NEAR-INFRARED SPECTROSCOPY: A RAPID NON-DESTRUCTIVE METHOD FOR MEASURING WOOD PROPERTIES, AND ITS APPLICATION TO TREE BREEDING*

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ABSTRACT

Near-infrared (NIR) spectroscopy provides a rapid, non-destructive method for the routine estimation of wood properties. NIR spectroscopy is increasingly being used to replace traditional methods of wood property assessment, and it provides a wealth of information to tree growers, tree breeders, and manufacturers of forest products. The technology is particularly well-suited to tree improvement programmes where large numbers of samples must be analysed, but it can be utilised in any forestry application where the rapid provision of wood property data is required.

The application of NIR spectroscopy to samples ranging in size from milled chips representing whole trees, to sections of wooden strips cut from increment cores, and, at the smallest scale, to wafers cut from the tangential face of cores was examined, along with studies that have utilised NIR spectroscopy for tree breeding purposes. The technology has some limitations and there are also obstacles to wider acceptance.

Keywords: near-infrared spectroscopy; tree breeding; wood properties; *Eucalyptus globulus*; *Eucalyptus nitens*; *Pinus radiata*; *Pinus taeda*.

INTRODUCTION

This paper is not intended to be a thorough examination of all the wood near-infrared (NIR) spectroscopy literature. Rather it is a review of selected articles that relate to the estimation of pulp yield, and properties related to pulp yield, by NIR spectroscopy. It will also describe how NIR spectroscopy has been used for the estimation of wood properties of increment cores and how data provided by NIR analysis have been used for the estimation of genetic parameters. Extensive

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reviews of NIR spectroscopy and its application to wood and forest products have been produced by Schimleck & Workman (2004), So *et al*. (2004), and Tsuchikawa (2007).

NIR spectroscopy is a relatively new analytical technique that has been utilised for the prediction of product properties since the 1960s. Prior to that time spectrophotometers were available that could measure NIR spectra; however, the NIR region of the electromagnetic spectrum was rarely utilised as it was considered that it contained no relevant structural information (Barton 2004). Burns & Margoshes (1992) described many of the problems encountered by spectroscopists who wanted to work with the NIR region; these included an absence of sharp peaks, an abundance of overlapping and shoulder peaks, a dramatic loss in sensitivity (2 to 3 orders of magnitude relative to the mid-infrared), and the difficulty of making band assignments owing to the presence of numerous overtone and combination bands.

The earliest analytical applications of NIR spectroscopy were reported in the early 1960s by Karl Norris of the U.S. Department of Agriculture (Barton 2004). For example, Ben-Gera & Norris (1968a,b) used NIR spectroscopy to determine the moisture, crude protein, and oil concentrations of cereal grains and oil-bearing seeds. By the early 1990s the analytical applications of NIR spectroscopy were widespread, with many different industries (agriculture, food, paper, petro-chemical, polymer, and textile) utilising the technology (Ciurczak 1992).

The rapid growth of the technology can be attributed to the emergence of high-precision spectroscopic instruments with very high signal-to-noise ratios to detect minute differences in the reflectance spectra, and high-speed computers to carry out the complex calculations involved in multivariate analysis (Norris 1989). In addition, NIR spectroscopy has several advantages over traditional laboratory techniques, including:

- rapid speed and reliability of determinations;
- rapid and easy sample preparation;
- multiplicity of analysis with one operation;
- operation by unskilled personnel; and
- non-destructive analysis (Norris 1989; Schultz & Burns 1990).

Origin of Near-infrared Spectra

The infrared region of the electromagnetic spectrum ranges from 700 to 10^6 nanometers (nm) (10 to 14 300 cm^{-1}) and is divided into near-, middle-, and far-infrared. A summary of each region is given in Table 1 (Osborne *et al*. 1993). The most useful region for quantitative (and to a lesser extent qualitative) analysis by reflectance in the near-infrared is 1200 to 2500 nm (8333-4000 cm^{-1}). For wavelengths

TABLE 1–A summary of the infrared region of the electromagnetic spectrum

Region	Characteristic transitions	Wavelength range (nm)	Wavenumber range (cm^{-1})
Near-infrared (NIR)	Overtones, combinations	700–2500	14300–4000
Middle-infrared (MIR)	Fundamental vibrations	2500–5×10^4	4000–200
Far-infrared	Rotations	5×10^4–10^6	200–10

below 1200 nm the weak absorption bands make reflectance measurements difficult and for those above 2500 nm the bands become too strong (Norris 1989). Spectra that occur in the near-infrared region consist largely of overtone and combination bands of the fundamental stretching vibrations of O-H, N-H, and C-H functional groups (Osborne *et al*. 1993; Shenk *et al*. 1992).

RAPID ESTIMATION OF PULP YIELD BY NIR SPECTROSCOPY

Despite the numerous advantages that NIR spectroscopy presents, its utilisation for the estimation of wood properties has a relatively short history, with the earliest work being reported as recently as the late 1980s. Preliminary studies concentrated on the feasibility of NIR spectroscopy to predict wood chemistry, specifically pulp yield, and two wood components (cellulose and lignin) that together have a large influence on pulp yield (Birkett & Gambino 1988; Schultz & Burns 1990; Wright *et al*. 1990; Michell 1995; Olsson *et al*. 1995).

A long-term objective of tree breeding programmes has been to improve the pulp yield of plantation-grown trees as pulp yield is an important factor in determining plantation profitability (Dean *et al*. 1990; Borralho *et al*. 1993; Greaves *et al*. 1997). However, traditional assessment of pulp yield is time-consuming and expensive, limiting the number of trees that can be analysed (Downes *et al*. 1997; Raymond & Schimleck 2002). In addition, it is destructive because trees identified for assessment must be felled. Hence, the estimation of pulp yield by NIR spectroscopy was particularly important as a rapid, non-destructive method had long been sought for this purpose.

These studies were generally based on whole-tree wood property data (cellulose, lignin, pulp yield, etc.) and NIR spectra collected from milled chips that were usually a subsample of the chips used to provide the wood property data. The composite chip sample could have represented a whole-tree or multiple trees and was obtained from either chipped billets or discs from several different heights. A regression equation, based on multiple linear regression or Partial Least Squares (PLS) regression, was obtained using the available wood property data and

corresponding spectra. The calibration was then used to predict the wood property for a set of validation samples based on the NIR spectra of these samples.

It was quickly established that NIR spectroscopy could be used to predict pulp yield, cellulose content, and lignin content, albeit with variable accuracy which could be related to the quality of the laboratory data used to develop the calibrations and the wood property variation present in the calibration and prediction sample sets. However, without exception the validation samples were drawn from the same population (pseudo-validation) as the calibration samples and from a practical viewpoint several questions were raised; for example:

- Is it possible to have multiple-species, or multiple-site calibrations?
- Could a calibration for one site be used to accurately estimate the wood properties of samples from a different site (a truly independent validation)?
- Can whole-tree wood properties be estimated using NIR spectra from cores?

The following sections examine attempts to answer these questions.

Multiple-species Multiple-site Calibrations

Multiple-site multiple-species calibrations are common in agriculture where extremely large spectral databases containing in excess of 1000 spectra per commodity have been created over many years (Dardenne 2004). In an extreme example Berzaghi *et al.* (2002) reported the development of multiple-site forage calibrations based on a set of 20 000-plus samples from Australia, Belgium, Canada, Germany, Italy, Sweden, and the USA.

The scale of this work has yet to duplicated with wood. This can be attributed to several factors, including the short history of wood-NIR research, the failure of NIR spectroscopy to replace traditional methods of wood property determination on a large-scale (in comparison, NIR spectroscopy has replaced many traditional methods of analysis in the agricultural industries), and the difficulty and cost of determining properties such as pulp yield. Another problem with a property such as pulp yield is that laboratories are not consistent with the pulping methods that they employ, making it impossible to share characterised samples between laboratories, unlike the work described by Berzaghi *et al.* (2002) on forage characterisation.

Garbutt *et al.* (1992) provided an early example of cellulose and lignin NIR calibrations based on mixed species (13 eucalypts, one hybrid), and Michell (1995) reported excellent calibration statistics for a number of wood and pulp quality parameters using native-forest-grown *Eucalyptus globulus* Labill. from 10 locations in Tasmania. The sample set utilised by Michell (1995) is worth noting. The samples, representing up to 10 trees per location, were collected by Orme in

1976 (Eldridge *et al*. 1993) and were part of an extensive investigation into the genetic variation of Tasmanian *E. globulus* (Turner *et al*. 1983). The variation that the set represented was extreme, with tree age ranging from 38 to 370 years and soda pulp yields ranging from 37.6 to 60.2% (Turner *et al*. 1983). The extreme variation that existed for this set explains the very high correlation coefficients reported by Michell (1995). Equivalent correlation coefficients cannot be expected for plantation-grown woods, as demonstrated by Michell & Schimleck (1998) and Schimleck *et al*. (2000) who reported examples of multiple-site kraft pulp yield calibrations (based on milled chips representing a whole-tree composite) for plantation-grown *Eucalyptus nitens* (Deane et Maiden) Maiden from Tasmania and multiple-site *E. globulus* and *E. nitens*, also from Tasmania.

Recent studies have attempted to include more species from more locations. For example, Schimleck, Rezende, Demuner, & Downes (2006) obtained calibrations for whole-tree basic density, lignin, pentosans, pulp yield, and specific consumption using seven different eucalypt species and five hybrids, sampled from three different locations in Brazil. Calibration statistics for pulp yield were poor; however, the pulp yield data utilised by Schimleck, Rezende, Demuner, & Downes (2006) had variable Kappa numbers (range = 16.7 to 18.8) and, therefore, was not directly comparable between samples, explaining the poor results.

Another study demonstrating the ability of NIR spectroscopy to provide multiple-site multiple-species calibrations was reported by Hodge & Woodbridge (2004) who developed calibrations for lignin content using five species of tropical and sub-tropical pines (*Pinus caribaea* Morelet, *P. maximinoi* H. E. Moore, *P. oocarpa* Schiede, *P. patula* Schltdl. & Cham., and P. *tecunumanii* F. Schwerdtf. ex Equiluz & J. B. Perry) grown in Brazil and Colombia. A lignin calibration based on the five species from both countries had a coefficient of determination (R^2) of 0.90. It should be noted that the calibration reported by Hodge & Woodbridge (2004) was based on small wedges cut from breast-height discs that had been split into juvenile and mature sections and not whole-tree chips; the wedges were also used to determine lignin content.

Efforts to develop multiple-site multiple-species calibrations for pulp yield continue as the development of a calibration that encompasses wide variation and does not require continual refinement has great appeal to many users. The most notable effort is at Ensis (a CSIRO Forestry and Forest Products and Scion unincorporated joint venture) where a pulp yield calibration has been developed based on approximately 700 samples (a mix of milled individual whole-tree samples and composite chip samples of many trees) and including multiple eucalypt species from multiple sites in Australia (G. Downes pers. comm.) (Fig. 1). At an average cost of several hundred Australian dollars per sample the calibration represents a sizable investment. It is important to note that NIR spectra for the calibration were obtained from milled

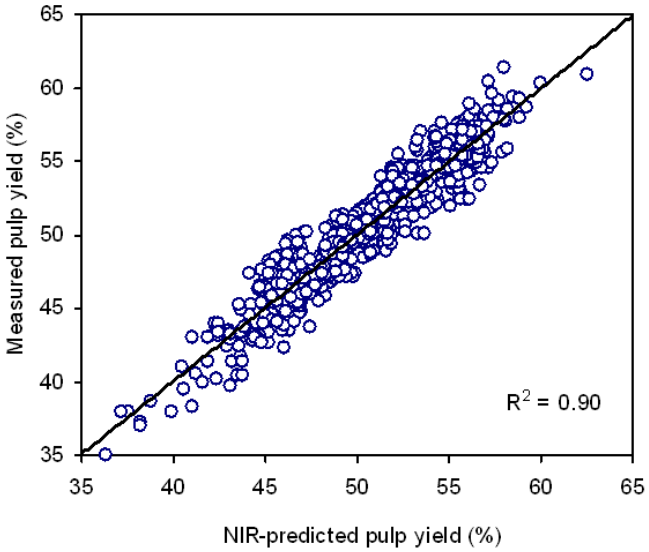

FIG. 1–Relationship between measured pulp yield and NIR-estimated pulp yield for the Ensis pulp yield calibration.
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chips representative of the chips from which the pulp data were obtained; therefore, from the perspective of the calibration, it does not matter if the sample is from part of a tree, a whole tree, or multiple trees.

Multiple-site multiple-species calibrations are a desirable objective for breeding programmes, particularly for a property such as pulp yield where the expense of laboratory measurements limits the number of samples that can be analysed; however, as noted by Murray (2004), predictions made by such calibrations will not be as precise as those obtained using a single-species, site-specific calibration. Typically these broad-based calibrations are referred to as GLOBAL calibrations. An alternative that has not been investigated for the estimation of wood properties is the LOCAL approach (Shenk *et al*. 1997) which involves developing a specific equation to predict a given property for a new sample. The new calibration is obtained using samples selected from a large database on the basis of their similarity to the unknown sample (Pérez-Marín *et al*. 2005). Several studies have demonstrated that LOCAL calibrations provide smaller predictive errors than GLOBAL calibrations (for example, Sinnaeve *et al*. 1994, Shenk *et al*. 1997, and Pérez-Marín *et al*. 2005). The advantage of the LOCAL approach is that it provides the benefits of using a GLOBAL strategy (i.e., a large database that encompasses the expected variation) with the accuracy of specific calibrations (Pérez-Marín *et al*. 2005). However, the establishment of a large database containing thousands of samples is critical (Pérez-Marín *et al*. 2005), explaining why the LOCAL approach has yet to be explored for estimating wood properties.

Application of Pulp Yield Calibrations to New Sites

The vast majority of the NIR-wood studies that reported test calibrations used them to predict the properties of a subsample from the same population (pseudo-validation). While this approach demonstrates that the calibration is applicable to the subset, it doesn't provide any indication of how it will perform if applied to samples from a new population. When a calibration is applied to samples from different sites, its performance can be expected to suffer owing to the myriad differences that exist between the sites represented in the calibration and prediction sets. An example of what can be expected was provided by Schimleck *et al*. (2000) when they used a northern Tasmania *E. nitens* pulp yield calibration based on trees from several sites to estimate the pulp yields of 38 plantation-grown *E. nitens* samples from southern Tasmania. It was found that the calibration over-estimated the pulp yields of the southern Tasmania *E. nitens* by approximately one unit of yield; however, the relationship between yields estimated by NIR spectroscopy and those determined using laboratory pulping was very good (R^2 = 0.87). Selections of the highest-yielding trees based on laboratory pulping and on NIR-predicted yields were also compared and it was found that of the top 12 pulp-yielding trees selected using laboratory data, nine would have been selected using NIR-predicted data. As noted by Schimleck *et al*. (2000) there would be little chance of missing an outstanding individual and, if tree selection was based on families rather than individuals, then it would be very unlikely that an outstanding family would be missed.

In a later study Schimleck, Kube, Raymond, Michell, & French (2005) applied a Tasmania-wide *E. nitens* pulp yield calibration to a set of 25 samples from a site in northern Tasmania (Gog). While a reasonable R_p^2 was obtained (0.70), prediction errors were large (SEP = 4.60%) as the Tasmania-wide calibration consistently under-estimated the pulp yields of the Gog samples.

One approach to improving the predictive performance of calibrations applied to samples from a site not represented in the calibration, is to include a small number of samples from the new site in the calibration set (Guthrie & Walsh 2002). Schimleck, Kube, Raymond, Michell, & French (2005) found that the addition of five Gog samples to the calibration set greatly improved the performance of the calibration when it was used to predict the pulp yields of the other Gog samples (R_p^2 = 0.77, SEP= 1.03%). Predicted yields (based on NIR spectra from whole-tree composites) before and after the addition of five Gog samples to the Tasmania-wide *E. nitens* pulp yield calibration are illustrated in Fig. 2.

Schimleck, Kube, Raymond, Michell, & French (2006) showed that the addition of a single Gog sample to the Tasmania-wide calibration set greatly reduced predictive errors and that the inclusion of at least three Gog samples in the Tasmania-wide set was sufficient to give relatively stable predictive errors. The practical implication

of these findings is that calibrations can be adapted to be directly applicable to new locations. However, concerns exist about the statistical robustness of a calibration if a relatively small number of samples can markedly change its performance.

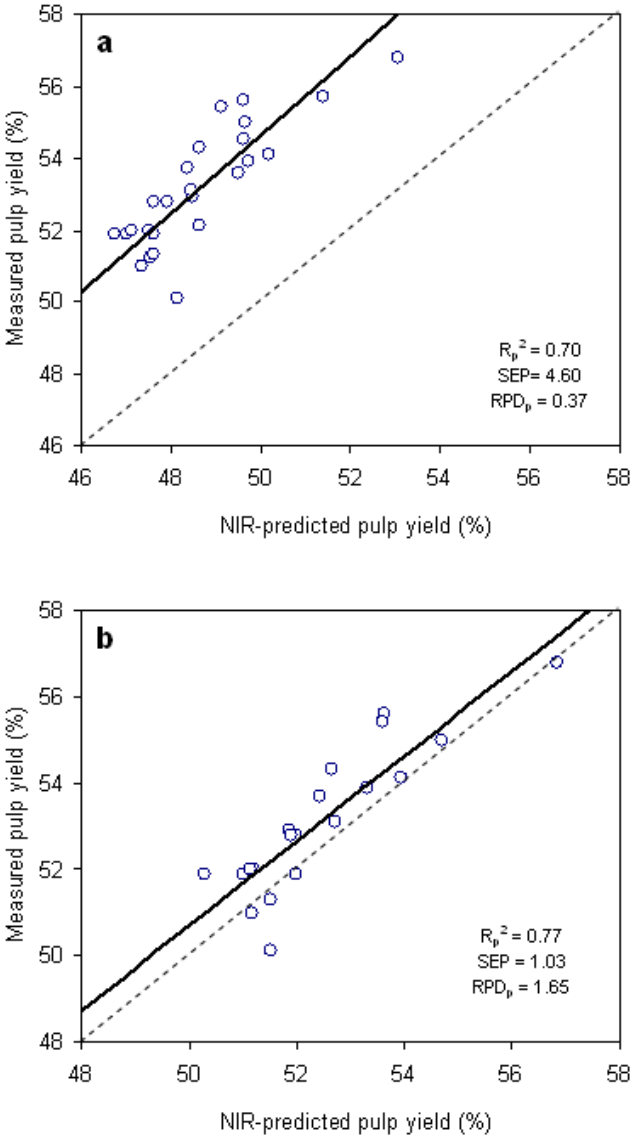

FIG. 2–Relationships between measured pulp yield and NIR-predicted pulp yield based on whole-tree composite chip samples from Gog (a) predictions made using a Tasmania-wide pulp yield calibration, and (b) predictions made using a Tasmania-wide pulp yield calibration after the addition of five samples from Gog. Note that the regression line has been plotted and that the thin broken line represents the line of equivalence in both figures.

ESTIMATION OF WHOLE-TREE WOOD PROPERTIES USING NIR SPECTRA OF MILLED CORES

The ability to accurately estimate whole-tree pulp yield based on NIR spectra collected from increment cores would greatly reduce the costs of plantation assessment and facilitate non-destructive sampling. In order to do this, calibrations between whole-tree pulp yield (obtained from destructively sampled trees) and NIR spectra collected from increment cores need to be developed. Schimleck, Rezende, Demuner, & Downes (2006) examined this approach in their study based on multiple eucalypt species and hybrids grown in Brazil. It was shown that wood property calibrations based on whole-tree data and NIR spectra collected from whole-tree composite chips and increment cores from 0.65 and 1.50 m provided similar calibration statistics (Fig. 3) and also performed in a similar manner when applied to a separate test set. A possible reason for the similar performance of the three sets of calibrations is that the rankings of the trees were in a similar order (for example, from highest to lowest yield) regardless of which set (whole-tree composite, 1.3 m core, 0.65 m core) was used, and for calibration purposes it is the relative order of samples that is important.

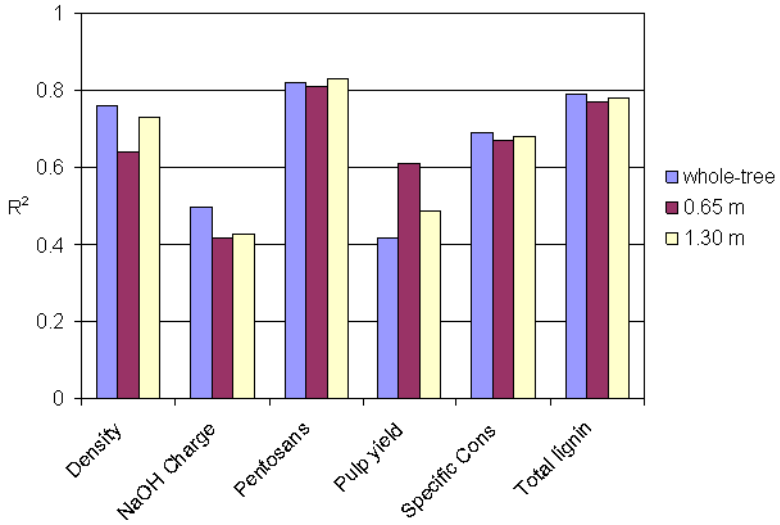

FIG. 3–Co-efficients of determination (R^2) of calibrations for each trait developed using milled whole-tree chips, 0.65-m cores, and 1.30-m cores. Reproduced with permission from *Appita Journal 59*: 231–236 (2006)

In a similar study based on hybrid poplar, Schimleck, Payne, & Wearne (2005) also found that calibrations for whole-tree pulp yield and cellulose based on whole-tree chip and increment core NIR spectra provided similar calibration statistics (whole-tree spectra $R^2 = 0.94$ (cellulose, five factors) and 0.96 (pulp yield, five factors) and core spectra $R^2 = 0.89$ (cellulose, five factors) and 0.90 (pulp yield,

six factors)). For the hybrid poplar examined by Schimleck, Payne, & Wearne (2005) a moderate relationship was found between whole-tree cellulose and core cellulose ($R^2 = 0.65$).

Alternatively, calibrations based on whole-tree chips can be used to estimate whole-tree yields, based on core NIR spectra. It can be expected that errors obtained using this approach will be greater because a core will not represent a whole tree in the same way as a composite chip sample, and variation specific to cores has not been included in the whole-tree calibration. In their study based on *E. nitens* Schimleck, Kube, Raymond, Michell, & French (2005) examined this approach and, in an unexpected result, found that predictive errors were actually lower (whole-tree $R_p^2 = 0.70$, SEP = 4.60%; cores $R_p^2 = 0.78$, SEP = 3.27%). They also found that the addition of five Gog cores greatly improved the predictive performance of the Tasmania-wide *E. nitens* pulp yield calibration when applied to the remaining cores from Gog ($R_p^2 = 0.76$, SEP = 1.04%).

The findings of these studies indicate that it is likely that a small number of trees would have to be destructively sampled and analysed to ensure variation unique to a particular site was included in the calibration. In selecting trees for destructive sampling it is preferable that the trees identified represent the variation in pulp yields (or wood chemistry in general); however, this may not always be possible. There are several different approaches and these are briefly described.

- Select trees without any prior knowledge of wood chemistry; the trees selected may still represent different diameter classes, provenances, clones, site conditions, etc.;
- Collect cores (for example, 50) from trees at the new location, predict the pulp yields of the 50 samples using an existing pulp yield NIR calibration (assuming an existing calibration is available), and make selections to represent the range of yields;
- Collect cores from the new location, collect NIR spectra from the cores, and use spectral analysis methods to identify the most spectrally unique samples. Advantages of using this method are that it can be used to select a specified number of samples that best represent the new population in terms of the spectral information, and an existing calibration is not required. There are several methods for selecting samples based on their spectral characteristics. Many spectroscopists use the Center and Select method developed by Shenk and implemented in WinISI software (Infrasoft International 2000) which uses a neighbourhood concept to identify spectrally unique samples. Other options include the Kennard-Stone (Kennard & Stone 1969), Duplex (Snee 1977), and OptiSim (Clark 1997) algorithms.

Assuming that an existing calibration exists (and this may be a big assumption) and that pulping facilities are available, there are several options in the way that

NIR spectroscopy can be used when estimating the pulp yield of trees from a new location. These options are summarised in Table 2.

TABLE 2–Advantages and disadvantages of each calibration option.

Option	Advantages	Disadvantages
Existing calibration	* Minimal cost as no additional pulping is required * Rankings generally OK * Able to identify majority of top trees	* Yield may be under- or over-estimated
Enhanced calibration	* Estimated yield closer to true yield * Improved ranking and identification of top trees	* Some additional pulping required (minimal)
Site-specific calibration	* Most accurate estimate of yield * Most accurate rankings	* Maximal cost as all trees must be pulp tested

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Estimation of the Wood Properties of Intact Radial Strips

An understanding of patterns of wood property variation from pith to bark with age, the effects of forest management practices on wood quality, and the minimum age of early assessment for various wood properties are all important questions for a breeding programme (Raymond 2002). Answers to these questions can only be obtained by the analysis of large numbers of increment cores and, until the recent development of the SilviScan instruments (Evans 1994, 1998), this was practical only for basic density on a large scale.

Recent studies (Schimleck & Evans 2002a,b, 2003, 2004) based on NIR spectra collected in 10-mm sections from the radial-longitudinal surface of strips cut from *Pinus radiata* D. Don increment cores have demonstrated that NIR spectroscopy can be used to examine the pith-to-bark variation of a range of physical properties and tracheid morphological characteristics. Using calibrations based on SilviScan-generated data, a range of properties were predicted by NIR spectroscopy including air-dry density, microfibril angle (MFA), stiffness, and tracheid coarseness, specific surface, and wall thickness. It has also been demonstrated that wood chemistry (cellulose, extractives, lignin, wood sugars) and fibre length can be estimated using NIR spectra collected from intact radial strips (Schimleck, Jones, Peter, Daniels, & Clark 2004; Jones *et al*. 2006; Poke & Raymond 2006). These studies were based on a limited number of samples compared to the studies that utilised SilviScan data.

The same questions as discussed for calibrations based on whole-tree composite chip samples apply for calibrations based on increment cores. Studies based on *Pinus taeda* L. have shown that multi-site calibrations (representing different site qualities and physiographic regions in Georgia, USA) could be obtained for the physical and morphological properties reported in the studies of Schimleck & Evans, and that the calibrations could be successfully applied to cores from different sites (Jones *et al*. 2005a,b). The development of multi-species calibrations has also been explored. Schimleck *et al*. (2001) obtained reasonable calibrations for air-dry density and stiffness using spectra collected from 54 different commercial species (both hardwoods and softwoods) from around the world.

The provision of wood property data in 10-mm increments makes it possible to examine radial variation and the estimation of core averages. However, it has limited use for examining the variation of wood properties within rings, which is important if a goal is the estimation of genetic parameters of individual rings (Sykes *et al*. 2003). Increasing the resolution of NIR measurements to 2 mm would make the estimation of wood properties within rings possible, but the findings of a recent study (Jones *et al*. 2007) suggest that existing NIR spectrometers are not suitable for scanning radial strips at high spatial resolution. An alternative method for collecting NIR spectra at high spatial resolution is to obtain NIR spectra from the tangential surface of samples cut from increment cores or radial strips. Depending on the thickness of the tangential section, reflectance (relatively thick sections) or transmission NIR spectra can be collected. The first approach (reflectance spectroscopy) has been utilised by Schimleck, Sussenbach, Leaf, Jones, & Huang (2007) to estimate earlywood and latewood microfibril angle within rings, using sections cut from *P. taeda* radial strips. The second approach (transmission spectroscopy, in the general range of 600–1900 nm) has been used by Yeh *et al*. (2004, 2005) and Sykes *et al*. (2005) who collected their spectra from thin wafers cut from *P. taeda* increment cores and estimated α-cellulose, lignin, and tracheid coarseness and length. Calibration statistics were relatively good and indicated that transmission NIR spectroscopy could provide a method for examining wood property variation at high spatial resolution within individual rings. Sykes *et al*. (2005) based their calibrations on the earlywood and latewood of two rings (Rings 3 and 8) and found that Ring 3 calibrations for α-cellulose and fibre coarseness predicted Ring 8 properties with R^2 values of approximately 0.6, indicating the potential for early selection. It should be noted that for the work based on transmission NIR spectroscopy sample preparation was particularly intensive and limited the number of samples that could be analysed quickly.

Compared to the studies based on whole-tree composite samples, the studies based on increment cores present an interesting contrast, particularly when SilviScan data are used for calibration development. SilviScan provides an abundance of data,

making the development of calibrations based on thousands of spectra possible. In addition, SilviScan provides wood property data that have been measured by a machine, largely negating the human element and minimising errors that can occur in wet chemistry data. Unlike a NIR spectrum collected from a whole-tree composite, a NIR spectrum collected from a section of a radial strip relates exactly to the wood property data available for calibration. In combination these factors provide calibration statistics for properties such as stiffness and microfibril angle that are generally superior to those reported for calibrations based on whole-tree composites. However, there are also problems unique to working with NIR spectra collected from radial strips and SilviScan data. It is imperative that a NIR spectrum for a given section of a radial strip corresponds exactly to the wood property data. Mismatched data will provide poor calibration statistics. If this occurs for a small number of samples, the mismatched spectra are obvious and the problem can be rectified, but if the problem is widespread little can be done. Similar problems can arise if the spectra and wood property data are slightly offset. The problem of mismatched or offset data may be relatively minor for spectra collected at a resolution of 10 mm, but as the resolution of spectral measurements increases matching the two data sets becomes more problematic. In addition, the number of spectra collected increases rapidly as resolution is increased. For example, increasing the resolution from 10 mm to 2 mm gives a five-fold increase in the number of spectra collected, compounding the problem. Recently Schimleck, Tyson, Jones, Peter, Daniels, & Clark (2007) have demonstrated that for calibration development it is unnecessary to use NIR spectra from every section of a core. A single spectrum per core is adequate provided that sections from other cores representing juvenile, mature, and juvenile/mature transition wood are included in the calibration set. Calibration and prediction statistics can be marginally improved by increasing the number of spectra per core to between three and five; the addition of further spectra is unnecessary. The practical implication of this finding is that the problems associated with creating calibrations based on radial strips scanned at high spatial resolution can be greatly reduced, as can the cost of SilviScan analysis.

Field-based NIR Spectroscopy of Wood Properties

Increasingly NIR spectroscopy is being investigated as a field-based measurement technique with the aim of providing data in real time. In agriculture this has become a reality, with NIR spectrophotometers being fitted on to harvesters for the estimation of forage quality (Haeusler *et al*. 2002; Paul & Pfitzner 2004; Sinnaeve *et al*. 2004). Ideally the estimation of wood properties by NIR spectroscopy would also be conducted in the field in real time and this would reduce the need to collect increment cores. Efforts are under way to make this goal a reality, but preliminary results for *P. taeda* (Jones *et al*. in press) have not been promising. These authors

cited as two obstacles the difficulty of designing a suitable fibre-optic probe for NIR analysis, and the performance of fibre-optic probe systems compared to bench-top NIR spectrometers. The high resin content of some softwood species is also problematic as it bleeds into the core hole, and will adhere to the fibre-optic probe (Jones *et al*. in prep.).

In a recent study, Acuna & Murphy (2006) used NIR spectra collected from chainsaw chips in the laboratory to predict the basic density of *Pseudotsuga menziesii* (Mirb.) Franco stems. Calibration R^2 ranged from 0.89 to 0.95, while R^2 obtained on a validation set (0.56 to 0.85) were weaker. The authors concluded that their results indicated that NIR technology could be used by mechanical harvesters to segregate logs based on basic density.

ESTIMATION OF GENETIC PARAMETERS BASED ON NIR-GENERATED DATA

The focus of this review has been largely pulp yield and, to a lesser degree, increment core properties, but the range of properties that can be assessed and incorporated into a breeding programme is not limited to the properties discussed. This is demonstrated by the work of Baillères *et al*. (2002), who examined a wide range of properties including extractive and lignin content, lignin composition (syringyl guaiacyl ratio), shrinkage (longitudinal, radial, and tangential), and surface longitudinal growth strain.

There is interest in using NIR spectroscopy for tree breeding purposes as it provides a rapid, non-destructive method for determining multiple properties for a large number of samples. As already discussed, non-destructive sampling can be achieved by removing increment cores and NIR spectroscopy can be used to predict whole-tree pulp yield based on core spectra. For convenience, breast height is generally used; however, for young (5 to 9 years) *E. globulus* and *E. nitens* Raymond *et al*. (2001a) recommended the use of sampling heights of 1.1 m for *E. globulus* growing on all sites and 0.9 m for *E. nitens* growing on high-quality sites (for *E. nitens* growing on poor-quality sites cores were not good predictors of whole-tree yield). In later studies these sampling recommendations were followed and provided data that were used to estimate genetic parameters and genotype-×-environment interactions for pulp yield and pulpwood productivity, and genetic parameters for cellulose content, extractives, lignin content, and susceptibility to decay in *E. globulus* (Raymond *et al*. 2001b; Raymond & Schimleck 2002; Poke *et al*. 2006).

The genetic gains in cellulose content for *E. nitens* have also been compared using cellulose content determined with wet chemistry and predicted by NIR calibrations based on different sampling intensities (Schimleck, Kube, & Raymond 2004). Cellulose content was examined as it is strongly related to pulp yield (Dillner *et*

al. 1970; Wallis *et al*. 1996; Kube & Raymond 2002) and provides a low-cost alternative for pulp yield improvement in a breeding programme (Kube & Raymond 2002). It was found that genetic gains based on NIR-predicted cellulose content were high, and very similar to the gains achievable using a direct measure of cellulose. In addition, NIR-predicted cellulose was found to be highly heritable, with heritabilities comparable to direct measures of cellulose. Schimleck, Kube, & Raymond (2004) also compared methods for selecting calibration samples (20, 40, and 60 samples based on pulp yields predicted using an existing calibration, and samples selected on the basis of spectral variation). Calibrations based on samples selected for their spectral variation provided higher genetic gains (90% of the maximum possible gain) than samples selected based on their predicted kraft pulp yields (see Table 3 for gains for a forward selection strategy, and Table 4 for gains for a backward selection strategy). The study of Schimleck, Kube, & Raymond (2004) demonstrated that NIR spectroscopy can be used to provide estimates of

TABLE 3–Gain in cellulose content (%) with different NIR calibration methods using a forward selection strategy. Gain is expressed as a proportion of the gain that would be achieved by assessing every tree for cellulose content using wet chemistry methods.

Type of calibration model	Selection of samples for calibration model			
	20/site	40/site	60/site	WinISI*
Local site model	73	83	88	89
Off-site model 1 (Dial calibration)	61	86	87	82
Off-site model 2 (Gog calibration)	64	73	70	83
Off-site model 3 (Kamona calibration)	80	90	91	91

* The number of samples used by WinISI was 37 at Dial, 59 at Gog, and 45 at Kamona.
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TABLE 4–Gain in cellulose content (%) with different NIR calibration methods using a backward selection strategy. Gain is expressed as a proportion of the gain that would be achieved by assessing every tree for cellulose content using wet chemistry methods

Type of calibration model	Selection of samples for calibration model			
	20/site	40/site	60/site	WinISI*
Local site model	93	94	87	100
Off-site model 1 (Dial calibration)	84	95	81	100
Off-site model 2 (Gog calibration)	83	97	79	100
Off-site model 3 (Kamona calibration)	90	91	89	99

* The number of samples used by WinISI was 37 at Dial, 59 at Gog, and 45 at Kamona.
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genetic parameters that agree very well with those determined using data obtained with conventional laboratory techniques but at a reduced cost.

CONCLUSIONS

NIR spectroscopy provides a rapid alternative to traditional methods of wood property assessment and potentially has a very important role in the future of tree breeding programmes. Many organisations have explored using the technology but few are using it on a regular basis. There are many issues related to the analysis of wood by NIR spectroscopy that have been overcome by other research areas where NIR spectroscopy is commonly used. One of the most important issues is the applicability of calibrations to new sites. Even with a multi-site calibration there is no guarantee that it will perform well when applied to samples from a new location, and there is no way of testing if the calibration will perform adequately. Samples from the new location can be added to the calibration to improve its performance on the new site, but most NIR users do not want to continually update their calibrations. The generation of large spectral databases is under way with the aim of providing calibrations that can be applied to new locations with confidence, but for properties such as pulp yield, the expense associated with determining it accurately prohibits the analysis of thousands of samples. Possibly networks of users will be formed, similar to those in agriculture, and the sharing of calibration sets will become common; however, this seems unlikely at present. Probably the best chance for the development of large spectral databases of characterised samples will be using samples analysed by SilviScan. The creation of large sets of spectra will also make it possible to explore the LOCAL approach for predicting wood properties.

In terms of commercialisation NIR spectroscopy, as a technique for estimating wood properties, lags behind both SilviScan and the new generation of acoustic tools presently available. Recently Paprican and Analytical Spectral Devices announced that they will develop an instrument based on NIR spectroscopy to estimate Kappa number and pulp brightness (TAPPI 2007). For the commercialisation of wood property estimation by NIR spectroscopy to become a reality on a large scale, the issues regarding the application of existing calibrations to new sites must be overcome and the estimation of wood properties in the field by NIR spectroscopy must become a reality.

Important questions also relate to the best way to utilise NIR spectroscopy for the estimation of wood properties, and how and by whom the analysis should be conducted. Expectations have consistently been over-optimistic about the estimation of wood properties by NIR spectroscopy and many NIR predictions of wood properties have not been considered sufficiently accurate for practical purposes. If, for example, NIR spectroscopy was used to estimate the pulp yield of

a single tree, then chances are it would provide an estimate that would disappoint the user. But if it is used to estimate population parameters based on a reasonably large sample set then it will provide excellent data. Likewise for the estimation of genetic parameters and identification of superior genotypes NIR spectroscopy is well suited owing to its ability to analyse large numbers of samples quickly and to predict multiple properties.

Presently several organisations and companies have investigated using NIR for research purposes and there has been considerable repetition of effort, hindering progress. For companies that do not want to continually update calibrations, a possible alternative is to establish a collaborative research centre devoted to NIR spectroscopy where member companies contribute to an operating budget, develop research objectives, and share research findings. Alternatively, companies can continue to develop their NIR applications internally or pay to have samples analysed on a per sample basis by a research provider.

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