

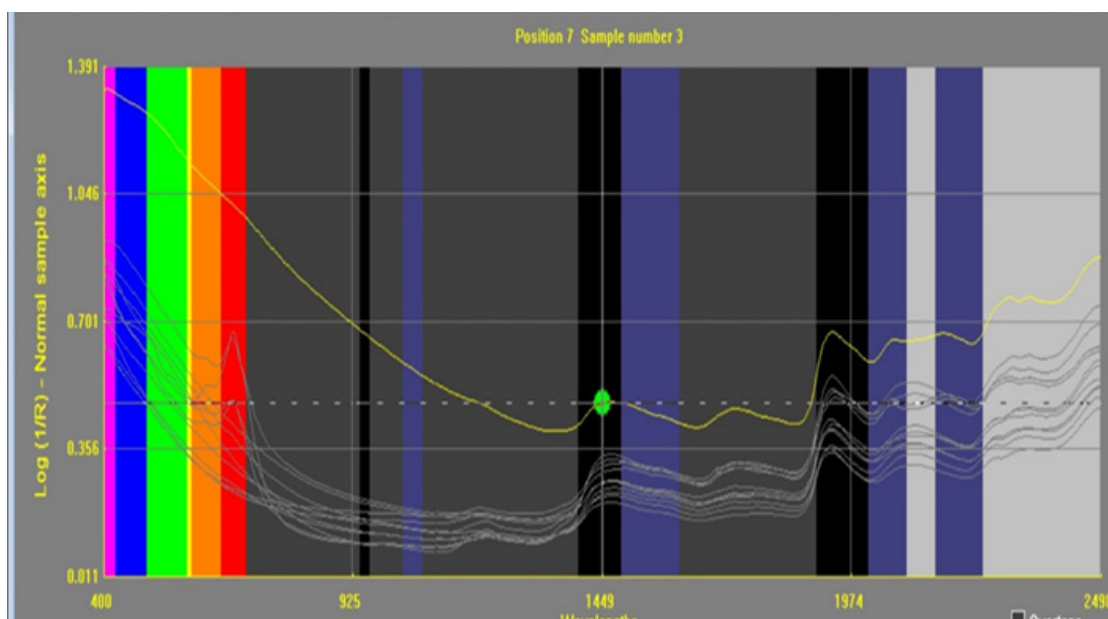
# Training report

## Advanced training on NIRS and build CGIAR-NARES forage and fodder crops breeding networks for common data management and exchange

Bayissa Hatew<sup>1</sup>, Prasad KVS<sup>2</sup>, Khadija Aouzal<sup>3</sup> and Chris S. Jones<sup>4</sup>

1. International Livestock Research Institute, Addis Ababa, Ethiopia
2. International Livestock Research Institute, Hyderabad, India
3. International Center for Agricultural Research in the Dry Areas, Morocco
4. International Livestock Research Institute, Nairobi, Kenya

November 2024





# ILRI

INTERNATIONAL  
LIVESTOCK RESEARCH  
INSTITUTE




©2024 International Livestock Research Institute (ILRI)

ILRI thanks all donors and organizations which globally support its work through their contributions to the [CGIAR Trust Fund](#)



This publication is copyrighted by the International Livestock Research Institute (ILRI). It is licensed for use under the Creative Commons Attribution 4.0 International Licence. To view this licence, visit <https://creativecommons.org/licenses/by/4.0>.

Unless otherwise noted, you are free to share (copy and redistribute the material in any medium or format), adapt (remix, transform, and build upon the material) for any purpose, even commercially, under the following conditions:

 **ATTRIBUTION.** The work must be attributed, but not in any way that suggests endorsement by ILRI or the author(s).

NOTICE:

For any reuse or distribution, the licence terms of this work must be made clear to others.

Any of the above conditions can be waived if permission is obtained from the copyright holder.

Nothing in this licence impairs or restricts the author's moral rights.

Fair dealing and other rights are in no way affected by the above.

The parts used must not misrepresent the meaning of the publication.

ILRI would appreciate being sent a copy of any materials in which text, photos etc. have been used.

Editing, design and layout—ILRI Editorial and Publishing Services, Addis Ababa, Ethiopia.

All photos by ILRI/Prasad K.V.S.V.

Citation: Hatew, B., Prasad, K.V.S.V., Aouzal, K. and Jones, C.S. 2024. *Advanced training on NIRS and build CGIAR-NARES forage and fodder crops breeding networks for common data management and exchange: Training report*. Nairobi, Kenya: International Livestock Research Institute (ILRI).

*Patron: Professor Peter C. Doherty AC, FAA, FRS*

*Animal scientist, Nobel Prize Laureate for Physiology or Medicine–1996*

Box 30709, Nairobi 00100 Kenya  
Phone +254 20 422 3000  
Fax +254 20 422 3001  
Email [ilri-kenya@cgiar.org](mailto:ilri-kenya@cgiar.org)

[ilri.org](http://ilri.org)  
*better lives, better planet through livestock*

ILRI is a CGIAR research centre

Box 5689, Addis Ababa, Ethiopia  
Phone +251 11 617 2000  
Fax +251 11 667 6923  
Email [ilri-ethiopia@cgiar.org](mailto:ilri-ethiopia@cgiar.org)

*ILRI has offices in East Africa • South Asia • Southeast and East Asia • Southern Africa • West Africa*

# Contents

Acknowledgements	iv
Background	1
Objectives	2
Participants	3
Content of the training	4
Data management and calibration	6
Gaps identified and opportunities	7
Conclusion	8
Training agenda	9

# Acknowledgements

We would like to thank all who contributed to, and helped in facilitating, the training workshop. This training was conducted by the International Livestock Research Institute (ILRI) in collaboration with the Alliance Bioversity and CIAT and the International Center for Agricultural Research in the Dry Areas (ICARDA) under the Crops to End Hunger (CtEH) project 'Improved fodder crops and forage grasses for Africa'. CtEH is a CGIAR Initiative to accelerate and modernize the development, delivery and widescale use of a steady stream of new crop varieties. We also would like to thank CtEH funders and partners. Any opinions, conclusions or recommendations expressed here are those of the authors alone.

# Background

As a follow up to the first training on the use and application of near-infrared reflectance spectroscopy (NIRS) (<https://hdl.handle.net/10568/151969>) an advanced training on NIRS technology was conducted with the following objectives.

# Objectives

1. To calibrate the FOSS DS2500 NIRS machine installed at the International Center for Agricultural Research in the Dry Areas (ICARDA), Rabat, and standardize the instrument to ILRI Hyderabad master NIRS for predicting quality traits using International Livestock Research Institute (ILRI)-developed global calibration models.
2. To train ICARDA and the International Institute of Tropical Agriculture (CIAT) NIRS team on the machine to scan, spectral selection, and export to WinISI 4.10, trim and standardize the spectra to the master NIRS for analyzing the samples using ILRI global calibration models.
3. To discuss the next step on NIRS uses in ICARDA and maintain the platform by adding wet chemistries periodically, and to expand the global models for more robust calibrations for the harmonization of the ILRI NIRS network and its platform use.

# Participants

Key livestock, forage and feed experts, researchers, analytical and technical staff from CGIAR centres and national partners of the three CGIAR centres (ILRI, the Alliance Bioversity and CIAT, and ICARDA) came from Colombia, Ethiopia, Kenya, Morocco, Senegal, Tunisia and Zambia (6 men and 5 women) met at the ICARDA Rabat campus for NIRS technology advanced training and its applications in feed and forage evaluation.

# Content of the training

ILRI, in collaboration with ICARDA and the Alliance Bioversity and CIAT, hosted a four-day training that involved presentations/lectures and demonstrations, and practical sessions that emphasized on the principles of NIRS instrumentation (both benchtop and handheld), the use and application of NIRS, data collection and processing, data management, modelling, calibration equation development and validation and monitoring calibration equations. The training also covered some forage mill maintenance and troubleshooting sessions intended to address problems that users are experiencing within their own applications.

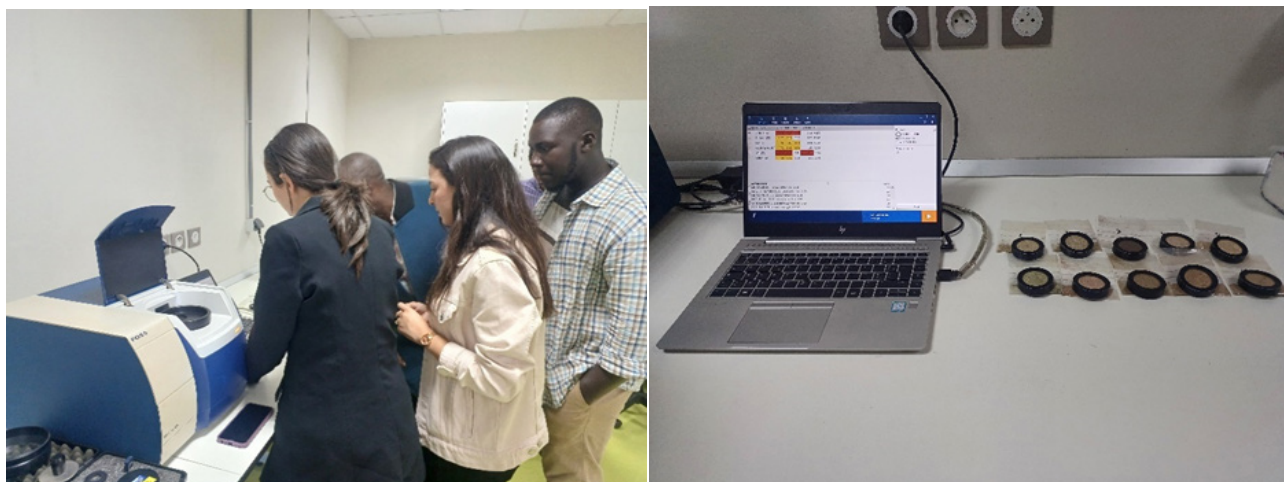
The training workshop specifically covered:

- A revisit of the electromagnetic spectrum (EMS) which is also a parental line for NIRS technology and its theoretical applications.



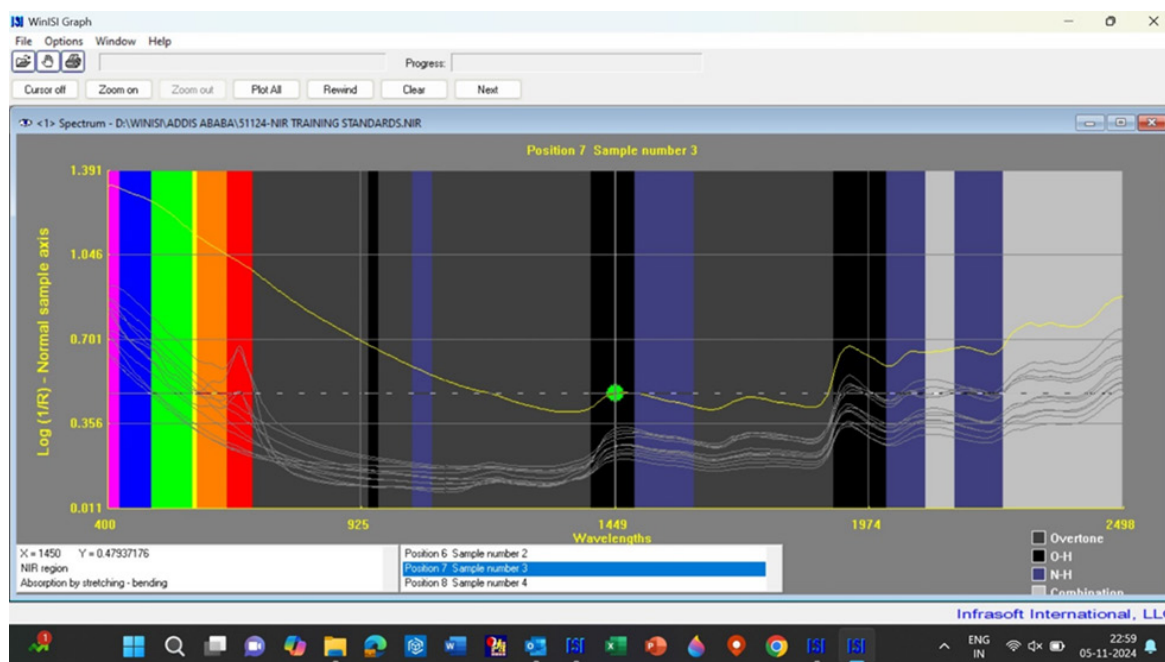
*Trainees following FOSS NIRS advanced training with chemometric modelling (photo credit: ILRI/Prasad K.V.S.V.).*

- NIRS technology advancements and their applications in feed and forage analyses and other allied areas of science supported by Powerpoint and video presentations.
- Demonstration of the most widely used NIRS software's with latest versions of ISIScan and WinISI calibration software for similar make of machines or different makes with spectral transformation techniques.
- Creation of the huge spectral library database for all available food-feed crops across countries and continental bases to export them to the NIRS network.
- Population structuring of the huge spectral database signatures with 2 and 3 dimensional structures to detect the spectral variance components for debugging the unwanted errors and noise.



ICARDA NIRS standardization and proficiency testing by ILRI feed standards (photo credit: ILRI/Prasad K.V.S.V.).

- Development of principal component analysis using the Mahalanobis (GH&NH) distances and fine tuning with available reference data from chemistry labs with conventional wet chemistry facilities, or high-throughput instrumentation laboratories.
- Selecting samples for wet chemical laboratory procedures to see the matrices compositions of the selected sample sets using the spectral selection algorithms.
- Conversion of the spectral files into calibration files, thereby to achieve good and robust calibration models by using the different regression models of Multi-Protocol Label Switching (MPLS), partial least squares (PLS) and principal component analysis (PCA) methods for future unknown sample predictions.
- Validation of unknown samples using a monitor results approach and the expansion of calibration models with new set of samples as and when they are received to see the chemical variance inclusions in calibration models.
- Standardization of the NIRS instruments and creating neural networks linking inherently compatible NIRS instruments for sharing of calibration models between same makes and different makes.



Ring test samples scanned on ICARDA FOSS DS2500 (host) stationary NIRS (photo credit: ILRI/Prasad K.V.S.V.).

# Data management and calibration

In addition, detailed and step-by-step training on data management and calibration was provided based on tools developed by ICARDA. In the beginning, an introduction on a general NIRS data management pipeline was given, and then the three phases of this pipeline were explained one by one. These phases are:

1. Proper data collection: data to be collected includes metadata, NIRS and wet chemistry data.
2. Storing data in an online and centralized database.
3. Running multivariate data analysis. This includes data pre-processing, modeling and predicting.

It was highlighted that data should be stored in a secured centralized database which is crucial so that the user can easily access the data, share and feed it programmatically into their analysis pipeline. For NIRS data extraction, a set of application programming interface (API) functions has been developed, and this helps to extract the metadata, wet chemistry data and NIRS data from the centralized database. After data storage and extraction, and prior to modeling or calibration, it was emphasized that it is of paramount importance to apply preprocessing techniques to the NIRS data to remove variations induced by physical phenomena in the spectrum. It was stated that there are two main preprocessing techniques: scatter correction methods and spectral derivatives. Multiplicative Scatter Correction (MSC) and Standard Normal Variate (SNV) are the most common scatter methods, and as for the spectral derivatives methods, Savitsky-Golay and Norris-Williams are the most common ones.

The calibration is then applied to the preprocessed NIRS data and the trait of interest reference values to find the mathematical correlation between the NIR spectrum and a component of interest. This showcases the importance of NIRS data in predicting unseen values of e.g. nutritional quality traits, viability, etc. The calibration techniques are mainly either descriptive or predictive. Although many descriptive models result in high values of  $R^2$ , this statistic is generally calculated on the training data which will systematically lead to biased results.

To avoid this problem, it was explained that it is important to apply the predictive, also known as Machine Learning, models for the NIRS calibration to produce more robust models. It is worth noting that the calibration models are affected by many factors, such as trial, year, location, the machine used, etc. and these should be considered in the analysis pipeline.

Lastly, it was mentioned that to build confidence in the calibration method, the errors that may occur due to repeatability and reproducibility should be measured within each laboratory, and the precision and accuracy metrics recorded. Also, participants were introduced to a web tool for NIRS data extraction, preprocessing and calibration which helps to shape the NIRS calibration pipeline.

## Gaps identified and opportunities

1. Addressing infrastructure gaps: The participants discussed the need of basic wet chemistry facilities to be strictly standardized as per the guidelines of animal nutrition protocols or the Association of Official Agricultural Chemists (AOAC) methods to obtain the most accurate and precise reference chemical data for having robust NIRS calibration models for future predictions of new germplasm samples.
2. Uniform grinding for uniformity: It was suggested that each organization needed to procure the Retsch SM100 or Wiley Mill heavy duty grinding machines to acquire uniform particle size (1mm sieve) of samples (especially for cereal crop stover grinding, e.g: maize, sorghum and millets) and a Cyclotech type grinder for all food grains and monogastric/aquaculture feed samples (micronized form) for repeatable biochemical laboratory (ring test between wet laboratories) data across the regions and to develop a valid regional spectral library.
3. Wet chemistry support: The training sessions underscored the need for basic wet chemistry capacity in the regions. Important areas of ruminant feed are the Van Soest Systems for fibre analysis, in vitro systems for rumen kinetics analysis and in vitro organic matter digestibility etc. Each organization and country should explore ways to acquire these systems or collaborate with ILRI to arrange support mechanisms. In the context of monogastric and aquaculture nutrition, capacity for amino and fatty acid analysis is a pertinent area to be addressed. The most economically feasible options are: 1) to acquire calibration equations that are available in the market for short-term applications; and 2) to collaborate for such services from established laboratories, e.g. ILRI in this context.
4. Exploiting existing capacity at ILRI for unique services: This includes capacity for micro-nutrient analysis to support NIRS applications in micronutrient analysis in emerging plant foods and derivatives. Pertinent examples include biofortification of beans and rice with iron and zinc, beta carotene in sweetpotatoes and retinol in yam beans, dhurrin potential in early maturity sorghum forages and tannins in some of the legume crop forages.
5. NIRS community of practice: The idea of establishing an NIRS community of practice (CoP) and updating the platform was also highly stressed as a good opportunity to strengthen collaboration, widen access and use of the NIRS platform.

## Conclusion

All the participants were given one calibration file and one demo version of WinISI III calibration development software for their practice at their respective workstations. The trainee participants have promised that they will practice all the training exercises given to them and were asked to raise any queries that they come across during the practical sessions at their laboratories. The Senegal Foss Stationary NIRS DS3F installed in ENSA agricultural research station, in Thies. The Cereal Quality Lab of INRAT, Tunisia received and installed the FOSS DS3F stationary NIRS machine after the training was finished and require standardizing the machine and validation by ring test samples. The Zambia partners do not have a FOSS stationary NIRS machine, they will raise the issue to their institute director to make use of the ILRI-developed global calibrations for Zambia food-feed-forage work. The CIAT Kenya team is sending the forage samples to ILRI mycotoxin and nutrition laboratory and may purchase the FOSS NIRS in future to integrate the NIRS in the community of practice (CoP). Till then they can make use of ILRI laboratory results for their forage quality testing.

Finally, all the trainee participants agreed on the need to establish an NIRS CoP to effectively make use of the ILRI-developed NIRS global calibration models. However, before establishing this CoP the following activities were identified as necessary.

1. Once all the purchases of NIRS equipment have been completed, the equipment (particularly the benchtop instruments) and their operations need to be standardized.
2. Further on-site training will be required to familiarize the users with the equipment and enable them to comfortably to use it, this is particularly important as most of the trainees have not used NIRS before and previous experience has shown that they will need further support until their NIRS machines are standardized and ring test validation is done on their new machines. For example, the ISRA Senegal machine was purchased recently and needs to have all the validation tests performed which is tentatively planned for year 2025.
3. The ring test was completed on ICARDA Rabat FOSS DS2500 NIRS machine and the results shared with the ICARDA and CIAT breeders to connect them in the network of NIRS CoP.

Overall, the training workshop ignited the trainee minds, and they would like to establish this technology platform in their respective countries for forage-feed quality testing for rapid phenotyping and to support their breeding/nutrition programs.

# Training agenda

4–8 November 2024, ICARDA Rabat campus, Morocco

Date	Time	Agenda	Facilitator/ Presenter
Day 1 Monday 4 November 2024	9:00–9:30 am	Registration	Fatima/ Essahli
	9:30–10:00 am	Welcome to ICARDA & overview of forage feed quality traits	Miguel
	10:00–10:30 am	Coffee/tea/group photo	
	10:30 am–12:30 pm	<ul style="list-style-type: none"> <li>Use &amp; maintenance of forage mills.</li> <li>Laboratory visit and sample analysis demonstration</li> </ul>	Khalwa
	12:30–2:00 pm	Lunch	
	2:00–3:00 pm	Theoretical & technology behind NIRS <ul style="list-style-type: none"> <li>What is NIRS, how does it work?</li> <li>Component of NIRS &amp; explanation of each part</li> <li>NIRS measurement processes</li> </ul>	Prasad
	3:00–3:30 pm	Coffee/tea	
	3:30–5:00 pm	Theoretical & technology behind ... (continued)	Prasad
Day 2 Tuesday 5 November 2024	9:00–10:00 am	NIRS analysis <ul style="list-style-type: none"> <li>Description of WinISI software, Installation &amp; Setup</li> <li>WinISI software functions / File view</li> </ul>	Prasad/ Khalwa
	10:00–10:30 am	Coffee/tea	
	10:30 am–12:30 pm	NIRS ... (continued)	Prasad
	12:30–1:30 pm	Lunch	
	1:30–2:30 pm	<ul style="list-style-type: none"> <li>Data collection/Spectra evaluation</li> <li>Develop calibration equation.</li> <li>Validation &amp; monitoring of calibration equations</li> </ul>	Prasad
	2:30–3:00 pm	Coffee/tea	
	3:00–5:00 pm	Data collection ... (continued)	Prasad
Day 3 Wednesday 6 November 2024	9:00–10:00 am	Database management & modelling	Khadija Aouzal
	10:00–10:30 am	Coffee/tea	
	10:30 am–12:30 pm	Database mgt ... (continued)	Khadija Aouzal
	12:30–1:30 pm	Lunch	
	1:30–2:30 pm	Handheld NIRS	Prasad/ Miguel
	2:30–3:00 pm	Coffee/tea	
	3:00–5:00 pm	Handheld ... (continued)	Prasad/ Miguel

Date	Time	Agenda	Facilitator/ Presenter
Day 4 Thursday 7 November 2024	9:00–10:00 am	Develop & use of NIRS SOP & Ring test	Prasad/ Khawla
	10:00–10:30 am	Coffee/tea	
	10:30 am–12:30 pm	NIRS DS2500 Standardization	Prasad/ Khawla
	12:30–1:30 pm	Lunch	
	1:30–2:30 pm	NIRS instrument maintenance & troubleshooting	Prasad/ Khawla
	2:30–3:00 pm	Coffee/tea	
	3:00–5:00 pm	NIRS instrument...Ring test results discussion	Prasad/ Miguel
Day 5 Friday 8 November 2024	9:00–10:30 am	Practical session: Put the acquired knowledge into practice & perform NIRS analysis in the lab	Khawla/ Prasad
	10:30–11:00 am	Coffee/tea	
	11:00 am–12:30 pm	Practical ... (continued)	Khawla/ Prasad
	12:30–1:30 pm	Lunch	
	1:30–3:00 pm	Discussion & conclusion of the training	Prasad/ Michael



The International Livestock Research Institute (ILRI) works to improve food and nutritional security and reduce poverty in developing countries through research for efficient, safe and sustainable use of livestock. Co-hosted by Kenya and Ethiopia, it has regional or country offices and projects in East, South and Southeast Asia as well as Central, East, Southern and West Africa. [ilri.org](http://ilri.org)



CGIAR is a global agricultural research partnership for a food-secure future. Its research is carried out by 15 research centres in collaboration with hundreds of partner organizations. [cgiar.org](http://cgiar.org)