DISEASE SURVEY OF *CNIDOSCOLUS ACONITIFOLIUS* (MILL.) I. M. JOHNSTON IN RIVERS STATE, NIGERIA

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ABSTRACT

Field survey was carried out in three senatorial zones in Rivers State to determine the disease incidence and disease severity of *Cnidoscolus aconitifolius*; a leafy vegetable mostly consumed for its haematological properties in the South-eastern part of Nigeria. The survey parameters observed and recorded were: location of sample collection, survey collector/Accession number, date of sample collection, total number of diseased plants, total number of diseased leaves on plant, disease symptoms observed on plants, disease Incidence (DI) and disease severity (DSV). Emuoha Local Government Area (LGA) showed the highest disease incidence and disease severity (53.31%) across all the LGAs surveyed while Rivers East showed the highest disease incidence and disease severity (42.71% / 43.18%) among the three senatorial zones surveyed. The symptoms that occurred mostly in all the three senatorial zones include leaf spots, chlorosis, mosaic, leaf crumple, necrosis and wilting. The level of disease incidence and disease severity shows that *Cnidoscolus aconitifolius* provides a favourable environment for the survival and growth of microorganisms some of which may be detrimental to the plant health. In this study, the methods used to carry out the survey and the recommendations made on how to develop appropriate disease management strategies are outlined.

Keywords: *Cnidoscolus aconitifolius*, Disease survey, Disease incidence, Disease Severity, Symptoms

Introduction

Cnidoscolus aconitifolius (Mill.) I. M. Johnston commonly called "tree spinach" is a leafy perennial shrub that is resistant to drought. It originated from southern Mexico and Central America (Ross-Ibarra & Molina-Cruz, 2002). *Cnidoscolus aconitifolius* is widely distributed in the temperate and tropical zones of the world. In some African countries, *Cnidoscolus aconitifolius* are not found growing in the wild, it is only cultivated locally at experimental stations (PROTA, 2019) Even though tree spinach has been in existence in the Philippines since 1954, it has not yet spread into the wild as only two collections have been made outside cultivation (Van-Welzen & Fernández-Casas, 2017). Today, *Cnidoscolus aconitifolius* is found growing in the wild and also in cultivation in all parts of Nigeria. Tree spinach is drought-tolerant and can grow in all soil types including infertile soils, but cannot tolerate high soil salinity. Although it can tolerate a pH between 4.5 and 7.5, it grows better at a pH between 5.5 and 6.5, but can also grow in highly acidic and alkaline soils. It can grow at temperatures of 12 to 38°C. *Cnidoscolus aconitifolius* occasionally tolerate inundations and requires a mean annual rainfall of 500 to 2500mm (PFAF, 2019; Useful Tropical Plants, 2019).

aconitifolius is commonly used as a vegetable in soups in south-western Nigeria (Oyagbemi & Otedola, 2013). *Cnidoscolus aconitifolius* leaves have been reported by many authors to have hepatoprotective, and anti-hypertensive properties (Lucky & Festus, 2014; Chukwu *et al*., 2018). Hamid *et al*. (2016) reported that the aerial parts of *Cnidoscolus aconitifolius* exhibit antimicrobial activities. The plant is also used as a blood booster in the Eastern and Southern parts of Nigeria where it is usually consumed by pregnant women and anemic children. Young leaves and shoot are squeezed with water, the water is sieved and drank. Mordi & Akanji (2012) reported that *Cnidoscolus aconitifolius* have antihaemorrhagic, antihypertensive and cardiac depressant properties. According to Orji *et al*. (2016), *Cnidoscolus aconitifolius* contains phenols which are well-known anti-cancer agents with high ability to fight against cancer. *Cnidoscolus aconitifolius* is used as a hedge to protect crops from farm animals. It is also used in the rural areas as a biological fence to barricade homes and mark boundaries between compounds and farm lands. Tree spinach is also used as an animal feed. The leaves can be used as an ingredient in poultry diets. The edible leaves have the potential to be canned or frozen (Growables, 2019).

Plant disease is a deviation from the normal functioning of a plant. It hinders plants from attaining optimum growth by negatively affecting their vital functions. Plant diseases lead to reduction in both the quality and quantity of agricultural products and this may result in starvation and hunger especially in under developed countries (Vipinadas & Thamizharasi, 2015). Visually observable abnormal patterns on plants are referred to as plant disease symptoms. In the case of leafy vegetables, the detection of diseases on leaves is very important be[these diseases generally lead to reduction in crop yield. Reduced production of vegetables is largely due to disease incidences (Patel *et al*., 2020). To a great extent, increase in the yield of crops can be attributed to enhanced plant disease and pest management. Sustainable crop protection is of great economic importance for animal feed and food production for man (Flood, 2010). The aim of this study is to carry out a disease survey of *Cnidosolus aconitifolius* growing in the wild and in cultivation in Rivers State, Nigeria.

Experimental

Disease survey and data collection

The disease survey was carried out in the three senatorial zones in Rivers State; Rivers East, Rivers West and Rivers South East. The Local government areas (LGAs) surveyed in each senatorial zone are shown in Figure 1. Surveys were carried out during the rainy season (June to August) in fields where *Cnidoscolus aconitifolius* were growing. The following survey parameters were observed and recorded; location of sample collection, LGA, survey collector/Accession number, Date of collection, Total number of diseased leaves on plant, Disease symptoms observed on plants, Disease Incidence (DI) and Diseases Severity (DSV). *Cnidoscolus aconitifolius* plant is presented in Figure 1. A map of Rivers State showing the LGAs and the three Senatorial zones surveyed for diseases is shown in Figure 2.

Fig. 1: *Cnidoscolus aconitifolius* plant

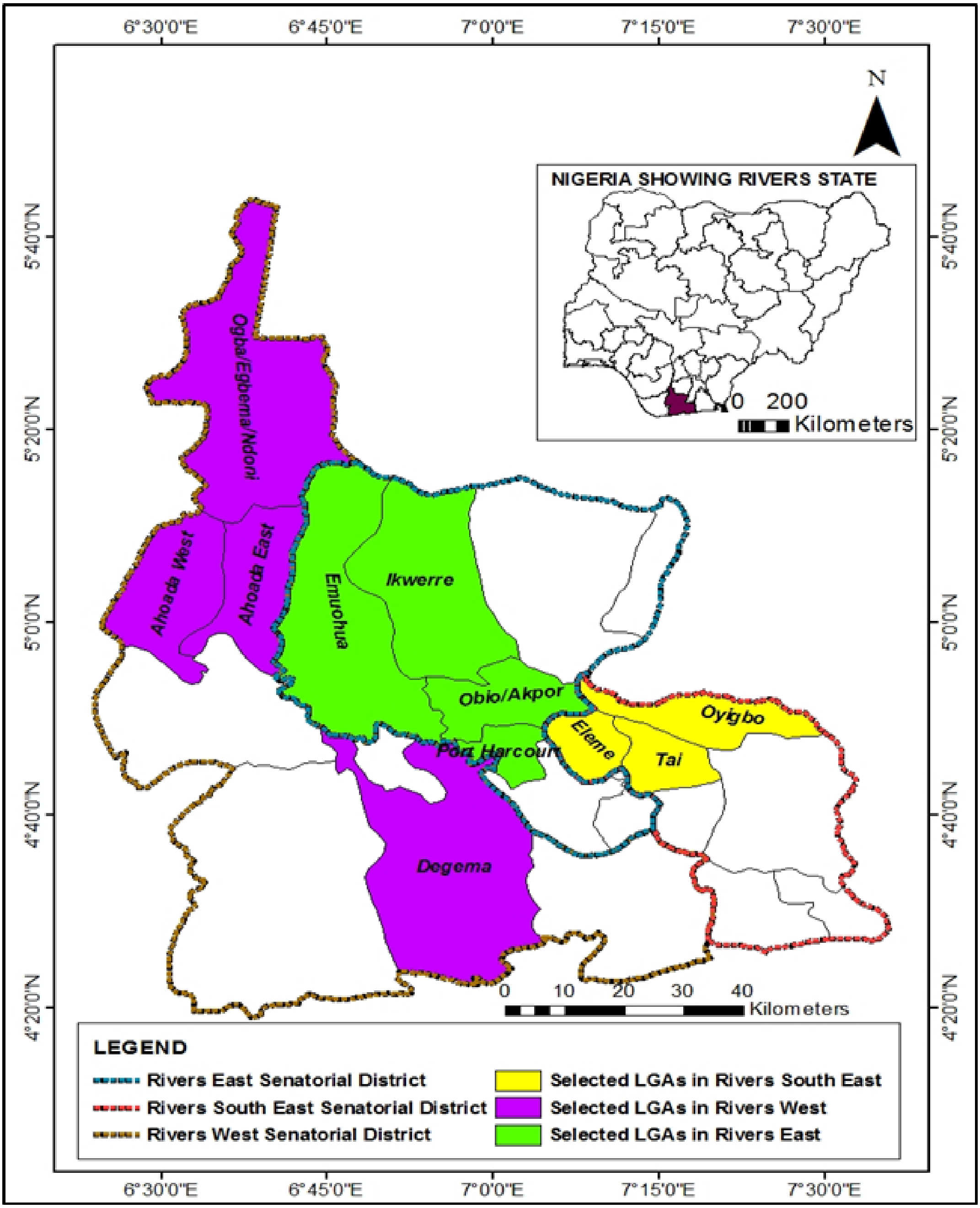


Fig. 2: Map of Rivers State showing the LGAs and the three Senatorial zones surveyed for diseases

The disease symptoms observed on plants were recorded. The disease incidence and disease severity were calculated based on the method employed by (Wheeler, 1969).

Disease incidence (DI) = n/N x 100%

$$DI = \frac{\text{Number of plants affected (n)}}{\text{Number of plants observed (N)}} \times 100\%$$

Where; DI = Disease Incidence,

n = number of plants showing symptoms, and

N = the total number of plants observed

$$\text{Disease severity (DS)}: \frac{\Sigma(ni.vi)}{N \times V} \times 100\%$$

ni = Number of plants affected at category level i

vi = Damage category level i

N = Number of plants observed

V = Highest damage category value

Damage category value:

0 = No symptom

1 = 0 < X 25%

2 = 25 < X <50%

3 = 50 < X < 75%

4 = X > 75%

Data analysis

Other data obtained from the study were analyzed using One-way Analysis of Variance (ANOVA) ($P<0.05$) and multiple comparisons using Least Significant Difference (LSD) with the aid of Statistical Package for Social Science (SPSS) software version 23.

Results

Disease Symptoms Observed on Cnidoscolus aconitifolius Plants

Different symptoms were observed on *Cnidoscolus aconitifolius* plants in the senatorial zones surveyed. The symptoms include: mosaic (mottling of dark and light green or yellow patches on leaves), wilting (vein clearing, drooping of leaves), necrosis (death of plant tissues), chlorosis / yellowing of leaves (pale yellow discolouration of green tissues), swollen leaves, reduced leaf size, hollows on leaves, anthracnose, leaf crumple, leaf spots, leaf curl and powdery mildew. The symptoms observed on *Cnidoscolus aconitifolius* are presented in Figure 3.

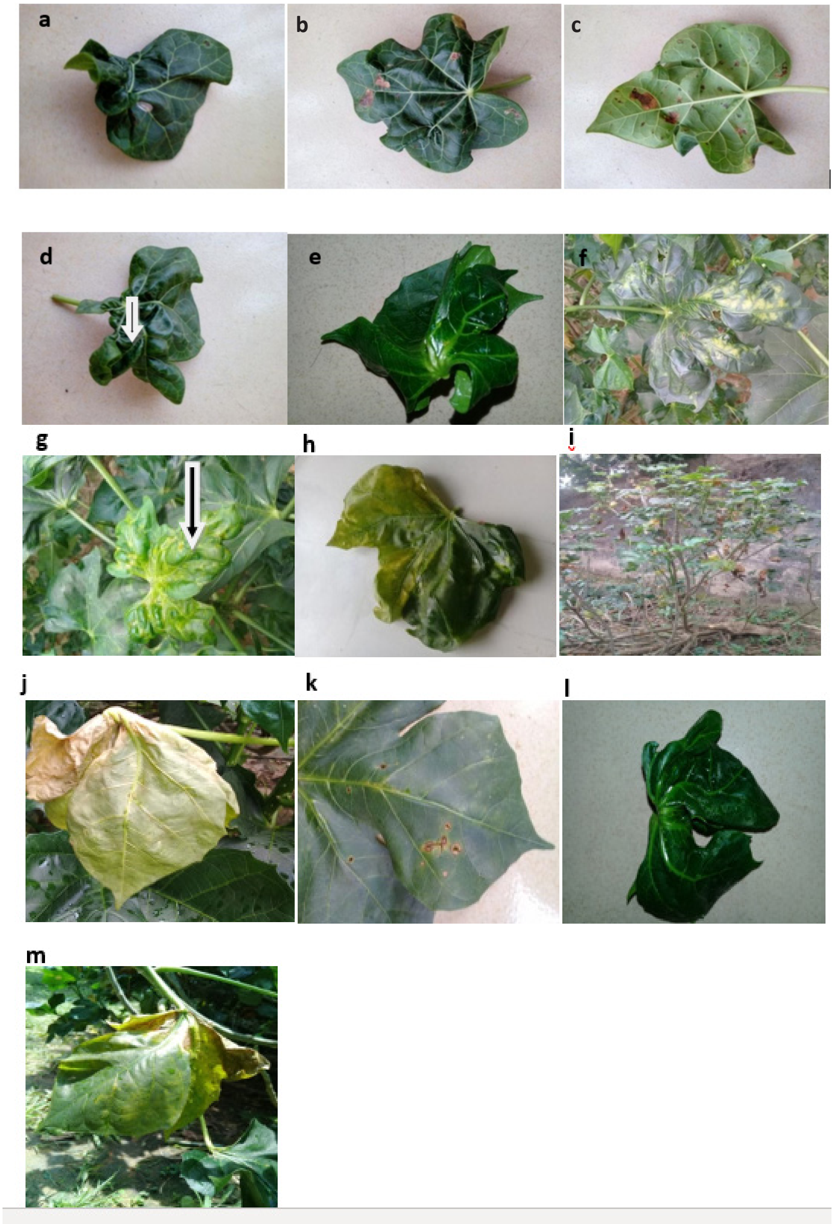


Fig. 3: Disease symptoms observed on *Cnidoscolus aconitifolius* leaves :-(a) Powdery mildew (b) Anthracnose (c) Necrosis (d) Leaf crumple (e) Leaf curl (f) Mosaic pattern (g) Swollen areas (h) Chlorosis (i) Defoliation/ leaf dropping (j) Leaf blight (k) Brown leaf spots with hollows (l) Reduced leaf size (m) wilting (vein clearing, drooping of leaves)

Disease Incidence and Disease Severity of Cnidoscolus aconitifolius in Rivers State

The disease incidence and disease severity was calculated for each local government in the three Senatorial zones where disease surveys were carried out. There was a significant difference (P < 0.05) in the incidences within the LGAs and within the Senatorial zones (Figure 4).

In Rivers East Senatorial zone (Emohua, Obio-Akpor, Ikwere and Port Harcourt Local government areas), Emohua LGA showed the highest disease incidence of 53.3%, while Port Harcourt LGA (32.5%) had the lowest disease incidence. In Rivers West Senatorial zone, Ogba/ Egbema/Ndonni LGA (29%) had the highest disease incidence followed by Ahoada West (24.4%), Degema (23.5%) and Ahoada East (16.6%). In Rivers South-east Senatorial zone (Tai, Eleme and Oyigbo LGAs), Eleme LGA had the highest disease incidence while Tai (19.4%) had the lowest disease incidence.

There was a significant difference (P < 0.05) in disease severity within the LGAs and within the senatorial zones. In Rivers East senatorial zone, Emohua LGA showed the highest disease severity of 53.3%, Ogba/Egbema/Ndoni LGA showed the highest disease severity of 29% in Rivers West senatorial zone while Eleme LGA showed the highest disease severity of 42.4% in Rivers South East senatorial zone (Figure 4).

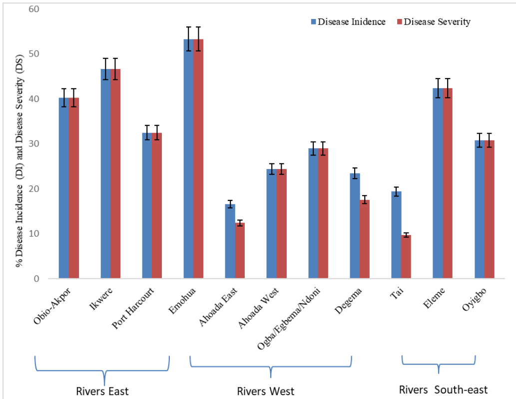


Fig. 4: Disease incidence and severity for each Local Government Area in Rivers State Disease Severity Indexes (DVI) was recorded for the three senatorial zones using a severity scale presented as heatmaps in Tables 1 to 3.

TABLE 1

Heatmap of Disease Severity Index (DVI) of disease survey in Rivers East Senatorial zone

S/N	Disease Symptoms observed	15-25%	26-35%	36-45%	46-55%	56-65%	66-75%	76-100%
1	Blight							
2	Chlorosis							
3	Hollows							
4	Leaf crumple							
5	Leaf dropping							
6	Leaf spots							
7	Mosaic							
8	Necrotic lesion							
9	Powdery mildew							
10	Reduced leaf size							
11	Swollen leaf							
12	Wilting							

15-25%
26-35%
36-45%
46-55%
56-65%
66-75%
76-100%

TABLE 2

Heatmap of Disease Severity Index (DVI) of disease survey in Rivers West Senatorial zone

S/N	Disease Symptoms observed	15-25%	26-35%	36-45%	46-55%	56-65%	66-75%	76-100%
1	Blight							
2	Chlorosis							
3	Hollows							
4	Leaf crumple							
5	Leaf dropping							
6	Leaf spots							
7	Mosaic							

8	Necrotic lesion							
9	Powdery mildew							
10	Reduced leaf size							
11	Swollen leaf							
12	Wilting							

Key:

15-25%
26-35%
36-45%
46-55%
56-65%
66-75%
76-100%

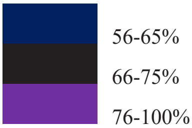
TABLE 3

Heatmap of Disease Severity Index (DVI) of disease survey in Rivers South-east Senatorial zone

S/N	Disease Symptoms observed	15-25%	26-35%	36-45%	46-55%	56-65%	66-75%	76-100%
1	Blight							
2	Chlorosis							
3	Hollows							
4	Leaf crumple							
5	Leaf dropping							
6	Leaf spots							
7	Mosaic							
8	Necrotic lesion							
9	Powdery mildew							
10	Reduced leaf size							
11	Swollen leaf							
12	Wilting							

Key:

15-25%
26-35%
36-45%
46-55%



There was no significant difference between the disease incidence and disease severity in all the three Senatorial zones surveyed. It was observed that an increase in disease incidence lead to a corresponding increase in disease severity. Rivers East Senatorial Zone had the highest disease incidence and severity as presented in Figure 5.

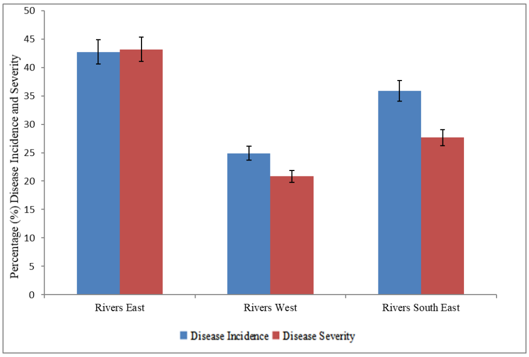


Fig. 5: Disease incidence and severity in Rivers State

Discussion

Disease survey of *Cnidoscolus aconitifolius* revealed that several symptoms were associated with the plant. The various symptoms observed on *Cnidoscolus aconitifolius* leaves include: mosaic, wilting, necrosis, chlorosis/ yellowing of leaves, swollen leaves, reduced leaf size, hollows on leaves, anthracnose, blight, leaf crumple, leaf spots, leaf curl and powdery mildew.

Emuoha Local Government Area showed the highest disease incidence and disease severity (53.31%) across all the LGAs surveyed while Rivers East showed the highest disease incidence and disease severity (42.71% / 43.18%) among the three senatorial zones surveyed. The symptoms that occurred mostly in all the three senatorial zones include leaf spots, chlorosis, mosaic, swollen leaves, leaf crumple, necrosis and wilting. To our knowledge, this stands as the first detailed report of these symptoms on *Cnidoscolus aconitifolius* in Rivers State.

Mosaic, yellowing, leaf rolling, and curling are virus-like symptoms observed on plants (Mwakosya *et al*., 2021). Symptoms of mosaic disease includes: vein clearing of new trifoliolate leaves, localized lesions on leaves, downward curling of leaf margins and upward curling of leaf tips, infected leaves become leathery and brittle, stunted growth and mottling of infected leaves (Brand *et al*., 1993; Hajimorad *et al*., 2005). Mosaic disease has been reported on Columbus grass, pumpkin plants and *Canna* sp. in United States and China (Mollov *et al*., 2016; Zhao *et al*., 2019; Tang *et al*., 2019).

Plant diseases such as wilt, anthracnose, rust, mildew, blight, leaf spot, damping-off, coils, scab, canker, gall, root rot and dieback are caused by fungi (Jain *et al*., 2019). Wilt disease is also caused by some bacterial pathogens. Timely identification of the pathogens responsible for the manifested symptoms on plants or plant produces may help in preventing the spread of the disease. The harmful effect of pathogens on plants is a component of plant disease management process (Baiyee *et al*., 2019).

Leaf spots occur as white or grayish-white areas on the edges, enclosed by brownish, reddish brown or yellowish margins (Sarsaiya *et al*., 2019). They are first visible on the upper (adaxial) leaf surface. The spots later degenerate into holes or hollows of different shapes and diameter. The younger leaves are first infected and show symptoms in the form of lesions. Overtime, some of the spots may merge and form blotches (Mattihalli *et al*., 2018). Leaf spot disease caused by *Cercospora sojina*, *Fusarium* sp. and *Puccinia arachidis* has been recorded on soyabean, mango and peanut respectively (Zeng *et al*., 2017; Mahmud *et al*., 2019; Chen *et al*., 2019).

Wilt is an economically important disease of crop plants. It is caused by both fungal and bacterial pathogens. It occurs as a result of water loss in plant stems and leaves. In the early stage of the disease, the older leaves become yellow while the younger leaves still retain their green colour. As the disease advances, the pathogen spreads throughout the vascular tissues of the host and plant parts like the leaves loose turgidity and droop. Disease advancement is facilitated under water stress (Devappa & Archith, 2019). *Fusarium oxysporum* and *F. solani* have been reported to be responsible for wilt disease on cucumber, melon, pumpkin, pea, bean, cotton, eggplant, tomato, potato and other Solaneceae (Nalini & Parthasarathi 2018; Singh *et al*., 2017; Devappa & Archith, 2019; Cox *et al*., 2019). The fungal spores spread through equipment, soil, air, plant debris, and seeds. The spores are difficult to eliminate from infested fields and plants as they can persist in the soil for several years.

The bacterial wilt pathogen, *Pseudomonas solanacearum* (*Ralstonia solanacearum*) is spread through water and soil. Infection also occurs through plant roots (usually through wounds or during lateral roots development) and transplanting infected plants. (Jones *et al.*, 2017). Disease spread from plant to plant occurs when the bacterium moves from the roots of infected plants to the roots of close by healthy plants usually through irrigation practices (Swanson *et al*., 2005). In tomato plants, disease spreads rapidly leading to plant death within 4 – 7 days after the first manifestation of symptoms (Jones *et al.*, 2017). Symptoms develop and the plant rapidly dies under favorable conditions (Coutinho, 2005; Cook and Floyd, 2020).

Leaf blight disease is indicated by yellowish (chlorotic) leaf margins and tips. Necrosis develops and spreads, forming dark-brown patches of various sizes. As the tissues die, the lamina eventually dies. Blight disease usually affects older leaves than younger leaves (Li *et al*., 2019). The leaf blight pathogens, *Bipolaris maydis*, *Cochliobolus heterostrophus* and *Xanthomonas oryzae* have been reported on maize, southern corn and rice respectively (Kumar *et al*., 2016; Mubeen *et al*., 2017; Wongkhamchan *et al*., 2018).

The oomycete, *Phytopthora infestans* is responsible for late blight disease in plants. Early infection is in form of gray and green spots on older leaves surfaces. The spots become darker as the disease advances leading to the formation of white mycelial masses on the abaxial (lower) leaf surfaces until the whole plant is colonized and completely infected. The pathogen is spread through contaminated planting materials such as seeds and tubers (Liljeroth *et al*., 2016). Late blight disease has been reported on potato in Netherlands, Sweden and Hungary (Small *et al*., 2015; Liljeroth *et al*., 2016; Hajianfar *et al*., 2016).

Early blight disease caused by *Alternaria solani* usually affects tomato and potato plants. Following infection, symptoms manifest as lesions on older leaves. With a progress in disease development, the lesions widen and the leaves turn yellow, droop (wither) and die. The infection may be systemic where it spreads to other plant parts. The pathogen is transmitted by irrigation, rain, farm implements, contaminated plant tissues, planting materials (tubers and seeds) and insects. Plants undergoing stress and malnourished plants are usually at a higher risk of this disease (Babu *et al*., 2015). Early blight disease has been recorded on potato in Basel and Denmark (Nassar *et al*., 2016; Abuley & Nielsen, 2019).

The fire blight pathogen, *Erwinia amylovora* infects apple, pear, cotoneaster, and other plants of Rosaceae family (Flachowsky *et al*., 2012; Gaucher *et al*., 2013). The bacterium infects the host through natural openings (eg. nectarthodes) and also through wounds on humid aerial organs. Some plant leaves may have scorched appearance. The pathogen only causes local infection and manifests as necrotic areas on plant tissues (Flachowsky *et al*., 2012).

Powdery mildew is caused by various fungi, each with its own host range. The infection starts with budding leaves which develop blisters making them to curl upwards. Infected leaves become covered with white to grayish dusty or powdery mass. As the diseases progresses, the leaves become necrotic and fall off. Flower buds infested with powdery mildew may be inhibited from opening. The pathogen usually infects younger leaves with high moisture content (Bai *et al*., 2019). Powdery mildew caused by *Oidium neolycopersici*, *Sphaerotheca macularis* and *Erysiphe necator* have been reported on cucumber, strawberry and grapevine respectively (Bai *et al*., 2019; Mahmud *et al*., 2019; Goyal *et al*., 2019).

Systemic foliar pathogens are major causes of low yield, commercial crop losses and diminished crop quality (Iqbal *et al*., 2018). A better understanding of the organisms responsible for plant disease symptoms and the application of various control measures will go a long way in harnessing the problems associated with crop losses due to plant diseases. Molecular tools which generate vast data can be used to accurately identify the organisms responsible for disease symptoms in plants, although these tools require sophisticated equipment and well-trained personnel to use them (Jain *et al*., 2019). A number of preventive and control measures can be employed to manage diseases in plants and control crop pests in order to increase crop yield. This includes the use of phytosanitation involving the selection of cuttings for propagation exclusively from symptomless plants, adjusting cropping practices to decrease the risk of infection, the removal (roguing) of diseased plants from partly infected stands, proper disposal and burning of crop debris to decrease the risk of infection, the use of resistant varieties, bio-stimulants (or defense activators) and the use of plant extracts.

Conclusion

Various disease symptoms were observed on *Cnidoscolus aconitifolius* during the survey period in the field. The level of disease incidence and disease severity shows that *Cnidoscolus aconitifolius* provides a favourable environment for the survival and growth of microorganisms.

It is important to carry out surveys in other States in areas where *Cnidoscolus aconitifolius* is grown to cover the six geo-political regions of the country. Other methods of detection of disease symptoms can also be used. In this study, visual method was used to detect leaf diseases; in further studies, we shall consider using other types of vegetable leaf disease detection techniques such as: image processing and deep learning. The survey should also be carried out in different seasons to determine whether there would be a significant difference between the two seasons. The relationship between disease severity and plant yield loss would also be ascertained in further studies.

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