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Role of Near - Infrared Spectroscopy in Seed Quality Evaluation: A Review

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ABSTRACT

The use of high-quality seeds is one of the most important elements for increasing agricultural production in any farming system. This element has become more crucial than ever for providing enough food security for the rising population, which is expected to exceed nine billion by year 2050. Selecting high yielding varieties of disease, insect, lodging and shattering resistance, along with other desirable characteristics are the basic keys for satisfactory crop performance and yield. The production of high-quality seed is the cornerstone of any successful agriculture program. It is also a good marketing tool for increasing the potential sale of crops, especially in today's competitive market. Therefore, adopting an efficient method to evaluate the seed quality non-destructively is the need of hour. One such technique or method is the use of NIR which helps to assess seed quality non-destructively and sort out seeds based on seed health, seed deterioration, viability, vigour including protein, starch and fatty acid composition as well as abiotic and biotic seed damage. It is a non-destructive analytical technique requires little sample preparation time and high-throughput, which makes it as a seed analysis tool.

Key words: High quality seeds, Near-infrared spectroscopy, Viability, Vigour.

Seed is one of the basic inputs for enhancing production. An optimum plant density per unit area is one of the keys to improve crop productivity. Hence, each and every seed should readily germinate and produce a vigorous and healthy seedling thereby ensuring its high productivity. (Kumar *et al*., 2017). Availability of good quality seed is the key for successful agriculture and their use is an important factor in the production of higher yield (Rejeendran *et al*., 2015). NIR absorption spectra are useful for the identification and quantitative analysis of compounds containing functional groups made up of hydrogen bonding to carbon, nitrogen and oxygen. Karl Norris, pioneer of NIRS, developed the first applications of NIRS for grains and seeds analysis in 1960s (Hart *et al*., 1962; Norris *et al*., 1965). Since then, instrumentation, statistical methods and software have been improved and number of applications has exponentially grown. NIRS is now a mature analytical method for grains and seeds, recognized by the American Association of Cereal Chemists (AACC) and the American Oil Chemist Association (AOCS). Near-infrared spectroscopy, working either in reflectance (NIRS) or transmittance (NITS), is widely used for the analysis of quality traits of intact seeds from different crops, especially cereals and oilseeds (Williams and Sobering, 1992). This technique is nondestructive, fast, cost-effective and permits the simultaneous analysis of many traits in a single measurement (Shenk and Westerhaus, 1993). NIRS technology is based on the absorption of near infrared light by organic compounds and water, which has been used in several applications for single seed analysis with notable success (Kusama *et al*., 1997; Cogdill *et al*., 2004; Pearson *et al*., 2001). NIRS is also known to play a role in simplification of the analysis of chemical and physical properties without sample preparation. It is based on the overtone and combination bands of specific functional groups, e.g., C-H, N-H and O-H bands, which are the primary structural components of organic molecules (Cozzolino *et al*., 2004). NIRS has become as a powerful tool due to chemometrics. Which does not require use of chemicals or reagents for estimation of the physical and chemical properties of samples.

NIRS can assess several constituents at the same time; therefore, it is also efûcient for mass-screening. Harmonic and combination vibration of the basic vibration can be observed in the wave length range of NIR spectroscopy. Unlike radiation in the medium infrared range (MIR), the short wave, energy-rich radiation has the crucial advantage that only the basic vibrations can be observed here. Harmonic vibrations can result from the simultaneous intake of several light quanta. Combination vibrations occur

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when a light quantum stimulates two different vibrations at the same time. In comparison, only a little energy is used when higher order vibrations are stimulated. This results in the much greater penetration depth of the near infrared light compared to the medium infrared range where only a few micrometres are possible. This means that a sample pre-treatment usually isn't required and NIRS is fast and automatic, reproducible, easy, reliable and non-destructive. NIRS calibration is less accurate than wet chemistry. NIRS Measurement outside of range of calibration samples is invalid and Small calibration sample sizes can lead to over confidence.

This review gathers relevant aspects of seed quality analysis by NIRS by both quantitative and qualitative means using NIRS. Its application in seeds will enable large screenings of low-quality seeds at single seed level and also bulk of seeds, which opens up a new way to access the seed quality by non-destructive manner.

The application of NIR Spectroscopy in Seed Science is reviewed here under

Reference methods and detection limit of NIR spectroscopy

NIRS is not an analytical method for direct measurement of trace elements or compounds found at part per million (mg/kg) or part per billion in the seed. The small size of seeds, the detection limit is a limitation of NIRS on single seed analysis. NIRS was adapted for analysis of single kernels in the mid 1990's. Advantages of single-kernel analysis include that it can detect attributes that may only be present in a few kernels from a bulk sample and it can provide information about distribution of attributes within a sample. In addition, specific kernels can be preserved for further analysis or used to propagate specific traits in breeding programs.

Dowell and Maghirang, (2002) suggested that weight less than 0.1 percent of the seed cannot be accurately measured. For instance, Patrick and Jolliff, (1997) observed that predicting meadow foam seeds with oil content below 5 mg were consistently overpredicted so the detection limit was fixed. But on the other hand, Janni *et al*., (2008) obtained large errors when predicting oil in corn kernels when including kernels with oil content above 8 percent on dry mass.

Seeds with abnormally high concentrations of any compound (specific hybrids or genetically modified seeds) are a problem for conventional NIR calibrations as those new seeds may present additional genetical or morphological changes that make them different from the rest and add an additional source of variability to be modelled and which may not always be properly done by linear methods like PLS (Partial least squares). The low detection limit of NIR may be even higher than 0.1 percent depending on the initial size of the seeds and the characteristics of the compound to be measured. The impact of kernel size on calibration accuracy was described by Tajuddin *et al*., (2002) when developing PLS oil calibrations for large (≥6mm) and small (≤6mm) soybean seeds. Larger seeds lead to better calibrations than smaller seeds standard error of prediction 0.09 and 0.14 percent respectively. On the other hand, attributes such as moisture often lead to much better accuracies than compounds such as protein or oil due to the strong absorption of water in the NIR region. This also means that the detection limit for moisture is lower compared to other compounds.

Accuracy and Feasibility of near Infra-Red spectroscopy for Single-Kernel Measurement

Single kernels can be measured by reflectance and transmittance spectroscopy with standard errors that are usually suitable for screening purposes. While the standard error of single-kernel analysis may be twice that of bulk-sample analysis, the benefit of knowing the distribution of attributes within a sample outweighs this loss in accuracy for many applications. The increase in sensitivity and reduction in cost of this technology and increases its acceptance in all industry segments.

While many attributes can be measured with this technology, it is limited to those that are present in sufficient quantities to significantly affect NIR absorption. Thus, it may not be possible to detect attributes that comprise ≤ 0.1 percent of kernel mass. It is useful for detecting the presence of internal insects and predicting the level of protein or oil, but it may not be useful for detecting minute traces of attributes such as those associated with some transgenic traits and are present at the ppm or ppb level. While literature shows that levels of transgenic traits, fumonisin and aflatoxin in corn (Kramer *et al* 2000; Pearson *et al* 2001; Dowell *et al*., 2002a) and vomitoxin in wheat (Dowell *et al* 1998) can be predicted, the standard errors of these predictions are very high. The ability of NIRS to predict levels of transgenic traits or toxins is likely due to the high correlation of these attributes to changes in other intrinsic characteristics such as protein levels, changes in the protein-starch matrix, etc.

High-Speed Sorting

High-speed sorters have commonly been used to remove visible defects in seed lots and recently gas sensors have been incorporated into these systems. Inspection and sorting with these instruments can occur at a rate as high as 10,000 kernels/s, or about 1100 kg/h. Dowell *et al*., (2002a) used a ScanMaster II at Texas to remove infested seeds from uninfected seed with 100% accuracy. They have also recently used this technology to remove red wheat from white wheat stock (Pasikatan and Dowell, 2002) and to sort into high and low protein groups. While these systems cannot quantify levels of attributes in single kernels and are limited to one or two wavelengths, they can rapidly sort samples into two groups that have distinctly different traits. Breeders and seed technologist are now using this technology to purify seed stock and it may have additional applications in the seed industry.

NIR-based prediction of seed germination

Standard germination percentages provide an estimation of a seed lot's potential for germination and seedling establishment under favourable conditions. Amery *et al*., (2018) reported that differences in spectral absorbance were observed for low and high germinating seed lots in soybean (Fig 1). This was observed over the entire 950–1650 nm wavelength range with more pronounced differences found beyond 1400 nm, indicating the potential to differentiate between seed lot categories using NIR spectroscopy.

The partial least square statistics for quantitative and qualitative models using GRAMS/AI software developed for predicting germination are summarized in Table 1. The potential for using NIR spectroscopy for quantitative measurement was relatively limited as shown by R^2 values, both for the training and validation data sets, ranging from 0.575 to 0.673 and 0.549 to 0.659, respectively. Qualitative discrimination models for three sample set for predicting seed germination. When the mid-predicted score of 1.5 was selected as the classification cut-off or threshold level, correctly predicting high germination seed lots (97.4-100%) but were very low for predicting low germination seed lots (28.6–47.6%). Shifting the classification threshold level from 1.5 to 1.7 allowed for better balance in correct classification of low and high germination seeds. The correct classification for low germination seeds improved from 81.0 to 85.7%, while the high germination soybean seeds went down to 82.1 to 89.7% (Amery *et al*., 2018).

NIR-based prediction of seed vigour

Seed vigour is a more sensitive indicator of seed lot quality compared with standard germination, as it relates to field establishment (TeKrony and Egli, 1977; Egli and TeKrony, 1995). It is common for seed lots with the same standard germination to have different vigour levels resulting from variable seed production systems or storage conditions. Vigour is also commonly reduced during seed storage prior to any loss in standard germination (Delouche and Baskin, 1973). Seed vigour tests fall roughly into three general categories (Geneve, 2005). The first set of tests measure standard germination percentage after a pre-germination stress imposition. The second group indirectly measures germination potential utilizing an accepted physiological or biochemical aspect of germination. The third set of vigour tests utilizes visual post germination analysis. The major stress-related vigour tests include accelerated ageing, controlled deterioration and cold test, while the most utilized indirect method uses electrolyte leakage (Marcos-Filho, 2015). Therefore, to genuinely identify the ability for NIRS to predict seed lot vigour.

For soybean seed vigour, three analyses were done for the 44-high germination (≥94% germination) samples (132 spectral data) were obtained: (a) quantitative, (b) 3-category qualitative and (c) 2-category qualitative. The quantitative model used the accelerated ageing germination percentage values. The 3-category qualitative model assigned dummy numerical scores for: (a) low, <74 % accelerated ageing germination (score=1), (b) medium, 74–89% AA germination (score= 2) and (c) high, ≥90 % accelerated ageing germination (score=3) vigour level. 2-category qualitative model, the medium and high Accelerated ageing germination categories in the 3-category qualitative model were combined and were assigned dummy numerical scores of (a) score=1 for low, <74 % accelerated ageing germination and (b) score=2 for combined medium and high, ≥74 % accelerated ageing germination (Amery *et al*., 2018) .

There were differences in spectral absorbance for the entire wavelength range for low, medium and high vigour soybean samples (Fig 2).

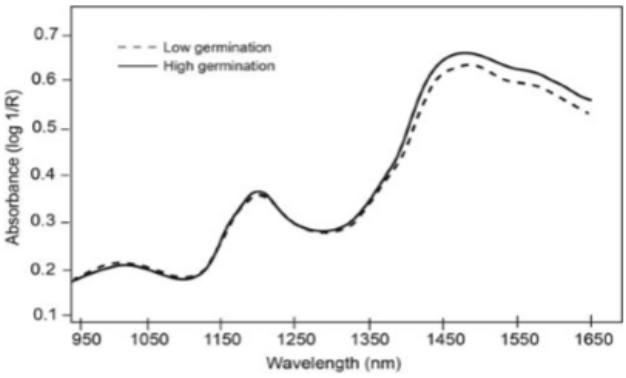


Fig 1: Average absorbance spectra (log 1/reflectance) for samples differentiated into low and high germination. (Amery *et al*., 2018).

Table 1: Statistical measure for prediction models developed for quantitative and qualitative determination of germination using training data set and the resulting classifications of validation sample.

Prediction model	Training data set				Validation data set Correct classification (%)		
	Sample set	Factors	R^2	SECV*	Predicted score cut-off	Low	High
Qualitative germination (%)	1	5	0.3944	0.2904	1.5	47.6	100.0
	2	6	0.4720	0.2712	1.7	85.7	89.7
	3	8	0.5214	0.2586	1.5	28.57	100.0
Quantitative germination (%)	1	12	0.6034	12.55	1.7	85.71	82.05
	2	10	0.5748	11.42	1.5	42.86	97.44
	3	10	0.6733	11.42	1.7	80.95	89.74

*SECV, standard error of cross-validation. (Amery *et al*., 2018).

Table 2: Statistical measures for prediction models developed for quantitative and qualitative determination of vigour (accelerated ageing) using training data set and the resulting classifications of validation samples.

Prediction model	Training data set				Validation data set Correct classification (%)		
	Sample set	Factors	R^2	SECV*	Low	Medium	High
Qualitative vigour 3-category	1	4	0.224	0.633	40.0	54.5	66.7
	2	7	0.228	0.616	0	63.6	61.6
	3	4	0.201	0.638	50.0	66.7	55.5
Quantitative vigour 2-category	1	6	0.418	-	100.0	-	96.6
	2	7	0.574	-	80.0	-	96.6
	3	6	0.536	-	83.3	-	96.6
Quantitativevigour	1	4	0.320	14.40	-	-	-
	2	6	0.439	12.81	-	-	-
	3	4	0.280	13.95	-	-	-

*SECV, standard error of cross-validation. (Amery *et al*., 2018).

Table 3: Classification results for the calibration set of the viable and non-viable (aged) watermelon seeds using the PLSDA model with various pre-processing methods.

Preprocessing	Viable seeds (n=315)		Non- Viable seeds (n=315)		Total seeds (n=665)		Accuracy
	Correct	Incorrect	Correct	Incorrect	Correct	Incorrect	(%)
Min normalization	315	0	350	0	665	0	100
Max normalization	315	0	350	0	665	0	100
Range normalization	315	0	350	0	665	0	100
MSC	315	0	350	0	665	0	100
SNV	315	0	350	0	665	0	100
Raw	315	0	350	0	665	0	100

MSC: Multiple scatter correction SNV: Standard normal variant. (Lohumi *et al*., 2013).

The qualitative prediction of vigour was evaluated using the 3-category and 2-category models. The 3-category model grouped samples into low, medium and high vigour and were assigned the score of '1', '2' and '3', respectively. Discriminant analysis showed overall poor correct classifications. Qualitative vigour 2-category models showed correct classification compared to other 3-category and quantitative models (Table 2).

NIR-based prediction of seed viability

Conventional methods used to evaluate seeds viability are destructive, time consuming and require the use of chemicals, which are not feasible to implement to process plant in seed industry. Where, NIR Spectroscopy effectively distinguish between viable and non-viable seeds. Lohumi *et al*., (2013) reported that FT-NIR reflectance spectra of both viable and non-viable watermelon seeds spectrum were collected in the range of 1000-2500nm. The calibration and validation set derived from the PLS-DA model classified viable and non-viable seeds with 100 % accuracy with pre-processed spectra (Table 3 and 4). The reason for non-viable seeds that change in chemical composition of the seed membrane such as lipids and proteins might be responsible for germination ability of the seeds. At 1570nm range N-H first overtone stretching vibration, which represents protein content, 2252nm range of O-H stretching which represents starch content and another absorption band also presents

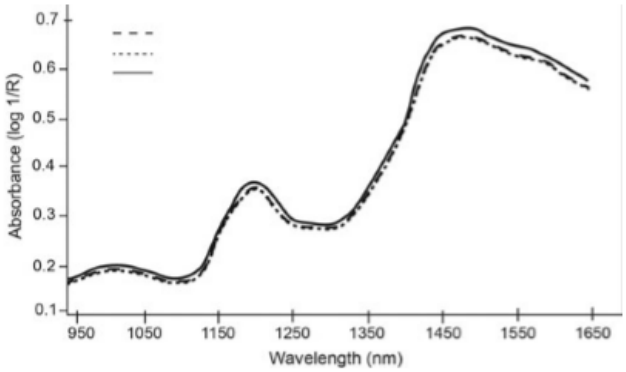


Fig 2: Average absorbance spectra (log 1/reflectance) of high-germination soybean seeds differentiated into low, medium and high vigour (accelerated ageing). (Amery *et al*., 2018).

near 2345nm, which was related to CH_2, a functional group being assigned to lipids (Fig 3).

Shrestha *et al*., (2017) reported that NIR spectroscopy in classifying viable and non-viable tomato seeds of two cultivars using chemometrics. The data exploration was performed by principal component analysis (PCA). Subsequently, viable and non-viable seeds were classified by partial least squares discriminant analysis (PLS-DA) and interval PLS-DA (iPLS-DA). The indication of clustering of viable and non-viable seeds was observed in the PCA of each cultivar and the pooled samples. However, the PCA

Table 4: Prediction results for the validation set of the viable and non-viable (aged) watermelon seeds using the PLSDA model with various preprocessing methods.

Preprocessing	Viable seeds (n=135)		Non- Viable seeds (n=150)		Total seeds (n=285)		Accuracy
	Correct	Incorrect	Correct	Incorrect	Correct	Incorrect	(%)
Min normalization	135	0	150	0	285	0	100
Max normalization	135	0	150	0	285	0	100
Range normalization	135	0	150	0	285	0	100
MSC	135	0	150	0	285	0	100
SNV	135	0	150	0	285	0	100
Raw	135	0	150	0	285	0	100

MSC: Multiple scatter correctionSNV: Standard normal variant. (Lohumi *et al*., 2013)

Table 5: Misclassified corn kernals in the viability study for each tested algorithm (mis-classified seeds/total seeds belonging to that class).

Algorithm	Raw spectra			SNV preprocessing		
	Non-viable	Viable	Total (%)	Non-viable	Viable	Total (%)
PLS-DA	125/242	30/82	47.8	80/242	44/82	38.3
SIMCA	112/242	37/82	46.0	126/242	32/82	48.8
LS-SVM	133/242	28/82	49.7	124/242	43/82	51.5
K-NN	116/242	37/82	47.2	119/242	42/82	49.7

(Agelet *et al*., 2012).

did not exhibit a pattern of separation among the early, normal and late germinated tomato seeds. The NIR spectral regions of 1160–1170, 1383–1397, 1647–1666, 1860–1884 and 1915–1940 nm was identified as important for classification of viable and non-viable tomato seeds by iPLS-DA. Spectral region from 1910 to 1980nm found protein moieties and protein bound water–viable seeds, 1800-1884nm found cellulose or carbohydrates and it relationship with non-viable seeds. Mannose (68%), glucose (21%) and galactose (5%) is present in tomato endosperm cell wall. Mannose creates hardness and control radicle protrusion. Increase soluble sugars during ageing reduction in seed viability and hydrolysis of sucrose it leads to increase soluble sugars and 1336 and 1655nm found major fatty acids found positively relationship with non-viable seeds. The sensitivity i.e. ability to correctly identify the positive samples and specificity i.e. ability to reject the negative samples of the (iPLS-DA) model on identified spectral regions for prediction of viable and non-viable seeds were 0.94 and 0.94 respectively and were higher than from (PLS-DA) model on original spectra. The iPLS-DA model predicted samples with classification error rate of 6.29 percent as compared to the 13.10 percent by the PLS-DA.

Agelet *et al*. (2012) recorded that discrimination of non-viable seeds from viable seeds using different statistical methods such as PLS-DA, SIMCA, LS-SVM and K-NN models and found that it is not possible to distinguish in both raw and SVN pre-processing spectra to get more misclassified seeds in corn and soybean (Table 5 and 6).

NIR-based prediction of infected seeds

There are few studies using NIRS for discriminating sound and damaged soybeans and wheat kernels. Wang *et al*. (2001) analyzed heat-damaged kernels using NIRS and could accurately discriminate heat-damaged kernels (>95% correctly classified) with just two wavelengths and partial least squares discriminant analysis (PLS-DA). They suggested that the classification was driven by differences in light scattering and color change in heat-damaged kernels. A classification model for wheat based on vitreous and non-vitreous endosperms, including defective kernels such as bleached, cracked and sprouted, found that bleached kernels were misclassified (Wang *et al*., 2002a). Wang *et al*. (2002b) also classified soybean seeds according to the type of damage (sprout, heat, frost, mold, or weather) using artificial neural networks (ANN) with discrimination success rates over 90% for most of the damaged types. In that study heat damage classifications achieved lower accuracies

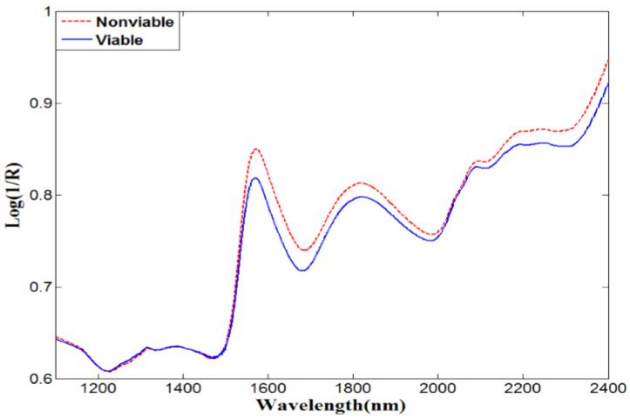


Fig 3: FT-NIR mean spectra of the original pre-processed watermelon seeds using the Standard normal variant method. (Lohumi *et al*., 2013)

Table 6: Misclassified soybean seeds in the viability study for each tested algorithm (mis-classified seeds/total seeds belonging to that class).

Algorithm	Raw spectra			SNV preprocessing		
	Non-viable	Viable	Total (%)	Non-viable	Viable	Total (%)
PLS-DA	150/299	30/80	48.0	142/299	44/80	47.2
SIMCA	157/299	37/80	49.1	247/299	32/80	63.4
LS-SVM	166/299	28/80	53.6	133/299	43/80	44.3
K-NN	178/299	37/80	54.9	120/299	42/80	43.3

(Agelet *et al*., 2012)

Table 7: Misclassified corn kernels in the heat-damage study for each tested algorithm (misclassified seeds/total seeds belonging to that class).

Algorithm	Raw spectra			SNV preprocessing		
	Damaged	Sound	Total (%)	Damaged	Sound	Total (%)
PLS-DA	0/51	1/52	1.0	0/51	2/52	1.9
SIMCA	3/51	2/52	4.9	3/51	13/52	15.5
LS-SVM	6/51	9/52	14.6	2/51	3/52	4.9
K-NN	10/51	8/52	17.5	10/51	8/52	17.5

(Agelet *et al*., 2012).

Table 8: Misclassified corn kernels in the frost-damage study for each tested algorithm (misclassified seeds/total seeds belonging to that class).

Algorithm	Raw spectra			SNV preprocessing		
	Damaged	Sound	Total (%)	Damaged	Sound	Total (%)
PLS-DA	6/12	4/13	40.0	6/12	2/13	32.0
SIMCA	6/12	4/13	40.0	10/12	0/13	40.0
LS-SVM	6/12	4/13	40.0	11/12	0/13	44.0
K-NN	5/12	5/14	40.0	3/12	7/13	40.0

(Agelet *et al*., 2012).

compared to the ones achieved in heat-damaged wheat kernels (64%). Kusama *et al*. (1997) used near infrared spectroscopy (NIRS) for analysing aging of soybeans. They classified with 60% accuracy sound seeds vs 3-day artificially aged soybean seeds, 80% accuracy when aged for 5 days and 100% accuracy when aged for 8 days.

Discrimination of frost-damaged corn kernels was not possible with NIRS, even with the use of non-linear methods such as Support vector machines (LS-SVM). Frost-damaged soybean discrimination by NIRS has been reported to be successful (accuracy over 90%) using artificial neural networks (ANN) (Wang *et al*., 2002b). It could be due to the embryonic tissue in corn is only a relatively small volume of the seed while in soybeans it occupies the entire seed for that reason soybean seeds can be easily prone to any damage. So that, NIRS may accurately classify the seeds based on damage and without any correlation with germination (Agelet *et al*., 2012) (Table 7 and 8).

Several studies have demonstrated the capability of NIRS combined with multivariate analysis for the discrimination of sets of similar biological materials. For instance, NIRS has successfully discriminated uninfested and infested wheat kernels (Ridgway and Chambers 1996; Ghaedian and Wehling 1997; Dowell *et al*. 1998). NIRS has been applied to identify stored-grain insects (Dowell *et al*., 1998) and to detect parasitized rice weevils in wheat kernels (Baker *et al*. 1999). Recently, NIRS has been applied to distinguish between dead-filled and viable-filled seeds of *Pinus sylvestris* L. (Lestander and Oden, 2002), empty and viable seeds of *Pinus patula* (Tigabu and Oden 2003a) and sound and insect-damaged seeds of *Albiziaschimperiana* (Tigabu and Oden 2003b).The feasibility of this technique to distinguish between sound and internally insect infested seeds in a seed lot of *Cordia africana* (Tigabu and Odén 2002) was studied. However, a single seed population was used in that study, hence it did not help to determine whether the physical and chemical properties of different seed lots affected the detection sensitivity.

NIR-based prediction of biochemical parameters

Seed protein content of rapeseed is a quantitative trait governed by additive gene action (Grami and Stefansson, 1977) that highly influenced by the environment and, therefore, exhibiting a low heritability (Grami *et al*., 1977). Velasco *et al*., (2002) validaed set of 117 additional seeds from three individual plants from the cultivars Bristol, Lirajet and Maplus for protein content both by NIRS and combustion methods. The coeficient of determination between NIRS and

combustion method values in the validation set was 0.94, with a standard error of performance of 0.77% and a ratio of the SEP to the standard deviation (SD) of the validation set of 0.28.The coefficient of variation (CV) for seed protein content in individual plants, as determined by the combustion method, was 11.7% for Bristol, 8.9% for Lirajet and 9.5 % for Maplus. The comparison of such variation with the standard error (SE) of NIRS analysis, defined as the combination of the SE of the combustion method and the SEP of NIRS calibration equation, revealed that the maximum variance within individual plants that can be detected using NIRS analysis of protein content in single seeds was 0.86 for Bristol, 0.83 for Lirajet and 0.85 for Maplus (Fig 4).

Near-infrared analyses of intact seeds are commonly made on bulk samples of variable size, depending on the instrument and device used. Furthermore, instruments including single-seed sample holders are available (Downey, 1994). The use of these devices has permitted the development of single-seed calibration equations for the analysis of oil and protein content in corn and soybean (Orman and Schumann, 1992; Dyer and Feng, 1995; Abe *et al*., 1995), protein content in wheat (Abe *et al*., 1995) and oil content in meadow foam (Patrick and Jolliff, 1997). Furthermore, Sato *et al*. (1995) pointed out the potential of NIRS to predict linoleic acid content in husked sunûower seeds. NIRS technique has not been applied to the analysis of single seeds of rapeseed. NIRS is currently used for the analysis of bulk samples of intact seeds for oil, protein and glucosinolate content and the fatty acid composition of the seed oil (Velasco and Becker, 1998).

Near-Infrared Spectroscopy was used for the first time to measure moisture concentration in soybean by Ben-Gera and Norris, (1968) and since then has been used to measure moisture, protein, oil and starch concentration in forage, legume and cereal crops, as well as other food commodities. It has also been applied for fatty acid profiling in oilseeds like Velasco *et al*. (1999) in rapeseed, Tillman *et al*. (2006) in peanut, Sato *et al*. (1995) in sunflower, Sato *et al*. (2003) in sesame and (Kovalenko *et al*., 2006; Pazdernik *et al*., 1997; Sato *et al*., 2002) soybean. The correlation between standardized NIR absorbance at 1708 nm and linoleic, oleic acid (0.85 and 0.88 respectively) in soy flour was examined by Sato *et al*. (2002). The palmitic (r 2 = 0.93), stearic (r 2 = 0.89), oleic (r 2 = 0.95) and linoleic acid (r 2 = 0.93) of soybean cotyledons has also been estimated using NIR spectroscopy Roberts *et al*. (2006). Using NIR technique, determination coefficients from 0.38 to 0.71 and from 0.18 to 0.56 (n = 90) were reported for all fatty acids of groundnut and whole soybean samples respectively Pazdernik *et al*., (1997). Near-Infrared Transmittance (NIT) Spectroscopy was used to develop chemometric models for fatty acids in whole soybean samples and performance of linear (PLS) and nonlinear calibration Support vector machines (SVM) and Artificial neural networks (ANN) methods were compared Kovalenko *et al*. (2006).

Pande *et al.* (2014) revealed that rapid and simple FT-NIR procedure to estimate phytic acid content in green gram seeds was developed using a single calibration model. The model was developed using the spectral region 4000–12,000 cm^{1}. This study demonstrated the suitability of FT-NIR spectroscopy to determine the level of phytic acid in green gram seeds samples. The maximum value of coefficient of determination (R^2) for the prediction of phytic acid was 593 mg/100 g and 591 mg/100 g for calibration and validation (Fig 5).

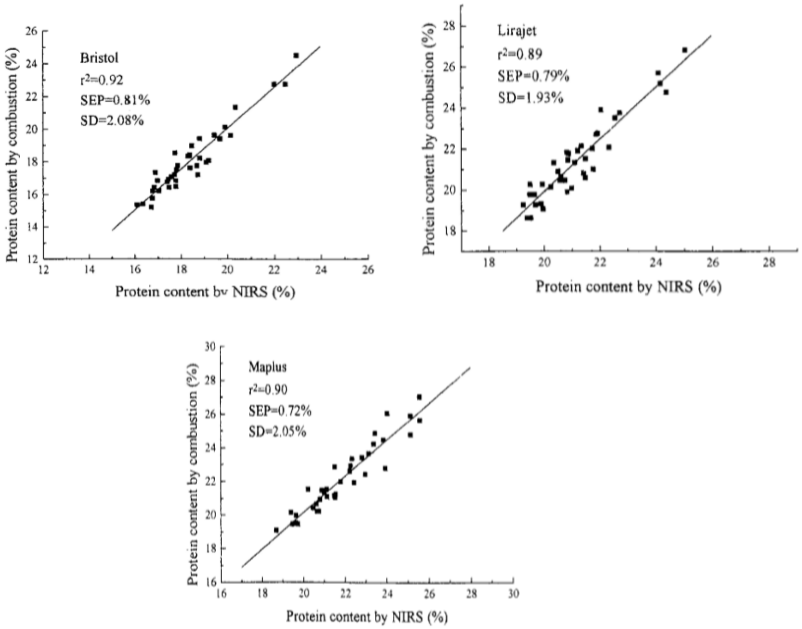


Fig 4: Scatter plots of combustion vs. NIRS values for protein content (% of fresh seed weight). (Velasco *et al*., 2002).

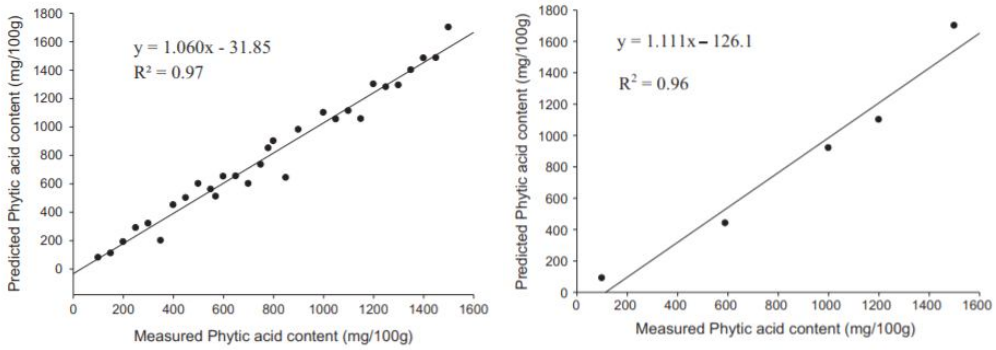


Fig 5: Linear regression plots of measured vs. predicted phytic acid content values for cross and test validated data set. (Pande *et al*., 2014).

Nadaf *et al*., 2014 observed that near infrared reflectance spectroscopy (NIRS) for large scale screening of fatty acid profile in peanut. The fatty acid profile was obtained by Gas Chromatography (GC) and Near-Infrared Spectroscopy. Modified partial least square regression model (mPLS) is the best calibration and satisfactory prediction abilities. The r^2 between NIRS and GC was 0.79 (palmitic acid), 0.91 (oleic acid) and 0.89 (linoleic acid) in cross validation, demonstrating the high reliability of NIRS in determining these fatty acid concentrations in intact single seeds. NIRS permitted analysis of about 40 samples per hour as against 1-2 samples in GC.

Plant breeders constantly looking to improve their varieties every year and to obtain new special trait seeds. This can be achieved by careful selection of best individual traits. Use of bulk seed samples in selection process for large seed production only a fraction with desired trait. Heritability of a desired characteristic may be low in large seed production. So that, analysing individual seed trait to understand the future plant characteristic and characteristics of its next generation. One of the best ways to predicting the individual seed trait is Near-Infrared Spectroscopy. It will analyse the individual trait (All seed quality parameters such as physiological and biochemical aspects) within a minute. This technology is very useful to farmers in the way of predicting seed quality parameters before sowing of seeds in the field so it will control the crop losses in the field. Seed should be physically and genetically pure and it should satisfy minimum seed certification standards. In seeds respiration rate and other physiological and biological processes should be kept at low level during storage, In the case of grain not such quality maintenance can happen. In that way Near-Infrared Spectroscopy can be evaluating the quality traits. physiological and biochemical properties are altered in seed and grain because seed can be treated with pesticides and fungicides to protect seed against storage pest and fungi and maintain the moisture content low so that physiological and biochemical properties are different in seed and grain in that way NIRS is useful for predicting the quality in seed and grain.

Qualitative application in NIR spectroscopy

NIR spectroscopy focused on detecting and sorting by mold damage and insect infestation have the most extensive related literature. Chemical imaging technologies allowed early detection and tracking of fungi development on kernels when wavelengths from the visible region were also used (Polder *et al*., 2005; Williams *et al*., 2012). Discrimination of insect-damaged or infested kernels is well studied. Some insect-damaged kernels are easily removed by cleaning, but insects growing inside the grain are invisible for methods based on visual inspections Mendoza *et al*. (2004) but NIRS helps to segregate these seeds. Accuracies in segregating kernels contaminated with insects depended of the size of the larvae. Kernels with large larvae could be discriminated with accuracies up to 94 percent, small larvae lead to accuracies of only 63% Maghirang *et al*. (2003). When the larvae are large enough, high discrimination accuracies were possible using a short wavelength range Ghaedian *et al*., (1997).The most relevant wavelengths in insect infestation were the ones related to water (involved in the metabolic processes of the insects), protein, lipids, phenolic compounds and carbohydrates because of the absorption of chitin from the insect cuticle and a decrease of starch levels in the grain (Ghaedian and Wehling 1997; Baker *et al*., 1999; Dowell *et al*., 1998).

CONCLUSION

NIR spectroscopy is helps in seed quality evaluation using single seed itself. The predictive ability of the calibration is mainly given by the commodity (kernel size and heterogeneity) and the instrumentation characteristics. When dealing with heterogeneous seeds reflectance is the best working mode. Partial least squares (PLS) regression is the best calibration model for predicting the sample quality. The NIRS is a very useful tool for breeders and seed technologist interested in vigour genetics and to germplasm preservation programmes and also useful tool for predicting seed quality non-destructively.

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