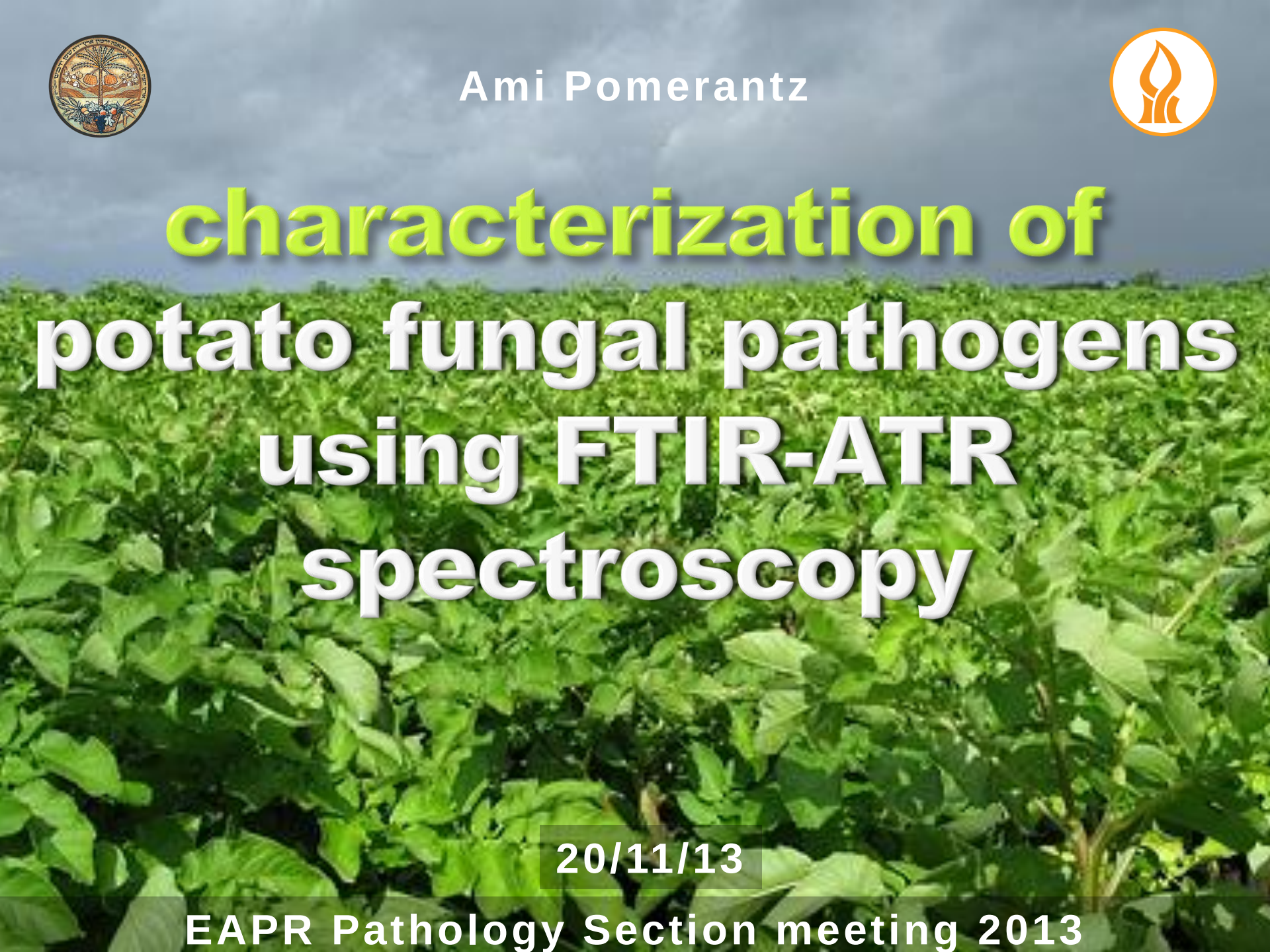

Ami Pomerantz
characterization of
potato fungal pathogens
using FTIR-ATR
spectroscopy
20/11/13
EAPR Pathology Section meeting 2013

Overview

- **Commercial Potato crops are susceptible to a large variety of fungal pathogens.**
- **Each year fungal pathogens cause great crop damages worldwide, resulting in losses of billions of dollars and euro.**

Research approach

- **Spectral analysis may help evaluate the association of different fungus isolates to various properties.**
- **We aim to determine different characteristics of fungal pathogens using their absorption spectrum.**

Research goals

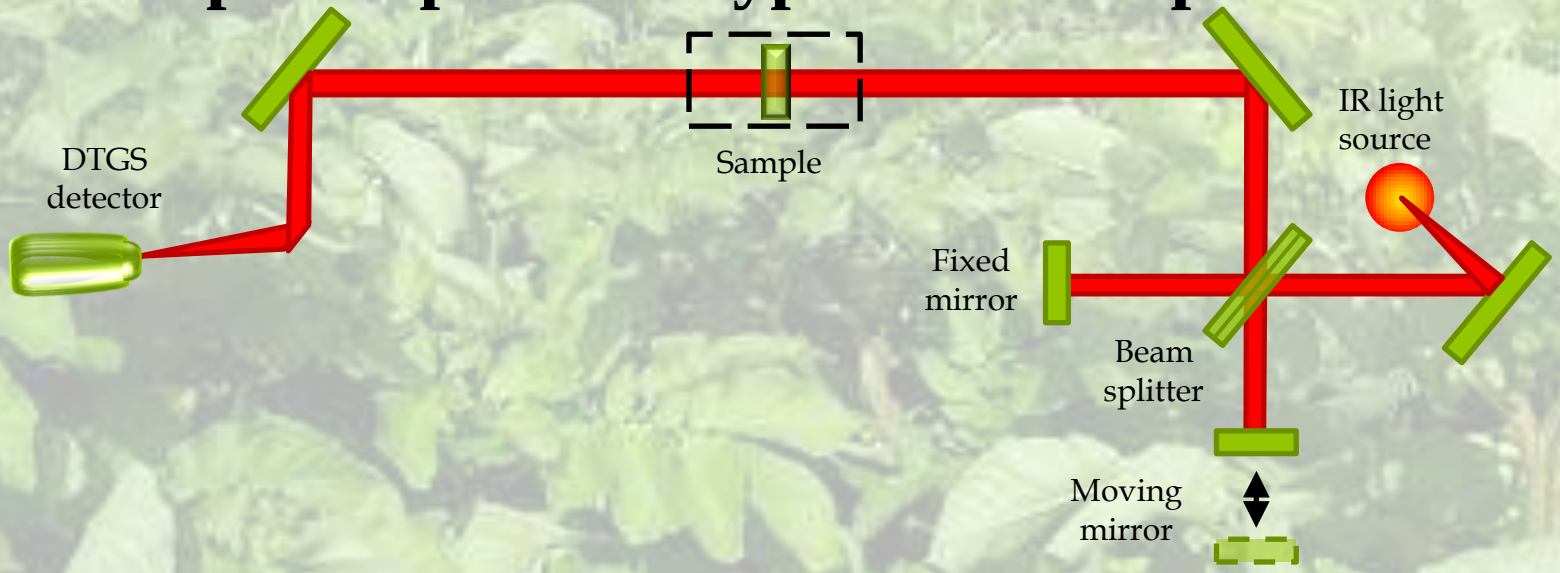
- **Classification of fungi isolates according to their characteristics will allow better treatment.**
- **Developing the Fourier Transform Infra-Red spectroscopy (FTIR) as a quicker and cheaper method (comparing to the biochemical and molecular methods used today).**

FTIR Spectroscopy

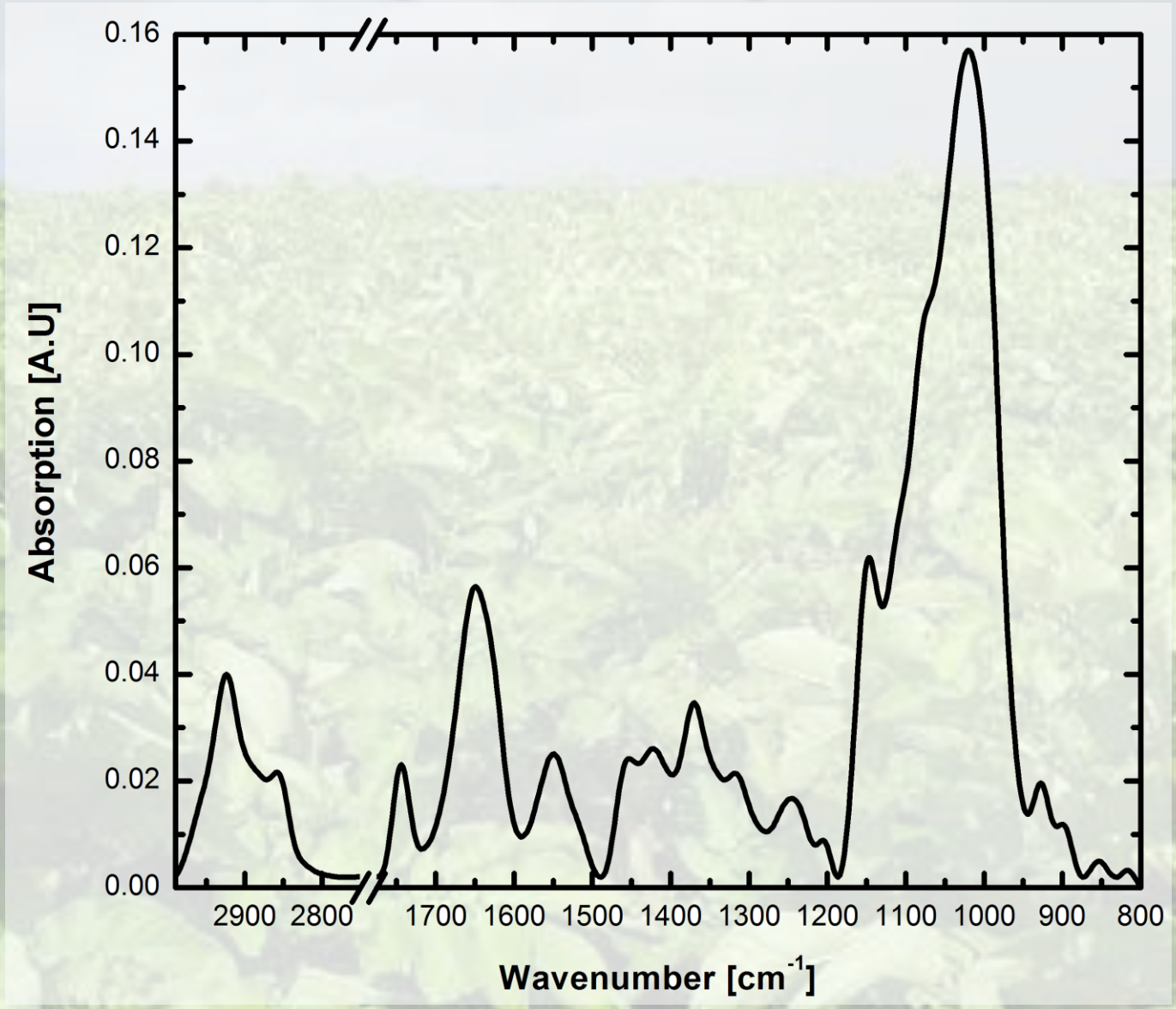
- **Each matter absorbs part of the electromagnetic waves in a specific way. The absorption depends on the materials functional groups and its structure and can be considered as a “finger print” of the matter.**
- **Biologic materials absorb infra-red (IR) radiation through changes in the vibrational and rotational levels of the molecules in the matter.**

FTIR spectrometer

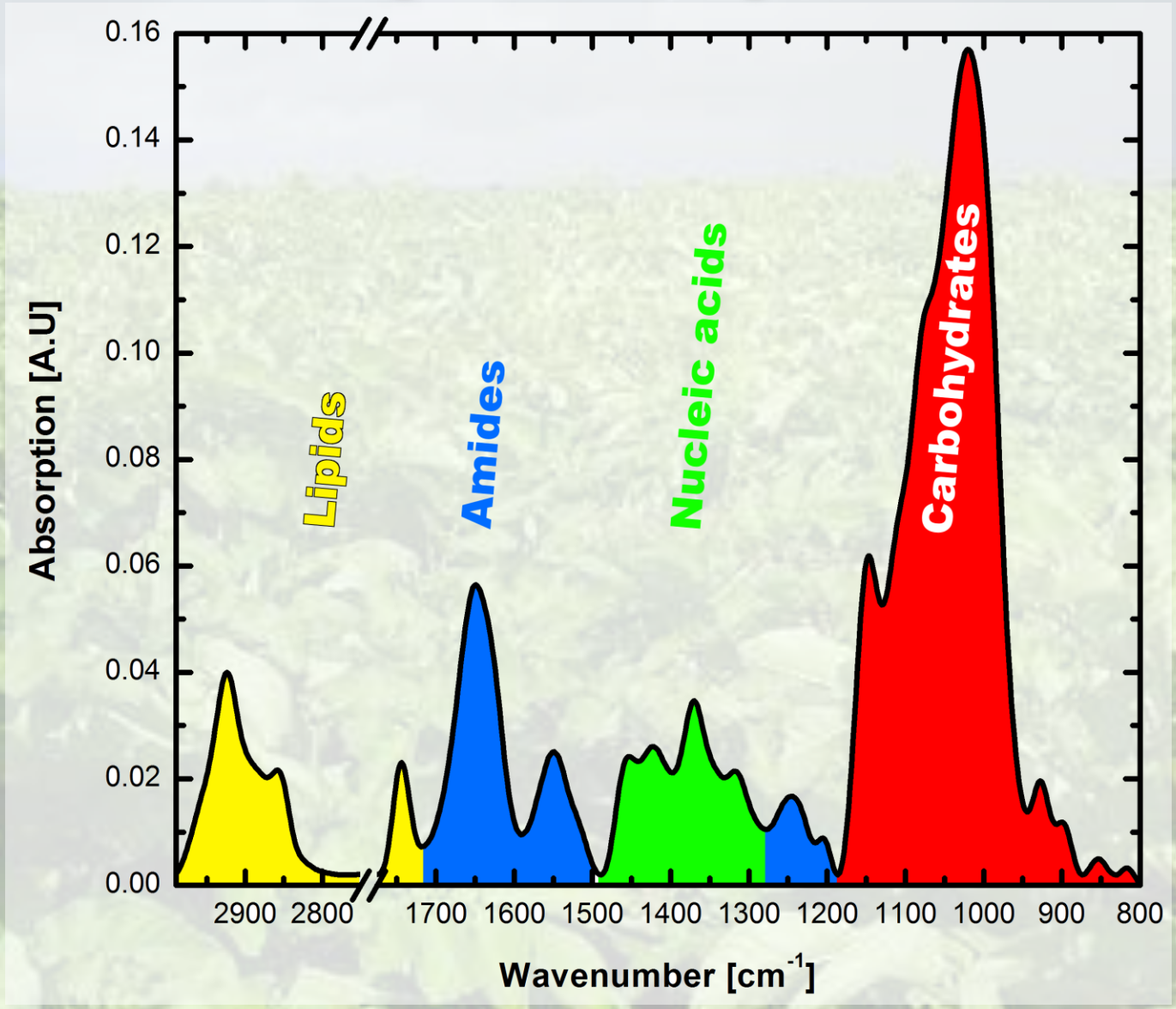
- **The FTIR spectrometer transmits a broad ray of IR light through a specimen and measures the amount of light absorbed in each frequency. Applying Fourier transform on the measured data provides an absorption spectrum typical to the specimen.**



Absorption spectrum

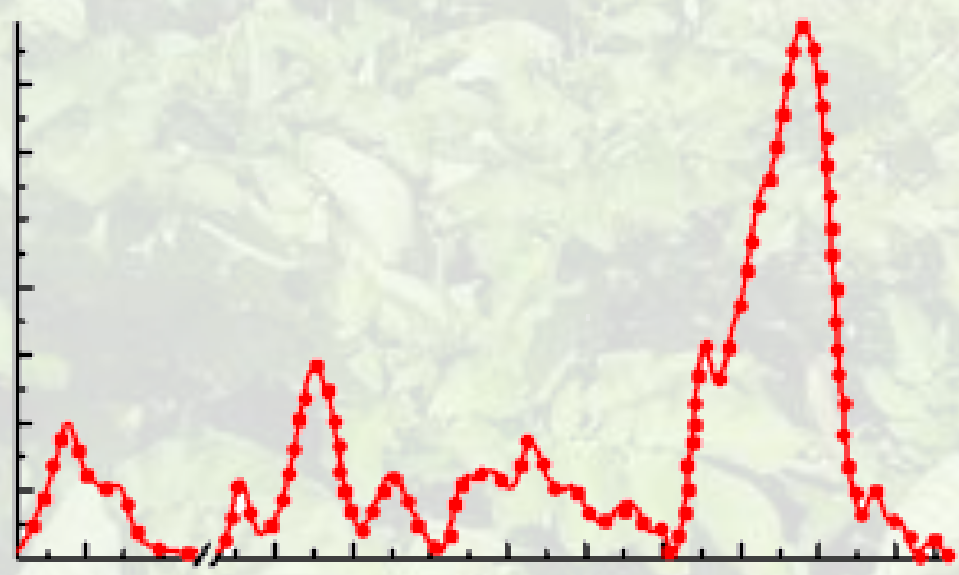


Absorption spectrum



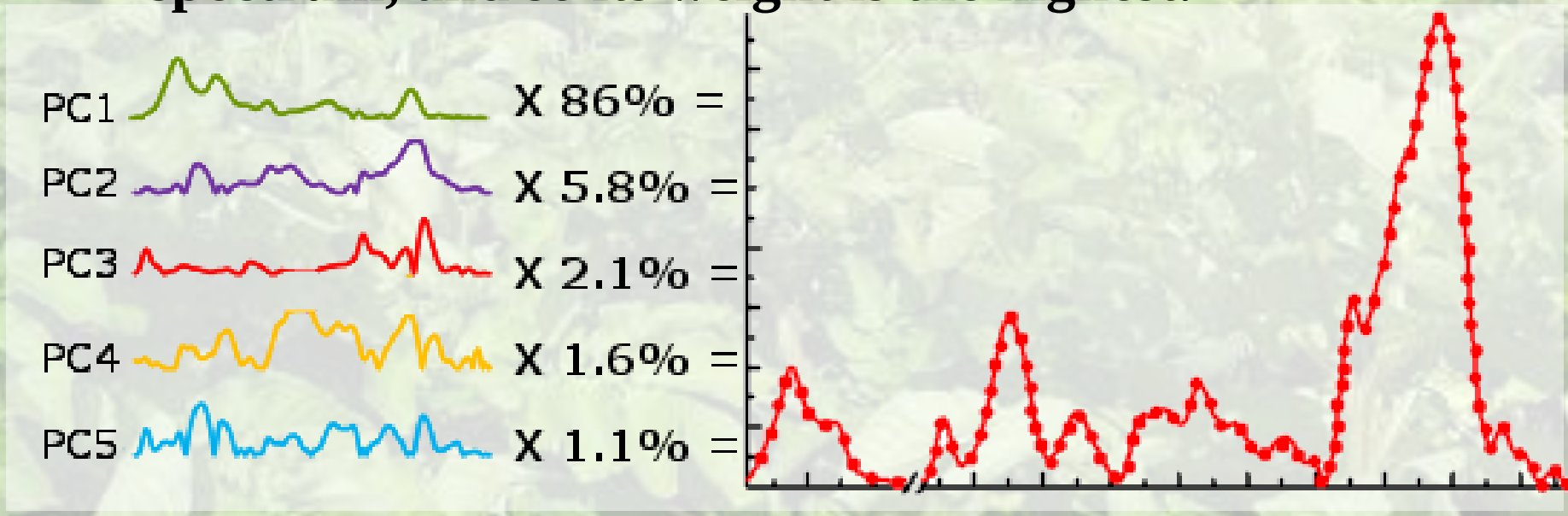
Principal component analysis

- **Original spectrum is composed of about 500 points (dimensions).**
- **To decrease the magnitude of the problem we define a small number of Principal Components (PC's).**



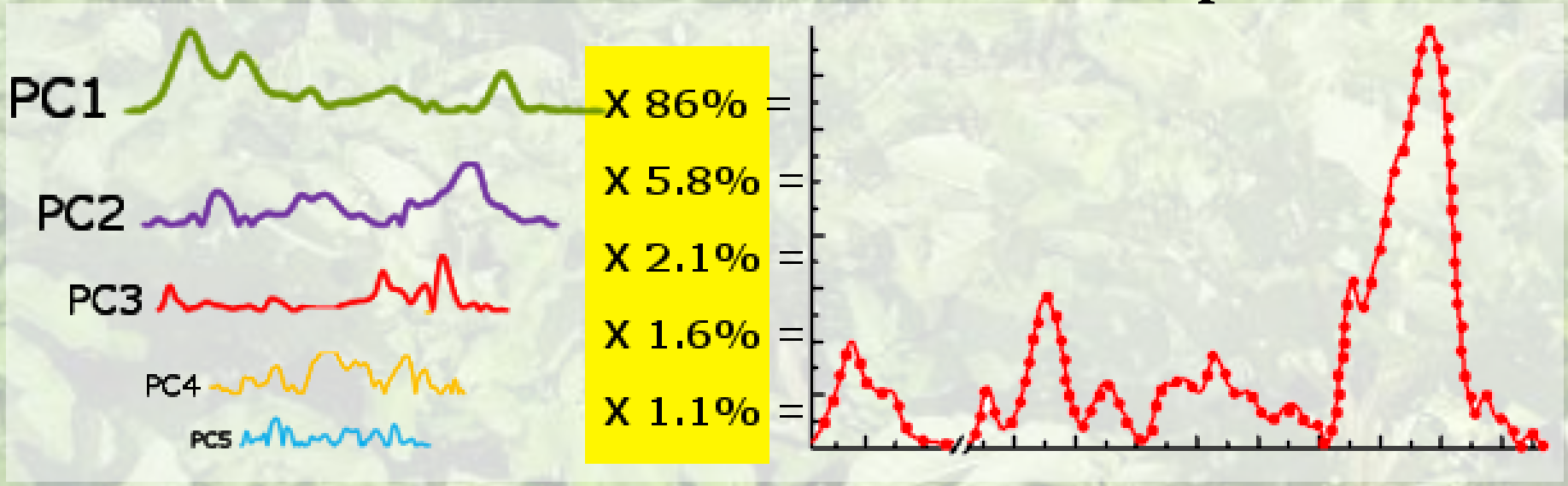
Principal component analysis

- In order to restore the original spectrum each PC must be multiplied with a certain weight (coefficient) and be added to the rest of the PC's.
- PC1 has the major characteristics of the original spectrum, and so its weight is the highest.

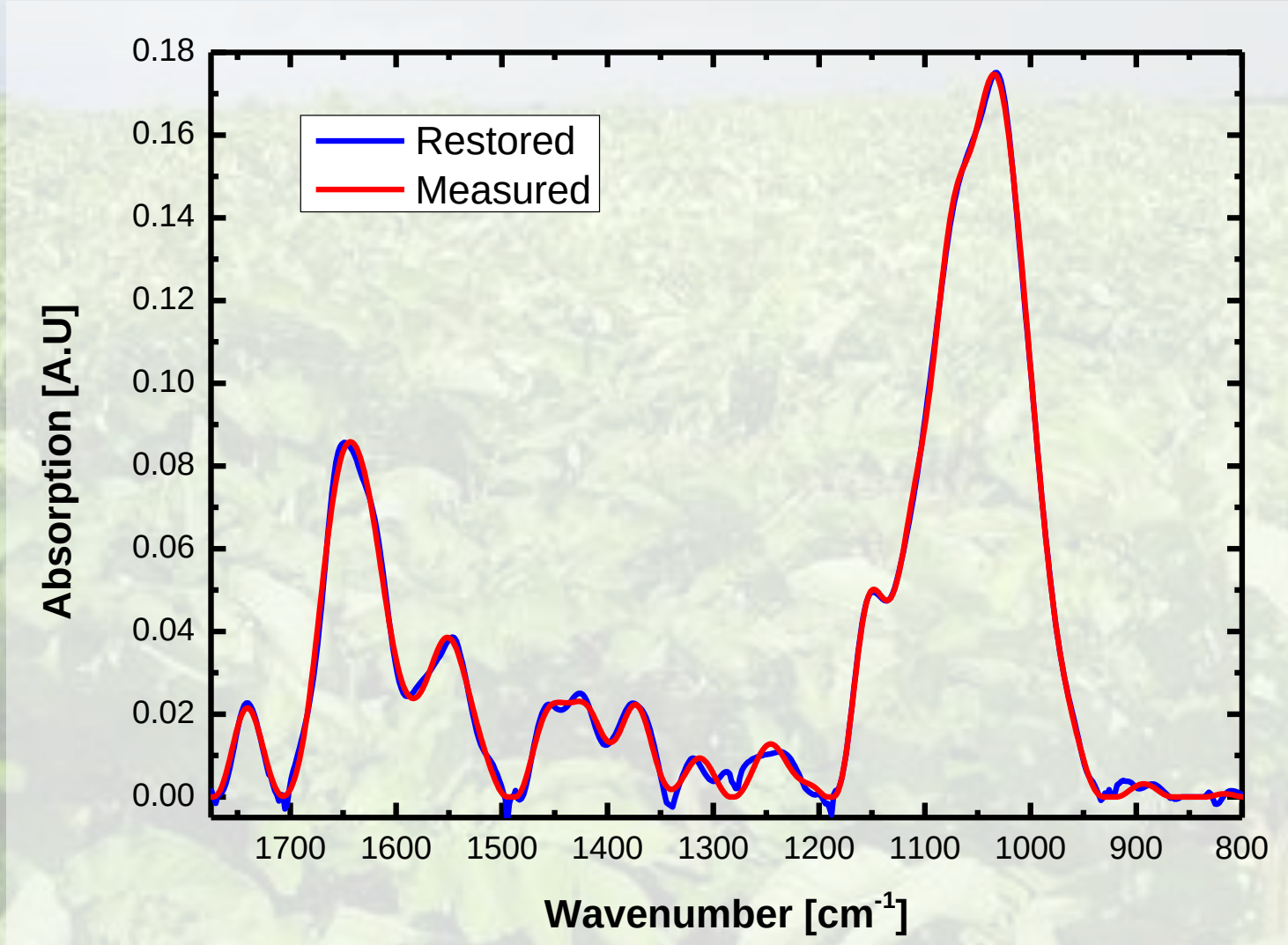


Principal component analysis

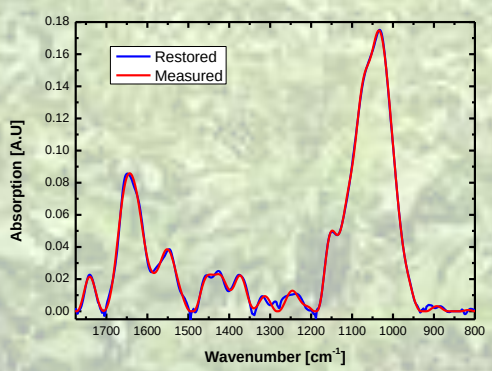
- These weights or coefficients, are now the describers of the spectrum.
- Instead of the 500 dimension spectrum we started with, we now have a 10-50 dimension spectrum.

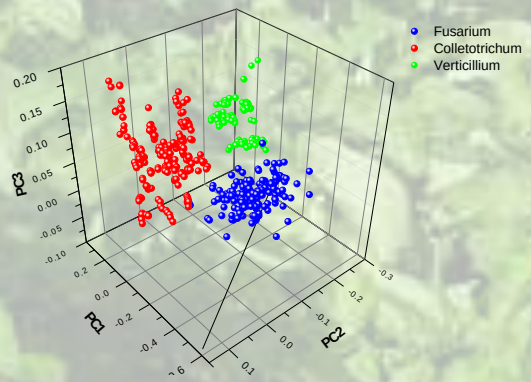


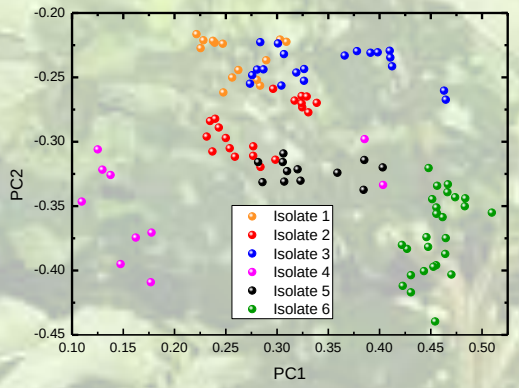
Spectrum PCA restoration



Results


Restored
Measured
Absorption [A.U]
Wavenumber [cm-1]


Fusarium
Colletotrichum
Verticillium
PC1
PC2
PC3


Isolate 1
Isolate 2
Isolate 3
Isolate 4
Isolate 5
Isolate 6
PC1
PC2

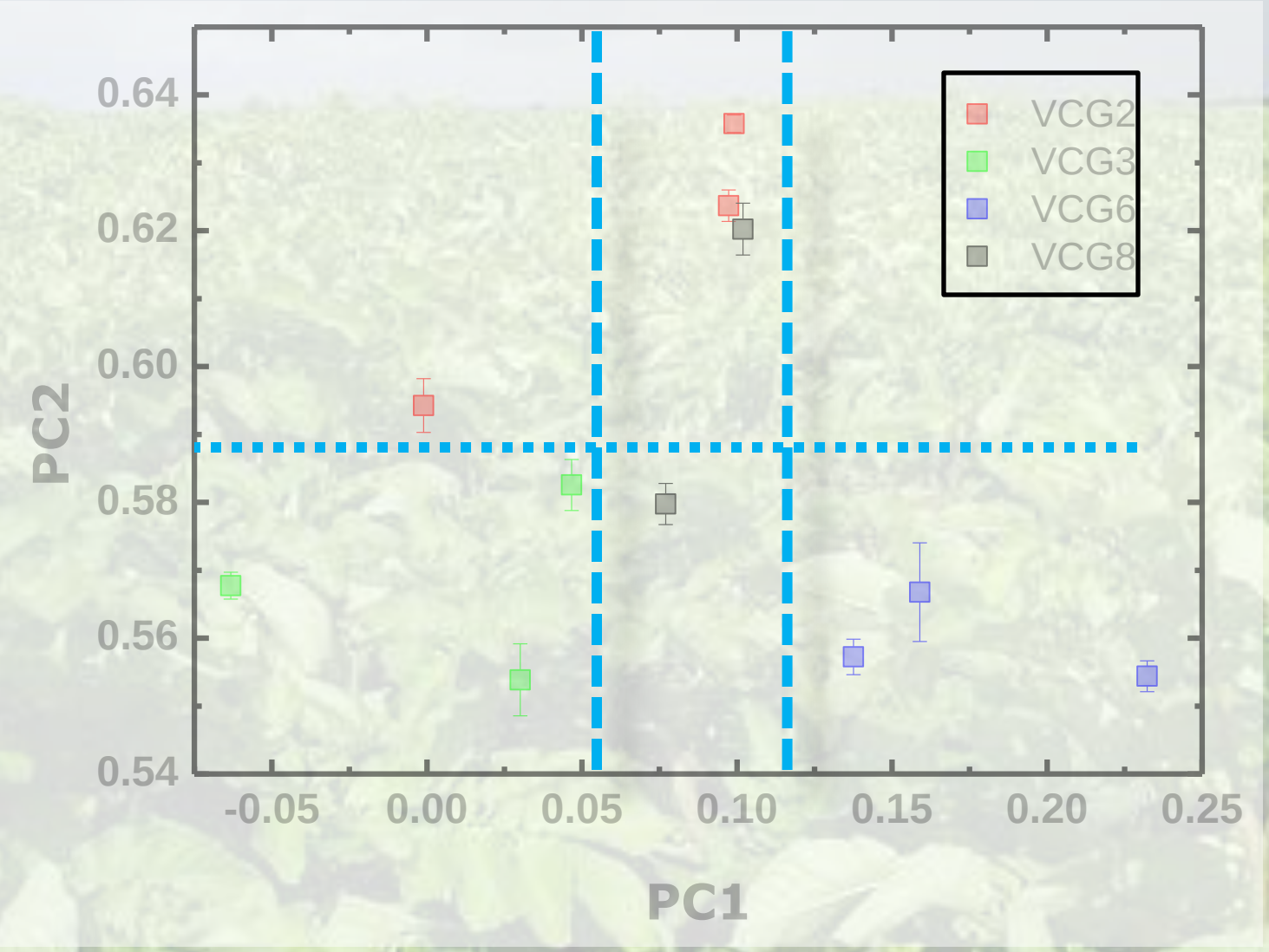
VCG classification

- First characteristic we examined is classification of isolates into Vegetative Compatibility Groups or VCGs.
- VCGs are isolate groups which have a common genetic similarity. These groups may make sub-population of common physiologic and pathogenic characteristics.
- This information is helpful in assessing the pathogenicity level of a certain isolate found in the field.

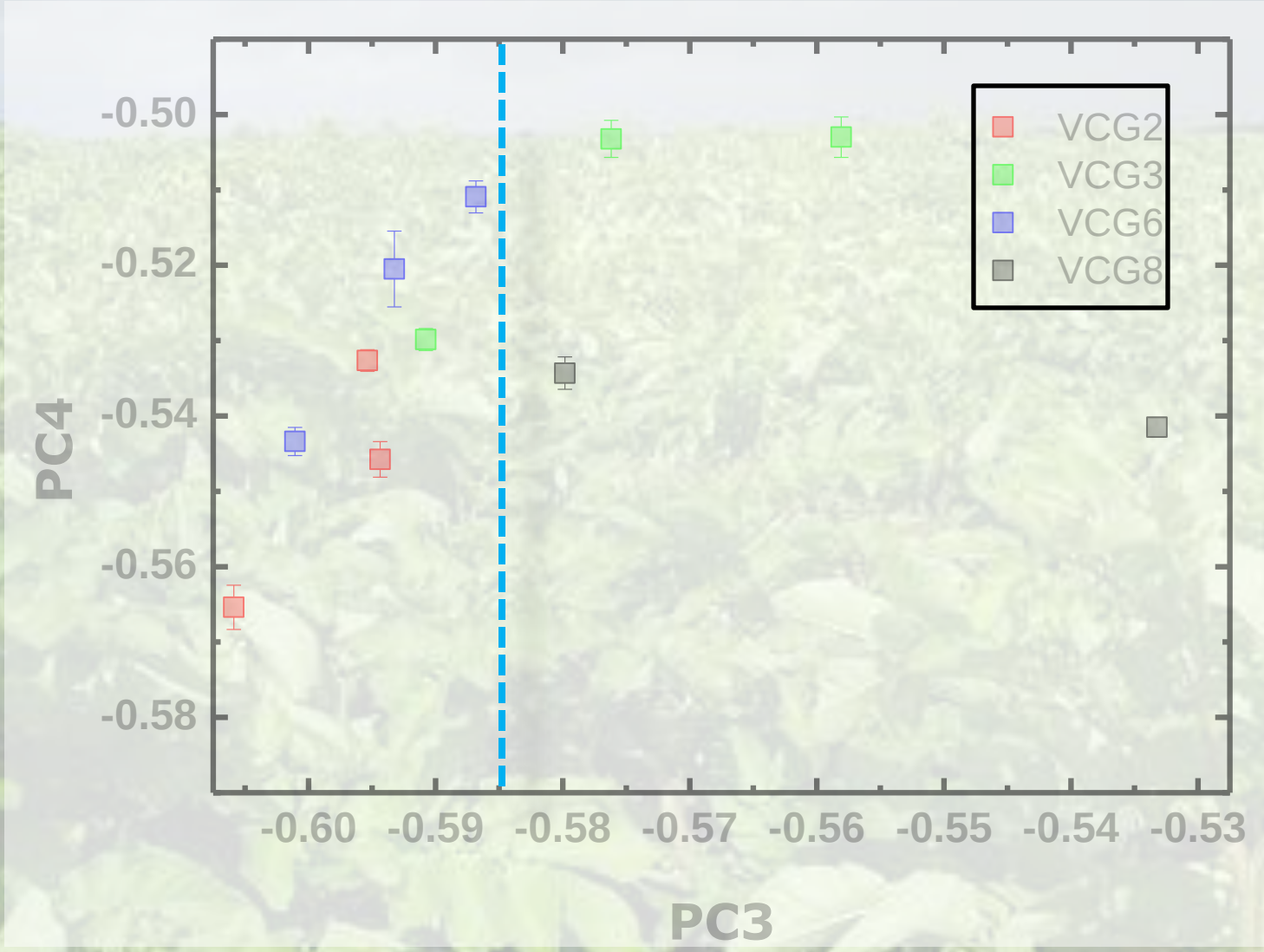
Fungi examined

- ***Colletotrichum coccodes*** **has been reported worldwide on many different hosts, primarily Solanaceae and Cucurbitaceae. It is the causal agent of black dot disease on potatoes.**
- ***Verticillium dahliae*** **which is the causal agent of Verticillium wilt in over 400 plant species including herbaceous, perennials, and woody species.**

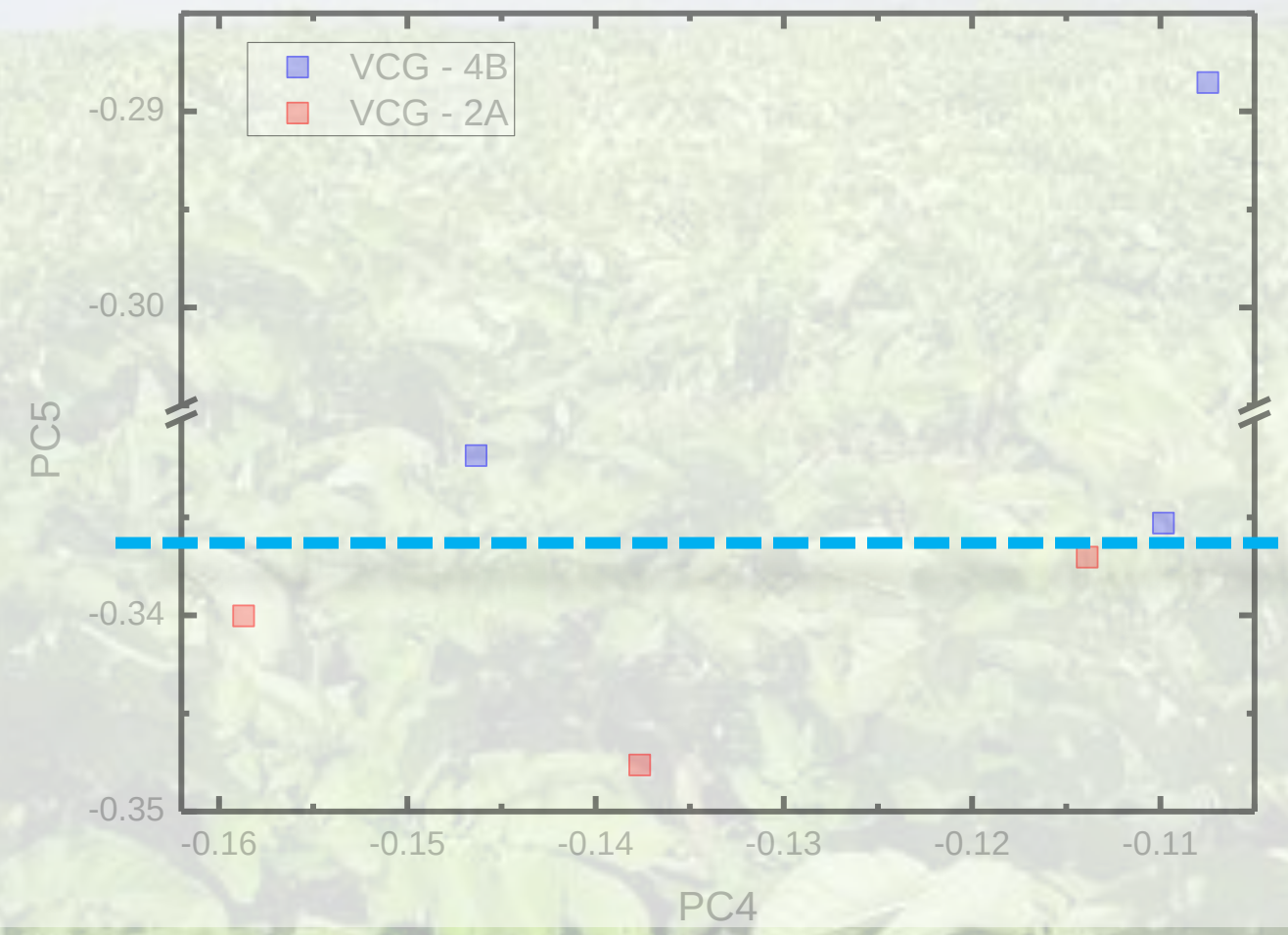
VCG classification – *C. coccodes*



VCG classification - *C. coccodes*



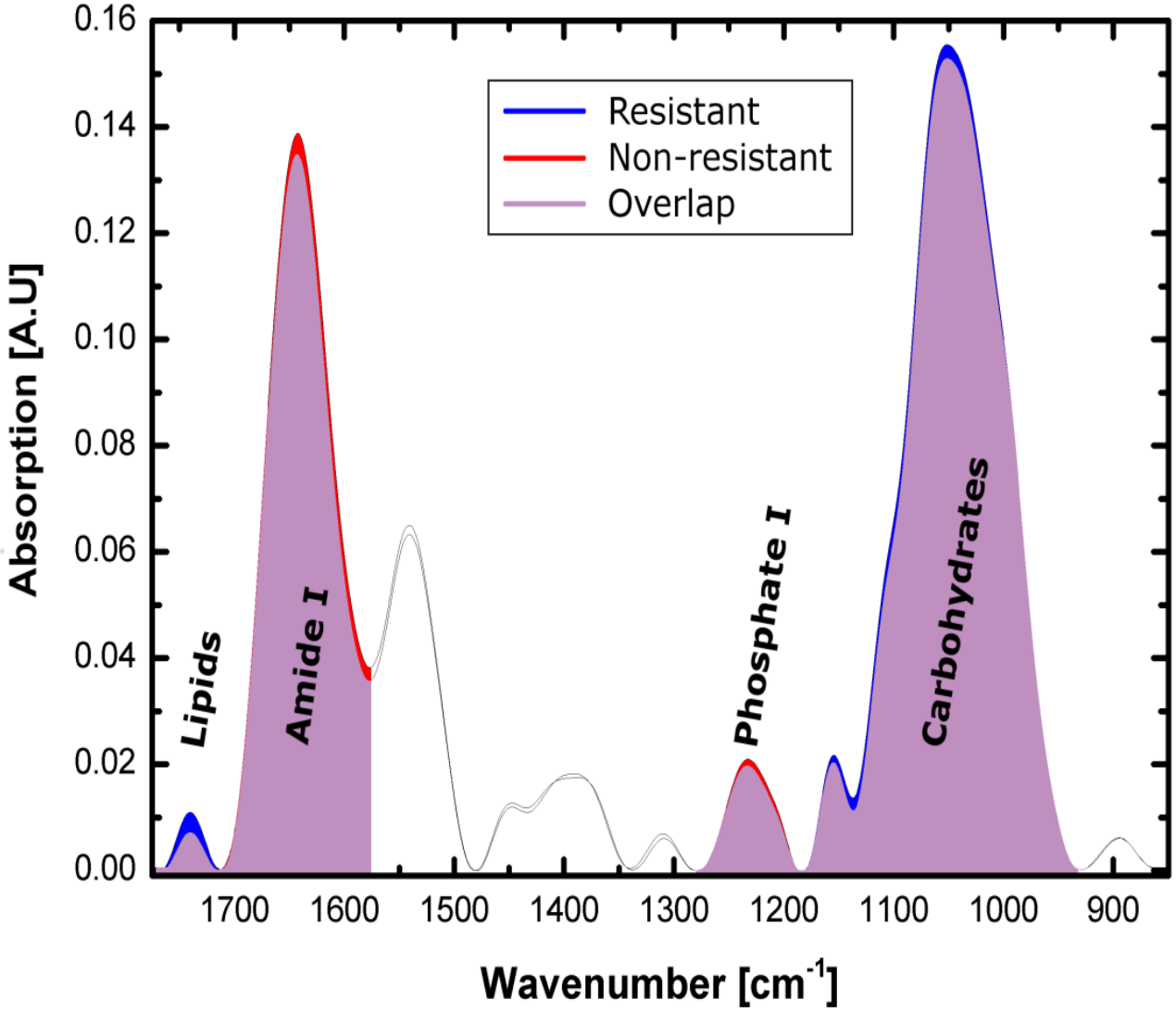
VCG classification – V. dahliae



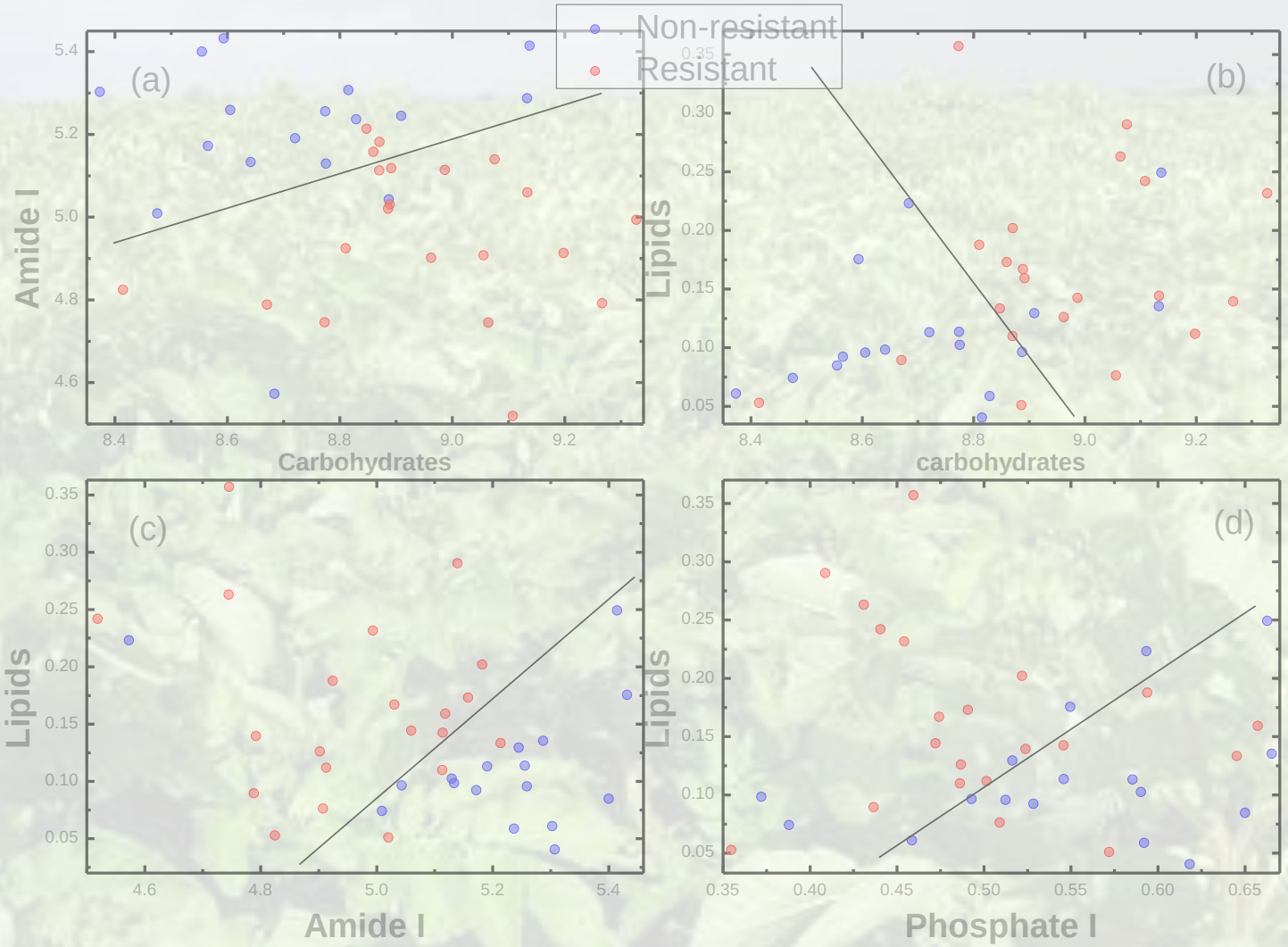
Fungicide resistance

- ***Phytophthora infestans*, is the causal agent of late blight in potato. This pathogen has devastated the potato crops in Ireland 150 years ago and is still causing great losses worldwide.**
- **Although fungicides controlling *P. infestans* are used successfully for almost 100 years, some strains have developed resistance to most common fungicides.**
- **Identification and characterization of these resistant strains, is required for better control of the disease.**

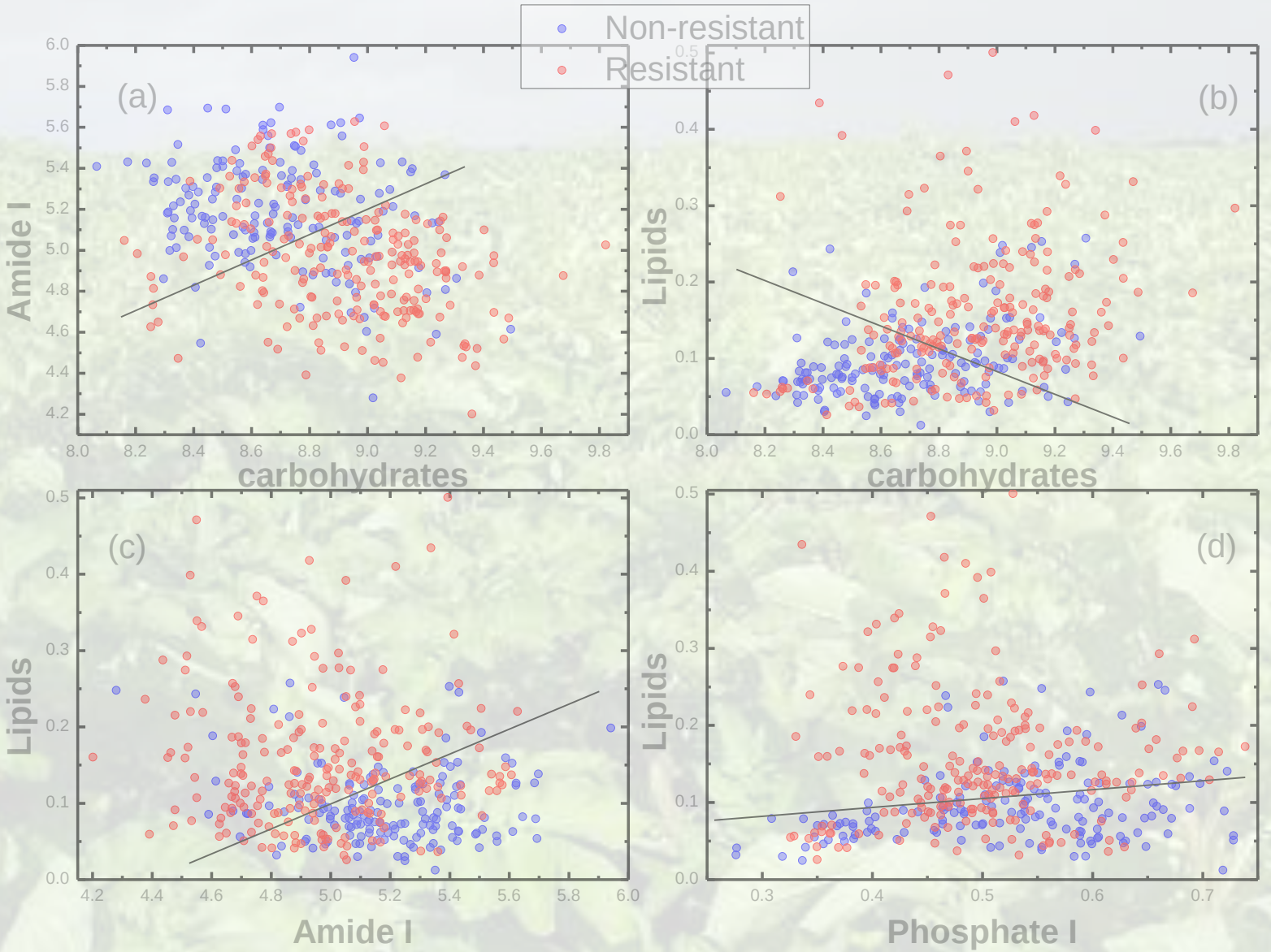
Fungicide resistance - Bands



Fungicide resistance - Average



Fungicide resistance - Actual



Fungicide resistance

Pairs	Resistant		Non-Resistant		sensitivity	specificity
	True Positive	False Negative	True Negative	False Positive		
Amid I - Lipids	163	58	126	37	**0.74**	**0.77**
Carbohydrates - Amid I	156	65	123	40	**0.71**	**0.75**
Carbohydrates - Lipids	161	60	120	43	**0.73**	**0.74**
Phosphate I - Lipids	153	68	122	41	**0.69**	**0.75**

Fungicide resistance

- **Applying Linear Discriminant Analysis (LDA) on the PCA manipulated data allows better distinction between the two groups of isolates.**
- **Using LDA has resulted in increasing the sensitivity to 81% and the specificity to 83%.**

Conclusions

- **It is possible to determine characteristics of various fungi isolates, using FTIR spectroscopy.**
- **This can be done almost “at a push of a button”.**
- **Today we continue the research and aim to look into other fungi characteristics.**
- **We aim to diagnose fungi *in situ* in the field directly on the plant, using a portable FTIR probe.**
- **We believe that FTIR technology could be used as a diagnostic tool for recognition and classification of phytopathogenic fungi.**

Acknowledgments

Prof. Mahmoud Huleihel
Prof. Shaul Mordechai
Prof. Yigal Cohen
Dr. Leah Tsror
Dr. Elad Shufen
Dr. Ahmad Salman

Thank you
for listening
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Questions
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