

# Article Mapping Foliar Nutrition Using WorldView-3 and WorldView-2 to Assess Koala Habitat Suitability

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Abstract: Conservation planning and population assessment for widely-distributed, but vulnerable, arboreal folivore species demands cost-effective mapping of habitat suitability over large areas. This study tested whether multispectral data from WorldView-3 could be used to estimate and map foliar digestible nitrogen (DigN), a nutritional measure superior to total nitrogen for tannin-rich foliage for the koala (*Phascolarctos cinereus*). We acquired two WorldView-3 images (November 2015) and collected leaf samples from Eucalyptus woodlands in semi-arid eastern Australia. Linear regression indicated the normalized difference index using bands "Coastal" and "NIR1" best estimated DigN concentration (% dry matter,  $R^2 = 0.70$ , RMSE = 0.19%). Foliar DigN concentration was mapped for multi-species *Eucalyptus* open woodlands across two landscapes using this index. This mapping method was tested on a WorldView-2 image (October 2012) with associated koala tracking data (August 2010 to November 2011) from a different landscape of the study region. Quantile regression showed significant positive relationship between estimated DigN and occurrence of koalas at 0.999 quantile ( $R^2 = 0.63$ ). This study reports the first attempt to use a multispectral satellite-derived spectral index for mapping foliar DigN at a landscape-scale (100s km<sup>2</sup>). The mapping method can potentially be incorporated in mapping and monitoring koala habitat suitability for conservation management.

Keywords: remote sensing; Eucalyptus; digestible nitrogen; koalas; habitat mapping

# 1. Introduction

Mapping and monitoring habitat suitability for threatened species at the landscape and regional scale ( $\geq 100$ s km<sup>2</sup>) is crucial for developing effective conservation management [1–4]. This is especially important for conserving specialist folivores such as the koala, *Phascolarctos cinereus*, the distribution of which at the landscape scale is determined largely by the occurrence of nutrient-rich foliage of food tree species [5,6]. Koalas are listed as 'Vulnerable' in the IUCN red list [7] and are threatened by disease and habitat destruction and fragmentation. Current published mapping of koala habitat suitability relies mainly on vegetation community mapping, especially the relative proportion of primary food tree species [2]. Although the primary food tree species vary in different regional landscapes with different eucalypt species communities, the more frequent koala use of these primary food tree species is related to leaf nutritional quality [4,8] which shows spatial variation [9] and could be affected by water availability and soil fertility [10,11]. Incorporating leaf nutritional information is one of the suggested enhancements to this method [2] because of the patchy distribution of antifeedant chemistry



and forage nutrients found in previous studies [9,10]. Koala food selection is a key determinant of tree use and is influenced by foliar water content, nitrogen, and concentrations of several groups of plant secondary metabolites (e.g., tannins and formylated phloroglucinol compounds) [8,11]. In the past, plant nutritional value to herbivores has been estimated using total nitrogen (N) or protein-to-fibre ratios [12,13]. However, for browse or foliage diets, protein (and hence N) digestibility can be significantly reduced by tannins. Tannins are widespread plant secondary metabolites which form insoluble tannin-protein complexes which resist digestion in the mammalian gut. Digestible N (DigN) concentration can be estimated by an in vitro digestion assay [14] and better reflects the nutritional value of tannin-rich foods, such as eucalypt foliage, to mammalian folivores. DigN has been shown to influence koala feeding decisions [10] and breeding success in eucalypt-feeding common brushtail possums [15]. Because DigN integrates the nutritional information of both attractants and deterrents to feeding, mapping DigN across an area would be another step forward from mapping attractants and deterrents individually [16] to meet the need of more specific measured food quality for herbivore conservation and habitat management.

Determining foliar quality over large areas using field-sampling and chemical analysis involves very large sample sizes and is unavoidably expensive [17–19]. Remote sensing applications offer new opportunities to address this problem by providing proximal data for estimating plant chemistry [20]. An ideal remote sensing data source for mapping foliar DigN would have adequate spatial resolution to isolate individual tree spectra, an appropriate spectral response to provide sensitive indices and a relatively lower cost. It is encouraging that foliar DigN was accurately modelled from airborne hyperspectral data in Australian temperate eucalypt woodlands [19]. However, mapping large areas with airborne remote sensing is generally more expensive than multispectral satellite remote sensing [21]. There is a need to find satellite-derived data that have high spatial resolution and can provide sensitive spectral indices for foliar DigN.

No studies mapping DigN with satellite remote sensing were found during our literature search, but a wide range of studies have used satellite-derived data (from WorldView-2, SPOT, Hyperion, Aster, Sentinel-2, and IKONOS) to map N in crops and trees [20,22–26]. Several suitable spectral indices have been found for mapping N using satellite-derived multispectral data such as Aster [21], Sentinel-2 [26], and WorldView-2 [20]. Testing potential spectral indices may identify appropriate indices for estimating and mapping DigN. Yet the requirement of high spatial resolution for mapping foliar DigN in individual trees narrows the range of suitable sensors. For example, many eucalypt canopies [27] are smaller than the spatial resolution of Hyperion (30 m), Aster (15 m), and SPOT (10 m). On the other hand, DigN results from the interactions of N, tannins and fibre in leaves, which have important absorption features in the shortwave infrared (SWIR) region [19]. Previously, most high-resolution satellite-borne sensors did not collect information in the SWIR spectrum, but since 2014, the multispectral sensor WorldView-3 (WV3) has collected eight SWIR bands at 3.7 m spatial resolution, in addition to eight visible and near-infrared (VNIR) bands (at 1.24 m spatial resolution). Compared to the WorldView-2 (WV2, eight VNIR bands at 1.84 m spatial resolution) and the other space-borne sensors mentioned above, WV3 has higher spatial resolution and an additional eight SWIR bands. A higher spatial resolution can potentially provide more pure pixels with reflectance from trees and reduce the impact of bare ground. WV3 also has very high resolution (0.31 m) for the panchromatic band which allows higher accuracy in spatial correction of field-measured location data. Therefore, WV3 may be suitable for mapping foliar DigN in the eucalypt trees used by koalas.

In this study, we identified the best spectral index from WV3 images to estimate and map foliar DigN in semi-arid eucalypt woodlands of the Mulgalands bioregion, Australia. We applied this mapping method to WV2 imagery of the same area collected contemporaneously with a previous koala tracking study [28,29], allowing us to test the usefulness of this method for understanding koala habitat suitability. This region supports a widely distributed, low-density koala population which is concentrated in riparian habitats dominated by eucalypt species [30]. The area is suitable but also challenging for testing spectral mapping methods because it has varied tree species and soil types.

We addressed three research questions: 1) is the WV3 data able to accurately estimate and map foliar DigN; 2) does the WV3 sensor provide better spectral indices to estimate foliar DigN when including SWIR bands; 3) does the mapped DigN associate with koala tree use?

## 2. Materials and Methods

# 2.1. Study Area

The study area was in the Mulgalands bioregion of semi-arid southwest Queensland, Australia. In summer, average temperature ranges are from 19–35 °C and in winter from 3–19 °C. Average annual rainfall is about 550 mm in the northeast and decreases to 292 mm in the southwest. Annual rainfall is highly variable and summer dominant. The major land use is cattle grazing.

Two landscapes (Landscape A and Landscape B) located ~200 km apart were studied (Figure 1). The main landforms are riparian strips dominated by *Eucalyptus camaldulensis, Eucalyptus populnea*-dominated floodplains and *Acacia aneura* (mulga) or *Acacia harpophylla*-dominated plains (Figure 2). The riparian areas and floodplains in Landscape B also have *Eucalyptus coolabah* and some *Eucalyptus melanophloia*. The four eucalypt species provide forage and shelter to koalas, while the acacia species provide shelter only. The primary food tree species is *E. camaldulensis* in the study area.



**Figure 1.** WorldView-3 satellite images in approximate true colour at Landscape A and Landscape B captured on 21 November 2015. The map of Queensland at the bottom-left corner shows the Mulgalands bioregion in grey and the locations of two landscapes as black dots.





**Figure 2.** Field colour photos of Landscape A showing a riparian habitat dominated by *Eucalyptus camaldulensis* (left) and a floodplain habitat dominated by *Eucalyptus populnea* (right) in October 2015.

## 2.2. Survey Design

There were two types of data collection: foliage sampling for leaf spectra and leaf DigN measurement, and acquisition of WV3 data for image spectra and spectral indices calculation.

In each landscape, two on-creek sites were chosen along creeks (ephemeral watercourse) then two corresponding off-creek sites were located between 200 m and 300 m from the creek (Figure 3). On-creek sites and off-creek sites had different tree species and were located in potential koala habitat. Ten trees from each on-creek site and six trees from each off-creek site were marked for foliage sampling. Off-creek sites had less trees selected because of its low tree density hence a lower number of trees available at the site scale (1000s m<sup>2</sup>). From these eight sites, 64 trees were selected in total, including 35 *E. camaldulensis*, 7 *E. coolabah*, 21 *E. populnea*, and 1 *E. melanophloia*.



**Figure 3.** Survey design of the study showing the position of two on-creek sites (black dots) and two off-creek sites (black dots) in a landscape. Corresponding off-creek sites were located between 200 m and 300 m from on-creek sites.

Ream [29] found that image acquisition in the Mulgalands faced different limitations in wet and dry seasons. Ground cover (i.e., grass and shrubs) thrives in wet seasons creating mixed pixels with reflectance from trees and ground vegetation, whereas in dry seasons, senescent ground cover, bare ground, and sparse tree canopies can create mixed pixels with stronger influence of soil [29]. To minimize the influence of grass, both field sampling and WV3 image acquisition were undertaken in October and November 2015, the late dry season. Capturing cloud-free satellite images was also more likely in the dry season. From October 2012 to November 2015, average annual rainfall was 342 mm in Landscape A and 317 mm in Landscape B. That was much lower than the long-term average annual rainfall (400–600 mm) and a severe rainfall deficit was reported for this period [31].

# 2.3. Field Sampling and Leaf Spectral Sampling

From 19–24 October 2015, the tree species and the location of each selected tree were recorded using a handheld GPS (Garmin eTrex®30) with a 3 m accuracy on average. To check and correct the tree GPS location when overlaying on the image, five distinct and widely dispersed locations (e.g., grid corners, fence corners, and road signs) in each landscape were selected as GPS control points for image geo-referencing. GPS coordinates and a description of each GPS control point were recorded by the same GPS device. Leaf samples were collected using a pruning pole from the lower canopy of all 64 selected trees between 7:00–10:30 a.m.

Approximately 50 g of leaves were collected from each tree and placed in paper bags to avoid sunlight for leaf spectral sampling within 30 mins. Fresh leaf spectra were acquired with an ASD FieldSpec<sup>®</sup> high-resolution spectrometer between 350 and 2500 nm with 1.1–1.4 nm bandwidth. A white reference panel was used to standardise and convert relative radiance to reflectance. A leaf clip and a plant probe with halogen bulb as light source were used to collect leaf surface reflectance. For each tree, 3 leaves were randomly selected, and 10 spectra were collected from each side. A total of 60 spectra were collected for each tree and the average spectra were calculated. These averaged ASD leaf spectra were later resampled to simulate WV3 spectra based on the spectral response of WV3 bands following the methods in Eitel, Long, Gessler, and Smith [17].

## 2.4. Foliar Chemical Analysis

After spectral sampling, leaf samples were frozen immediately, and transported frozen to the University of Queensland where they were freeze-dried and ground (ZM200 Retsch®). Near-infrared reflectance spectroscopy (NIRS) was used to assist in analysing foliar chemicals. The 64 leaf samples in this study were collected along with 719 additional leaf samples for a larger koala study carried out in southwest Queensland. Hence NIRS was done using the 783 leaf samples following Moore, Lawler, Wallis, Beale, and Foley [9].

N was quantified using a combustion method with Leco TruMac CN analyser (Leco Corporation, St. Joseph, MI, USA). DigN was quantified using a two-stage in vitro digestion with pepsin and cellulose, following the method of DeGabriel, Wallis, Moore, and Foley [14]. DigN concentration was presented as % dry matter (DM).

# 2.5. WV3 Image Acquisition and Pre-Processing

The WV3 multispectral images (DigitalGlobe, USA) used in this study comprised eight multispectral and eight SWIR bands (Table 1). SWIR bands were collected at 3.7 m spatial resolution by WV3 but were only made available at 7.5 m due to restrictions imposed by the US Government. One image was captured for each landscape at 10:40 a.m. AEST, 21 November 2015. Each image covered an area of 100 km<sup>2</sup> and cost AU\$109 per km<sup>2</sup>. The geo-referencing quality was assessed by overlaying the satellite images with the field-measured GPS control points in ArcMap (Version 10.4). Minor deviations were corrected to the nearest 0.31 m (the spatial resolution of WV3 panchromatic band). Satellite images were atmospherically corrected using Quick Atmospheric Correction in ENVI

(Version 5.2). The eight VIS-NIR multispectral bands were resampled to 7.5 m spatial resolution to calculate indices incorporating both VIS-NIR and SWIR bands.

Band Name	Wavalangth (nm)	Emotial Decolution		
Danu Name	wavelength (iiii)	Spatial Resolution		
MUL1: Coastal	400-450	1.24 m		
MUL2: Blue	450-510	1.24 m		
MUL3: Green	510-580	1.24 m		
MUL4: Yellow	585-625	1.24 m		
MUL5: Red	630-690	1.24 m		
MUL6: Red Edge	705–745	1.24 m		
MUL7: NIR1	770-895	1.24 m		
MUL8: NIR2	860-1040	1.24 m		
SWIR1	1195-1225	7.5 m		
SWIR2	1550-1590	7.5 m		
SWIR3	1640-1680	7.5 m		
SWIR4	1710-1750	7.5 m		
SWIR5	2145-2185	7.5 m		
SWIR6	2185-2225	7.5 m		
SWIR7	2235-2285	7.5 m		
SWIR8	2295-2365	7.5 m		

Table 1. Details of the sensor bands of WorldView-3 images acquired for this study

# 2.6. Collecting Image Spectra of Sampled Trees

Image spectra were extracted from the WV3 image pixel corresponding to the GPS position of each tree (Figure 4). Tree canopies were identified based on GPS position, field photos and the 0.31 m panchromatic band. In the 1.24 m WV3 images, trees with only epicormic growth were not used if their canopies were covered by or mixed with neighbouring canopies. For an identified tree canopy, a set of pixels were selected and the averaged spectra was used. In the 7.5 m WV3 images, most pixels covered more than one tree hence the trees covered by the same pixel were combined as one averaged tree.



**Figure 4.** WorldView-3 image displayed in RGB using bands Blue, Green, Red (**a**,**b**) and bands NIR1, Red and Blue (**c**,**d**) showing the same sample sites in Landscape B at 1.24 m (**a**,**c**) and 7.5 m (**b**,**d**) spatial resolutions. Yellow dots denote tree GPS locations.

The normalized difference vegetation index (NDVI) was calculated using Red and NIR1 bands for both field leaf spectra and image spectra [32]. The WV3 NDVI values of some trees were below 0.6, which was lower than the average NDVI of ASD leaf spectra. These trees were not used for this study as their WV3 spectra were likely to be mixed spectra with other features such as soil, tree trunks, or big stems rather than pure tree reflection.

As a result, out of the total 64 trees, 49 trees were used for the 1.24 m WV3 images (29 *E. camaldulensis*, 5 *E. coolabah*, 14 *E. populnea*, and 1 *E. melanophloia*) and 27 averaged trees were used for the 7.5 m WV3 images (19 *E. camaldulensis*, 2 *E. coolabah*, 5 *E. populnea*, and 1 *E. melanophloia*).

# 2.7. Calculating and Assessing Spectral Indices

Single bands, first derivative spectra and spectral indices were calculated from resampled ASD spectra, WV3 1.24 m spectra and WV3 7.5 m spectra, to compare the indices' performance with leaf-acquired hyperspectral spectra and satellite-derived multispectral data.

These spectral indices were selected from recent studies estimating plant N using remotely-sensed spectra (Table 2). Normalized difference indices (NDIs) from WorldView-2 (WV2) spectra were used to estimate grass N concentration [20]. The ratio of transformed chlorophyll absorption in reflectance index and optimized soil adjusted vegetation index (TCARI/OSAVI) was used to measure chlorophyll content which is related to nitrogen content [33]. The TCARI/ OSAVI was then modified by Herrmann, et al. [34] into TCARI<sub>1510</sub>/OSAVI<sub>1510</sub> because the 1510 nm band is directly related to N content. Four SWIR-based indices including TCARI<sub>1510</sub>/OSAVI<sub>1510</sub> from ASD field spectra showed better estimation for N content of potato fields [34]. Another ratio of modified chlorophyll absorption ratio index and the second modified triangular vegetation index (MCARI/MTVI2) showed better performance than TCARI/OSAVI in estimating wheat N content using multispectral data [17].

**Table 2.** Spectral indices used for estimation of foliar DigN in this study. Spectral bands in the formula are based on WorldView-3 (WV3) sensor bands.  $R_a$  and  $\lambda_a$  represents the reflectance and the average wavelength of a certain band.  $N_{1,24}$  is the number of an index from WV3 1.24m spectra.  $N_{7.5}$  is the number of an index from WV3 7.5m spectra or resampled ASD spectra.

Spectral Index	Formula	N <sub>1.24</sub>	N <sub>7.5</sub>	Reference
Single band	Ra	8	16	
First derivative	$D_{a-b} = (R_a - R_b) / (\lambda_a - \lambda_b)$	7	15	
Normalized difference indices (NDI)	$NDI_{a-b} = (R_a - R_b) / (R_a + R_b)$	28	120	[20]
Transformed chlorophyll absorption in reflectance index (TCARI)	TCARI = 3[(Red edge - Red) - 0.2(Red edge - Green) (Red edge / Red)]	1	1	[33]
Optimized soil adjusted vegetation index (OSAVI)	OSAVI = 1.16[(NIR1 - Red) / (NIR1 + Red + 0.16)]	1	1	[35]
Transformed chlorophyll absorption in reflectance index 1510 (TCARI <sub>1510</sub> )	TCARI <sub>1510</sub> = 3[(Red edge - SWIR2) - 0.2(Red edge -Green) (Red edge / SWIR2)]	0	1	Adjusted
Optimized soil adjusted vegetation index 1510 $(OSAVI_{1510})$	OSAVI <sub>1510</sub> = 1.16[(NIR1 - SWIR2) / (NIR1 + SWIR2 + 0.16)]	0	1	from [54]
Modified chlorophyll absorption in reflectance index (MCARI)	MCARI = [(Red edge - Red) - 0.2(Red edge - Green)] (Red edge / Red)	1	1	[36]
Modified triangle vegetation index 2 (MTVI2)	$MTVI2 = [1.8(NIR1 - Green) - 3.75(Red - Green)] / sqrt [(2NIR1 + 1)^2 - 6NIR1 + 5sqrt(Red) - 0.5]$	1	1	[37]
	TCARI / OSAVI	1	1	[17]
Combined index	MCARI / MTVI2	1	1	[17]
	TCARI <sub>1510</sub> / OSAVI <sub>1510</sub>	0	1	[34]

The correlation coefficients between the spectral indices and DigN concentrations were used to estimate the performance of indices. For the index with the best correlation coefficients, linear and second-order regression equations were used to determine the best estimation of DigN. The coefficient of determination ( $\mathbb{R}^2$ ), *p*-value and root mean square error ( $\mathbb{R}MSE$ ) were used to compare the performance of the spectral indices. All data analysis was performed in Excel.

#### 2.8. Mapping DigN with Selected Index

Because the average NDVI of ASD leaf spectra was 0.6, a mask of NDVI > 0.6 was applied to the WV3 images to subset the areas of vegetation. Using the region of interest (ROI) tool in ENVI, a plot of average spectra of eucalypt trees, non-eucalypt trees (*A. aneura* and *Geijera parviflora* mixed) and dense grass patches (mixed species) was created to find ways to mask out grass and non-eucalypt trees. The accuracy of the vegetation masks was visually assessed in ArcMap. A set of 100 random pixels were created within the final masked image. Then they were checked visually using the 0.31 m panchromatic image and the 1.24 m VNIR image to confirm whether they cover trees. The equation of the selected index for DigN was applied to the masked image in ENVI. The foliar DigN concentrations of eucalypt tree areas were mapped and displayed in colour.

#### 2.9. Application of DigN Mapping Method to WV2 Image

The WV2 image was collected at 10:58AM AEST, 16 October 2012, which was a similar time and season as the WV3 images. It covered a landscape (270 km<sup>2</sup> at 2 m spatial resolution) adjacent to the south of Landscape A which has the same landform and vegetation. The WV2 image was atmospherically corrected using Quick Atmospheric Correction in ENVI (Version 5.2). The mapping method described above was applied to this image. Vegetation masks were applied to the image to subset areas of trees. The accuracy of the vegetation masks was visually assessed in ArcMap. A set of 100 random pixels were created within the final masked image. Then they were checked visually using the 0.46 m panchromatic image and the 2 m VNIR image to confirm whether they cover trees. The equation of the selected index for DigN was applied to the masked image using band math in ENVI to produce a map of foliar DigN concentrations.

# 2.10. Associating Mapped DigN from WV2 Image with Koala Tree Use Data

Koala tracking and data collection was described by Davies, Gramotnev, Seabrook, Bradley, Baxter, Rhodes, Lunney, and McAlpine [28]. The locations of five koalas in this landscape were recorded as GPS points from August 2010 to November 2011. All GPS points were overlaid in ArcMap. Because there were no field-measured GPS control points, we reduced potential errors by observing and checking the GPS points against koalas' habits (i.e., spend more time in trees than on the ground, do not swim). If a group of neighbouring points were on ground/water surface and have a tree to the same distance and direction nearby on the WV2 image, they were manually spatially adjusted to tree canopies Only nocturnal locations (5:00 p.m. to 06:00 a.m.) were used because koalas mainly feed during this time and their tree use during the night was more likely to be influenced by foliar DigN [38]. These points were then converted to a raster layer (sum of koala tree use) with the same pixel size as the DigN map which corresponds with the WV2 image (2 m spatial resolution). As the accuracy of most GPS units was within a 5 m radius, an averaging convolution filter for a  $11 \times 11$ pixels moving window was applied to the layer in ENVI to calculate the density of koala tree use. The same convolution process was applied to the DigN map. DigN and density of koala tree use data were queried and extracted in TerrSet (Version 18.3), then analysed using quantile regression in XLStat (www.xlstat.com) at different quantiles (0.200–0.999). Quantile regression can estimate potential causal relationships between variables at certain quantiles when not all predictive factors are measured for variable ecological responses [39]. Hence quantile regression is suitable to this case because koala tree use can be affected by DigN and other factors, such as plant secondary metabolites and tree size [8].

# 3. Results

#### 3.1. Laboratory Measures of Foliar Chemistry

Table 3 summarises the measured DigN and N concentrations for all samples (n = 64), samples for 1.24 m WV3 images (n = 49) and 7.5 m WV3 images (n = 27). The Pearson correlation coefficient between DigN and N concentration was r = 0.46 (P < 0.001).

**Table 3.** DigN and N concentrations (% dry matter) of all samples, sample subsets for WorldView-3 images at 1.24 m and 7.5 m spatial resolutions

Response Variable	Samples Size	Mean	Min	Max	SD	CV (%)	
N	64	1.65	1.09	2.20	0.22	13.12	
N 1.24 m	49	1.65	1.09	2.20	0.22	13.62	
N 7.5 m	27	1.66	1.20	2.20	0.23	13.56	
DigN	64	1.12	0.35	1.86	0.33	29.12	
DigN 1.24 m	49	1.14	0.35	1.86	0.34	29.86	
DigN 7.5 m	27	1.20	0.35	1.86	0.35	29.46	

The best index to estimate foliar DigN concentration was NDI<sub>Coastal-NIR1</sub> using WV3 7.5 m spectra. Its regression relationship with DigN concentration ( $R^2 = 0.70$ , P < 0.001, RMSE = 0.191% DM) is shown in Figure 5.



**Figure 5.** Regression of the relationship between digestible nitrogen (DigN; % dry matter) and NDI of MUL1 (Coastal) and MUL7 (NIR1) bands. NDIs were calculated from WorldView-3 images of Landscape A and B at 7.5 m spatial resolution.

The performance of indices of DigN concentration in WV3 spectra was different from that in resampled ASD spectra (Table 4). The first derivative including SWIR bands ( $D_{SWIR6-7}$  and  $D_{SWIR7-8}$ ) were better than  $D_{Red \ edge-NIR1}$  in resampled ASD spectra but this was reversed in WV3 spectra. Similarly, single band SWIR3 performed well with resampled ASD spectra while coastal was better with WV3 7.5 m spectra. The NDIs were poor at estimating DigN from resampled ASD spectra but performed well with WV3 7.5 m data. The NDIs using only VNIR bands performed better than those containing the SWIR band in WV3 7.5 m spectra.

<b>Table 4.</b> Coefficient of determination $(R^2)$ , p-value and RMSE (% dry matrix)	atter) for various spectral
indices used as regression estimators of foliar digestible nitrogen concentra	tion (DigN; % dry matter)
in eucalypts. Spectral indices were calculated from three datasets including	g resampled ASD spectra,
WorldView-3 (WV3) 1.24 m spectra and WV3 7.5 m spectra. $R^2$ over 0.40 are	e shown in bold text.

Spectral Index		Resampled ASD		WV3 1.24 m			WV3 7.5 m			
		$R^2$	_p	RMSE	$\mathbb{R}^2$	p	RMSE	$R^2$	p	RMSE
	MCARI	0.00	0.674	0.324	0.03	0.242	0.332	0.18	0.026	0.312
	MTVI2	0.02	0.254	0.324	0.13	0.011	0.315	0.42	< 0.001	0.264
D	MCARI/MTVI 2	0.02	0.214	0.320	0.00	0.692	0.336	0.04	0.327	0.339
Previous	TCARI	0.12	0.005	0.304	0.14	0.009	0.313	0.08	0.147	0.331
Indices	TCARI <sub>1510</sub>	0.05	0.069	0.316	-	-	-	0.01	0.612	0.344
	TCARI/OSAVI	0.07	0.031	0.312	0.20	0.001	0.302	0.23	0.012	0.304
	TCARI <sub>1510</sub> /OSAVI <sub>1510</sub>	0.02	0.304	0.322	-	-	-	0.01	0.694	0.345
C' 1	Coastal	0.14	0.002	0.300	0.34	< 0.001	0.274	0.45	< 0.001	0.257
Single	Blue	0.11	0.007	0.305	0.29	< 0.001	0.284	0.36	< 0.001	0.277
band	SWIR3	0.48	< 0.001	0.235	-	-	-	0.06	0.222	0.335
	D <sub>Red edge-NIR1</sub>	0.04	0.136	0.319	0.05	0.137	0.329	0.36	< 0.001	0.276
First D	D <sub>SWIR6-7</sub>	0.52	< 0.001	0.225	-	-	-	0.15	0.049	0.319
	D <sub>SWIR7-8</sub>	0.51	< 0.001	0.226	-	-	-	0.00	0.937	0.346
	Coastal-NIR1	0.13	0.004	0.303	0.40	< 0.001	0.262	0.70	< 0.001	0.191
	Coastal-NIR2	0.13	0.004	0.303	0.45	< 0.001	0.250	0.66	< 0.001	0.202
	Coastal-SWIR1	0.12	0.005	0.305	-	-	-	0.32	0.002	0.285
	Coastal-SWIR3	0.08	0.027	0.312	-	-	-	0.12	0.074	0.324
NDI	Blue-NIR1	0.09	0.018	0.301	0.32	< 0.001	0.277	0.60	< 0.001	0.218
NDI	Blue-NIR2	0.09	0.018	0.310	0.36	< 0.001	0.269	0.55	< 0.001	0.232
	Blue-SWIR1	0.08	0.025	0.311	-	-	-	0.27	0.006	0.296
	Green-Yellow	0.03	0.143	0.319	0.19	0.002	0.303	0.34	0.001	0.281
	Red Edge-NIR1	0.02	0.294	0.321	0.19	0.002	0.302	0.46	< 0.001	0.255
	Red Edge-NIR2	0.02	0.298	0.321	0.34	< 0.001	0.274	0.34	0.001	0.281

#### 3.3. Mapping DigN with Selected Indices

Based on the average spectra of eucalypt trees, non-eucalypt trees and grass (Figure 6), vegetation areas with  $R_{NIR1} > 0.455$  were masked out to exclude grass, whereas the remaining tree areas with ( $R_{NIR1}-R_{Red}$ ) < 0.16 were masked out to exclude the known non-eucalypt trees. Among the 100 random pixels from the masked image, 99 pixels in Landscape A, and 98 pixels in Landscape B were visually confirmed that they cover trees.



**Figure 6.** Average reflectance of *Eucalyptus camaldulensis* and *E. populnea* (solid lines), non-eucalypt tree and grass (dashed lines) at 16 spectral bands from WorldView-3 images at 7.5 m spatial resolution. The reflectance of *E. coolabah* and *E. melanophloia* were similar to these eucalypt species and not shown.

The regression equations from NDI<sub>Coastal-NIR1</sub> were applied to the pixels of trees in the WV3 7.5 m images to map foliar DigN concentration (Figure 7) across Landscape A and Landscape B.

In both landscapes, the foliar DigN along creek lines stood out on the maps where the tree density was relatively higher (Figure 7). Overall foliar DigN concentration was lower in Landscape B. The trees on the floodplain (around 300 m away from the creek) in Landscape A showed high DigN concentrations but the species of these tree were uncertain because of a lack of field reference observation. GPS collar data showed koalas in the Mulgalands spent more than 80% of their time on average within riverine habitat [28] and koalas tend to be found within 300 m of creek lines [40]. Therefore, the trees 300 m away from creek were of less importance in this study and were not observed in the field. The cluster of higher DigN in the zoomed-in area of Landscape B was a house garden with frequent irrigation, which contained mature *E. camaldulensis* (grown from local seedlings), non-eucalypt trees, bushes, and grass.



**Figure 7.** Maps of foliar digestible nitrogen (DigN) at Landscape A and B in colour. Black boxes in full maps indicate locations of zoomed-in areas. Red circles in zoomed-in areas indicate locations of sample sites. Features in non-tree areas (soil, water and grass) are white.

#### 3.4. Relationship between Mapped DigN from WV2 Image with Koala Tree Use

There were 1,428 GPS records of the occurrence of koalas at night time. After averaging convolution, there were 66,926 tree pixels (2 m) with the frequency of koala tree use which was between 1 to 43 (mean = 2.58).

For DigN mapping, a mask of NDVI>0.57 was applied to the WV2 images to subset the areas of vegetation. Based on average spectra of grass and different tree species (Figure S1), vegetation areas with  $R_{NIR1} > 0.59$  were masked out to exclude grass, whereas the remaining tree areas with  $(R_{NIR1}-R_{Red}) < 0.28$  were masked out to exclude the known non-eucalypt trees. All 100 random pixels from the masked image in Landscape C were visually confirmed that they cover trees. Foliar DigN concentration of each pixel (2 m) was estimated using the WV2 spectra and the NDI<sub>Coastal-NIR1</sub> spectral index. The estimated DigN concentration of these 66,926 tree pixels was between 0 to 1.11% DM (mean = 0.21% DM).

Quantile regression showed improvement in fit being at higher quantile (Figure S2). The highest coefficients of determination between the frequency of koala tree use and the estimated DigN concentration from WV2 spectra was in the 0.999 quantile (Figure 8,  $R^2 = 0.63$ , P < 0.001).



**Figure 8.** Correlation between koala tree use frequency (as mapped in the field in 2010 and 2011) and log10 transformed estimated DigN concentration (% dry matter) from the WV2 image of 2012 with quantile regression fitted line for the 0.999 quantile (dark grey) and 95% confidence bounds (light grey).

#### 4. Discussion

This study assessed the potential of using WV3 multispectral data to map foliar DigN concentrations of eucalypt trees from two landscapes in the semi-arid Mulgalands bioregion, Australia. The spectral index NDI<sub>Coastal-NIR1</sub> was found to satisfactorily estimate foliar DigN concentrations. The DigN mapping method using this index were tested by linking previous koala tree use frequency with mapped DigN from a WV2 image of an independent landscape in the study area. Results indicated

greater koala tree use for trees with higher estimated DigN, which is consistent with previous studies [8,41]. Therefore, it is possible to estimate and map the variation in eucalypt foliar DigN over large areas using WV3- or WV2-derived multispectral data and these estimates are meaningful for the evaluation of koala habitat suitability.

#### 4.1. Spectral Indices to Estimate and Map Foliar DigN

This is the first attempt we know of to estimate foliar DigN using satellite-derived spectra. Previously, Youngentob, Renzullo, Held, Jia, Lindenmayer, and Foley [19] successfully estimated foliar DigN using airborne hyperspectral spectra. DigN is an integrated foliar nutritional measurement influenced by concentrations of nitrogen, tannins, cellulose, and lignin [14]. Consequently, the relationship between spectral indices and foliar DigN concentration would be indirect. The best index from WV3 spectra, NDI<sub>Coastal-Blue</sub>, showed good estimation capability. Regarding the first and second research questions (whether WV3 can be used for DigN mapping and whether SWIR bands can improve the mapping), this study suggested that the WV3 sensor can provide spectral indices to estimate and map foliar DigN in eucalypt across a landscape but the best index does not include SWIR bands.

It was interesting that the ASD and WV3 spectra both returned useful, but different spectral indices, from distinct regions of the spectrum. In the resampled ASD spectra, the best-performing spectral indices ( $R^2 > 0.4$ ) were obtained from the SWIR region but the best spectral indices from WV3 spectra were from the VNIR region (Coastal, Blue, NIR1, and NIR2) instead. Because the best-performing spectral indices from ASD and WV3 spectra used bands of absorption features of DigN associated chemicals, we suggest that such differences may be related to issues of (1) sampling scale, with the leaf scale of ASD spectra versus the canopy scale of WV3 spectra, (2) atmospheric effects which apply to WV3 spectra but not to ASD spectra, and 3) the potential signal integrity and signal to noise ratio differences between the two different sensors [42]. In the SWIR region, SWIR3, 6, and 8 contain nitrogen absorption features at 1645, 2180, 2240, 2300, and 2350 nm [19,43]. SWIR3 also contains tannin and lignin absorption features (1658, 1675, and 1668 nm), whereas SWIR7 has absorption features of cellulose (2280 nm) and lignin (2272 nm) which influence dry matter digestibility [43,44]. In the VNIR region, apart from the absorption features of chlorophyll at 430 and 460 nm or nitrogen at 808, 868, 910, and 1020 nm [43,45,46], spectral indices using the four VNIR bands also contain absorption features of tannins (803 nm in NIR1, 948 and 993 nm in NIR2) and lignin (478 nm in Blue) [47,48].

#### 4.2. Better Performance of WV3 7.5 m Spectra than 1.24 m Spectra

When estimating foliar DigN, most spectral indices performed better (higher  $R^2$ , lower RMSE) with WV3 7.5 m spectra than with WV3 1.24 m spectra. This may be caused by the difficulty of identifying pure pixels for a single tree canopy from WV3 1.24 m images. The canopies of dense *E. camaldulensis* along creeks overlap and one pixel may contain the leaf reflection of more than one tree. Therefore, defining the tree spectra of one tree from WV3 1.24 m images based on tree trunk GPS risks including the reflection of its neighbours. In addition, the accuracy of the handheld GPS is about 3 m which is less than the spatial resolution of WV3 1.24 m spectra. The pixel selected based on a GPS coordinate may not precisely represent the pure spectra of a specific tree, giving a lower correlation with foliar DigN. In contrast, at 7.5 m spatial resolution, when one pixel covered more than one tree GPS, its spectra were defined as the combined tree spectra of the trees within this pixel. The combined tree spectra were correlated with the combined foliar DigN of these trees.

This does not mean WV3 1.24 m data is less useful. Tree sampling in this study was designed primarily to select koala food trees for foliar chemical analysis, rather than to be compared with satellite imagery. A better sampling design for a similar remote sensing study would have sampled more trees and chosen isolated trees with denser and bigger canopies that were identifiable in the image [19] and used higher-accuracy differential GPS location data. These measures may improve spectral indices

performance in WV3 1.24 m data. This requirement for isolated trees constrains the development and validation of spectral indices, but need not constrain subsequent estimation of habitat suitability, where pixels capturing more than one tree return an estimate of mean nutritional quality which is nonetheless useful for habitat mapping purposes.

# 4.3. Mapped DigN to Evaluate Koala Habitat Suitability

Our results show that trees with higher foliar DigN concentration can have any level of koala use, but low DigN trees only have low koala use. Although koalas spend more time feeding in trees with higher DigN concentrations, koala tree use is also influenced by other factors such as foliar plant secondary metabolites, tree species, and tree size [8]. Low koala densities in the study region also mean that many suitable trees may never be encountered by koalas. That is why the estimated DigN concentration only explained part of the variation of koala tree use frequency. For example, some trees with very high estimated foliar DigN concentration but very low koala tree use frequency were located on the flood plain where koalas spent less than 20% of time during the tracking period [28]. Although there were unmeasured factors, the mapped DigN concentration from WV2 proved to be a limiting factor for koala tree use frequency. In responding to the third research question, the mapped DigN is associated with koala tree use and this DigN mapping method is useful in assessing koala habitat suitability.

In addition, testing the relationship between mapped DigN and koala tree used worked as a validation of the DigN mapping method. The validation is independent for using an image of a different sensor, landscape and time, however it is indirect by using koala tree use instead of field measured foliar DigN. Therefore, we were not able to measure the accuracy of applying the DigN mapping method on WV2.

#### 5. Conclusions

We have reported the first attempt to correlate satellite-derived multispectral data with foliar DigN, which is an important measure of food quality for koalas. The foliar chemical variations of koala food tree species indicate food availability and quality more precisely than the presence or absence of food tree species [8]. Hence, the DigN mapping method can improve the mapping of high quality habitat at the landscape scale for effective koala conservation and habitat management. Because this DigN mapping method was developed across two landscapes and four eucalypt species, then tested for a new landscape with similar eucalypt species, we concluded that it can be applied to new open eucalypt woodlands with similar tree species which are, for example, widely distributed in the Murray–Darling basin in Australia. This technique, once tested to assess its reliability with different tree species, could be used to validate the accuracy of coarse koala habitat mapping based on vegetation communities and improve koala habitat classification by adding finer foliar nutritional information.

**Supplementary Materials:** The following are available online at http://www.mdpi.com/2072-4292/11/2/215/s1, Figure S1: Average reflectance of eucalypt tree (solid line), non-eucalypt tree and grass (dashed lines) at eight spectral bands from the WorldView-2 image at 2 m spatial resolution, Figure S2: Coefficients of determination for quantile regression for 30 values of quantiles (from 0.970 to 0.999), where y is the frequency of koala tree use and x is estimated DigN concentration from WorldView-2 spectra.

**Author Contributions:** Conceptualization, H.W., N.L., L.S., and C.M.; Methodology, H.W. and N.L.; Validation, H.W. and N.L.; Formal analysis, H.W. and N.L.; Investigation, H.W., N.L., and B.M.; Resources, L.S. and B.M.; Writing—original draft preparation, H.W.; Writing—review and editing, N.L., L.S., B.M., and C.M.; Visualization, H.W.; Supervision, C.M.; Project Administration, H.W.; Funding Acquisition, H.W., N.L., and C.M.

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