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## Rapid measurement of moisture content in groundnut kernels using non-destructive near infrared reflectance spectroscopy

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**Abstract**

The Near Infrared Reflectance Spectroscopy (NIRS), a non-destructive and robust tool was calibrated for rapid estimation of moisture content (MC) in whole groundnut kernels. A set of 8 groundnut genotypes were soaked overnight, followed by drying in hot air oven at 60<sup>o</sup> C. Data were recorded after every 2 hrs drying using a moisture meter followed by scanning in NIRS, till constant MC was obtained. NIR absorption spectral data from 400 to 2500 nm in 2 mm intervals were collected. Modified partial least squares (MPLS) regression was applied to scatter-corrected spectra (SNV and detrend). Calibration equation with high values for the coefficient of determination ( $R^2$ ), the coefficient of determination for cross-validation (1-VR) and low values for the standard error of calibration or standard error of cross-validation were estimated. Among the various models employed, model 2 with pretreatment 2,4,4,1 was best with an  $R^2$  of 0.99 in the calibration set, 1-VR value of 0.99 in the cross-validation set, lowest values for the standard error of calibration (0.33) and standard error of cross-validation (0.55). Calibration equations for moisture content showed a close relationship between NIRS predicted and lab values in this model. Thus, the selected model can act as the best models for prediction of moisture content in groundnut kernels with high accuracy. This study shows the potential of NIRS to predict the moisture content of groundnut seeds as a routine method in breeding programs, processing industries and for farmer's advice.

**Keywords:** Groundnut, moisture content, near infrared reflectance spectroscopy, coefficient of determination, cross-validation

**Introduction**

The moisture content (MC) in groundnut (Peanut; *Arachis hypogaea* L.) seeds is an important criterion in marketing, storing and processing. Storing groundnuts with high moisture content can prime to increased risk of mold fungi growth and aflatoxin contamination. Traditionally, groundnuts after harvesting are allowed to dry in the field for 1-2 days which reduces the MC by about 20%, followed by further drying of stripped pods in the sun for 1 or 2 days which brings down the moisture content to around 8-10%. Pods should be well dried to below 5% moisture for seed purpose. The storage of groundnut at an optimum moisture level is necessary for processing industry. There could be many lots of groundnut moving around industry yard at various drying stages. The average MC of samples from each load has to be periodically examined to determine the desired MC. It is desirable that the MC measurement should rapidly and non-destructively measured on whole kernels during the drying process to avoid over-drying of groundnut kernels, as over-drying not only increases the cost of drying but also affects the quality of the groundnut [1]. Moisture determinations involving the removal of water from seeds can be considered as the basic methods of moisture determination. When the dry matter is determined it is assumed that no-volatile material other than water was driven off. Where the moisture was trapped by some means and measured the assumption is that all the water was driven off and trapped. Much work was done to develop basic methods of moisture determination. The absolute methods like drying without heat, lyophilization, reversibility method and hot air oven method (dry weight/wet weight methods) are proven to be requiring much time, equipment, high degree of skill for operation and may lead to seed vigour/viability loss. So development of an accurate, rapid, non-destructive method to determine MC of whole groundnut kernels could save considerable time and labour during the drying process and prevent the loss of large quantities of edible groundnuts used for MC measurements [2].

Near-infrared reflectance spectroscopy (NIRS) is employed for the estimation of moisture content in grains and seeds [3]. Due to its rapidity (about 60-70 seconds per test), favorable economics, simplicity of sample preparation and absence of chemicals, it has become an extremely important adjunct to the grain and food industries. At present, NIRS technology is being used as an analytical method for the estimation of the composition of foods, feeds, grains, oilseeds, pharmaceuticals and in medical research. Previously, the NIRS was used at ICRISAT to develop calibration equations for rapid estimation of oil, protein and fatty acids (oleic, linoleic and palmitic acid) (unpublished data), which are being applied in groundnut breeding for selection and recording of quality traits in advanced breeding lines. The objective of this study was to develop calibration models for estimating MC of whole groundnut kernels and to use it as a tool for recording MC along with other kernel quality parameters.

## Material and Methods

### Groundnut Samples

Eight groundnut genotypes (ICGV 00441, ICGV 92195, ICGV 00440, ICGV 00308, ICGV 91114, ICGV 87846, ICGV 03043 and ICGV 89280) developed at ICRISAT were used in the study for estimation of moisture content. The sufficient seeds (~800 g) of eight genotypes were soaked in water for overnight. The soaked seeds were divided into ten batches to make the diverse gradient in moisture content. All the ten batches were kept in hot air oven for drying and each batch was removed after every two hours of drying starting first batch at 3 hours and last batch with 23 hours of drying. The data was recorded for every two hours drying using a moisture meter. The experiment was conducted during February 2018 to April 2018.

### Spectrum Collection

All the samples were scanned on a NIR Systems model XDS monochromator (model XDS RCA, FOSS Analytical AB, Sweden, Denmark). About 100g of sample was loaded into a rectangular cup, and reflectance spectra ( $\log 1/R$ ) from 400 to 2498 nm were recorded at 2 nm intervals. Each sample was subsequently scanned 32 times and the average spectrum was collected. In order to develop the calibration equation, about 100 g of each groundnut sample was scanned in a rectangular cup. The cup was filled up sufficiently to allow good absorption of the incident light. In each scan, NIR light was allowed to fall on the bottom of the sample holder containing the groundnut kernels, where it penetrated and interacted with the samples. The reflected energy spectrum over the wavelength range of 400-2,498 nm that carried absorption information of the samples was collected. The eight groundnut genotypes with recorded 80 data points were divided into a calibration.

### Spectral Data Analysis

NIRS spectral data were analyzed using WinISI4 (version 4.3) software (Infrasoft International, Port Matilda, PA, USA). Reflectance spectra between 400-2500 nm with 2 nm intervals were taken as independent variables and the MC of the sample as dependent variable. Modified partial least square (MPLS) regression analysis was performed on the

calibration set to develop an empirical equation, suitable for estimating the MC in unknown samples. For performing MPLS the number of parameters was set to 'default' and the number of cross-validation groups set to 8; with samples with a ' $H$ ' value larger than 4 (spectral outliers) and a (Student) ' $T$ ' value larger than 2.5 (sample which did not fit the calibration model) being eliminated [4].

Different mathematical pretreatment methods were tested on the calibration set and the best method was chosen based on the optimum results obtained for  $R^2$  (determination coefficient of calibration) and 1-VR (coefficient of determination in cross-validation). The mathematical treatment uses the raw data or the first or second derivatives of  $\log 1/R$  data to remove background differences while enhancing spectral differences; combined with gap sizes in data points over which the derivative is calculated [5]; and a smoothing algorithm that reduces random noise in the spectral data [6]. Calibrations were performed with five different mathematical treatments (1,2,2,1; 1,4,4,1; 1,8,8,1; 2,4,4,1; 2,8,8,1) using SNV + D (Standard Normal Variate + Detrend) scatter correction option. Scatter corrections are useful in reducing differences in the spectra related to physical characteristics such as particle size and path length of reflectance from the particle surface [7]. Four cycles of outlier elimination were allowed. Calibration models were assessed using statistics that included the standard error of calibration (SEC), the coefficient of determination in calibrations ( $R^2$ ), the standard error of cross-validation (SECV), and the coefficient of determination in cross-validation (1-VR) [4]. The optimum calibration equations were obtained based on the highest  $R^2$  or 1-VR and the lowest SEC or SECV values.

## Results and Discussion

The MC of eight groundnut samples estimated using moisture meter at different time intervals is given in Table 1. The results revealed great variation in MC among the samples ranging from 3.7 to 23.3% indicating their suitability for developing calibration equation using NIRS.

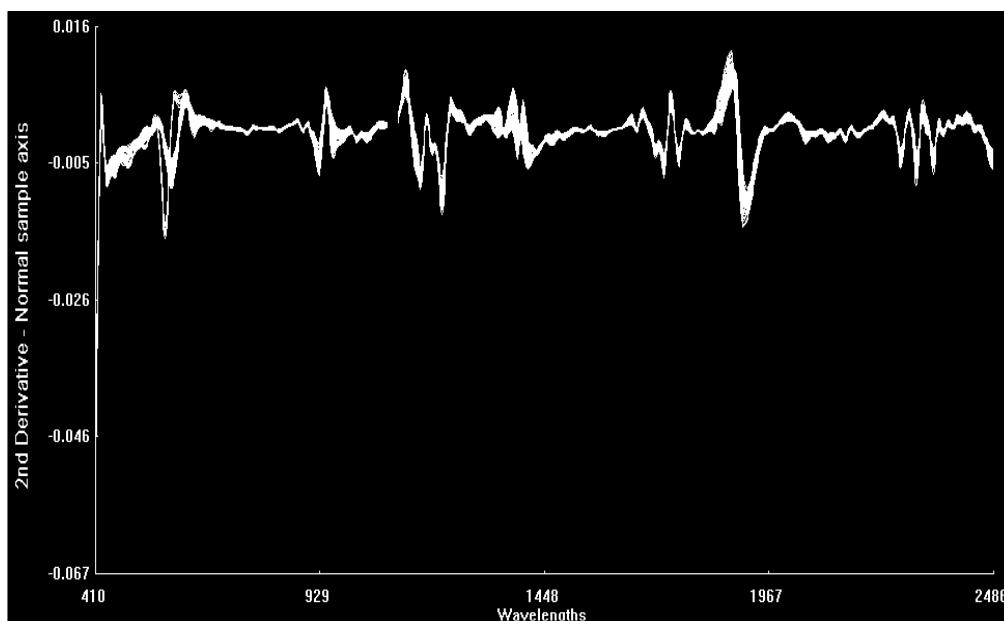
The reflectance moisture spectrum of groundnut for different MC levels is depicted in Fig 1. The averaged spectra of the different moisture levels have similar shapes, but different amplitudes. The peaks in the spectra represent high reflection of NIR electro-magnetic energy, which was influenced by differences in sample particle size, the reflective nature of the sample surface and by the spectrum baseline. In NIR reflectance spectroscopy, a beam of radiation is bombarded on the sample, penetrates a few millimeters, is diffused, and is then reflected back to the detector. Since the radiation penetrates and interacts with the sample, it carries absorption information and the representative spectra are returned as NIR absorption curves. The overall spectra of a wide range of MC in groundnut show strong absorption bands that differ based on MC.

The values for the mean, the range of moisture content and standard deviation in the calibration set was shown in Table 1. The variability for the moisture content generally varies from 20% in freshly harvested kernels to about 5% in well-cured groundnut [2]. In the current set of calibration set, a wide range in moisture content (3.7-23.3) enabled to develop a strong calibration equation for predicting MC in unknown samples.

**Table 1:** The moisture content of whole groundnut kernels in the calibration set

Genotype/ Per cent moisture content after oven dry	5 HAD	7 HAD	9 HAD	11 HAD	13 HAD	15 HAD	17 HAD	19 HAD	21 HAD	23 HAD
ICGV 87846	23.3	19.7	18.8	14.9	10.6	7.8	6.3	5.2	4.6	4.2
ICGV 89280	21.0	20.7	17.9	14.1	11.0	7.6	4.6	4.6	4.2	4.0
ICGV 91114	23.1	21.7	18.9	13.4	10.4	8.1	7.1	6.0	5.2	4.8
ICGV 00440	20.1	17.7	16.6	10.1	6.8	5.2	4.4	4.0	3.7	3.7
ICGV 03043	16.1	14.3	11.1	7.7	5.7	4.6	4.0	3.8	3.7	3.7
ICGV 92195	21.8	19.6	17.6	12.8	10.2	5.6	4.8	4.8	4.3	4.1
ICGV 00441	21.8	19.5	19.4	15.4	12.2	10.7	8.2	6.7	5.5	4.7
ICGV 00308	21.7	21.1	19.6	13.7	10.7	6.2	5.2	4.6	4.1	4.1
Total samples				80						
Range			3.7- 23.3							
Mean				10.6						
SD <sup>a</sup>				6.51						

HAD: Hours after oven-dry

<sup>a</sup> Standard deviation**Fig 1:** Near- infrared absorbance spectrum of groundnut kernels at different moisture levels

The calibration and cross-validation statistics obtained using the different mathematical pretreatments are depicted in Table 2. It was clearly evident that the model 2 with pretreatment 2,4,4,1 gave the optimum performance for MC. Using this pretreatment the calibration equations for MC showed a close relationship between NIRS predicted and lab values, with an  $R^2$  of 0.99 in the calibration set and 1-VR value of 0.99 in the cross-validation set. In earlier investigations, the reported

determination coefficient of calibration ( $R^2$ ) for moisture was 0.93 [2], 0.99 [8] and 0.98 for in-shell groundnuts [8]. The obtained  $R^2$  value (0.99) in the present investigation is as good as the earlier existing prediction model. Among five pretreatments, the model 2 showed the lowest values for the standard error of calibration (0.33) and standard error of cross-validation (0.55).

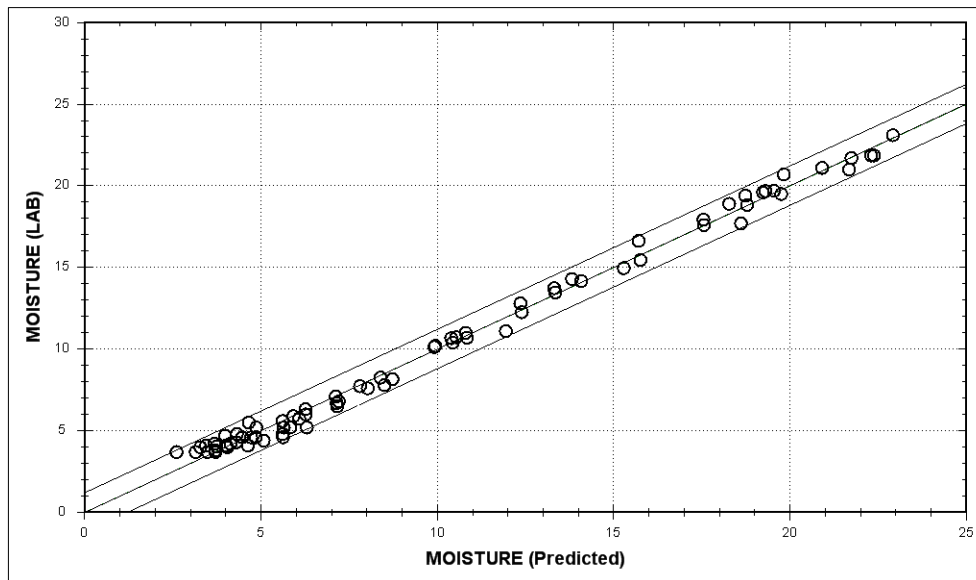
**Table 2:** Calibration and cross-validation statistics for groundnut kernels using different mathematical treatments by NIRS

Constituent	Model	Pretreatment	Calibration <sup>a</sup>				Cross-validation <sup>b</sup>	
			Mean	Range <sup>c</sup>	$R^2$	SEC	SECV	1-VR
Moisture	1	1,2,2,1	10.51	0-29.8	0.99	0.49	0.61	0.99
Moisture	2	2,4,4,1	10.38	0-29.6	0.99	0.33	0.55	0.99
Moisture	3	1,4,4,1	10.51	0-29.8	0.99	0.50	0.61	0.99
Moisture	4	2,8,8,1	10.23	0-29.1	0.99	0.39	0.58	0.99
Moisture	5	1,8,8,1	10.34	0-29.3	0.99	0.42	0.56	0.99

<sup>a</sup> $R^2$ , determination coefficient of calibration; SEC, standard error of calibration;<sup>b</sup>SECV, standard error of cross-validation; 1-VR, the coefficient of determination in cross-validation<sup>c</sup> Range represent for estimated values between minimum and maximum in the calibration set as determined by a moisture meter

Figure 2 depicts the cross-validation efficiency of NIR values for MC with that of the lab estimated values using the 2,4,4,1 pretreatment. Good correlation between the predicted and

reference MC values of groundnut was achieved. Both NIR absorption and reflection data and their derivatives resulted in  $R^2$  values exceeding 99%, which is at most accurate.



**Fig 2:** Cross-validation plots of the calibration equations for moisture content (n=77)

There are several reports on application of NIR reflectance spectroscopy for analysis of MC in in-shell <sup>[9, 10]</sup> and shelled groundnut <sup>[2]</sup> including different botanical types. Although several rapid non-destructive MC estimating tools such as moisture meter are available, what separates NIRS from other devices is its ability to predict multiple traits in a single scan. For example, at ICRISAT the NIRS is being used for predicting oil, protein and fatty acid concentrations (palmitic, oleic and linoleic acid concentrations) of unknown samples, to be used for making selection decisions and advancing genotypes with desired trait features. In the market and food processing industry, use of equipments such as NIRS instead of moisture meter, for estimating MC of groundnut kernels can be beneficial as it helps them to judge the quality of their produce on different quality parameters, and hence pay a premium price to farmers for produce that fit with their quality guidelines.

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