

# UC Riverside

## International Organization of Citrus Virologists Conference Proceedings (1957-2010)

### Title

Immunoblot Detection of Citrus Psorosis in Israel Using Citrus Ringspot Antiserum

### Permalink

<https://escholarship.org/uc/item/83b6j0z1>

### Journal

International Organization of Citrus Virologists Conference Proceedings (1957-2010), 12(12)

### ISSN

2313-5123

### Authors

Da Graça, J. V.  
Bar-Joseph, M.  
Derrick, K. S.

### Publication Date

1993

### DOI

10.5070/C583b6j0z1

Peer reviewed

# Immunoblot Detection of Citrus Psorosis in Israel Using Citrus Ringspot Antiserum

J. V. da Graça, M. Bar-Joseph and K. S. Derrick

**ABSTRACT.** Antiserum to citrus ringspot virus-4 was used to detect the presence of a 48-Kd protein in extracts of citrus and *Chenopodium quinoa* infected with Israeli strains of psorosis by immunoblot. Healthy plants and impietratura-infected citrus did not contain such a protein. These results confirm the serological relationship between ringspot and psorosis.

Citrus ringspot virus (CRSV) was shown to be a two-component RNA virus with short (300-500 nm) and long (1500-2000 nm) highly flexible filamentous particles, with a 48-Kd putative capsid protein (2, 3). Antiserum to the protein of CRSV-4, a non-bark-scaling isolate from Florida (6), was used to trap the two types of particles from infected citrus and *Chenopodium quinoa* by serologically-specific electron microscopy (2) and to detect the viral protein on nitrocellulose blots of electrophoresis gels (4). Other strains of citrus ringspot, as well as symptomatically similar citrus psorosis from Argentina, Spain and Florida also have infectivity associated with two components and possess a 48-Kd protein (1, 5, 8). Although these results all suggest that citrus psorosis and ringspot are related, there are also indications that psorosis in California is associated with a carlavirus (7).

Citrus psorosis is a worldwide disease (9), and an investigation on the nature of the causal agent in Israeli citrus was undertaken.

Various citrus types (grapefruit, sour orange and *Citrus macrophylla*) were inoculated with psorosis, and young symptomatic leaves were used to inoculate seedlings of *C. quinoa*, a local lesion host for both psorosis and ringspot (7). Typical lesions appeared on the inoculated leaves after 10 days (Fig. 1).

Extracts of symptomatic and healthy citrus and *C. quinoa* leaves were made as previously described (2). Extracts of impietratura-infected citrus leaves were similarly prepared to con-



Fig. 1. Local lesions on a *Chenopodium quinoa* leaf inoculated with Israeli psorosis.

firm the absence of CRSV-related protein (1). After separation in sucrose gradients, fractions previously shown to contain the CRSV 48-Kd protein were concentrated and separated on a 12% SDS-polyacrylamide gel, and then transferred to a nitrocellulose membrane in a Bio-Trans Mini Gelman Sciences semi-dry blotter. Immunostaining of the blot was done using antiserum to CRSV-4 produced in rabbits (4), and after reaction with anti-rabbit Ig-horse radish peroxidase conjugate the blot

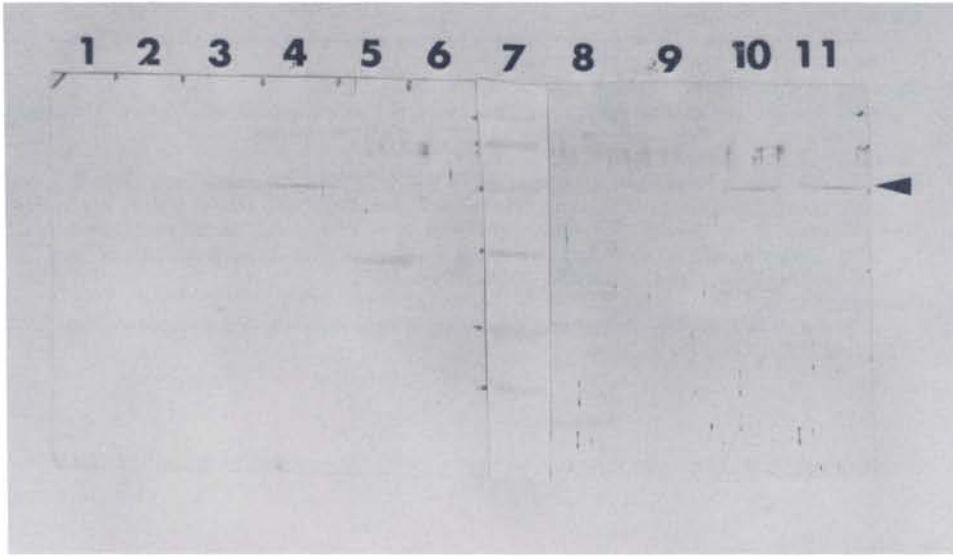


Fig. 2. Immunoblot of SDS-polyacrylamide gel of top and bottom fractions of extracts of healthy and psorosis-infected citrus and *Chenopodium quinoa*, and impietratura-infected citrus. (Arrow indicates 48-Kd)

- Lane 1 - healthy citrus (top fraction)
- Lane 2 - healthy citrus (bottom fraction)
- Lane 3 - psorosis-infected citrus (top)
- Lane 4 - psorosis-infected citrus (bottom)
- Lane 5 - impietratura-infected citrus (top)
- Lane 6 - impietratura-infected citrus (bottom)
- Lane 7 - molecular weight standards (97.4, 66.2, 45, 31, 21.5 & 14.4 Kd)
- Lane 8 - healthy *C. quinoa* (top)
- Lane 9 - healthy *C. quinoa* (bottom)
- Lane 10 - psorosis-infected *C. quinoa* (top)
- Lane 11 - psorosis-infected *C. quinoa* (bottom)

was developed in 4-chloro-1-naphthol. A sharp band corresponding to 48-Kd was observed in both top and bottom fractions of psorosis-infected citrus and *C. quinoa*, but not in any healthy plant extracts, nor in the impietratura sample (Fig. 2).

These results provide further support for the proposal that citrus ring-spot and psorosis are caused by similar

serologically related agents and that impietratura is an unrelated disease (1).

#### ACKNOWLEDGEMENTS

The visit of the senior author to Israel was funded by the South African Foundation for Research and Development, and the Israeli Ministry of Science and Technology.

#### LITERATURE CITED

1. da Graça, J. V., R. F. Lee, P. Moreno, E. L. Civerolo and K. S. Derrick  
1991. Comparison of isolates of citrus ringspot, psorosis, and other viruslike agents of citrus. *Plant Dis.* 75: 613-616.
2. Derrick, K. S., R. H. Brlansky, J. V. da Graça, R. F. Lee, L. W. Timmer and T. K. Nguyen  
1988. Partial characterization of a virus associated with citrus ringspot. *Phytopathology* 78: 1298-1301.
3. Derrick, K. S., R. H. Brlansky, R. F. Lee, L. W. Timmer and T. K. Nguyen  
1988. Two components associated with citrus ringspot virus, p. 340-342. *In: Proc. 10th Conf. IOCV. IOCV, Riverside.*
4. Derrick, K. S., R. F. Lee, B. G. Hewitt, G. A. Barthe and J. V. da Graça  
1991. Characterization of citrus ringspot virus, p. 386-390. *In: Proc. 11th Conf. IOCV. IOCV, Riverside.*

5. García, M. L., E. L. Arrese, O. Grau and A. N. Sarachu  
1991. Citrus psorosis disease agent behaves as a two component ssRNA virus, p. 337-344. *In: Proc. 11th Conf. IOCV. IOCV, Riverside.*
6. Garnsey, S. M. and L. W. Timmer  
1980. Mechanical transmissibility of citrus ringspot virus isolates from Florida, Texas and California, p. 174-179. *In: Proc. 8th Conf. IOCV. IOCV, Riverside.*
7. Levy, L. and D. J. Gumpf  
1991. Studies on the psorosis disease of citrus and preliminary characterization of a flexuous virus associated with the disease, p. 319-336. *In: Proc. 11th Conf. IOCV. IOCV, Riverside.*
8. Navas-Castillo, J., P. Moreno, J. F. Ballester-Olmos, J. A. Pina and A. Hermoso de Mendoza  
1991. Detection of a necrotic strain of citrus ringspot in Star Ruby grapefruit in Spain, p. 345-351. *In: Proc. 11th Conf. IOCV. IOCV, Riverside.*
9. Roistacher, C. N.  
1975. Psorosis A. *In: Description and illustration of virus and virus-like diseases of citrus. Vol. II, SETCO-IRFA, Paris. 10 pp.*