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Predicting botanical composition of grass-clover pastures mixtures using near-infrared reflectance spectroscopy

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Key words : NIRS , botanical composition , binary mixtures , predication equation

Introduction The development of a reliable and rapid approach for determining botanical composition has been a research goal for many years . Several researchers have attempted to use NIRS as an approach to determine botanical compositions . Some researchers have already proven the capability of NIRS to determine species content in legume-grass mixtures (Pitman et al ., 1991) . Our objective was to compare NIRS to other methods for separating complex mixtures of species .

Materials and methods Twenty-four cool season clover and grass cultivars were used for this study . Clover species included three red clover(*Trifolium pratense* L) cultivars , four white clover (Trifolium repens L) , and one kura clover (Trifolium ambiguim . Bieb) cultivar . The grass species included two festuloliums (*Festulolium braunii* , *K . A .)* cultivars , four orchardgrass (*Dactylis glomerata* L .) , seven perennial ryegrass (*Lolium perenne* L) and three tall fescue (*Festuca arundinacea* Schreb) . The experiments were arranged in a randomized complete block design (RCBD) with three replications . Each replication consisted of 67 entries of different combinations of grass and clover cultivars . Samples were hand clipped within a 0 .25 m^2 quadrat in each plot when plants were 20 cm in height and dried at 65 °C then ground to pass through a 1mm-screen . Calibration equations were developed using 1-artificially pure mixed samples , 2-hand-separated samples from one location and year and 3-hand-separated samples from multiple locations and years . Standard deviation of calibration (SD) , standard error of calibration (SEC) and calibration coefficient of determination (R^2) were used to evaluate the calibration equation . Prediction coefficient of determination (R^2) , standard error of prediction (SEP) , slope , and bias were used as validation criteria of the equations .

Results Data from artificially pure mixtures shows a very strong calibration equation with R^2 of 0 .99 and standard error of calibration (SEC) of 1 .4 . However , regression analyses of the spectral data with botanical composition resulted in prediction coefficient of determination (R^2) of 0 .31 and 0 .24 and SEP of 28 .1 and 26 .7 for grass and clover , respectively . The variation in plant maturity and weed content observed in the field samples were not represented in the lab mixed samples . Thus , the calibration equation developed from the artificially mixed samples of several grass and clover species did not accuracy predict the species composition clipped from three different locations over three growing seasons . A higher prediction may obtain if the calibration equation was derived from samples representing all locations and years (Shaffer et . al ., 1990) .

Equations developed from hand-separation from one location and year had R^2 of 0 .97 for grass and 0 .95 for clover . However , when this equation was applied to predict the botanical composition of samples collected from three locations and three years , similar predictions were obtained to the results of the artificial pure mixed samples with validation R^2 of 0 .25 for grass with somewhat improvement prediction for clovers with R^2 of 0 .37 . This clearly reflects the problem of obtaining enough representative samples that can include all spectra data from other locations .

On other hand Calibration equations developed from hand-separated samples collected from three locations over three years had R^2 of 0 .82 for grass and 0 .84 for both clovers and weeds . The validation statistics for these equations resulted in an R^2 of 0 .67 to 0 .72 , SEP of 6 .9 to 12 .8 , and a bias of 0 .3 to 0 .6% . Slopes were close to one for the grass and clover . In spite of there being multiple species in the weeds portion , SEP for weeds was lower than grass and clover components . It is possible the weed species were more similar in reflectance values than the different species of grasses .

Conclusions Even though we had some success in predicting grass-clover mixtures based on multiple locations and years as compared with the equation created from a single location and year , this study did not result in as high R^2 as those observed in other studies (Locher et al ., 2005) . The lower R^2 observed were due to various factors . Environmental conditions and the grinding and storing processes can affect NIRS prediction . This study , which presented two different strategies for developing a calibration equation , has shown that creating an equation from representative samples of a larger database of previously hand-separated mixtures can increase the prediction accuracy .