

# APPLICATION OF ADVANCED MOLECULAR TECHNIQUES FOR CHARACTERIZING TYLCV IN CUCUMBER

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# Introduction

- Viral diseases of plants are causing significant economic losses in many crops, mainly tomato and, recently, cucumber.
- Cucumber growers have reported economic losses as high as 90-95% for the last several years.
- In Kuwait, tomato yellow leaf curl virus (TYLCV) was reported as a major pest of tomato and cucumber, but it was not fully characterized at the molecular level.
- Common symptoms on cucumber plants infected with TYLCV are: yellowing, upward leaf cupping, stunting, mosaicking, and leaf and fruit deformation.



## Common symptoms on cucumber plants infected with TYLCV





# Objective

- The aim of this study was to adapt and optimize a very sensitive, rapid method for the detection of tomato yellow leaf curl virus (TYLCV) in Cucumber.



# Methodology

## Sampling:

- 90 Cucumber leaf samples were collected monthly during the growing season.
- The collections were made from greenhouse farms in Wafra and Abdally.
- **Kuwait Map showing two agricultural areas.**
- **Al- Wafra** ★
- **Al-Abdally** ★







**Sampling**

Cucumber leaves

# Detection of TYLCV in Cucumber using PCR

## DNA Extraction:

DNA was extracted from 90 infected cucumber leaf samples using Dellaporta method for total genomic extraction (Dellaporta et al.,1983).

## PCR Amplification

Two pairs of primers TY1(+) and TY2(-) (Accotto et al., 2000), and TYC1R and TYC1F were used in the PCR protocol to detect TYLCV on 50 samples.





# Results

## Symptomatology

- Viral symptoms recorded included leaf yellowing, silvery, upward cupping, leaf deformation, leaf curling, and stunting.





## Plate 1. Mosaicking in cucumber plants.





## Plate 2. Yellowing in a cucumber plant.





## Plate 3. Mottling in a cucumber plant.



**Plate 4. Healthy and infected cucumber fruits in Al-Falah greenhouses, Abdally**





# Plate 5. Production of cucumber in greenhouse from Al- Jazy Farm



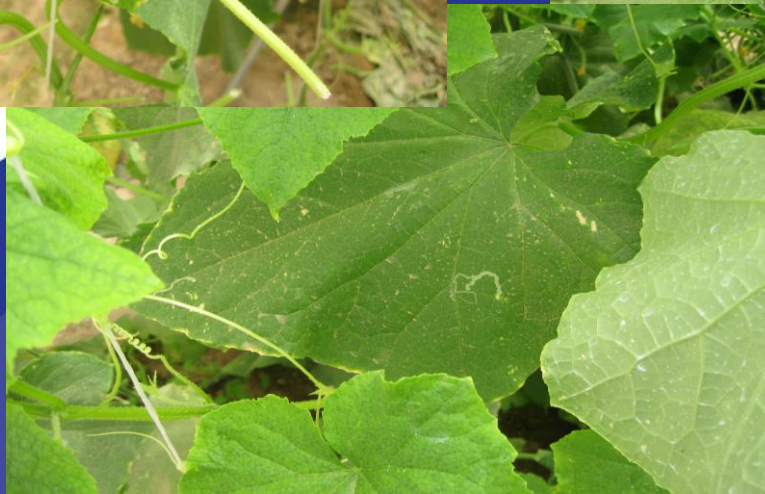


## Plate 6. Abnormality of cucumber fruit grown in Al-Falah Greenhouse, Abdaly





## Plate 7. Leaf minor tunnels and yellowing of cucumber leaves from Al-Thawab farm





## Plate 8. Cucumber plants at Dloo Grenhouses, wafra

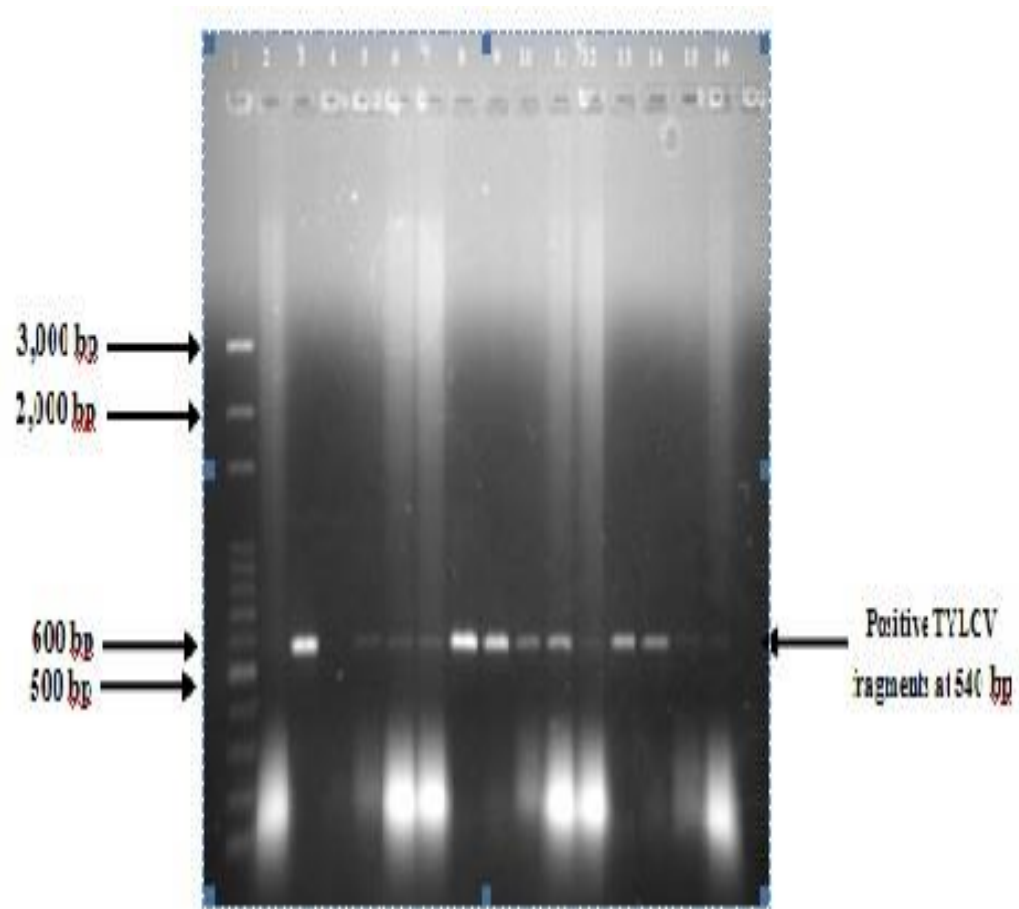




## **Detection of TYLCV in Cucumber using PCR**

- PCR tests revealed that 35 out of 50 samples were positive that gives a band with a size of 540 bp, which ensures the presence of TYLCV in cucumber.
- Best results were performed by TY1 and TY2 primer pair.
- Positive samples were stored for further analysis and sequencing for studying the genetic characterization of the virus.

## PCR Gel





# Conclusion

- Best results were performed by TY1 and TY2 primer pair, as the band with a size of 540 bp was noticed very clearly. Positive samples were stored for further analysis.

# Thank You

