Near infrared hyperspectral imaging as a decision support tool for the sugar beet breeder

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Introduction

The aim of this study is to show that Near Infrared (NIR) hyperspectral imaging spectroscopic technique (Figure 1) combined with chemometrics could be used as a tool to help the sugar beet breeder. Two case studies were conducted in collaboration with SESVANDERHAVE N.V/S.A., to assess the contamination of plants by pathogens (*Heterodera schachtii* and *Cercospora beticola*) in order to select tolerant from susceptible plants.

Figure 1 : NIR line scan (pushbroom) hyperspectral camera equipped with conveyor belt (Burgermetrics) .

Parameters

- Wavelength range: 1000-2500 nm by step of 6 nm
- 1 line = 320 pixels = 320 spectra
- 1 image = 150 000 spectra
- Time of acquisition for 1 image = 1 min

Case study 1: Beet Cysts Nematodes cysts counting on sugar beet roots

Figure 2: BCN cysts at different development stages observed by optical microscopy.

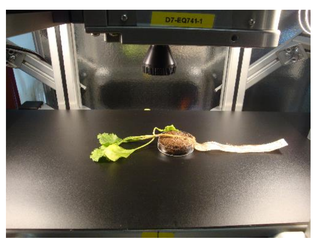

Figure 3: Image acquisition of one plant using a NIR line scan hyperspectral imaging system.

The damage caused by Beet Cyst Nematodes (BCN), *Heterodera schachtii*, on the sugar beet root leads to a yield reduction and is related to the cysts number. The current method to count the number of cysts on roots consists of visual observations using an optical microscope (Figure 2). The objective of the study is to discriminate between cyst, root and soil support as well as to quantify the BCN cysts presence using the NIR line scan hyperspectral imaging system.

For this experiment, 10 tolerant and 10 susceptible to nematodes sugar beet plants were grown in plastic boxes. The plants were inoculated with suspension of Heterodera schachtii juveniles. Then, boxes were analysed (Figure 3).

For the counting of cyst, four spectral libraries (cyst, root, soil support and background including plastic box) have been built by selecting pixels in the images of 4 plants. Those libraries were used for the building of 3 discrimination equations : "background *vs.* soil support + root + cyst", "soil support *vs.* root + cyst" and "root *vs.* cyst". The equations have been applied successively to all the pixels in the images of the 20 plants in order to isolate the pixels detected as cysts. Finally, groups of pixels were defined to quantify the number of BCN cysts in each box (Figure 4).

The results show a discrimination between tolerant and susceptible plants, even if the NIR hyperspectral imaging method underestimates the number of BCN cysts on some plants (Figure 5).

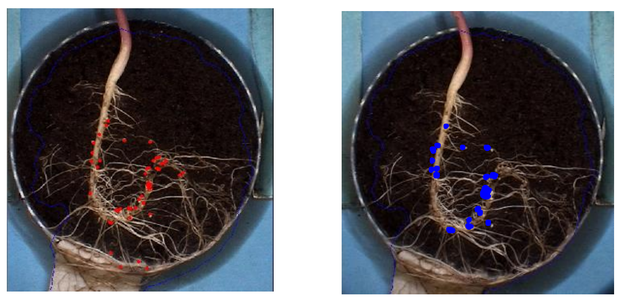

Figure 4: Number of BCN cysts counted by optical microscopy in red (49 cysts) and predicted by NIR hyperspectral imaging in blue (31 cysts) on a tolerant plant.

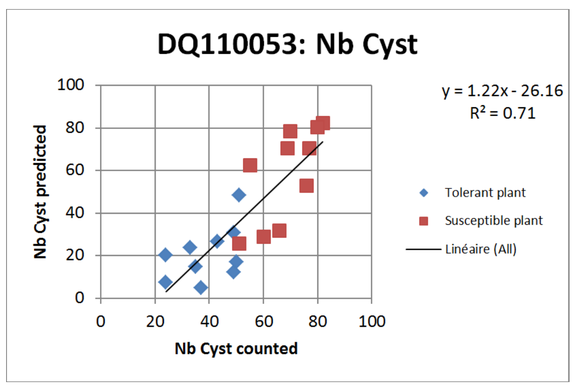

Figure 5: Number of BCN cysts counted by optical microscopy and predicted by NIR hyperspectral imaging on tolerant and susceptible plants.

Case study 2: Assessment of Cercospora Leaf Spots development on sugar beet leaves

Figure 6: CLS susceptible plant at a early stage of the plant development.

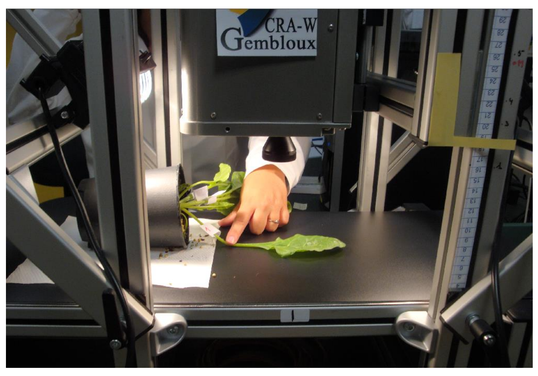

Figure 7: Image acquisition of sugar beet leaves using a NIR line scan hyperspectral imaging system.

The damage caused by Cercospora Leaf Spot (CLS), *Cercospora beticola*, on sugar beet leads to a yield reduction. The current method to assess the necrosis level on leaves consists of visual observations. The objective of the study is to discriminate between cercospora leaf spots and healthy leaf as well as to quantify the disease area using the NIR line scan hyperspectral imaging system.

For this experiment, 4 tolerant and 4 susceptible to CLS sugar beet plants were grown in plastic pots in a greenhouse (Figure 6). The plants were infected by spraying with *Cercospora beticola* spores suspension. Then, some leaves were analysed during 8 days (Figure 7).

For the necrotic area assessment, discrimination model was built using two spectral libraries corresponding to the cercospora leaf spots and the healthy leaves. This model was applied to all the individual pixels in the images of the leaves in order to isolate and quantify the cercospora leaf spots (Figure 8). As it can be observed, the cercospora leaf spots became detected by the discrimination models only when they are visually observed on the leaves. The results show clear differences between tolerant and susceptible plants (Figure 9). The disease development is slower on tolerant plants and the necrosis covers less than 20% of the leaf area. For the susceptible plants, the infection is faster and 100% of the leaf area can be infected on the same period.

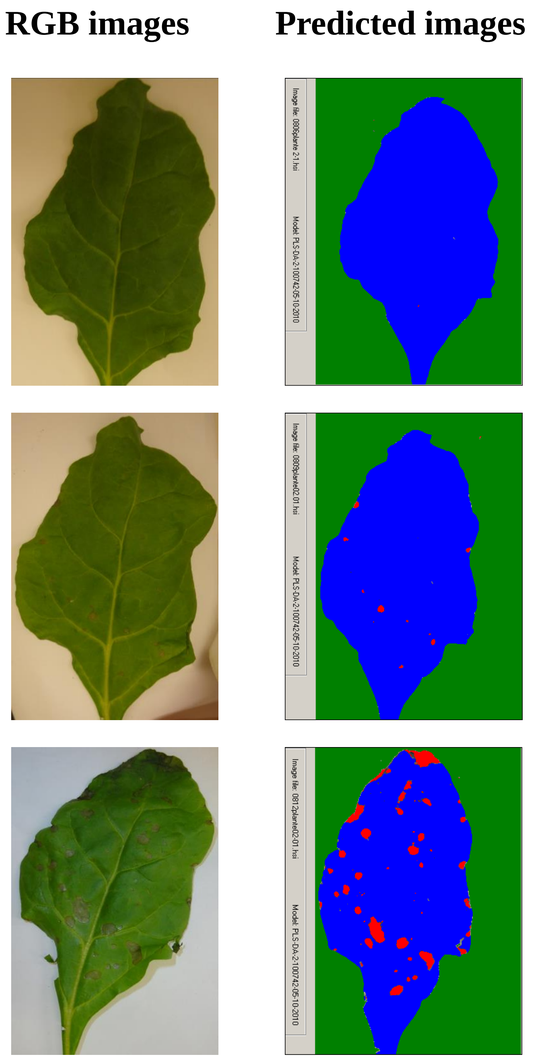

Figure 8: RGB picture, classification results, showing the cercospora leaf spots in red observed at 3 dates (3 days before the first symptoms, the day of the first symptoms and 3 days after the first symptoms) on a leaf from a tolerant plant inoculated by spray with CLS and analysed using the NIR line scan hyperspectral imaging system.

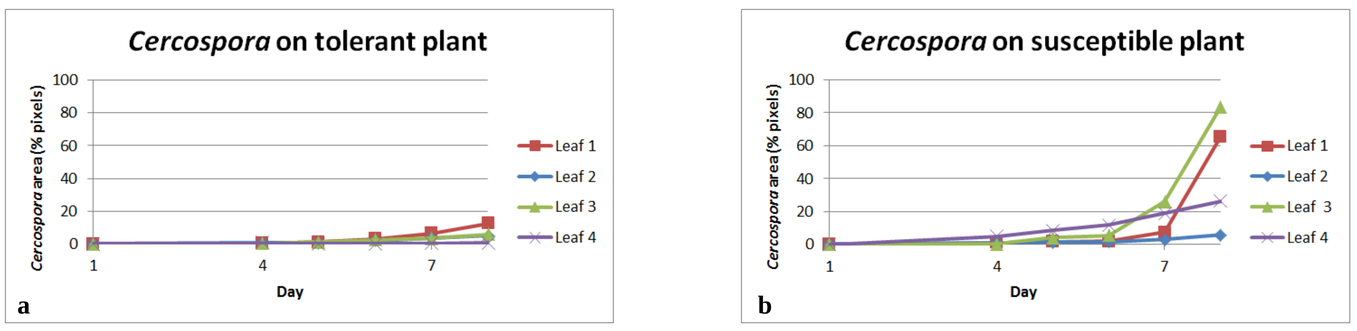

Figure 9: CLS development on leaves from tolerant (a) and susceptible (b) plants.

Conclusion

These 2 case studies have shown the potential of the NIR hyperspectral imaging methods combined with chemometric tools as a decision support tool for the plant breeding. It allows to select tolerant from susceptible plants by assessing either the number of BCN cysts in roots or the leaf area infected with the CLS.

Reference

Fernández Pierna J.A. , Vermeulen P. , Amand O. , Tossens A. , Dardenne P. & Baeten V. (2012). NIR hyperspectral imaging spectroscopy and chemometrics for the detection of undesirable substances in food and feed. Chemometrics and Intelligent Laboratory Systems. 117 : 233-239.

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