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# Prediction of tree and shrub leaf chemistry and digestibility by the use of Near Infrared Reflectance Spectroscopy

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## Introduction

In the Mediterranean area, farmers are graze their herds on a diversity of pastures, including wooded rangelands. At certain periods, some of them expect the rangeland diet to meet up to 3/4 of total energy for high requirement ruminants such as lactating goats[1,2]. The sheperds therefore design their grazing route by combining native and cultivated pastures, and fenced paddocks often include a wide heterogeneity of fodder resources[3]. To upgrade pasturing management recommendations, one needs to assess the nutritive value of native foliages including their changes over time and spatial diversity[4]. As an exemple, one can see on figure 1 the wide range in fodder resources quality in a Provence goat farmland (Meuret, *unpublished*). In such diverse environments, assessing the nutritive value of fodder resources, including native and cultivated tree and shrub foliages, by the conventional wet chemical analysis of samples is time-consuming and too expensive, as very large samplings are required. In addition, some results are declared uncertain due to the particular composition of woody foliages as compared with cultivated forages for which standard analytical methods were developed[5,6,7,8]. Fiber assay is disturbed by the possible insolubilization of proteins caused by their interaction with phenolic compounds[9]. In the last ten years, simultaneoulsy with studies that tried to improve wet chemical analysis for ligneous products, the near-infrared reflectance spectroscopy (NIRS) has been tested as a tool to describe diverse pastoral resources including woody foliages[10,11,12]. Within the same period, remote sensing and ecological studies have used this technique to predict leaf canopy chemistry[13,14,15,16,17]. Using a wide range of native and cultivated Mediterranean tree and shrub foliages, our objective was to evaluate the potential of NIRS to determine foliage chemistry and *in vitro* digestibility for ruminants.

## Material and Methods

Samples were collected from typical wooded rangeland and fodder tree plantations in the French Mediterranean area. The data base comprises 25 species, representing the diversity of regional ligneous fodder resources (botanical names[18]) : *Arbutus unedo L., Amorpha fruticosa, Buxus sempervirens L., Calycotome spinosa L., Cistus albidus L., Cistus monspeliensis L., Cistus salviifolius L., Cytisus villosus Pourr., Colutea arborescens L., Coronilla emerus L., Erica arborea L., Erica scoparia L., Hedera helix L., Juniperus communis L., Juniperus oxycedrus L., Morus alba L., Phillyrea angustifolia L., Phillyrea latifolia L., Pistacia lentiscus L., Pistacia terebinthus L., Quercus ilex L., Quercus pubescens Willd., Rhamnus alaternus L., Robinia pseudoacacia L., Ruscus aculeatus L.* Most species were sampled at different periods in the year, when they were actually being browsed by animals. The samples were taken from plant parts selected within the different eating bites[19]. The date base comprises 222 samples, including leaves and stems only, divided before analysis into the successive growths parts.
All samples were placed directly after cutting in a refrigerated container at +10°C before being frozen at -20°C. They were then air dried in a ventilated oven at 60°C. Drying kinetics were monitored for each sample, and the drying times were calculated to obtain 93% dry materials.

Drying times were much shorter than the conventional "to constant weight" durations. We had checked that this technique avoids excessive heating of foliages and denaturation of soluble compounds[1]. All samples were then ground into a cyclone mill trough with a 1-mm mesh. Since, for financial reasons, all the samples were not analyzed for all constituents and in vitro digestibility, the size of each group is indicated under brackets : Moisture (222) (103°C for 24h); Organic matter (OM : 222) (550°C for 24h); Nitrogen (N : 222) (micro-Kjeldahl analysis); Neutral Detergent Fiber (NDF : 98), Acid Detergent Fiber (ADF : 122) and Acid Detergent Lignin (ADL : 122) from Fibertec procedure[20]; in vitro digestibility of the dry matter with a pepsin-cellulase method developed for forages[21] (IVDMD : 83).

All the samples were scanned with a near-infrared reflectance spectrophotometer (NIRSystem 5000). Each sample was packed into a sample cell having a quartz-glass cover. The reflectance measurement of monochromatic light was made from 1100 to 2500 nm to produce a spectrum with 700 data points at 2 nm intervals over this range. The band-pass used 10 nm and the wavelength accuracy is 0.5 nm. Reflectance (R) is converted to absorbance (A) using the following equation : A = log (1/R). Data analysis was conducted using ISI software system[22].

Partial Least Squares Regressions (PLSR) were developed for OM, N, NDF, ADF, ADL and IVDMD with each calibration using 9 math treatments, corresponding to raw spectrum, first and second derivative. Whereas the number of sample is rather low, any independent validation set has been created. The number of PLS factors to include in the model is choosen by the minimum error obtained by cross-validation. SECV (standard error of cross-validation) gives a more realistic value of accuracy than SEC (standard error of calibration). Calibrations for the different constituents and in vitro digestibility were carried out on the whole 222 sample data base (broad-based equations).

NIR prediction for IVDMD were applied to describe the consequences of phenological changes in *Quercus pubescens Willd.* foliage on the digestibility of the edible plant parts at browsing height for a small ruminant. From April 1991 to May 1992, 12 samplings were taken periodically in a coppice near Avignon. Each sampling was a mix of 20 leafy branches with representative proportions of plant parts, that were cut on a 20 m long transect within the coppice. Plant parts were collected and conditioned as described above. The edible parts consisted of the current year's growth leaves ($L_0$) and the three most recent growth stems ($S_0$, $S_{-1}$, $S_{-2}$). Acorns were collected but not NIR analyzed. The prediction equation for IVDMD was applied to the individual plant parts, after checking equation reliablity for this spectral data base. As the various plant parts collected were considered from the opening of buds stage, the age of each part was known within the limits of a few days. A calibration was made on the age of current year's growth leaves and stems.

## Results and Discussion

The foliage base is very heterogeneous. The standard deviation spectra of the raw spectra of the foliage are twice wider than the standard deviation of a reference herbaceous forage bases[23]. The 222 foliage samples data base is characterized by an extremely wide range of chemical components concentration and in vitro digestibility (Table I). The lowest standard errors of calibration (SEC) and cross validation (SECV) are comparable to the literature data for nitrogen (SEC slightly up to 0.1) and are better for lignin (SECV is 1.48 compared to SEC 2.5 - 2.9 in previous studies with foliages[13,14,16] (Table II). Calibration Rsquares ranged from 0.96 to 0.99. Concerning IVDMD, SECV is close to 2.0 and appears satisfying because quite similar to usual standard errors concerning in vitro digestibility predictions[23,24] determined on narrower sets than than the current one. Graphic comparisons between wet chemistry values and NIR predicted values for N, ADL and IVDMD are shown in figure 2. One can see that the prediction equations are all effective.

The broad-base equation for IVDMD was applied to the 46 collected *Quercus pubescens Willd.* plant parts. Since oak species are well represented in the calibration data base, all the samples are predictible whereas the distances of these samples *vs* the calibration set remain less than the conventional limit of 3. As maturing progress, leaf IVDMD decreases from 65 to 15 % dry matter with a +15 % difference with the last stem value until October (figure 3). The two older growth stems have a regular and very similar variation around 30 % IVDMD. This type of information, with a high frequence of sampling on individualized edible plant parts, should be linked with the intake models of small ruminant browsing on oak foliages[1]. From these models, one can estimate that the nutritive value of this oak during the summer period (mid-June to September) varys from 55 to 52% OM digestibility.

## Conclusion

The NIRS is a performing technique to predict constituent contents for nutritive value estimations of Mediterranean foliages from trees and shrubs. The NIR analysis could point out which samples have been deteriorated during the conditioning process. It is a highly valuable tool for international networks that conduct woody fodder surveys and have to compare samples from various origins[25]. This study shows that broad-based calibrations could be made on extremely diverse sets of data, including high nutritive leguminous shrubs such as *Citisus* and *Robinia* and very coarse *Ericacea* species, grouping leaves and stems within the same data base. The calibration for fiber lignified fractions appears quite satisfactory. That confirms the relevance of our specific oven-drying technique for woody foliages. Prediction of pepsin-cellulase in vitro digestibility could be reliably used for these foliages, as is the case with more conventional green forages and herbage, to determine the effect of maturation on nutritive value of plant parts.

## References


1. M.Meuret M, Feuillages, fromages et flux ingérés. *PhD Thesis Agron.Sci.*, Fac. Gembloux, INRA-SAD Avignon (1989).
2. M.Napoleone and T.Gillet, Profil de production du troupeau et système d'élevage caprin de la région Provence Côte d'Azur. *INRA Prod. Anim.* 3, 347-354 (1990).
3. B.Hubert, Comment raisonner de manière systémique l'utilisation du territoire pastoral ? in *Proc. IVth International Rangeland Congress*, Montpellier (France) : (1992).
4. M.Meuret, J.BOZA, H.NARJISSE and A.NASTIS, Evaluation and utilizatión of rangeland feeds by goats. in *Goat Nutrition*, Ed. by P.MORAND-FEHR, Wageningen, Pudoc III, EEAP Pub 46, 160-171 (1991).
5. M.N.McLeod, The digestibility and the nitrogen, phosphorus and ash contents of the leaves of some australian trees and shrubs. *Aust.J.Exp.Agric.Anim.Husb.*, 13, 245-250 (1973).
6. P.J.Urness, A.D.Smith and R.K.Watkins, Predicting dry matter digestibility of mule deer forages, *J.Range Manage.*, 30, 119-121 (1977).
7. M.Lachaux, M.Meuret et M.de Simiane, Composition chimique des végétaux ligneux pâturés en région méditerranéenne française : problèmes posés par l'interprétation des analyses, in *La forêt et l'élevage en région méditerranéenne française*, Ed. by B.Hubert, *Fourrage* Hors-série : 231-267 (1987).
8. J.D.Reed JD, Relationship among soluble phenolics, insoluble proanthocyanidins and fiber in East African browse species. *J.Range Manage.*, 39, 5-7 (1986).
9. J.Ho Ahn, B.M.Robertson, R.Elliott, R.C.Gutteridge, and C.W.Ford, Quality assessment of tropical browse legumes : tanin content and protein degradation. *Anim.Feed Sci.Technol.*, 27, 147-156 (1989).
10. J.L.Holechek, J.S.Shenk, M.Vavra and D.Arthun, Prediction of forage quality using near infrared reflectance spectroscopy on esophageal fistula samples from cattle on moutain range. *J.Anim.Sci.*, 55, 971-975 (1982).
11. P.A.Duncan, D.C.Clanton and D.H.Clark, Near Infrared reflectance spectroscopy for quality analysis of sandhills meadow forage. *J.Anim.Sci.*, 64, 1743-1750 (1987).
12. J.J.Waelput, R.Biston and M.Meuret, Study of the potential of near infrared reflectance spectroscopy in the analysis of the tree foliage intake of goats. *Reprod.Nutr.Dev.*, S2, 166s (1990).
13. D.H.Card, D.L.Peterson and P.A.Matson, Prediction of leaf chemistry by the use of visible and near infrared reflectance spectroscopy. *Remote Sens.Environ.*, 26, 123-147 (1988).
14. C.A.Wessman, J.D.Aber, D.L.Peterson and J.M.Melillo, Foliar analysis using near infrared reflectance spectroscopy. *Can.J.For.Res.*, 18, 6-11 (1988).
15. C.D.Elvidge, Visible and near infrared reflectance characteristics of dry plant materials. *Int.J.Remote Sensing.*, 11, 1775-1795 (1990).
16. T.M.McLellan, M.E.Martin, J.D.Aber, J.M.Melillo, K.J.Nadelhoffer and B.Dewey, Comparison of wet chemistry and near infrared reflectance measurements of carbon-fraction chemistry and nitrogen concentration of forest foliage. *Can.J.For.Res.*, 21, 1689-1693 (1991).
17. P.J.Curran, J.L.Dungan, B.A.Macler, S.E.Plummer and D.L.Peterson, Reflectance spectroscopy of fresh whole leaves for the estimation of chemical concentration. *Remote Sens. Environ.*, 39, 153-166 (1992).
18. T.Tutin, *Flora Europea*, CUP, London (1980).

19. M.Meuret, Feasibility of in vivo digestibility trials with lactating goats browsing fresh leafy branches. *Small Rumin.Res.*, 1, 273-290 (1988).
20. P.J.Van Soest and J.B.Robertson, Analysis of forage and fibrous foods : a laboratory manual for animal science, 613. Cornell Univ Pub. (1985).
21. J.Aufrère, Etude de la prévision de la digestibilité des fourrages par une méthode enzymatique. *Ann.Zootech.*, 31, 111-130 (1982).
22. J.S.Shenk and M.O.Westerhaus, ISI-NIRS-2 : Software for near-infrared instruments. Infrasoft International , User Manual (1991).
23. R.Biston et P.Dardenne, Application de la spectrométrie dans le Proche Infrarouge. *Bull.Rech.Agron.Gembloux*, 20,: 23-41 (1985).
24. J.Aufrère and C.Demarquilly, Predicting organic matter digestibility of forage by two pepsin-cellulase methods. in *Proc XVI Intern.Grassld.Congr.*, Nice (France), 877-878 (1989).
25. H.Guérin, B.Toutain and S.T.Fall, Nutritive value of ligneous forage in West-Africa. in *Proc. CEE Workshop on Natural Resource Development and Utilization*, Wageningen (The Netherlands), 106-110 (1992).

Table I
Wet chemical range (%) for each constituent within the calibration sample set.

| *Variable* | *n* | *Mean* | *STD* | *Range* |
|---|---|---|---|---|
| OM | 222 | 93.8 | 3.2 | 82.6 - 98.3 |
| N | 222 | 1.84 | 0.86 | 0.39 - 4.17 |
| NDF | 98 | 52 | 13 | 85.0 - 23.8 |
| ADF | 122 | 31.5 | 10.7 | 12.7 - 54.1 |
| ADL | 122 | 13.9 | 6.2 | 3.0 - 26.9 |
| IVDMD | 83 | 64 | 19.8 | 28.2 - 94.0 |

Table II
Modified partial least squares regression
Equation calibration statistics for OM, N, NDF, ADF, ADL and IVDMD without outliers and with scatter correction

| *Variable* | *n* | *SEC* | *$R^2$* | *SECV* | *Math treatment ** |
|---|---|---|---|---|---|
| OM | 204 | 0.54 | 0.97 | 0.63 | 1 5 5 |
| N | 206 | 0.11 | 0.98 | 0.14 | 1 5 5 |
| NDF | 90 | 1.36 | 0.99 | 2.06 | 1 5 5 |
| ADF | 110 | 1.85 | 0.97 | 2.36 | 1 10 5 |
| ADL | 108 | 1.04 | 0.97 | 1.48 | 2 5 5 |
| IVDMD | 75 | 1.51 | 0.99 | 2.09 | 2 20 5 |

** Math treatment indicates the mathematical transformation of spectral data : the first number is the order of the derivative function, the second is the lenght in data points over which the derivative was taken and the third the segment lenght over which the function was smoothed.*

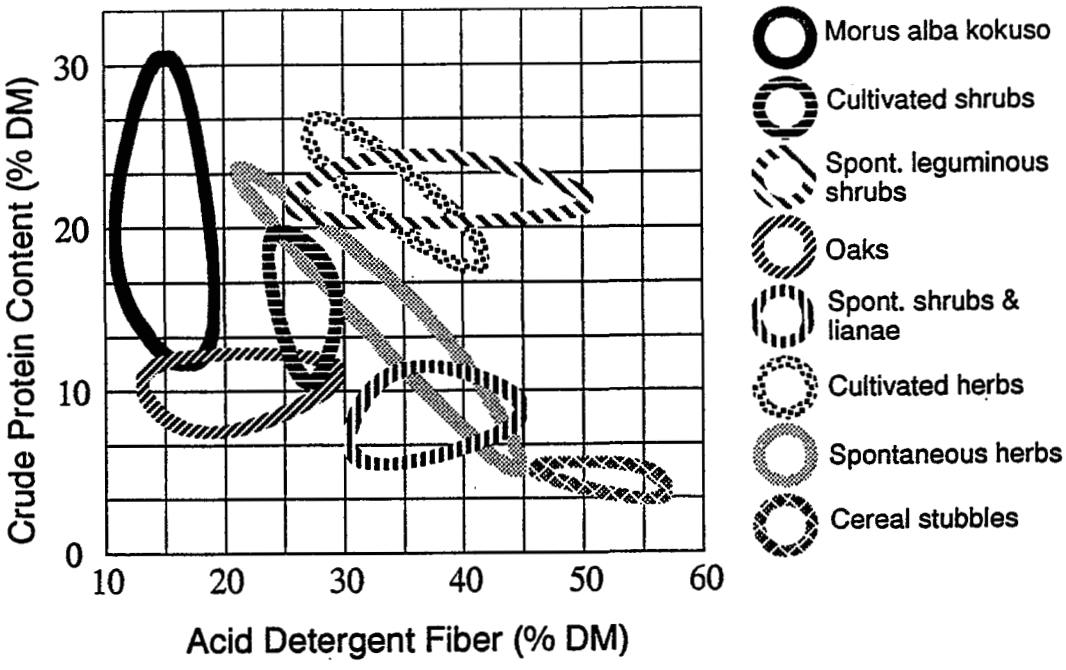


Figure 1

Range of fodder resources quality within a diversified goat farmland in Provence (Meuret, *unpublished*)

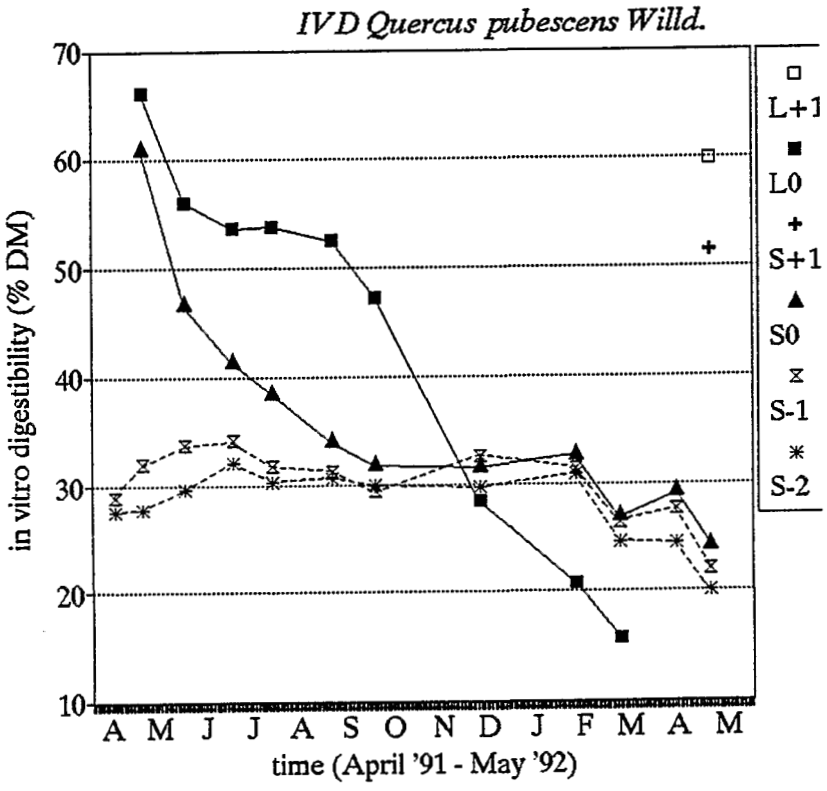


Figure 3

Variation over time in the in vitro pepsine-cellulase digestibility for the edible plant parts of Quercus pubescens Willd. $L_0/S_O$ is the current year's growth leave to stem ration (% dry matter).

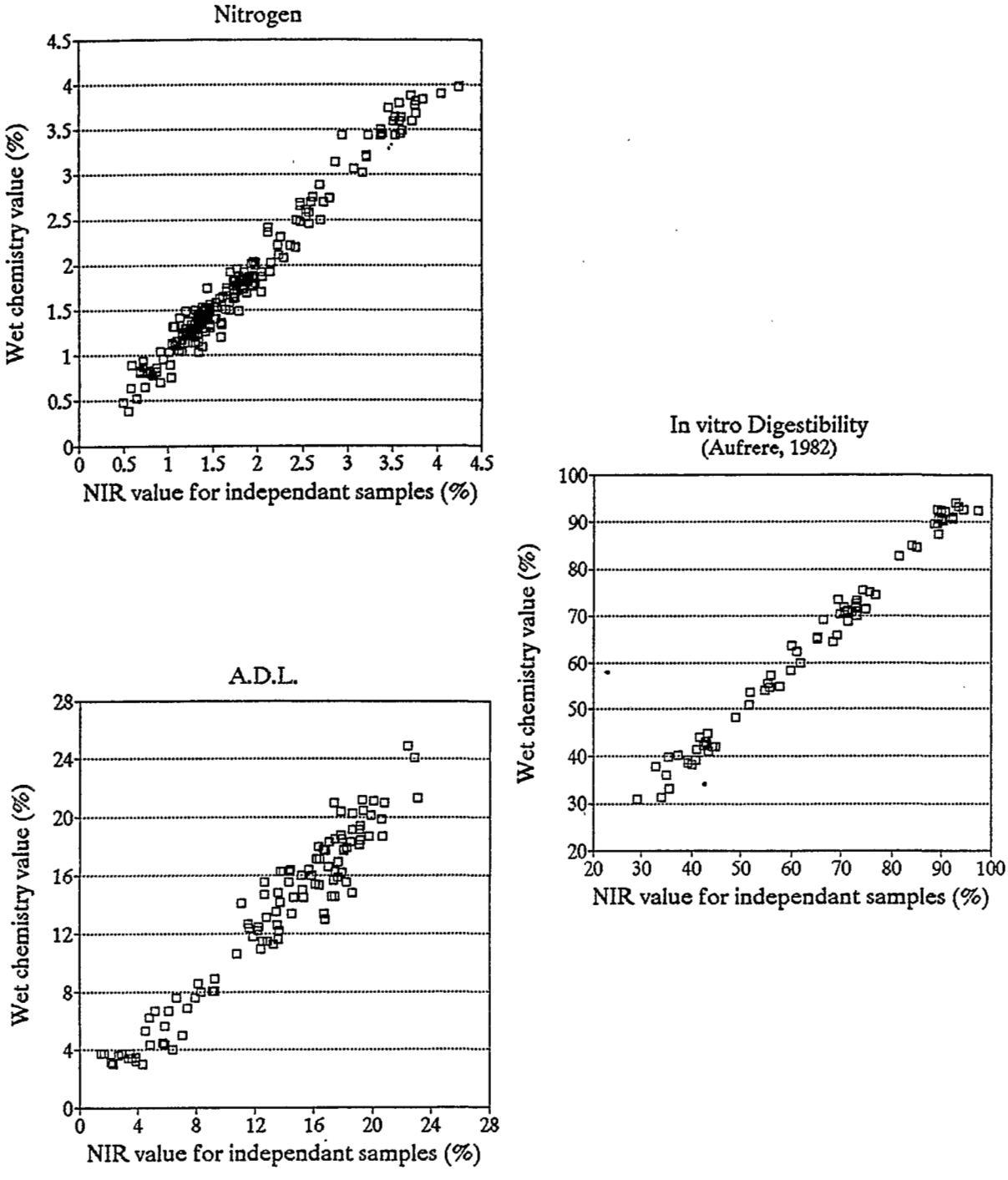


Figure 2

Relationship between NIR predicted value for independant samples set and wet chemistry value (% dry matter) for nitrogen, lignin and *in vitro* digestibility.