Characterizing canopy biochemistry from imaging spectroscopy and its application to ecosystem studies

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1. Introduction

The first studies reporting on the application of airborne imaging spectrometer data to quantify non-pigment biochemical components of vegetation canopies were published by Wessman et al. (1988) and Peterson et al. (1988). Since then, remotely sensed data from imaging spectrometers have continued to be improved and applied to quantify vegetation constituents such as water, nitrogen, protein, cellulose, and lignin (Card et al., 1988; Wessman et al., 1989; Matson et al., 1994; Zagolski et al., 1996; Martin & Aber, 1997; Roberts et al., 1997; Ustin et al., 1998; Serrano et al., 2002; Smith et al., 2003). In recent work, maps of canopy chemistry have revealed patterns in key ecosystem processes, such as net primary production, at fine-scale over large areas (Smith et al., 2002; Ollinger & Smith, 2005), as well as the spatial distribution of plant functional types and species (Asner & Vitousek, 2005). Concurrent with advances in imaging spectroscopy applications, the airborne instruments have improved substantially (e.g., Green et al., 1998) and have become more readily available. Some of the most widely used sensors include the Airborne Visible/InfraRed Imaging Spectrometer (AVIRIS; Green et al., 1998), the HyMap Imaging Spectrometer (HyMap; Cocks et al., 1998), and the Compact Airborne Spectrographic Imager (CASI; ITRES Research Limited, Alberta, Canada). Imaging spectrometers have also been deployed aboard satellite platforms (e.g., Earth Observing-1 Hyperion; Ungar et al., 2003). Improved measurements from imaging spectrometers, and increased availability of data, have greatly expanded the opportunities to use remote sensing to investigate a variety of ecosystem processes (Ustin et al., 2004).

Estimation of canopy chemical properties from imaging spectrometer data grew from laboratory research on animal feed and forage quality (e.g., Norris et al., 1976). These studies were conducted under controlled conditions using statistical relationships, mainly stepwise multiple linear regression (SMLR), between laboratory spectra and biochemical assays of nitrogen and protein (e.g., Marten et al., 1989). Procedural limitations in the estimation of nitrogen and protein included statistical overfitting, having too many independent variables to predict a single dependent variable (Martens & Naes, 2001), and the difficulty in extending predictive equations to other data sets (Grossman et al., 1996). In recognition of these problems, there has been an increase in the number of studies applying statistical methods that rely on mathematical treatments of the full spectrum, e.g. Partial
Leastsquares (PLS) regression, instead of a few selected and highly-correlated channels chosen by SMR (e.g., Williams & Norris, 2001; Smith et al., 2002). Another focus has been on the detailed examination of leaf and canopy spectra in wavelength regions where biochemical constituents of interest display strong absorption features. Photon transport models have also been increasingly used to predict spectroscopic reflectance signatures of vegetation and to estimate canopy chemistry via model inversion using actual or simulated airborne imaging spectrometer data (e.g., Jacquemoud et al., 1995, Cecatto et al., 2002).

In this paper we focus our review of recently published literature to those that take a “spectroscopic” approach from the laboratory to the remote sensing level of measurement. In these studies, changes in reflectance spectra are examined at wavelengths within the range of absorption features caused by the chemical bonds in the biochemical of interest. These studies apply algorithms that are sensitive to, and dependent upon, changes in reflectance over a series of contiguous channels, and thus reflectance as a function of wavelength, either across a subset of the spectral range or across the full spectral range measured by the sensor. At the remote sensing level, we term such approaches “spectroscopic remote sensing.”

We restrict our discussion to non-pigment biochemical constituents of plants. We recognize that leaf pigment composition, including chlorophyll and accessory pigments, is tremendously important in vegetation remote sensing, and is reviewed by Ustin et al. (this issue). The absorbance spectrum of extracted leaf pigments in Fig. 1 shows that pigments absorb strongly across the visible region from 0.35 to 0.70 μm. Thus, our focus in this paper will be on wavelengths greater than 0.70 μm, where important non-pigment leaf constituents, specifically water, nitrogen, cellulose and lignin, have measurable absorption and scattering features. Our goal is to summarize recent articles that have advanced our understanding of how vegetation reflectance spectra represent multiple biochemical properties at leaf and canopy scales. Furthermore, we present recent studies showing how maps of leaf constituents facilitate an examination of ecosystem processes over large areas. We have partitioned the review into three topics: 1) empirical and modeling studies on the effects of plant biochemical constituents on reflectance spectra, 2) remote sensing applications of imaging spectrometers to characterize canopy chemistry, and 3) the use of imaging spectrometers to study primary production and nutrient cycling.

### 2. Leaf spectra and biochemical constituents

Table 1 shows a summary of the average concentrations of these constituents, as well as the range in their concentrations, for deciduous, coniferous, and graminoid leaves compiled by the Accelerated Canopy Chemistry Program (ACCP, 1994). ACCP was a program developed during the original NASA High Resolution Imaging Spectrometer (HIRIS; Goetz & Davis, 1991) to advance canopy chemical studies for that mission and beyond. Although we recognize that the ACCP dataset does not represent the full range of leaf biochemical concentrations found in ecosystems (e.g., Wright et al., 2004), it has associated leaf reflectance spectra that can be examined to better understand spectral variations caused by changes in biochemical constituents. It is useful to note that other leaf spectral–chemical data sets are available to the community as well (Hosgood et al., 1994).

Water is often the most abundant chemical in leaves. Looking at the spectra of fresh versus dried vegetation, we can clearly see the effects of water on reflectance spectra. Fig. 1 shows the reflectance spectrum of fresh pine needles (thick line) in comparison to their spectrum after drying (thin line). The peaks in the absorption coefficient of water at 0.97, 1.20, 1.45, and 1.93 μm are also expressed as local decreases in the reflectance spectrum of the fresh leaf centered near 0.98, 1.19, 1.44, and 1.92 μm. These dips in the water absorption coefficient are centered near 0.98, 1.19, 1.44, and 1.92 μm. These dips in the water absorption coefficient are centered near 0.98, 1.19, 1.44, and 1.92 μm.

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### Table 1

Average percentage by mass leaf biochemical constituents in deciduous and coniferous leaf samples from the Accelerated Canopy Chemistry Program data set (ACCP, 1994).

<table>
<thead>
<tr>
<th>Vegetation type</th>
<th>n</th>
<th>Biochemical concentration (%)</th>
<th>Mean (std. dev.), minimum to maximum</th>
</tr>
</thead>
<tbody>
<tr>
<td>Water (% fresh leaf weight)</td>
<td>Nitrogen (% dry leaf weight)</td>
<td>Water (% dry leaf weight)</td>
<td>Cellulose (% dry leaf weight)</td>
</tr>
<tr>
<td>Deciduous 366</td>
<td>59.9 (4.9)</td>
<td>2.20 (0.53)</td>
<td>21.47 (4.36)</td>
</tr>
<tr>
<td>41.9 to 70.3</td>
<td>1.02 to 3.51</td>
<td>12.42 to 33.41</td>
<td>24.20 to 67.57</td>
</tr>
<tr>
<td>Coniferous 268</td>
<td>55.6 (4.04)</td>
<td>1.18 (0.31)</td>
<td>23.56 (3.77)</td>
</tr>
<tr>
<td>35.4 to 66.3</td>
<td>0.62 to 2.09</td>
<td>13.75 to 31.00</td>
<td>23.69 to 49.89</td>
</tr>
<tr>
<td>Cultivated grass 69</td>
<td>–</td>
<td>0.91 (0.19)</td>
<td>14.70 (1.06)</td>
</tr>
<tr>
<td>0.54 to 1.29</td>
<td>11.62 to 16.65</td>
<td>50.83 to 62.69</td>
<td>3.16 to 5.40</td>
</tr>
<tr>
<td>Wild grass 8</td>
<td>–</td>
<td>0.26 to 0.85</td>
<td>15.10 to 20.73</td>
</tr>
</tbody>
</table>

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reflectance spectrum indicate areas of relatively stronger absorption and are referred to as absorption features. The wavelength positions of the band centers of absorption features are defined as the minimum value in the continuum-removed reflectance spectrum (Clark & Roush, 1984). The spectrum of the dried leaf shows three absorption features, centered near 1.7, 2.1, and 2.3 µm, that were not easily discernible in the fresh leaf spectrum. These three features are caused by several leaf biochemical constituents, the most abundant and widely studied of which are nitrogen (in proteins), cellulose and lignin (Curran, 1989; Elvidge, 1990; Wessman, 1990; Kokaly, 2001). As leaves and plants vary in the concentrations of these constituents, their reflectance spectra vary by changing strengths of the related absorption features. In the next sections of this paper, we will examine recent advances in our knowledge of how these non-pigment leaf constituents alter leaf and canopy reflectance.

2.1. Water

Water is one of the most important factors regulating plant growth and development in ecosystems (Kramer & Boyer, 1995). The effects of water limitation are most evident in species found in arid and semi-arid regions, where adaptations in plant form and physiology function to conserve water, e.g., the Crassulacean Acid Metabolism (CAM) photosynthetic pathway (Bhagwat, 2005). Additionally, water limitation is now widely recognized as a major control even over humid tropical forest dynamics (Asner et al., 2004a, Nepstad et al., 2004). At the leaf level, water is required for the maintenance of leaf structure and shape, thermal regulation, and for photosynthesis. The loss of water from leaves is regulated by cells that control gas exchange between the leaf and the atmosphere (Zeiger, 1983).

In the ACCP dataset, leaf water content averages 60% and 56% by fresh weight for deciduous and coniferous leaves, respectively (Table 1). Fig. 2 shows the modeled effect of increasing water content on the dry leaf absorption features (Kokaly & Clark, 1999). Shown for comparison is the modeled absorption of a 0.2 mm thickness of water. As the water content increases to 60%, the wavelength positions of the dry leaf absorption features shift as the water absorption increases, and the modeled leaf spectrum approaches the spectrum containing dry matter and maximum water concentration (Fig. 2). Early research modeled leaf reflectance as largely arising from the influence of leaf water (Gates et al., 1965). Gao and Goetz (1990) expanded our ability to measure and model a combination of absorptions of atmospheric water vapor, liquid water, and dry vegetation in reflectance spectra measured by airborne spectrometers. Green et al. (1991) used similar spectroscopic approaches to establish leaf and canopy Equivalent Water Thickness (EWT) that integrate reflectance from 0.867 through 1.049 µm, whereas Jacquemoud and Baret (1990) developed one of the first physically-based leaf optical models explicitly sensitive to water content. Roberts et al. (1997) further demonstrated the ability to estimate leaf water thickness by analyzing reflectance from 0.867 through 1.686 µm and by modeling atmospheric water vapor and liquid water in imaging spectrometer data. With these methods, canopy water content has been repeatedly quantified (reviewed by Ustin et al., 2004).

Spectroscopic water features have been quantified and used to map vegetation water content and canopy water stress in a variety of ecosystems. For example, Ustin et al. (1998) and Serrano et al. (2000) used the near-infrared (NIR) water features to detect and map canopy water content in chaparral shrubland vegetation. Similarly, Roberts et al. (2004) used the NIR water features to map canopy water content in a temperate forest, and found that canopy water content was a better metric of canopy leaf area index (LAI) than the traditional normalized difference vegetation index (NDVI). Asner et al. (2004a) used the 1.19 µm water feature to estimate canopy water content in a humid tropical forest in the Brazilian Amazon. The authors related decreases in the dry-season spectroscopic water feature to a decrease

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Fig. 2. Modeled leaf reflectance as a function of increasing water content based on dry white pine needles (the 0% water spectrum). The band center wavelength positions of strong absorption features are given. Spectra are offset by 0.3 for clarity. Figure is adapted from Kokaly and Clark (1999).

Fig. 3. Reflectance spectra for a single oak leaf on a black background (thick line) and for a stack of four oak leaves (thin line). Large differences in spectral shape are seen for near-infrared plateau (the shaded area from 0.78 to 1.3 µm), for the depths and shapes of the 0.98 and 1.19 µm leaf water absorption features, and for the slope of the reflectance curve in the leading edge region (the dark shaded area from 0.78 to 0.91 µm) of the NIR plateau. Reflectance levels at 0.82, 0.86, 1.24, and 1.6 µm are labeled.
in field-measured canopy water content (the convolution of LAI and leaf water concentration). In contrast, NDVI was found to be insensitive to these subtle dry-season effects on canopy properties.

Many empirical studies have evaluated water content algorithms based on analyses of leaf spectra, where leaves were measured in an integrating sphere or as a stack of foliage (Kokaly & Clark, 1999; Sims & Gamon, 2003; Stimson et al., 2005; Yu et al., 2000). It is important to recognize that leaf spectra measured using either of these methods are not direct representations of whole-canopy spectra (Myneni et al., 1989). In Fig. 3, the spectral measurement made with an integrating sphere displays relatively weak 0.98 and 1.19 µm absorption features on the NIR plateau compared to the stacked leaf spectrum. In addition, the leading edge of the near-infrared plateau (0.78 to 0.91 µm) changes from flat and level in reflectance for the single leaf spectrum to a line with positive slope in the stacked leaf spectrum. In contrast to the single leaf spectra, the stack of leaves has a NIR reflectance in the 70% range, a value that is rare in canopy spectra, where NIR reflectance typically falls in the range of 25–50%. This exaggerated reflectance occurs because stacked leaves do not allow sufficient inter-leaf attenuation of NIR light, caused by scattering out of the optical path of the sensor as well as reabsorption of scattered light by the undersides of foliage (NIR reflectance of canopies is dominated by scattering processes not absorption; Baret et al., 1994). In reality, canopy spectra of live vegetation have spectral properties that differ from the stacked leaf spectrum and the single leaf measurement in Fig. 3. Scattering of radiation in the canopy, as influenced by plant and canopy architecture, enhances the leaf-level water absorption features and raises the reflectance level of the NIR relative to other wavelength regions (Gao, 1996; Asner, 1998).

Problems associated with scaling measurements from leaf to canopy levels remains a major challenge in the remote sensing community. Equally important is the recognition that there often exist major covariances between vegetation structure and chemistry, which make the deconvolution of these vegetation properties very challenging with imaging spectroscopy. To address this issue, leaf hemispherical reflectance measurements taken in conjunction with leaf transmittance measurements are often combined and scaled up to canopy reflectance using a number of canopy radiative transfer models. Using this approach, Jacquemoud et al. (2000), and many others, have shown how water is expressed throughout the NIR and shortwave-infrared (SWIR) spectral range (~800 nm to 2500 nm), and how radiative transfer models can be used to estimate leaf equivalent water thickness via model inversion. Cecatto et al. (2001) also used a radiative transfer model to develop NIR and SWIR indexes sensitive to leaf and canopy properties.
water content. Many other examples are available in the literature showing that both forward and inverse model simulations can be used to simulate and to estimate leaf and canopy water content (see Jacquemoud et al. this issue for an extensive modeling review).

2.2. Nitrogen

Nitrogen (N) is a relatively small component of leaf dry weight, covering a range of as low as 0.26% in some grasses to 3.5% in broadleaf deciduous samples from the ACCP dataset (Table 1). Despite its small contribution to leaf mass, field studies have found N to be strongly linked to ecosystem functions such as photosynthesis and net primary production (Field & Mooney 1986, Schimel et al., 1997, Smith et al., 2002) as well as biogeochemical properties such as soil C:N ratios and nitrate production (Ollinger et al., 2002). Foliar N is also widely used as a parameter in ecosystem models (e.g., Parton et al., 1995; Ollinger & Smith, 2005). Nitrogen has been quantified at leaf and canopy scales using reflectance measurements (e.g. Wessman et al., 1988, Curran et al., 1997, Smith et al., 2002), which may seem surprising because it is such a small component of the leaf. Nitrogen occurs primarily in
proteins and chlorophylls in the leaf cells. Proteins are the major nitrogen-containing biochemical constituent of plants. A single protein, ribulose-1,5-bisphosphate carboxylase-oxygenase (rubisco) accounts for 30–50% of the N in green leaves (Elvidge, 1990), where it is found in high concentration in the stroma of chloroplasts (Sainsi & Melzer, 2005; Douce & Heldt, 2000); because of its abundance in plants, rubisco is considered the most plentiful protein on the planet. Rubisco is the principal CO₂-fixing enzyme in C₃ plants and the ultimate CO₂-fixing enzyme in C₄ and CAM plants. Nitrogen is also 6.5% (by weight) of chlorophylls, which are the primary light harvesting molecules in the photosynthesis process that converts carbon dioxide and water into carbohydrate.

Many researchers have associated spectroscopic estimation of nitrogen to that of chlorophyll pigments, based on the fact that the two variables are moderately correlated within and across ecosystems (r² usually ranging from 0.4–0.6; Wright et al., 2004). Mutanga et al. (2003) studied the effects of increasing N on the chlorophyll absorption feature centered near 0.68 μm. Results of their experiment showed that increased fertilization with N resulted in increased chlorophyll absorption that deepened and widened the chlorophyll absorption feature until eventually reaching a point of saturation. Nonetheless, this experiment illustrated a frequent observation that changes in non-pigment constituents such as nitrogen are also correlated to channels in the visible wavelength region where the reflectance levels are dominated by pigments (LaCapra et al., 1996; Martin & Aber, 1997; Serrano et al., 2002).

A direct connection between N content and pigments that influence reflectance at visible wavelengths results from the fact that four N atoms in the central tetra-pyrrole head of the chlorophyll molecule act to stabilize the central magnesium ion. However, this link can directly explain only a small portion of the total contribution of N to leaf and canopy reflectance because only about 19% of leaf N in C₃ plants is allocated to light harvesting complexes, and only 1.7% of leaf N is directly held in chlorophyll (Evans, 1983). In contrast, roughly 70% of leaf N is tied up in molecules that support carbon fixation (Chapin et al., 1987). These N-containing compounds include biosynthetic and CO₂-fixing molecules such as rubisco. Although nitrogen-limited ecosystems, such as many temperate forests, show about a 50% covariance between leaf chlorophyll and total N (Sterner & Elser, 2002), variation in this relationship is expected to result from variation in environmental factors (most notably, light) that affect optimal N allocation between light harvesting compounds and carboxylating enzymes. Further, ecosystems such as the humid tropics, where N limitations are less important, show a pronounced decoupling of leaf chlorophyll and total N (Asner & Martin, 2008), likely due to other competing evolutionary factors such as N allocation to defense compounds.

Recent studies have provided additional insight into why nitrogen, despite being a small component by leaf weight, is successfully estimated from reflectance measurements of leaves and canopies. Kokaly (2001) showed that as N increases, changes in leaf reflectance of dry leaves occur in the NIR absorption feature centered at 2.1 μm (Fig. 4A). The changes were shown to be caused by two absorption features at 2.055 and 2.172 μm that are situated on the shoulders of the 2.1 μm absorption (Fig. 4B), corresponding in wavelength position and shape with the absorption features of proteins (Fig. 4C). These protein absorptions arise from vibrations of N-containing amide bonds that form the backbone of the protein structure and are repeated along the length of each molecule. Furthermore, as Fig. 4C shows, these absorptions are offset from the more centrally located absorptions of lignin and cellulose at 2.102 and 2.144 μm, respectively, accentuating the influence of this biochemical constituent so that it has an observable impact on reflectance spectra (Kokaly 2001).

Modeling the spectral contributions of leaf N has been challenging. The original version of PROSPECT (Jacquemoud & Baret, 1990) attempted to incorporate N into the absorption and scattering processes represented in the model. This was later abandoned due to inconsistencies in the retrieval of N via model inversion. This is not surprising given the enormous range of leaf compounds containing N and the varied functional properties of those compounds (Chapin et al., 1987). The LIBERTY model also attempted to incorporate leaf N concentration as a parameter for needleleaf simulations (Dawson et al., 1998), showing that it could be done but that there were strong covariances between N and other leaf properties.

2.3. Lignin/cellulose

Cellulose is a polymer of glucose molecules and is the main constituent of plant cell walls. Cellulose received its name in 1839 (Brognaert et al., 1839), following the work of French chemist Anselme Payen who described the resistant fibrous solid remaining after treating plant tissues with acids, ammonia, and solvents (Payen, 1838). Cellulose concentration averages from 37% in conifer to 65% in grasses in the ACCP dataset (Table 1). It is an end-product of the carbohydrate produced by plant metabolic pathways of photosynthesis, and is considered to be among the most abundant forms of living terrestrial biomass (Crawford, 1981). Lignin, one of the plant polyphenolic compounds, is a complex, hydrophobic molecule of aromatic nature, primarily comprised of oxyphenylpropae units assembled in a large macromolecule polymer with molecular mass in excess of 10,000 unified atomic mass units (u). Cellulose is used by plant leaves for the wall of parenchyma cells, whereas lignin is used in...
the secondary cell walls of xylem and sclerenchyma (vascular fibers). Differences in the ratio of lignin to cellulose are largely due to differences in the amounts of tissue types within a leaf, rather than a single tissue with varying amounts of lignin. Lignin is responsible, along with cellulose, for the rigidity of plant cell walls, and averages from a low of 15% in cultivated grass species to a high of 24% in conifer single tissue with varying amounts of lignin. Lignin is responsible, along with cellulose, for the rigidity of plant cell walls, and averages from a low of 15% in cultivated grass species to a high of 24% in conifer 

Field studies have shown that lignin is resistant to decomposition and that lignin concentrations in plant litter exert a strong influence on soil nutrient cycling (Aber & Melillo, 1982). Quantification of leaf cellulose and lignin in the plant canopy, before leaves fall to the ground as litter, offers a potential link to understanding rates of nutrient cycling in soils. Motivated by its demonstrated effect on rates of N mineralization, lignin (along with N) was quantified in the earliest studies of canopy biochemistry (Wessman et al., 1988; Peterson et al., 1988; Wessman et al., 1989). In general these studies of lignin, while successful, have shown lower correlation coefficients and higher errors of estimation when compared to the results for nitrogen concentration. In part, this has been explained by the error in quantifying lignin in the laboratory (e.g. LaCapra et al., 1996), as its chemical structure is variable in the relative content of precursor alcohols, coniferyl, sinapyl, and r-coumaryl, from which lignin is polymerized (Crawford, 1981). Furthermore, cellulose and lignin are intertwined in a complex manner, along with other polysaccharides, in plant cell walls (Crawford, 1981), and their spectra have broad and overlapping absorption features (Fig. 5A). Most researchers have therefore lumped these two materials into a more general quantity of non-photosynthetic vegetation (NPV; Roberts et al., 1993; Wessman et al., 1997), cellulose-lignin (Daughtry et al., 2004) or ligno-cellulose (Elvidge, 1988), and dry matter (Jacquemoud et al., 1995; Faret et al., 2008).

Only a few studies have attempted explicit quantification of cellulose as a separate quantity at the canopy level (Castellu-Etchegorry et al., 1995; Zagolski et al., 1996; Curran et al., 1997), achieving moderate success comparable to N quantification. As a primary component of plant litter, the ligno-cellulose feature may be important in quantifying plant litter (e.g. crop residue) contributions to carbon pools (e.g. Nagler et al., 2003). Still, cellulose is an abundant biochemical constituent of plant foliage, and its absorption features are strong and overlapping with lignin and protein; continued research into its quantification as a separate constituent may assist in developing better algorithms for quantifying the biochemical constituents of plants that are more strongly linked to ecosystem processes and for characterizing the qualities of dry plant matter and crop residue.

Recent research has continued to examine the influence of dry plant matter on remotely-sensed vegetation spectra. In a study of dryland ecosystems, Serrano et al. (2002) estimated leaf lignin concentrations in areas with a considerable fraction of dry, senescent or woody vegetation cover and found that lignin is correlated with spectral reflectance at 1.754 μm ($R^2 = 0.44–0.58$). In analyses that included sites with a high fraction of dry plant matter, the correlation between reflectance and lignin concentration decreased. Kokaly et al. (2007) examined dry vegetation spectra and found consistent differences between the spectra of dry grass and dry conifer samples (Fig. 5) caused by biochemical composition. Table 1 shows that conifer needles have a much lower average cellulose to lignin ratio (1.56) than grasses (3.89) in the ACCP data set. The overlapping spectral features of these two constituents contribute to the general shapes of the 2.1 and 2.3 μm absorption features in dry plants. The band center (defined as the wavelength of the minimum value in continuum removed reflectance) of the 2.1 μm absorption feature shifted from a longer wavelength in spectra of dry conifers to a shorter wavelength in spectra of dry grasses, a shift consistent with the absorption of cellulose at a shorter wavelength compared to lignin (Fig. 5). The shape of the 2.3 μm absorption feature was also found to be distinct between spectra of dry grass and dry conifer samples. The triplet absorption feature of dry conifer (with weaker shoulder absorptions at 2.27 and 2.35 μm around the central 2.31 μm absorption) shifts to a doublet shape in the spectra of dry grasses, which is consistent with the doublet absorption feature of cellulose (Fig. 5). In the dry pine spectrum (dominated by lignin), the left shoulder of the 2.3 μm

![Fig. 8. Reflectance spectra of (A) dry vegetation and (B) soils, and tied-SWIR spectra of dry vegetation (C) and soils (D), used for automated spectral mixture analysis of AVIRIS imagery. Figure adapted from Asner and Heidebrecht (2002).](image)
feature is much lower than the right shoulder, consistent with lignin having its absorption centered at this left shoulder position (Fig. 5A). The changes in shape between dry grass and dry conifer features are preserved in canopy level measurements made with AVIRIS (Fig. 5C). Fig. 5 shows how the shifts in spectral shape from high cellulose/low lignin grass to higher lignin/lower cellulose pine needles match the shifts in the absorption features of pure cellulose and lignin.

In contrast to the problems of scaling the reflectance features associated with water from leaf to canopy levels in the NIR (see previous section), scaling of lignin features in the 2.3 µm range is more direct since reflectance is low and thus scattering effects are weak (Asner, 1998). In fact, chemical absorptions in the visible (pigments) and SWIR (lignin, cellulose, protein-N) ranges are more directly scalable to the canopy level simply because scattering caused by canopy structural variation is low in these wavelength regions as compared to the NIR where water dominates. Modeling studies that support these findings have continued to evolve in parallel to the empirical research by incorporating the specific absorption spectra of lignin and cellulose (combined) in simulations of leaf and whole-canopy reflectance (Dawson et al., 1998; Ceccato et al., 2001).

3. Ecosystem composition from imaging spectroscopy

Along with improved characterization of non-pigment biochemical constituents, airborne and space-based imaging spectrometers have been applied in ways that broaden our understanding of the basic composition of ecosystems. Here we limit the review to examples where detection of plant species or functional groups have been based on broad differences in biochemistry and the influence of various biochemical constituents on reflectance. Later, we will highlight studies that have dissected the broad spectroscopic signals into more detailed chemical determinations. In each example, analyses of imaging spectrometer data were dependent upon the shape of the reflectance spectra as a function of wavelength; in other words, they took a “spectroscopic” remote sensing approach.

Vegetation composition and species dominance have been intensively studied using the biochemical absorption and scattering features measurable with imaging spectroscopy. For example, Kokaly et al. (2003) applied AVIRIS data to map forest cover types in Yellowstone National Park using the USGS Tetracorder algorithm (Clark et al., 2003) in an analysis of the 0.68 µm chlorophyll absorption feature with the 0.98 and 1.20 µm leaf water absorption features. They showed that the spectral features of four forest cover types, lodgepole pine, whitebark pine, Douglas-fir, and a mixed Engelmann Spruce-Subalpine Fir class, differed significantly in the shape and depth of these three absorption features. By analyzing these absorption features in each spectrum, Kokaly et al. (2003) mapped the distribution of whitebark pine in Yellowstone (Fig. 6). Townsend et al. (2003) and Plourde et al. (2007) used Hyperion and AVIRIS, along with spectral analysis of nitrogen concentrations, to map canopy compositions in the Yellowstone Park area.

Fig. 9. Fractional cover map of photosynthetic vegetation (PV, in red), non-photosynthetic vegetation (NPV, in green), and soil (in blue) of the Nacunan Biosphere Reserve (yellow lines) in central Argentina, derived from AVIRIS SWIR spectral signatures. Figure adapted from Asner and Heidebrecht (2002).
species in an eastern deciduous forest. Fuentes et al. (2001) used AVIRIS data to derive indices of leaf pigments and water content, which were in turn used to map boreal forest vegetation in Canada. The use of canopy chemistry in mapping species composition stems from earlier studies that showed ecological patterns emerging from canopy chemical mapping to be highly correlated with taxonomic composition (e.g. Wessman et al., 1988). In one study, Martin et al. (1998) found that most of the common tree species in a north temperate forest could be identified by their unique combinations of N and lignin concentrations.

In rainforest canopies of Hawaii, Asner and Vitousek (2005) used airborne imaging spectroscopy and a new photon transport modeling approach to quantify canopy water content and upper-canopy leaf N concentration. Their water and N maps indicated how biological invasion altered the chemistry of forest canopies across a Hawaiian montane rain forest landscape (Fig. 7). They found that the N-fixing tree *Morella faya* doubled canopy N concentrations and water content as it replaced native *Metrosideros* forest. Furthermore, they found that the understory herb *Hedychium gardnerianum* reduced N concentrations in the native forest overstory and substantially increased aboveground water content. In this case, spectroscopic remote sensing provided fundamentally new information on ecosystem processes that had not been previously detected or measured in the field; subsequent field studies confirmed the remotely sensed measurements.

Another broad use of non-pigment biochemical analysis with imaging spectroscopy has focused on surface cover of live and dead tissues. The goals here have been to provide highly automated rapid analysis of changing ecosystem conditions without the detailed chemical and taxonomic properties of the vegetation becoming overly expressed. In a sense, this is a conservative use of biochemical mapping techniques, but one which can form a foundation for more detailed analyses with imaging spectrometer data (e.g., Wessman et al., 1997, Roberts et al., 1998). For example, Asner and Heidebrecht (2002) showed that by analyzing reflectance spectra in the 2.03–2.50 µm region, after normalization to the reflectance at 2.03 µm, the amount of dry vegetation cover could be quantified with high precision and accuracy throughout arid and semi-arid regions. Fig. 8 shows the spectra of the soil, green vegetation, and dry vegetation endmembers used in the study. The dry vegetation endmember shows the strong 2.1 and 2.3 µm absorption features caused by dry leaf and
dry woody material, independent of species composition or many other confounding factors. In a subsequent dryland study of Argentina, Asner et al. (2003) used the same method to map an aridlands region subjected to long-term grazing. The resulting maps of sub-pixel fractional photosynthetic and NPV cover revealed spatial patterns related to aboveground and belowground carbon storage and nutrient cycling in this semi-arid ecosystem (Fig. 9).

Fire is a widespread disturbance to forest ecosystems and the global carbon cycle, impacting net primary production (Randerson et al., 2006), releasing stored carbon to the atmosphere as CO₂ (Andreae and Merlet, 2001), and increasing release of carbon through post-fire decomposition of scorched vegetation (Auclair and Carter, 1993). Roberts et al. (1998) used spectral mixture analysis to separate fire fuels and dominant species in chaparral ecosystems of California. This study illustrated how the spectroscopic signatures of non-pigment biochemical composition, seen in remotely sensed spectra as shifts in spectral shape from high cellulose/low lignin grass to lower cellulose/higher lignin woody plant foliage, can be used to better understand the distribution of fire-prone species across a landscape. Similarly, Kokaly et al. (2007) were able to differentiate scorched conifer trees from other non-photosynthetic vegetation and map their distribution on the post-fire landscape at the Cerro Grande fire, which occurred in the summer of 2000 near Los Alamos, New Mexico (Fig. 10). Direct analysis of the 2.1 and 2.3 µm absorption features in AVIRIS data was used to define the width of scorched conifer zones along the edges of severely burned areas.

4. Spectroscopic studies of ecosystem processes

Early applications of airborne imaging spectrometry to quantify specific chemical constituents in foliage by Peterson et al. (1988) and Wessman et al. (1988) were conducted with the broader objective of examining spatial patterns of N cycling across forested landscapes. Subsequent studies have continued to develop and improve these methods (Matson et al., 1994; Castellu-Etchegorry et al., 1995; Zagolski et al., 1996; LaCapra et al., 1996; Martin and Aber 1997; Serrano et al., 2002; Ollinger et al., 2002). Recently, Smith et al. (2002) extended the application of imaging spectrometers to quantify canopy N content for net primary production (NPP) studies over a large area of U.S. deciduous and coniferous forest. Using a PLS regression approach with AVIRIS data, they developed robust N predictions, validated by field measurements with $r^2 = 0.71$ and standard error of the estimate (SEE) of 0.19% N.

Ollinger and Smith (2005) used imaging spectrometer-derived estimates of canopy N to conduct ecosystem model simulations for Bartlett Experimental Forest, NH. Combining remotely sensed N estimates with climate variables and a deciduous-evergreen unmixing model, the PnET-II ecosystem model (Aber et al., 1997) was run to produce NPP maps over an area containing an unusually dense set of field-based growth estimates (Fig. 11). Modeled aboveground NPP was partitioned into foliar NPP ($r^2 = 0.77$; SEE = 12.6%) and wood NPP ($r^2 = 0.74$; SEE = 11.9%). Predictions generated by running the model with either AVIRIS- or Hyperion-derived N estimates yielded far greater accuracy than those generated under the more common method of using mean field-based values for dominant vegetation types.

In a related study that focused on spatial patterns of N cycling in the White Mountain National Forest, Ollinger et al. (2002) examined species-level leaf N concentrations with respect to soil N availability and found that both vary as a function of stand age and disturbance history. For deciduous forests, leaf N concentrations were typically higher in old and relatively undisturbed stands than in younger stands of similar composition and in earlier stages of recovery from disturbance (clear-cutting or fire). This was attributed to disturbance...
effects on N mineralization in the soil and resulted in a positive relationship between N mineralization and foliar N concentrations. Interestingly, this trend was not observed in evergreens, which maintained consistently lower foliar N concentrations. Combining field results with AVIRIS-derived estimates of foliar N and lignin:N ratios yielded detailed spatial estimates of soil C:N ratios and the fraction of the landscape subjected to N loss through nitrification.

5. Discussion

The biochemical properties of vegetation canopies are directly expressed in the reflectance signatures that can be derived from measurements made by imaging spectrometers. A variety of elemental and molecular interactions with shortwave radiation cause scattering and absorption features in the 0.4 to 2.5 \( \mu m \) range associated with water, carbon, nitrogen, pigment and other chemicals. Our knowledge of the fundamental expressions and controls over spectral signatures continues to increase, allowing for more quantative application of imaging spectroscopy to ecosystem questions on land and in aquatic environments. To date, canopy chemical studies based on a spectroscopic remote sensing approach have yielded reflectance signatures consistent with the absorption features of individual biochemical constituents such as: (a) water reducing reflectance at longer wavelengths, (b) nitrogen increases causing greater chlorophyll absorption near 0.68 \( \mu m \) and protein-related absorption at 2.05 and 2.17 \( \mu m \), (c) cellulose-to-lignin ratio changing the 2.1 and 2.3 \( \mu m \) features. These have been observed at both the leaf and canopy levels.

Our review of the literature revealed technical problems in the use of field and airborne spectrometers for conducting spectroscopic research. Many published studies show field spectra that are noisy at wavelengths greater than 1.7 \( \mu m \); this noise limits the full exploitation of the 2.1-2.4 \( \mu m \) absorption features of dry plants and the comparison of observations in these studies to others with higher signal-to-noise ratio (SNR). For spectrometers with high noise levels in this region, measurement procedures should include more replicate measurements and/or greater averaging times in order to increase SNR. In addition, recent studies have used channels within wavelength regions of strong atmospheric absorption to correlate to biochemistry. This should be avoided because local variations in atmospheric conditions and uncertainty in atmospheric correction greatly impact reflectance levels in the wavelength regions of water vapor and carbon dioxide absorption, not the chemicals of interest. Atmospheric correction in general needs to be improved, as most radiative transfer models show strong atmospheric residuals in the derived surface reflectance spectra (Green et al., 1998). These residuals can be reduced by ground calibration using field spectrometer measurements to derive empirical correction factors (Clark et al., 2002), but widespread application of imaging spectrometer data to ecosystems requires, in particular, better correction for radiative absorption by water vapor (as the water vapor residuals corrupt the leaf water features) and for atmospheric scattering (as errors in scattering correction can affect shorter wavelengths, less than 0.6 \( \mu m \), where absorption features of accessory pigments are centered).

Despite advances in our understanding and application of spectroscopic remote sensing to ecosystem questions, a wide range of technical uncertainties and issues still need to be addressed. First, the community should consider the complexities of related chemicals (e.g., tannin vs. lignin, and cellulose/starch/hemi-cellulose) that have very similar spectral absorption features. A similar issue exists for the 12–16 pigments that are actively expressed in the visible portion of the spectrum. That is, we need to understand the expression of specific elements (C and N) in the molecules that have signatures in the SWIR spectrum. In addition, an increased understanding of leaf and canopy reflectance that accounts for the overlapping absorption features of these elemental and molecular constituents may allow a better approach to quantifying non-pigment biochemical constituents. Coupled leaf and canopy radiative transfer models have already helped untangle overlapping spectral features, and thus to derive suites of biochemicals from spectral signatures (Jacquemoud et al., this volume; Jacquemoud et al., 1995; Ceccato et al., 2002). These models are evolving to include more biochemical constituents (Faret et al., 2008), but those efforts are also hampered by lacking field and laboratory data on multiple element and molecular stoichiometries.

Beyond the basic chemical-spectroscopic linkages, we do not yet have a clear understanding of the biological and ecological controls over biochemical composition and their potential covariation with other plant traits that can influence reflectance. Much progress has been made in areas such as the development of linkages between leaf nitrogen and physiology of plant canopies (e.g., Field and Mooney 1986, Reich et al., 1997). Similarly, we know a great deal about the co-variance between leaf nitrogen and phosphorus (McGroddy et al., 2004, Townsend et al., 2007), and the N-P link has been applied to hyperspectral reflectance data in montane tropical forests (Porder et al., 2005). In that case, N was estimated by way of its stoichiometric link to N, which was the remotely sensed element. Nonetheless, linkages among elements and molecules are rare in remote sensing, and a wide range of biochemicals such as lignin, cellulose, hemi-cellulose, starch, sugars, nutrients, and pigments have not been systematically explored across environmental gradients or taxonomic lines.

In this paper, we emphasized the role that spectroscopic remote sensing plays in studies of ecosystem form and function. Spectroscopic remote sensing relies on high-performance measurements of scattering and absorption features caused by chemical bonds in materials — in our case, leaf biochemical constituents. Although many applications have involved a reduction of the data, to decrease the dimensionality of the information provided by spectrometers, a growing number have also shown the utility of obtaining full spectral properties and being able to identify distinct reflectance features in a variety of spectral regions.

Existing and planned spectrometers face the issue of sensor fidelity, which refers to the quality and usability of a spectroscopic signature. Sensor fidelity relies on the signal-to-noise performance, uniformity, and stability of an imaging system. Whereas many imaging spectrometers have been built, a small fraction provides high fidelity data required for true spectroscopic remote sensing studies. Spectroscopic remote sensing can act as a bridge between plot level sampling and 10–30 meter-scale remote sensing over 100–10,000 km² areas, and could also serve as a bridge to continental and global-scale instruments, where large pixel size precludes direct comparison with plot measurements of important ecosystem properties (Turner et al., 2004). AVIRIS and HyMap are among the very few airborne sensors to provide the requisite data; no spaceborne spectrometers (including EO–1 Hyperion, Ungar et al., 2003) have done so. With that in mind, the Flora imaging spectrometer mission was conceived (Asner et al., 2004b), and successfully passed through the U.S. National Academy of Sciences Decadal Survey (NAS, 2007) review. Today, this effort has evolved into the new HyspIRI satellite project, which remains at an early science-planning stage at NASA (http://hyspiri.jpl.nasa.gov/). Earlier science and sensor design developments have also been taken place, and are continuing today. HySpIRI and similar programs, such as the European Union’s EnMap mission (Stuffer et al., 2006), hold great promise for advancing the science of spectroscopic remote sensing to the global arena, with high-fidelity spectrometers mapping ecosystems into the future.

6. Conclusions

In the past 20 years, a large and growing body of literature has demonstrated the successful use of imaging spectroscopy to quantify water, nitrogen, cellulose, and lignin concentrations in plants. These quantifications have been made across measurement scales, from leaf reflectance measurements made in the laboratory, to whole plant


