

# Comparative Transmission of Potato Virus X (PVX) in Water-Based Medium between NFT and Soil Cultivation of Pepper Plants

Running Title: Comparative Transmission of PVX in NFT and Soil Cultivation

Haneen H. Mohammed Ali1,  
Raghad N. Mheedi 2

1Department of Plant Protection, College of Agriculture and Forestry, University of Mosul, Mosul, Iraq, 41001,

Corresponding author: haneenshingaly2@gmail.com  
(Duhok, Iraq, +9647515449199 )

Second author : zrnaif20055@uomosul.edu.iq  
(mosul,iraq, +9647740901702)

## Abstract

Hydroponics is a method of growing plants without the need for soil, using a synthetic growth medium and a solution of mineral fertilizers. It fights diseases and pests spread by the soil, boosts output, and facilitates better management of the plant environment. While the ability to regulate the distribution of nutrients may be a benefit of the NFT system, plants are also exposed to possible viral contamination when soil-borne infections are reduced. If nutrient solutions are used in hydroponic systems, water-borne plant diseases may spread more quickly, increasing the risk of epidemics if improper management is not practiced. Numerous plant viruses are spread by water, as evidenced by the comparatively well-researched studies on plant virus prevalence in irrigation water, irrigation water source, and aquatic environment causes. The goal of the study was to make comparisons between soil systems and hydroponic systems (NFT, or nutrient film technology), which illuminate the complex dynamics of Potato Virus X transmission.

In the fall of 2021–2022, a field survey was carried out for many potato farms in the governorates of Nineveh, Dohuk, and Erbil, which are located north of Iraq, where the infection's signs, which included mild leaf deformation and tiny, alternating-color dots in a modest mosaic shape, were noticed. Depending on the type and field, these symptoms changed.

The Double Antibody Sandwich Enzyme-linked Immunosorbent (ELISA) test was used to confirm the virus's existence. The absorbance of the healthy sample was 0.319, whereas the infected sample had a yellow hue of 2.896. In order to maintain the isolation and function as a inoculation for the virus in this investigation, mechanical injection was performed on pepper and potato seedlings following the Immunostrip Assay's confirmation of the virus's existence.

This study examined the impact of pepper virus infection (PVX) on plant development and how it spreads among pepper plants grown in soil and under NFT hydroponic systems. Two investigations were carried out in the hydroponics experiment: (1) a test to determine if PVX can spread through the roots (using healthy seedlings and water polluted with PVX) and (2) a comparison of the growth of the seedlings (using pure water and healthy seedlings).

The findings show that mosaic symptoms and development are impacted by waterborne viral transmission. Using the AutoCAD software, the incidence and severity of the disease were assessed by comparing healthy and infected leaves. On the third day, the infection rate was (3 days/16%), and on the fifth day, it was (5 days/50%). On the ninth day, (9 days/46%), symptoms worsened to reach On the sixteenth day, it hit 100%, marking its



pinnacle.

The hydroponics experiment was finished before the field experiment, which consists of three experiments plus two comparative experiments, started.

In the first field experiment, an artificial infection was used to induce symptoms ranging from mosaic to mild to moderate leaf deformation, with mottling and clear swelling between the veins and moderate color change in a section. Pepper seedlings were inoculated and planted alternating with healthy seedlings in the same planting line (maraz) and watered with regular irrigation. Fairly noticeable indications started to show up on the fourth day, according to the report, marking the beginning of the plant's reaction to the experimental settings.

In the days that followed, more precisely until the twelfth day, the intensity of these symptoms demonstrated a rising trajectory. The critical stage of this evolution took place on day 12, which also happened to be the day when irrigation was applied, which controls the evaluation of virus transmission.

Regarding the second field trial, drip irrigation was employed. When the seedlings were treated with water infected with the virus through fertiliser, symptoms started to show on the eighth day, when they were at a rate of 8 days/24%. They then increased, reaching 15 days/27%) on the fifteenth day. In addition to mottling and swelling between the veins for some of the seedlings, it caused mosaic symptoms with color exchange between the veins. These symptoms persisted until the twenty-second day, when they were seen (22 days/34%). Two comparison studies were carried out: the first used drip irrigation and the second ordinary irrigation. The findings of both trials revealed normal growth.

This study helps to emphasize how crucial it is to shield aquatic plants from the virus. However, growing agricultural pepper in the ground produced comparable outcomes, and various irrigation systems were the means by which the virus spread. The NFT system's ability to display illness signs demonstrated how vulnerable this innovative technology is to plant diseases.

However, we have now documented on the Internet the exact mechanism of interaction between the many chemicals that allowed the virus to spread. In order to prevent the spread of PVX, it is vital to provide legal aid services to those who use water for sanitary irrigation and drip systems. This underscores the significance that infection practices play in the virus's propagation. This attests to the severity of the virus's propagation, which is influenced by a number of detrimental administrative, agricultural, and environmental aspects. This study intends to avoid pathogens from causing any symptoms in vegetables (apart from water) and assess the system prior to hydroponics growing in order to slow down the propagation of the virus.

**Keywords:** Disease management; plant pathogen; plant viruses; potato virus x; viral transmission.

## Introduction

Hydroponics is a method of growing plants in a solution of mineral nutrients without the need for soil, but with the use of an artificial growth medium (Bhattarai et al., 2008). This technique improves access to sustainable food supplies in both developed and developing countries, makes it easier to control the plant environment, increases production, and resists soil-borne pests and diseases (Lommen, 2007). Additionally, it is a more sensible use of water resources (Daniel, 2014).



The use of nutrient solutions in hydroponic systems may facilitate the rapid spread of waterborne plant pathogens throughout the crop, which may increase the potential for epidemics if not managed properly. However, the production of soil-less crops is growing globally, and this has given plant growers who face soil-related issues an alternative (Stewart-Wade, 2011). The roots of the plants that are produced hydroponically—also referred to as liquid farming—are submerged straight into the fertilizer solution.

It is further split into two categories:

The first kind is closed systems, which include various hydroponic systems, such as:

1) Deep flow method (DFT) and 1) nutrient film technology (NFT)

The open system, which is the second kind, also includes the following: 1) The capillary action technique; 2) The floating technique; and 3) The root dipping technique (Maharana & Koul, 2011). The Solanaceae family has provided food and medicine to human civilizations for thousands of years. More recently, these plants have become beautiful.

Several Solanaceae species have been the focus of traditional genetic studies and molecular research for the past century (Gebhardt, 2016). Numerous species, including tobacco, pepper, and tomatoes, belong to the Solanaceae family. The crop is indigenous to tropical America and has more than thirty kinds. The intake of peppers by humans has a minimum of 6,000 years of history, while pepper farming has a minimum of 2,500 years of history.

Hot and sweet peppers are grown all over the world and play an important role in Asian cuisine. Hot and sweet peppers come in five main types, the most commonly cultivated species, *C. annum*. And *C. chinense*, *C. baccatum*, and *C. pubescens*. (Daunay et al., 2008; Perry et al., 2007) Numerous diseases, including viruses like Potato, attack pepper crops. A crop variety of significant commercial importance, virions are flexible filaments with a pitch of 3.3–3.7 nm, helical symmetry, and dimensions of 470–580 nm in length and diameter. Occasionally, a central axial channel with a diameter of around 3 nm can be identified. Each turn of the basic helix has significantly less protein subunits than 9.0. According to (Atabekov et al., 2007), the radial location of the RNA backbone is 3.3 nm. They cross across. Due to their mostly asymptomatic nature in the host, potexviruses can co-infect, which frequently leads to much lower output and more severe symptoms (Hameed et al., 2014).

Because of its great biological stability, the PVX virus is easily spread by contacting infected plant leaves with healthy plant leaves (Smith, 1933). The virus that infects potatoes According to (Bawden et al., 1948), the buds were infected with infectious juice, and the virus was also passed from infected tubers to healthy tubers housed in the same bag.

Another researcher's results showed that the infection in the rows at the bottom of the tractors was 3-4 times higher than in the rows on the outer sides of the wheels, and that after the tractor passed 20 meters of infected plants, new infections appeared at a distance of 150 meters. This is one way of transmission of potato virus X via contaminated instruments (Winther-Nielsen, 1972). Potato virus X cannot be spread by seeds, as stated by the researcher (Alemu et al., 2002)

The virus classification :according to ICTV

Realm: Riboviria

Kingdom: Orthornavirae



Phylum: kitrinoviricota

Class: Alsuviricetes

Order: Tymovirales

Family: Alphaflexiviridae

Genus: Potexvirus

Species: Potato Virus X

## 2- Material and method

### 2-1 Field survey, collecting samples potentially infected with potato virus X, and preserving them:

Leaf samples were collected from a potato crop infected with symptoms of small, alternating color spots in a slight and moderate mosaic shape and slight leaf deformation. It includes the entire leaf area, as a field survey was conducted with the aim of collecting leaf samples in several areas of Nineveh and Dohuk Governorates. The first survey included in the Simil district of Dohuk Governorate in the fall of 2021-2022 for potato fields, while the second survey, for the same procedure, included several visits to areas (Dughat village), , Rabia, Mosheh village, Sharekhan, Al-Kubba, Khorsibat, Alam Luk, Jarish), where samples potentially infected with Potato Virus The third survey included several visits to the Bardarash areas in cooperation with KH Agricultural Company and the Rufia district in Ariel Governorate.

The vegetative parts of the potato crop, represented by the tubers potentially infected with the PVX virus, were collected in the same way and from the same areas mentioned above. All samples taken from the survey areas were placed in clean paper bags and all information was written on them for the purpose of preserving the sample (varieties, region, date of collection, time of collection, field area). , farmer's name and load number), taking into account collecting samples of healthy potato plants for comparison(fig.1 a and b).

All leaf samples were kept in the refrigerator at 4C, and the vegetative parts (tubers) were stored in the laboratory at 25 C degrees until all subsequent tests began.

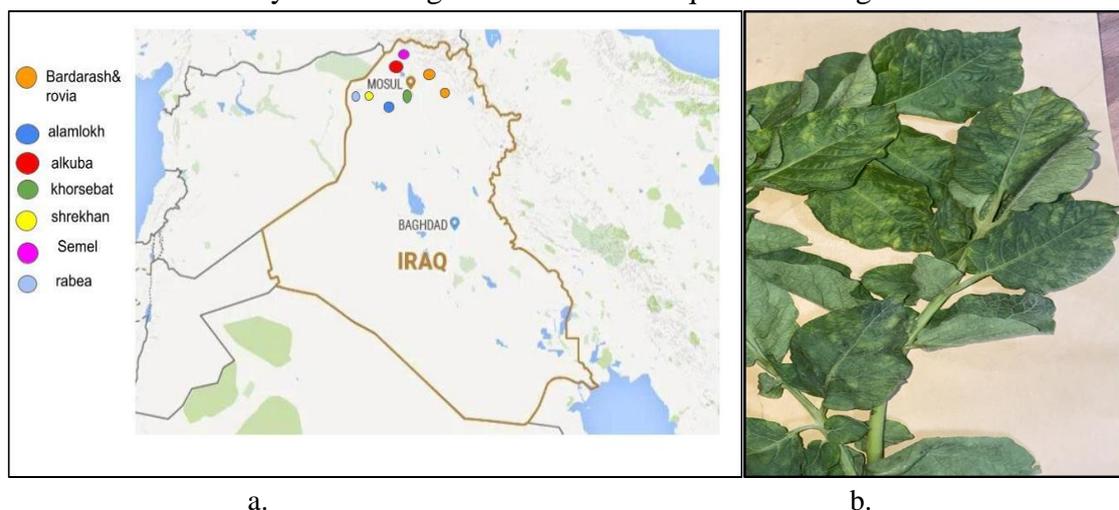


Fig.1 . a. Field survey areas: Mosul governorates and its outskirts, Dohuk and Erbil  
b. Symptoms of moderate mosaic are yellow mottled spots between the veins on potato plants.



**2-2 Diagnosis of potato virus X (PVX) by the Double Antibody Sandwich Enzyme-linked Immunosorbent (ELISA) test:**

Each representative sample was taken from the leaves collected from the field survey areas and kept in a refrigerator at 4 C, and a serological test was conducted on them using the dual containment ELISA test (DAS-ELISA) according to the method mentioned by Adams & Clark 1977 and used by the producing party, which is Agdia Company. In its American headquarters (fig.2) (table .1 )

Table .1 . Components of the ELISA test for the PVX virus from Agdia.

	Component	size	number
1	Antibody-coated 96-well microtiter plates	N/A	1 plate
2	Alkaline phosphatase enzyme conjugate	0.150 mL	1 bottle
3	RUB6 enzyme conjugate diluent, 1X ready to use	11 mL	1 bottle
4	PNP substrate tablets, 5 mg each	12 count	1 bottle
5	PNP substrate buffer, 5X concentrate	12 mL	1 bottle
6	Positive control	N/A	1 vial
7	PBST buffer, 20X concentrate, 50 mL	50ml	3 pouches
8	TWEEN® 20	15ml	1 bottle
9	General extract buffer, GEB	16.5 g	1 bottle



a. b.

Fig.2. Potato Virus Diagnostic Serology Kit (PVX) X provided by the American company Agdia

- a- Boxes of preserved chemicals prepared by the company and the binding solution
- b- Polystyrene dishes pre-treated with anti-virus serum.



### 2-3 Diagnosis of potato virus (PVX) X using ImmunoStrip test:

The Potato virus X pathogen ImmunoStrip is used to detect the presence of PVX in potato leaves. ImmunoStrips are the perfect screening tool for use in the field, greenhouse, and the lab.

This test used according to the user guide from agdia company (fig.3)

Steps for employing immunostrips to examine samples potentially infected with the virus, according to the methodology of the American firm Agdia:

1. Collect a sample of the contaminated leaves and clean them of dust by cutting off a part of the leaf likely to be infected with Potato Virus X using scissors, measuring 0.15 g or the equivalent of 1 inch.
2. The examination bag is opened with sterile scissors and the bag is cut below the label.
3. The 0.15 g sample is introduced into the bag by inserting it in the mesh at the bottom of the bag.
4. The sample is ground and crushed gently while still within the bag, using a tiny hand mortar or the end of a pen. After crushing, the sample is placed in the bag for three (3) minutes before adding the PVX virus immunity strip.
5. Insert the end of the strip into the section of the bag at an angle (without the mesh) until it reaches the crushed sample, following the direction of the arrows on the strip.
6. After inserting the strip, the bag is fixed upright and behind it is any solid material that prevents it from tilting or falling. You must wait for (30) minutes to take the result. The appearance of the two lines means the presence of the virus in that sample.

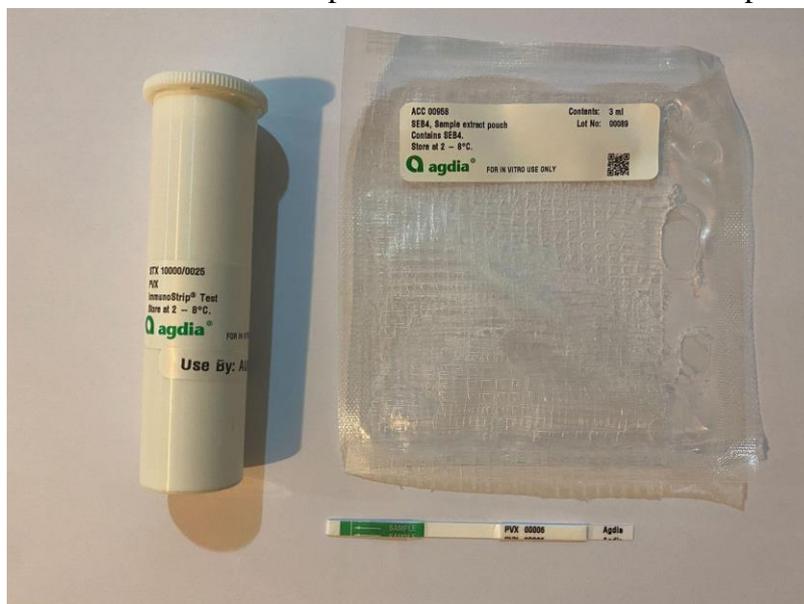


Fig.3 Virus detection immunoassay kit (PVX).

### 2-4 Potato virus X isolation preservation:

Potato virus X isolation preservation: Several leaves were obtained from the potato plant and from the same plant on which the serological diagnostic was done using the immuno-strip test and the dual containment ELISA test (DAS-ELISA). After washing the leaves, extracting the main vein, and putting them in a clean and sanitary ceramic mortar with a solution, they were crushed. KH<sub>2</sub>PO<sub>4</sub> phosphate buffer with a weight:volume ratio



of 1:2 (leaves to buffer solution) and a pH of 7.6.

The extract was put in a sterile beaker after being filtered through two layers of muslin fabric. Following that, the filtrate was utilized in mechanical inoculation stages using 400-600 mesh carborundum powder as an abrasive for fifty pots of Lucinda variety potato plants and fifty pots of California variety pepper plants. Both plants were chosen to protect the viral isolation, and the plants were then kept.

In the Plant Protection Department's greenhouse at the College of Agriculture and Forestry, in addition to regular fertilization and watering. In addition to spraying and watering with the insecticide and fungicide Commando 20%, water 10-25mL/20L, and benquil with a water content of 2g/1L to prevent the occurrence of any possible fungal or insect infection. Every two months, the isolation was restarted on new potato and pepper plants of the same two types and in the same manner as indicated above. Juice made from the leaves of plants where the diagnosed isolate was found(fig.4)



Fig.4. Maintaining isolation on potato and pepper plants.

### **2-5 Testing for the presence of potato virus X (PVX) using hydroponics**

The primary objective of this research is to scrutinize the transmission dynamics of Potato Virus X (PVX) within the context of the Nutrient Film Technique (NFT) hydroponic

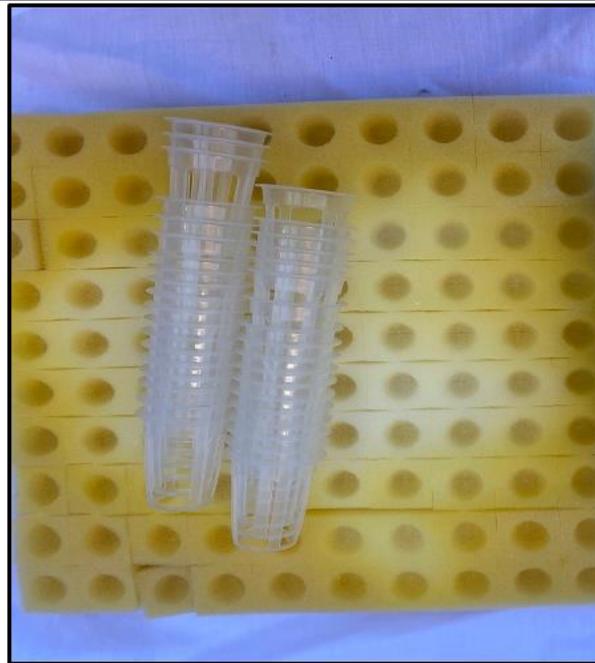
system. To this end, a meticulously structured experimental design was employed to unravel the intricate interplay between PVX and the hydroponic

The construction of the Nutrient Film Technique (NFT) hydroponic system involved assembling four water pipes, each with dimensions of 96 cm in length, 80 cm in width, and 50 cm in height. These pipes were equipped with a total of nine holes, each having a diameter of 5 cm. The setup comprised essential components, including a water pump, a water reservoir, and a 1-meter-long water hose. This system had a capacity of accommodating 13 liters of water, which underwent a continuous recycling process ( fig.5). The water pump was positioned within the water reservoir and directed water upward through the water hose to the upper part of the device. Subsequently, the water cascaded down the platforms, eventually returning to the water reservoir to be recirculated within the NFT system. The device was also provided with a sterile sponge piece containing 100 holes with a length and width of 20 x 20 cm, and each square of one hole had a length and width of 2 x 2 cm, in addition to the plastic bases attached to the hydroponics device for the purpose of installing pepper seedlings inside each square sponge piece.(fig.5

Following the conclusion of the comparative experiment, the hydroponic system underwent a thorough disinfection process. This involved the removal of recirculated water, including hydroponic pads. A cleansing procedure using a solution of 5% bleach mixed with water (Clorox) was conducted, encompassing the entire system. The water reservoir was subsequently filled with 13 liters of tap water infused with 5% bleach (Clorox). The circulation of this cleansing solution was sustained for an entire day. Upon achieving complete disinfection, the system underwent a meticulous rinse with tap water, followed by recirculating 13 liters of tap water for an additional day to eliminate any residual traces of chlorine.



a.



b.

Fig.5.a. Utilized Hydroponic Configurations in the Current Investigation

**b. The sponge and the seedling holder**

The Nutrient Solutions used in this experiment was from netherland (pokon company ) (fig.6). As a precautionary measure, the water was also tested using rapi test samples imported from the American company Luster Leaf. Each sample represents the following tests (PH, N, P, K tests). They were tested separately and using compressed capsules for all four containers. The water was placed inside this container designated for it. One capsule per examination (fig.6). Also the EC was tested .



a.



b.

Fig.6 . a.the NPK and PH test

b. The nutrient solution that used in haydroponics system .

### 2-5-1 Inoculation of circulating water

In this research, pepper seeds were initially planted in seedling trays, and then approximately 36 seedlings were planted, each containing 10 leaves, and transferred to a hydroponic system to conduct two experiments on them, which included 36 seedlings for each experiment separately. The first experiment was a comparison experiment without the virus vaccine, while the second experiment was by adding the viral vaccine for potato virus X, according to the dilution end point of the virus  $10^5$  (Awasthi & Verma, 2017). The progression of symptoms was monitored at four specific time points: 3, 5, 9, and 15 days. (fig.7).



Fig.7. The seedling that transferd to the hydroponics system with its holder .

### **2-6 Potato virus X bio transmission test**

The virus's existence was evaluated scientifically utilizing two structured and coordinated field lines. The experiment was conducted out in fields belonging to the Department of Horticulture in Dohuk Governorate, with a total area of 50 m<sup>2</sup>, and 25 m<sup>2</sup> of the field was used. To prepare the land for the experiment, the area was cleared of shrubs and pollutants. In addition to providing two more lines for comparison.

#### **2-6-1 Virus transmission test using artificial infection**

The presence of the virus was determined by artificially infecting 26 pepper seedlings with 5-6 leaves of the California variety. 13 pepper seedlings were physically infected with carbonidum 100-400 mesh using the authorized mechanical inoculation method. the first pepper seedling that inoculated with virus mechanically and left the second unpollinated, and so on for the remainder of the seedlings in the rotation.

The soil line is 3m<sup>2</sup> in length, and I allowed a space of 20 cm between each seedling. They were watered and fertilized on a regular basis, and the plants were observed in the field until symptoms arose. The Autocad tool was also utilized to compare the findings of a healthy pepper plant leaf and a virus-infected pepper plant leaf.(fig.8)



Fig.8. The feild of experiments in duhok city north of iraq .

#### **2-6-2 Virus transmission test using contaminated water using drip irrigation**

The virus's transmission was tested using contaminated water, and it was verified that the virus was present in it based on the previous diagnosis. 40 healthy pepper seedlings were planted and fastened in the field beds, with a length of 3m<sup>2</sup> for each bed and a gap of 30cm between one seedling and another.

The pepper seedlings, which were 5-6 leaves old, were irrigated with polluted water using drip irrigation. It is linked to pipes that have a fertilizer in them to which the virus was put at a concentration of 300mL per 13L of water. The virus was added in the same amount for each watering, with fertilizer being applied alternately between waterings. Depending on the final dilution degree for potato virus X, one liter requires 30mL of virus. The plants were left in the field until they developed symptoms.

A comparative experiment was also carried out with healthy pepper seedlings and clean water, utilizing the same drip watering method, the AutoCAD application, and comparing healthy and infected leaves to see whether there was a difference in leaf growth.(fig.9).





Fig.9. Fertilizers used in a drip irrigation experiment containing potato virus X (PVX) .

### 3- Result and dicussion

#### 3-1 Field survey and determination of the presence of potato virus PVX (X)

During field survey visits, manifestations of viral infection on the potato crop (*solanum tuberosum*) were observed, and these symptoms and their degree of clarity varied from region to region, as well as from variety to variety, which began in 2022 in some of its cultivation areas in the governorates of Nineveh, Dohuk, and Erbil. Symptoms were documented in general. The presence of potato virus is indicated by mosaic and mild mottling, as well as dwarfism and malformed leaves.

#### 3-1-1 Diagnosis of potato virus PVX (X).

The results of diagnosing potato plant leaf samples brought from the targeted areas and from which leaf samples were collected in the fields were shown using the Double Antibody Sandwich Enzyme-linked Immunosorbent Assay (DAS-ELISA) method of work described in the kit, and the PVX serological kit for the potato virus was used for this purpose. purchased from the American firm Agdia.

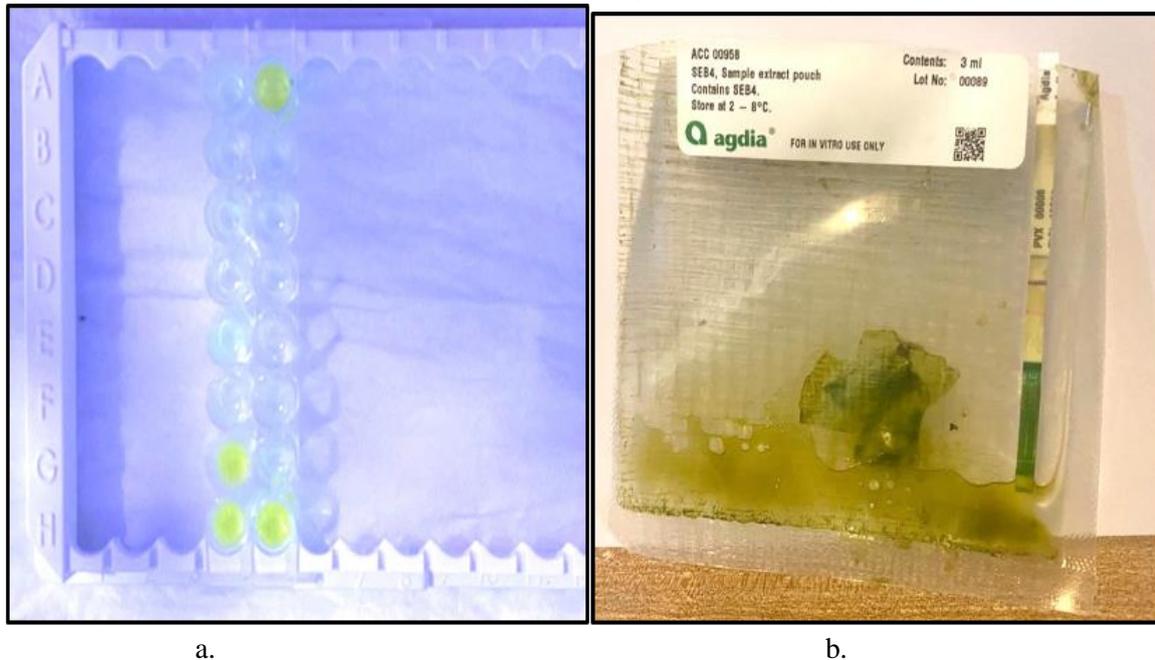
The ELISA result was calculated. The yellow tint was evident to the naked eye in the ELISA sample pits but not in the comparison samples of healthy plants (healthy plant juice). The yellow hue was likewise found in the drill column of the specified ELISA dish. The absorbance was measured at 405 light-staining lenses, where the absorbance of the sample properly was 0.319, and the sample with yellow color was 2.896, and the absorbance was measured at 405 light-reflecting lenses and using an optical telescope reader, where the absorbance was indirect 0.319 for a fast lens and slow light color yellow was 2.896., which is considered a pure solution of the potato virus Viruses spread on potato crops and crops of the Solanaceae family in Nineveh Governorate (Qasim and Muhammad,



2010). (fig.10)

This virus is also common in numerous Arab and international nations, according to (Miha et al., 1993). ELISA is a commonly used test that has been used to diagnose numerous viruses, including the papaya ringspot virus (Hartati et al., 2020). Tomato spotted wilt virus (Gao & Wu, 2022) is also present. Also, according to the source (Mehle & Ravnkar, 2012), diagnosing viruses with ELISA is one of the most used approaches for identifying viruses in aquatic settings.

additionally, The virus was identified using immunostrips imported from AGdia in accordance with the kit's instructions, and the findings were likewise positive, as shown in (Fig. 10). Immunological diagnosis is regarded as one of the simplest and fastest techniques to identify viruses, as demonstrated by (Alfadhil & Zagier, 2017), who utilized immunological strips to diagnose Cucumber mosaic virus (CMV). Immunological strips have also been utilized in other studies, such as identifying tomato chlorosis virus (ToCV) in the field (Chaudhary et al., 2023). There are also various ways for identifying plant illnesses in general and viral infections in particular, as well as several additional sources, such as (Dyakov & Elansky, 2019; Narayanasamy, 2011; Pandey et al., 2015).



a.

b.

Fig.10. Diagnosis of potato virus x (pvx) by :

**a. (DAS-ELISA) test**

**b.immunostrips test**

### **3-2 Transmission of potato virus X in a hydroponic system using Nutrient Film Technique (NFT)**

The findings revealed the existence of the potato virus. The symptoms developed after adding the viral inoculum to the water and establishing four observations as a consequence of monitoring the plants on a regular and daily basis. The symptoms appeared on the third day in a slight and moderate mosaic form between the veins of the leaf, in addition to the abnormal growth observed on the plants, where the infection rate reached (3days / 16%), and on the fifth day it was (5days / 50%), and the symptoms increased on the ninth day



(9days / 46%), reaching 100% on the fifteenth day.(fig.11)

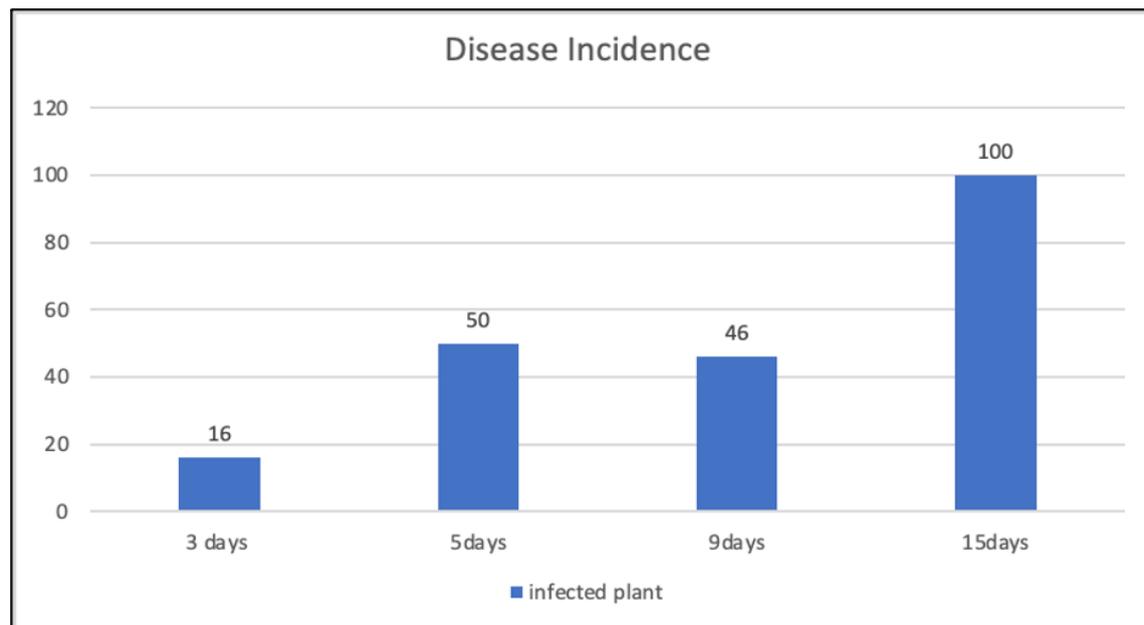


Fig.11. The disease incidence of transmission of potato virus X through the NFT system .

The symptoms were severe mosaic in nature, occurring between veins and alternating color patches on one leaf (Fig. 12). The presence of disease symptoms on 36 pepper seedlings mounted on the device resulted in the development of the yellow color indicating the presence of the virus in the holes of the ELISA dish in the double-containment ELISA test. For infected samples obtained from hydroponically produced pepper leaves contaminated with the virus. The seedlings were most likely infected through the root, which is consistent with what other researchers have said, as infectious plant-pathogenic viruses from at least seven distinct genera have been detected in aquatic settings.

The majority of plant-pathogenic viruses found in ambient waters thus far are thought to be... Stable viruses may infect plants straight through the roots and frequently infect several hosts (Mehle & Ravnikar, 2012). Another research that supports our findings found the presence of the pvv virus in the roots and leaves of a tomato crop cultivated in a hydroponic system with the virus injected to the nutrient solution (Mehle et al., 2014). Only if viruses can enter the plant via the roots, which is the most convenient route, can their presence in the water stream of a hydroponic system be considered epidemiologically significant. When employing sprinkler irrigation systems, however, there is a risk of viral infection through the leaves or other upper parts of the plants (Mandahar, 1990).

The PVX virus can spread from the roots of infected plants to the nutrient solution, and in the end it can reach the roots of healthy plants to the green parts of these plants and infect them, and this is consistent with what was mentioned in several sources(Mehle et al., 2014; Park et al., 1999; Wang & Kniel, 2016).In addition, ELISA was used to detect PepMV in one research, where the result showed the presence of the virus in all parts of the plant after transmission through a contaminated nutrient solution(Schwarz et al., 2010).There is more in another study. After about three days of inoculating plant leaves with tomato mosaic



virus, virus particles were detected in the nutrient solution. It was then found that the amount had increased to a point that could be easily seen with an electron microscope. Viral particles could infect the target plants and remain infectious. In nutrient solutions for at least six months, it begins to cause systemic symptoms in 10 days (Pares et al., 1992). In addition, the 15-week observation period of one of the research reveals that PFBV (Pelargonium Flower Break Virus) spreads rapidly. Two weeks after starting the hydroponic system, contamination of the recirculating nutrient solution was discovered, which occurred approximately one month later. The virus concentration continued to rise and six weeks later, a systemic infection developed and all plants were infected by the time the experiment ended (Berkelmann et al., 1993).

Also, in one study, it was proven that the MNSV (Melon necrotic spot virus) was transmitted in water through nutrient solutions in a closed hydroponic system and soil as well (Gosalvez et al., 2003).



Fig.12. The symptoms of potato virus x in NFT system .

Also among the results of hydroponics with the NFT system was the result of measuring the electrical conductivity of pepper ( $0.84 \text{ dsm}^{-1}$ ). The degree of electrical conductivity, or what is called the degree of salinity, is considered an important measure.

Very high in hydroponics, where the percentage being higher than the required limit leads to burning the plant or poisoning the plant, and lowering it leads to a severe deficiency of elements and their dissolution in the agricultural water, where the normal limit for the degree of EC is (0.8–1.8) (<https://planteli.com2022>) (Van et al., 2021; Yang et al., 2021), The results of measuring the percentage of nutrients in the nutrient solution were as shown in (Table 2). Agricultural crops have different requirements for the nutrient solution based on the crop; alterations in certain minerals can affect the nutritional content, texture, flavor, and size of the fruits, among other aspects. The nutrient solution used in hydroponic agriculture has an impact on each of these qualities (Levine & Mattson, 2021).

Furthermore, as hydroponically grown crops primarily rely on nutrient solutions,

controlling the nutrient solution is essential to the amount and caliber of fruits produced (Valentinuzzi et al., 2015).(Tabela 2).

Additionally, the pH test result was as indicated in (Table 2). For the plant to benefit from the nutrients added to the water in hydroponics, the nutrients must have a certain pH level. An unstable pH balance between high and low can cause a change in the quantity of nutrients in the body. For instance, when the pH falls below 5.0, plants may experience iron toxicity or a shortage of magnesium and calcium, whereas a pH level above 6 or 6.5 may cause an iron insufficiency. (Gillespie et al., 2021; Mayavan et al., 2017).

Table 2. The result of test for the PH,EC,N,P,K and Temperature .

parameter	value
PH	6.0
EC	0.84 dsm <sup>-1</sup>
N+	3.75_4.0
P+	1.20
K+	4.80
Temperature	19 ±

Additionally, two studies were conducted using pepper seedlings: one without the vaccination and the other with it added. The measurements of the leaves' length, breadth, circumference, and area are displayed in (Table 3).The AutoCAD software was then used to compare the development of the virus-infected leaf to the growth of the healthy leaf, and the results indicated a difference in growth, as seen in (Fig.13)

This application was used. The leaf area of the infected plant was 707.7 mm<sup>2</sup> with a diameter of 120.1 mm<sup>2</sup>, while the leaf area of the healthy plant measured 1345.3 mm<sup>2</sup> with a diameter of 154.3 mm<sup>2</sup>. These results and growth differences were successfully acquired in this study. The researcher (Alemu et al., 2002) concurs that there are obvious differences between the infected and healthy leaves in this instance, since the virus results in leaf deformities and changes in leaf area relative to the healthy leaf.



Table 3. The result of compare healthy and infected plant by use AutoCad .

No.	Leaf sample	Length	Width	Perimeter (MM.L)	Area ( MM <sup>2</sup> )
1	healthy	66	31	154.3	1354.2
2	Infected	53	22	120.1	707.7
3	Comparison	-13	-9	-34.2	-646.5

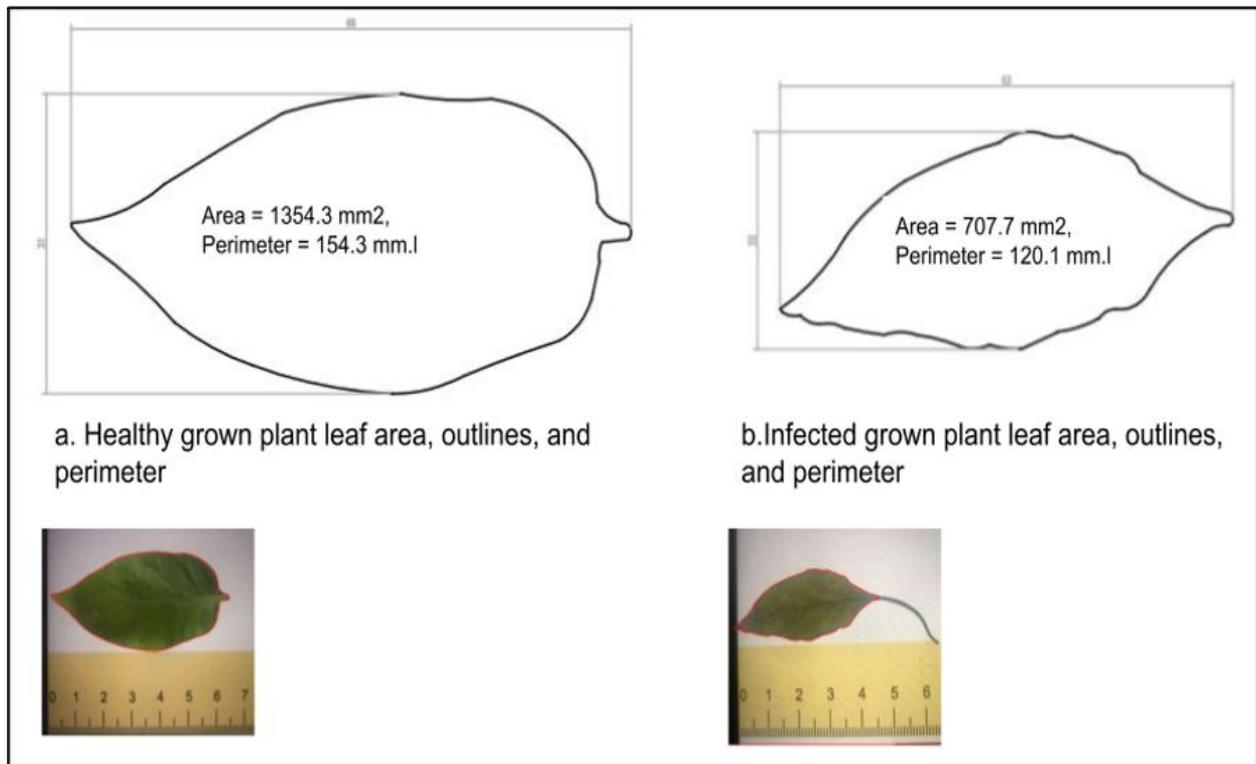


Fig.13 . Using AutoCAD, the measurement results were as follows:

- a. Measuring the leaf area of a healthy pepper plant is 1354.3 mm
- b. Leaves of infected plants mm 707.7

### 3-3 Transmission of potato virus X using artificial infection (mechanical inoculation)

In pepper plants treated with the virus (PVX) as an artificial infection by mechanical inoculation of the leaves using carborundum as a spread on the leaves, the findings of the irrigation experiment utilizing artificial infection showed distinct symptoms.

The symptoms were apparent swelling between the veins, mosaicism, mild to moderate deformation of the leaves, and moderate color change in the leaves. Certain seedlings were



stunted in comparison to healthy seedlings, and additional symptoms such as mosaics and deformation of the newly formed leaves were visible in the leaf region where the disease symptoms were evident. This part of the leaf was distinct from the healthy plants (fig 14). Among the findings was the discovery of a variation in plant lengths, considering the existence of several variations amongst the infected pepper plants without We sought to measure the lengths of plants cultivated in the field that were not affected. With a centimeter (cm) measuring tape in hand,

Two experimental circumstances were used to measure the length of the pepper plant: the first experiment used artificial infection, and the second experiment was a comparison. whereby pepper plant lengths are assessed.

In a negative contrast, Potato Virus It acts as a measure of the height of pepper plants that are not infected with the PVX virus in the experimental setting. In the PVX virus experiment, however, height measurements varied from 25 to 39 cm . This range represents the average height of pepper crops exposed to the PVX virus. These data demonstrate the effect of the PVX virus on pepper crop height.

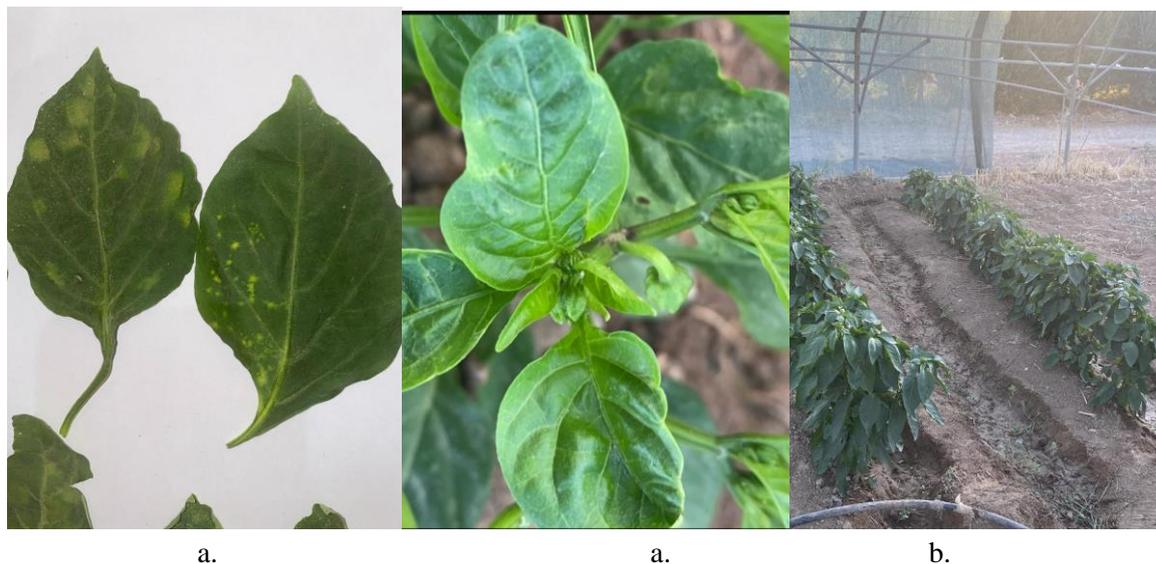


Fig.14 . a. Symptoms of pepper plants using artificial infection with the virus (PVX)

b. Healthy pepper plants without virus

The incidence rate rule was also used to calculate the disease's incidence rate. After the commencement of cultivation in the framework of an artificial irrigation experiment employing artificial infection on a pepper crop, relatively strong symptoms occurred on the fourth day, showing the early signs of plant response to the experimental circumstances. The intensity of these symptoms increased over the next few days, particularly up to the twelfth day. On day 12, the crucial phase of this development occurred, coinciding with the administration of irrigated irrigation, which determines the assessment of PVX transmission (fig.15).

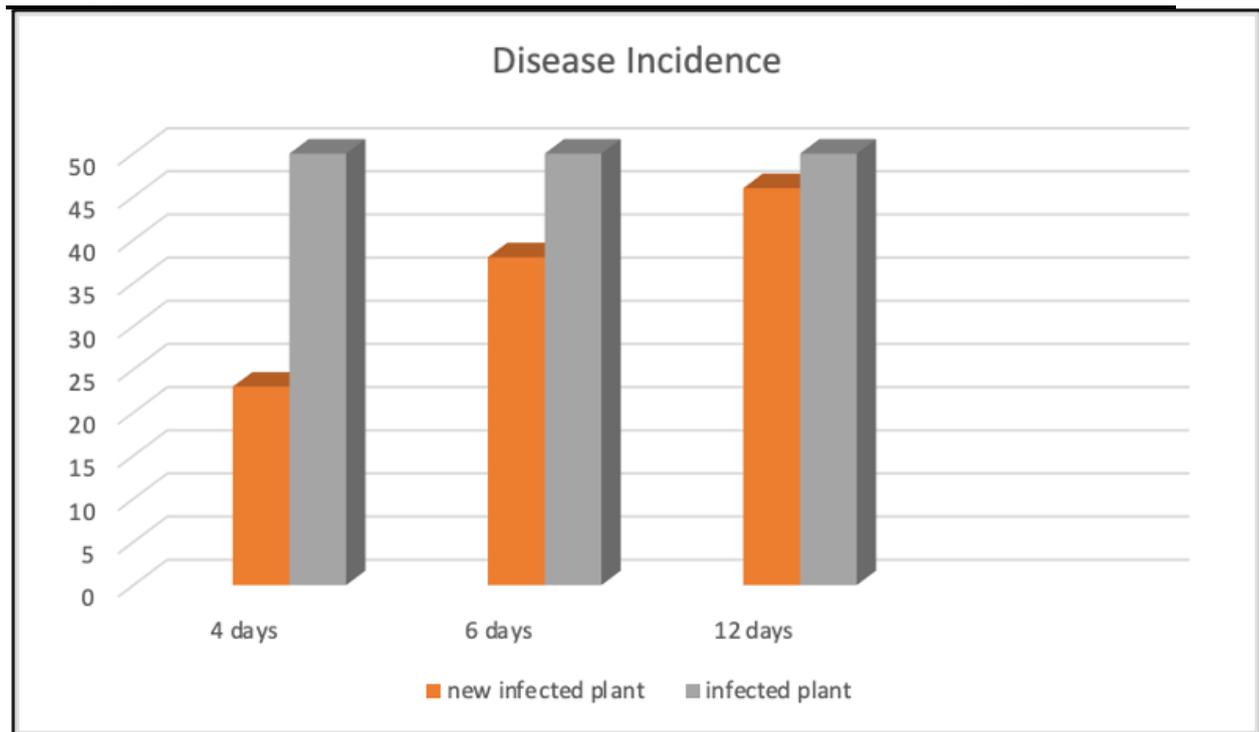


Fig.15. The figure shows the infection rates of potato virus (PVX).

The lead color represents seedlings infected before planting (mechanically inoculated) that showed symptoms of the virus, while the orange color represents seedlings that were infected after planting and the number of seedlings that showed symptoms during three dates, 4, 12, and 6 days, respectively.

A vast array of viruses can proliferate via irrigation water. These viruses can spread from the root of an infected plant to the root of another plant by irrigation water, according to several authorities (Tomlinson & FAITHFULL, 1984; Yarwood, 1960).PVX causes a variety of symptoms such as mottling and severe necrosis of leaves and stems (Arogundade et al., 2020).

Furthermore, despite their small quantity, a large variety of viruses that cause plant illnesses can infect the roots through irrigation water without the need for vectors. Via the root, these viruses have the ability to spread epidemics (Sevik, 2011). Water sources contain pathogens, which have the potential to spread. Reusing irrigation water through systems raises the possibility of disease outbreaks, as noted by Moorman in remarks from Pennsylvania State University Extension for greenhouse growers of ornamental crops(Dixon, 2015).

It was shown in a different study that viruses are often regarded as the most hazardous possible pathogens in water (Mehle et al., 2014).

Using the AutoCAD application, measurements were taken of a leaf sample from both a healthy and an infected plant (Fig. 16) (Table 4). With a leaf area of 2088.7 mm<sup>2</sup> and a diameter of 212.03 mm<sup>2</sup>, the infected and healthy leaves, as well as the plant, showed growth disparities when this program was executed successfully in this study. The width of the healthy leaf was 245.8 mm<sup>2</sup>, and its area measured 2985.3 mm<sup>2</sup>. Here, the afflicted leaf



and the healthy leaf are clearly different from one another. After the experiment was finished, a confirmatory double-containment ELISA test was performed on each seedling.

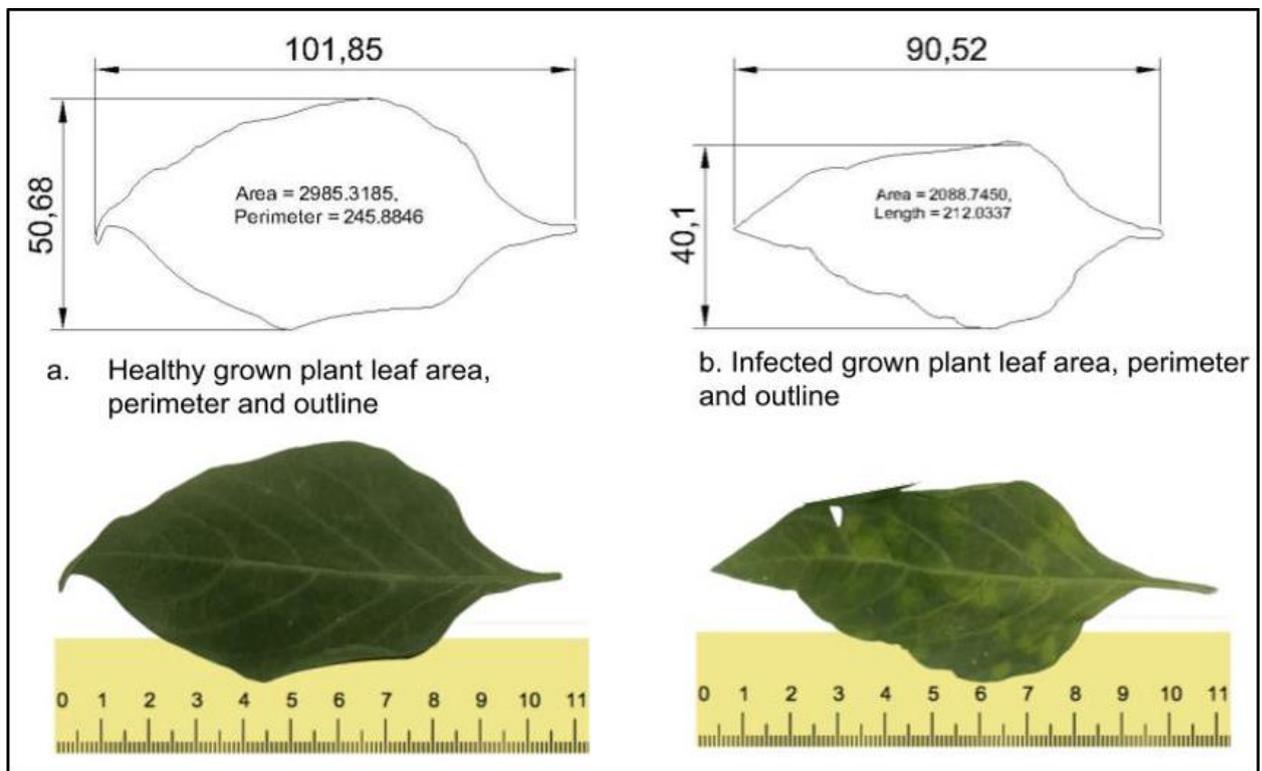


Fig.16. Using AutoCAD, the measurement results were as follows:

a. Measuring the leaf area of a healthy pepper plant mm<sup>2</sup> 2985.3

b. Leaves of infected plant mm<sup>2</sup> 2088.7

Table .4. Results of the AutoCAD program, where the number 1 represents the length, width, and surface area of the healthy leaf, the number 2 represents the growth dimensions of the leaf infected with the virus, and the number 3 represents the degree of difference.

No	Leaf sample	Length	Width	Perimeter (MM.L)	Area ( MM <sup>2</sup> )
1	healthy	50	101	245.8	2985.3
2	Infected	40	90	212.0	2088.7
3	Comparison	-10	-11	-33.8	-896.6

### 3-4 Transmission of the virus using contaminated water using drip irrigation

The outcomes of the potato virus-contaminated water drip irrigation experiment It reached (15 days/27%) on the fifteenth day, exhibiting mosaic symptoms with color changes between the veins. The symptoms persisted until the twenty-second day, at which point they were present in addition to some of the seedlings' mottling and swelling between the



veins (22 days/34%).(Fig. 17).



a.



b.

Fig.17. a. Symptoms of drip irrigation experience  
b. Comparison experiment without adding the virus.



The infection rate was calculated (fig.18) according to:

$$\text{Disease incidence} = \frac{\text{number of infected plant}}{\text{Total number of plant}} \times 100$$

Total number of plant

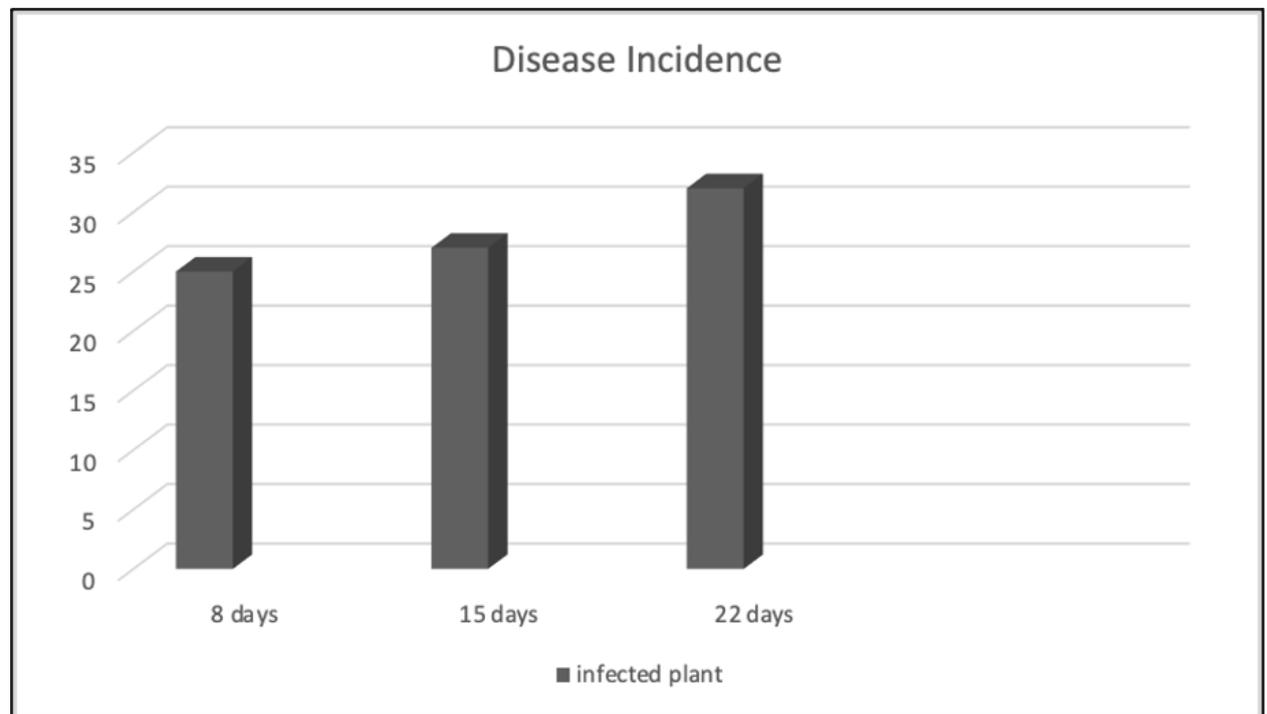


Fig.18. The figure represents the infection rate for the experiment of contaminating irrigation water with drip irrigation, where the black color represents the plant infection rate with the PVX virus during three dates of 8, 22, and 15 days respectively, giving a rate of 84%.

The possibility that the seedlings were infected through the root is supported by a very recent study that used water containing the TMV virus to irrigate tobacco seedlings. The study found that symptoms started to show up 14 days after watering and got worse until day 28. The study concluded that it is important to comprehend the risk of epidemics and water contamination with plant viruses because they can affect agricultural productivity (Zhang et al., 2023).

It was shown in another study that irrigation water can be a useful tool for viral dissemination. Drip irrigation system was used to water cucumber and tomato plants. Drip irrigation caused viral contamination every time since the most impacted organs were the roots, which were then followed by leaves and fruits. Viral contamination of the above- and below-ground portions of both crops resulted from drip irrigation (Alum et al., 2011).

Additionally, the researcher concurs that one of the most significant symptoms noted is the deficiency in both quantity and quality of fruits (Abdalla et al., 1991). According to the findings of an additional experiment, irrigation water, or the cucumber green mosaic virus (CGMV), tomato mosaic virus (ToMV), tobacco necrosis virus (TNV), and lettuce large



vein factor (LBYA), is the means by which these viruses are spread (Paludan, 1985).

Using the AutoCAD software, it is also possible to measure the growth rates and differences in size between an infected and a healthy leaf (Fig. 19) (Table 5). We acquired findings and growth disparities on the infected and healthy leaves, which was the leaf area of the infected leaf, thanks to the efficient use of this program in this study.

The leaf area of the healthy plant measured 4552.3 mm<sup>2</sup> with a diameter of 2325.4 mm<sup>2</sup>, while its measured 2347.8 mm<sup>2</sup> with a diameter of 226.0 mm<sup>2</sup>. Here, the diseased and healthy leaves differ noticeably from one another. After the experiment was finished, a confirmatory double-containment ELISA test was performed on each seedling.

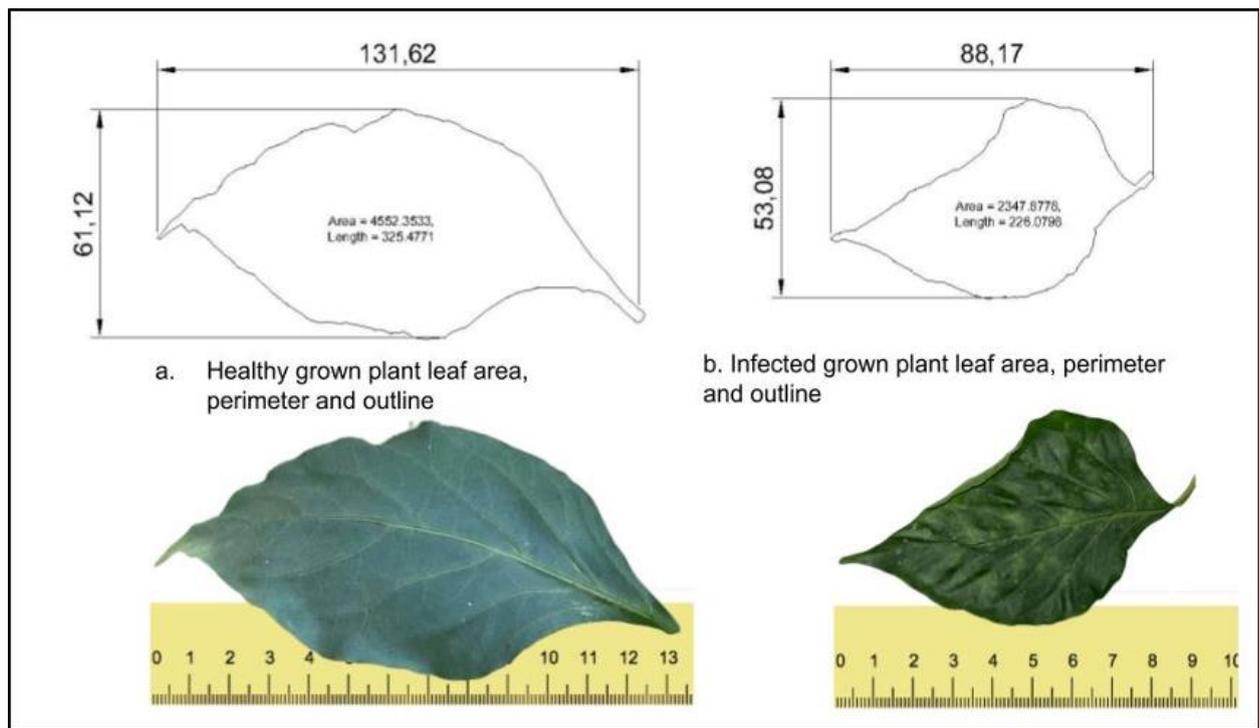


Fig.19. Using AutoCAD, the measurement results were as follows:

- a. The leaf area of a healthy pepper plant is 4552.3 mm<sup>2</sup>
- b. Leaves of the affected plant 2347.8 mm<sup>2</sup>

Table . 5 . Results of the AutoCAD program, where the number 1 represents the length, width, and surface area of the healthy leaf, the number 2 represents the growth dimensions of the virus-infected leaf, and the number 3 represents the degree of difference.

No	Leaf sample	Length	Width	Perimeter (MML)	Area ( MM <sup>2</sup> )
1	healthy	61	131	325.4	4552.3
2	Infected	53	88	226.0	2347.8
3	Comparison	-8	-43	-99.4	-2204.5



### conclusion

1. Since hydroponics with this technology recycles water, we may deduce that the Nutrient Film Technology (NFT) system has low consumption since it doesn't need a lot of nutrients or water to keep running. Furthermore, it is difficult for salts to build up on the surface due to the constant, steady flow. All crops produced hydroponically within the greenhouses are supported by the roots of the plants placed in the device, as well as by the large-scale standard design that makes it easy to expand the membrane networks inside and open several channels instead of only one.
2. The findings demonstrate that water recycled in hydroponic systems is utilized again in agriculture to reduce water usage and preserve resources. It also helps regulate growth rates by regulating nutrient levels.
3. Dual containment ELISA test findings shown a high degree of efficacy in identifying Potato Virus X (PVX). A spectrophotometer with a wavelength of 405 nm was used to measure absorption. A clear difference could be seen between the infected and healthy samples. The absorbance of the healthy samples was found to be 0.319, but the infected ones showed a considerable rise to 2.896. The data and favorable findings demonstrated the spread of the potato virus.
4. A comparison of healthy and infected pepper seedlings revealed the significant impact of PVX on the morphology and development of the plants. With the use of AutoCAD software, it was possible to see that the affected plants' leaves were smaller in length, breadth, perimeter, and area. For instance, in the hydroponics experiment, the area of infected leaves dropped from 1354.2 mm<sup>2</sup> (healthy) to 707.7 mm<sup>2</sup>, but in the field experiment including fake infection, a healthy pepper plant's leaf area measured 2985.3 mm<sup>2</sup>. Regarding the drip irrigation experiment, the measurement of the leaf area of the healthy pepper plant was 4552.3 mm<sup>2</sup>, whereas the leaves of the infected plant were 2347.8 mm<sup>2</sup>. The leaves of the infected plant measured 2088.7 mm<sup>2</sup>.
5. All of these findings add to a thorough knowledge of how PVX affects pepper plants grown in different irrigation and hydroponic systems. This study highlights the complex consequences of PVX on crop health by evaluating the virus's capacity to travel via root systems, impede development, and jeopardize physiological features. Additionally, its agricultural production in the field and on the water

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