

# Foliage moisture content and spectral characteristics using near infrared reflectance spectroscopy (NIRS)

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ABSTRACT: Foliage moisture content (FMC in % dry-weight) was measured in seven Mediterranean evergreen species during summer 2001. All samples were dried, ground and scanned using a NIR spectrophotometer. Two methods of calibration for FMC were compared, one using stepwise regression on selected wavelengths and the other using partial least squares (PLS) regression that takes into account all the spectral information. The PLS regression method provided a better accuracy and was thus chosen. For every species except one, PLS calibrations between spectral data and FMC values were accurate:  $r^2$  of the linear regression between measured values and values predicted by the model varied from 0.93 to 0.99, standard error of cross validation (SECV) values were 3 to 6 times lower than the standard deviation (SD) of the reference values. PLS Calibration of FMC in all seven species combined was also accurate:  $r^2 = 0.94$ ,  $SD/SECV > 3$ . It is thus possible to measure the initial moisture content of a fresh foliage sample from its spectral characteristics when dried, whatever the species.

## 1 INTRODUCTION

Under a Mediterranean-type climate, hot dry summers make forests and shrublands very susceptible to wildfires. Current fire prevention systems aim at forecast and locate fire risk, and one of the important parameters to take into account is fuel moisture content. Satellite data could be a prime tool to measure spatial and temporal variations in vegetation moisture content. Nowadays, Earth surveillance satellites explore the environment with a better spatial, spectral and temporal accuracy that should allow the assessing of plant physiological parameters like moisture content using the visible and infrared parts of the electromagnetic spectrum. This study takes place in the larger framework of a research aiming at improving the prediction of foliage moisture content from satellite data, and thus the estimation of the short-term fire risk.

In this study, relations between moisture content of foliages and their spectral characteristics in the visible, near infrared and part of short wave infrared regions have been studied. Near infrared reflectance spectroscopy (NIRS) is a technique that originally measured spectral absorbance in the NIR domain only (hence its name), but now covers a wider spectral band ranging from 400 to 2500 nm. In this range, light reflected by the organic matter gives a unique signature with important biochemical information about character and number of functional groups such as -CH, -OH, and -NH chemical bonds. Chemometric developments allow to unravel spectra and to calibrate NIRS

signal, i.e., to relate the spectra of samples to their reference values. As the spectral reflectance in the visible and near infrared regions is closely related to the biochemical composition of a sample, NIRS is widely used to determine plant biochemical composition (Norris *et al.* 1976, Card *et al.* 1988, McLellan *et al.* 1991a & b, Joffre *et al.* 1992, Lacaze & Joffre 1994, Bolster *et al.* 1996, Foley *et al.* 1998, Gillon *et al.* 1999a). It has also been used to measure some more integrative parameters, like leaf tissue thickness and leaf mass per area (Ourcival *et al.* 1999), leaf age (Meuret *et al.* 1993), calorific value of forest fuels (Gillon *et al.* 1997), litter quality (Gillon *et al.* 1999b, Joffre *et al.* 2001) and stage of decay (Gillon *et al.* 1993), or some soil biological properties (Nilsson *et al.* 1992, Fritze *et al.* 1994). However, this method has never been used to measure foliage moisture content, a physiological state of leaves which is related to their biochemical characteristics.

To this aim, the spatial and temporal changes in the foliage moisture content (FMC) of seven Mediterranean species have been measured during summer 2001, on different sites near Montpellier and in the Maures massif, southern France. NIRS analyses were conducted on these samples when dried. Our objectives were therefore (1) to evaluate if the NIR spectral data of these samples when dried were related to the initial moisture content of the fresh foliage, in each species, and whatever the species, and (2) if the determination of the foliage moisture content from the spectral data was accurate and sensitive to spatial and seasonal variations.

## 2 MATERIAL AND METHODS

### 2.1 Data sets

Foliage samples from seven Mediterranean evergreen species, *Quercus ilex* (QI), *Q. coccifera* (QC), *Cistus albidus* (CA), *Juniperus oxycedrus* (JO), *Spartium junceum* (SJ), *Arbutus unedo* (AU), and *Erica arborea* (EA), were collected once or twice a week on the same sites during summer 2001. QI, QC, CA, JO and SJ samples were collected in a limestone region near Montpellier. AU and EA samples were collected in three 1,000 m<sup>2</sup> sites (A, B and C) lying 300 m apart on a granite upland of the Maures massif. Foliage samples were cut at midday, immediately put into hermetic boxes, rapidly brought back to the laboratory and weighed; they were then oven-dried at 60°C during 24 h, and weighed again. Foliage moisture content (FMC) was calculated in % dry-weight. At each sampling date in each site, FMC of one sample of QI and QC, of five replicate samples of CA, JO and SJ, and seven replicate samples of AU and EA were measured. For each of the QI, QC, CA, JO and SJ series, NIRS analyses were conducted on at least 50 samples selected to cover the whole range of the FMC values in each species. For the AU and EA series, NIRS analyses were conducted on the mixed seven replicate samples. Thus, NIRS analyses took into account a total of 416 samples belonging to seven specific sets.

### 2.2 NIRS analysis

All these 416 dried samples were ground in a cyclone mill through a mesh size of 1 mm, and scanned using a NIR spectrophotometer (NIRSystems 6500). For each measurement, 32 scans were made, at 2-nm intervals over a range from 400 to 2500 nm, to produce a mean spectrum with 1050 data points. The spectrum of the apparent reflectance ( $R$ ) was evaluated by internal software relative to a ceramic standard. The software further processed the data and stored them in absorbance units ( $A$ ) equal to  $\log(1/R)$ . Data analysis was conducted using the ISI software system (Shenk & Westerhaus 1991a).

### 2.3 Calibration procedures

The calibrations involve search for predictive relationships between spectral data and FMC reference values. Calibration equations are mathematical transfer functions built using reference and spectral values of the calibration sample set and used to predict an unknown quantitative value  $Y$  from available spectroscopic measurements  $X$  (Martens & Naes 1989).

### *Comparison of different calibration methods*

Two methods of calibrations for FMC were compared, one using stepwise regression on selected wavelengths and the other using partial least squares (PLS) regression that takes into account all the spectral information. All samples from the seven species were used and the data set was split into a calibration set containing two-thirds of the samples (n=269) over which the calibrations were performed allowing the calculation of a standard error of calibration (SEC), and a validation set containing one-third of the samples (n=135) over which the calibration equations were applied to obtain a standard error of prediction (SEP). Stepwise regression calibrations and PLS calibrations were developed and compared. For each regression 36 models, representing four pre-treatments x three transformations x three spectral regions, were applied to the data. The four pre-treatments correspond to no pre-treatment, standard normal variate (SNV), de-trending transformation, and SNV + de-trend (Barnes *et al.* 1989). Pre-treatment of the spectra by calculation of the SNV transformation scales each spectrum to have a standard deviation of 1.0 to help to reduce particle size effects. De-trending removes the linear and quadratic curvature of each spectrum with the use of a second-degree polynomial regression. The three transformations applied correspond to raw absorbance data, first-order and second-order derivative. In addition, three series of calibrations using different spectral regions were produced, the first on the entire spectrum, (400-2500 nm), the second on the near-IR and short waves IR (1100-2500 nm), and the third on the visible and near-IR (400-1100 nm).

Stepwise regression is performed by selecting the wavelength that is most highly correlated with the reference values, and adding it to the equation. The second wavelength is added by calculating partial correlations with all other wavelengths and selecting the wavelength with the highest partial correlation. The process continues until the addition of a wavelength makes no further improvement to the explanation of variation in the reference values (F value significant at 0.01). After each wavelength is added to the equation, the program re-evaluates all wavelengths in the equation before continuing (Windham *et al.* 1989, Shenk & Westerhaus 1991b). To avoid over-fitting, the number of selected wavelengths was limited to 14.

The PLS method (Martens & Jensen 1982, Shenk & Westerhaus 1991b) uses all the spectral information, unlike the stepwise regression method which uses only a small number of wavelengths (Windham *et al.* 1989). PLS is a combination of PCA and multiple linear regression. By reducing the large set of raw spectral data into a small number of orthogonal factors, PLS avoids problems of over-fitting and collinearity (Martens & Naes 1989). The number of terms used in the equations was limited to 14.

### *PLS calibration of FMC*

After comparison of the results of the two methods of calibration, the PLS regression method was chosen. Calibrations were then developed for FMC on whole sets, first for every specific set (specific calibrations), then for all species combined (global calibration). Cross validation was used to estimate the optimal number of terms in the calibration to avoid over-fitting. This consisted in selecting three-quarters of the samples to develop the model and one-quarter for the prediction. The algorithm was repeated four times, and all the residuals of the four predictions were pooled to provide a standard error of cross validation (SECV) on independent samples. The minimum SECV determines the number of terms to be used. Samples with large cross-validation residuals are usually omitted, and the cross-validation performed again. The cycle of omitting samples with large residuals, followed by another cross-validation, was performed twice. The final model was then recalculated with all the samples to obtain the standard error of calibration (SEC). For each calibration, six mathematical treatments, corresponding to the first and second derivative and a gap of 4, 8, 12 data points, were compared. After comparison of the results of various treatments, the calibration equation that gave the best results in terms of standard error of cross validation (SECV) was selected.

SEC is not the best predictor of accuracy because it only describes how well the reference values are fitted with the calculated regression line (Shenk & Westerhaus 1996). In contrast, SECV is a true estimate of the prediction capability of the equation because it is calculated on independent

samples. According to Williams (1987), if the SECV approaches the standard deviation (SD) of the reference values, NIR calibration is not very efficiently predicting; a ratio  $SD/SECV > 3$  seems a reasonable threshold above which to use NIRS calibration.

To compare the accuracy of the global equation to that of the specific equations, the FMC values of each specific set were predicted using the global equation and were compared to the measured values, allowing the calculation of the standard error of prediction (SEP). These SEP values can then be directly compared to the SECV values of the specific calibrations.

To evaluate the precision of the NIRS method for measuring FMC, SECV was compared to the reference (weighing) method error, i.e. the standard deviation of differences between duplicate samples measured using the reference method (Williams 1987). To this aim, we used the AU and EA sets where seven duplicate samples were collected in each site at each sampling date and measured using the reference method.

To evaluate the sensitivity of the NIRS method for measuring FMC variations, we compared the spatial and seasonal variability of FMC calculated from NIRS data to that calculated from measured data. To this aim, we used the same AU and EA sets, and we compared their standard deviation in the three sites at each sampling date, and in each site at the 25 sampling dates, respectively.

### 3 RESULTS

#### 3.1 *Foliage moisture content*

During the summer, the measured FMC values for all seven species varied from 38% to 146% i.e., a range of variation of 108%; but this range depended on the species (Table 2 and Fig. 2). It was low in QI (23%), moderate in QC, CA, JO and SJ (42 to 64%), and large in AU and EA (81 to 85%).

#### 3.2 *Relation between FMC and spectral absorbance data*

There were close relationships between FMC and spectral data from all species combined (Fig. 1). Several spectral regions in the visible, near-near-IR and near-IR domains were significantly correlated with FMC.

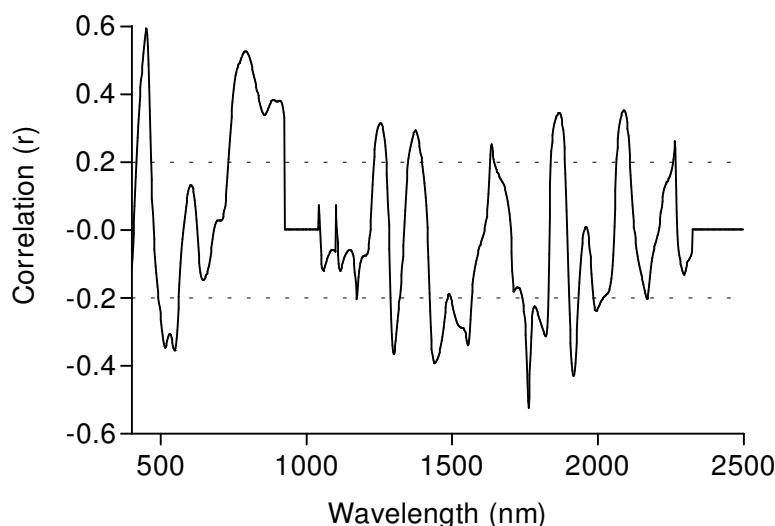


Figure1. Correlogram of FMC with spectral data (Mathematical treatment of the spectral data: 2,20,20).

### 3.3 Comparison between stepwise and PLS regression calibrations

PLS regression procedures gave better results than stepwise regression procedures (Table 1). In PLS regressions, the standard error of calibration (SEC) ranged from 5.42 to 9.89 (7.25 to 13.19 in stepwise regressions),  $r^2$  from 0.80 to 0.94 (0.65 to 0.89), and SEP from 6.60 to 12.12 (7.97 to 14.99). For the best combination of pre-treatments and spectral regions, the ratio between SEC obtained by stepwise regression and that obtained by PLS regression was 1.34. In the stepwise regression calibrations, the best calibration was obtained from the first derivative data of the entire spectrum after a complete pre-treatment, and the best prediction from the raw data of the near-IR region without pre-treatment (Table 1).

In the PLS regressions, using the first or the second derivative in all cases improved the results as compared to regressions calculated from raw spectral data (Table 1). For the best combination of pre-treatments and spectral regions, the ratio between SEC obtained using raw spectral data and that obtained using first and second derivative data were 1.20 and 1.37, respectively. The results were almost always better when the full spectrum was taken into account, rather than just the near-IR region or the visible region. In some cases (raw data and second derivative), the results of prediction obtained in the near-IR region were slightly better than over the entire spectrum. By contrast, the use of visible region alone did not allow calibration with comparable accuracy. Any pre-treatment (SNV, de-trend, or SNV + de-trend) always improved the results of the predictions. As a

consequence of these different tests of methodologies, we then performed PLS regression calibrations using the first-order and second-order derivative data from the whole spectrum after a complete pre-treatment (SNV + de-trend).

### 3.4 Specific PLS calibrations

PLS calibrations for FMC were first monitored on each species. Except for QI, the specific calibrations between spectral data and FMC values were accurate:  $r^2$  of the linear regression between measured values and values predicted by the model varied from 0.93 to 0.99, SECV values from 2.0% to 5.7%, i.e. values 2.9 to 5.8 times lower than the standard deviation (SD) of the measured values (Table 2 and Fig. 2). For QI, probably due to the low range of FMC values, the calibration of FMC was less accurate:  $r^2 = 0.85$ , SECV = 2.7%, SD/SECV = 1.9.

### 3.5 Global PLS calibration

PLS calibration of FMC in all seven species combined was also accurate:  $r^2 = 0.94$ , SECV = 6.3%, SD/SECV = 3.6 (Table 2 and Figure 2). However, the SECV value was larger than all the SECV values of the specific calibrations.

### 3.6 Global PLS equation applied to the specific sets

For every species, the FMC values predicted using the global equation were generally less accurate than those predicted using the specific equations. When applying the global equation to the specific sets (Table 3),  $r^2$  values of the linear regression between measured and NIRS predicted values were lower than in specific calibrations (Table 2), and the SEP values were larger than the SECV values of the specific calibrations. However, in each species, FMC values predicted using the global equation was of the same range as the measured values and these predicted values changed with time during summer like the measured values (Fig. 3 & 4). In AU and EA, whose FMC markedly recovered after the rains of end September, NIRS predicted values also re-increased, but with a little delay compared to the measured values, especially in the drier B and C sites (Fig. 4).

### 3.7 Precision of NIRS predicted FMC

The reference method error was calculated from the seven field replicate samples of AU and EA collected in each site at each sampling date (Table 4). Compared to the reference method errors, the SECV values of the two specific calibrations (Table 2) were 1.8 and 2.1 times larger for AU and EA, respectively, and the SEP values obtained by applying the global equation to the same sets

(Table 3) were 1.8 and 2.4 times larger. NIRS prediction of FMC was then about twice less precise than the reference (weighing) method.

Table 1. Statistics for the stepwise regression and PLS regression calibrations of FMC (% DW). Calibration sample set n = 269 (range 37.80-139.45 ; mean 88.85 ; SD 22.19), validation sample set n = 135 (range 41.30-146.16 ; mean 86.71; SD 22.39). Math treatment indicates the transformation of spectral data : the first number is the order of the derivative function, the second is the segment length (nm) over which the derivative was taken, and the third the segment length over which the function was smoothed. SEC, standard error of calibration ; SEP, standard error of prediction. Wgths, number of wavelengths used in the stepwise regression model; Terms, number of terms used in the PLS calibration model.

Math treatment	Spectral region/ Pre-treatment	Stepwise regression calibration				PLS regression calibration			
		SEC	r <sup>2</sup>	SEP	Wgths	SEC	r <sup>2</sup>	SEP	Terms
<b>0,4,4</b>	<b>400-2500 nm</b>								
	None	8.12	0.87	9.28	10	9.11	0.83	11.20	12
	SNV	9.88	0.80	11.78	8	8.07	0.87	9.11	14
	De-trend	9.86	0.80	10.93	8	8.25	0.86	8.38	14
	SNV & De-trend	7.82	0.88	9.16	11	7.43	0.89	8.55	14
	<b>1100-2500 nm</b>								
	None	8.34	0.86	7.97	12	8.66	0.85	8.28	14
	SNV	8.67	0.85	8.70	14	8.64	0.85	8.52	14
	De-trend	9.30	0.82	9.75	13	8.75	0.85	8.03	14
	SNV & De-trend	13.19	0.65	14.99	6	8.61	0.85	8.42	14
	<b>400-1100 nm</b>								
	None	8.81	0.84	11.93	14	9.89	0.80	11.47	12
	SNV	11.99	0.71	12.89	4	9.67	0.81	11.25	13
	De-trend	11.53	0.73	12.30	6	9.40	0.82	11.23	14
	SNV & De-trend	9.04	0.83	10.91	11	8.82	0.84	10.93	14
	<b>1,4,4</b>	<b>400-2500 nm</b>							
None		9.85	0.80	11.06	7	6.33	0.92	7.68	14
SNV		7.74	0.88	8.55	10	6.17	0.92	6.60	14
Detrend		10.41	0.78	12.02	6	6.56	0.91	6.67	14
SNV & De-trend		7.25	0.89	8.91	13	6.41	0.92	6.76	14
<b>1100-2500 nm</b>									
None		7.79	0.88	8.94	14	6.76	0.91	7.14	14
SNV		8.70	0.85	9.02	9	6.94	0.90	7.09	14
De-trend		10.98	0.76	13.00	6	6.96	0.90	7.19	14
SNV & De-trend		7.98	0.87	8.95	12	6.95	0.90	7.15	14
<b>400-1100 nm</b>									
None		8.50	0.85	11.22	10	8.07	0.86	10.79	13
SNV		7.88	0.87	11.00	13	7.96	0.87	10.54	12
De-trend		11.89	0.71	13.03	4	8.54	0.85	10.76	12
SNV & De-trend		8.34	0.86	10.01	14	7.81	0.88	9.84	13
<b>2,4,4</b>		<b>400-2500 nm</b>							
	None	7.86	0.88	10.67	14	6.01	0.93	8.16	13
	SNV	8.30	0.86	10.68	12	5.88	0.93	7.84	13
	De-trend	7.41	0.89	9.32	14	5.42	0.94	8.23	14
	SNV & De-trend	7.79	0.88	10.99	13	5.74	0.93	7.81	13
	<b>1100-2500 nm</b>								
	None	8.67	0.85	8.91	12	6.67	0.91	7.77	14
	SNV	9.03	0.83	9.94	12	7.02	0.90	7.83	13
	De-trend	9.91	0.80	11.04	10	6.67	0.91	7.76	14
	SNV & De-trend	8.67	0.85	9.40	12	7.02	0.90	7.84	13
	<b>400-1100 nm</b>								
	None	8.50	0.85	11.51	14	8.90	0.84	11.86	7
	SNV	9.43	0.82	12.00	14	8.86	0.84	11.78	6
	De-trend	9.18	0.83	11.19	10	8.94	0.84	12.12	7
	SNV & De-trend	8.70	0.85	11.62	12	8.68	0.85	12.07	7

Table 2. Statistics of the specific PLS calibrations for FMC (% DW) in *Quercus ilex* (QI), *Q. coccifera* (QC), *Cistus albidus* (CA), *Juniperus oxycedrus* (JO), *Spartium junceum* (SJ), *Arbutus unedo* (AU) and *Erica arborea* (EA), and of the global PLS calibration (all species combined). SD, standard deviation of measured values ; SEC, standard error of calibration, ; SECV, standard error of cross validation ; Math, indicates the mathematical transformation of spectral data : the first number is the order of the derivative function, the second is the segment length in data points over which the derivative was taken, and the third is the segment length over which the function was smoothed.

Species	n	Mean	Range	SD	SEC	r <sup>2</sup>	SECV	SD/SECV	Math
QI	53	67.07	55.58-78.17	5.05	1.95	0.85	2.72	1.86	1,8,4
QC	50	81.75	63.48-108.71	9.80	1.41	0.98	3.41	2.87	2,4,4
CA	50	90.29	60.19-123.69	18.45	2.43	0.98	5.19	3.55	2,4,4
JO	47	87.36	68.41-110.11	10.99	1.44	0.98	2.02	5.44	2,12,8
SJ	50	109.88	88.67-133.04	12.55	3.28	0.93	3.78	3.32	1,12,8
AU	73	106.21	61.59-146.16	22.40	4.25	0.96	5.69	3.94	1,4,4
EA	73	73.10	37.80-119.01	23.12	2.70	0.99	3.95	5.84	1,4,4
All sp.	404	88.14	37.80-146.16	22.35	5.55	0.94	6.28	3.56	2,8,8

Table 3. Prediction statistics of FMC (% DW) in each specific set (same samples than in Table 2) using the global PLS calibration equation. SEP, standard error of prediction.

Species	n	SEP	r <sup>2</sup>
QI	53	5.62	0.38
QC	50	4.47	0.79
CA	50	8.91	0.77
JO	47	5.12	0.80
SJ	50	5.22	0.83
AU	73	5.70	0.94
EA	73	4.62	0.96

### 3.8 Sensitivity of NIRS predicted FMC

Using the measured data, FMC spatial variability in AU and EA was about 4 times higher than the reference method error (Table 4), and seasonal variability was higher than spatial variability; this was particularly demonstrative in EA. Using the NIRS predicted data, spatial and seasonal variability was of the same magnitude as that calculated from the measured data. However, this variability was generally a little lower when FMC was predicted using the global equation than the specific equations.

Table 4. Variability of FMC calculated from the measured values and from the NIRS predicted values using the specific PLS equations or the global PLS equation. Reference method error = SD of the measured FMC values of the 7 replicates collected in each site at each sampling date. Spatial variability = SD of the values in the three sites at each sampling date. Seasonal variability = SD of the values at the 25 sampling dates in each site.

	Measured values	NIRS predicted values	
		Specific equations	Global equation
<b>Arbutus unedo</b>			
Reference method error	3.26		
Spatial variability	13.13	13.60	11.72
Seasonal variability	19.87	20.31	19.02
<b>Erica arborea</b>			
Reference method error	1.89		
Spatial variability	7.36	6.64	6.88
Seasonal variability	22.02	22.01	21.16

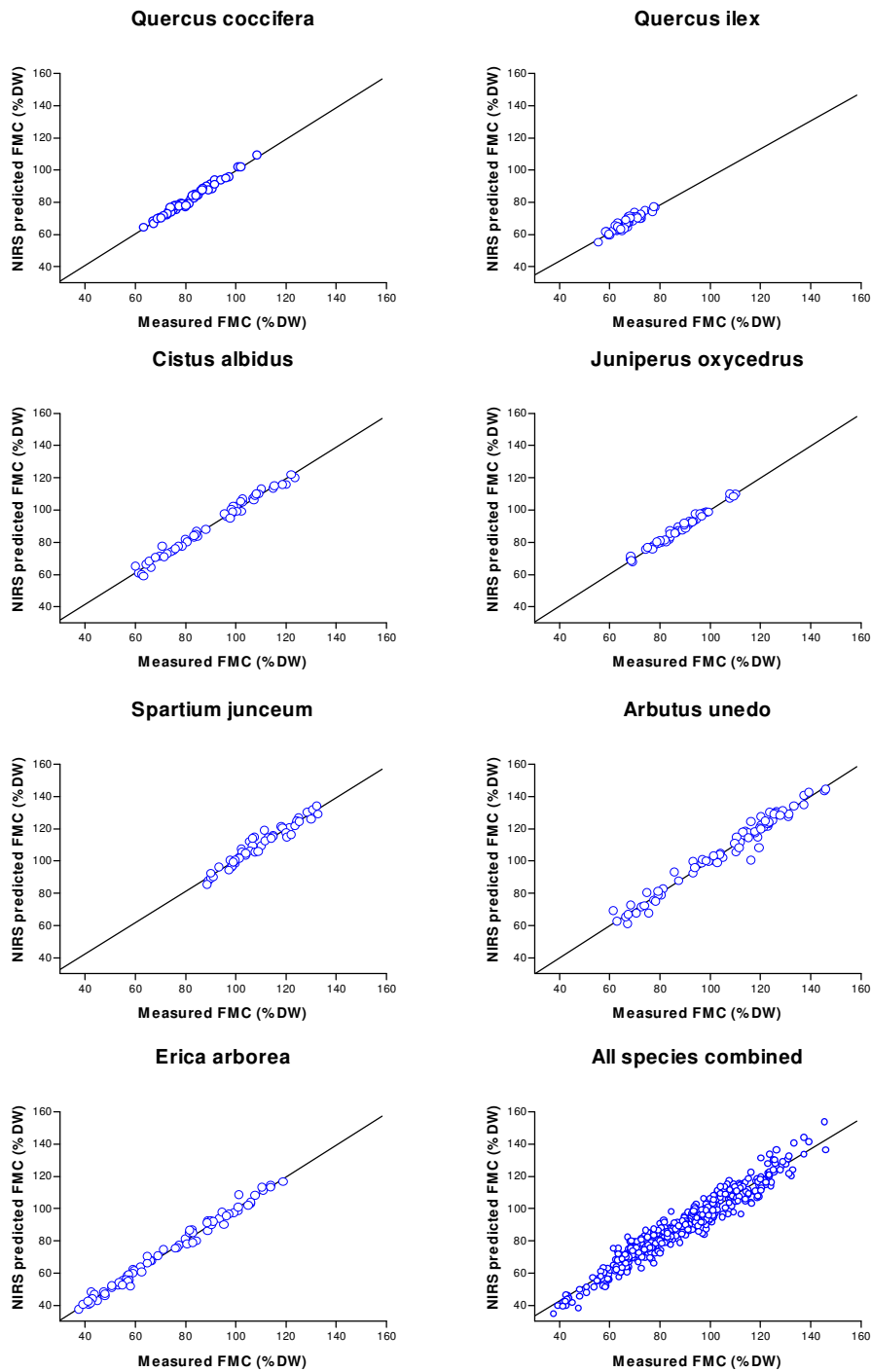


Figure 2. Relationship between measured values of FMC and NIRS predicted values using the specific PLS calibration equations for each species and the global PLS calibration equation for all species combined.

#### 4 DISCUSSION

Foliage from the seven species studied allowed to take into account a large spectral variability range, quite representative of the Mediterranean plant cover. The variation of the FMC values measured during this study was particularly large, from 38% to 146%, especially in the Maures region where a poor rainfall was recorded during summer 2001 (Fig. 4). Sampling of two different species, *Arbutus unedo* and *Erica arborea*, from June to October in three 300 m distant sites with

different soil water availability, allowed to estimate spatial and seasonal variability in foliage moisture content and the sensitivity of its measurement by NIRS.

It was possible to accurately relate the initial moisture content of a fresh foliage sample to its spectral characteristics when dried. As already pointed out by Joffre *et al.* (1992), Bolster *et al.* (1996) and Ourcival *et al.* (1999), our results showed that the PLS method of calibration consistently provided a better accuracy than stepwise methods. Theoretical chemometrics studies demonstrated that the stepwise regression predictor has deficient performance when there is collinearity in spectral data (Martens & Naes 1989). Using all the spectral information through multivariate analysis of derivative spectra allowed us to solve the collinearity problem by using a small number of orthogonal regressors, and to build efficient equations for FMC.

Using PLS methods, the calibration of FMC in each species was accurate when a sufficient range of values was available. In *Quercus ilex* only, leaf water content did not vary enough during summer to perform an accurate relation between the spectral absorbance and FMC. Interestingly NIRS calibration of FMC was also accurate in all the seven species grouped. This means that some spectral characteristics of the leaves vary in the same way in the different species according to their moisture content. It could be objected that the calibration equations take into account the spectral characteristics related to the ageing process in leaves because, in most samples used in the calibrations, FMC values decreased with time during summer, thus with leaf age. However, the recover of NIRS predicted FMC after the rains of end September in *Arbutus unedo* and *Erica arborea* (Fig. 4), without new leaves appearance, demonstrated that NIRS equations were actually related to FMC. These results show that it is possible to measure the initial moisture content of a fresh foliage sample from its spectral characteristics when dried, and this is true whatever the species.

These results mean that some biochemical properties of the leaves vary in the same way in the different species according to the physiological state induced by water limitation. However, a same level of soil water does not lead to a same level of foliage moisture content whatever the species; *Arbutus unedo* and *Erica arborea* for example, both sampled in the same sites at the same dates, had different foliage moisture contents (Fig. 4). It is well-known that leaf moisture content at equilibrium varies according to the species, that's why water stress is often expressed as leaf water content relative to that measured in a water saturated situation. Drought tolerance and resistance also vary and each species responds to water limitation using different mechanisms, like growth inhibition, stomatal closure to reduce water loss, decrease in photosynthesis, respiration and transpiration, leaf area adjustment, changes in physical processes and in carbohydrate and nitrogen metabolism (Smith & Griffiths 1993, Kramer & Boyer 1995). In particular, in plants experiencing water deficits, many changes in physiology and metabolism occur, related to adjustment of metabolism to conditions within the cell and tissue, like synthesis and accumulation of osmotically active solutes, protein and enzymes composition (Lawlor & Cornic 2002). The change observed in the spectral characteristics of the leaves was the expression of these biochemical changes in the plant tissues induced by the progressive water limitation and the decreasing foliage moisture content.

For each species, NIRS prediction of FMC was generally more accurate using the specific equation than the global equation performed from all the seven species grouped. Moreover, the FMC values predicted using the specific equations were more sensitive to spatial and seasonal variability. This result is common in NIRS studies; the spectro-chemical model is simpler when the group of samples have similar spectral characteristics but it must also be representative of all the forms of variation in the material and must possess as wide a range of reference values as possible (Abrams *et al.* 1987, Shenk & Westerhaus 1993, Gillon *et al.* 1999a). Thus, if the aim is to use satellite spectral data, only the global equation is relevant since it takes into account the spectral variability of a range of Mediterranean species and it could be applied to a more spectrally diverse range of material. Moreover, this global calibration equation is effectively predictive according to Williams (1987), since the standard error of cross validation on independent samples was 3.6 times lower than the standard deviation of the reference values, and the standard error of calibration, when the final model was calculated, 4.0 times lower.

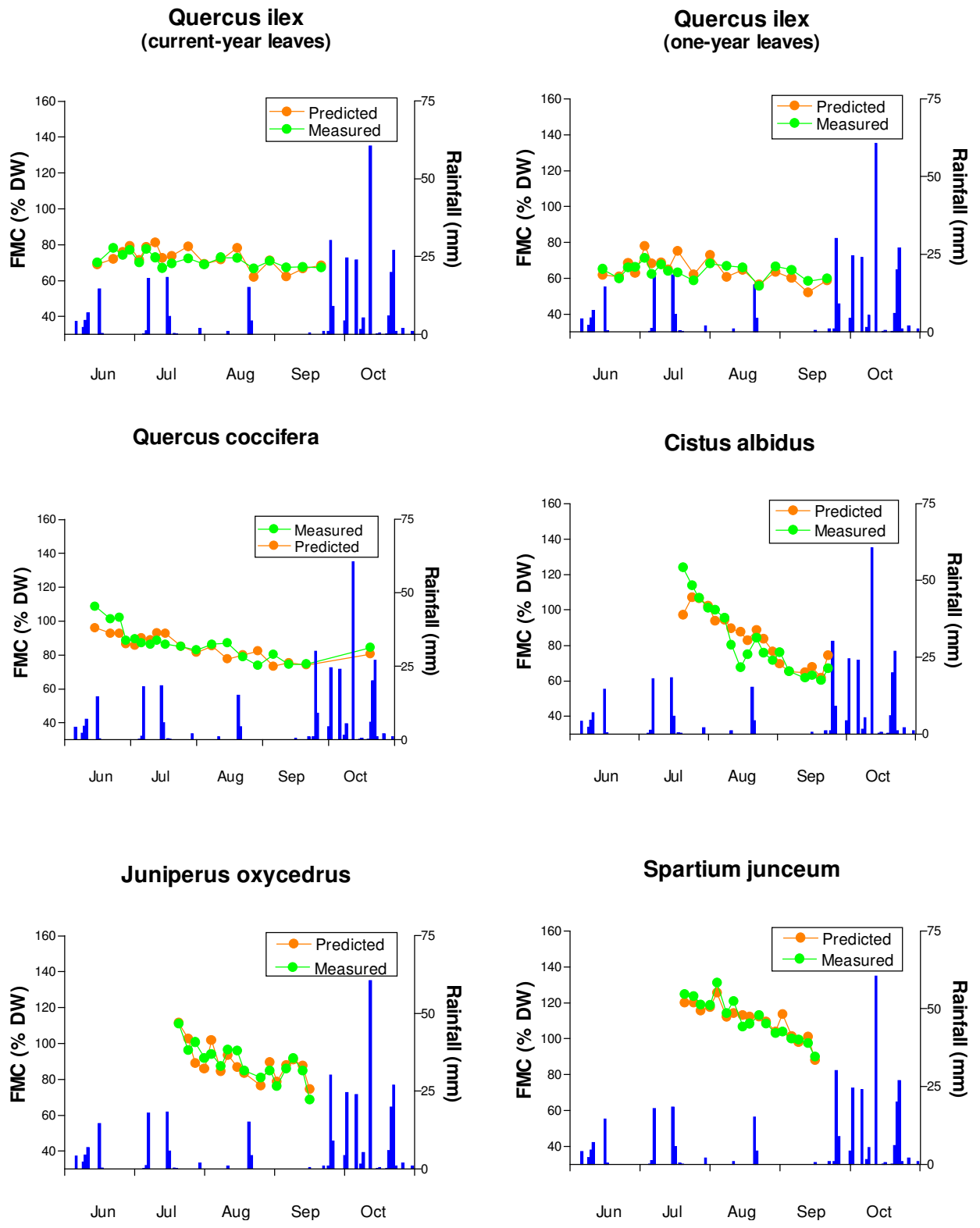


Figure 3. Change with time of the measured values of FMC and the NIRS predicted values using the global PLS calibration equation in the five species sampled near Montpellier.

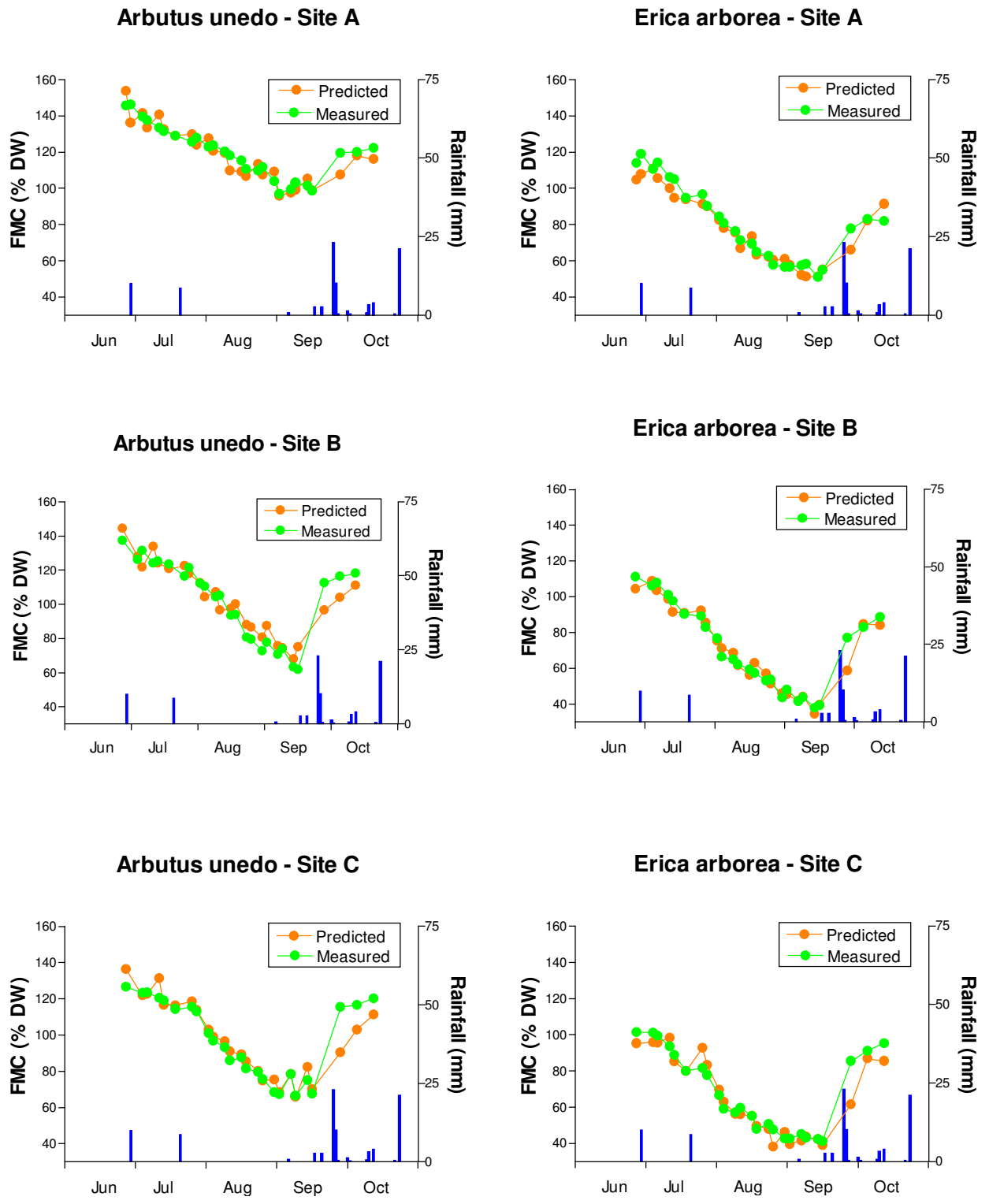


Figure 4. Change with time of the measured values of FMC and the NIRS predicted values using the global PLS calibration equation in *Arbutus unedo* and *Erica arborea* both sampled in three different sites of the Maures massif.

Using the global equation, NIRS measurement of FMC was about twice less precise than the conventional weighing method. However, NIRS provided the same information on foliage moisture content as the conventional method. (1) NIRS measured the same ranges of foliage moisture content in each species as the conventional method; and (2) it measured the same decrease in foliage moisture content with decreasing soil water availability during summer, and the same recover after the first rains at the end of summer. This was particularly demonstrative in *Arbutus unedo* and *Erica arborea*. However, compared to the measured values, we could observe a little delay in the re-increase of the NIRS predicted values in September-October, as if the restoration of the spectral characteristics of the leaves after the first rains, i.e. their biochemical properties, was processing more slowly than the input of water in the leaf tissues. Last, (3) NIRS also demonstrated that it was sensitive to FMC variations; it provided the same information as the conventional method on the spatial and seasonal variability of foliage moisture content.

The aim of this study was not to compare NIRS measurement of FMC with the conventional method –it was about twice less precise, and not faster-; our aim was to evaluate the potentiality of using satellite spectral data, from 400 to 2500 nm, to measure foliage moisture content. It is now clear that, in this spectral range, there are accurate relations between the spectral absorbance (or reflectance) of the leaves and their moisture content. It is highly likely that it would be the same from fresh whole leaves; Card *et al.* (1988), Curran *et al.* (1992) and Lacaze and Joffre (1994) showed that the same biochemical information could be extracted from near infrared spectroscopy of fresh and dried leaves, despite the dominant spectral features related to the absorption of photosynthetic pigments and water. It is thus liable that satellite spectral data do not only contain information on the quantity of water in the plant cover, but also on the vegetation biochemical composition, directly related to its moisture content.

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