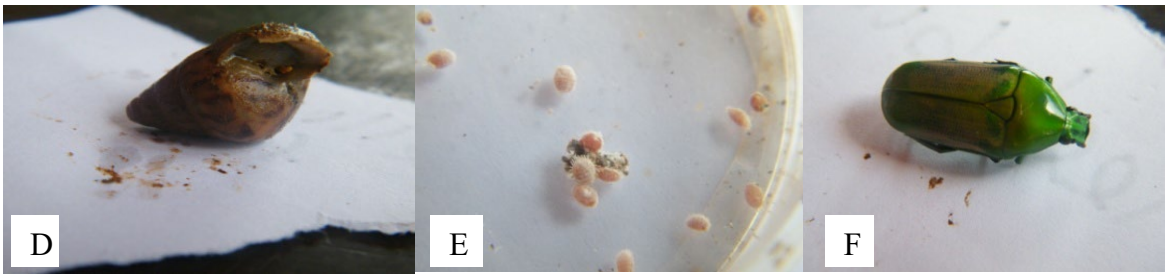


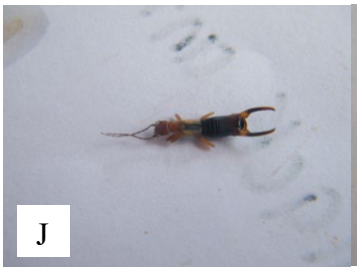
Fig.2: Means percentage abundance of pest phyla identified from sweep net and hand picking in three experimental field sites.

Polyommatus icarus *Lithobius forticatus* *Diachlonis ferrugatis*



Helix aspersa *Phenacoccus solenopsis* *Diphucephala colaspidoides*

Pterostichus melanarius *Lasius niger* *Cordyceps sinensis*



Forficula auricularia

Gryllus rubens

Omocestus viridulus

Julus terristris

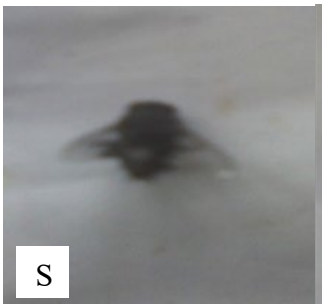
Aphis pomi

Zonocerus variegatus

Simulium spp

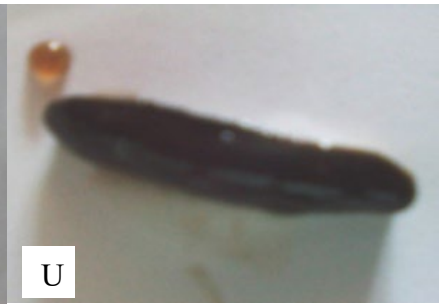
Solenopsis geminata

Miranda aurantia



Musca domestic

Epidendrum magnoliac

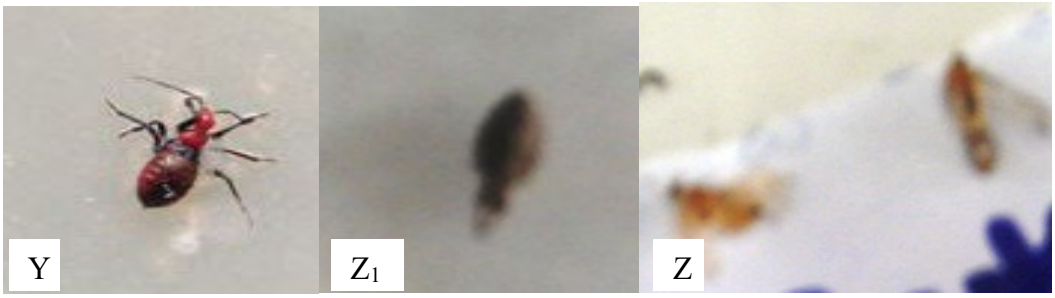


Deroceras reticulatum

Tetropium fascum

Lumbrius terrestris

Gryllus rubens



Lygaeus kalmii *Caenurgina erechtea* *Glenurus gratus*

Fig.3: Pest of taro (A to Z_2) harvested in three experimental fields (Ekona, Yaounde, Bambui)

DISCUSSION

All the pests' culture inV6 juice agar medium showed no fungi growth except Aphids with very little fungi growth. No disease symptom was expressed by plants inoculated with spores from the aphid culture, indicating that the concentration of the pathogen in the inoculum could not cause infection. However, in the field the insect pest feed on taro leaves, stems and corms, creating wounds on the plant organs on which they feed. These wounds enable pathogen to penetrate these organs causing taro diseases. When pests feed from one plant to the other, they carry the pathogen along and the wounds which they create favour the growth of *P. colocasiae* which is transmitted by wind and rain water from infected plants in to these plants. The disease manifestation of *Phytophthora colocasiae* in taro is a leaf blight disease characterized by irregular lesions which expand and coalesced to destroy large areas of leaves (Manju *et al.,* 2017). Pests found on taro corms and soils surrounding corms createwounds on taro corms before harvesting, on storage there is much deterioration of corms due to attack by pathogens like fungi and bacteria which enter the wounds. Van der Bruggen et al. (1987) found fungus penetrating through insect puncture wounds on young cassava stems, and extend through the cork layer, the epidermis and other tissues. Pests live in soil and some feed on roots of plants damaging the roots and causing wilting of plants. The synergistic effect of roots damaged and leaves damaged by fungi lead to crop failure. Reports from Deo et al. (2009) states, the adult beetles create tunnels into the soil just at the base of the taro corm, and feed on the growing corm leaving large holes that reduce the crop quality, cause wilting, death and the wounds they create during feeding promote the attack by rot-causing organisms. Other insect pests include taro leafhopper (*Tarophagus proserpina*), which transmits viruses and causes wilting and death of the plant in heavy infestations. The hawk-moth larva causes defoliation of plants and the armyworm or cluster caterpillar, damage the leaves extensively (Carmichael *et al.,* 2008). Aphids are sucking pests that were abundant and frequently encountered. There exist a mutual association between the aphids and ant where aphids feed on plant sap from leaves, petiole and secrets honey dews which sweetish, ants feed on this and also render protection for aphids. Aphids can have a dramatic negative impact on their host plants partly due to their capacity for extremely rapid population growth (Dixon 1987). Aphids reproduce rapidly and have been shown to adapt quickly to host-plant phenology and ecology, as well as plant physiology and biochemistry (Pattersson *et al.,* 2007). They can result in direct damage to crops through feeding on phloem tissue; they can also contribute to severe indirect damage by acting as primary vectors of many plant viruses. They ingest phloem sap from their hosts through narrow piercing–sucking mouthparts called stylets. During probing, aphids' stylets transiently puncture epidermal, mesophyll and

parenchyma cells, and this mechanical damage may influence plant responses to infestation (Delahau, 2005).

CONCLUSION AND APPLICATION OF RESULTS

Pests associated in taro fields that were screened, have the potential to feed on leaves, stems and corms of taro plant, thus creating wounds on the plants which predisposes the penetration of pathogens (*P.colocasiae*) carried by wind, rain and bodies of pests. Thus data from this current study can enhance integrated pest management practices, by facilitating identification of pest that transmits *P.colocasia*e, to inform best management practices that can reduce diseases severity and incidence in *Colocasia* crop production.

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Conflict of interest

Authors have declared no conflict of interest

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