

Non-Destructive Assessment of Edible Seed Chemical Content by Near-Infrared Hyperspectral Imaging for the Improvement of Nutritional Traits in Bottle Gourd (*Lagenaria Siceraria*)

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ABSTRACT

The edible seeds of bottle gourd [*Lagenaria siceraria* (Molina) Standl.] are rich in oils, proteins and minerals of high nutritional quality. They are highly prized in pan tropical regions where they constitute valuable resources for food and nutrition security. In this study, near-infrared hyperspectral imaging (NIR-HSI) was combined with chemometrics to assess the variability of seed chemical content of African cultivars for the selection of nutritional traits. Six hundred seeds of four accessions belonging to two cultivars were collected from the Ivory Coast (West Africa) and analysed. The NIR-HSI spectra collected on whole seeds in the 1100-2400 nm range revealed that the main absorption bands of the seed chemical content were associated with water, lipids and proteins. The absorbance values between seeds of the same accession in these spectral regions varied up to 1.8 folds. Among the two chemometric tools used, principal component analysis (PCA) did not separate the accessions while Partial Least Squares Discriminant Analysis (PLS-DA) discriminated the accessions with 87.33 % to 94.67 %, and the cultivars with 90 % to 92 % correct classification. Seed oils from bottle gourd are for instance rich in linoleic acid which is an essential fatty acid for human health. The non-destructive and qualitative determination of the content of single seeds was demonstrated in the study and provides the opportunity to select superior seeds for the improvement of key nutritional traits in bottle gourd.

Keyword: *Lagenaria siceraria*, near-infrared hyperspectral imaging, seed chemical content, PCA, PLS-DA, nutrition security

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INTRODUCTION

Bottle gourd (*Lagenaria siceraria*) is a pan-tropical species belonging to the Cucurbitaceae family (Jeffrey, 1990). It is also known as white flower gourd, or simply gourd. Bottle gourd originated from Africa (Decker-Walters et al., 2004; Kistler et al., 2014; N'dri et al., 2016) and spread to pan-tropical regions of the world including Eurasia, Oceania and the Americas. This bi-hemispheric distribution positions the edible types of bottle gourd among the crops ablest to contribute to food security, especially in sub-Saharan Africa where it is widely cultivated and where the edible seeds are highly prized (Zoro et al., 2003; Achigan-Dako et al., 2006; Zoro et al., 2006). Indeed, the oleoproteaginous edible seeds of gourds can contain more than 50 % lipid and 35 % protein (Badifu, 1993; Loukou et al., 2011), as well as

minerals of good nutritional qualities (Ogunbusola et al., 2010; Loukou et al., 2011; Ogundele et al., 2013). Also, detailed analyses have shown that oil from the seeds of bottle gourd is a good source of linoleic acid, an essential fatty acid for humans. The bioavailability of proteins from bottle gourd seeds is higher than the bioavailability of proteins from groundnut (Loukou et al., 2011). Further, the amino acid profile of *L. siceraria* seed protein showed the presence of 18 amino acids among which 11 essential amino acids (Badifu and Ogunsua, 1991; Ogundele and Oshodi, 2010; Loukou et al., 2011). Despite these qualities, the lack of improved varieties is one of the constraints for increased production of bottle gourd, especially for large-scale or industrial exploitation of the seeds for food uses. The variability for seed

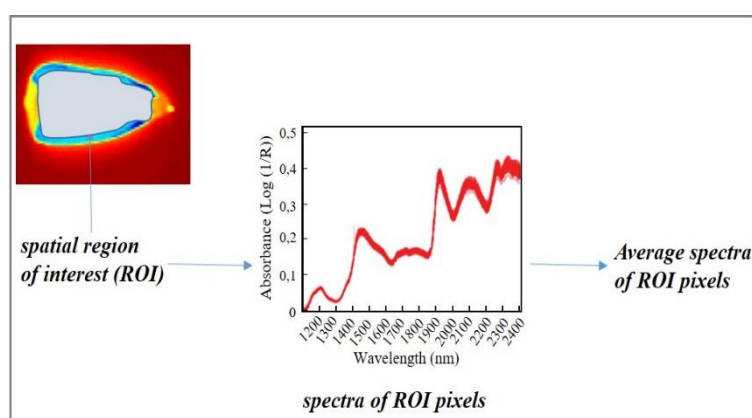


Figure 1. Extraction of spectra from NIR hyperspectral image of a seed of bottle gourd showing spatial region of interest (ROI) and average raw spectra.

chemical content of bottle gourd has been studied previously at the population levels using chemical methods of bulk seeds (Fokou et al., 2009; Ogundele and Oshodi, 2010; Loukou et al., 2011; 2012). Although efficient, these methods do not capture the variability of chemical content at the single seed level where the genetic variability can be significant in landraces. Indeed, chemical methods are destructive and do not allow the selection of the best individual seeds within an accession. A single seed of bottle gourd can produce thousands of seeds when sown, therefore providing the opportunity to capture the variability within an accession and to select the best progenies from single seeds of landraces for the improvement of nutritional traits. In this study, near-infrared hyperspectral imaging (NIR-HSI) was used to assess the intra and inter variability of landrace accessions for seed chemical content, with the objective of finding effective tools for the assessment and the improvement of seed nutritional qualities in bottle gourd.

The conventional near-infrared spectroscopy (NIRS) is based on the vibration properties (overtones and combination vibrations) of major X-H chemical bonds within organic molecules (C–H, O–H and N–H) and their interactions with infrared radiation. Thus, NIRS spectroscopy methods can be used to capture the spectral information which allows the assessment of the chemical content of intact biological materials. Near-infrared hyperspectral imaging (NIR-HSI) is a non-destructive technique that combines the conventional NIR spectroscopy and imaging techniques to obtain both spectral and spatial information from a sample (Chang, 2003; Baeten et al., 2010; Manley, 2014), thus, allowing the analysis of the chemical content of different parts of the same biological materials such as single seeds.

MATERIAL AND METHODS

Plant materials

Six hundred intact and healthy seeds belonging to two cultivars (round and oval fruits) of the edible oleoproteaginous seed type of bottle gourd were

collected from farmers in the localities of Korhogo (9° 23' N, 5° 33' W) and Manfla (7° 30' N, and 5° 53'W) situated respectively in northern and Midwest of Ivory Coast (West Africa). These seed lots consisted of four accessions (150 seeds per accession) including two accessions of the oval fruit cultivar LS-BAK (collected at Korhogo) and LS-BAM (collected at Manfla) and two accessions of the round fruit cultivar LS-BRK (collected at Korhogo) and LS-BRM (collected at Manfla). Each seed was labeled with a number before the spectrometric analysis in order to be linked to its own chemical content data.

NIR hyperspectral image acquisition and data analysis

NIR hyperspectral images were acquired using a push-broom imaging system (Burger Metrics SIA, Riga, Latvia). The instrument used is a SWIR ImSpector N25E HSI system (Specim Ltd) that contains a cool, temperature-stabilized Mercury-Cadmium-Telluride (MCT) detector (Xenics), combined with a conveyor belt (Burger metrics). The parameters used were described by Vermeulen et al. (2012). The NIR hyperspectral images were acquired from each seed at 209 wavelength channels in the 1100–2400 nm range, with an interval of 6.3 nm between channels. Each image consisted of 320-pixel lines and the image acquisition time was 0.05 sec/frame, translating to 20 frames/sec, or 25 sec/image. Before performing the seed scanning, the spectral imaging system was calibrated with a dark image and a white image to compensate for offset due to the dark current, the light source temperature drift and the lack of spatial lighting uniformity (Vermeulen et al., 2012). The dark image was obtained by blocking the lens entrance and the white image (background) was collected from a standard white reference board (empty Teflon plate) as described by Vermeulen et al. (2012). The spectra were then automatically corrected accordingly. Absorbance values were obtained by converting reflectance values.

About 1000 pixels extracted from the hyperspectral image of the regions of interest of each seed were used to produce the raw spectra of each seed (Figure 1) and

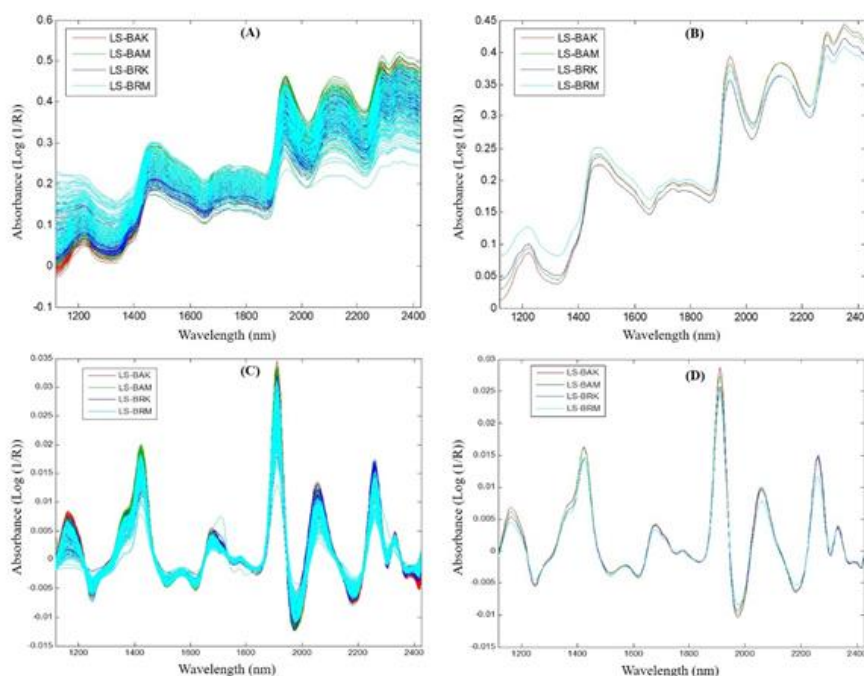


Figure 2. NIR-HSI spectra of bottle gourd seeds: **(A)** raw spectra of 600 seeds: **(B)** raw spectra of the average of 150 seeds per accession (LS-BAK, LS-BAM, LS-BRK and LS-BRM): **(C)** pre-processed spectra of 600 seeds: **(D)** pre-processed spectra of the average of 150 seeds per accession.

averaged. The average spectra of the 150 seeds of each accession (one spectrum per seed) were grouped to constitute the average spectrum of the accession. The absorption bands identified from the spectra were then assigned to specific functional groups and components according to the absorption table of Osborne et al. (1993) and assignment bands from some previous studies (Cho and Iwamoto, 1989; Hourant et al., 2000; Westad et al., 2008). Standard normal variate (SNV) and first derivatives were used as pre-treatment methods to remove unwanted information such as additive and multiplicate effects on the spectral measurement and to remove the baseline effect (Barnes et al., 1989; Heise and Winzen, 2002; Gowen et al., 2007; Manley, 2014). The pre-processing spectra were used in all the subsequent calculations. Principal component analysis (PCA) and the partial least squares discriminant analysis (PLS-DA) were tested for the classification of the accessions and the cultivars based on their spectral profiles. The PLS-DA was fit using the measurements at the 209 wavelengths mentioned earlier. To estimate the performance of PLS-DA to classify bottle gourd accessions, a confusion matrix was generated with the training set in a cross-validation method using the leave-one-out cross-validation (a version of k-fold cross-validation). All the analyses of this study were carried out using Matlab v 6.5 R13 software (MathWorks, Natick, MA, USA).

RESULTS AND DISCUSSION

Analysis of seed spectra and the chemical assignment of absorption bands

The raw spectra of 600 seeds are shown in Figure 2A. The data obtained on each seed within the same accession were grouped to obtain the average spectrum of the accession (Figure 2B). As expected, all the averaged spectra exhibited the same profiles, meaning that the seeds of the four accessions contain the same types of chemical content (in terms of presence/absence). Differences were observed in the levels of absorbance for specific absorption bands which relates generally to differences in the quantity of chemical content detected (raw and pre-processed spectrum) (Figure 2). In these spectral regions, the level of absorbance between the seeds of the same accession (within accession) varied up to 1.8 folds. These results could indicate a quantitative variability of the chemical content of seeds within the same accession. This must be confirmed by further chemical analyses.

As indicated in Figure 2, the main absorption bands of whole seeds of bottle gourd are observed in the following NIR regions: 1150-1250 nm, 1430-1480 nm, 1910-1960 nm, 2080-2180 nm, 2259-2297 nm and 2320-2366 nm. The bands associated with fatty acids and lipids are primarily due to the vibration modes of the C-H bonds attributable to three functional groups: $-\text{CH}_2$ (methylene), $-\text{CH}_3$ (methyl) and $-\text{CH}=\text{CH}-$ (ethylene) (Workman, 1996; Hourant et al., 2000; Westad et al., 2008). These functional groups are associated with the regions 1150-1250 nm, 1430-1480 nm, 2259-2297 nm and 2320-2366 nm of the absorption bands of the analysed seed (Figure 2A). The region around 1160 nm (band ranging from 1150-1250 nm) is due to the second overtone of elongation vibration of the C-H bond of the functional groups $-\text{HC}=\text{CH}-$. Similar results were obtained for peanut with an absorption peak at 1162 nm (Fox and

Cruickshank, 2005) and canola seed at 1164 nm (Westad et al., 2008). The absorption band between 1160-1250 nm observed in the NIR spectra of *L. siceraria* is due to the absorption of the ethylene functional group, which indicates the presence of unsaturated fatty acids (Cho and Iwamoto, 1989, Hourant et al., 2000). Unsaturated fatty acids of bottle gourd seeds are mainly linoleic acid and oleic acid (Loukou et al., 2011). Linoleic acid is a polyunsaturated omega-6 fatty acid which is one of the essential fatty acids for humans (cannot be synthesized by the human body and must be obtained through human diet), and oleic acid which is a monounsaturated fatty acid classified as an omega-9 fatty acid. The polyunsaturated linolenic acid which is an essential omega-3 fatty acid represents a very small proportion of bottle gourd seed oils. In addition, previous studies have shown a relationship between the 1150-1250 nm region and the presence of unsaturated fatty acids. A band was identified around 1180 nm in pure fatty acid spectra containing cis double bonds (Sato et al., 1991). In the studies of Hourant et al. (2000), oils rich in polyunsaturated and monounsaturated fatty acids showed higher absorption than saturated fatty acids around 1164 nm. The regions 2259-2297 nm and 2320-2366 nm correspond to the combination of the C-H stretching vibration and bending modes of methyl (-CH₃) and methylene (-CH₂) functional groups associated with oil (Osborn, 1993; Hourant et al., 2000; Westad et al., 2008). The bands ranging from 2080-2180 nm with a maximum around 2110 nm are due to the stretching vibration of the N-H bond of the CONH₂ and CONHR functional groups associated with the proteins. The region around 1450 nm (band 1430-1480 nm) is attributed to O-H first overtone stretching vibration bound to water and the band from 1910-1960 nm also associated with the absorption of water is due to the combination of O-H stretching vibration and deformation. Main absorption bands of spectra were attributed to water, lipids and proteins. These results are consistent with previous studies on the chemical composition of *L. siceraria* seeds, which showed that lipids and proteins are the major constituents (Badifu and Ogunsua, 1991; Badifu, 1993; Loukou et al., 2011). The bands (1430-1480 nm and 1910-1960 nm) attributed to water represent moisture content since water is present even in dry bottle gourd seeds (Ogundele and Oshodi, 2010). Due to the fact that each of the seed chemical content (water, lipids or proteins) can be detected at different spectral locations in the 1100-2400 nm range, one of the contributions of this study is that it provided specific spectral locations at which to detect and to conduct non-destructive measurement of the main seed chemical content of bottle gourd, thereby reducing the number of wavelengths to screen in future studies.

Hyperspectral data are usually affected by many factors such as light scattering which may cause baseline variation and multiplicative effects on spectral data. These additional effects on data are not useful for the chemical characterization of the sample and can be considered as noise with respect to the data interpretation (or modelling). Hence, SNV and first-derivative pre-processing techniques were used on the raw spectra extracted from the images to reduce noise

and to remove any irrelevant information, which resulted in the spectra presented in Figures 2C and 2D. These pre-processing techniques have reduced the differences between the four accessions and similar profiles were obtained also for all seeds and all accessions. Chemometric tools were then employed on pre-processed spectra for the discrimination of the four accessions.

Discrimination of the bottle gourd accessions by PCA and PLS-DA

Principal component analysis (PCA) was performed on the pre-processing spectra to evaluate the qualitative variability within and between the accessions. Figure 3 shows the scores plot of the first two principal components (PCs) of the NIR-HSI spectra from four accessions. There was no clear separation between the four accessions using PCA, meaning that individuals composing the four accessions of bottle gourd share as expected some similarities in chemical content composition. The first two PCs explained 93.48 % of the variation between the accessions, with 72.58 % and 20.90 % of the variation for PC1 and PC2 respectively. In PCA plots, samples located far from each other may indicate different patterns of variables in general. The inspection of the score plot showed a spread of seed of the same accession (Figure 3), pointing at differences in seed content within each accession. The intra accession variations in the absorbance levels (up to 1.8 folds) detected in these samples of landraces may also support the intra accession variability in the quantity of seed content.

The loadings plot (Figure 4) of the first two PCs showed the principal wavelengths responsible for the sample distribution. The variations exhibited by PC 1 are mainly due to the wavelengths around 1150 nm (C-H stretching vibrations, second overtone), 1900 nm (O-H stretching and deformation combination mode), 2000 nm (N-H stretching vibrations and amide II), 2246 nm (N-H stretching and NH₃⁺ deformation) associated to lipids, water, proteins and amino acids respectively. The loading line plot of PC 2 indicated variations corresponding also to lipids, water and proteins. The PC 2 showed an absorption peak at 1326 nm (2 x C-H stretching vibrations and C-H deformation), 1900 nm and 1950 nm (O-H stretching and deformation combination mode), 2183 nm (2 x amide I + amide III combination mode), 2246 nm (N-H stretching and NH₃⁺ deformation), 2290 nm (N-H and C=O stretching), 2322 nm (C-H stretching vibrations).

The pre-processing spectra were subjected to the PLS-DA to classify the seeds according to their respective accessions. Table 1 summarizes the performance of PLS-DA for the inter-accession variability. The confusion matrix allows the visualization of the performance of an applied algorithm (here PLS-DA). When the PLS-DA model is applied to the training set, 90 % to 96 % of the seeds are correctly classified in their respective accessions. Applying cross-validation, seeds from the three accessions LS-BAK, LS-BAM and LS-BRM were correctly identified as belonging to their respective accessions with more than 90 % of correct classification,

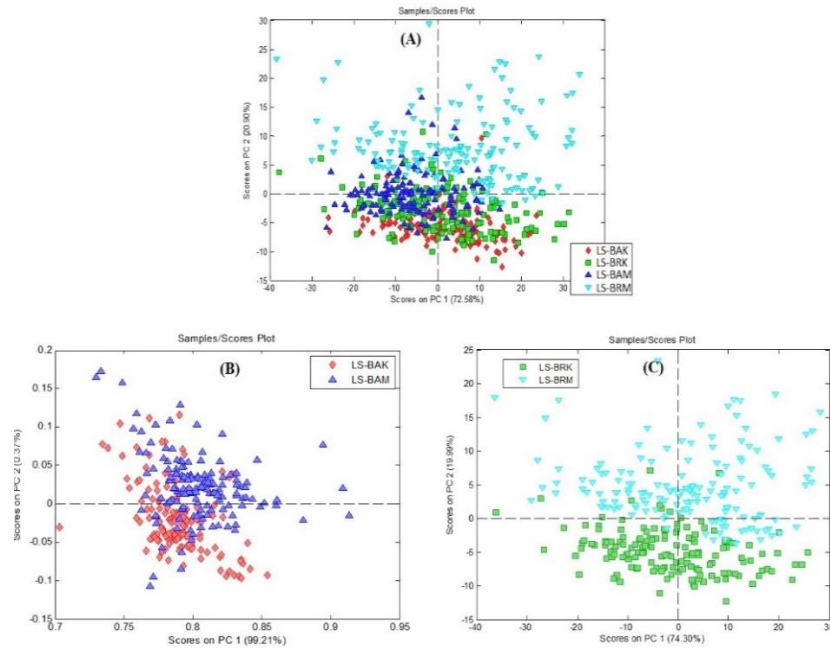


Figure 3. PC1 $\times$ PC2 score plot of (A) four accessions (LS-BAK, LS-BAM, LS-BRK and LS-BRM), (B) oval fruit cultivars and (C) round fruit cultivars. Each accession contains 150 seeds of oleoproteagenous edible seeds of *L. siceraria*. The data used are NIR-HSI pre-processed spectra (SNV and first-derivative) in the spectral range of 1100–2400 nm.

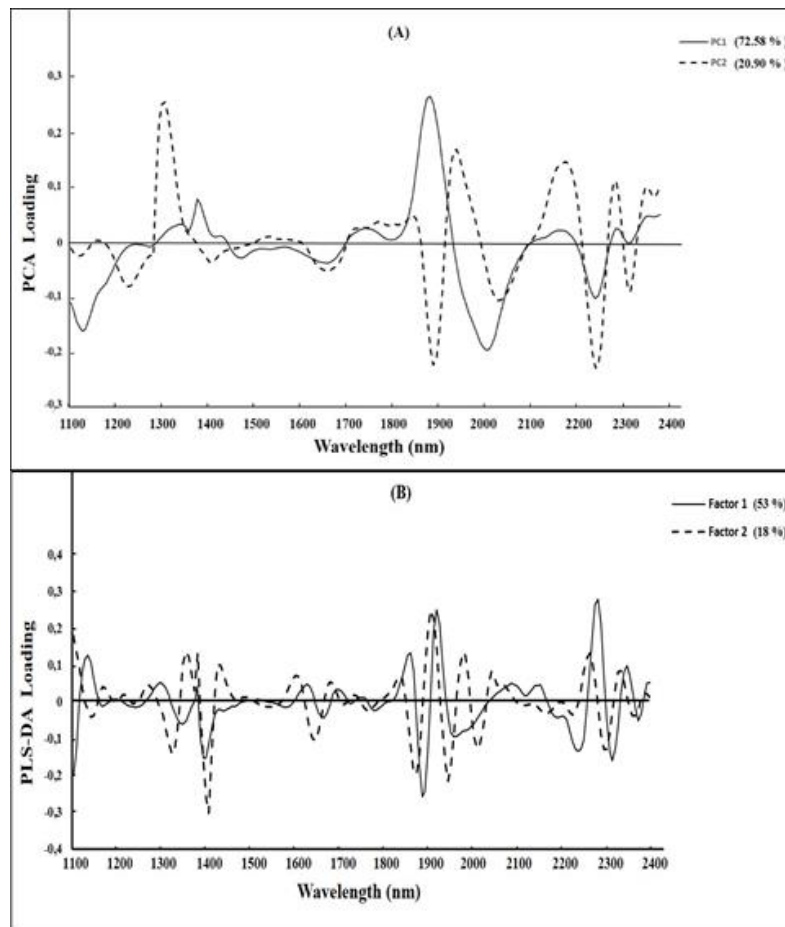


Figure 4. (A) PCA loading line plot of PC1 and PC2, and (B) PLS-DA loading line plot of Factor 1 and Factor 2 generated with the NIR spectra of oleoproteagenous edible seeds of *L. siceraria*. The spectral regions contributing the most to the variation are captured by the importance of the height of peaks and valleys at the corresponding wavelengths.

Table 1. Confusion matrix from Partial Least Squares-Discriminant Analysis (PLS-DA) for the classification of bottle gourd seed accessions using near infrared hyperspectral imaging data

Predicted class (%)	Accessions	Actual class				Cross-validation			
		Training set							
		LS-BAK	LS-BAM	LS-BRK	LS-BRM	LS-BAK	LS-BAM	LS-BRK	LS-BRM
	LS-BAK	95.34	1.33	0.67	1.33	94.67	1.33	0.67	2.67
	LS-BAM	1.33	93.34	7.33	2	3.33	92.00	10	2.67
	LS-BRK	3.33	4	90	0.67	2	4	87.33	0.66
	LS-BRM	0	1.33	2	96	0	2.67	2	94.00

Table 2. Confusion matrix from Partial Least Squares-Discriminant Analysis (PLS-DA) for the classification of bottle gourd cultivars using near infrared hyperspectral imaging data.

Predicted class (%)	Cultivars	Actual class		Cross-validation	
		Training set			
		Round fruits	Oval fruits	Round fruits	Oval fruits
	Round fruits	93	9	92	10
	Oval fruits	7	91	8	90

while 87.33 % of LS-BRK seeds were correctly classified as belonging to that accession. Thus overall, the PLS-DA showed a good performance to classify *L. siceraria* accession with a classification accuracy of 87.33 % to 94.67 %. Table 2 presents the results of the PLS-DA through the confusion matrix obtained with NIR-HSI data of *L. siceraria* round fruit cultivars versus the oval fruit cultivars. The results indicated that 7 % of round fruit cultivar seeds are misclassified as seeds of the oval fruit cultivars, and 9 % of the oval fruit cultivar seeds are incorrectly classified by PLS-DA as the seeds of the round fruit cultivars. The cross-validation results (Table 2) showed that the seeds of the two cultivars were discriminated with an average of 91 % correct classification (90 % and 92 % averaged).

To identify the wavelengths responsible for the discrimination between accessions, all the factors explaining at least 90 % of the total variation were used. The first two factors explained 53 and 18% of the total variation respectively (Figure 4). The main wavelengths identified in this study are primarily found in the ranges of 1380-1452 nm, 1850-2020 nm and 2227-2325 nm. These spectral regions are related to lipids, water and proteins.

The variability among accessions based on seed chemical content composition can have various origins. It may be related to genetic factors, fruit maturation, environmental conditions and/or post-harvest treatments. However, in this work, the intra and inter accession variations are due mostly to genetic variations as the fruits of all accessions were harvested mature, underwent the same treatments (post-harvest treatments), and accessions of the same regions were exposed to the same environmental factors. The variation in seed chemical content has previously been assessed on bulk seeds of bottle gourd using chemical analysis methods such as GC, HPLC, Kjeldahl, Lowry and hexane extraction (Fokou et al., 2009; Ogundele and Oshodi, 2010; Loukou et al., 2011). To our knowledge, the present study is the first non-destructive single seed

assessment of intra and inter accessions variability of seed chemical content combined with the identification of specific spectral absorption bands of the seed chemical content in bottle gourd. This approach constitutes a major step towards the development of selection schemes for the improvement of nutritional traits in edible seeds of bottle gourd. The success of breeding programs depends largely on the availability of genetic variation and the efficacy of evaluating large numbers of genotypes. The NIR-HSI spectroscopy is adapted to assessing the chemical variability of single seed content which may be the result of genetic variation. Thus, this approach is suitable for screening certain aspects of genetic diversity.

The results of this study showed that among the two chemometric methods used, NIR-HSI coupled with PLS-DA is adapted for the assessment of inter accession variability in bottle gourd. At the intra accession level, the spread of individual seeds in the PCA score plots and the intra accession variations of the absorbance levels point to possible differences in seed chemical content within the same accession. These approaches are therefore adapted to the collection and management of genetic resources and the improvement of nutritional traits in bottle gourd.

CONCLUSION

Africa is the center of origin of bottle gourd (*L. siceraria*). Thus, unraveling and using the genetic diversity of African landraces expressed through the variation of seed chemical content is of prime importance for the improvement of nutritional traits in bottle gourd of Africa, Eurasia, Oceania and the Americas. This study conducted on seeds extracted from four accessions belonging to two African cultivars showed that near-infrared hyperspectral imaging (NIR-HSI) combined with chemometrics can be used for the classification of accessions and seeds of bottle gourd based on their

chemical content. The study showed that PLS-DA is particularly adapted for the discrimination of gourd seeds according to their accessions of origin with 87.33 % to 94.67 % ; and the cultivars with 90 % to 92 % exact classification of the seeds according to the accessions. The PCA did not separate the accessions but showed a spread of individuals in the score plots. This, together with the absorbance differences observed between seeds of the same accession indicate likely quantitative variability in seed content within the same accession. Further analyses such as the calibration and quantitative assessment of seed chemical content will be however needed to confirm the intra accession variability. The finding of specific spectral regions which constitute the optimal absorption bands of the major nutritional reserves of the seed (lipids and proteins) is a desirable outcome that will reduce the number of NIR spectral regions and absorption bands analysed in subsequent studies. The importance of the non-destructive nature of the study resides in the fact that it allows the assessment of the variability of seed lipid and protein content and the use of the best accessions and the best seeds for subsequent cycles of varietal selection for nutritional content, thereby increasing the efficiency of breeding for nutritional traits. Although this was a qualitative analysis, intra and inter accession differences observed in the levels of absorbance of the spectra for lipids, proteins and water indicate variations in the quantity of the chemical content according to the Beer-Lambert law (linear relationship between the concentration and the absorbance). This can be further confirmed through a calibration for a non-destructive and quantitative evaluation of the seed content to improve the accuracy of the selection for nutritional qualities.

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REFERENCES

- Achigan-Dako GE, Fanou N, Kouke A, Avohou H, Vodouhe SR, Ahanché A (2006). Agronomic evaluation of three species of Egusi (Cucurbitaceae) used for food in Benin and development of a yield prediction model. *Biotechnol. Agron. Soc. About.*, 10: 121–129.
- Badifu GIO (1993). Food potentials of some unconventional oilseeds grown in Nigeria: a brief review. *Plant Foods Hum. Nutr.*, 43: 211–224.
- Badifu GIO, Ogunsua AO (1991). Chemical composition of kernels from some species of *Cucurbitaceae* grown in Nigeria. *Plant Foods Hum. Nutr.*, 41: 35–44.
- Baeten V, Fernandez Pierna JA, Vermeulen P, Dardenne P (2010). NIR hyperspectral imaging methods for quality and safety control of food and feed products: Contributions to four European projects. *NIR News* 21: 10–13.
- Barnes JR, Dhanoa SM, Lister JS (1989). Standard normal variate transformation and detrending of near-infrared diffuse reflectance spectra. *Appl. Spectrosc.*, 43: 772–777.
- Chang CI (2003). Hyperspectral imaging: techniques for spectral detection and classification. Springer Sci. Business Media, 1:370 p.
- Cho RK, Iwamoto M (1989). The purity identification of sesame oil by near-infrared reflectance spectroscopy. *Proceedings of the 2nd International NIRS conference*. Tsukuba, Japan. 142–147 pp.
- Decker-Walters DS, Wilkins-Ellert M, Chung S-M, Staub JE (2004). Discovery and genetic assessment of wild bottle gourd [*Lagenaria siceraria* (Mol.) Standley; Cucurbitaceae] from Zimbabwe. *Econ. Bot.*, 58: 501–508.
- Fokou E, Achu MB, Kansci G, Ponka R, Fotso M, Tchiegang C, Tchouanguep FM (2009). Chemical properties of some Cucurbitaceae oils from Cameroon. *Pak. J. Nutr.*, 8 (9): 1325–1334.
- Fox GP, Cruickshank A (2005). Near infrared reflectance as a rapid and inexpensive surrogate measure for fatty acid composition and oil content in peanuts (*Arachis hypogaea* L.). *J. Near Infrared Spectrosc.*, 13: 285–291.
- Gowen AA, O'Donnell PC, Cullen JP, Downey G, Frias JM (2007). Hyperspectral imaging an emerging process analytical tool for food quality and safety control. *Trends Food Sci. Technol.*, 18: 590–598.
- Heise HM, Winzen R (2002). Chemometrics in near-infrared spectroscopy. In: *Near-infrared spectroscopy: Principles, instruments, applications*. Siesler HW, Ozaki Y, Kawata S, Heise HM (Eds). Weinheim, Germany: Wiley-VCH Verlag GmbH; pp. 25–161.
- Hourant P, Baeten V, Morales M, Meurens M, Aparicio R (2000). Oil and fat classification by selected bands of near-infrared spectroscopy. *Appl. Spectrosc.*, 54: 1168–1174.
- Jeffrey C (1990). Systematics of the *Cucurbitaceae*: An overview, In: *Biology and utilization of the Cucurbitaceae*. Bates DM, Robinson RW and C Jeffrey (Eds.). Cornell University Press, Ithaca, pp: 3–28.
- Kistler L, Montenegro A, Smith BD, Gifford JA, Greene RE, Newsoma LA, Shapiro B (2014). Transoceanic drift and the domestication of African bottle gourd in the Americas. *Proc. Natl. Acad. Sci.*, 111: 2937–2941.
- Loukou AL, Lognay G, Barthelemy JP, Maesen P, Baudoin JP, Zoro BIA (2011). Effect of harvest time on seed oil and protein content and compositions in the oleaginous gourd *Lagenaria siceraria* (Molina) Standl. *J. Sci. Food Agric.*, 91: 2073–2080.
- Loukou AL, Lognay G, Baudoin JP, Kouamé LP, Zoro BIA (2012). Effects of fruit maturity on oxidative stability of *Lagenaria siceraria* (Molina) Standl Seed oil extracted with hexane. *J. Food Biochem.*, 37: 475–484.
- Manley M (2014). Near-infrared spectroscopy and hyperspectral imaging: non-destructive analysis of biological materials. *Chem. Soc. Rev.*, 43: 8200–8214.
- N'dri ANA, Zoro BIA, Kouame LP, Dumet D, Vroh-Bi I (2016). On the dispersal of bottle gourd [*Lagenaria siceraria* (Mol.) Standl.] out of Africa: a contribution from the analysis of nuclear ribosomal DNA haplotypes, divergent paralogs and variants of 5.8S protein sequences. *Plant Mol. Biol. Rep.*, 34: 454–466.
- Ogunbusola EM, Fagbemi NT, Osundahunsi FO (2010). Amino acid composition of *Lagenaria siceraria* seed flour and protein fractions. *J. Food Sci. Technol.*, 47: 656–661.
- Ogundele JO, Oshodi AA. 2010. Proximate composition and some functional properties of three varieties of *Lagenaria siceraria* melon seeds. *Res. J. Agric. Biol. Sci.*, 6: 108–112.
- Ogundele JO, Oshodi, AA, Fakunle CO, Oyebanji AO, Amoo IA (2013). Evaluation and comparison of the amino acid composition of three varieties of *Lagenaria siceraria* melon seed flours. *Food Sci. Qual. Manage.*, 15: 36–42.
- Osborne BG, Fearn T, Hindle PH (1993). *Practical NIR spectroscopy with applications in food and beverage analysis*. Essex, England: Longman Scientific & Technical, Harlow, 2nd edn, pp: 29–33.
- Sato T, Kawano S, Iwamoto M. 1991. Near infrared spectral patterns of fatty acid analysis from fats and oils. *J. Am. Oil Chem. Soc.*, 68: 827–863.
- Vermeulen P, Fernandez Pierna JA, Egmond VPH, Dardenne P, Baeten V (2012). Online detection and quantification of ergot bodies in cereals using near infrared hyperspectral imaging. *Food Addit. Contam.*, 29: 232–240.
- Westad F, Schmidt A, Kermit M (2008). Incorporating chemical band-assignment in near infrared spectroscopy regression models. *J. Near Infrared Spectrosc.*, 16: 265–273.
- Workman J (1996). Interpretative spectroscopy for near infrared. *Near infrared spectroscopy: the future waves*, A.M.C. Davies and Phill Williams, NIR Publication, Chichester, West Sussex, UK.
- Zoro BIA, Koff KK, Dje Y, Malice M, Baudoin JP (2006). Indigenous cucurbits of Cote d'Ivoire: a review of their genetic resources. *Sci. Nat.*, 3: 1–9.
- Zoro BIA, Koffi KK, Dje Y (2003). Botanical and agronomic characterization of three species of cucurbits eaten in sauce in West Africa: *Citrullus* sp, *Cucumeropsis mannii* Naudin and *Lagenaria siceraria* (Molina) Standl. *Biotechnol. Agron. Soc. About.*, 7: 189–199.