Evaluation of pesticide coating efficiency on cereal seeds by combining NIR Hyperspectral Imaging and Chemometrics

Ph. Vermeulen[1]*, P. Flemal[2], O. Pigeon[1], P. De Vos[1], J.A. Fernández Pierna[1], P. Dardenne[1] and V. Baeten[1]
[1]Walloon Agricultural Research Centre (CRA-W), Belgium; [2]Catholic University of Louvain (UCL), Belgium
*Contact person: p.vermeulen@cra.wallonie.be; FoodFeedQuality@cra.wallonie.be

Introduction

A good pesticide treatment requires that the active substances are homogeneously distributed on seeds coming from the same batch. Indeed, lower doses may lead to insufficient protection for seeds while an overdose can increase the risk of phytotoxicity and establish an economic loss for the seed producer. The objective of this study is to assess the quality of the treatment by near infrared hyperspectral imaging (Burgermetrics). In total, three cereals species (wheat, barley and spelt) and three groups of pesticides (Prochloraz/triticonazole, Prothioconazole and Fludioxonil) have been studied. Five criteria were assessed: i) identification of seed species, ii) identification of the type of pesticide applied on seeds, iii) the uniformity between seed batches based on the average dose of pesticides, iv) the consistency of treatment between seeds from the same batch and v) the homogeneity of the pesticide coating at the seed level. The results are compared to the results obtained with classical near infrared spectroscopy (Bruker MPA) and chromatography (Waters UPLC).

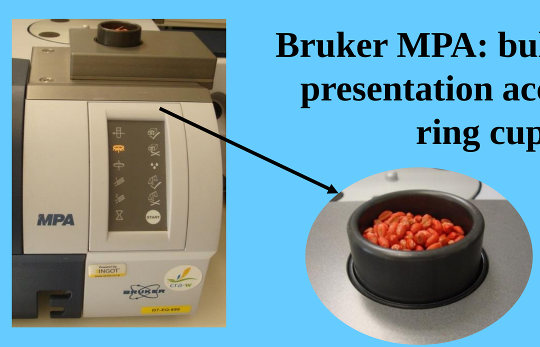
Reference method

Chromatographic methods are the reference methods to assess seed quality. These methods are selective, sensitive, accurate and repeatable, but also time consuming, expensive, destructive and require a substantial amount of solvents.

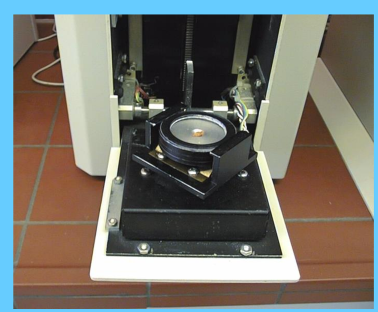
Waters Ultra performance liquid chromatography (UPLC)

NIR spectroscopy

Alternative methods, rapid, non destructive, requiring no sample preparation and no solvent are needed. Near infrared (NIR) spectroscopy seems to be an interesting technique for the determination of the quality of seed treatment. Previous studies have proved that NIR used with specific seed by seed sample presentation can determine quantitatively the active substance concentration on a treated sample and permits as well to evaluate the distribution of the treatment between seeds.



Bruker MPA: bulk sample presentation accessory: ring cup



Foss NIRSystems 6500 : single seed presentation: rotating aluminium cup with one seed in the hole in the middle

Pigeon O. (2003). Study of the quality of seed treatments with plant protection products using near infrared spectroscopy (PhD thesis). Faculté Universitaire des Sciences Agronomiques de Gembloux, Belgium, 194 p.

NIR hyperspectral imaging

The method developed in this study consists to assess the quality of the seed treatment by near infrared hyperspectral imaging (NIR HIS), by analyzing several seeds simultaneously.

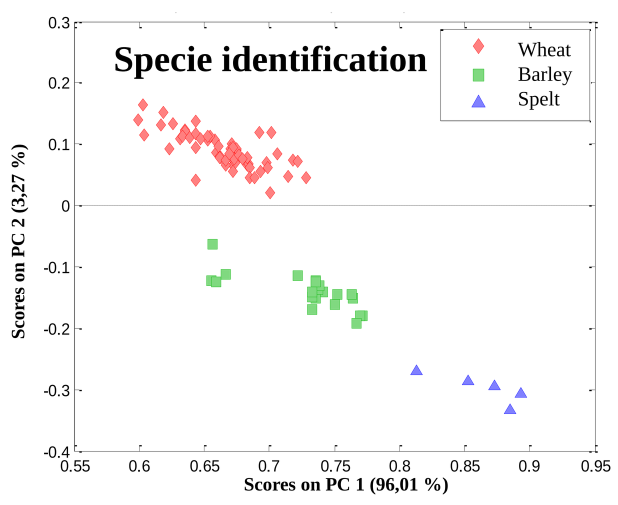
Burgermetrics NIR HIS: multi seeds presentation on conveyor belt

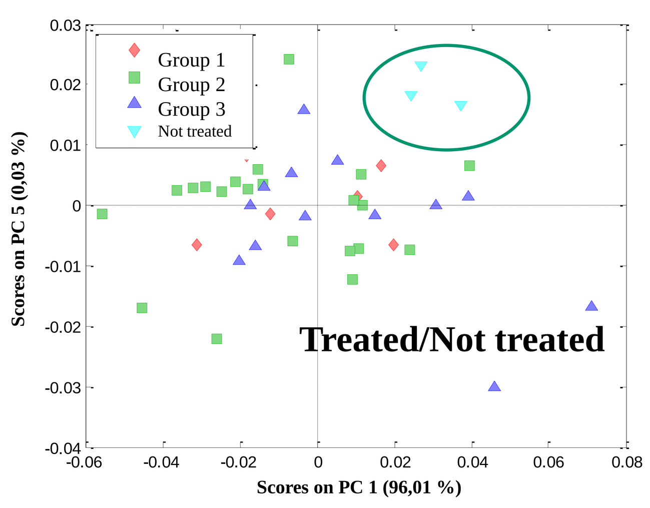
Flemal, P. (2015). Développement de méthodes en spectroscopie proche infrarouge pour l'étude de la qualité du traitement des semences (TFE). Université catholique de Louvain La Neuve, Belgium, 91 p.

Chemometrics

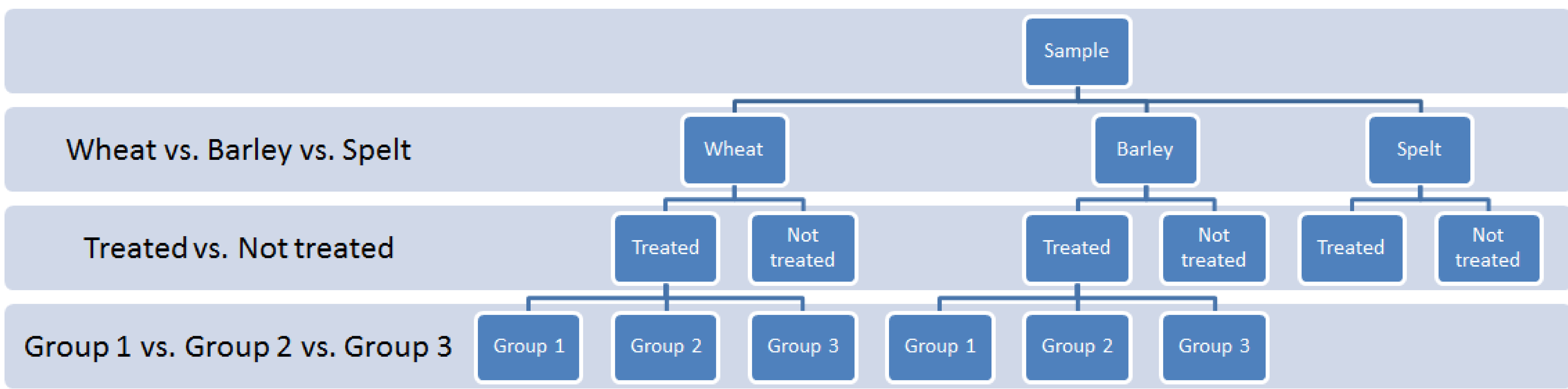
Various chemometric methods were applied in order to extract the maximum amount of information from the spectral data. As a first step, the unsupervised principal component analysis (PCA) was applied to the data to get some indication about the natural grouping of the seeds. Based on this information, a dichotomist classification tree was built where each node of the tree corresponded to a PLS discrimination model (PLS-DA) for a specific group of seeds. In addition, calibration models were built using the partial least squares regression (PLS) in order to assess the quantity of pesticide applied on the seeds.

Exploratory analysis : PCA





Dichotomist PLS-DA classification tree



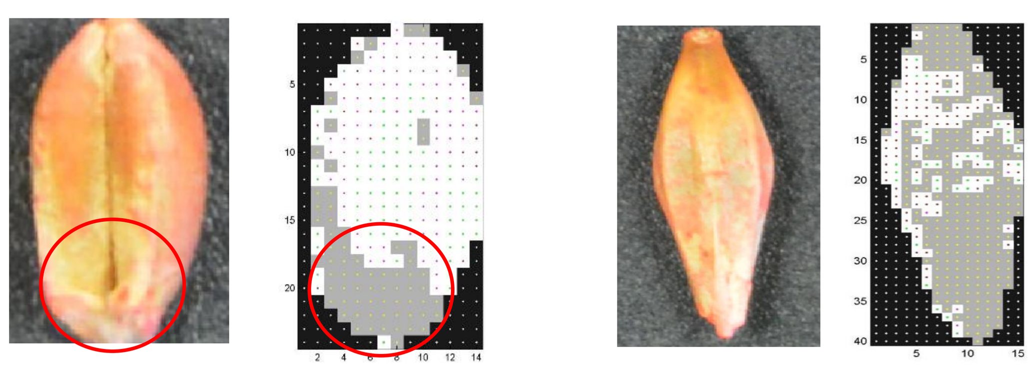
Homogeneity inter seeds batches: Bulk analysis

Models	Classes	Calibration			Cross-validation		Validation		
		n_c	TP	FP	TP	FP	n_v	TP	FP
NIR spectroscopy									
Wheat									
Treated vs. Not Treated	Treated	48	100	0	100	0	8	100	NaN
	Not Treated	3	100	0	100	0	0	NaN	0
Barley									
Treated vs. Not Treated	Treated	14	100	0	100	0	4	100	NaN
	Not Treated	3	100	0	100	0	0	NaN	0
Spelt									
Treated vs. Not Treated	Treated	2	100	0	100	0	1	100	NaN
	Not Treated	2	100	0	100	0	0	NaN	0
NIR hyperspectral imaging									
Wheat									
Treated vs. Not Treated	Treated	48	97	4	96	4	8	97	NaN
	Not Treated	3	96	3	96	4	0	NaN	3
Barley									
Treated vs. Not Treated	Treated	14	89	17	87	17	4	87	NaN
	Not Treated	3	83	11	83	13	0	NaN	12
Spelt									
Treated vs. Not Treated	Treated	2	100	7	94	14	1	100	NaN
	Not Treated	2	93	0	86	6	0	NaN	0

Performance of the PLS-DA equations discriminating the T/NT status of the seeds bulks (wheat, barley and spelt) using NIR-HIS in comparison to NIR spectroscopy

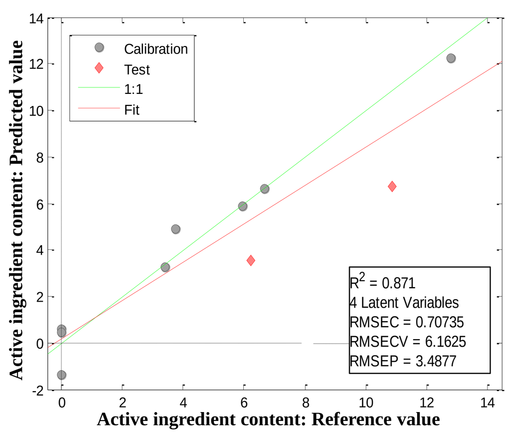
Homogeneity intra seeds batch: Seed by seed analysis

Discrimination PLSDA models



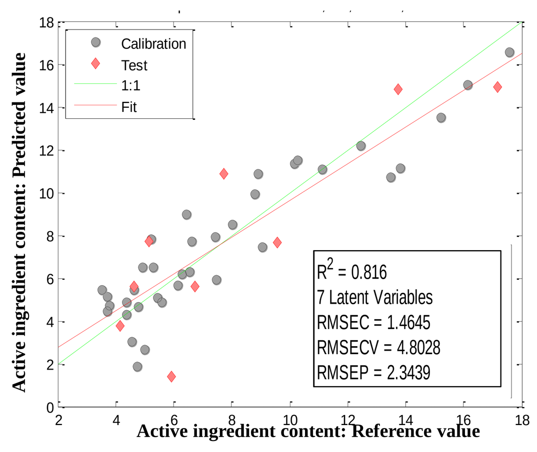
Predicted images showing the T/NT status on single seeds (wheat and barley)

Calibration PLS models



			% Target dose					% Samples		
Groups of pesticide	Active ingredient	n	Mean	SD	Min	Max	CV	<30%	>30%	± 30%
Ultra performance liquid chromatography										
Group 1	Prochloraz + triticonazole	2	73,9	8,4	68	79,9	11,4	50	0	50
Group 2	Prothioconazole	5	52,1	15,1	34,2	66,9	29	100	0	100
NIR spectroscopy										
Group 1	Prochloraz + triticonazole	2	58,9	19,2	45,3	72,4	32,5	50	0	50
Group 2	Prothioconazole	5	58,1	19	40,1	83,2	32,7	60	0	60
NIR hyperspectral imaging										
Group 1	Prochloraz + triticonazole	2	59,3	24,4	42,1	76,6	41,1	50	0	50
Group 2	Prothioconazole	5	48,4	14,5	32,4	66,1	30	100	0	100

PLS model and results for barley showing the classification of the treated seeds bulks in 2 groups: underdosing (<30%) and overdose (>30%) using NIR-HIS in comparison to NIR spectroscopy and UPLC



		% Target dose					% Seeds		
Sample	n	Mean	SD	Min	Max	CV	<30%	>30%	± 30%
Ultra performance liquid chromatography									
Wheat									
20	24	86,6	30,3	35,8	159	37,6	33	4	37
51	24	239,7	147,5	143,7	795,7	61,5	0	100	100
Barley									
74	24	49,6	30,5	23,1	149,2	61,6	83	4	87
75	24	55,2	27,5	22	107,2	50,0	71	0	71
NIR hyperspectral imaging									
Wheat									
20	24	93,6	26,3	40,7	165,6	28,1	12	8	21
51	24	187,3	35,7	115,2	269,4	19,0	0	96	96
Barley									
74	24	44,5	22,1	8,5	103,5	49,6	87	0	87
75	24	49,5	25	17,1	93,7	50,4	79	0	79

PLS model (barley) and results (wheat and barley) showing the classification of the treated single seeds in 2 groups: underdosing (<30%) and overdose (>30%) using NIR-HIS in comparison to UPLC.

Conclusions

The PLS-DA models allow classifying the seeds based on the specie and the treated/untreated status with a sensitivity of 100%. The discrimination according to the type of pesticide allows differentiating two out of three groups with a sensitivity of 100%. Hyperspectral imaging also allows to provide information on the presence of cereal seeds mixture in a batch but also the presence of untreated seeds in a treated seeds batch. The homogeneous distribution of the treatment between the seeds of one batch or within each seed is also assessed. On another hand, the calibration models allow classifying the treated seeds in 2 groups: underdosing and overdose.

The application of chemometrics on near infrared hyperspectral images offers new future prospects for the quality control of the coating efficiency of pesticides on seeds.

Acknowledgements

The authors wish to thank Guillaume Jacquemin from the Research Unit "Crop Production Systems" at the CRA-W for providing samples and Olivier Pigeon's team from the Research Unit "Plant Protection Products and Biocides Physico-chemistry and Residues" for providing reference analysis.

Walloon Agricultural Research Centre