

Table 1: Chemical analysis: Acidity, Peroxide Value, K232, K270, Total carotenoids and total chlorophylls

		Monovarietal Oil (N=49)			
		BM	Zw	TZ	FBS
Acidity (%AO)	Rang	1.14-2.54	1.06-2.46	1.23-2.30	1.46-2.12
	Mean	1.83	1.65	1.7	1.82
	S.D	0.51	0.38	0.33	0.21
PV (mg/Kg)	Rang	3.22-13.72	3.27-13.33	3.27-11.94	4.73-11.33
	Mean	8.7	10.11	8.79	7.81
	S.D	3.52	2.62	2.66	2.38
K232	Rang	1.02-2.98	1.96-2.98	2.29-2.95	1.87-2.96
	Mean	2.63	2.49	2.67	2.68
	S.D	0.53	0.38	0.18	0.3
K270	Rang	0.3-1.14	0.13-1.14	0.16-0.67	0.17-0.62
	Mean	0.24	0.46	0.34	0.28
	S.D	0.12	0.28	0.2	0.13
Total carotenoids (mg/kg)	Rang	3.42-7.45	3.42-6.26	3.63-4.82	3.81-5.51
	Mean	4.78	4.16	4.07	4.6
	S.D	1.07	0.75	0.44	0.48
Total Chlorophylls	Rang	4.40-11.10	4.17-9.87	4.69-8.03	5.28-8.07
	Mean	7.28	6.09	5.91	6.84
	S.D	2.13	1.63	1.09	1.02

Carotenoid and Chlorophylls Determination: The observed levels of pigments for all studied samples are in the range of [0-20ppm]. The olives are characterized by a significant reduction in chlorophyll and to a lesser extent of the carotenoids with the evolution of the mature.

3.2 Means Infrared (MIR) Spectra

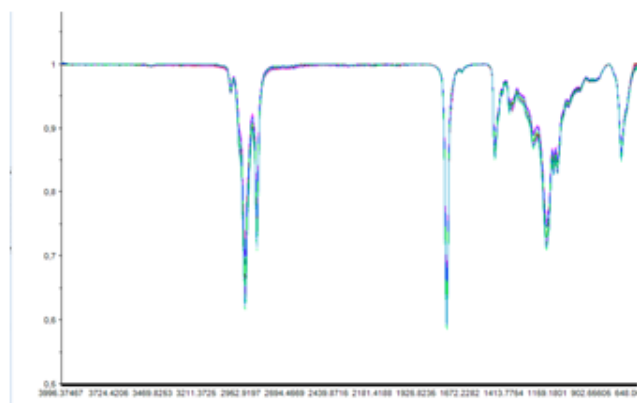


Figure 1: MIR spectra of the virgin olive oil samples in the 4000-600 cm^{-1} spectral range

Fig. 1 shows MIR spectra of monovarietal olive oil respectively. The spectra did not evidence an obvious difference from visual inspection according to the geographic origin. The MIR spectra are dominated by some peaks at 2924, 2852, 1743, 1463, 1377, 1238, 1163, 1114, 1099 and 721 cm^{-1} . Absorbance at 2924 and 2852 cm^{-1} are due to bands arising from CH_2 stretching vibrations, asymmetric and symmetric, respectively. The major peak at 1743 cm^{-1} arises from $\text{C}=\text{O}$ stretching vibrations. In particular this peak is associated with the triglyceride ester-linkage (COOR) band and the $\text{C}=\text{O}$ absorption of free fatty acid present in olive oil.

The band at 1460 cm^{-1} arises from asymmetric stretching in methyl and methylene groups, while the peak at 1160 cm^{-1} is associated with the stretching of the $\text{C}-\text{O}$ bonds of aliphatic esters [18] [19].

3.3 Chemometric Methods

a- Principal component analysis (PCA):

After analyzing of 49 samples of olive oils, Principal Component Analysis with full cross validation was applied to the first data set of 20 classification samples exploring the full acquired data.

The PCA model with two components already explained 99% of the total data variance (PC1 captured 100% and PC2 captured 1% of the variance respectively). PC1 vs. PC2 scores plot of the spectra of the first data set given in Figure 2, distinguished four major clusters of samples according to conformity FBS: Fkih Ben Saleh area/ ZW: Zawit Cheikh area / TZ: Tagzirt area and BM: Béni Mellal. The rapprochement between the samples of TZ and ZW is probably due to geographical and climatic rapprochement of these two areas.

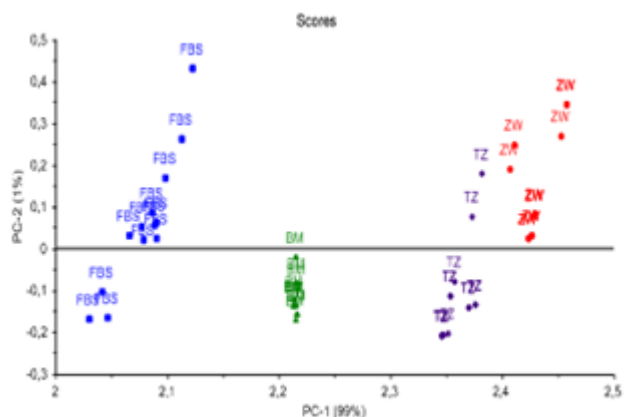


Figure 2: PCA scores plot (PC1 vs. PC2) in the analysis of the MIR spectra

b- Partial least squares discrimination PLS-DA

This PLS2-DA model has been built considering the MIR spectra as X variables, while the Y variables have been associated with the four different areas (one different y variable for each region, with 1 or 0 depending on whether it belongs or not to the considered data group). The model obtained in this way has been able to discriminate among the four areas (BM, ZW, FBS and TZ), as it can be seen from the PLS2-DA scores plot in Figure 3.

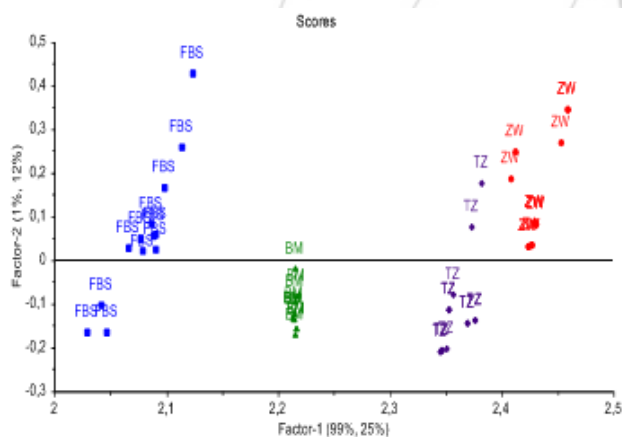


Figure 3: PLS2-DA scores plot (LV1 versus LV2) in the analysis of the MIR spectra.

These calibration models were first validated by internal full cross validation. Comparison between different models was done considering some figures of merit such as R2, RMSEC and RMSECV. The obtained model was able to distinguish satisfactory the four geographical origins (TG,ZW, FBS and BM), as it can be seen from the PLS2-DA scores plot in Figure 3, where the four groups of samples are more distinguishable. The first latent variable, LV1, explains 25 % of Y variance with 99 % of X variance, discriminates among the four origins. The second latent variable explains a rather small percentage of

variance of Y (12%) and also a small amount of variance in X (1%).

Table 2: Statistical parameters by chemometric elaboration of olive oils MIR spectra in calibration and internal validation step

Classes ^a	Figure of merit ^b		
	Rc ²	RMEC	RMECV
BM	0.904	0.140	0.193
ZW	0.974	0,082	0.147
TZ	0.945	0,082	0,147
FBS	0,984	0,063	0,115

^a Investigated classes by PLS-DA.

^b Reported model figures of merit: R2c – R-square in calibration; RMSEC-Root Mean Squared Error in Calibration; RMSECV-Root Mean Squared Error in cross validation.

Table 2 shows the calculated figures of merit of the results obtained by the PLS2-DA model using the calibration samples. A correlation between measured and predicted classes (R2 around 0.96 and 0.98 in all cases) and low prediction errors (RMSEC between 0.049 and 0.08) were obtained.

c- Predicting geographical origin of new samples

The predictive ability of PLS2-DA model using MIR data was tested on 14 new samples, not used in the calibration step. These include four samples from TAGZIRT, three samples from Zawit Cheikh, three samples from BENI MELLAL and four samples from FEKIH BEN SALAH. PLS-DA assigns an oil sample to a particular oil classes if the predicted value is comprised between 0.5 and 1.5 for that class. Table 3 shows the classification results with the comparison between known and predicted values for the four origins.

Table 3 shows that all samples from TZ, FBS, ZW and BM for the validation data set were correctly classified. This means that a 100% accurate classification was achieved, i.e. all oil olive spectra of the validation data set matched correctly to the four corresponding classes. PLS2-DA predicted values were always very close to 0 or 1. These results confirm that the predictive ability of the developed PLS2-DA model has been satisfactory. Therefore, it has been concluded again that MIR spectroscopy coupled to the PLS2-DA chemometric method could be successfully used to discriminate olive oils origins.

Table 3: Prediction of olive oil regions by chemometric analysis of MIR spectra

Samples	Class 1: ZW		Class 2: BM		Class 3: TZ		Class 4: FBS	
	y-predicted	y-reference	y-predicted	y-reference	y-predicted	y-reference	y-predicted	y-reference
TZ	0.0854	0.000	-0.1098	0.000	0.9621	1.000	0.0660	0.000
TZ	0.0792	0.000	-0.1055	0.000	0.9364	1.000	0.0925	0.000
TZ	0.0607	0.000	-0.0950	0.000	0.9727	1.000	0.0637	0.000
TZ	-0.0320	0.000	0.0076	0.000	1.0800	1.000	-0.0619	0.000
ZW	1.0185	1.000	0.0566	0.000	-0.0138	0.000	-0.0649	0.000
ZW	1.0448	1.000	0.0519	0.000	-0.0575	0.000	-0.0417	0.000
ZW	1.0448	1.000	0.0519	0.000	-0.0575	0.000	-0.0417	0.000
FBS	0.0676	0.000	0.2555	0.000	0.0859	0.000	0.5429	1.000
M2FBS	0.0934	0.000	-0.0930	0.000	-0.0327	0.000	1.0322	1.000
FBS	-0.1256	0.000	0.1798	0.000	0.0567	0.000	0.8922	1.000
FBS	-0.0383	0.000	-0.0489	0.000	0.1252	0.000	0.9595	1.000
BM	-0.0045	0.000	0.8558	1.000	0.0316	0.000	0.1263	0.000
BM	0.0015	0.000	0.8311	1.000	0.0246	0.000	0.1522	0.000
BM	0.0035	0.000	0.8672	1.000	0.0066	0.000	0.1305	0.000

4. Conclusion

The discrimination between olive oils from four Moroccan geographical areas was performed by a chemometric modeling procedure using the MIR data recorded by analysis of olive oils. Each area was identified on the basis of different spectral information spread along the selected 4000-600 cm⁻¹ spectral range. MIR analysis has proved to be rapid and simple, requiring no chemical pre-treatment of the samples. In fact, the infrared spectrum is able to give a complete called "finger print" of olive oil, which contains its intrinsic quality influenced by several factors such as environment, horticultural techniques and cultivar origin. The application of the PLS-DA algorithm on the samples of a prediction set has allowed a classification with an accuracy of 100%. In conclusion, the spectroscopic methods could represent a reliable, cheap and fast classification tool, able to verify the origin of olive oils and without requiring chemical analyses for discrimination.

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