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# Short Notes: First Report of a Co-Infection of Squash Vein Yellowing Virus and Tomato Leaf Curl Palampur Virus in Cucumber Plants in Iraq

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**Abstract:** Mosaic disease causes a serious epidemic of cucumber plants in Iraq. To investigate causal agents of such symptoms, whole genome and metatrancriptomic sequencing data were collected from pathogenic cucumber leaves. The result shows that *Tomato leaf curl Palampur virus* DNA-A and B were existed with 2,756 and 2,719 bp sequences that deposited under acc. numbers ON229618 and ON229620 respectively, and the isolate named Babylon1. Further, *Squash vein yellowing virus* was found with 9,832 bp sequence that deposited under acc. number ON229619, and the isolate called Iraq. Mixed infection reported here for the first time in cucumber in Iraq, highlighting the impact of such infection that threat the cucumber fields.

**Keywords:** Bioinformatics, Cucumber genome, Geminiviridae, Potyviridae, RNA seq, Total DNA reads.

*Tomato leaf curl Palampur virus* (TLCPV), as a member of the family Geminiviridae causes mosaic disease in Cucurbitaceae plants in low tunnels, greenhouses, and open fields. The disease causes a devastating epidemic of melon, cucumber, and squash. Throughout the Middle East, this virus has spread rapidly, threatening cucurbites production (Heydarnejad *et al*., 2013; Dhkal *et al*., 2020; Adhab & Alkuwaiti., 2022). *Squash vein yellowing virus* (SqVYV) was first detected in Florida in 2007 (Hernandez *et al*., 2021). The virus belongs to the Potyviridae, genus *Ipomovirus*, known to infect cucurbits (Jailani *et al*., 2021; Inoue-Nagata *et al*., 2022). There is an interesting correlation between circulatory persistence and semi-persistent transmission of TLCPV and SqVYV by whiteflies *Bemisia tabaci* (Kumar & Kumar 2018; Kavalappara *et al*., 2021). The leaves of an infected cucumber plant in Babylon Province were collected on 22nd October 2021 (Fig. 1A), and then cut into squares of 0.5×0.5 cm. A single Eppendorf tube containing five times as much RNAlater (2 ml) was then sent for sequencing to the company of DNAlink in South Korea. A whole genome sequencing was performed on the extracted DNA and RNA (Platform: Novaseq6000; Applications: WGS Nano550 and WTS/mRNA, respectively). DNA reads accounted for 89,893,674 clean reads, whereas RNA reads accounted for 53,458,772. In order to create one representative reference sequence, 5040 suspected viruses were downloaded from NCBI-GenBank and were concatenated to form one 76,145,671 nucleotide sequence. A reference genome-wide clean read mapping using Geneious software was performed on

whole DNA and RNA (http://www.geneious.com/). As a result, 1,055,827 and 1,347,785 reads were assembled against TLCPV DNA-A and TLCPV DNA-B to produce 2,756 and 2,719 bp consensus sequences, respectively, which have been deposited under accession numbers ON229618 and ON229620. The isolate has been named Babylon1. As shown in Fig. 1 C and D, TLCPV DNA-A encodes six protein domains (AV1, AV2, AC1, AC2, AC3, and AC4), and TLCPV DNA-B encodes two proteins (BV1, BC1). Further, 196,675 reads were assembled against SqVYV and 9,832 bp consensus was produced and then deposited in GenBank under accession number ON229619, and the isolate named Iraq (Fig. 1B). SqVYV has ten protein domains involved in one open reading frame (P1a, P1b, P3, 6K1, C1, 6K2, VPg, Pro, Replicase and CP). The analysis of phylogeny shows that SqVYV was very close to Israeli isolate (Fig. 2), while TLCPV DNA-A and TLCPV DNA-B were in high similarity with Iranian isolates (Figs. 3 and 4). For the first time in Iraq, the co-infection of the two viruses has been reported in cucumber, highlighting the impact of double virus infections that spread epidemically across the cucumber fields.

## Contributions of authors

Project supervision was provided by HA, OA, and FF. In collaboration with HA, OA carried out the experiments and analyzed the data. OA and FF drafted the final version of the manuscript, which was revised and approved by all authors.

## Conflicts of interest

The research was not conflicted by any commercial or financial relationships.

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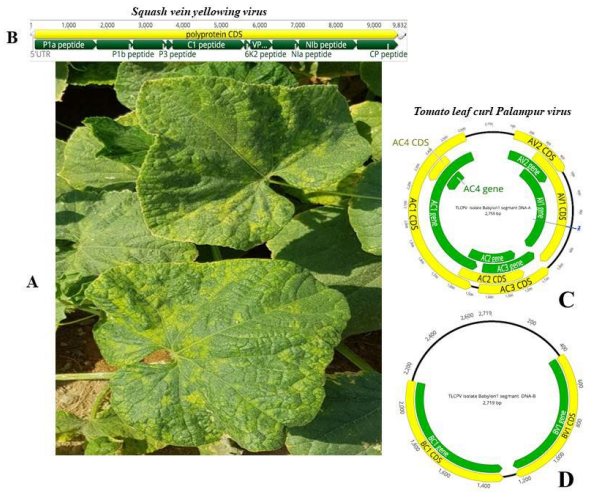


**Fig. (1): Symptomatic cucumber leaves show typical mosaic symptoms caused by co-infection of SqVYV and TLCPV (A). Complete sequence of SqVYV shows ten protein domains (B). Two DNA segments of TLCPV, A (C), and B (D).**

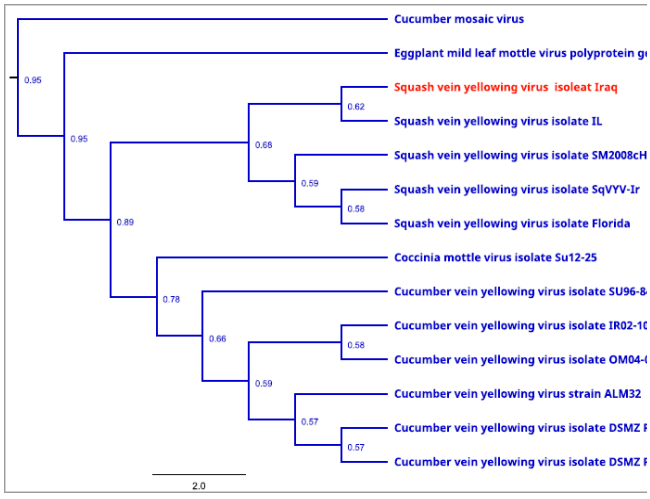


**Fig. (2): Geneious tree builder was used to build the tree of SVYV, and ClustalW was used to align the nucleotide sequences. The out group member was *Cucumovirus Cucumber mosaic virus*.**

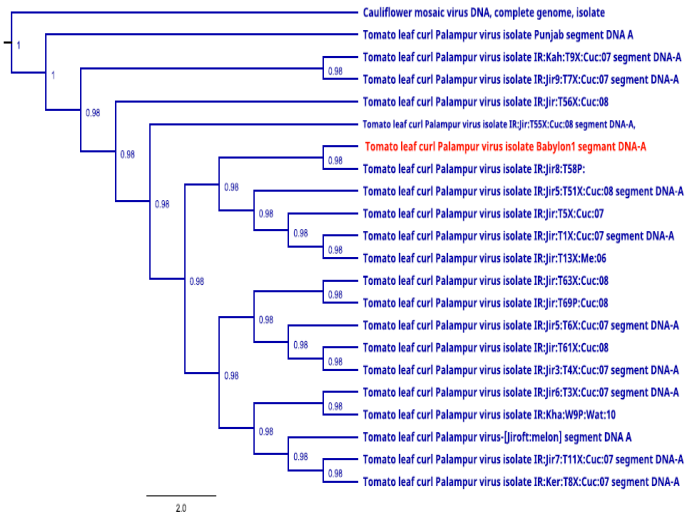


**Fig. (3): ClustalW was used to align full-genome nucleotide sequences of TLCPV DNA-A and build the tree using Geneious tree builder. The out group member was *Caulimovirus Cauliflower mosaic virus.***

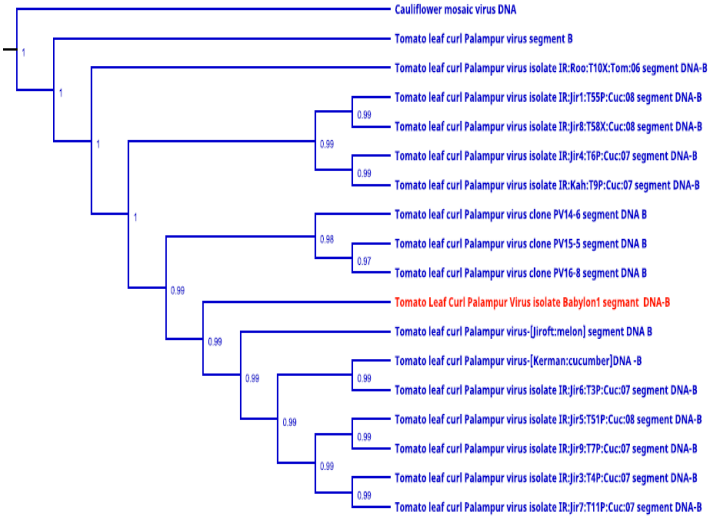


**Fig. (4): Full genome nucleotide sequences of TLCPV DNA-B were aligned using ClustalW using Geneious tree builder. The out group member was *Caulimovirus Cauliflower mosaic virus*.**

# التقرير الأول عن الإصابة المشتركة بفايروس اصفرار عروق الشجر وفايروس تجعد أوراق الطماطة البالامبوري في نباتات الخيار في العراق

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**المستخلص**: يسبب مرض الموزائيك وباء خطير على نباتات الخيار في العراق. و للتحقق من العوامل المسببة لمثل هذه الأعراض، تم جمع بيانات الجينوم الكامل وبيانات الاستنساخ الكلية من أوراق الخيار المصابة. أظهرت النتيجة وجود فايروس تجعد أوراق الطماطه البالامبوري بتسلسلين 2,756 و2,719 زوج قاعده. تم إيداع التسلسلين وتم تسمية العزلة العراق . علاوة على ذلك، تم العثور على فيروس اصفرار عروق الشجر بتسلسل 9,832 تم إيداع التسلسل تحت رقم إيداع وسميت العزلة بابيلون1. تم تسجيل الإصابة المختلطة هنا لأول مره في الخيار في العراق ON229619 ON229618 و ON229620 تحت الأرقام اعلاه، مما يسلط الضوء على تأثير هذه العدوى التي تهدد حقول الخيار.

**الكلمات المفتاحية:** برامج المعلوماتية الحياتية، جينوم الخيار، عائلة الفايروسات التوأمية، عائلة واي البطاطا، التسلسل الكامل للجينوم، تسلسل الكامل لبيانات الاستنساخ.