OPEN ACCESS Int. J. Phytopathol. 09 (03) 2020. 179-186 DOI: 10.33687/phytopath.009.03.3457

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Available Online at EScience Press

International Journal of Phytopathology

ISSN: 2312-9344 (Online), 2313-1241 (Print)

https://esciencepress.net/journals/phytopath

IMPACT OF FARM MANAGEMENT PRACTICES ON DOWNY MILDEW DISEASE OF CUCUMBER IN HIGH TUNNELS

[a]Muhammad N. Shahid, [a]Muhammad Abid, [a]Sobia Chohan, [b]Muhammad Binyameen, [a]Muhammad Sajid

[a] *Department of Plant Pathology, Faculty of Agricultural Sciences & Technology, Bahauddin Zakariya University, Multan, Pakistan.*

[b] *Department of Entomology, Faculty of Agricultural Sciences & Technology, Bahauddin Zakariya University, Multan, Pakistan.*

ARTICLE INFO

Article history

Received: October 12, 2020

Revised: December 23, 2020

Accepted: December 27, 2020

Keywords

Downy mildew

Pseudoperonospora cubensis

Cucumber

Plant resistance

Sowing time

High tunnels

ABSTRACT

Downy mildew of cucumber caused by *Pseudoperonospora cubensis*is is a serious threat to the cucurbits production in high tunnels. In this study, the disease incidence and severity were recorded from nine high tunnel farms, located at Vehari District, Pakistan. The results revealed that downy mildew incidence was ranged from 24 to 80%. The disease development was correlated with plant genotype, sowing time, commonly used fungicides, and plant growth stages. "Kaan" was the most resistant while "Yayla" was the highly susceptible plant genotype against the disease. Cucumber sowing at mid-November showed significantly less disease over early and late sowing. Flowering was the most vulnerable stage of plants for disease development. 'Champion' (Copper hydroxide) followed by 'Score' (Difenoconazole) fungicides reduced the disease to 55% and 43%, respectively, which increased the yield 8-15 %. We conclude that the use of resistant genotype "Kaan", protectant fungicide 'Champion' and appropriate sowing time Mid-November may reduce the disease incidence appreciably in high tunnels.

Corresponding Author: Muhammad Abid

Email: muhammad.abid@bzu.edu.pk

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INTRODUCTION

Cucumber (*Cucumis sativus* L.) is a widely cultivated member of the Cucurbitaceae family in various regions of the world. It is one of the widely consumed vegetables and is common for its plentiful nutritional value, dominant health care benefits and its wonderful taste (Ando *et al.*, 2012; Cohen *et al.*, 2011). It was cultivated at more than 2 million hectares in 2019 around the globe with 87810000 tons production and ranked 4th among other vegetable production (FAO, 2019). Downy mildew of cucurbits is one of the most important foliar diseases in the tunnels due to their humid environment (Shetty *et al.*, 2002). This disease is caused by obligate parasitic oomycete fungi *Pseudoperonospora cubensis* (Berk and Curt.) Rostow (Palti and Cohen, 1980).

Downy mildew of cucumber first appears as water-soaked small lesions on the underside of leaves. Later, these lesions turn chlorotic and dark coloured with fungal spore on the underside of leaves. These chlorotic spots of bad skin turn necrotic and these lesions are often angular shape where they edge leaf veins, and gradually the whole leaf becomes necrotic and die (Palti and Cohen, 1980; Thomas, 2016). Environmental conditions and farm practices play a very important role in disease intensity (Cohen, 1977; Cohen *et al.*, 2011). Leaf wetness is also a serious threat to the development of disease (Gupta and Gupta, 2018). Unavailability of free moisture on the leaf will suppress the sporangia germination. Rainfall, dew development or irrigation, epitome hotness is good for sporulation and subsequent infection at a temperature ranging between 5 °C and 30 °C (Lebeda and Widrlechner, 2003). The environmental conditions are highly important in downy mildew disease spread and influence its intensity (Ding *et al.*, 2016).

Downy mildew of cucurbits has been reported in seventy countries on various species of *Cucumis* genus all around the world (Shetty *et al.*, 2002). This disease causes about 40% yield losses annually around the globe. Colucci (2008) reported 32 different hosts of *Pseudoperonospora cubensis* from USA and Canada during 2006 and 2007, and *Cucumis* and *Cucurbita* as the most susceptible genera. The management of disease using pesticides increases resistance in pathogens, increase production cost and pollute the environment. The current requirement of our agriculture is to design integrated pest management (IPM) strategies to provide an eco-friendly solution to the diseases. The use of resistant genetic resources, planting time, preference and efficacy of pesticides, physical and biological measures are an important parameter to develop integrated pest management strategies (Juroszek and von Tiedemann, 2011; Ketta *et al.*, 2016; Thomas, 2016).

Farming of vegetables in high or low tunnels is a highly efficient way to produce greenhouse vegetables, which increase the vegetable supply to the market in extended season time and save the energy resources (Ali *et al.*, 2019). Cucumber production in tunnels has a strong role for gaining more yield in a small area. This crop is having a great challenge of diseases and insects. Downy mildew of cucumber is the major disease of cucumber in high tunnels. Diseases can be managed efficiently by adopting good farming and cultural practices in the high tunnels by adopting good farming and cultural practices in the high tunnels. It is important to study the dynamics of downy mildew disease and develop its management strategy. The current study was focused on: 1-determination of downy mildew disease incidence and severity in cucumber high tunnels; 2- determination of plant resistance resource against the disease; 3- the impact of downy mildew disease management practices in high tunnels on disease incidence and severity; 4- the influence of disease incidence on crop yield.

MATERIALS AND METHODS

Experiment site

The study was conducted at District Vehari (30.0452° N, 72.3489° E), Punjab, Pakistan, for two consecutive years 2015-16 and 2016-17. Nine farms cultivating cucumber in high tunnels were selected and named as Farm 1, Farm 2, Farm 3, Farm 4, Farm 5, Farm 6, Farm 7, Farm 8 and Farm 9. Each tunnel farm was 10 to 15 km away from the other farm.

Disease incidence and severity assessment

Fifty plants were selected randomly and tagged for the study. The crop was regularly visited and the disease incidence (%) of downy mildew was recorded from the time of sowing till harvesting with the formula given below:

$$\text{Disease Incidence (\%)} = \frac{\text{Number of infected plants}}{\text{Total number of plants observed}} \times 100$$

For the determination of disease severity, 200 diseased leaves were collected randomly from 20 different plants. Percentage leaf damage for the downy mildew disease was calculated and given scores as 1 = 1–3% leaf area damage, 2 = 3-6% leaf area damage, 3 = 6–12%, leaf area damage 4 = 12–25% leaf area damage, 5 = 25–50% leaf area damage, 6 = 50–75% leaf area damage, 7 = 75-87% leaf area damage, 8= 87-93% leaf area damage, 9= 93-100% leaf area damage. Mean percentage leaf area damage was calculated as disease severity percentage. Disease severity was determined at three stages of plant growth including flowering, early fruiting and late fruiting stages.

Cucumber cultivars, sowing times and fungicides

Six cultivars including "Yayla", "Joud", "3776", "9786", "Jawad", "Kaan" were sown in different farms given in Table 1. These varieties were sown about 15-day intervals i.e. Early Sowing (ES)=1-5 November in Farm 1, Farm 4 and Farm 7, Mid Sowing (MS)=13-15 November in Farm 3, Farm 6 and Farm 8, and Late Sowing (LS) = 28-30 November in Farm 2, Farm 5 and Farm 9 as shown in Table 1.

Four fungicides were used by the farmers in different doses in all the tunnel farms. The name of fungicides, their active ingredients and their doses are given in Table 2. These fungicides were used at the time of flowering, and early and late fruiting stages.

Crop yield

On maturity, the crop was harvested, and the total yield of cucumber was recorded from each of nine farms. The total production of each form was compared with a disease incidence of that farm.

Data analysis

All the collected data sets were statistically analyzed and subjected to analysis of variance (ANOVA). Treatments means of all data sets were compared by the least significant difference (LSD) and Duncans Multiple Range (DMR) test for multiple mean comparisons at ($P \leq 0.05$) by using the statistical software package (SAS 2002, (Statistical analysis system, V.8.0, SAS Institute, Cary, NC).

RESULTS

Impact of sowing time on disease development

Those plants sown in the mid of November, showed less disease incidence 28% than early (42%) and late (66%) sowing time. So, the sowing time demonstrated a great impact on disease development.

Relationship between plant stages and disease attack

We calculated the disease severity at the flowering, early fruiting, and late fruiting stages of crops. The mean results were compared with the percentage of diseased plants (Figure 2). We observed all the stages of plants showed noteworthy disease. The disease severity increased over time. Maximum disease incidence can be seen in March and April when in high tunnels the crop was at late fruiting stage. The flowering stage disease severity was relatively lower and increased later.

Impact of Crop genetics on disease development

The results indicate that the variation in genotype also produced a high impact on the disease incidence (Figure 3). We found “Yayla” was the most susceptible variety showing 91% disease incidence followed by “9786” variety showing 60% disease incidence. “Kaan” variety showed the minimum disease incidence 22%. No variety was found immune to downy mildew disease.

Downy Mildew Disease incidence in high tunnels

Overall disease incidence, in Vehari district in high tunnels of nine selected farms, is shown in Figure 1. Every farm was found infected with downy mildew disease. We observed maximum disease incidence 81% on Farm-2 followed by Fram-5 with 60% and Farm-9 with 57% incidence of the disease. Minimum disease incidence was recorded on Farm-8 with 23 % disease incidence.

Table 1. Six different varieties cultivated at nine different farms at three sowing times.

Study Sites	Sowing Time			Crop Genotype
	Early November	Mid November	Late November	
Farm 1	1-5	---	---	Yayla, Joud, 3776
Farm 2	---	---	28-30	Yayla, 9786, 3776
Farm 3	---	13-15	---	Yayla, Joud, 3776
Farm 4	1-5	---	---	Yayla, 9786
Farm 5	---	---	28-30	Yayla, 9786, Jawad
Farm 6	---	13-15	---	Yayla, 9786, Jawad
Farm 7	1-5	---	---	Yayla, 9786, Kaan
Farm 8	---	13-15	---	Yayla, 9786, Kaan
Farm 9	---	---	28-30	Yayla, 9786

Table 2. Four fungicides their formulation, active ingredients and their doses used in high tunnels.

Common Name	Formulation	Active ingredient	Dose	Source
Cabriotop	60% WDG	Metiram +Pyraclostrobin	4 g/L	FMC, Pakistan Ltd.
Ridomil gold	68 WP	Metalaxyl + Mancozeb	2.5 g/L	Syngenta, Pakistan Pvt Ltd.
Score	25 % EC	Difenoconazole	2 ml/L	Syngenta, Pakistan Pvt Ltd.
Champion	77 % WP	Copper hydroxide	2 g/L	Jafar Brothers Pak. Pvt Ltd.

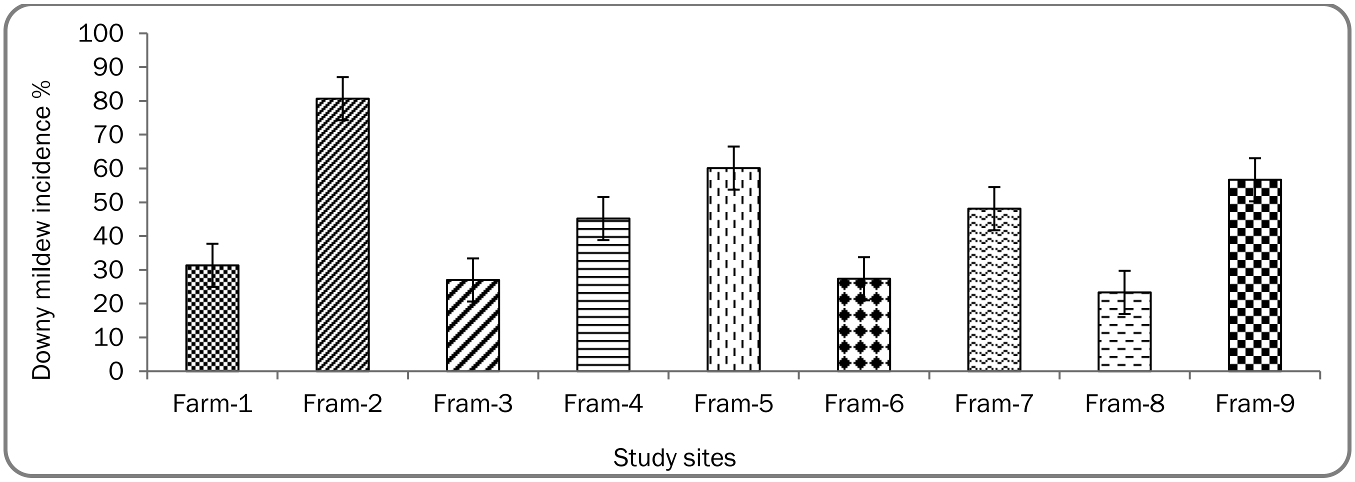

Figure 1. Disease incidence of downy mildew of cucumber in high tunnels in District Vehari. ES= Early Sowing; MS= Mid Sowing; LS= Late Sowing.

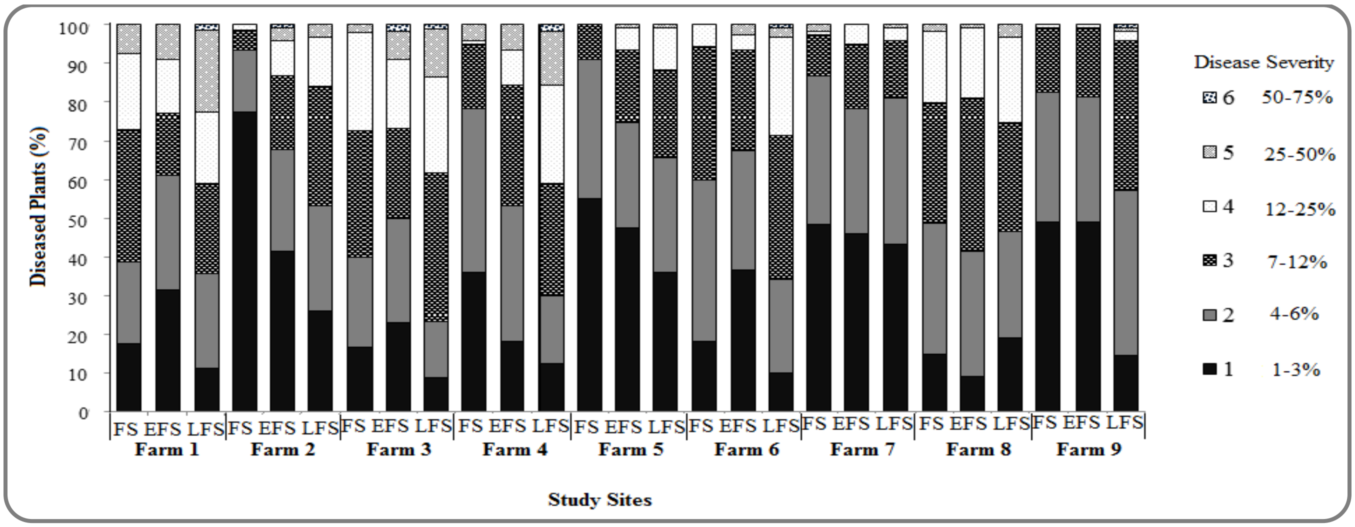

Figure 2. Percentage of infected plants and disease severity of downy mildew at different growth stages of plants in high tunnels. FS= Flowering stage; EFS= Early fruiting stage; LFS= Late fruiting stage.

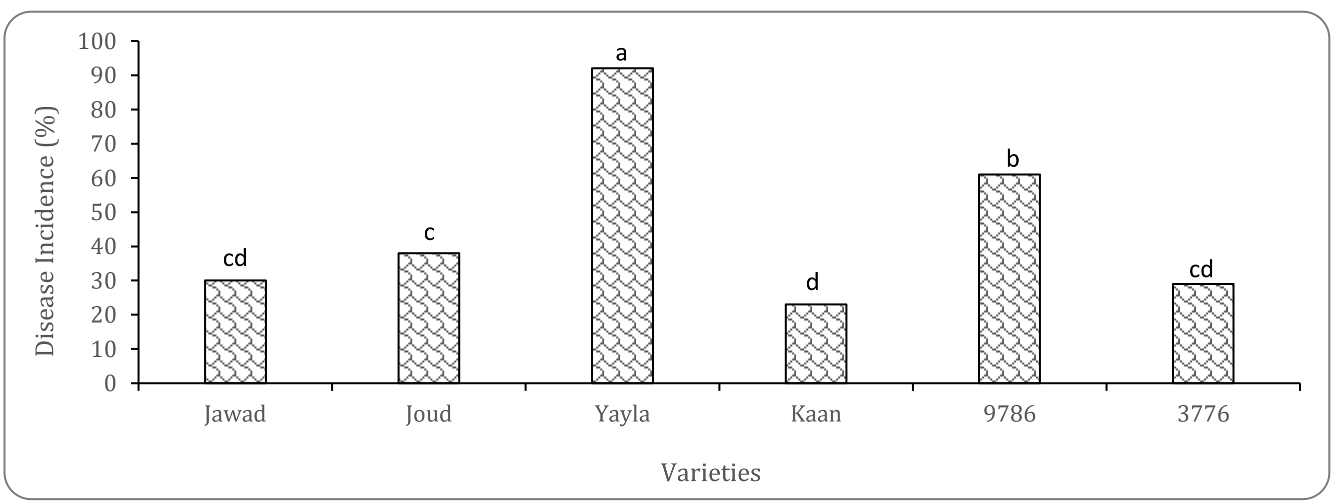

Figure 3. Disease incidence (%) on six local varieties. Different small letters showing the statistical difference in among mean disease incidence on different varieties at a 95% level of confidence.

Influence of fungicides on disease development

In the current study, four fungicides were used and their relationship with the disease incidence was calculated to find out their influence in controlling disease spread (Figure 4). We found that "Copper hydroxide" showed the maximum disease reduction of 54.5% over control. The second most effective fungicide against downy mildew "Difenoconazole" showing disease reduction up to 43% followed by "Metiram +Pyraclostrobin" 38% and then "Metalaxyl + Mancozeb" (33%). None of the fungicides showed complete effectiveness against the fungal pathogen and failed to give complete disease control.

Impact of disease on crop yield

The cucumber production (Kg/acre) was recorded and correlated with disease incidence (%) as shown in Figure 5. The results indicate that crop yield was less where the disease incidence was higher. The maximum disease incidence (81%) with very crop production about 75600 Kg/Acre was recorded in Farm 2. While Farm-1 showing very low disease incidence (31%) gave very good crop yield (88800 Kg/Acre). Farm1, Farm 3, Farm 6 and Farm 8 are showing lesser disease incidence and higher yield. Farm 2, Farm 4, Farm 5 and Farm 7 are showing higher disease incidence and lower yield. Farm 9 is an exception, where the disease is higher and crop production is also higher.

DISCUSSION

High tunnels save the crops from frost and extreme environmental conditions and give the high yield before the open field crop if managed well. Crop production is affected by many factors including diseases which are the core factor liable for low yield of cucumber in high tunnels. Farmers take various measures for the high crop yield and combat with the problems including diseases. These cultural, biological, and chemical factors used in high tunnels may also affect directly or indirectly the disease incidence in the field (Juroszek and von Tiedemann, 2011).

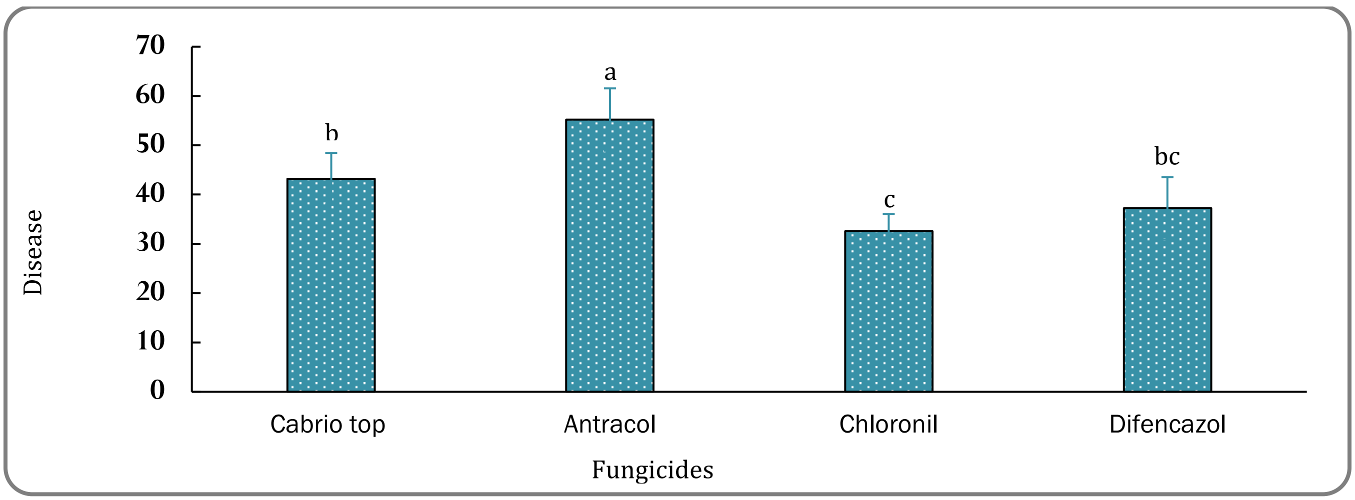

Figure 4. Means values of disease inhibition (%) by four fungicides used in high tunnels for controlling downy mildew of cucumber.

Overall, the farms with late sowing time showed highest attack of the disease, followed by early sowing time, while the mid-November sowing showed the least disease incidence (Figure 1). The time of sowing in the high tunnels showed a huge impact on disease incidence. This may be due to the impact of temperature and variation in humidity which led to the adverse conditions for the pathogen survival (Srivastava *et al.*, 2010). The changing environmental conditions by altering planting dates were reported to reduce or increase the disease (Wolfe *et al.*, 2007).

The change in planting and harvesting dates is dependent on region, crop variety and market conditions (Garrett *et al.*, 2006; Wolfe *et al.*, 2007). According to Srivastava *et al.* (2010), changing the sowing dates and cultivar should be explored to reduce the vulnerability of crop production to climate change.

The potato growers in African highlands choose such sowing time to avoid the rainy season to decrease the danger of late blight and the risk of drought stress (Hijmans *et al.*, 2000).

In this study, the impact of disease severity at flowering, early and late fruiting stages were assessed (Figure 2). Maximum disease incidence was seen in March and April when in high tunnels the crop was at late fruiting stage. The disease severity was relatively lower till flowering stage and increased with time. Similarly, VandenLangenberg and Wehner (2016) showed in an experiment also described that the old stage of the plant showed a higher intensity of disease than an early flowering stage.

In host-pathogen interaction, the resistance cultivars played a significant role (Legrève and Duveiller, 2010). In this study, the variation in genotypes produced a high impact on disease incidence (Figure 3). It was found that "Yayla" was the most susceptible variety showing 91% disease incidence and "Kaan" variety was the most resistant with 22% disease incidence. No variety was found immune to downy mildew disease. The field history described that "Yayla" and "9786" varieties were the most sown for many years. Therefore, the cultivar became more susceptible to the pathogen due to continuous sowing for many years (Savory *et al.*, 2010). Our findings showed that variability in the crop cultivars also produced variation in disease incidence. The use of resistant varieties is an ecofriendly and cheap method to manage a disease (Dodds and Rathjen, 2010).

Fungicides are expensive and unfriendly to our environment but used to control pathogenic fungi and oomycetes as they are quick and precise. In the current study, four fungicides were used and their relationship with the disease incidence was calculated to find out their efficiency against the disease spread (Figure 4). It was found that "Copper hydroxide" showed the maximum disease reduction of 54.5% over control. The second most effective fungicide against downy mildew as "Difenoconazole" showing disease reduction up to 43% followed by "Metiram +Pyraclostrobin" 38% and then "Metalaxyl + Mancozeb" (33%) as shown in Figure 4. None of those fungicides showed the complete effectiveness against the fungal pathogen. This might be due to emerging disease resistance in *Pseudoperonospora cubensis* fungus (Urban and Lebeda, 2006).

Predominantly, fungicides are applied foliar against this pathogenic fungus and the fungicide treatments may be influenced by climate change (Ghini *et al.*, 2007). Application of fungicides and the use of genetic resistance are the core strategy of integrated pest management approaches (Delmas *et al.*, 2017; Garrett *et al.*, 2006).

It is a fact that the increase in disease will diminish the crop yield and quality by affecting plant health. We also observed the same results in the current study when yield and disease incidence were compared (Figure 5). In the current study, cucumber production (Kg/acre) was recorded and correlated with the disease incidence (%). Overall, an average yield was documented as 80933.34 Kg/Acre and the disease incidence was 44.42%. We found a highly negative correlation between disease incidence and the cumulative crop yield. These results describe that the increased incidence decreased the crop yield in all the farms.

We conclude that downy mildew disease had a keen role in yield reduction in cucumber in high tunnels. Sowing time, genetic variation and plant age significantly affected the disease incidence. The use of disease resistant varieties of cucumber, appropriate fungicides usage may be helpful in disease reduction. We also observed that the old stage of the crop was more sensitive to the disease intensity than flowering or early fruiting stage.

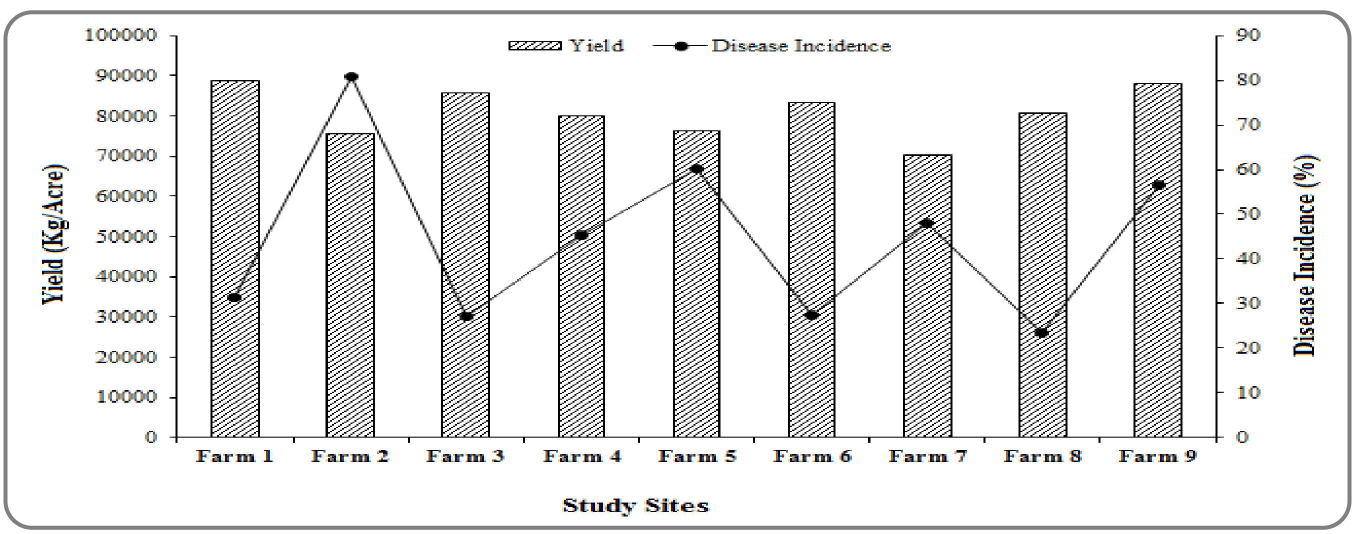

Figure 5. Impact of disease incidence (%) of downy mildew of cucurbits on yield (Kg/Acre) at nine different tunnel farms.

ACKNOWLEDGMENT

Authors are thankful to all progressive growers of the district Vehari for their cooperation during research work.

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CONFLICT OF INTEREST

There is no conflict of interest among authors.

AUTHORS CONTRIBUTIONS

All the authors equally contributed in this research and manuscript editing.

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