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₁ 1 Introduction

- 2 This document provides detailed field and analytical methods for the study of tree canopy struc-
- ture in southern African woodlands. The study aimed to understand the effects of tree species di-
- 4 versity and stand structure on tree canopy structure and grass biomass. Chapter XXX contains
- 5 the same methods in brief.

₆ 2 Sampling

- ⁷ Fieldwork was conducted at two sites, the first in Bicuar National Park, southwest Angola (S15.1°,
- 8 E14.8°), and the second in and around Marure Forest Reserve, southeast Tanzania (S9.0°, E39.0°).
- 9 Fieldwork was conducted during the peak growth period of each site, in order to capture the high-
- est foliage volume in the canopy and grass volume in the understorey.

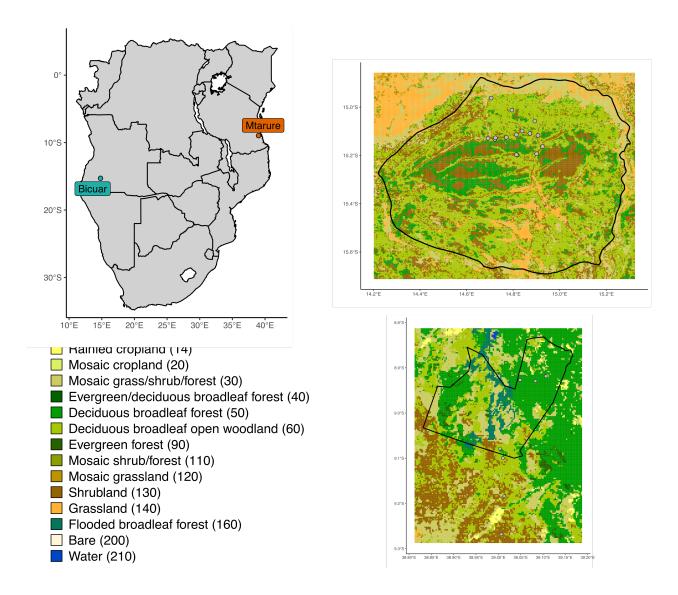


Figure 1: Location of study sites within southern Africa (a), and of 1 ha plots within each site. The black polygons denote the boundaries of protected areas which encompass the majority of study sites, Bicuar National Park in Angola (b), and Mtarure Forest Reserve in Tanzania (c). Each site map is coloured according to the GlobCover global land cover classification.

Site	MAT	MAP	Temp. range	CWD				
	$(^{\circ}C)$	$(mm y^{-1})$	$(^{\circ}C)$	$(mm y^{-1})$				
Bicuar	20.8 (0.70)	825.9 (52.01)	24.5 (0.90)	-844.8 (44.29)				
Mtarure	25.7 (0.24)	958.4 (25.19)	$12.0 \ (0.33)$	-739.6 (8.06)				

Table 1: Climatic data for each site, extracted from WorldClim at 2.5 minute resolution. Values are the mean and standard deviation (in brackets) of all pixels intersecting each protected area. MAT = Mean Annual Temperature. MAP = Mean Annual Precipitation. Temp. range = Temperature range, calculated as the mean of annual difference between highest temperature of hottest month and lowest temperature of coldest month. CWD = Climatic Water Deficit, calculated as the sum of the difference between monthly rainfall and monthly evapotranspiration when the difference is negative, sensu Chave et al. (2014).

- 11 At each site, a number of 1 ha permanent plots were sampled. In Angola, 15 plots were sampled,
- while in Tanzania, only seven were sampled, following the curtailment of fieldwork due to COVID-
- 19 travel restrictions. Permanent plots were located in areas of homogeneous vegetation type,
- away from roads and undisturbed by humans. Plots were established following the SEOSAW pro-
- tocol (version 3.0, SEOSAW 2020). Plots were located quasi-randomly by first locating areas from
- satellite imagery expected to comprise savanna woodland vegetation. At each site, plots were de-
- 17 liberately located along a gradient of stem density.
- Each permanent plot was further subdivided into nine 10 m diameter circular subplots arranged in
- a regular grid, with a buffer from the plot edge (Figure 2).

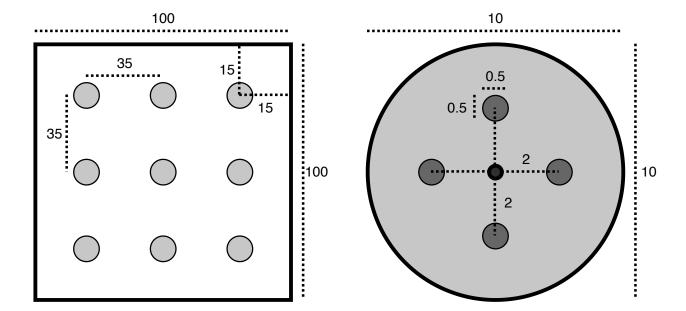


Figure 2: The layout of 10 m diameter subplots within each 1 ha square plot. Each subplot is situated inside a 15 m buffer from the plot edge, with 35 m between subplot centres. Subplots are arranged in a 3x3 grid. Disc-pasture measurements and biomass samples are located in cardinal directions 2 m from the centre of the subplot. All distances are in metres.

3 Field measurements

21 3.1 Trees

- 22 For each subplot, we measured all woody stems >5 cm stem diameter with canopy material inside
- the subplot. For each stem we recorded:
- Tree identity
- Stem diameter (diameter at breast height 1.3 m)
- 26 For each tree, which may be composed of multiple stems joined at the base, we recorded:
- Species
- Height to top of canopy
- Canopy area, ellipse from two perpendicular measurements (Figure 3)
- Distance from subplot centre
- Compass direction from subplot centre

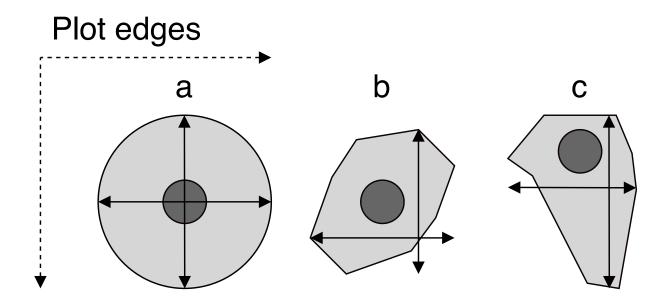


Figure 3: Examples of tree crowns as viewed from above to demonstrate how crown extent measurements are located. Darker grey circles show the main stem while pale grey polygons show the maximum extent of the crown. Extent measurements are taken parallel to the plot edges. a) shows a perfectly circular tree crown, b) and c) show irregular tree crowns, demonstrating that maximum crown extent in a given orientation be offset from the stem.

32 3.2 Grass biomass

- 33 Grass volume and biomass within each subplot was estimated from four sample points located 2
- m from the subplot centre in cardinal directions (Figure 2). At each point, a disc-pasture meter
- measurement was taken with a 45.8 cm radius disc weighing exactly 1.5 kg (Bransby and Tain-
- ton, 1977). Small woody stems were removed from disc-pasture sample points before the disc-
- pasture measurement was taken. The location of the sample point was moved if the designated
- point intersected with coarse woody debris, rocks, shrubs, or standing trees. Within each 1 ha
- ³⁹ plot, biomass harvesting was conducted at nine randomly allocated disc-pasture sample points.
- 40 Tree leaf litter was removed from biomass samples. Biomass harvesting involved clipping all grass
- material within the 45.8 cm radius to ground level, taking care not to include roots. Grass sam-
- ples from Angola were dried until the mass remained constant $(\pm 5 \text{ g})$ for >48 hours, then weighed
- 43 to ascertain the grass biomass. Grass samples from Tanzania could not be processed due to cur-
- tailment of fieldwork due to COVID-19 travel restrictions.

5 3.3 Hemispherical photography

- 46 At the centre of each subplot a single photograph was taken with a Nikon D750 full-frame DSLR
- camera, with a circular fisheye lens. The lens had an equisolid (equal area) projection, which avoids

48 image distortion. The projection function is given by:

$$R = 2f\sin\left(\theta/2\right) \tag{1}$$

- Where R is the radial position of a point on the image on the sensor, f is the focal length of the
- lens, and θ is the angle in radians of the desired angular radius of the cropped image.
- 51 The photo was taken facing directly to zenith, with the top of the camera facing magnetic north,
- at a height of 1.3 m or above understorey vegetation, whichever was higher. Table 2 shows de-
- scribes the camera settings for each hemispherical photo.

Table 2: Description of camera settings used for each hemispherical photo. Note that the values of shutter speed and ISO are deliberately variable within sensible thresholds to adapt to light conditions.

-							
Setting	Value						
Camera model	Nikon D750						
Lens model	Sigma 8 mm f/3.5 EX DG Circular Fisheye						
Pixel pitch	$5.95\mu\mathrm{m}$						
Sensor resolution	24.3 MP						
Shutter speed	$> 1/60 \mathrm{s}$						
Aperture	5-7						
ISO	100-200						
Exposure compensation	-0.7 (Brusa and Bunker, 2014)						
Focus	∞ (Hu and Zhu, 2009; Frazer et al., 2001)						
Image size	Large Fine JPEG - circular image 4016x4016 px						
Orientation	Landscape						

- Photos were captured under uniform light conditions as much as possible, either under overcast
- 55 skies or early in the day before direct sunlight could be seen on the photo.
- ImageJ (Fiji version 2.1.0/1.53c) was used to binarize hemispherical photos (), to separate plant
- 57 material from sky. We first split each image into red, green and blue channels. We used the Huang
- algorithm to automatically threshold images, using the blue channel only, under the assump-
- tion that plant material reflects little blue light, while the sky reflects much more (). Images were
- saved as PNG at the original pixel resolution.

61 3.4 Stand structure

- From the stem measurements we calculated a number of indices to characterise whole-plot and
- 63 subplot stand structure.
- We calculated the spatial mingling index (M) according to von Gadow and Hui (2002) at the plot
- 65 level. The spatial mingling index is a spatially explicit estimate of the degree to which species are
- 66 spatially mixed within a plot:

$$M = \sum_{i=1}^{N} \left(\frac{S_i}{S} \frac{1}{k} \sum_{j=1}^{k} v_j \right)$$
 (2)

with
$$v_j = \begin{cases} 0, & \text{neighbour } j \text{ same species as reference } i \\ 1, & \text{otherwise} \end{cases}$$
 (3)

(4)

- where k is the number of nearest neighbours considered for each reference tree, S_i is the number
- of species found among the k nearest neighbours of tree i, S is the total number of species in the
- plot, and N is the total number of trees in the plot. In our case we used the conventional value of
- 70 k = 4.
- 71 We also calculated the Winkelmass W according to von Gadow and Hui (2002) at the plot level.
- 72 The Winkelmass estimates the degree of spatial uniformity in stem spatial distribution:

$$W = \sum_{i=1}^{N} \frac{1}{k} \sum_{j=1}^{k} v_j \tag{5}$$

with
$$v_j = \begin{cases} 0, & \alpha_j \le \alpha_0 \\ 1, & \text{otherwise} \end{cases}$$
 (6)

(7)

where α_j is the angle between consecutive neighbours and α_0 is the principal angle, where $\alpha_0 = 360/k$.

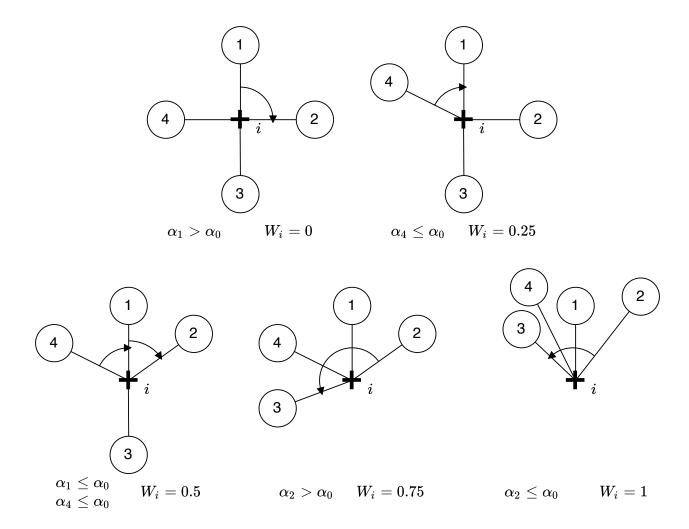


Figure 4: Possible values of W_i at a sample point i, denoted by a cross. Neighbours are represented as circles numbered sequentially from 1 to 4, where k = 4. The angles of arrows in each example are given below, along with the Winkelmass for that example.

To estimate tree spatial structure in subplots we used an adapted version of the Iterative Hegyi index (H_i) (Hegyi, 1974). Our adapted formula allows the index to be based on a point rather than a focal tree, transforming it from a tree-centric competition index to a point-centric crowding index:

$$H_i = \log \sum_{j=1}^{n} \left(\frac{1}{L_{ij}} D_j\right) \tag{8}$$

where D_j is the stem diameter of neighbour tree j and L_j is the distance of the neighbour from the subplot centre.

81 4 Terrestrial LIDAR

- Within each subplot, a variable number of scans were recorded using a Leica HDS6100 phase-
- shift terrestrial laser scanner (TLS). The number and position of scans within a subplot was de-
- termined by the arrangement and density of canopy material in the subplot. Scan positions were
- arranged to minimise shadows within the canopy, and to maximise canopy penetration. Number of
- scans per subplot ranged between one and five in both Angola and Tanzania (Table 3).
- Five Leica 6" planar tilt and turn targets were used at each subplot to align scans. To allow reg-
- 88 istration of scans among subplots, the location of each target was registered using a Leica VIVA
- 689 GS10 GNSS unit, set up in post-processed kinematic (PPK) configuration with a base-station lo-
- ocated ~100 m from the edge of each 1 ha plot. The location of each target was measured for at
- 91 least 4 minutes. Further, we used the TrimbleRTX GNSS post-processing service to precisely lo-
- cate each target (Chen et al., 2011). When registering scans we discarded targets with location
- 93 accuracy of >3 cm.

Table 3: Description of scan settings used for each scan.

Sotting	Value
Setting	value
Scanner model	Leica HDS6100
Wavelength	$650\text{-}690~\mathrm{nm}$
Spot size at exit	3 mm
Beam divergence	0.22 mrad
Range	79 m @90%; 50 m @18% albedo
Azimuth range	0-360°
Zenith range	0-155°
Increments	0.018°
Point spacing over 25 m $$	7.9 mm
Pixels per line	20000
Lines	10000
Compressed file size	~800 MB
Duration of scan	6 minutes 44 seconds
Lines Compressed file size	10000 ~800 MB

94 4.1 Registration

- 95 Scan registration for each subplot was conducted in Leica Cyclone (version 9.1). Targets from
- each scan were aligned using Cyclone's automatic target acquisition.
- After registration, scan scenes were exported from Cyclone as PTX files, one per subplot.

98 4.2 Voxelisation

- 99 PTX files were converted to compressed LAZ files using PDAL (). The exact code used to ex-
- tract and apply the PTX rotation matrix to each point in the PTX file can be found IN THIS
- 101 APPENDIX HERE.
- LAZ files were voxelised to different voxel sizes depending on the application of the data. For
- grass biomass estimation, we used 2 cm³ cubic voxels, while for subplot height profile estimation
- we used 5 cm³ voxels, and for whole plot canopy rugosity we used 10 cm³ voxels. WHY THO

105 4.3 Noise reduction

Outlier detection and noise reduction was conducted in PDAL using the filters.outlier filter, using the "statistical method" (sensu Rusu et al. 2008), with k = 8 (mean number of neighbours),

and m = 1.96 (standard deviation threshold, approximating a 95% confidence interval):

$$\overline{\mu} = \frac{1}{N} \sum_{i=1}^{N} \mu_i \tag{9}$$

$$\sigma = \sqrt{\frac{1}{N-1} \sum_{i=1}^{N} (\mu_i - \overline{\mu})^2}$$
 (10)

$$t = \mu + m\sigma \tag{11}$$

$$outlier_i = \begin{cases} true, & \text{if } \mu_i >= t \\ false, & \text{otherwise} \end{cases}$$
 (12)

where N is the number of points in the scene, $\overline{\mu}$ is the mean distance to nearest neighbour points, and σ is the standard deviation of these distances. t is the threshold distance used to define an outlier.

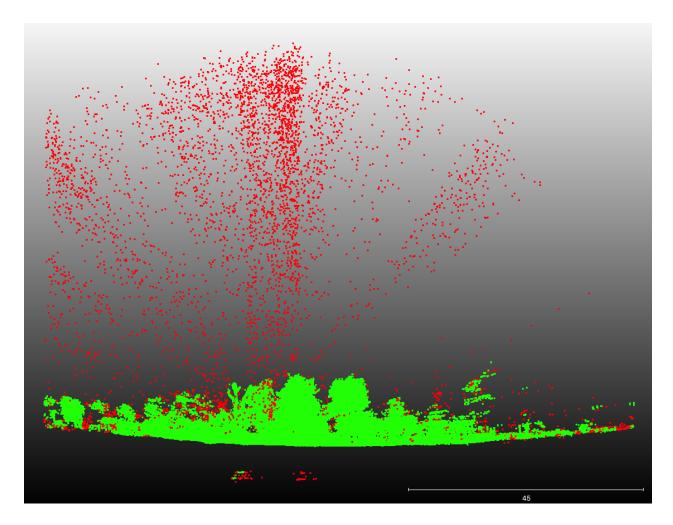


Figure 5: 2 m deep cross section of subplot showing the efficacy of the noise reduction and voxelisation process. Red points are points excluded by this cleaning process, while green points are used in further analysis.

112 4.4 LiDAR analysis

4.4.1 Foliage density profiles

- To estimate subplot foliage density profiles, first the point cloud was cropped to a 10 m diame-
- ter cylinder of infinite height. Then the filters.pmf (Progressive Morphological Filter PMF)
- PDAL function was used to identify ground points (sensu Zhang et al. 2003). The filters.hag_nn
- (Nearest Neighbour) PDAL function was used to generate height above ground of each point within
- the cylinder. Points below ground level were then discarded. Height profile points were exported
- to a XYZ file then imported into R for further processing.
- We excluded points above the 99.9th percentile of height, under the assumption that these often
- constituted noise that had not been adequately removed by PDAL.
- In R, within each 5 cm width vertical layer, we calculated the gap fraction as the proportion of

unfilled 5 cm³ voxels. We filtered the point cloud data to the tree canopy, excluding grass. We identified the breakpoint between the grass understorey and the tree canopy as the first local minima above 1.3 m from the ground.

We extracted statistics from the foliage density profile for use in statistical analysis. We first smoothed the density profile using a loess model with a span of 0.1. We then calculated the number of local maxima and minima along the profile. We defined local maxima and minima as points where the gap fraction of the surrounding 50 cm of 5 cm bins was lower or higher, respectively.

We calculated the effective number of layers (ENL), using the true-numbers equivalent of the Shannon diversity index (sensu (Ehbrecht et al., 2016)). We also calculated the conventional Shannon diversity index on the gap fraction of 50 cm bins:

$$H' = -\sum_{i=1}^{N} p_i \ln p_i \tag{13}$$

Where N is the number of 50 cm bins in the height profile, and p_i is the proportion of filled voxels in layer i (gap fraction).

135 We calculated the area under the curve of foliage density using trapezoid estimation.

We extracted the height of the maximum foliage density peak, and calculated the difference between the highest and lowest local maxima. We also extracted the maximum canopy height within the subplot.

139 We calculated the coefficient of variation of the point cloud height distribution.

To describe the uniformity of the foliage density distribution we used Ripley's L function, which is more commonly used in describing spatial variation across a 2 dimensional surface. Ripley's L is an adjustment to Ripley's K, defined as:

$$\widehat{K}(t) = \lambda^{-1} \sum_{i \neq j} \frac{I(d_{ij} < t)}{n} \tag{14}$$

$$\widehat{L}(t) = \left(\frac{\widehat{K}(t)}{\pi}\right)^{1/2} \tag{15}$$

We also used the standard error of a linear model of foliage density and height as a simple single number method of describing the uniformity of foliage density. Under a completely even distribution of foliage material through the canopy, the standard error should be zero, while clumping causes deviations from this uniform distribution and increases the standard error.

147 4.4.2 Canopy gap fraction

Due to terrestrial LiDAR measurement locations being spread over the subplot to avoid occlusion 148 of canopy material, we simulated a scan position at the centre of the subplot using the point cloud data from all scans per subplot. Similar to the processing chain for the foliage density profiles, 150 PDAL was used to crop the point cloud to a 20 m cylinder around the subplot centre, then used 151 filters.hag_nn to classify ground points and recalculate height above ground. We cropped the 152 point cloud to points above 1.3 m, with a 50 cm exclusion sphere around the scan position at 1.3 153 m above the ground. The point cloud was converted to a POV-Ray object, where each point was 154 transformed to a 1 cm³ cube. POV-Ray was then used to produce a ray-traced image. As with 155 the hemispherical photos, we used a fisheye lens with an equisolid projection and a view angle of 156 180°, located at the subplot centre, at the same height as the hemispherical photo, with the top 157 of the camera facing magnetic north and the camera facing straight up. Each cube was set as a 158 non-reflective object, and the sky had an equal gamma of 1.0. POV-Ray produced an image of 159 4016x4016 px, identical to the cropped circular dimensions of the images produced by the hemi-160 spherical photos. 161 Simple canopy gap fraction as seen from the ground was measured using two methods: 1) hemi-162 spherical photography and 2) terrestrial LiDAR. Hemiphot () was used to estimate gap fraction 163 from both the hemispherical photos and the TLS POV-Ray simulation. Hemiphot calculates canopy 164 gap fractions in 90 evenly sized concentric rings. To obtain the total gap fraction of an image: 165

$$G_{\text{tot}} = \sum_{\alpha=0.5}^{\alpha=89.5} (G_{\alpha} A_{\alpha} / A_{\text{tot}})$$
(16)

Where G_{α} is the fraction of unfilled pixels in ring α , A_{α} is the sky area of the ring segment, and A_{tot} is the total sky area of the hemisphere.

We compared gap fraction estimates from both the TLS and hemispherical photo using a linear mixed model which accounted for variation among plots and between the two sites. While plots in Mtarure had a marginally steeper slope, this difference was not significant. We found that hemispherical photography almost exclusively over-estimated gap fraction, except in the most open subplots. Additionally, at lower gap fractions (greater canopy cover) the over-estimation of gap fraction by hemispherical photography was larger (Figure 6).

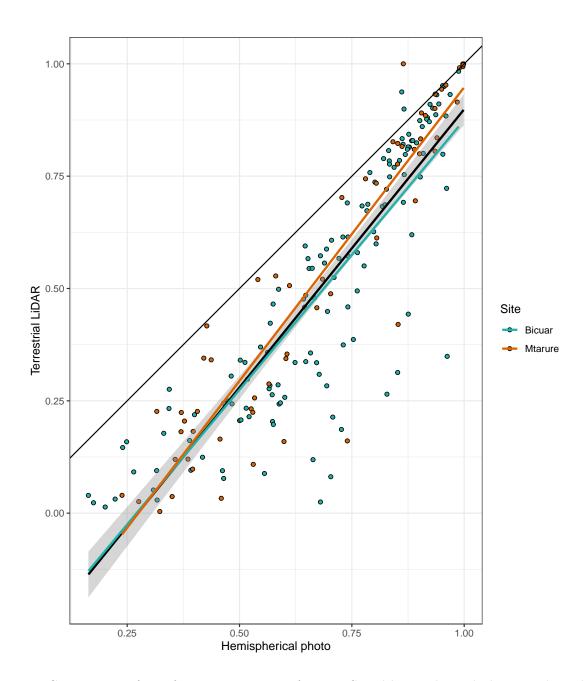


Figure 6: Comparison of gap fraction estimation from TLS and hemispherical photography. The black line of best fit is a linear model of all points ± 1 standard error, while the coloured lines are site specific linear models.

174 4.4.3 grass biomass estimation

- An allometric model was developed to estimate grass biomass at every disc-pasture sample point using the grass biomass sample masses. This model was only developed for Angola where grass biomass samples were weighed. The model consisted of a linear mixed effects regression testing the relationship between disc-pasture height (independent) and grass biomass (dependent), with a random slope term for each 1 ha plot.
- grass volume was measured from TLS point cloud data following the methodology of. First the

point cloud was cropped to points below 2 m. The point cloud was then aggregated to cubic voxels of 2 cm³. Within each vertical 2 cm² column, the mean height of points was calculated, then the volume below the mean was assumed to be entirely filled with grass material.

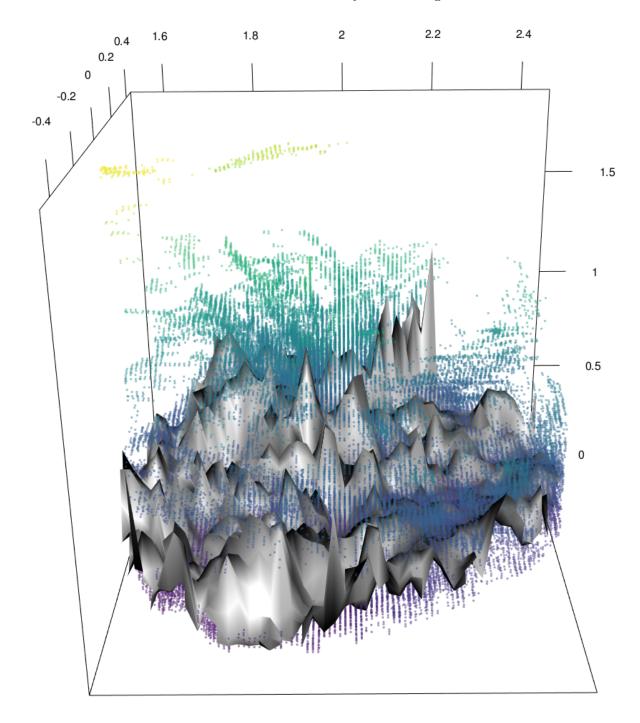


Figure 7: Point cloud with mean heights for each $2~\mathrm{cm}^2$ column labelled and the estimated grass volume below.

84 4.4.4 Canopy rugosity

The canopy rugosity of each 1 ha plot was estimated. All scans from each plot were merged to a single point cloud, and noise reduction was performed as described above and the cloud was vox-

elised to 10 cm³ cubic voxels. The point cloud was cropped to the plot boundaries, which were 187 located with dGPS similar to the LiDAR targets. 188 A canopy height model was produced to describe the upper canopy surface. The 99th percentile of 189 height in each 10 cm² vertical column was extracted. The maximum height was not used as this 190 occasionally constituted a severe outlier which skewed further canopy height model smoothing. We used the pit-filling algorithm described in Khosravipour et al. (2014) to smooth the canopy height 192 profile by removing gaps within trees caused by incomplete penetration of the LiDAR beam. 193 From the canopy height profile we extracted a number of statistics for use in statistical modelling. 194 We calculated the mean and coefficient of variation of canopy height across the plot (canopy ru-195 gosity), following (Parker and Russ, 2004). We calculated the Topographic Ruggedness Index 196 (TRI) as the mean of absolute differences between the heights of each column and the height of 197 its eight surrounding cells (Wilson et al., 2007). From this we estimated the plot level mean TRI 198 and coefficient of variation. 199 We also calculated a second measure of canopy rugosity (R_c) following Hardiman et al. (2011),

$$R_c = \sigma(\sigma G_z)_x \tag{17}$$

Where G_z is the vertical height axis z, x is the horizontal axis, and σ is the standard deviation.

using all point cloud data rather than just the top surface:

₂₀₃ 5 Statistical analysis

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All linear mixed effects models werre conducted using the {lmer} package in R version 4.0.2 (R Core Team, 2020).

₂₀₆ 5.1 Foliage density profiles

We conducted a number of linear mixed effects models to assess the effects of tree diversity and 207 stand structure on various aspects of canopy structure measured at the 10 m subplot scale. Lin-208 ear mixed effects models were used to account for the non-independence of samples caused by 209 the nested sampling structure of subplots within plots, and plots within sites. For each subplot 210 canopy structure measure, we created a linear mixed effects model with fixed effects of subplot 211 species richness, and tree spatial structure using the adapted Hegyi index (H_i) and the coefficient of variation of stem diameter. We compared the standardized effect sizes of each fixed effect to 213 understand the relative effect of species richness and spatial structure on canopy structure. We 214 compared models with all combinations of fixed effects to understand which combination of fixed 215

effects best explained variation in each subplot canopy structure measure. We also compared models to a null model including only random effects of plot and site to evaluate whether this 'best' model explained real variation in canopy structure.

5.2 Grass biomass

To estimate the correlation between grass volume estimated by TLS and grass biomass estimated 220 from the allometry of DPM height and grass biomass samples, we conducted a linear mixed ef-221 fects model of grass biomass vs. grass volume, with nested random slope terms for each 1 ha plot nested within site. 223 We conducted a linear mixed effects model to assess the effects of canopy structure on grass volume, with random slope terms for each 1 ha plot nested within site. We began with a maximal 225 model which included fixed effects of subplot tree species richness, stem density, TLS gap fraction, 226 layer diversity, height of maximum foliage density, standard deviation of the foliage density pro-227 file, and our simple measure of foliage density uniformity. We re-fitted the model with all possible combinations of fixed and random effects and compared AIC, BIC, and log-likelihood to determine 229 which combination of explanatory variables best accounted for variation in grass volume. Once 230 this 'best model' had been identified we extracted standardized effect sizes for each fixed effect to 231 compare their relative contribution to the model. We also compared random effects for each fixed effect to understand how the relationship differed between the two sites.

234 5.3 Canopy rugosity

To understand the effect of species composition and stand structure on whole-plot canopy rugosity, we conducted a linear mixed effects model with fixed effects of tree species shannon diversity index, stem density, spatial mingling index and winkelmass, with random intercept terms for
each site. We extracted slopes for each fixed effect to compare their effect sizes and compared our
model with a null model which consisted only of the random effect of site and the fixed effect of
stem density.

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