IDENTIFICATION AND QUANTIFICATION OF CYST NEMATODE IN SUGAR BEET SEEDS BY HYPERSPECTRAL NEAR INFRARED IMAGING

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Introduction

Yield reduction in relation to the cyst nematode number in the sugar beet roots

The damage caused by nematode on the sugar beet root lead to a yield reduction and is related to the cyst number. The current work, carried out in collaboration with SESVANDERHAVE Company, aims to assess by hyperspectral NIR imaging the presence of cyst nematode on sugar beet root. The objective of the study is to discriminate between cyst, root and soil support as well as to quantify the cyst nematode presence.

Brown and white cyst nematode (*Heterodera schachtii*)

Materiel and methods

For this experiment, 30 plants of sugar beet with different level of resistance, were grown in a soil support spread in plastic plates: 20 plants were infested with cyst, 10 plants were not infested and were used as control. The number of cyst nematodes was previously counted by optical microscopy at SESVANDERHAVE. Then, to cover the root area, 4 images by plant were acquired with the hyperspectral imaging system installed at CRA-W (Figure 1). The instrument used is a MatrixNIR® Chemical Imaging System (Malvern instruments Ltd) recording sequential images with an InGaAs array detector (240x320 pixels) active in the 900-1700 nm range, that means 76 800 spectra per image. The data treatment was carried out under Matlab 7.5.0 (R2007b).

For the identification of cysts, 30 spectra have been selected for each type of structure that can be found in the plate (root, cyst and soil support). Spectra were preprocessed with 1st Derivative and a PCA was performed.

For the quantification, four spectral libraries (cyst, root, soil support and background including plastic plate and teflon support) have been built by selecting pixels in the images of 4 plants. Those libraries were used for the building of discrimination equations in order to identify cysts from root, soil support particles and background. SVM, Support Vector Machines, was used as classification method for the construction of these models. Three equations have been built: "background *vs.* soil support + root + cyst", "soil support *vs.* root + cyst" and "root *vs.* cyst". Then, these equations have been applied successively to all the pixels in the images of the 30 plants in order to build a mask, by isolating the cysts, and then calculate the number of pixels detected as cysts by surface unit.

Instrument parameters:

- Wavelength range: 900-1700 nm by step of 10 nm
- Average on 16 coadds/wavelength and 4 scans/image
- 1 acquisition = 1 image = 240 * 320 pixels = 76800 spectra
- 1 pixel = 80µm * 80 µm = 0.0064 mm2
- Analysed surface = +/- 5cm2
- Time of acquisition = 5 min/image

Cyst analysis:

- root system including cysts in a soil support spread in a plastic plate
- 1 cyst = +/- 25 pixels = +/- 25 spectra
- Analysed surface = +/- 5cm2 of the soil support

Figure 1: Image acquisition on sugar beet seedling with hyperspectral imaging system

Results

Regarding the identification, the PCA performed on the preprocessed data shows that the cysts can be clearly discriminated from the root. Results were showed on a poster presented at EASIM-2009, the 2nd general conference in spectral imaging.

Regarding the quantification, four spectral libraries have been built by selecting pixels in the four images of four plants: 3300 pixels corresponding to background (teflon + plastic), 20200 pixels corresponding to soil support, 7400 pixels corresponding to roots and 680 pixels corresponding to cyst nematodes. Figure 2 shows the picture of one seedling with root system and cysts as well as the results of the data treatment applied on the 4 images acquired to cover the full plastic plate. The data treatment on each image consists of 4 steps (a to d) allowing to detect the cysts. Figure 2a shows the image at 1000 nm without preprocessing. Figure 2b shows the pixels detected as soil support, roots and cysts after "background *vs.* soil support + root + cyst" equation application on the image of Figure 2a. Pixels corresponding to background (plastic plate and teflon support) are displayed in white color. Figure 2c shows the pixels detected as roots and cysts after "soil support *vs.* root + cysts" equation application on the not white pixels of Figure 2b and after removing of pixels with intensity > 0.15. Those pixels correspond to a light color. Pixels classified as soil support are displayed in white. Figure 2d shows the pixels detected as cysts after "root *vs.* cysts" equation application on the not white pixels of Figure 2c. All the pixels not classified as cysts are displayed in white color. The rest includes pixels detected as cysts displayed in black.

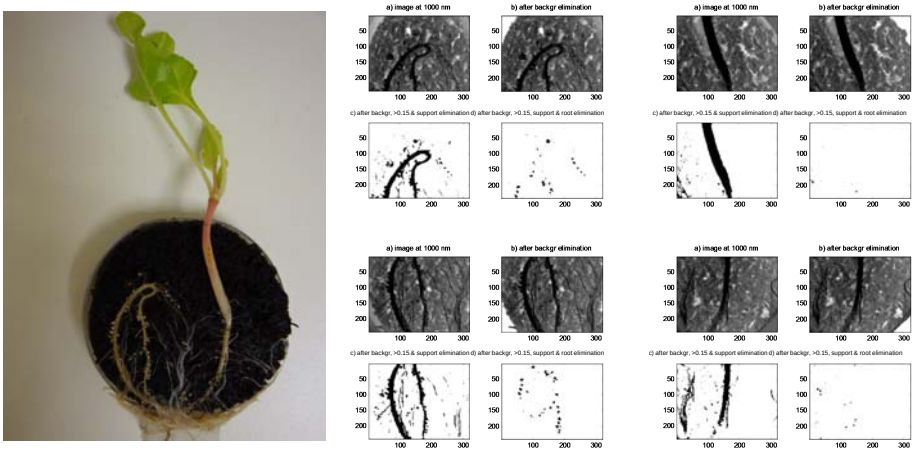

Figure 2: real picture and 4 corresponding acquired images with data treatment: (a) image at 1000 nm; (b) pixels detected as soil support, roots and cysts after "background (plastic plate and teflon support) *vs.* soil support + root + cyst" equation application on the image of Figure a; (c) pixels detected as roots and cysts after "soil support *vs.* root + cysts" equation application on the not white pixels of Figure b and after removing of pixels with color intensity > 0.15; (d) pixels detected as cysts after "root *vs.* cysts" equation application on the not white pixels of Figure c.

A correlation of 0.65 has been calculated between the number of cysts counted on the roots under optical microscopy and the number of pixels recognized as cyst nematode by the hyperspectral NIR imaging. Figure 3 shows this relation achieved with the results of the 30 plants. It has to be noted that cysts are counted on the total surface of the glass plate while NIR imaging analyses the total surface by acquisition of 4 images. Values have been corrected to take into account of the overlapping between some images.

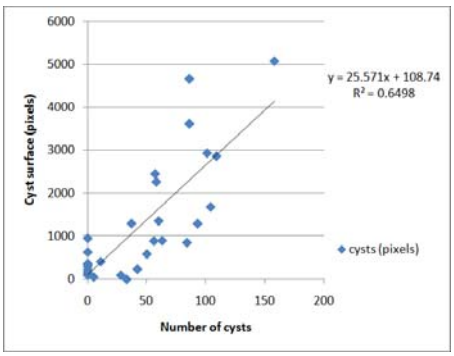

Figure 3: Relation between cyst number (Ref Val) and cyst surface (NIR imaging)

Conclusion

This study showed the potential of the hyperspectral NIR imaging to discriminate the cysts from the root and the soil support in a sugar beet root as well as to quantify the number of cysts. To avoid some overlapping between images taken on the same plant, additional images will be acquired covering the total surface of the glass plate using a different optical configuration.

References

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