# Investigation of wilting and decay factors of cucumber at greenhouses (Case Study: in Kohgiluyeh-va-Boyerahmad, Iran)

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## **Abstract**

In order to identify the wilting and decay factors of cucumbers at greenhouses in Kohgiluyeh-va-Boyerahmad, a number of samples were taken of slow-growing plants with general yellowing, necrotic and chlorotic leaves, withered farm bushes, and cucumber greenhouses during 2011-2012. After transferring the samples to the laboratory, the pathogens were immediately isolated and eliminated in the laboratory. The infected root and crown samples were disinfected and then cultured on a PDA medium. They were identified after purification through to the hyphal tip technique. The results showed that most infection in greenhouses was associated with downy mildew on cucumber caused by *pseudoperenospora cubensis*. The second fungus factor leading to leaf necrosis was detected to be a cucumbers powdery mildew fungus known as *Erisiphe cichoracerum*. Based on macroscopic and microscopic observations and confirmation on pathogenicity of the fungi, the necrosis of the plant and damping-off was associated with *Pythium* and *Phytophthora*.

Key words: wilting, cucumber, identification, Kohgiluyeh-va-Boyer-Ahmad, pseudoperenospora cubensis

### **INTRODUCTION**

ucumber (Cucumis sativus L.) is the fourth most widely grown vegetable crop in the world after tomato (Solanum lycopersicum L. var. lycopersicum [syn. Lycopersicon esculentum Mill.]), cabbage (Brassica oleracea L. var. capitata L.), and onion (*Allium cepaL*.) [1]. Many factors cause yellowing and drying of leaves, root and crown wilting and decay in cucumber. Cucurbit downy mildew, caused by the oomycete pathogen Pseudoperonospora cubensis (Berk. and Curt) is a major foliar disease of cucumber [2]. Based on the available resources, Pseudoperenospora cubensis in some regions creates brownish yellowing necrotic patches on the lower layer of the leaves, thus leading to wilting and decay in cucumber bushes. Another factor causing decay in the cucumber crown and root is the fungi family of Pythiaceae from the category of *oomycetes*. The sudden death of plants occurs within one or two days from first irrigation as the plant turns into a dry green color [3].

Cucurbit downy mildew, caused by the oomycete pathogen *Pseudoperonospora cubensis* (Berk. and Curt.) The cucurbit downy mildew pathogen is an obligate parasite and, with the rare exception of ospore production, can only survive and reproduce on living host tissue [4].

According to a study in Varamin Research Center in collaboration with Tehran University and the Plant Protection Research Institute on the pathogenic bacterial isolates, it was revealed that the burn-like V-shaped patches on the cucumber leaves accompanied by a yellow halo on cucumber at 27 to 30 °C give rise to expanding the bacterial blight [3].

Occasionally, Fusarium leads to wilt and decay of cucurbit bushes. In case the disease spreads, the plant will die. In this respect, the weeds in the cucurbit farms such as alhagi, licorice, wild radish, wild safflower and alfalfa play a key role in the pathogen survival and stability [5].

According to Macdonald andLeach (1976), Fusarium the factor responsible for wilting caused by the fungus *Fusarium oxysporum* FSP betae isolated from roots of weeds such as *Brassica nigra*, Chenopodium under natural conditions <sup>[6]</sup>.

However, in the province of Kohgiluyeh-va-Boyerahmad no research has been done on diseases related to greenhouse cucumber so far. Yellowing and wilting are among the most important limiting factors in the greenhouse cucumber cultures. In this study, effort was made to identify the potential pathogenic factors at several cucumber greenhouses in Kohgiluyeh-va-Boyerahmad.

# **MATERIALS AND METHODS**

#### **Sampling**

At first, the cucumber greenhouses in Kohgiluyeh-va-Boyerahmad were inspected during 2011-2012. The slowgrowing plants with general yellowing; necrotic and chlorotic leaves and withered farm bushes were collected and then transported to the laboratory. The stages were carried out based on standard practices, such as isolation, purification and identification of species. For this purpose, the leaf samples were immediately transferred to the laboratory to obtain microscopic slides and were examined under the microscope. Moreover, the parts of discoloration in leaves, roots and crowns were washed with tap water and then 3-5 mm fragments were prepared to be disinfected through commercial liquid bleach (Javelle water) 10% containing active ingredient sodium hypochlorite for 1-2 minutes. They were then cultured in NA and PDA mediums. The purification was conducted through a single spore procedure on the water agar medium 2% [5]. The diagnosis of species was based on macroscopic and microscopic observation as well as the available identification resources. Moreover, the pathogenicity of

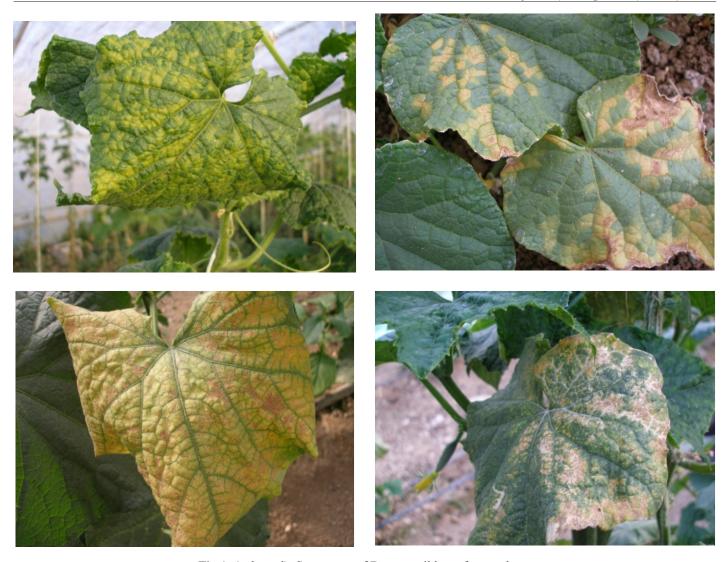


Fig 1: (a, b, c, d): Symptoms of Downy mildew of cucumbers.

agents isolated from the cucumber roots and crowns was approved during the experiment through dipping the root into Root dip suspension [7].

## **Isolation and purification:**

A. Examination of leaf samples: a number of leaf samples were immediately transferred to the laboratory and were then divided into small pieces of 1-2 cm. They were washed with tap water and placed on sterile filter paper. Then, they were incubated at 25 °C in wet desiccator. After 48 hours, the microscopic samples were examined.

B. number of leaf samples were transferred to the laboratory so as to immediately microscopic slides be taken from them and then be examined under the microscope.

C. The parts of discoloration in leaves, roots and crowns were washed with tap water and then 3-5 mm fragments were prepared to be disinfected through commercial liquid bleach (javelle water) 10% containing active ingredient sodium hypochlorite for 1-2 minutes. They were then cultured in NA and PDA mediums.

The purification was conducted through a single spore procedure on the water agar medium 2%. Having been purified, they were transferred to the PDA media and were stored at a

temperature of 20 °C for future studies [7].

Identification of species: It was based on the macroscopic and microscopic observations and the available resources:

## **Pathogenesis studies**

The pathogenicity of agents isolated from cucumber root and crown during the test was approved through dipping the roots in spore suspensions (rootdip) using cucumber susceptible cultivars. Then, the young seedlings were planted in pots and the occurrence of the disease was evaluated [8].

### **RESULTS**

From autumn 2011 to autumn 2012, samples were taken regularly from 20 farms and greenhouses in the province. The random sampling procedure from the infected farms and greenhouses was conducted [9].

Based on the morphological and microscopic traits, the cause of disease on the cucumbers bushes *Pseudoperenospora cubensis* was diagnosed to be Downy mildew, which averagely accounted for 75% of the pallor and drying of the cucumber bushes in the farms and greenhouses (Figure 1).

In some greenhouses, the prevalence of the disease amount to





Fig 2: (a, b): Severe infection with Downy mildew



Fig 3: Conidia of cucumbers powdery mildew



Fig 5: Sporangium of Phythium



Fig 4: Severe infection with powdery mildew

one hundred percent (Figure 2). These results are consistent with those obtained in other studies.

The second major damaging cause in cucumber plants can be found as little white spots on the leaves and stems, which are gradually covered by a white powder and can be seen later on both sides of the leaves (Figure 3).

The fungi in dry conditions can quickly produce conidia, which appear on the older leaves of reproductive organs as small as pinhead in black color.

Based on morphological and microscopic traits, the cause was identified to be *Erisiphe cichoracerum* on cucumbers.

The symptoms appeared on young cucumber seedling destroyed the soft and thin roots. Moreover, the microscopic observations in the laboratory showed that the sporangium was spherical and a tube comes out where there is a spherical object. Based on macroscopic and microscopic observations and confirmation of pathogenicity of the fungus, the cause of damping-off in the young cucumber seedling was identified to be *Phythium* <sup>[9]</sup>. (Figure 5).

The target plants were suddenly wilted while the leaves were green and dry up, which occurs often after irrigation (figure 6)



**Fig 6:** Symptoms of wilting caused by the fungus Phytophthora

#### **DISCUSSION**

The majority of farm and greenhouse infection were due to fungal factors leading to symptomatic mosaic pattern on the leaves. As soon as the disease spreads on the lower surface of the leaf, a purple patch appears right in front of the stains, which later turns into white to black. The leaves are quickly dried, which can be seen mostly in the mid leaves of the bush [7].

In other words, chlorosis and necrosis are the main characteristics of this fungus on leaves. The sporangium of the fungus is spherical and non-chain. Moreover, it entails binary branches leading to strigma on which sporangium can be seen [9].

## **CONCLUSION**

According to the obtained results, 75% of discoloration and drying cucumber plants was related to *pseudoperenospora cubensis* as a factor of downy mildew in fields and greenhouses. The second important leaf factor causing damage and disease in plants has been recognized in relation to *Erisiphe cichoracerum* as a factor of Powdery mildew. On the factors causing damage in the root and crown of cucumber, Phythium spp and Phytophthora were isolated and confirmed as factor causing the death of young seedlings of cucumber and suddenly fading cucumber plants, respectively.

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