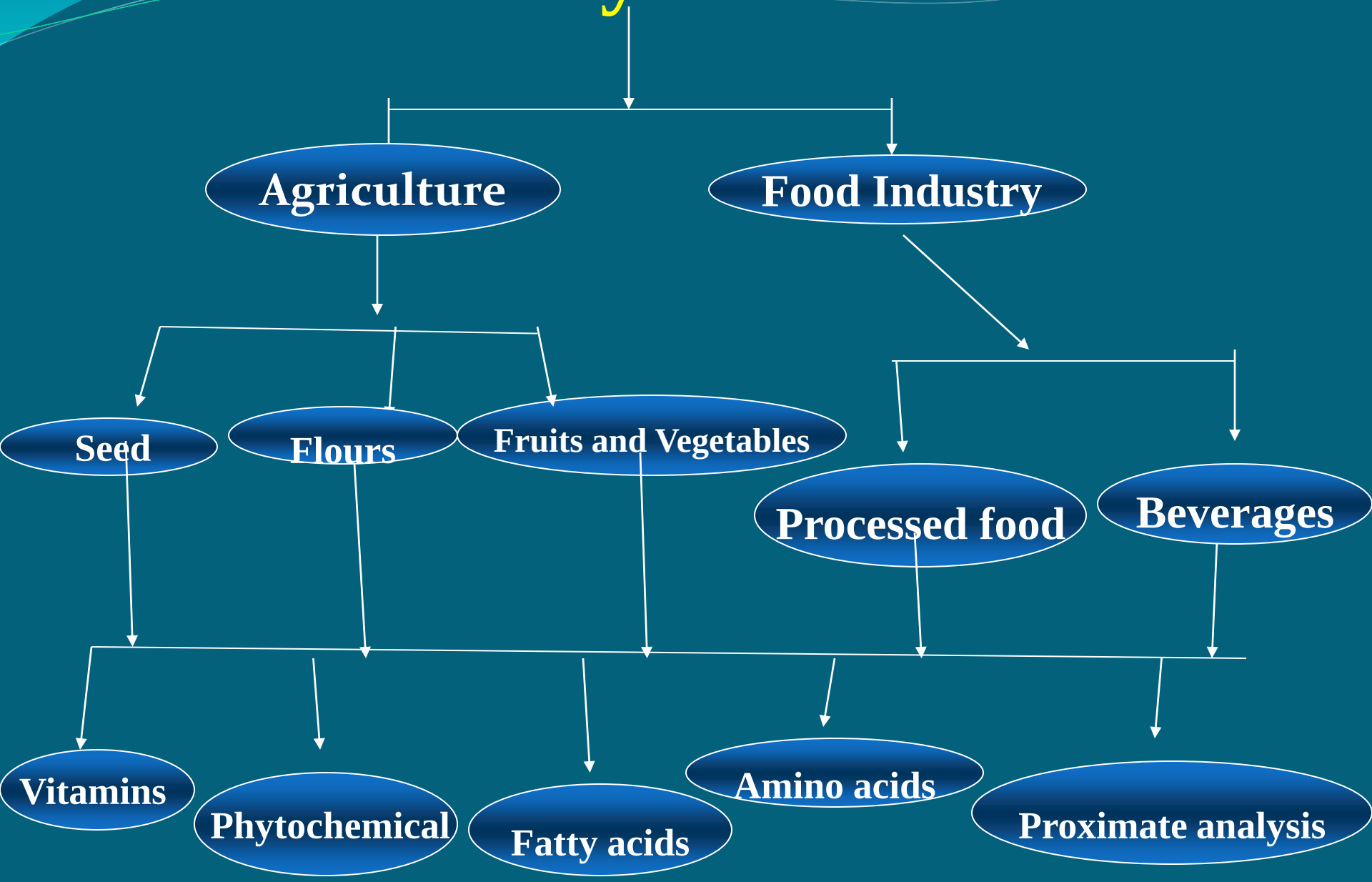
NIRS Based Food Quality Assessment Approaches for Cereals, Oilseeds, Pulses, Fruits and Vegetables

- *Rakesh Bhardwaj, Sangita Yadav[#] and Poonam Suneja
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Utility of NIRS

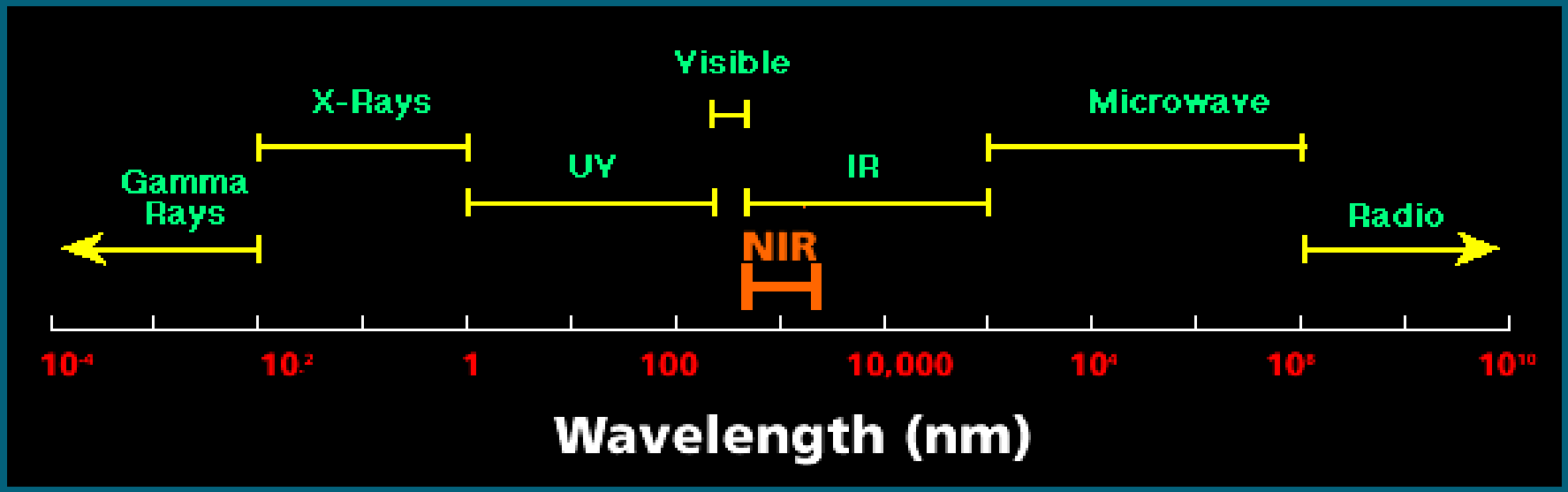


Near Infra-red spectrophotometer (NIRS) Technique



- Multi-trait technique
- Non- destructive
- Reliable & Cost effective
- Rapid screening of vast germplasm resources
- Limited sample preparation
- Replaces typical time consuming, expensive and hazardous primary analysis
- Eliminates the errors involved at each step of wet chemistry

Wavelength regions



SPECTRAL ABSORPTION

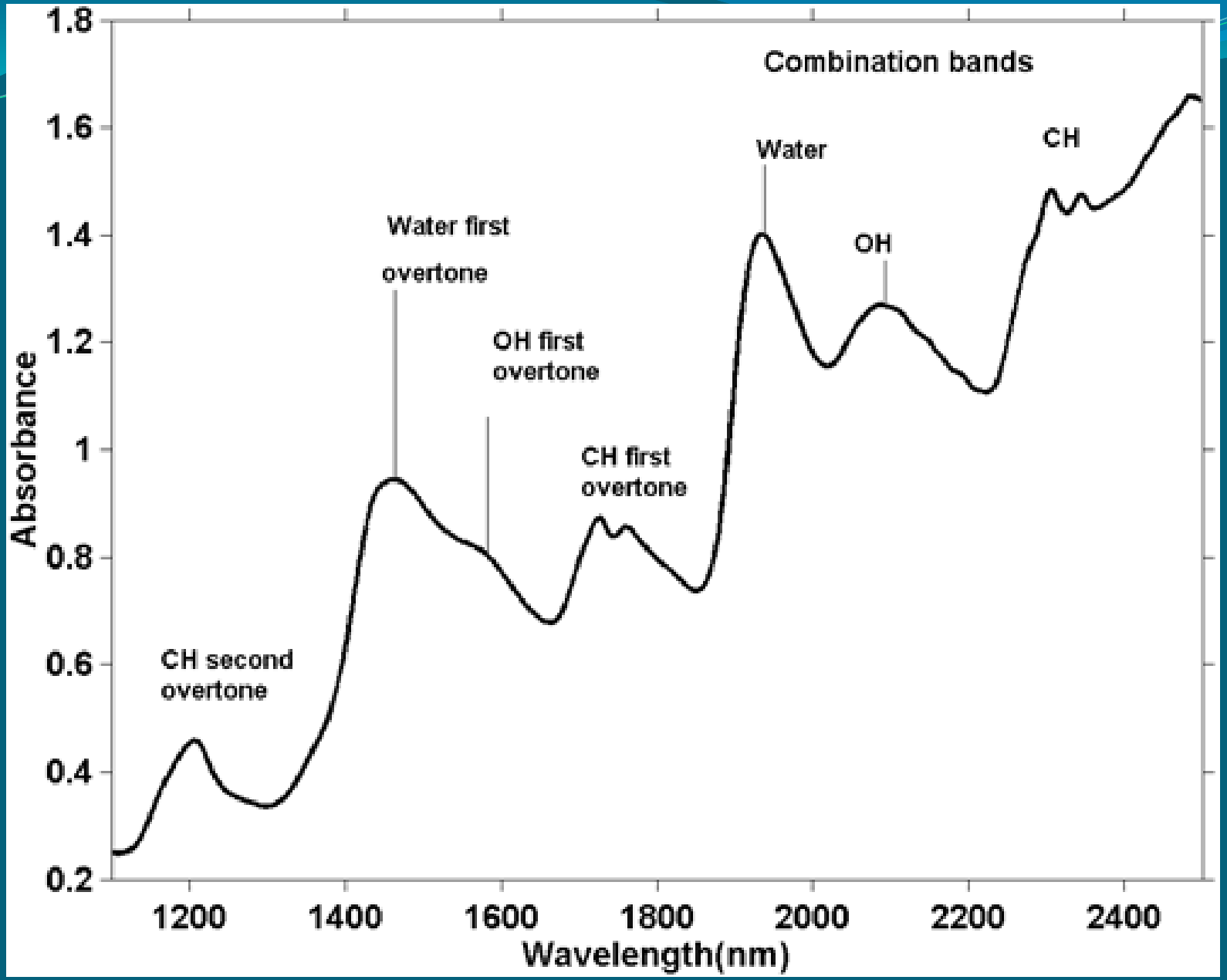
Radio wave : NMR
Microwave : Rotation of molecules (ESR)
IR : Fundamental Molecule Vibrations
NIR : Overtones and combinations of Mid –IR
UV/ Visible : Electronic transition, Energy of electron raised to an excited state
X-ray : Core electronic transition in atom (XRF)

NIRS Characteristics

- Fundamental O–H and C–H stretching absorption are different. So, the series of overtones generated by these absorptions are also different.
- The most common (and energetic) combination bands arise from stretch and bend combinations in the same group.
- R-H group have the strong overtones due to hydrogen's degree of anharmonicity.
- O-H, N-H, C-H, S-H bonds etc. have Strong Absorption.
- H_2 and O_2 has no change in dipole moment , hence no NIR absorption

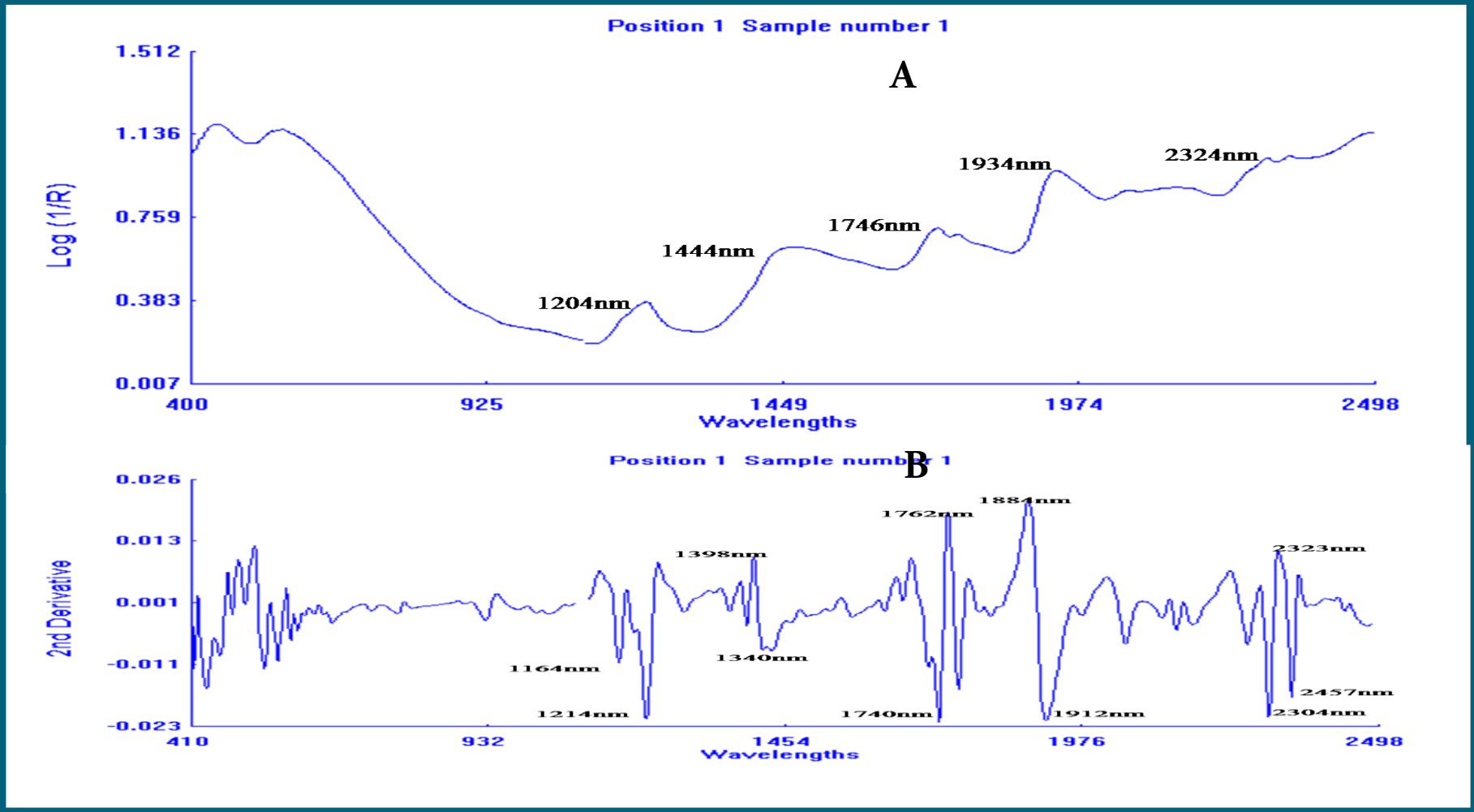
Absorption in the NIRS Region

- **Absorptions in the NIR region (780–2500 nm) are generated from fundamental vibrations - overtones and combinations.**
- **Overtones are like harmonics. Every fundamental wavelength produce a series of absorptions at (approximately integer) multiples of the frequency .**
- **Combinations arise from the sharing of NIR energy between two or more fundamental absorptions. Large number of combinations are observed.**
- **The effect of all these absorptions combine to make NIR spectra sloppy which consist of only a few broad peaks**



Spectrum of Biscuit dough

Spectral Analysis



Raw spectrum (log 1/R; A) and second derivative (B) of NIRS average spectrum of intact seeds of rapeseed – mustard germplasm

NIRS work at ICAR-NBPGR

- **NIRS model 6500, Foss NIR Systems Inc., MD, USA in the reflectance mode**
- **Intact seed samples for small seed like brassica, amaranth and grounded-sieved flour for bold seeds like chickpea, ground nut.**
- **All spectral data are recorded as the logarithm of reciprocal of reflectance (log 1/*R*)**
- **Wavelength range from 400 to 2500 nm, at 2 nm intervals**
- **WinISI II software - NIRS calibration, mathematical processing, and statistical analysis.**

- **Mahalanobis distance (*H* distance) - Ranks spectra according to *H* distance from the average spectrum, and provides spectral boundaries to eliminate outliers with $H > 2.5$**
- **Split samples randomly - calibration and validation sets.**
- **First sample set used to calibrate and cross-validate the derived equation; Internal cross validation to avoid over fitting of the equations (Shenk & Westerhaus, 1996).**
- **Second sample set for external validation to test the goodness of fit of the developed equations (Windham et al., 1989).**

Prediction models for non-destructive analysis developed in NBPGR

Oil seeds	***Brassica*, safflower, sunflower, linseed etc**	**Oil percent, Fatty acids**
Legumes	**Pigeon pea, chickpea, Pea, lentil etc**	**Protein, saponins, sugar, phenol, phytate, starch**
Cereals	**Maize, wheat etc**	**Oil percent, Protein content**
Psedo-cearals	**Amaranth, buckwheat etc**	**Oil percent, Fatty acids, Protein content**
Millets	**Faba bean , rice bean etc.**	**Protein content**
Vegetables	**Brinjal**	**Ash content, phenol, sugar, starch, mineral content**

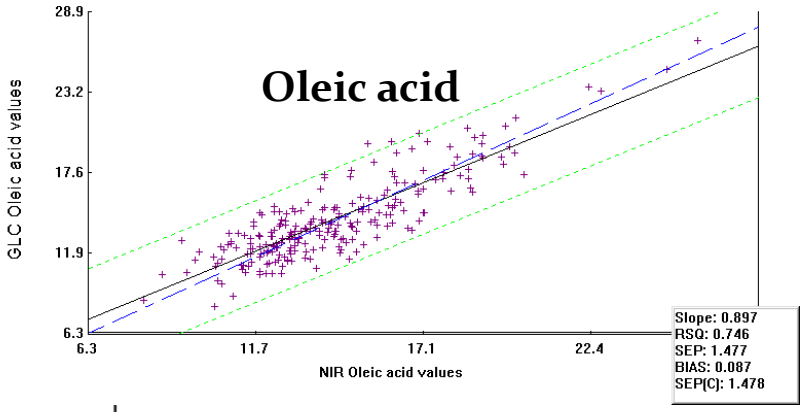
Performance of best predicted equation

- **Standard error of cross validation (SECV) : Minimum**
- **Coefficient of determination (R^2) : Maximum**
- **Corrected standard error of prediction (SEP(C)) : Minimum**
- **Ratios of standard deviation (SD) of reference data to SEPV : >3**

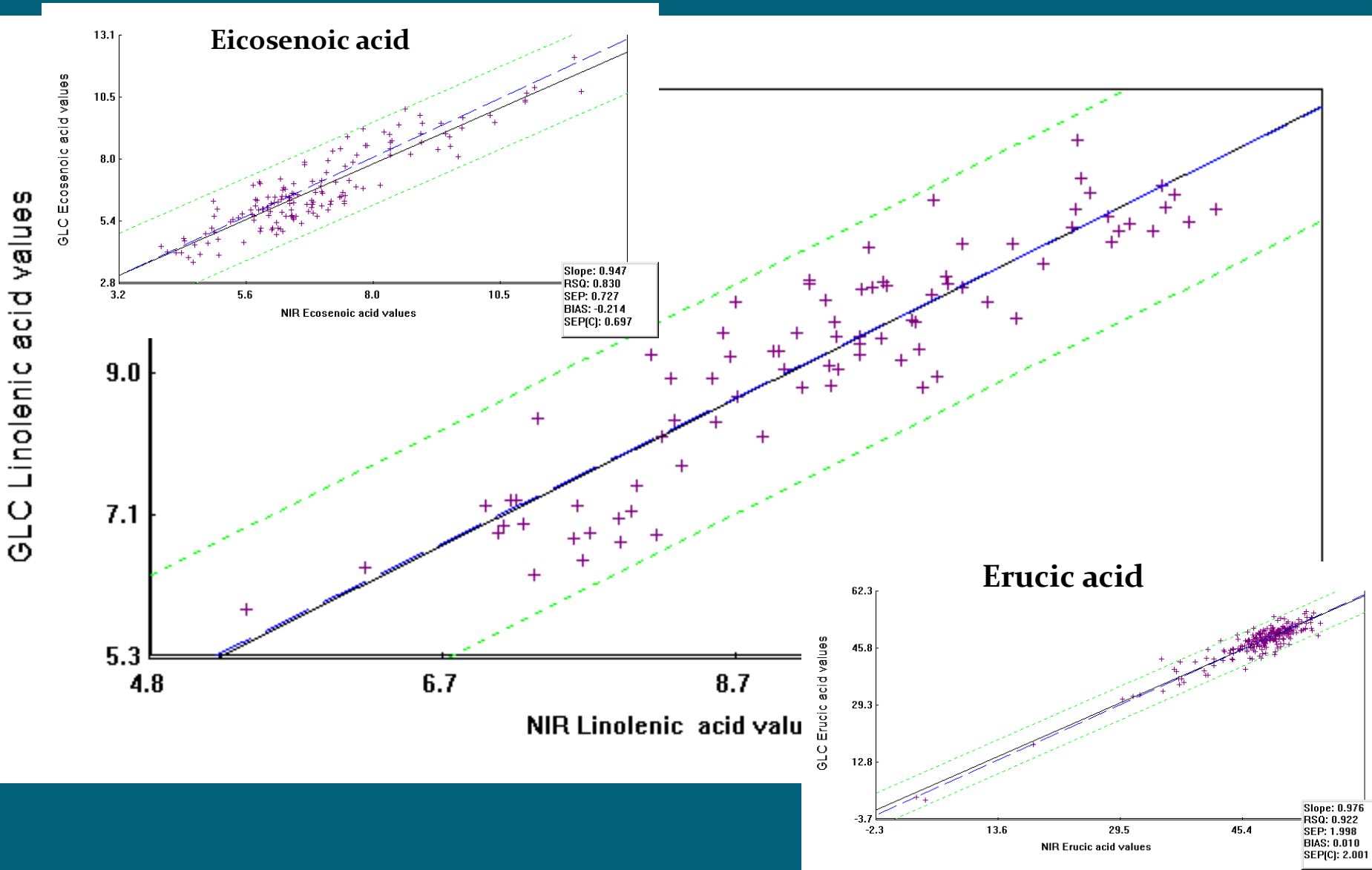
Validation statistics for oil and fatty acids of *Brassica*

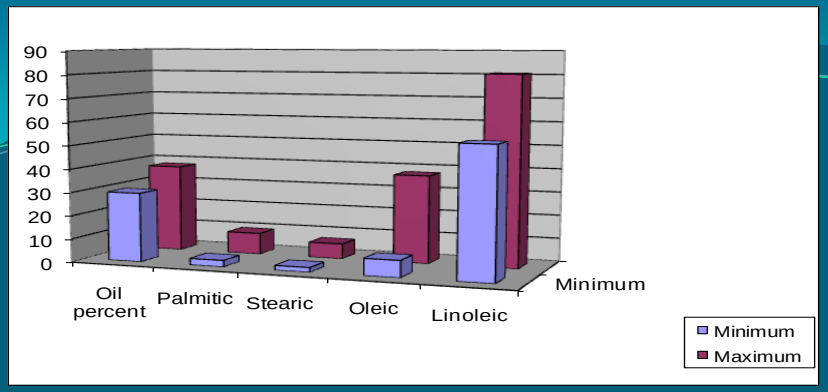
Parameters/Traits	oil	Palmitic acid	stearic acid	oleic acid	linoleic acid	linolenic acid	ecosenoic acid	erucic acid
No. of samples analysed	102	112	79	117	112	115	83	119
Mean	40.03	2.676	1.02	17.66	16.36	8.925	6.093	45.62
SD (Standard Deviation)	5.33	0.692	0.18	11.97	2.354	1.81	1.332	14.30
SEP (Standard error of Prediction)	1.612	0.197	0.089	1.82	0.90	0.772	0.605	2.33
RSQ (Coefficient of Determination of Cross Validation)	0.922	0.918	0.758	0.978	0.853	0.818	0.792	0.974
SEP(C) (Corrected Standard Error of Prediction)	1.52	0.198	0.089	1.825	0.903	0.774	0.608	2.33
Slope (the steepness of straight line curve)	1.071	1.006	1.056	1.032	1.008	0.971	0.998	1.034
Bias (Average difference between reference and NIRS values)	0.545	0.002	-0.005	0.112	-0.046	-0.026	-0.718	-0.18
RSP (Ratio of SD of Reference data to SEP(C)	3.51	3.49	2.02	6.55	2.60	2.34	2.19	6.13

Scatter plots of NIRS vs. reference values for palmitic, oleic acid and linoleic acid in calibration sets

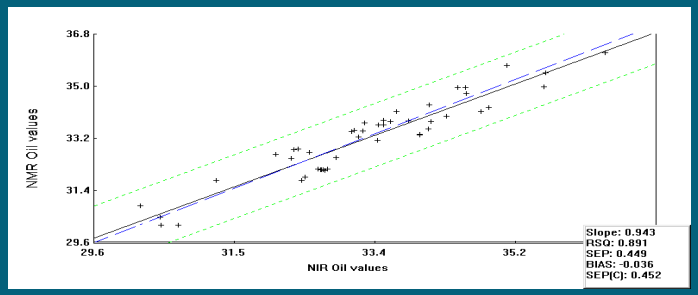


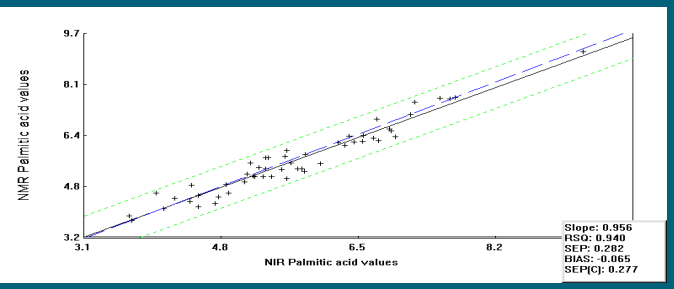
Scatter plots of NIRS vs. reference values of linolenic acid, eicosenoic acid and erucic acid in calibration sets

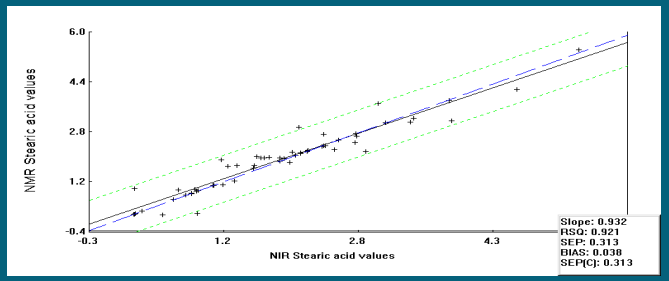


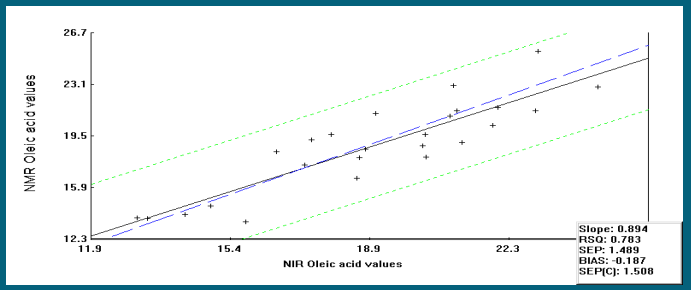


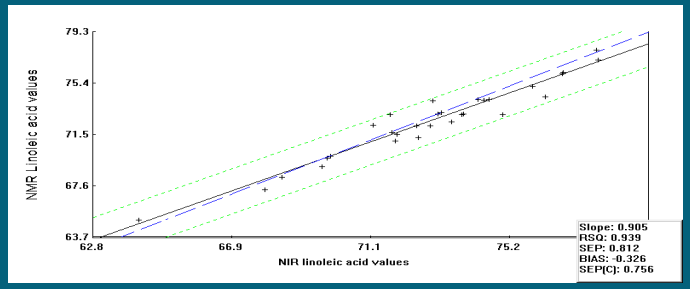
	Mean	SD	r^2	RPD
Oil	33.23	1.35	0.891	3.00
Palmitic acid	5.71	1.12	0.940	4.06
Stearic acid	1.86	1.11	0.921	3.58
Oleic acid	18.96	3.12	0.780	2.06
Linoleic acid	72.82	3.04	0.939	4.02









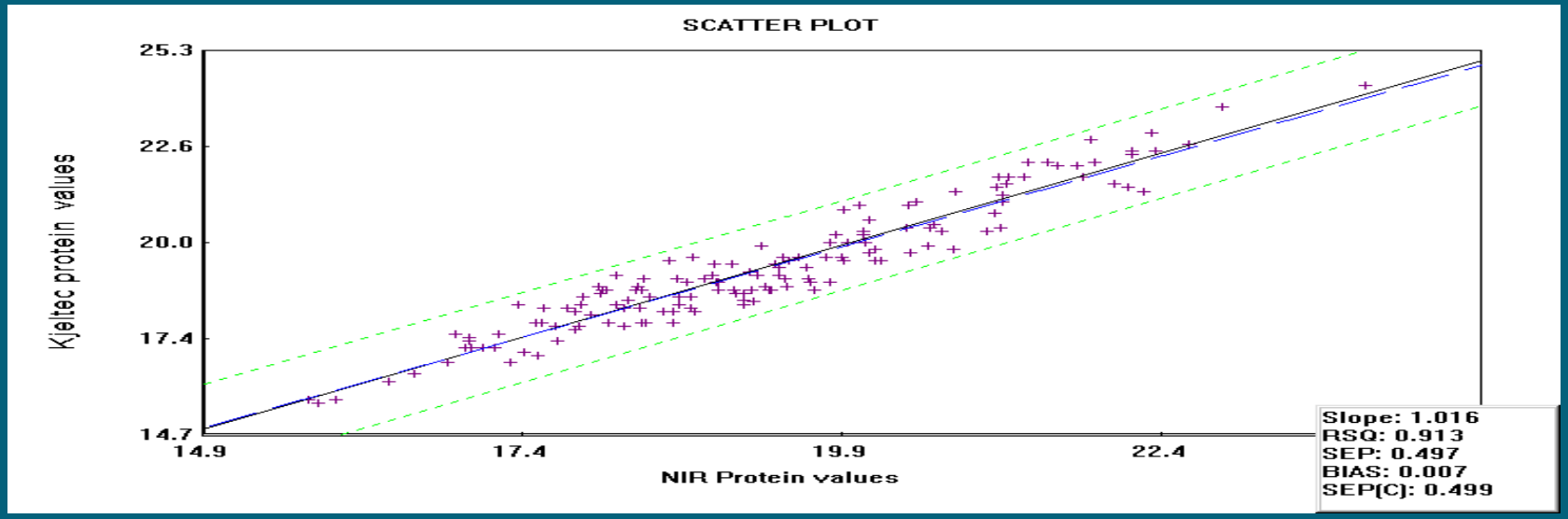


Calibration models for oil and fatty acid composition of Safflower

Communicated in *Indian J. of Plant Genet. Resour.*

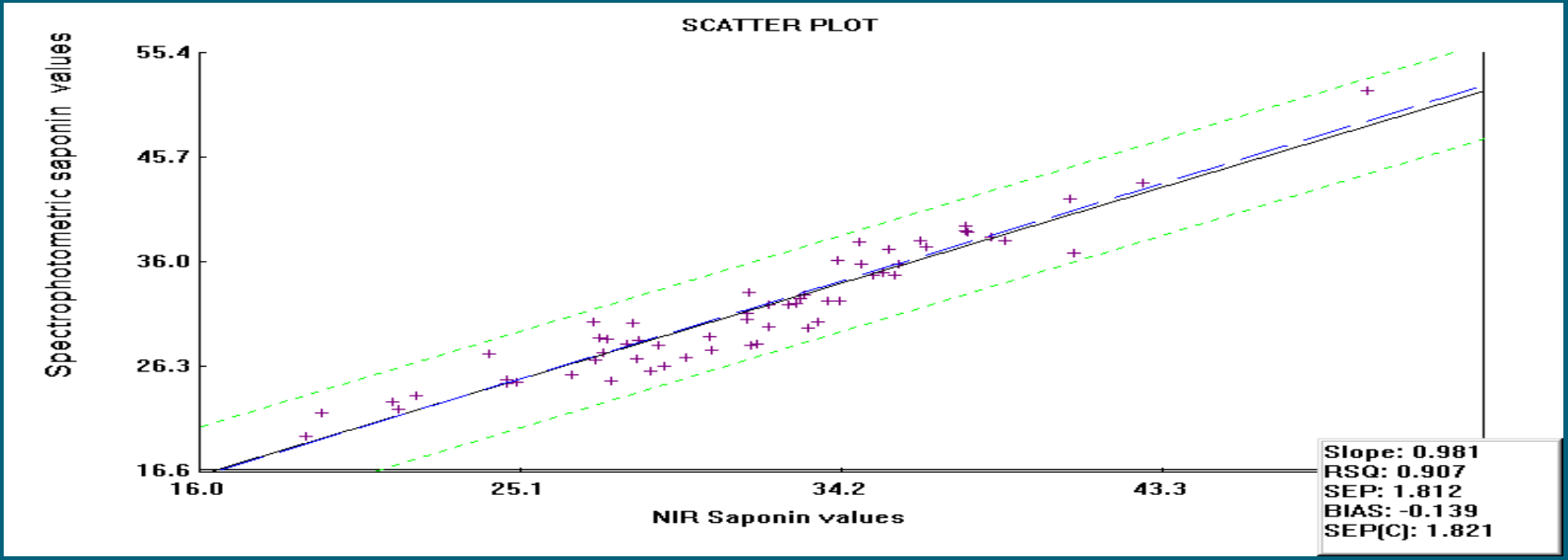
Calibration equation for protein in chickpea

No. of samples	148
Standard deviation (SD)	1.688
Standard error of prediction {SEP (C) }	0.499
Coefficient of determination in calibration (r^2)	0.913
slope	1.016
SD/SEP(C)	≥ 3



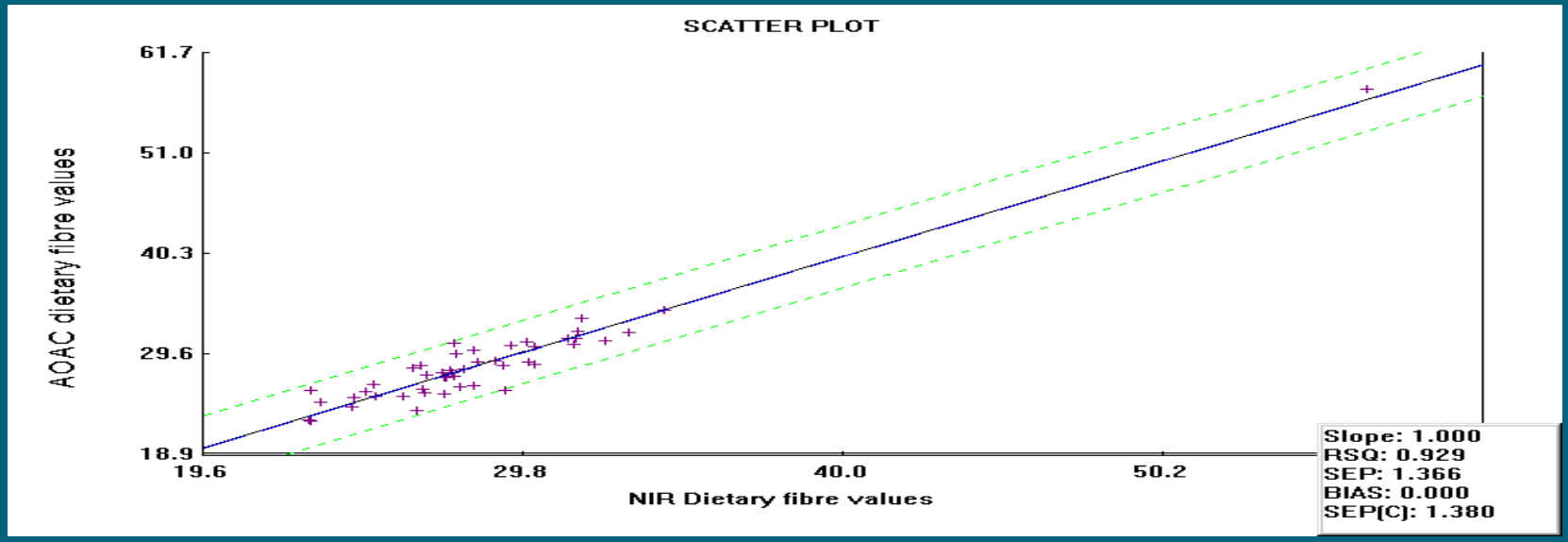
Calibration equation for Saponin in chickpea

No. of samples	60
Standard deviation	5.96
Standard error of prediction (C)	1.821
Coefficient of determination in calibration (r^2)	.0.907
slope	0.981
SD/SEP(C)	≥ 3



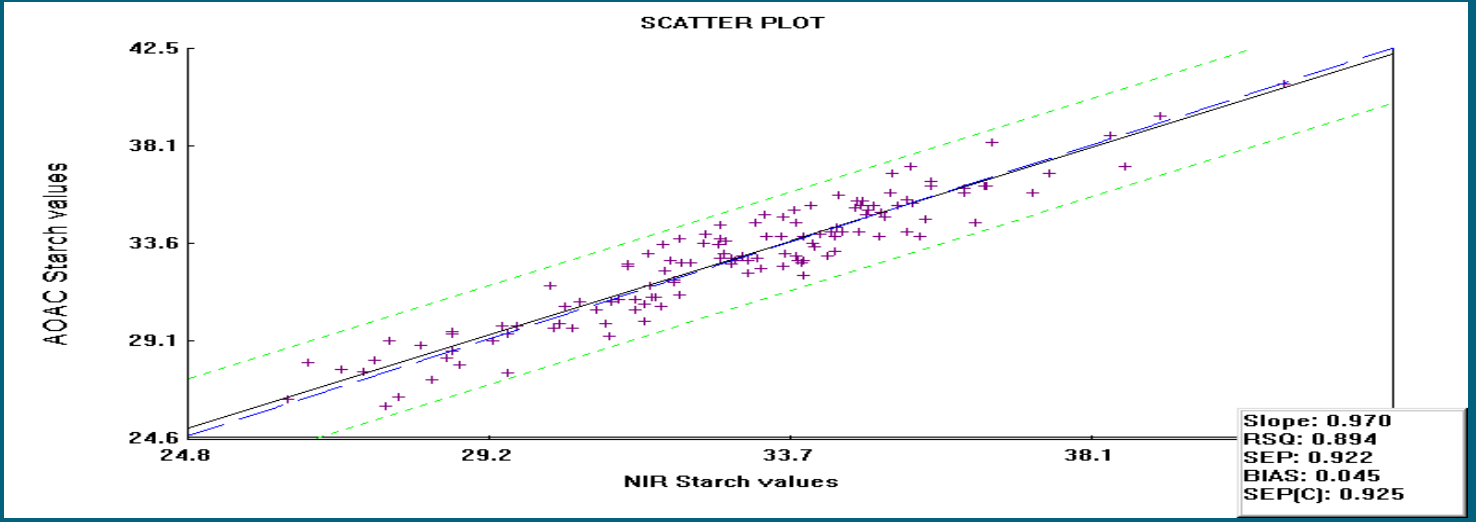
Calibration equation for dietary fibre in chickpea

No. of samples	47
Standard deviation	5.189
Standard error of prediction (C)	1.380
Coefficient of determination in calibration (r^2)	0.929
slope	1.0
SD/SEP(C)	≥ 3



Development of calibration equation for Starch content in chickpea

No. of samples	127
Standard deviation	2.829
Standard error of prediction (C)	0.925
Coefficient of determination in calibration (r^2)	0.890
slope	0.970
SD/SEP(C)	≥ 3

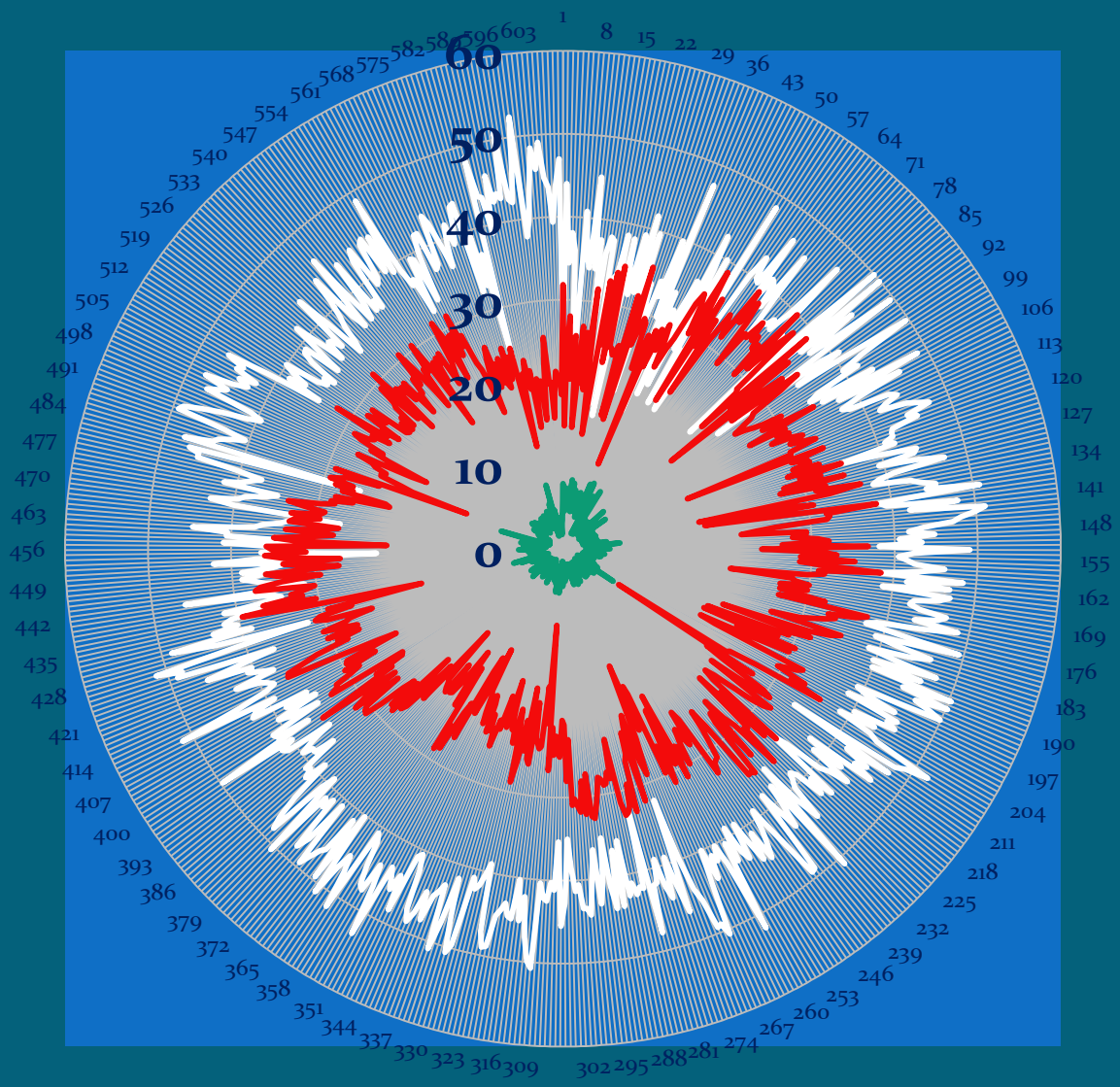


Selecting Trait Specific Appropriate Prediction Model for chickpea

Trait	Math treatment 2,4,4,1		Math treatment 3,6,6,2		Math treatment 4,6,6,2	
	(R^2)	(SEC)	(R^2)	(SEC)	(R^2)	(SEC)
Starch	0.9271	0.890	0.6050	1.8722	0.7762	1.5592
Protein	0.9338	0.4906	0.8918	0.627	0.5812	1.184
Oil	0.9235	0.2630	0.9208	0.2489	0.588	0.8748
Moisture	0.8831	0.1754	0.9436	0.1142	0.8866	0.1727
Dietary fiber	0.8749	0.9349	0.9681	0.7881	0.2496	2.4389
Phenol	0.8132	1.7414	0.8978	1.288	0.9367	0.7387

R^2 - Coefficient of determination in Prediction, SEC – Standard error of calibration

NIR based screening of chickpea germplasm (733 accn)

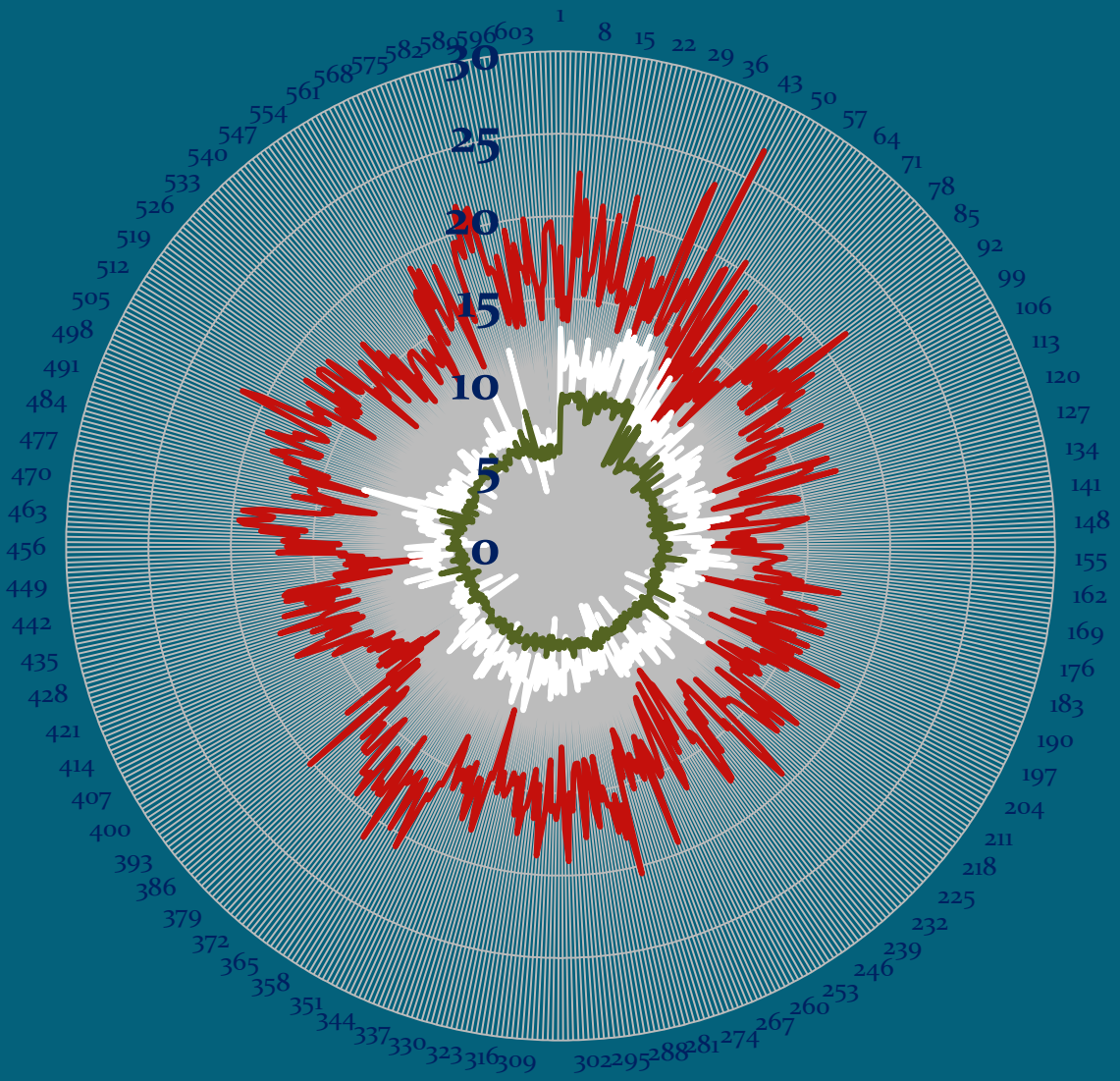


—Strach

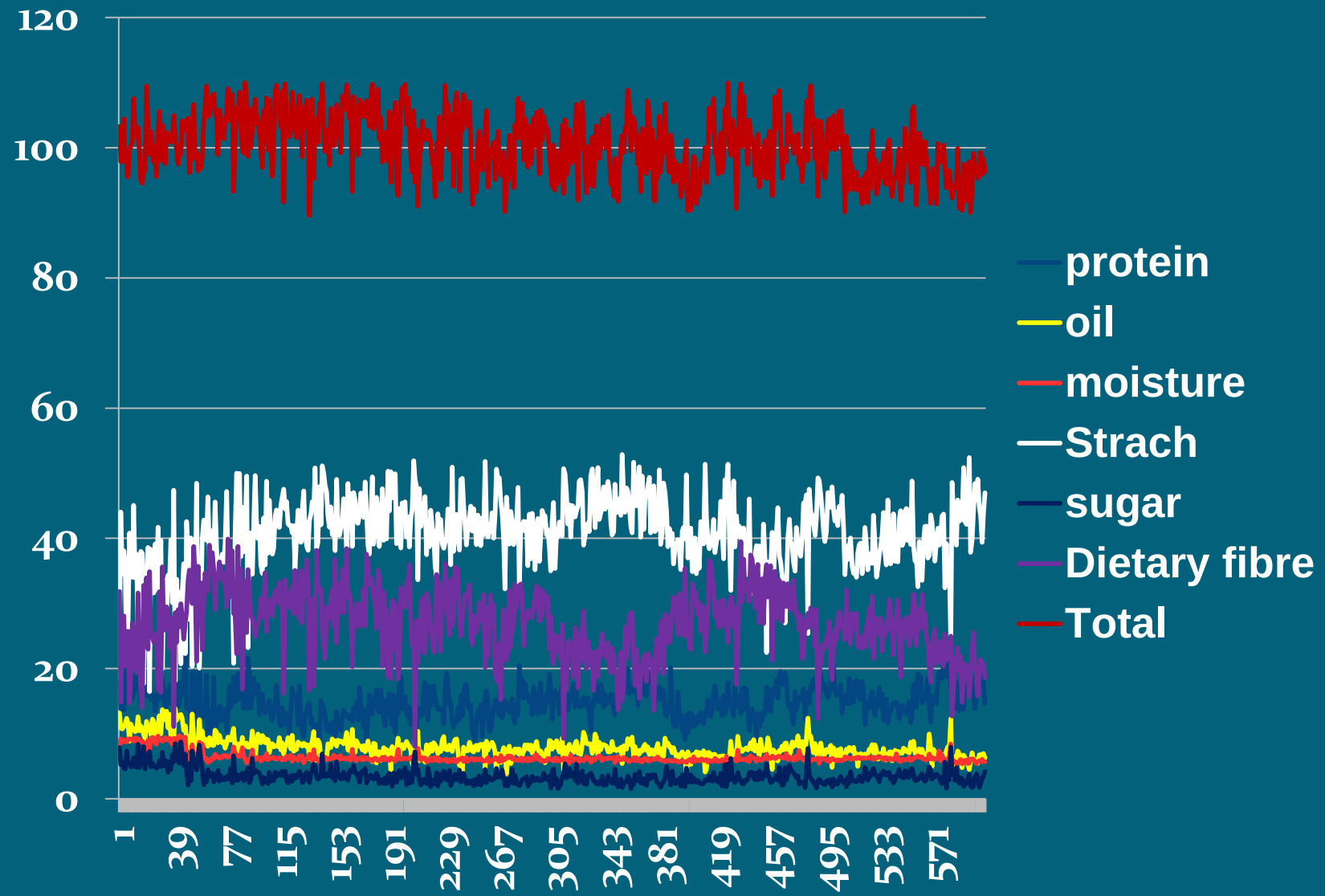
—sugar

—Dietary fibre

NIR based screening of chickpea germplasm (733 accessions)



NIR based screening of chickpea germplasm (733 accn)



NIR based screening of chickpea germplasm (733 accn)

Trait	Est. min	Est. Max	Mean	SD	SEC	RSQ	SD/SEC
Starch	22.23	42.08	32.12	3.29	0.89	0.93	3.7
Protein	13.75	25.19	19.47	1.91	0.49	0.93	3.9
Oil	1.68	7.39	4.54	0.95	0.26	0.92	3.6
Dietary fiber	15.38	41.85	28.62	4.41	0.79	0.97	5.6
Moisture	8.36	11.42	9.89	0.562	0.20	0.94	2.8
sugar	1.77	7.81	4.48	0.90	0.30	0.87	3.0

SD/SEC > 3.0, RSQ > 0.9

Utility of FOSS NIRS (6500) in Vegetable Analysis











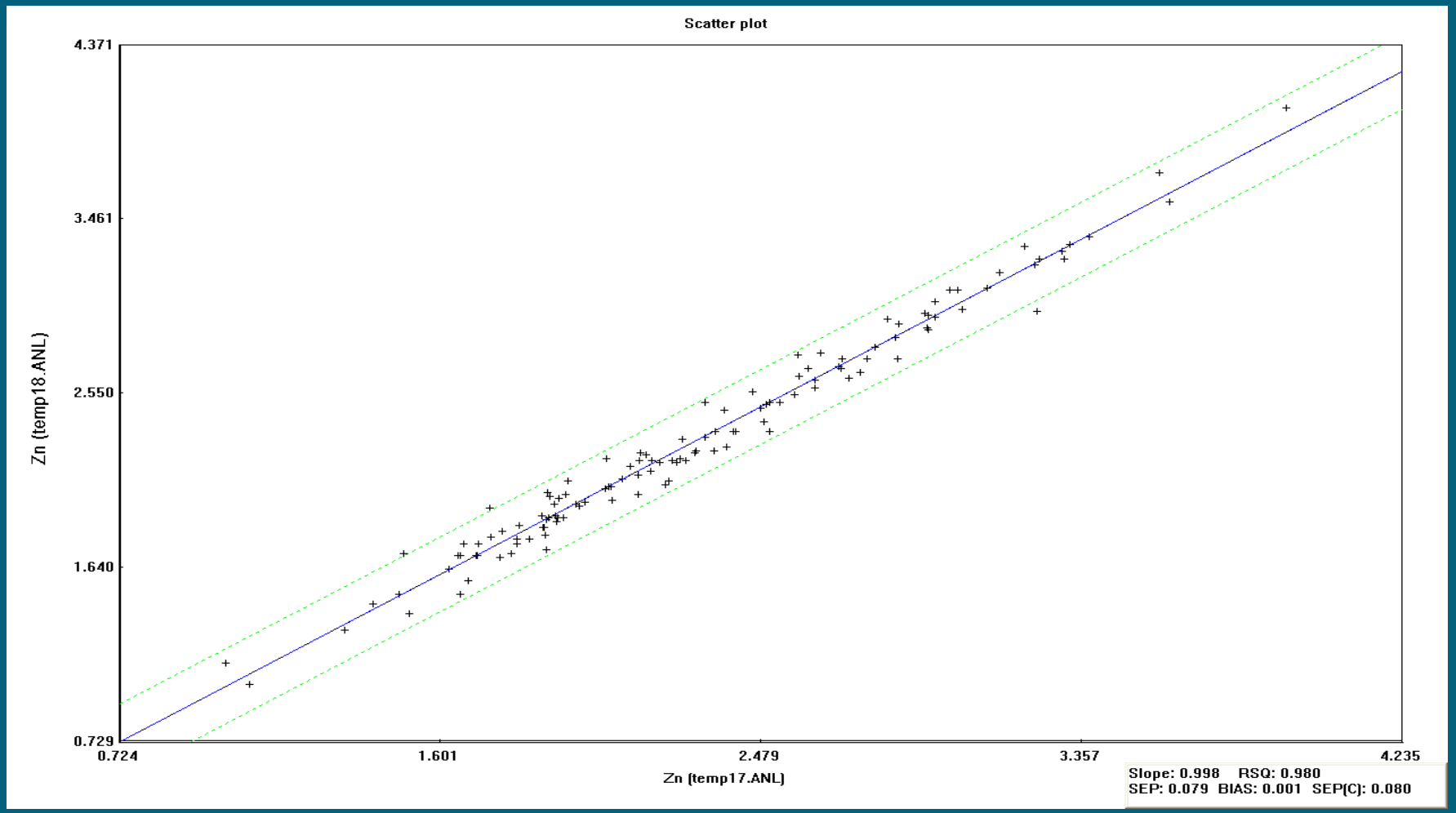
Selecting Trait Specific Appropriate Prediction Model for Eggplant

Trait	Math treatment 2,4,4,1		Math treatment 3,6,6,2		Math treatment 4,6,6,2	
	(R^2)	(SEC)	(R^2)	(SEC)	(R^2)	(SEC)
Ash	**0.975**	**0.116/0.018 (6.44)**	0.886	0.116/0.039 (2.97)	0.868	0.116/0.043 (2.69)
Fe	**0.978**	**5.07/0.76 (6.67)**	0.555	5.07/3.3387 (1.51)	0.592	5.07/3.25 (1.56)
Zn	**0.987**	**0.561/0.08 (7)**	0.868	0.561/0.204 (2.75)	0.917	0.561/0.161 (3.48)
Cu	**0.974**	**0.706/0.113 (6.72)**	0.744	0.706/0.359 (1.96)	0.847	0.706/0.279 (2.53)
Sugar	**0.973**	**0.537/0.088 (6.1)**	**0.928**	**0.537/0.144 (3.72)**	0.851	0.537/0.208 (2.58)
Phenol	**0.984**	**0.036/0.004 (9)**	**0.948**	**0.036/0.008 (4.5)**	**0.958**	**0.036/0.007 (5.14)**
Moisture	**0.977**	**0.895/0.136 (6.58)**	**0.947**	**0.895/0.207 (4.32)**	**0.973**	**0.895/0.147 (6.08)**

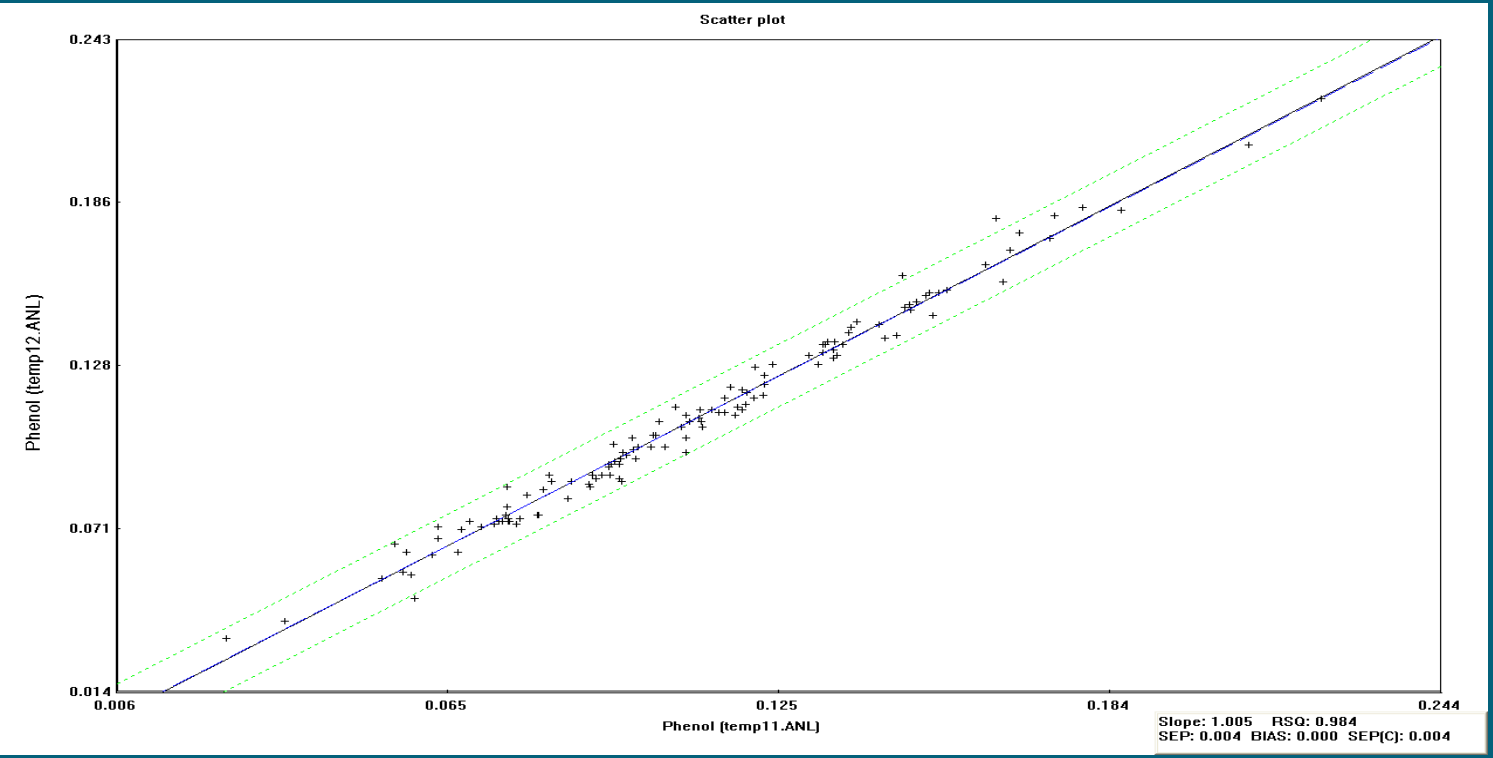
NIR prediction results for eggplant fruits

Traits	Wet chem. (126 sam)		NIR Estim. (126 sam)		Predicted (102 sam)	
	Range	Mean	Range	Mean	Range	Mean
Ash	0.47-1.21	0.75	0.40-1.11	0.75	0.37-1.08	0.79
Mg	25-345.7	79.14	31.6-242.1	83.2	8.07-232.9	103.0
Fe	2.62-29.8	9.55	2.56-26.3	9.92	1.34-20.0	11.7
Zn	1.03-4.04	2.31	0.64-4.03	2.34	1.36-4.67	2.69
Cu	0.88-4.96	2.00	0.88-4.08	1.97	1.09-4.98	2.52
Protein	0.94-2.04	1.42	0.78-2.06	1.42	0.93-1.89	1.46
Sugar	0.81-3.73	2.35	0.74-3.96	2.35	1.24-3.68	2.42
Phenol	0.03-0.22	0.11	0.03-0.22	0.11	0.05-0.24	0.14
Moisture	88.3-93.5	91.3	88.6-93.9	91.3	87.2-94.2	91.1

NIR prediction model for eggplant fruits (calibration scatter plot for Zn)



NIR prediction model for eggplant fruits (calibration scatter plot for Total Phenols)



Conclusion

- NIRS calibration models developed can accurately predict majority of traits.
- Can be helpful for improving efficiency of breeding programs aimed at altering fatty acid composition in oil seed crops, protein content and amino acid profiling in legume crops
- NIRS analysis of food and food product has found extensive use in all segment of Industry viz. processed food , Beverages, seed industry etc.
- NIRS prediction models on multiple nutritional traits for raw ingredients and processed products can help food industry to deliver quality products.
- NIRS predicted values are much more closer to reality than to calculated values used on food labels.