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in

D'Onghia A.M. (ed.), Djelouah K. (ed.), Roistacher C.N. (ed.). Citrus tristeza virus and Toxoptera citricidus: a serious threat to the Mediterranean citrus industry

Bari : CIHEAM Options Méditerranéennes : Série B. Etudes et Recherches; n. 65

2009 pages 165-171

Article available on line / Article disponible en ligne à l'adresse :

http://om.ciheam.org/article.php?IDPDF=801410

To cite this article / Pour citer cet article

Santoro F., Gualano S., Djelouah K., Guario A., D'Onghia A.M. **Remote sensing to support the monitoring of Citrus tristeza virus (CTV) infected areas.** In : D'Onghia A.M. (ed.), Djelouah K. (ed.), Roistacher C.N. (ed.). *Citrus tristeza virus and Toxoptera citricidus: a serious threat to the Mediterranean citrus industry.* Bari : CIHEAM, 2009. p. 165-171 (Options Méditerranéennes : Série B. Etudes et Recherches; n. 65)



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Remote sensing to support the monitoring of *Citrus tristeza virus* (CTV) infected areas

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Abstract. *Citrus tristeza virus* (CTV) is the most destructive pathogen in *Citrusspp.*. It is therefore necessary to promptly identify suspected infected trees on a large scale by the use of satellite imaging. To this aim, the vegetation indices of CTV-infected and CTV-negative citrus trees were preliminarily investigated in order to provide the tree spectral signature detected in the field by the HandHeld Post Dispersive Spectrometer. The study was carried out in selected commercial groves ('Precoce di Massafra' clementine and 'Navelina' orange) located in the CTV foci area, both showing a high CTV infection rate. The trees were previously tested by serological (DTBIA and ELISA) and molecular (PCR) tools to assess the virus presence. The preliminary results highlighted a difference in the spectral signatures of CTV-infected and CTV-free trees, allowing a discrimination of the canopy stress level based on these results, the use of vegetation indices from high-resolution multispectral satellite imaging may represent a valid tool backing up the pathogen monitoring in wide areas.

Keywords. Citrus – CTV – Spectroradiometer – Spectral signatures – PCA – Vegetation indices.

La télédétectiondans le suivideszonesinfestées par le CTV

Résumé. Le virus de la tristeza des agrumes est le pathogène le plus redoutable chez l'espèce Citrus. Il est donc nécessaire d'identifier rapidement, et sur une grande échelle, les arbres suspectés d'être infectés à l'aide des images satellites. A cet effet, les indices de végétation des arbres d'agrumes infectés ou non infectés par le CTV ont été évalués préliminairement afin de fournir les signatures spectrales décelées au champs par spectrométrie (HandHeld Post Dispersive Spectrometer). Cette étude a été menée dans des vergers commerciaux sélectionnés (où on cultivait le clémentinier 'Precoce di Massafra' et l'oranger 'Navelina'), dans la zone où étaient localisés les foyers de CTV, qui montraient un taux d'infection de CTV élevé. Les arbres avaient préalablement été testés par voie sérologique (DTBIA et ELISA) et moléculaire (PCR) en vue d'évaluer la présence du virus. Les résultats préliminaires ont mis en évidence une différence dans les signatures spectrales des arbres infectés et des arbres non infectés par le CTV, permettant ainsi de faire une distinction concernant le taux de stress du feuillage sur la base des indices de végétations estimés. Un niveau supplémentaire de variation a été déterminé selon l'espèce. Les résultats obtenus, l'utilisation des indices de végétation fournis par les images satellites multi-spectrales à haute résolution peuvent représenter un outil valable pour le suivi de ce pathogène sur de vastes étendues.

Mots-clés. Agrumes – CTV – Spectroradiomètre – Signatures spectrales – PCA – Indices de végétation.

I – Introduction

Citrus tristeza closterovirus (CTV) is one of the principal causes of damaging and of destruction of citrus in the world. The large scale monitoring of the virus in the citrus growing areas is necessary in order to prevent the entrance and spread of the virus in virgin areas and to control the virus where it is not established yet through the application of eradication measures.

The virus monitoring and eradication are usually supported by aerial photos, for the identification and mapping of the groves, followed by hierarchical tree sampling and virus detection by DTBIA or ELISA.

However, this activity encounters several constraints which make the control of CTV highly difficult to manage in a sustainable manner because of the virus natural spread by aphid vectors. These factors are mainly related to the difficulty in choosing the right area to be monitored, to the symptomless status of the trees, to the time-consuming sampling and laboratory detection assays. Therefore, the success of this program, which is focusing on the virus monitoring in combination with its eradication, is based on the rapid identification and elimination of the infected trees in order to avoid the large virus dissemination, thus eliminating or maintaining the virus inoculum at very low levels.

To this aim, the remote and the proximal sensing techniques, successfully applied for early detection of apple plant stress caused by *Venturia inaequalis* (Delalieux *et al.*, 2007), of spider mite damage in peach orchards in California and of water stress in an olive orchard with thermal remote sensing imagery (Sepulcre-Cantó *et al.*, 2006), showed to be very promising in the detection of CTV infected trees in commercial citrus groves.

The set of data provided by the above-mentioned methodologies may be synthesized through individual analytical parameters called vegetation indices. These parameters are calculated from the sum, difference, ratio, or other linear combinations of two or more spectral bands. They are widely used in modeling studies to estimate the greenness or productivity of vegetation based on the reflectance value (Asrar *et al.*, 1989).

The setting up of proximal and remote sensing in the monitoring of CTV was firstly carried out in the framework of a research project supported by Osservatorio Fitosanitario Regionale of Apulia region, Italy.

II - Proximal sensing

Proximal sensing was applied in CTV infected groves of 'Navelina' orange and 'Precoce di Massafra' clementine, where plants were virus tested by serological and molecular means.

The spectral signature was measured on selected CTV-infected and CTV-negative trees to highlight the *reflectance* differences at various wave lengths. Measurements have been made through a field spectroradiometer, FieldSpec® HandHeld by ASD, placed on the tree canopy at a height h and nadir direction (Fig. 1).



Figure 1. Geometrical scheme of spectroradiometer measurement.

For each measurement, the *white reference* (internal reference for the instrument) procedure has been applied, through a thermoplastic "disc" (Spectralon) and, later, through the acquisition of a set of points which measure the canopy *reflectance*. The results achieved are processed in a single spectral curve (signature), which is representative of the tree canopy spectrum (Fig. 2).



Figure 2. Results of spectral signatures measured in the clementine (a) and sweet orange (b).

For the analysis of the dataset collected in the field and for recognizing differences in the wave shapes of infected plants spectra, a multivariate statistical analysis has been conducted through the PCA (Principal Components Analysis) algorithm (Jolliffe, 2002).

PCA on spectral signatures, aiming at discriminating between CTV-positive and CTV-negative plants, has been based on the dataset of the studied species orange and clementine).

For 'Navelina' orange, discrimination between infected and healthy trees is clear-cut, thereby showing a high separability (Fig. 3).



Figure 3. PCA projection of cases for sweet orange.

In contrast, for 'Precoce di Massafra' clementine, the result has not been satisfactory, thereby showing a low separability (Fig. 4).



Figure 4. Scatter plot PC1-PC2 for clementine.

In order to synthesize and concentrate in single numerical parameters the wide array of information contained in the spectral signatures (reflectance values in the various wave lengths), two vegetation indices have been calculated: NDVI and mCAI (Fig. 5, 6).



Figure 5. Some vegetation indices of CTV-negative and CTV-positive clementines.

Normalized Difference Vegetation Index sweet orange



Figure 6. Some vegetation indices of CTV-negative and CTV-positive sweet orange.

The Normalized Difference Vegetation Index (NDVI) is widely applied to recognize vegetated areas; it is very sensitive to the presence of biotic symptomatologies (Fletcher *et al.*,2004).

The mCAI or mCARI analytical index (modified Chlorophyll Absorption Index or modified Chlorophyll Reflectance Absorption Index) is used to estimate the vigour and the health status of the vegetation (Oppelt and Mauser, 2001).

Both analytical indices, calculated on the basis of spectral signatures, show some variability with respect to laboratory results on the presence-absence of CTV infection. In particular, results concerning 'Navelina' orange matched laboratory assays. In contrast, for those concerning 'Precoce di Massafra' clementine, mCAI showed some correlation (but not on the whole sample); and the NDVI gave a reliability slightly exceeding 60%.

On the whole, both vegetation indices provide a good indication about the stress status of the canopy which is related to the presence of the virus.

III – Remote sensing simulation

In order to estimate the vegetation indices (e.g. NDVI) corresponding to the spectral bands of the high-resolution satellite QuickBird, a simulation study has been started.

Results report a satisfactory overlapping between the values of NDVI calculated with the spectroradiometer (blue) and those estimated by QuickBird (red) (Fig. 7).



Figure 7. Results of NDVI simulation from QuickBird imagery.

The result is backed up by a satisfactory correlation coefficient, expressed by a regression study, in which the NDVI calculated and that estimated (in all the citrus species under study) have shown a strong correlation averaging 99% (Fig. 8).



Figure 8. Correlation coefficient of the NDVI calculated and estimated.

The result will be downsized in that the simulation does not take into account the effect of the atmosphere on the image.

IV – Conclusions

These data, combined with adequate statistical analysis, could clearly separate citrus species as clementine and sweet orange. Moreover, the application of vegetation indices (NDVI, mCAI) showed a different variability according to the presence or the absence of the CTV infection in relation to the tested species. Applied to the sweet orange, these indices clearly discriminated between the CTV-positive and CTV-negative trees, whereas a very slight discrimination occurred in the case of Clementine. Therefore, these parameters showed a useful indication about the canopy stress which is apparently highly linked to the presence of CTV infection, even in infected asymptomatic trees.

This study provided preliminary results in the assessment of CTV infection through the reflectance variability; nevertheless, further investigation about the real interaction between the presence of the CTV in the tree as the main source of stress and the reflectance variability is needed.

The proximal sensing technique based on reflectance data could be combined with the satellite imagery and become a highly promising tool for the CTV monitoring on a large scale.

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