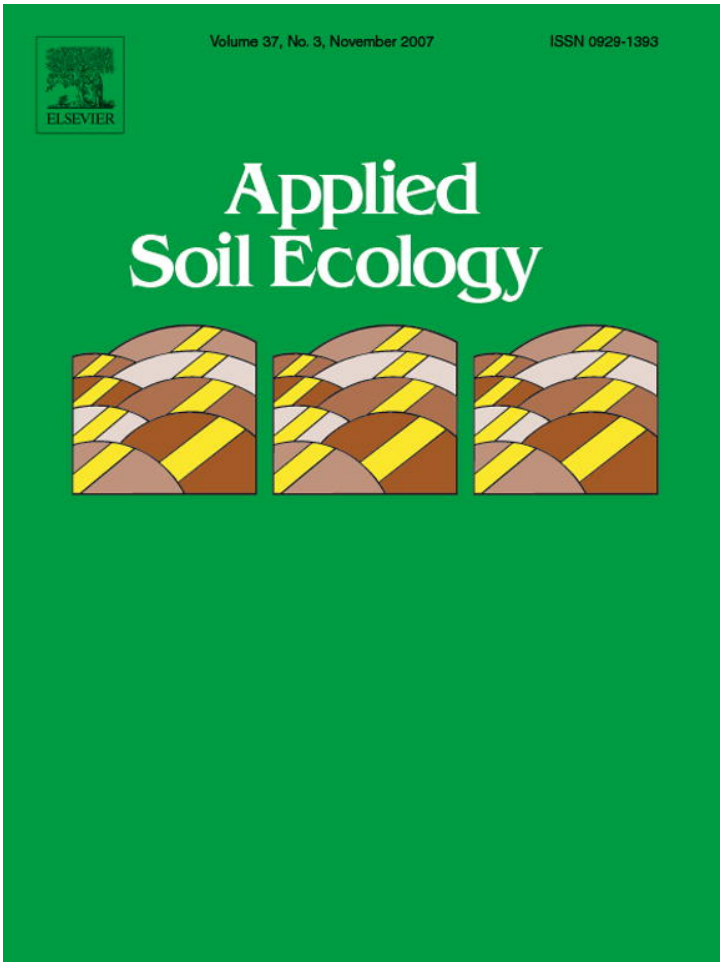

APPLIED SOIL ECOLOGY 37 (2007) 223–232

available at www.sciencedirect.com

ScienceDirect

journal homepage: www.elsevier.com/locate/apsoil

Evaluation of the recovery of microbial functions during soil restoration using near-infrared spectroscopy

Heidy Schimann [a,*], *Richard Joffre* [b], *Jean-Christophe Roggy* [a], *Robert Lensi* [b], *Anne-Marie Domenach* [a]

[a] *UMR EcoFoG, campus agronomique, BP 709, 97387 Kourou, French Guiana*
[b] *CEFE, 1919 routes de Mende, 34000 Montpellier, France*

ARTICLE INFO

Article history:
Received 2 July 2007
Received in revised form
2 July 2007
Accepted 3 July 2007

Keywords:
NIRS
Microbial activities
Respiration
Denitrification
Carbon
Nitrogen
Soil functioning
Restoration

ABSTRACT

Microbial-based indicators, such as C and N contents or microbial functions involved in C and N cycles, are currently used to describe the status of soils in disturbed areas. Microbial functions are more accurate indicators but their measurement for studies at the ecosystem level remains problematical because of the huge spatial variability of these processes and, consequently, of the large number of soil samples which must be analyzed. Our goal was to test the capacity of near-infrared reflectance spectroscopy (NIRS) to predict respiration and denitrification but also carbon and nitrogen contents of soils submitted to various procedures of restoration. To achieve this objective, we took advantage of an experiment conducted on a reforestation system established after open-cast gold mining in French Guiana. In this experimental station, plantations of various ages and various soil textures were at our disposal. Our results showed that both plantations and soil texture had a strong impact on the recovery of soil functioning: carbon and nitrogen contents, respiration and denitrification increased with age of plantation and clay content. Calibrations were performed between spectral data and microbial-based indicators using partial least squares regression (PLS). The results showed that C and N contents were accurately predicted. Microbial functions were less precisely predicted with results more accurate on clayey soils than on sandy soils. In clayey soils, perturbed or restored soils and the year of plantation were discriminated very efficiently through principal component analyses of spectral signatures (over 80% of variance explained on the first two axes). Near-infrared spectroscopy may thus be extended to the prediction of functional soil parameters, but the capacity of this method must be strengthened by expending the databases with other soils in other contexts. The possibility of using NIRS provides many opportunities for understanding both the temporal dynamics and the spatial variability of the recovery of key microbial functions during soil restoration.

1. Introduction

Soil governs plant productivity in terrestrial ecosystems and acts to maintain the equilibrium of biogeochemical cycles through biotransformations (or functions) mediated by living organisms. It has been recognized for many years that microbes are responsible for 80–90% of these functions (Nannipieri et al., 2003).

It is thus of great interest for the sustainability of our environment to assess if the procedures of restoration of sites

* *Corresponding author.*
E-mail address: heidy.schimann@cirad.fr (H. Schimann).
0929-1393/$ – see front matter
doi:10.1016/j.apsoil.2007.07.001

degraded by changes of land-use may allow the soil to partially or totally recover its microbial functions. Microbial-based indicators such as changes in total biomass or in the structure of the total microbial community or of a given group of microorganisms have been often used to describe the soil quality. As an example, dynamics of microbial soil quality along a temporal sequence of restoration have been assessed using these types of indicators (Carter et al., 1997; Schloter et al., 2003). Nevertheless, there is also – and may be overall – a need for microbial-based indicators which could directly account for the functional status of the soil (Dighton and Pierce, 1997; Anderson, 2003). Nevertheless, few studies aiming at evaluating the extent to which the soil has been affected by human activities and/or how it recovered its initial characteristics were based on the assessment of the recovery of soil microbial functions. For example, Carter et al. (1997) and Schloter et al. (2003) described the recovery of soil quality using variables such as C and N mineralization. Even in this case, there is often a lack of clear distinction between "real" function (i.e. the actual fluxes generated by the function at a given moment) and "potential" function (i.e. the maximal capacity of the soil to express a given function), these two types of variables leading different ways of interpretations. The first step of such approaches is to select appropriate microbial functions as relevant indicators of soil functioning. Nevertheless, one of the major limitations is that measurements of microbial functions are often time consuming and require a large number of analysis of soil samples to be representative of a given situation.

Near-infrared or visible–near infrared spectroscopy has been successfully used to predict soil or litter chemical properties with the aim to reduce time and cost in comparison with traditional analytical methods. As examples, these methods have been applied for the prediction of organic matter content (Ben-Dor and Banin, 1995; Couillard et al., 1997), total C and N contents (Chang and Laird, 2002), leaf litter quality (Joffre et al., 2001), litter mass remaining during decomposition stages (Gillon and Joffre, 1993), or microbial biomass (Chodak et al., 2002). On the contrary, there is little information about the capacity of NIRS spectra to predict more functional variables such as C and N mineralization of soils submitted to various disturbances (Ludwig et al., 2002), rates of C mineralization in different size classes of aggregates obtained by physical fractionation of two tropical soils in Kenya (Mutuo et al., 2006), or microbial respiration in soil structures resulting from animals activities (Hedde et al., 2005). Nevertheless, preliminary studies showed that the information contained in a single spectrum enables many different soil variables to be characterized simultaneously (Odlare et al., 2005). For instance, Velasquez et al. (2005) showed that NIRS was able to rank a soil submitted to different land-uses along two axis of PCA, according to physico-chemical characteristics (organic matter, nutrients contents, Al^{3+} saturation and total-P concentration).

Using a soil strongly altered by gold mining activity and subjected to different modes of restoration (reforestation), our objective was to assess the capacity of NIRS to simultaneously describe and predict the evolution of physical (soil texture), chemical (C and N contents) and functional (microbial functions: respiration and denitrification) characteristics. Gold-mining results in drastic modifications of the system and among others, a strong decrease in organic matter content and the creation of patches of contrasted soil granulometry. Therefore, areas predominantly sandy or clayey and submitted to the same reforestation treatments were at our disposal to assess the relationships between recovery of microbial functions and evolution of NIRS spectra.

Potentials of respiration and denitrification were chosen as the model functions because (1) they can be measured with accuracy, (2) both are performed by an abundant and diversified microflora and (3) they are considered as key-functions for the functioning of ecosystems and for the quality of our environment (both functions being involved in the exchange of gas-trace between soil and atmosphere).

2. Materials and methods

2.1. Experimental site

The experimental site was an open-cast gold mine (Mine Boulanger); located in the east of French Guiana, on the Coralie track (4°30N, 52°20W). This mine is part of an experimental reforestation project led by the Forest National Office (ONF) since 1996. The objectives of this project were to limit soil erosion and to recover environmental conditions favorable to plant regeneration in order to re-establish the forest cover. Two chronosequences corresponding to two areas differing in texture were studied (high sand or high clay contents, Table 1). For each chronosequence, plots of 1000 m^2 in average were designed. The fast-growing N-fixing *Acacia mangium* Willd (*Mimosaceae*) was chosen to facilitate the recovery process because of its exceptional rusticity and because its broad crown could rapidly cover the soil. These plots were planted with 3-month-old seedlings of *A. mangium* at different densities (1250, 1100 and 400 individuals ha^{-1}) – and/or different times (1996 and 1999) – Table 1). This result in four treatments: 5S–8S for 5 years old (5S) and 8 years old (8S) plantation on sandy soil, respectively, and 5C–8C for 5 years old (5S) and 8 years old (8S) plantation on clayey soil, respectively. As well, three control plots were sampled: (i) one which was not included on the restoration procedure on the sequence with high sand content (CS), (ii) one which was not included on the restoration procedure on the sequence with high clay content (CC), and (iii) one in the natural forest sandy soil near the mining zone (NF, Table 1).

2.2. Soil sampling strategy

Fifteen soil cores (10 cm × 10 cm × 10 cm and 10 cm depth from the surface) were randomly harvested in each plot in order to be representative of all the surface of one plot. We chose to sample only in the first 10 cm because in tropical forest soils, the majority of the microbial activity stand in the first 10 cm. Samples were air-dried, crushed and sieved at 2 mm, thoroughly homogenized and stored at room temperature (25 °C) until analyses. As sample drying and storage could weakly modify enzymatic activities, it has been verified that these modifications affect soil sample similarly and that, consequently, the relative range of values was not expected to be changed.

Table 1 – Soil properties and vegetation characteristics

Description		Control sand (CS) No plantation	5 years sand (5S) Plantation of *Acacia mangium* 1999	8 years sand (8S) Plantation of *A. mangium* 1996	Control clay (CC) No plantation	5 years clay (5C) Plantation of *A. mangium* 1999	8 years clay (8C) Plantation of *A. mangium* 1996	NF Natural forest
Plantation of *A. mangium*	Density (individuals ha^{-1})	0	1100	1250	0	400	1250	–
	Height (m)	0	12 (0.5)	21.5 (0.7)	0	8.5 (0.4)	20 (0.92)	–
	Mean DBH (cm)[a]	0	nd	21.4 (1.04)	0	nd	21.6 (1.3)	–
	Vegetation cover	25%	65%	97%	10%	60%	98%	–
	Age (years)	0	5	8	0	5	8	–
Soil texture (%)	Clay	19.8 ± 1.7 b	9.0 ± 2.2 b	11.0 ± 2.4 b	51.7 ± 1.4 a	40.9 ± 0.9 a	44.9 ± 1.5 a	25.8 ± 1.4 b
	Loam	16.2 ± 0.9 b	18.9 ± 5.4 b	18.9 ± 2.7 b	15.5 ± 0.9 b	23.2 ± 0.6 c	19.7 ± 1.5 b	5.7 ± 0.6 a
	Sand	64 ± 2.7 b	72.1 ± 7.3 b	70.1 ± 5.1 b	32.7 ± 1.8 a	35.9 ± 1.6 a	35.4 ± 0.8 a	68.5 ± 1.5 b
	%N	0.02 ± 0.02 a	0.06 ± 0.05 b	0.12 ± 0.14 c	0.03 ± 0.01 a	0.04 ± 0.02 a,b	0.23 ± 0.02 d	0.18 ± 0.09 d
	%C	0.32 ± 0.002 a	0.87 ± 0.004 b	1.81 ± 0.01 c	0.36 ± 0.001 a	0.51 ± 0.01 a,b	3.35 ± 0.013 d	2.67 ± 0.005 d
	C:N	11.9 ± 0.4 a	15 ± 1.5 c	15.6 ± 1.9 c	13.2 ± 0.9 b,c	12.1 ± 0.5 a,b	14.5 ± 1.2 c	14.6 ± 1.2 c

For soil texture, values for clay, sand, loam, %N, %C and C-to-N ratios are means ± standard error ($n = 5$), followed with different letters if significantly different (one-way ANOVA, unequal n HSD test of Tukey, at $p < 0.05$) CS: control on sandy soil; 5S: 5 years old plantation on sandy soil; 8S: 8 years old plantation on sandy soil; CC: control on clayey soil; 5C: 5 years old plantation on clayey soil; 8C: 8 years old plantation on clayey soil; NF: natural forest soil.

[a] Diameter at breast height.

2.3. Soil physico-chemical characteristics

Five samples by plots among the whole soil samples (i.e. 15 all in all) were randomly chosen to determine texture and carbon and nitrogen content. Soil texture was determined by the hydrometer method (Gee and Bauder, 1986). Carbon and nitrogen contents were determined with a Perkin-Elmer elemental analyzer (PE 2400 CHN) on 91 milled soil samples.

2.4. Denitrifying enzyme activity and substrate induced respiration measurements

Denitrifying enzyme activities of soil (DEA) were measured by the methods of Tiedje (1984) and Lensi et al. (1985). For each sample, 10 g of sieved and air-dried soil were placed in a 150 ml plasma-flask and sealed with rubber stoppers. In each flask, air was removed with a vacuum-pump and replaced by 90% of helium and 10% of acetylene to inhibit the N_2O-reductase. Six milliliters of a solution (quantity ensuring of 100% saturation of the soil) containing 100 μg of N (from KNO_3), 2 mg of C (glucose and glutamic acid) per g of soil was added. These C and N addition were considered as non-limiting for denitrifying enzyme functioning. The flasks were incubated 4 h at 28 °C. This short incubation time was used in order to avoid any de novo enzymatic synthesis or cellular growth. Two hundred microliters of gas sample were analyzed for N_2O on a gas chromatograph equipped with an electron capture detector (VARIAN 3800-CP, Les Ulis, France).

Substrate induced respiration (SIR) was measured using a method from Anderson and Domsch (1978) which we modified as follows. For each sample, 10 g of sieved and air-dried soil were placed into a 150 ml plasma-flask. A solution containing 2 mg of C (glucose) per g of soil was added to ensure 80% of the water holding capacity of the soil. The flasks were incubated 4 h at 28 °C. Two hundred microliters of gas sample were analyzed for CO_2 using a gas chromatograph equipped with a microcatharometer (VARIAN 4900-GC, Les Ulis, France).

2.5. Spectroscopic measurements

All soil samples were scanned with a NIRSystem 6500 spectrophotometer (NIRSystems Inc., Silver Spring, MD, USA). Each sample was packed into a sample cell with a quartz-glass cover and measured. The spectrophotometer has a spectral range of 400–2500 nm, with 2-nm sampling intervals, a band width of 10 nm and a wavelength accuracy ±0.5 nm. For each measurement, 32 scans were made to produce a mean spectrum with 1050 data points. The spectrum of apparent reflectance R is evaluated by internal software relative to a ceramic standard. The recorded spectral data were processed and stored as absorbance units (A, equal to log(1/R)). Data analysis was conducted using the ISI software system (Shenk and Westerhaus, 1991).

2.6. Spectral data analysis

Spectral data were processed using several pretreatments and transformations and considering different spectral regions. The three pretreatments corresponded to no pre-treatment, MSC (multiplicative scatter correction) and SNVD (standard

normal variate and de-trending transformation) (Barnes et al., 1989). The two transformations which we applied correspond to raw absorbance data and second order derivative. Three spectral regions were considered, the first one with the entire spectrum (VIS–NIR region from 400 to 2500 nm), the second one with the near-infrared region (NIR region from 1100 to 2500 nm), and the third one with the visible and near near-infrared region (VIS region from 400 to 1100 nm). For each analyze, we selected here the combination of pre-treatment, transformation and spectral region that gave the best results. Spectral data were first used to discriminate the soils according to their place in the restoration sequence. Principal component analyses (PCA) were carried out on the spectral data of all soils, clayed soils and sandy soils. For each sample set, 18 PCA were performed considering the three pretreatments, two transformations and three spectral regions. The second set of spectral data analyses correspond to the process of calibration. The calibration procedure involved search for predictive relationships between spectral data and 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 and Naes, 1989). Calibrations equations were built for carbon and nitrogen content, as well as substrate induced respiration and denitrifying enzyme activity using partial least squares regression (PLS) method (Martens and Jensen, 1982; Shenk and Westerhaus, 1991). The PLS method is a multivariate linear calibration technique that reduces large sets of raw data into small numbers of orthogonal (non-correlated) factors in order to minimize the error sum of squares among the values to be predicted. Furthermore, PLS avoids problems of overfitting and collinearity (Martens and Naes, 1989).

The PLS models were validated using internal cross-validation which helps to estimate the optimal number of terms without causing overfitting. This consists of selecting three-quarters of the samples to develop the model and one-quarter for the prediction. The algorithm is repeated four times and all the residuals of the four predictions are pooled to provide a standard error of cross-validation (SECV) on independent samples. The minimum SECV determines the number of terms to be used. The final model is then recalculated with all the samples to obtain the standard error of calibration (SEC). Calibrations were calculated over the entire set of soil samples and separately for clayey and sandy soils.

3. Results

3.1. *Reconstitution of soil and vegetation*

Plantation of the legume *A. mangium* allowed a very rapid reconstitution of vegetation with a vegetation cover reaching 60% after 5 years and around 100% after 8 years whatever the soil texture. At the same time, vegetation recovery of the control plots of the chronosequences (CC and CS) remained very weak with values lesser than 25% in the sandy soils and 10% in the clayey soils (Table 1). In both sequences (i.e. CS, 5S, 8S or CC, 5C, 8C, Table 1), soil C and N increased with time with the maximal value in the oldest plot and greater values in clayey soils than in sandy soils. Eight years after the plantation, soil C and N contents reached values slightly higher in the clayey soil compared to natural forest (2.67% and 0.18%, respectively). In contrast, dynamics of C and N contents of sandy soils were slower than in clayey soils with values corresponding to around 65% of the soil of natural forest established on the same texture. C-to-N ratios increased significantly in sandy soil with values varying between 11.9 and 15.6, but not significantly in clayey soil where values were similar and varied between 12.1 and 14.6.

3.2. *Microbial activities*

As indicated by the values of confidence intervals, substrate induced respiration (SIR) displayed a high heterogeneity within each plot, with the lowest values in the two control

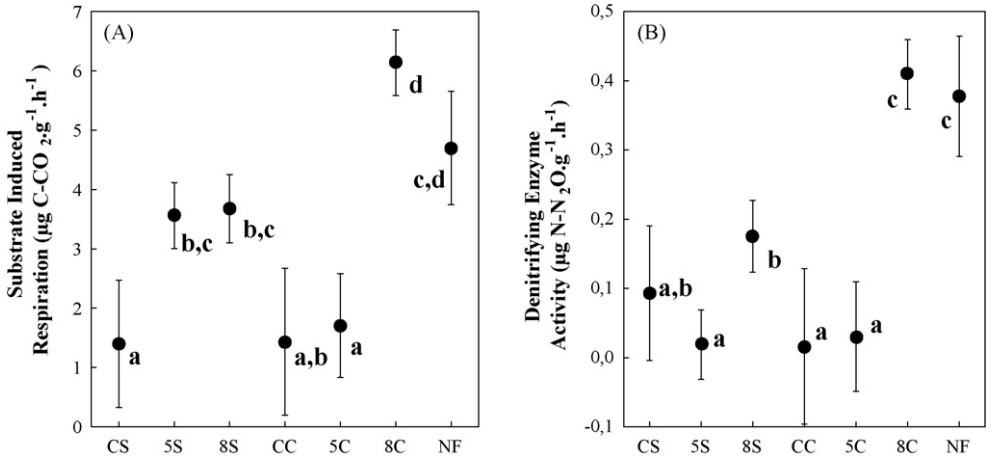

Fig. 1 – (A) Substrate induced respiration and (B) denitrifying enzyme activity in the plots. Plots are means ± 0.95 of confidence intervals ($n = 15$), followed with different letter if significantly different (one-way ANOVA, unequal *n* HSD post hoc test of Tukey, at $p < 0.05$). CS, 5S, 8S: control, 5 years old and 8 years old plantations in sandy soils. CC, 5C, 8C: control, 5 years old and 8 years old plantations in clayey soils. NF: natural forest soil.

plots (CC and CS, Fig. 1A). In the sandy soil plots (CS, 5S, 8S), SIR was significantly higher in 5S and 8S compared to CC, but with no significant difference between them. In the clayey soil plots (CC, 5C, 8C), SIR was similar in 5C plot and significantly higher in the 8C plot compared to the control, reaching a value close to the natural forest control (NF). Denitrifying enzyme activity (DEA, Fig. 1B) was low in most of the plots except in 8C which showed similar average values for DEA as in NF. In the sandy soil plots, there was a slight but not significant increase in DEA between CS and 8S, 5S displaying a value slightly inferior to CS, whereas in the clayey soil plots, only treatment 8C was significantly different from the others treatments.

3.3. *Discrimination of soils with NIRS spectra*

Principal component analyses were performed on the spectral data of all soils, clayey soils and sandy soils (Fig. 2). For each sample set (all soils, clayey soils and sandy soils), the highest proportion of total variance explained by the first two components was obtained using SNV pre-treatment on second derivative of the full VIS–NIR spectral region (1100–2500 nm). In all cases, the first two components explained more than 85% of the total variance.

When considering the entire sample set, factors 1 and 2 explained 89.2% of the variance and separated soils by age and texture (Fig. 2A). In each group of determined age and texture, factor 1 ordinated samples showing the large intravariance of each group except for the natural forest samples more grouped at the left (Fig. 2A). When considering the sandy soils (Fig. 2B), the situation was clearer with a stronger effect of the first and second factors (90.09% of the variance) discriminating the four plots CS, 5S, 8S and NF. In particular the 8S and NF plots are clustered in the 2nd factor but not in the first one. In the clayey soils (CC, 5C and 8C, Fig. 2C), the two first factors of the PCA (96.58% of the variance) separated the three plots with a gradient between the control plot extremely degraded (CC), the 5 years old plantation plot and the 8 years old plantation ones.

Wavelengths whose loading were high in the PCA are shown in Table 2. Whatever the sub-sample, it is noteworthy that visible region added information. As expected, wavelengths associated with carbon and nitrogen compounds as cellulose and protein played an important role in discrimination of soils in the factorial maps.

3.4. *Spectroscopic calibration and measurements*

Results are resumed in Table 3, Figs. 3 and 4. Whatever the sample set (all plots or clayey plots), carbon and nitrogen equations were precise with r^2 varying from 0.94 to 0.98 (Figs. 3

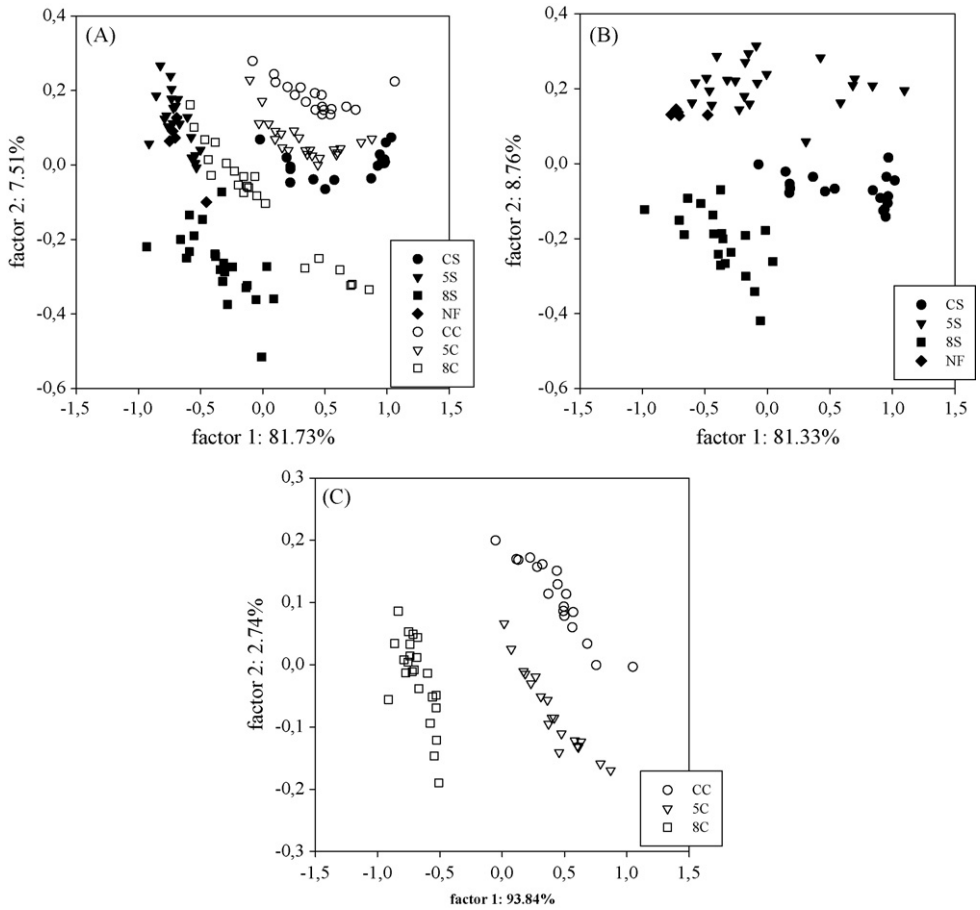

Fig. 2 – Principal component analysis (PC1 × PC2) plots generated from the NIRS spectra data with all soil samples (A); samples containing low clay content (B); samples containing high clay content (C). CS: control sand; CC: control clay; 5S and 8S: 5 and 8 years old plantations in sandy soils; 5C and 8C: 5 and 8 years old plantations in clayey soils; NF: natural forest soil.

Table 2 – Wavelengths with high value loadings on axis 1 and 2 of principal component analyses for all soils, clayey soils and sandy soils

	Axis 1		Axis 2	
	Wavelengths with high values of loadings	Associated to	Wavelengths with high values of loadings	Associated to
All soils	580	Yellow	450	Blue
			550	Green
	2100	C–O–O stretch cellulose	1420, 1450	CH aromatic
	2180	N–H bend and C–H stretch protein	1890	C═O stretch
	2130, 2200 and 2380	C–H stretch	2130, 2180	C–H stretch
	2350	Cellulose	2300	C–H bend
Clayey soils	430–580	Blue–yellow	460–500	Blue
			530–580	Green–yellow
			630–670	Orange–red
	1450	C–H aromatic	1450	C–H aromatic
	2100	C–O–O stretch cellulose	2140, 2200 and 2320	C–H stretch
	2180	N–H bend and C–H stretch protein		
	2350	CH_2 bend cellulose		
	2380	C–H stretch		
Sandy soils			540–560	Green
			1450	C–H aromatic
			1896	C═O stretch
	2100	C–O–O stretch cellulose	2136 and 2320	C–H stretch
	2140	C–H stretch		
	2180 and 2296	N–H bend and C–H stretch protein	2272	O–H stretch and cellulose
	2350	CH_2 bend cellulose		

and 4). The SECV values for C and N were three to four times lower than those of the S.D. of the measured values with better results from the clayey soils. In the sample set including all soils, residual prediction deviation (RPD = S.D./SECV) reached 3.1 for both C and N contents and were increased and reached 4 (4.1 and 4.5 for N and C, respectively) when considering the clayey soils (Table 3).

Calibration statistics for microbial activities (SIR and DEA) were on the whole less accurate than those of carbon and nitrogen (Table 3, Figs. 3 and 4). Moreover, they varied when

Table 3 – Calibration statistics for the constituents in this study

Constituent	*n*	Mean	S.D.	SEC	RSQ	SECV	RPD	Math	Number of terms
All plots									
N content ($\mu g\ g^{-1}$ soil)	91	0.09	0.08	0.02	0.94	0.02	3.1	2 5 5	8
C content ($\mu g\ g^{-1}$ soil)	89	1.24	1.14	0.24	0.96	0.32	3.14	2 10 10	8
SIR (μg C-$CO_2\ g^{-1}$ soil h^{-1})	85	3.59	2.41	1.26	0.73	1.39	1.73	2 10 10	4
DEA (ng N-$N_2O\ g^{-1}$ soil h^{-1})	125	122.97	156.19	59.66	0.85	72.13	2.15	2 10 10	7
Plots with high clay content (CC–5C–8C)									
N content ($\mu g\ g^{-1}$ soil)	63	0.09	0.09	0.01	0.97	0.02	4.14	2 5 5	6
C content ($\mu g\ g^{-1}$ soil)	62	1.20	1.31	0.20	0.97	0.29	4.51	2 5 5	6
SIR (μg C-$CO_2\ g^{-1}$ soil h^{-1})	42	4.28	3.24	1.03	0.89	1.45	2.23	2 5 5	4
DEA (ng N-$N_2O\ g^{-1}$ soil h^{-1})	73	181.34	205.74	81.77	0.84	100.04	2.05	2 5 5	4
Plots with low clay content (CS–5S–8S–NF)									
SIR (μg C-$CO_2\ g^{-1}$ soil h^{-1})	44	3.09	1.41	1.17	0.31	1.27	1.10	2 10 10	2
DEA (ng N-$N_2O\ g^{-1}$ soil h^{-1})	43	89.14	86.01	30.97	0.87	42.44	2.02	2 5 5	4

The values given in the columns refer to number of samples (*n*), the mean of measured values (mean), standard deviation of measured values (S.D.), standard error of calibration (SEC), R squared (RSQ), standard error of cross-validation (SECV) of a linear regression (measured against predicted values); residual prediction deviation (RPD = S.D./SECV). Math: mathematical treatment of the spectral data: the first number is the order of the derivative function, the second is the segment length data points over which the derivative was taken and the third is the segment length over which the function was smoothed; number of terms: number of terms of the PLS models. All plots: equation obtained with all points of the seven plots of sampling. Plots with high clay contents: equations with only the points of CC (control clay), 5C (5 years old plantation with high clay content) and 8C (8 years old plantation with high clay content). Plots with low clay content: equations with only the points of CS (control sand), 5S and 8S (5 and 8 years old plantations with high sand contents) and NF (natural forest soil). SIR: substrate induced respiration in μg C-$CO_2\ g^{-1}$ soil h^{-1}; DEA: denitrifying enzyme activity in ng N-$N_2O\ g^{-1}$ soil h^{-1}; N and C content in $\mu g\ g^{-1}$ of soil.

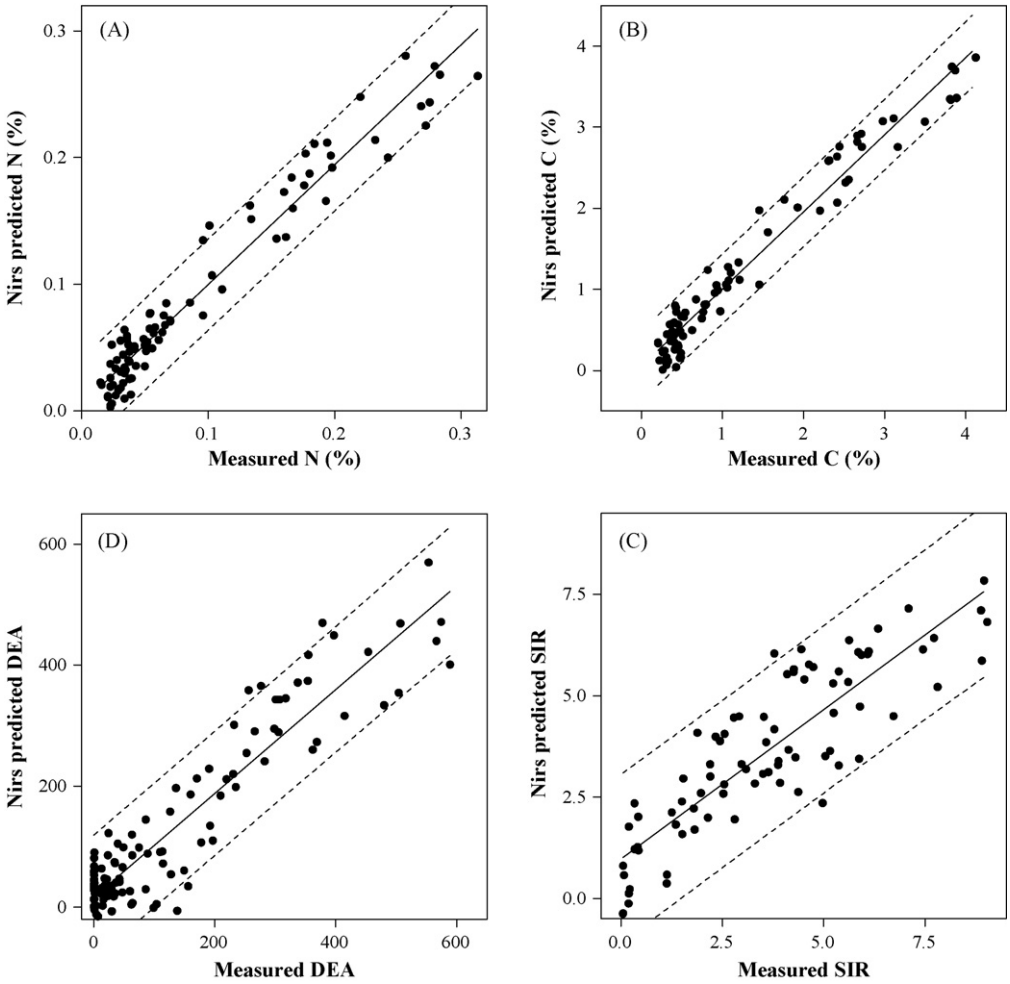

Fig. 3 – NIRS predicted values of N and C contents (A and B, respectively; in μg g^{-1} of soil), denitrifying enzyme activity (DEA) in μg N-N_2O g^{-1} h^{-1} (C) and substrate induced respiration (SIR) in μg C-CO_2 g^{-1} h^{-1} (D) for all soil samples. The broken lines correspond to the 95% prediction interval of the regression line.

considering all soils, clayey soils or sandy soils: (i) for DEA, calibrations were quite good whatever the sample set with r^2 = 0.85, 0.84 and 0.87; SECV = 72.1, 100.0 and 42.4; with RPD values always around 2 (all soils, clayey soils and sandy soils, respectively); (ii) for SIR, calibrations were good when considering all soils (r^2 = 0.72; SECV = 1.3903, and RPD = 1.7), better when using clayey soils samples (r^2 = 0.89; SECV = 1.4505; RPD = 2.2) and poor for sandy soils (r^2 = 0.31; SECV = 1.2762; RPD = 1.1).

4. Discussion

The results of this study tend to confirm our initial hypotheses that information contained in near-infrared soil spectra can be used to predict the dynamics of recovery of soil microbial functions. The planting of *A. mangium* on the soils influenced by mining enabled the degraded soils to achieve good vegetation cover, regardless of the soil texture. Moreover, it induced an increase of the soil carbon and nitrogen contents, with the maximal values in the oldest plots, particularly in the clayey soil, similar to the natural forest ones. These results suggest a strong impact of the Legumes planting on the reconstitution of the physico-chemical characteristics of the soils and are consistent with several works underlining the major improving effects of trees on degraded soils (see Lamb et al., 2005 for a review) through (1) an increase in soil organic matter *via* litters and roots turn-over; (2) an increase in soil N content through N-fixation. Mao et al. (1992) and Fisher (1995) also underlined an increase in N mineralization and microbial biomass by rhizospheric effect and modification of micro-climate (air and soil temperature, soil humidity). Microbial activities showed the same trends as for C and N with an increase from the unrestored areas to the natural forest conditions. These results strengthen previous data (for example Lamb et al., 2005) which show that N_2-fixing trees influence the process of reforestation because it allows the rapid recovery of a functioning microflora community. This recovery is of great importance because, as quoted by Loreau (2001) and Nannipieri et al. (2003), the efficiency of nutrient cycling performed by soil microbial communities plays a central role on an integrative ecosystem function such as productivity. The increase in SIR or DEA resulting from the Legume planting was lower in sandy than in clayey soils even

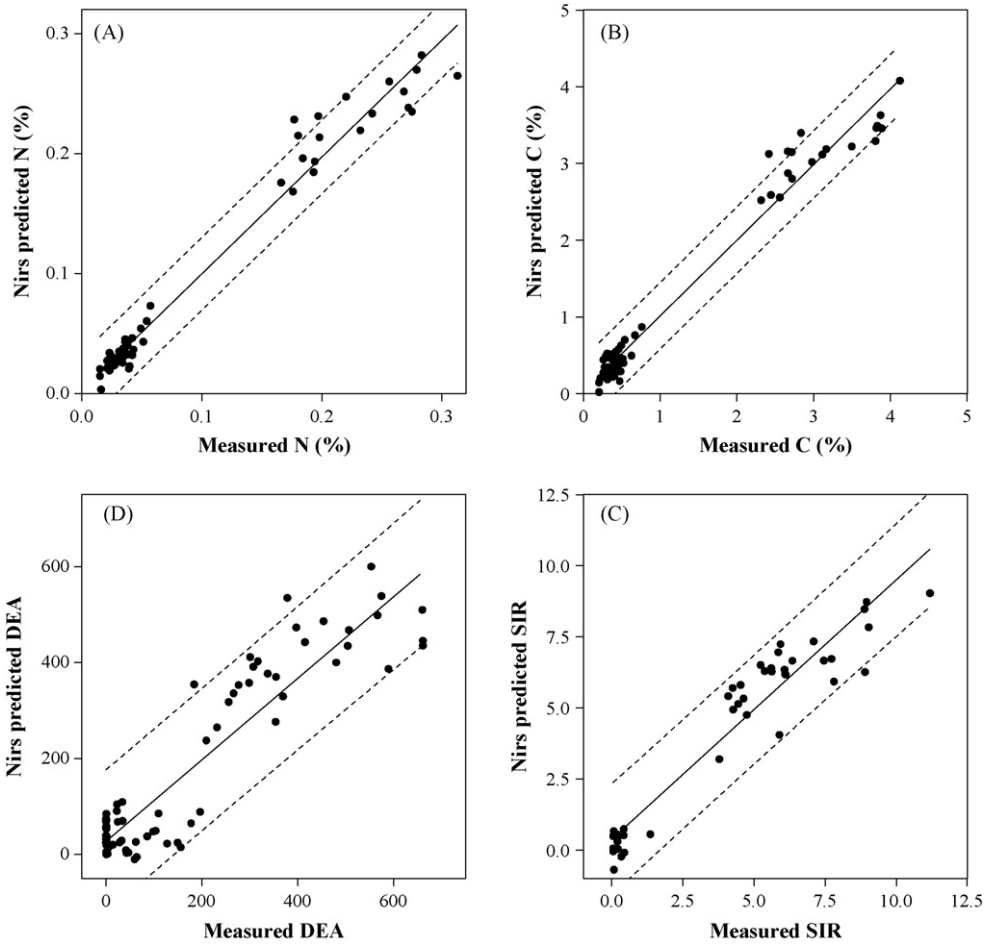

Fig. 4 – NIRS predicted values of N and C contents (A and B, respectively; in μg g^{-1} of soil), denitrifying enzyme activity (DEA) in μg N-N_2O g^{-1} h^{-1} (C) and substrate induced respiration (SIR) in μg C-CO_2 g^{-1} h^{-1} (D) of the clayey soils samples. The broken lines correspond to the 95% prediction interval of the regression line.

after 8 years of plantation. Indeed, in clayey soil Legumes induced a stronger effect, with microbial activities similar to those of the sandy soil of the natural forest. These results are consistent with those from Müller and Höper (2004) who showed significant positive correlations between soil basic respiration and soil clay content in forest and grassland soils. Surprisingly, only slight differences in C-to-N ratios were observed between the plots supporting very contrasted vegetation types (few grasses, Legumes or natural forest): for instance, after 8 years, the C-to-N values of natural forest, clayey and sandy soils were roughly similar (Table 1). High clay content can optimize the short-term turn-over of organic substrates (Saggar et al., 1999), even if C-to-N values observed (Table 1) suggest that a rapid turn-over of organic substrate is possible in all plots. Indeed, the C-to-N ratios found in our work are similar to ones published elsewhere and showing an easy mineralization of organic matter (Hodge et al., 2000). Principal component analyses of NIRS spectra were performed on the three groups of soil samples. More than 85% of the variance was explained by the first two factors in the three analyses; and the discrimination of soils in the factorial maps was based on C and N components as shown by the wavelengths associated with carbon compounds as cellulose and protein (Table 2). Grouping by age was observed on the factorial maps (Fig. 2) whatever the texture. Within each class of age, dispersion of the samples reflected the strong spatial heterogeneity of soil conditions. These results are consistent with those of Odlare et al. (2005) who showed that factor 1 of NIR spectra accurately described soil spatial variation. In our study, the influence of the planted Legumes (i.e. age of plantation) as well as the texture of soil (i.e. clay content) was reflected in the NIR spectra, more precisely in the first two factors of the PCA. Scanning a large set of soil samples resulting from a systematic sampling scheme could provide a very rapid and objective method to document spatial variability of initial soil after perturbations as mining activities. In the same manner, NIRS could quantify rapidly the spatial variability of soil properties after restoration procedures. Our NIR-PLSR calibration results show that C and N contents can be predicted reliably, which confirms others works (Ben-Dor and Banin, 1995; Couillard et al., 1997; Chang and Laird, 2002; Ludwig et al., 2002). Moreover, our results confirmed that functional variables as SIR or DEA can be satisfactorily predicted by NIRS (Chodak et al., 2002; Palmborg and Nordgren, 1993). However, such predictions exhibit different accuracies according to the soil texture.

Indeed, at least SIR is better predicted in clayey than in sandy soils, whereas DEA is equally predicted in the different treatments. Our results indicate that biological variations in soils can be revealed by near-infrared spectral information (Fritze et al., 1994; Viscarra Rossel et al., 2006). Van Waes et al. (2005) have shown the improvement of NIRS to predict soil organic carbon, when dividing samples in texture groups (silt, sand, clay). In our case, only a slight increase in calibration results was obtained for carbon and nitrogen contents when analyzing the clayey sample set instead of the total sample set. Results are less clear when analyzing microbial activities calibrations because the size of samples between textural groups was not equivalent and because the results of calibration for SIR in sandy soils are poor. For instance, RPD values were slightly increased for respiration and equivalent for denitrification when analyzing the clayey soils. Literature gives several concepts considering the residual prediction deviation values: Chang et al. (2001) considered that an acceptable RPD could be >2 for soil analysis; but others authors (Williams, 2001; Fearn, 2002) stated that RPD >3 are adequate for analytical purposes. Nevertheless, our results clearly showed the potential of this method to predict soil microbial activities with some accuracy, and are consistent with results of Pietikaïnen and Fritze (1995) or Fritze et al. (1994) who managed to explain over 80% of the variation in soil basal respiration by near-infrared spectroscopy; or those of Reeves et al. (2000) showing the capacity of NIR spectroscopy to determine biological activities (dehydrogenase, phosphatise or nitrification potential) to some degrees.

Our results showed the capacity of NIRS to establish diagnosis of soil quality in a context of ecosystem restoration. Moreover, it is well known that microbial processes (such as denitrification) generally shows high spatial variability at different scales (Dendooven et al., 1996; Parry et al., 1999). In this context, the capacity of NIR spectroscopy to predict soil microbial activities allows the elaboration of large database establishing metabolic fingerprinting of soils at larger scales. Using these databases, informations about the spatial variability of the soil processes will be assessed with lower cost and time on a larger number of samples.

5. Conclusion

NIRS spectroscopy is an accurate method that enables different parameters of soils to be predicted. Our main results are successful calibrations of NIRS spectra with parameters like respiration and denitrification and, consequently to extend the usefulness of NIR spectroscopy for the prediction of functional soil parameters. The capacity of this method must be strengthen by increasing the databases with other soils in other contexts. This will allow, for further questioning aiming to understand the role of microbial key-functions at an ecosystemic level (which systematically involve studies on spatial variability), to have at our disposal a very useful tool.

Acknowledgement

This work was supported by a grant from the French ''Ministère de la Recherche et de l'Education Supérieure''.

REFERENCES

Anderson, T.H., 2003. Microbial eco-physiological indicators to asses soil quality. Agr. Ecosyst. Environ. 98, 285–293.

Anderson, J.P.E., Domsch, K.H., 1978. A physiological method for the quantitative measurement of microbial biomass in soils. Soil Biol. Biochem. 10, 215–221.

Barnes, R.J., Dhanoa, M.S., Lister, S.J., 1989. Standard normal variate transformation and detrending of NIR spectra. Appl. Spectrosc. 43, 772–777.

Ben-Dor, E., Banin, A., 1995. Near-infrared analysis as a rapid method to simultaneously evaluate several soil properties. Soil Sci. Soc. Am. J. 59, 364–372.

Carter, M.R., Gregorich, E.G., Anderson, E.W., Doran, J.W., Janzen, H.H., Pierce, F.J., 1997. Concepts of soil quality and their significance. In: Gregorich, E.G., Carter, M.R. (Eds.), Soil Quality for Crop Production and Ecosystem Health. Elsevier, Amsterdam, pp. 1–19.

Chang, C.W., Laird, D.A., 2002. Near-infrared reflectance spectroscopic analysis of soil C and N. Soil Sci. 167, 110–116.

Chang, C.W., Laird, D.A., Mausbach, M.J., Hurburgh Jr., C.R., 2001. Near-infrared reflectance of soil properties. Soil Sci. Soc. Am. J. 65, 480–490.

Chodak, M., Ludwig, B., Khanna, P., Beese, F., 2002. Use of near infrared spectroscopy to determine biological and chemical characteristics of organic layers under spruce and beech stands. J. Plant Nutr. Soil Sci. 165, 27–33.

Couillard, A., Turgeon, A.J., Shenk, J.S., Westerhaus, M.O., 1997. Near infrared reflectance spectroscopy for analysis of turf soil profiles. Crop Sci. 37, 1554–1559.

Dendooven, L., Pemberton, E., Anderson, J.M., 1996. Denitrification potential and reduction enzymes dynamics in a Norway spruce plantation. Soil Biol. Biochem. 28, 151–157.

Dighton, J., Pierce, F., 1997. Is it possible to develop microbial test systems to evaluate pollution effects on soil nutrient cycling? In: van Straalen, N.M., Lokke, H. (Eds.), Ecological Risk Assessment of Contaminants in Soil. Chapman and Hall, London, pp. 51–69.

Fearn, T., 2002. Assessing calibrations: SEP, RPD, RER and R^2. NIR News 13, 12–14.

Fisher, R.F., 1995. Amelioration of degraded rain forests soils by plantations of native trees. Soil Sci. Soc. Am. J. 59, 544–549.

Fritze, H., Petri, J., Risto, H., 1994. Near-infrared characteristics of forest humus are correlated with soil respiration and microbial biomass in burnt soil. Biol. Fert. Soils 18, 80–82.

Gee, G.W., Bauder, J.W., 1986. Particle size analysis. In: Klute, A. (Ed.), Methods of Soil Analysis: Part I. 2nd ed. Agronomy Monograph, vol. 9. ASA and SSSA, Madison, WI, USA, pp. 383–411.

Gillon, D., Joffre, R., 1993. Predicting the stage of decay of decomposing leaves by near infrared reflectance spectroscopy. Can. J. Forest Res. 23, 2552–2559.

Hedde, M., Lavelle, P., Joffre, R., Jimenez, J.J., Decaëns, T., 2005. Specific functional signature in soil macro-invertebrate biostructures. Funct. Ecol. 19, 785–793.

Hodge, A., Robinson, D., Fitter, A., 2000. Are microorganisms more effective than plants at competing for nitrogen? Trends Plant Sci. 5, 304–308.

Joffre, R., Ågren, G.I., Gillon, D., Bosatta, E., 2001. Organic matter quality in ecological studies: theory meets experiment. Oïkos 93, 451–458.

Lamb, D., Erskine, P.D., Parrotta, J.A., 2005. Restoration of degraded tropical forests landscapes. Science 310, 1628–1632.

Lensi, R., Gourbière, F., Josserand, A., 1985. Measurement of small amounts of nitrate in an acid soil by N_2O production. Soil Biol. Biochem. 17, 733–734.

Loreau, M., 2001. Microbial diversity, producer–decomposer interactions and ecosystem processes: a theoretical model. Proc. Roy. Soc. B-Biol. Sci. 268, 303–309.

Ludwig, B., Khanna, P.K., Bauhus, J., Hopmans, P., 2002. Near infrared spectroscopy of forest soils to determine chemical and biological properties related to soil sustainability. For. Ecol. Manage. 171, 121–132.

Mao, D.M., Min, Y.W., Yu, L., Martens, R., Insam, H., 1992. Affect of afforestation on microbial biomass and activity in soils of tropical China. Soil Biol. Biochem. 24, 865–872.

Martens, H., Jensen, S.A., 1982. Partial least squares regression: a new two-stage NIR calibration methods. In: Holas,Kratochvil, (Eds.), Proceedings of the World Cereal and Bread Congress, Prague, June 1982. Elsevier, Amsterdam, pp. 607–647.

Martens, H., Naes, T., 1989. Multivariate Calibration. John Wiley & Sons.

Müller, T., Höper, H., 2004. Soil organic matter turnover as a function of the soil clay content: consequences for model application. Soil Biol. Biochem. 36, 877–888.

Mutuo, P.K., Shepherd, K.D., Albrecht, A., Cadisch, G., 2006. Prediction of carbon mineralization rates from different soil physical fractions using diffuse reflectance spectroscopy. Soil Biol. Biochem. 38, 1658–1664.

Nannipieri, P., Ascher, J., Ceccherini, M.T., Landi, L., Pietramellara, G., Renella, G., 2003. Microbial diversity and soil functions. Eur. J. Soil Sci. 54, 655–670.

Odlare, M., Svensson, K., Pell, M., 2005. Near infrared reflectance spectroscopy for assessment of spatial soil variation in an agricultural field. Geoderma 126, 193–202.

Palmborg, C., Nordgren, A., 1993. Modelling microbial activity and biomass in forest soil with substrate quality measured using near infrared reflectance spectroscopy. Soil Biol. Biochem. 25, 1713–1718.

Parry, S., Renault, P., Chenu, C., Lensi, R., 1999. Denitrification in pasture and cropped soil clods as affected by pore space structure. Soil Biol. Biochem. 31, 493–501.

Pietikaïnen, J., Fritze, H., 1995. Clear-cutting and prescribed burning in coniferous forest: comparison of effects on soil fungal and total microbial biomass, respiration activity and nitrification. Soil Biol. Biochem. 27, 101–109.

Reeves III., J.B., McCarty, G.W., Meisinger, J.J., 2000. Near-infrared spectroscopy for the determination of biological activity in agricultural soils. J. Near Infrared Spectrosc. 8, 161–170.

Saggar, S., Parshotam, A., Hedley, C., Salt, G., 1999. C-14-labelled glucose turnover in New Zealand soils. Soil Biol. Biochem. 31, 2025–2037.

Schloter, M., Bach, H.-J., Metz, S., Sehy, U., Munch, J.C., 2003. Influence of precision farming on the microbial community structure and functions in nitrogen turnover. Agr. Ecosyst. Environ. 98 (1–3), 295–304.

Shenk, J.S., Westerhaus, M.O., 1991. ISI NIRS-2. Software for Near-infrared Instrument. Silverspring.

Tiedje, J.M., 1984. Denitrification. In: Page, L.A., Miller, R.H., Keeney, D.R. (Eds.), Methods of Soil Analysis. Part 2. Chemical and Microbiological Properties. 2nd ed. Am. Soc. of Agr. Inc. (ASA); Soil Sci. Soc. Am. Inc. (SSSA), Madison, pp. 1011–1026.

Van Waes, C., Mestdagh, I., Lootens, P., Carlier, L., 2005. Possibilities of near infrared reflectance spectroscopy for the prediction of organic carbon concentrations in grassland soils. J. Agr. Sci. 143, 487–492.

Velasquez, E., Lavelle, P., Barrio, E., Joffre, R., Reversat, F., 2005. Evaluating soil quality in tropical agroecosystems of Colombia using NIRS. Soil Biol. Biochem. 37, 889–898.

Viscarra Rossel, R.A., Walvoort, D.J.J., McBratney, A.B., Janik, L.J., Skjemstad, J.O., 2006. Visible, near infrared, mid infrared or combined diffuse reflectance spectroscopy for simultaneous assessment of various soil properties. Geoderma 131 (1/2), 59–75.

Williams, P.C., 2001. Implementation of near-infrared technology. In: William, P., Norris, K.H. (Eds.), Near-infrared Technology in the Agricultural and Food Industries. American Association of Cereal Chemist, St Paul, Minnesota, USA, pp. 145–169.