Identification and quantification of cyst nematode in sugar beet seeds by hyperspectral near infrared imaging

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Introduction

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

Material and methods

For this experiment, 30 plants of sugar beet with different levels of resistance were grown in a soil support spread in plastic plates: 20 plants were infested with cysts, 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 on individual plants 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 (240×320 pixels) active in the 900–1700 nm range, which means 76,800 spectra per image.[1] The data treatment was carried out using Matlab 7.5.0 (R2007b).

For the identification of cysts, 30 spectra were 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.

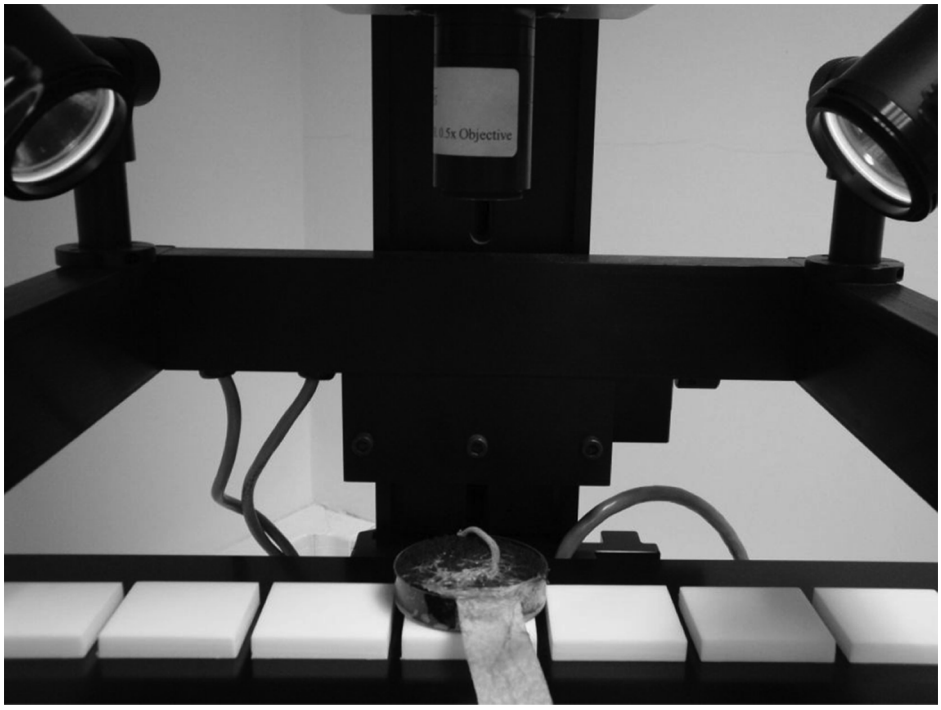

Figure 1. Image acquisition on sugar beet seedling with hyperspectral imaging system.

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

Results

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

Regarding the quantification, four spectral libraries were built by selecting pixels in the four images of four plants: 3,300 pixels corresponding to background (teflon + plastic), 20,200 pixels corresponding to soil support, 7,400 pixels corresponding to roots and 680 pixels corresponding to cyst nematodes. Figure 2 shows the picture of one seedling with root system and cysts.

Figure 2. Real picture of one seedling with root system and cysts.

Figure 3 shows the results of the data treatment applied on the 4 images acquired to cover the full plastic plate of the sugar beet seedlings corresponding to Figure 2. The data treatment applied on each image consisted of four steps (a to d) allowing detection of the cysts. Figure 3(a) shows the image at 1000 nm without preprocessing. Figure 3(b) shows the pixels detected as soil support, roots and cysts after "background *vs.* soil support + root + cyst" equation application on the image of Figure 3(a). Pixels corresponding to background (plastic plate and teflon support) are displayed in white. Figure 3(c) shows the pixels detected as roots and cysts after "soil support *vs.* root + cysts" equation application on the not white pixels of Figure 3(b) and after removing of pixels with intensity > 0.15. Pixels classified as soil support are displayed in white. Figure 3(d) shows the pixels detected as cysts after "root *vs.* cysts" equation application on the not white pixels of Figure 3(c). All the pixels not classified as cysts are displayed in white. The rest includes pixels detected as cysts displayed in black.

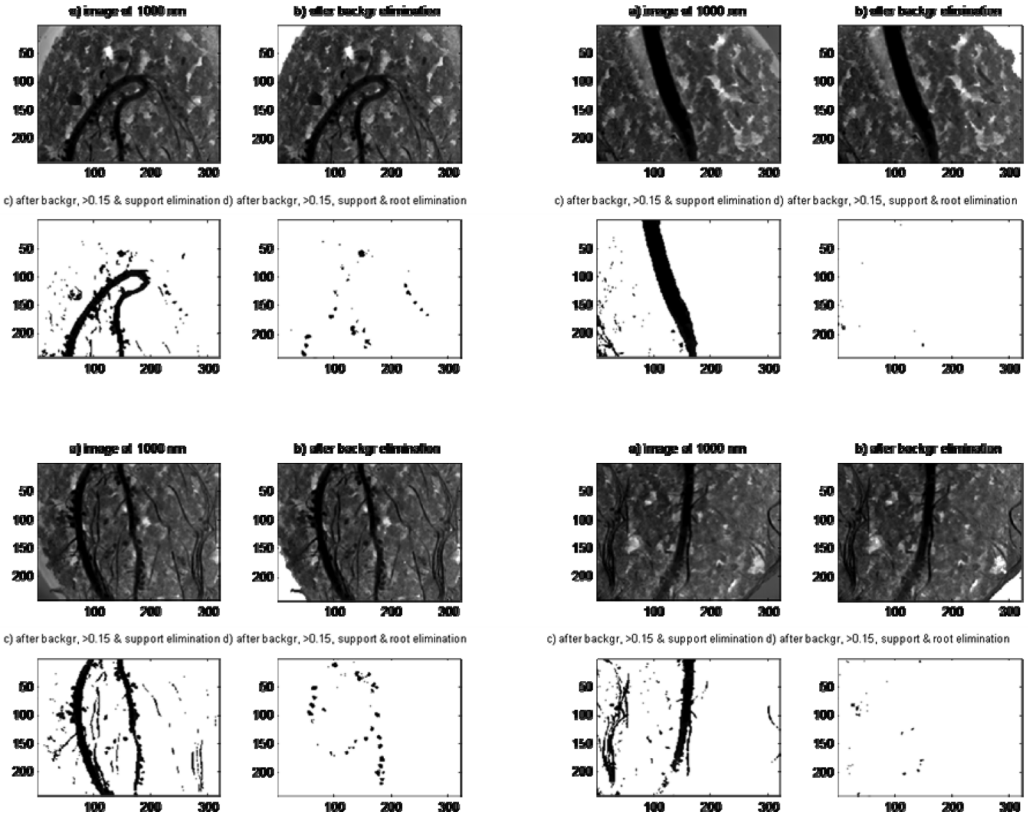

Figure 3. Four acquired images corresponding to Figure 2, with data treatment shown in four steps for each image: (a) image at 1000 nm; (b) pixels detected as soil support, roots and cysts after "background (plastic plate and teflon support) vs. soil support + root + cyst" equation application on the 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 was calculated between the number of cysts counted on the roots under optical microscopy and the number of pixels recognized as nematode cysts by hyperspectral NIR imaging. Figure 4 shows this relationship achieved with the results of the 30 plants. It has to be noted that for the reference value cysts were counted on the total surface of the glass plate, while NIR imaging analysed the total surface by acquisition of 4 images. Values have been corrected to take into account the overlapping between some images.

Conclusion

This study showed the potential of 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

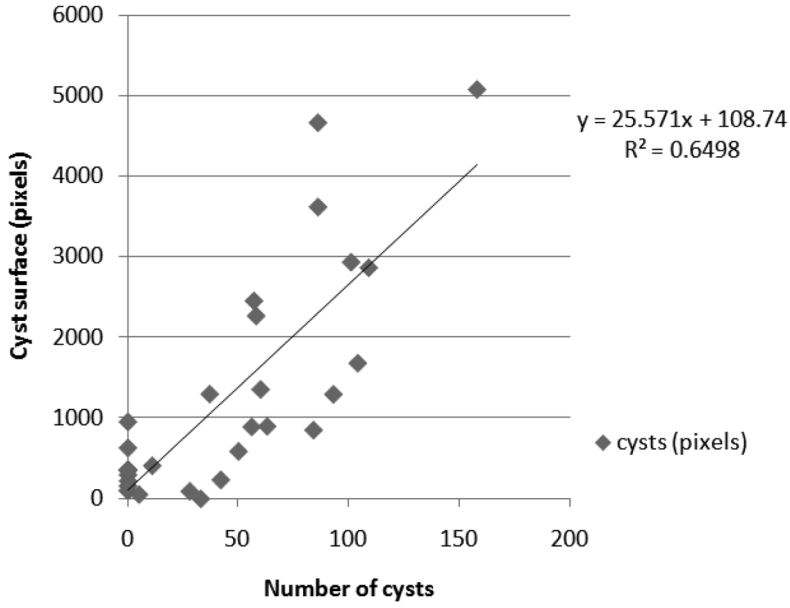

Figure 4. Relation between cyst number (Ref Val) and cyst surface (NIR imaging).

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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