

University of Tartu
Faculty of Science and Technology
Tartu Observatory
Department of Remote Sensing

Bachelor's thesis in environmental technology (12 ECT)

**Leaf inclination angle distribution in deciduous broadleaf and evergreen broadleaf
Eucalyptus forests under elevated atmospheric CO₂**

Anni Maria Pearson

**Supervisors: PhD Jan Pisek (TO)
PhD Margit Aun (TO)**

Tartu 2024

Leaf inclination angle distribution in deciduous broadleaf and evergreen broadleaf Eucalyptus forests under elevated atmospheric CO₂

The aim of this thesis was to compare two methodologies, leveled digital photography and a new software, AngleCam, in terms of their performance in accurately measuring and predicting leaf inclination angles. Additional aims were to see if the spherical distribution of leaf angles was appropriate for the two sites and if the elevated CO₂ environments affect the inclination angles of the present species. The sites are BIFoR FACE (U.K.) and EucFACE (Australia). The results found that the two methodologies agree at predicting possible vectors of change, while the absolute values and distribution types differ significantly. However, AngleCam requires some more training with specific species and distribution types. Furthermore, for this study, the assumption of a spherical distribution is overturned. Changes in leaf inclination under elevated CO₂ are noticeable but generally within the limits of their natural variation.

Keywords: leaf inclination, FACE, elevated CO₂, AngleCam, leveled digital photography approach

CERCS code: T181-- remote sensing, B270 – plant ecology

Lehe kaldenurga jaotus lehtpuu- ja igihaljas lehtpuumetsas kõrgendatud atmosfäärses CO₂ tingimustel

Antud töö eesmärk oli võrrelda kaht meetodit, tasapinnalist digitaalfotograafiat ja uut tarkvara AngleCam, ning nende võimekust mõõta täpselt lehe kaldenurki. Lisa eesmärkideks oli leida, kas sfääriline jaotus lehtede puhul on paikapidav antud uurimiskohtades leiduvate liikide puhul ning kas kõrgendatud CO₂ antud kohtades mõjutab lehe kaldenurka. Uurimiskohad on BIFoR FACE (Suurbritannia) ja EucFACE (Austraalia). Tulemusena leiti, et mõlemad meetodid suudavad ennustada muutuste vektoreid, kuid erinevad väärtuste ja jaotuste osas suurelt. AngleCami puhul on vaja veel programmi treenimist spetsiifiliste liikide ja jaotustega. Sfääriline jaotus antud töös kasutatud liikide puhul ei pea paika. Samuti leiti, et kõrgendatud CO₂ mõjutab lehe kaldenurka, kuid muutus jääb loodusliku variatsiooni piiridesse.

Märksõnad: lehe kaldenurk, FACE, kõrgendatud CO₂, AngleCam, tasapinnaline digitaalfotograafia lähenemine

CERCS kood: T181 – kaugseire, B270 – taimeökoloogia

LIST OF CONTENTS

| | |
|-------------------------------------|----|
| LIST OF ABBREVIATIONS | 4 |
| 1. INTRODUCTION | 5 |
| 2. MATERIAL AND METHODS | 7 |
| 2.1 SITE DESCRIPTION | 7 |
| 2.2 FIELD SAMPLING | 9 |
| 2.3 METHODOLOGY AND MATERIALS | 10 |
| 7. RESULTS | 15 |
| 8. DISCUSSION | 25 |
| 9. CONCLUSION..... | 29 |
| KOKKUVÕTE | 31 |
| ACKNOWLEDGEMENTS | 33 |
| REFERENCES | 34 |

LIST OF ABBREVIATIONS

| | |
|-----------------------|-------------------------------------|
| AC | AngleCam |
| ALIA | Average leaf inclination angle |
| CO₂ | Carbon Dioxide |
| FACE | Free Air Carbon Dioxide Enrichment |
| IJ | ImageJ |
| LIAD | Leaf inclination angle distribution |

1. INTRODUCTION

Trees play an essential role in our environment by capturing significant amounts of carbon dioxide (CO₂) from the atmosphere through photosynthesis. Given the ongoing climate changes, this ability is of profound interest. Trees are estimated to capture around 130 Gt of CO₂ from the atmosphere every year (Bala, 2014).

Several previous studies have highlighted the importance of leaf inclination angle distribution (LIAD) in regards to interactions between the environment and trees (e.g., Chianucci *et al.*, 2018; Pisek *et al.*, 2011, 2021). However, as mentioned in the studies before, there is a lack of a reliable methodology that is cheap and does not consume extensive amounts of time. In previous studies, Ryu *et al.* (2010) and Pisek *et al.* (2011) introduced leveled digital photography as practical and a cheap, reliable source of gathering images for such measurements. Recently, Kattenborn *et al.* (2022) have developed a machine learning approach called AngleCam that should be able to detect leaves in images automatically and provide the LIAD parameters automatically. AngleCam has been trained in specific environments in Germany (Kattenborn *et al.*, 2022), and thus, there was an interest if this program is also applicable in different climates and locations.

LIAD is a vital plant structural trait that determines radiation interception, biomass production, rainfall interception, and evapotranspiration (Oguchi *et al.*, 2018; Pisek *et al.*, 2021). LIAD has remained one of the most poorly constrained parameters and a significant source of uncertainty in ecological models due to difficulty in quantification (Chianucci *et al.*, 2018). LIAD is often assumed to be spherical (Pisek *et al.*, 2021). Only a little attention has been paid to possible changes in leaf orientation and inclination distribution with elevated CO₂ (Pisek *et al.*, 2023). Still, its effect on plant growth could alter the proportions of sunlit and shaded leaf areas and feedback on carbohydrates available for further development.

To study the impact of elevated CO₂ in the atmosphere and its relation to the environment, The Free Air Carbon Dioxide Enrichment (FACE) projects are providing valuable insight into understanding the response that forests might have to such conditions. FACE projects are a few from around the world. They follow and observe the effects that elevated CO₂ environments have on flora. Specific FACE experiments are being set up in different conditions and locations to test the effects of carbon dioxide, giving scientists a better understanding of the potential

and probable consequences of climate warming. It is conceivable that the elevated CO₂ also affects the leaf inclination angle distributions.

The lack of choice in the Free-Air Carbon Dioxide Enrichment facilities remarkably determined the selection of test sites. As to our knowledge, there is only one FACE project currently running in the Northern Hemisphere, that being BIFoR FACE in the U.K., and the other available data was from that of the Eucalyptus FACE (EucFACE) project in Australia. With the two chosen FACE projects, it is possible to explore if the trees react in similar trends to each other even though the species, as well as soils and other environmental factors such as sunlight and precipitation, are different.

Research Objectives

This thesis compares two methodologies for measuring leaf angle distribution in two Free-air CO₂ (FACE) experiments in mature forests (one in Australia and the other in the U.K.) under ambient and elevated CO₂ conditions.

Specifically, this thesis will address the following questions:

1. Do the two methods – the established leveled digital photography and the new, machine-learning-based AngleCam provide comparable results?
2. Is spherical leaf angle distribution suitable for present species at the two study sites?
3. How does leaf inclination angle distribution change under varying CO₂ concentrations?

2. MATERIAL AND METHODS

2.1 SITE DESCRIPTION

This thesis used two test sites: Birmingham Institute of Forest Research Free-Air Carbon Dioxide Enrichment (BIFoR FACE) and Eucalyptus Free-Air Carbon Dioxide Enrichment (EucFACE). The following sections provide detailed descriptions of each.

BIFoR site description

The site is in woodland in Staffordshire, England (52.801°N, 2.301°W), United Kingdom. It is a facility established to study the effects of elevated CO₂ levels on forest ecosystems.

The BIFoR FACE site is composed of mature woodland and consists of six circular plots referred to as 'arrays' (Fig.1). Each array, approximately 30 meters in diameter, serves as a designated research area within the BIFoR FACE facility (Hart *et al.*, 2020). The BIFoR FACE project started in 2017 and is currently the only such active project in the Northern Hemisphere.

Arrays 1, 4, and 6 are enriched arrays; these receive an extra 150 ppm (parts per million) of CO₂, while arrays 2, 3, and 5 are considered ambient and receive no extra CO₂ during the experiment. (BIFoR FACE, n.d.). The main species of trees are pedunculate oaks (*Quercus robur*) up to a height of 25 meters. The site also has some understory forest consisting of sycamore maple (*Acer pseudoplatanus*) and common hazel (*Corylus avellana*) (Hart *et al.*, 2020).



Figure 1. Aerial view of the BIFoR Free-Air Carbon Enrichment (FACE) site associated with Birmingham Institute of Forest Research. Photo: BIFoR FACE website, (BIFoR FACE, n.d.).

EucFACE site description

The second dataset was sourced from the Eucalyptus Free-Air CO₂ Enrichment (EucFACE) project facility at Western Sydney University. The site is located in Southern Australia, on the Cumberland Plain near Sydney (33°37'S, 150°44'E) on an old alluvial floodplain (Gimeno *et al.*, 2016). The predominant tree species is blue gum (*Eucalyptus tereticornis* Sm.) (Pisek *et al.*, 2021).

The site has been active since 2012 (Duursma *et al.*, 2016). It is composed of six circular plots located on the premise, each about 30 meters in diameter and 25 meters tall, with accessible towers to the top of the canopy (Fig. 2). These six plots are referred to as 'rings' for the EucFACE project.

Among the six rings designated as experimental units within the EucFACE project, rings 2, 3, and 6 are categorised as 'enriched rings' due to the added 150 ppm of CO₂. Conversely, rings 1, 4, and 5 are classified as 'ambient rings', maintaining natural CO₂ levels without supplementation (Pisek *et al.*, 2021).



Figure 2. View from a ring at EucFACE woodland, facing the rest of the rings. Photo: EucFACE website, (Gallery – EucFACE Experiment, n.d.).

2.2 FIELD SAMPLING

The site data have been divided into two sections. The EucFACE data was previously collected and used by Pisek *et al.* (2021). This thesis further analyses the data with a new approach – the AngleCam method by Katttenborn *et al.* (2022). BIFoR data are a new collection of photos that has been gathered for this work. In the following section, data collection procedures at each site are explained in detail.

BIFoR FACE site sampling

At the BIFoR site, samples were taken using a leveled digital photography approach described by Ryu *et al.* (2010). This method entailed capturing leveled photos of the canopy within each designated array at specific height intervals of 5-, 10-, 15-, 20-, and 25-meters.

The leveled photos were taken by operators Mr Peter Miles and Dr Kris Hart during four days, 8.08.-11.08.2023. The images were taken in a small timeframe to maximise the optimal weather conditions: very low to moderate wind, sunny and no rain. Photographs were taken using a Google Pixel 3XL Android camera phone and a Samsung Galaxy S9 Android, model number SM-G950F.

Operators climbed up each array tower structure and took pictures at the previously mentioned height levels; each level was marked on the posts. Usually, around 20-30 photos were taken at each height, including an all-around sample of the tree canopy. The images were separated and analysed by corresponding height and array, amounting to 30 sets of images (5 height intervals for each of 6 arrays).

EucFACE site sampling

Similarly, the same measurement procedure was used at the BIFoR site. The leveled photos were taken from each ring from the walkup tower in the centre of each ring. Here, no markings were made, nor were photos distinguished between heights, and they were allotted to the individual rings only.

Photos were taken using a Sony Xperia Z5 Compact phone with an F2.0 lens (Pisek *et al.*, 2021) on 14.02.2019 with little to no wind present.

Within the context of the EucFACE project, the rings were marked as elevated, meaning the CO₂ levels at those rings had been enriched with a surplus of 150 ppm CO₂, or ambient, where no extra CO₂ had been added.

2.3 METHODOLOGY AND MATERIALS

Theory

De Wit (1965) classical distribution types are used for classification of leaf angles. Six proposed primary distribution types are assigned to leaves based on their inclination angles (de Wit, 1965):

1. Planophile – leaves are predominantly horizontally inclined in the canopy
2. Plagiophile – canopies are dominated by inclined leaves away from the horizontal position
3. Uniform – equal proportion of leaf inclination angles for any angle
4. Spherical – same relative frequency of leaf inclination angles to form a spherical surface
5. Erectophile – leaves oriented predominantly in a vertical direction
6. Extremophile – leaves are oriented towards extremes (horizontal, vertical position) within the canopy.

In the following section, equations 1-5 show how the Beta distribution parameters μ and ν are calculated based on the inclination angles measured. The Beta distribution is calculated using Equation (5), that assigns the values to one of the six de Wit (1965) distribution categories as described above.

The angles measured are converted from degrees to radians marked as θ_L . It is then used to get $t=2*\theta_L/\pi$. From there equation (1) is used as follows:

$$f(t) = \frac{1}{B(\mu,\nu)} (1-t)^{\mu-1} t^{\nu-1} \quad (1)$$

For the Beta distribution $B(\mu,\nu)$ (Goel & Strebel, 1984), the parameters μ and ν are found using the equations (2) and (3), where the maximum standard deviation is σ_0^2 that consists of a mean of t ; σ_t^2 is the variance of t (Wang *et al.*, 2007):

$$\mu = (1 - \bar{t}) \left(\frac{\sigma_0^2}{\sigma_t^2} - 1 \right) \quad (2)$$

$$\nu = \bar{t} \left(\frac{\sigma_0^2}{\sigma_t^2} - 1 \right) \quad (3)$$

The parameters are then used in the integral equation to calculate the Beta distribution (Equation (4)) which includes the Gamma function Γ .

$$B(\mu, \nu) = \int_0^1 (1-x)^{\mu-1} x^{\nu-1} dx = \frac{\Gamma(\mu)\Gamma(\nu)}{\Gamma(\mu+\nu)} \quad (4)$$

As the last step, to make the distributions clearer and easier to understand, they are calculated to assign a classical de Wit (1965) distribution type using Equation (5).

$$\chi_{L=} \min \int_0^{\pi/2} |f(\theta_L) - f_{de\ Wit}(\theta_L)| d\theta_L \quad (5)$$

Figure 3 gives a visual understanding of how each of the classical distribution types are presented.

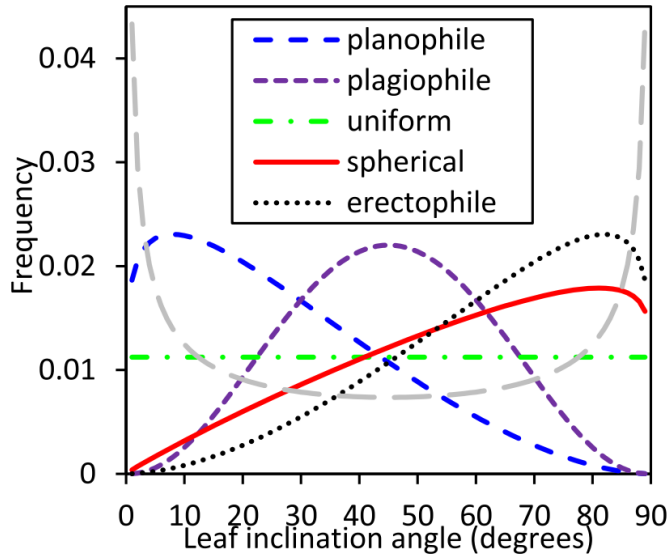


Figure 3. Beta distribution for the six theoretical leaf inclination angle distributions by de Wit (1965). (adopted from Pisek *et al.*, 2022)

ImageJ measurements

An open-source image processing tool, ImageJ (<http://imagej.nih.gov/ij/>, last accessed 20.04.2024), was used to measure and collect the data from the photos. Each usable image (not blurred or skewed) and the leaves perpendicular to the viewing point were measured with ImageJ. This is illustrated in Figure 4, an example of ImageJ being used on a BIFoR FACE array at the 15-meter height level. The selected leaf is shown perpendicular to the viewer and marked at both ends with magenta marking. Measurements were conducted separately for each height level at the BIFoR site, with approximately five measurements performed per photo. In total, 3491 angles were measured for BIFoR FACE arrays. Similarly, measurements were conducted for the EucFACE rings, with a total of 2535 angles measured during the study by Pisek *et al.* (2021).

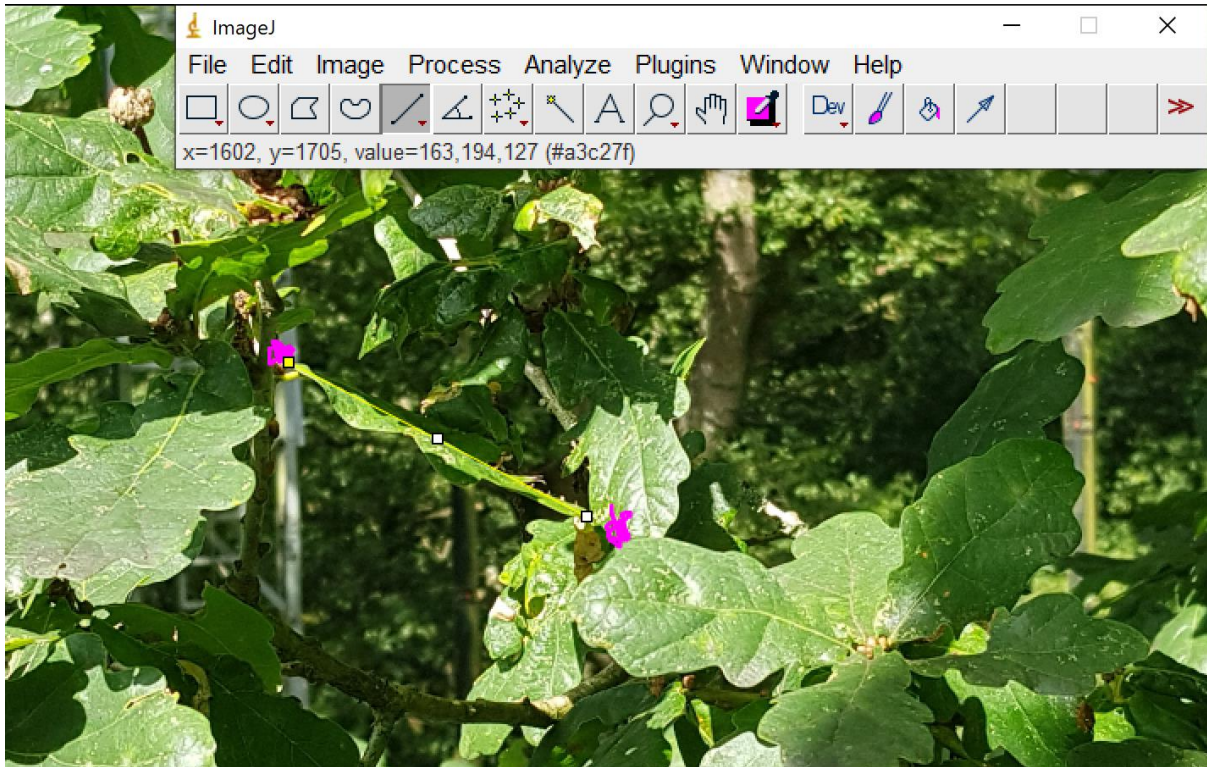


Figure 4. ImageJ open processing tool used on BIFoR FACE array two 15-meter height level. The selected leaf is perpendicular to the viewer and marked at both ends with magenta markings.

AngleCam Analysis and Validation

The AngleCam analysis involved feeding the photos of each site into the AngleCam program (tejakattenborn/AngleCAM: AngleCam; vers. 2022-04-07; zenodo.org; last accessed 20.04.2024) developed by Kattenborn *et al.* (2022). AngleCam uses a Convolutional Neural Network (CNN) in machine learning to analyse images and extract relevant data regarding LIAD. The main aim of this part of the research was to validate the methodology and compare the results with that of the ImageJ data.

In this study, the photos captured at each site were processed through AngleCam, which scans the images and identifies specific features, such as leaf edges. Before the analysis, the CNN within AngleCam was trained with labelled images as described by Kattenborn *et al.* (2022).

During the analysis, the leaf inclination distribution type, mean leaf inclination angle, beta distribution parameters μ and ν (see 2.3 Methods and Materials, Theory), and the standard deviation was determined (Kattenborn *et al.*, 2022).

Kattenborn *et al.* (2022) trained the program with 2770 images from the test site in Germany with the following species: European ash (*Fraxinus excelsior* L.), English oak (*Quercus robur* L.), Sycamore maple (*Acer pseudoplatanus* L.), European hornbeam (*Carpinus betulus* L.) and small-leaved lime (*Tilia cordata* Mill). It was initially tested with 60 different species of herbs, shrubs and deciduous trees (Kattenborn *et al.*, 2022).

7. RESULTS

Comparison of leaf inclination data from the BIFoR site using ImageJ and AngleCam

Figure 5 presents the data for BIFoR FACE site to show the differences between the two methodologies. While measurements with ImageJ estimate lower values (degrees) than AngleCam for both ambient and elevated arrays, AngleCam gives higher (steeper) angles as shown in the Figure 5. In Figure 5 the x-axis represents the arrays (1, 4, 6 are elevated; 2, 3, 5 are ambient), and the y-axis is for average leaf inclination angle (ALIA) in degrees.

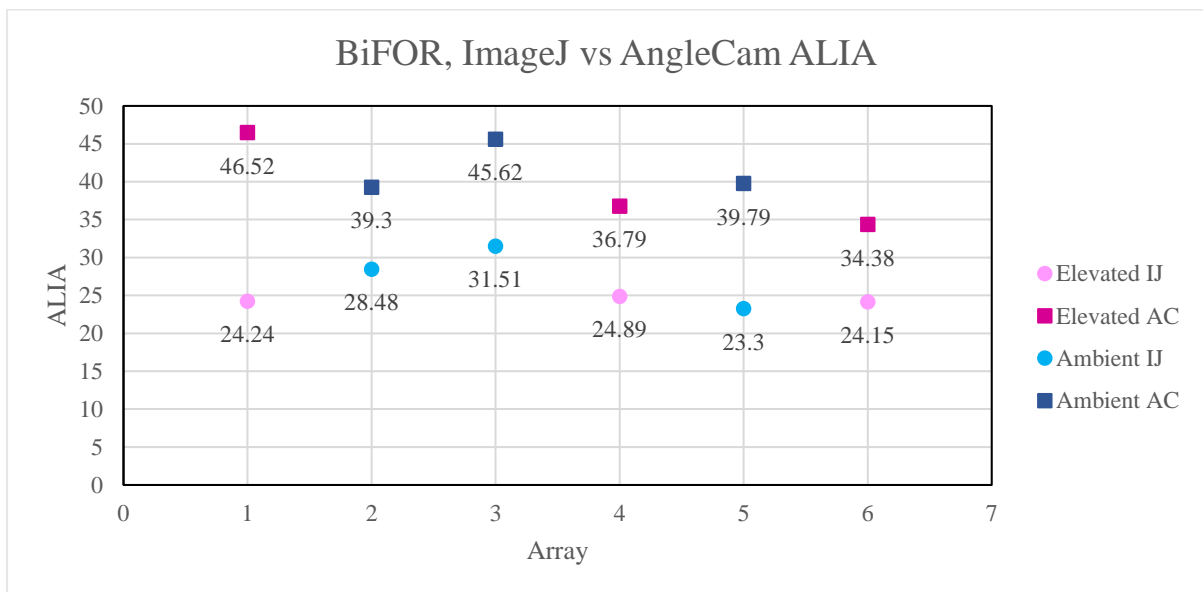


Figure 5. BIFoR site comparison of ImageJ and AngleCam data for both elevated and ambient arrays. Elevated arrays are marked with pink hues and ambient with blue hues. Squares represent the data measured by AngleCam (AC) and circles the data measured with ImageJ (IJ).

Below are two tables comparing leaf inclination data collected using ImageJ and AngleCam at the BIFoR site in more detail. Table 1 represents data from ImageJ, while Table 2 presents data from AngleCam.

Table 1 lists the six arrays and combined arrays, one where the data was used from the elevated arrays and one for when the ambient array data was used. For each array, the number of measurements, the mean leaf angle, the standard deviation, Beta parameters μ and ν , and the de Wit leaf angle distribution type are presented.

For arrays 1-6, between 529 and 615 measurements were done for each array. Mean inclination angles for the arrays were between 23.3 and 31.51 degrees, with standard deviations ranging from 16.24 to 21.27. The values for the parameters μ and ν were respectively between 2-3.66 and 0.93-1.34. All data unanimously indicates a planophile distribution type.

On separate lines are also the elevated arrays and the ambient arrays. For the elevated arrays (1, 4, 6), the total count of measurements done was 1810, the mean inclination angle of those measurements was 24.43 degrees, the standard deviation amounting to 18.12, μ and ν parameters being 2.83 and 1.05 accordingly, and the distribution type was planophile. For the ambient arrays (2, 3, 5), the count of measurements 1681 was done with the mean angle of 27.67 degrees, 20.03 standard deviation and the parameters μ and ν being 2.28 and 1.01 correspondingly, with the distribution of leaf angle being planophile once again.

Table 1. BIFoR site dataset of mean leaf inclination angles, standard deviation (s.d.), parameters μ and ν , and de Wit leaf distribution type as measured with ImageJ, including count of measurements done.

| Dataset | Count | Mean | s.d. | μ | ν | Type |
|------------------------------|-------|-------|-------|-------|-------|------------|
| Array 1 | 581 | 24.24 | 18.74 | 2.59 | 0.95 | planophile |
| Array 2 | 577 | 28.48 | 20.42 | 2.19 | 1.01 | planophile |
| Array 3 | 529 | 31.51 | 21.27 | 2 | 1.08 | planophile |
| Array 4 | 614 | 24.89 | 19.25 | 2.44 | 0.93 | planophile |
| Array 5 | 575 | 23.3 | 17.51 | 3.02 | 1.05 | planophile |
| Array 6 | 615 | 24.15 | 16.28 | 3.66 | 1.34 | planophile |
| Array 1+4+6 (elevated) | 1810 | 24.43 | 18.12 | 2.83 | 1.05 | planophile |

| | | | | | | |
|-----------------------------|------|-------|-------|------|------|------------|
| Array 2+3+5 (ambient) | 1681 | 27.67 | 20.03 | 2.28 | 1.01 | planophile |
|-----------------------------|------|-------|-------|------|------|------------|

In Table 2, the same photo collection was used as an input for AngleCam, and the results found for arrays 1-6 were somewhat different from Table 1. Compared to ImageJ results (Tab. 1), the mean inclination angles found were notably steeper, ranging between 34.38 and 46.52 degrees. The standard deviation for these was from 21.8 to 22.93 degrees De Wit distribution type for arrays 1-5 were mostly uniform and different from ImageJ results; only array 6 was planophile and agreed in de Wit classification type with ImageJ result.

On a separate line, both elevated and ambient arrays were brought out with the elevated (arrays 1, 4, 6) arrays having a mean inclination angle of 39.2 degrees, standard deviation of 22.21, parameters μ and ν 1.71 and 1.32 correspondingly and the distribution type being uniform. The ambient arrays (2, 3, 5) have a mean angle of 41.62 degrees, a standard deviation of 22.28 degrees, parameters μ and ν being 1.64 and 1.41, respectively, and the uniform distribution type.

Table 2. BIFoR site dataset of mean leaf inclination angles, standard deviation (s.d.), parameters μ and ν , and de Wit leaf distribution type as measured by AngleCam.

| Dataset | Mean | s.d. | μ | ν | Type |
|---------|-------|-------|-------|-------|------------|
| Array 1 | 46.52 | 22.5 | 1.45 | 1.55 | uniform |
| Array 2 | 39.3 | 22.04 | 1.75 | 1.36 | uniform |
| Array 3 | 45.62 | 21.8 | 1.61 | 1.65 | uniform |
| Array 4 | 36.79 | 22 | 1.8 | 1.24 | uniform |
| Array 5 | 39.79 | 22.93 | 1.56 | 1.24 | uniform |
| Array 6 | 34.38 | 22.15 | 1.79 | 1.11 | planophile |

| | | | | | |
|---------------------------|-------|-------|------|------|---------|
| Array 1+4+6 (elevated) | 39.2 | 22.21 | 1.71 | 1.32 | uniform |
| Array 2+3+5 (ambient) | 41.62 | 22.28 | 1.64 | 1.41 | uniform |

Figures 6 and 7 illustrate the frequency distribution of leaf inclination angles measured using ImageJ and AngleCam, respectively, at the BIFoR site, displaying the distribution pattern across different angle degrees.

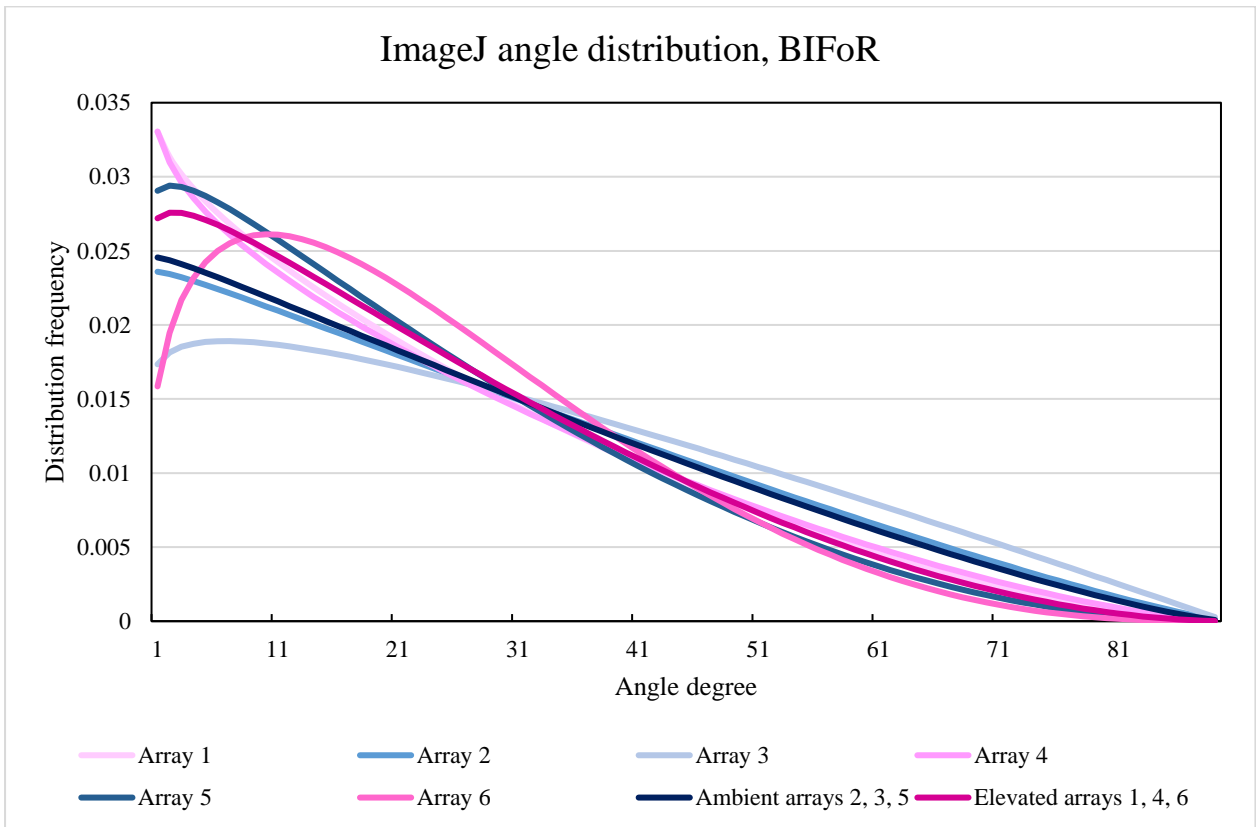


Figure 6. ImageJ leaf inclination angle distribution for the BIFoR site.

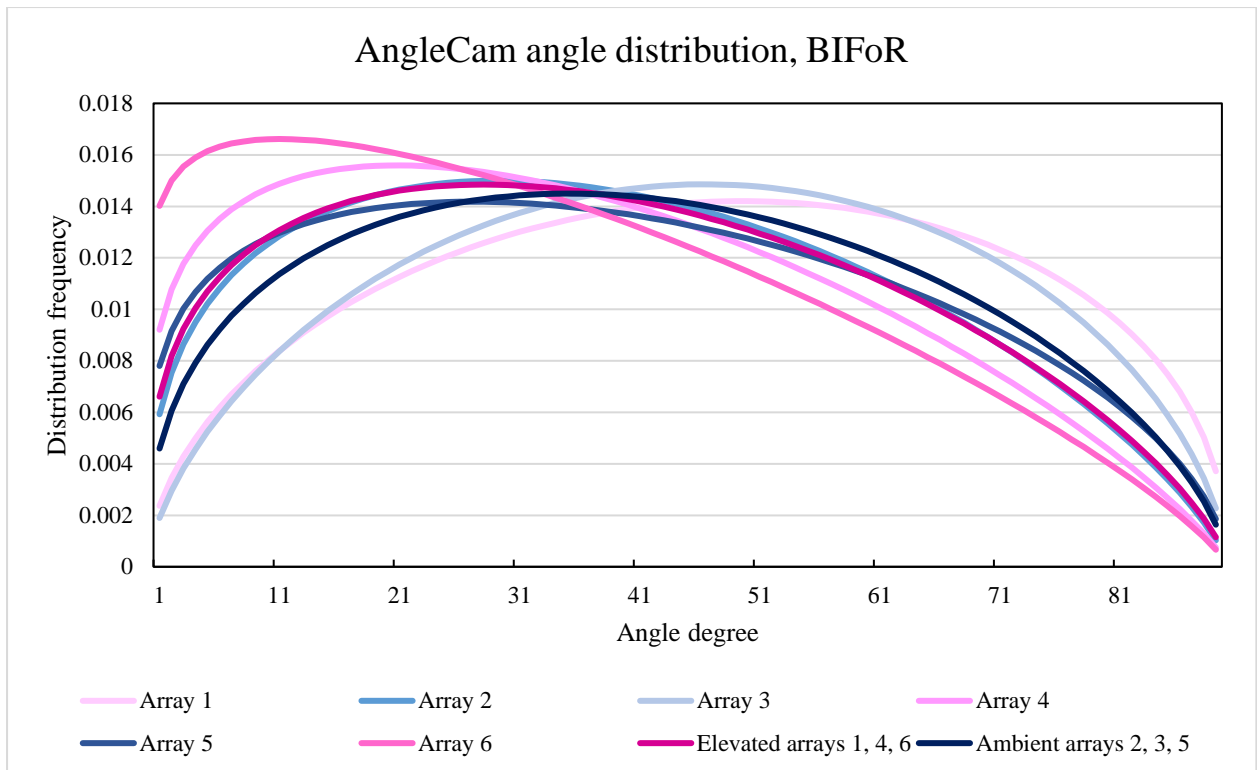


Figure 7. AngleCam leaf inclination angle distribution for BIFoR site.

Next, Figures 8 and 9 go more in-depth from the height perspective, as the average mean angles have been found using the height level data instead of the array data. For example, all 5-meter height measurements from elevated arrays (1, 4, 6) and ambient arrays (2, 3, 5) have been calculated together to get the mean angle for the 5-meter height level, regardless of the array. The same has been done for 10-, 15-, 20-, and 25-meter height measurements.

In Figure 8, the data from ImageJ has been used. The average difference between the elevated and ambient arrays can be seen with the calculated error bars showing the standard deviation for each height. The same premise applies for Figure 9 as for Figure 8, with the difference being that only the input is from AngleCam data.

It is noticeable that as the height increases, the inclination angles get steeper. This is true for both elevated and ambient arrays, leaves behave in the same pattern of going to a more vertical inclination the higher in the canopy they are. However, the absolute value differences between the lowest, 5-meter, and the highest, 25-meter, are near 20 degrees when measured with ImageJ. Yet, measured by AngleCam, the same heights exhibit a smaller value difference in the top and bottom of the canopies, approximately 10 degrees.

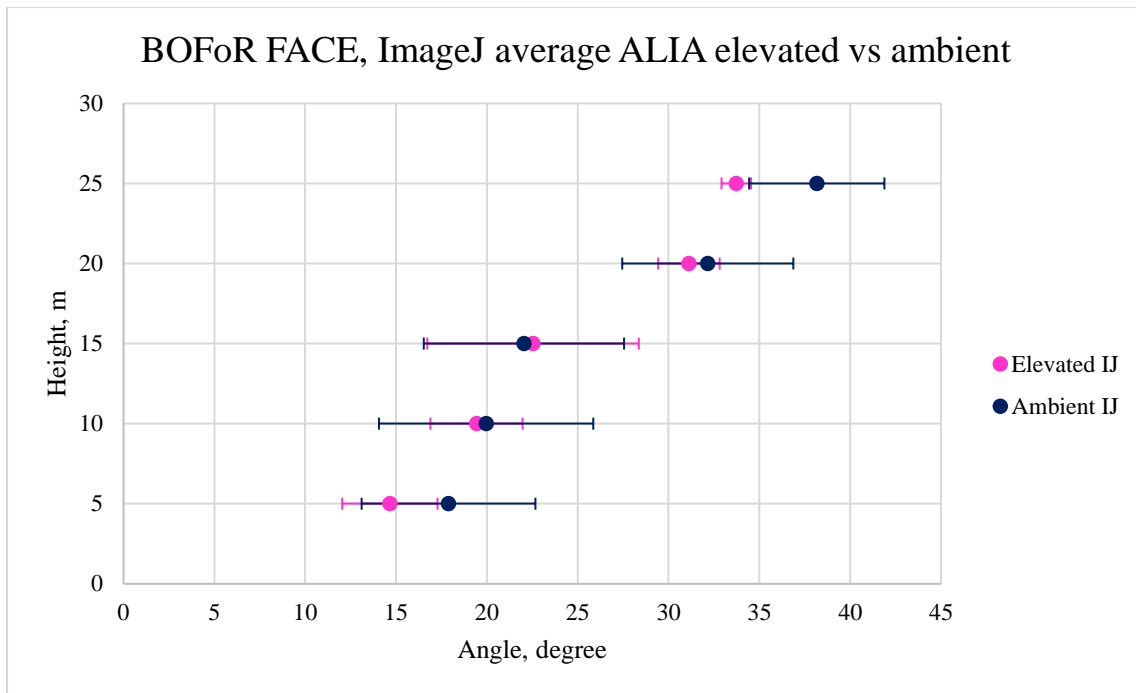


Figure 8. BIFoR site leaf inclination angles are measured according to height level with average elevated and average ambient degrees measured with ImageJ. Error bars are given with standard deviation values based on the averages for each height.

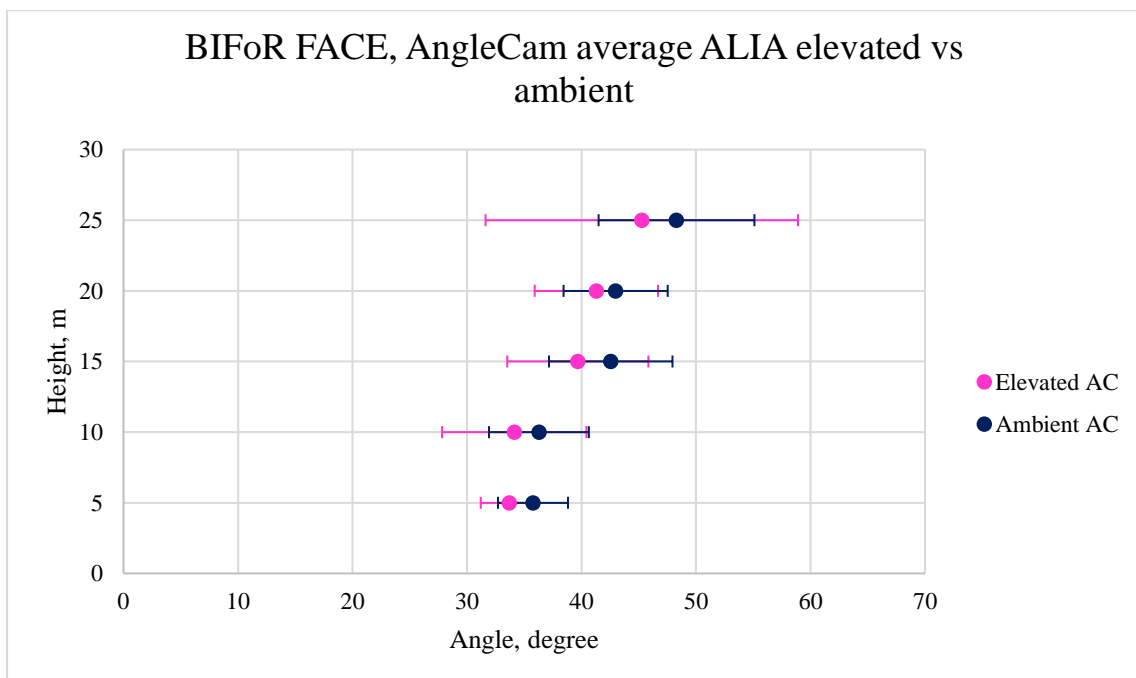


Figure 9. BIFoR site leaf inclination angles according to height level with average elevated and average ambient degrees measured by AngleCam. Error bars are given with standard deviation values based on the averages for each height.

Comparison of leaf inclination data from the EucFACE site using ImageJ and AngleCam

Figure 10 illustrates the differences between ImageJ and AngleCam measurements for the EucFACE site. Here it is visible that for both the elevated (1, 4, 5) and the ambient (2, 3, 6) rings the higher leaf inclination values are for the ImageJ measurements, while AngleCam shows consistently lower values for the same rings. This is opposite to that of BIFoR, where AngleCam predicted higher values than ImageJ (Fig. 5).

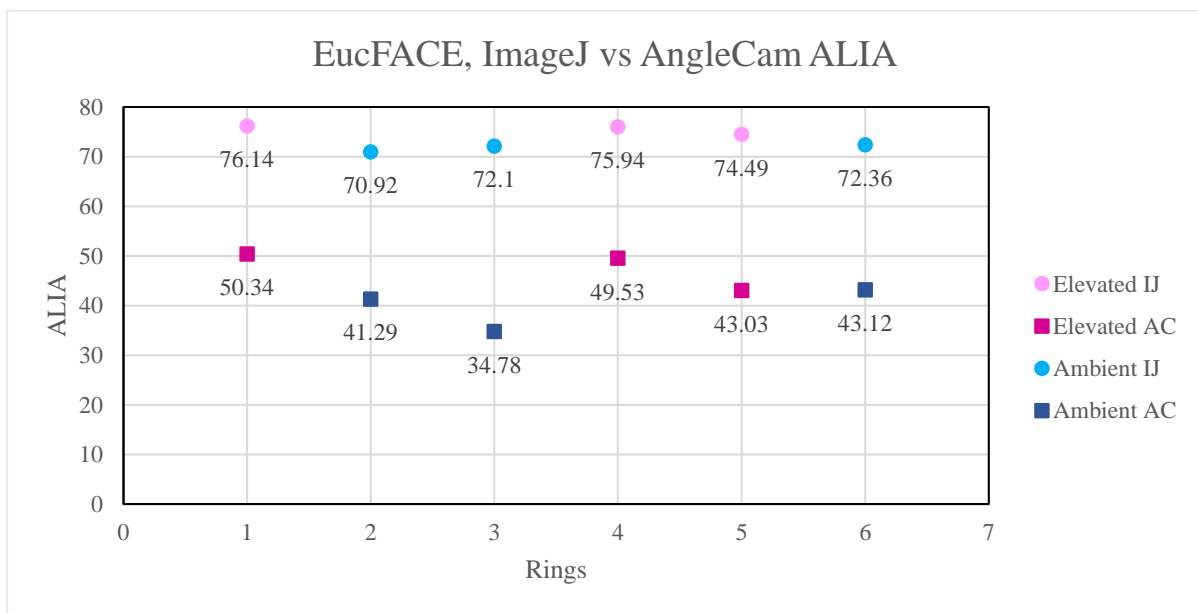


Figure 10. EucFACE site comparison of ImageJ and AngleCam data for both elevated and ambient rings.

Below are two tables comparing leaf inclination data collected using ImageJ and AngleCam at the EucFACE site. Table 3 presents data from ImageJ, while Table 4 presents data from AngleCam.

Table 3 shows visible results for rings 1-6, with the elevated and ambient rings listed separately. The count of measurements done by ring previously by Pisek *et al.* (2021) is between 411 and 434. The mean angles range from 70.92 to 76.14 degrees, with the standard deviation being 13.48-17.57 degrees from the mean. Parameters μ and ν are respectively from 0.57 to 1.07 and 2.66 to 4.39. The distribution type collectively is erectophile for all rings.

On a separate line are the elevated rings (1, 4, 5) with a count of 1282 measurements done and a mean angle of 75.53 degrees. The standard deviation from this is 14.48 degrees, with the parameters being μ 0.68 and ν 3.53. The type of leaf distribution was erectophile for the elevated rings. The ambient rings (2, 3, 6) accounted for 1253 measurement counts and had a collective mean inclination angle of 71.8 degrees. The standard deviation was calculated at 15.42, with parameter μ being 0.91 and parameter ν being 3.58. Ambient rings also demonstrated the erectophile-type distribution.

Table 3. *EucFACE site dataset of mean leaf inclination angles, standard deviation (s.d.), parameters μ and ν , and de Wit leaf distribution type as measured with ImageJ, including count of measurements done.*

| Dataset | Count | Mean | s.d. | μ | ν | Type |
|--------------------------|-------|-------|-------|-------|-------|-------------|
| Ring 1 | 425 | 76.14 | 13.48 | 0.74 | 4.07 | erectophile |
| Ring 2 | 411 | 70.92 | 17.57 | 0.72 | 2.66 | erectophile |
| Ring 3 | 420 | 72.1 | 14.45 | 1.03 | 4.15 | erectophile |
| Ring 4 | 434 | 75.94 | 15.12 | 0.57 | 3.1 | erectophile |
| Ring 5 | 423 | 74.49 | 14.77 | 0.74 | 3.56 | erectophile |
| Ring 6 | 422 | 72.36 | 14.06 | 1.07 | 4.39 | erectophile |
| Ring 1+4+5 (elevated) | 1282 | 75.53 | 14.48 | 0.68 | 3.53 | erectophile |
| Ring 2+3+6 (ambient) | 1253 | 71.8 | 15.42 | 0.91 | 3.58 | erectophile |

The same data (photographs) were used as input for AngleCam; the results can be seen in Table 4. For AngleCam, no count of measurements is available due to the program's build. For rings 1-6, the mean angles varied from 34.78 to 50.34 degrees, with the standard deviation ranging from 22.12 to 23.52 degrees. The parameters were μ and ν , respectively 1.27-1.79 and 1.13-

1.7. The leaf distribution type is uniform for rings 1, 2, 4, 5, and 6, with the outlier being ring 3 with a planophile distribution type.

Elevated rings (1, 4, 5) are brought out on a separate line with a mean angle of 46.56 degrees and a standard deviation of 22.75 degrees. The distribution type is uniform and in line with the individual rings. The mean angle for the ambient rings (2, 3, 6) was 39.68 degrees. The distribution type was classified as uniform.

Table 4. *EucFACE* site dataset of mean leaf inclination angles, standard deviation (s.d.), parameters μ and ν , and de Wit leaf distribution type as measured by AngleCam.

| Dataset | Mean | s.d. | μ | ν | Type |
|--------------------------|-------|-------|-------|-------|------------|
| Ring 1 | 50.34 | 22.66 | 1.27 | 1.62 | uniform |
| Ring 2 | 41.29 | 23.52 | 1.43 | 1.21 | uniform |
| Ring 3 | 34.78 | 22.12 | 1.79 | 1.13 | planophile |
| Ring 4 | 49.53 | 22.15 | 1.39 | 1.7 | uniform |
| Ring 5 | 43.03 | 23.11 | 1.45 | 1.33 | uniform |
| Ring 6 | 43.12 | 22.96 | 1.48 | 1.36 | uniform |
| Ring 1+4+5 (elevated) | 46.56 | 22.75 | 1.4 | 1.51 | uniform |
| Ring 2+3+6 (ambient) | 39.68 | 22.89 | 1.57 | 1.24 | uniform |

Tables 3 and 4 above showcase the collective data acquired through ImageJ and AngleCam programs.

To graphically visualise the results, Figure 11 gives an overview of the angle distribution frequency for EucFACE as estimated by the ImageJ approach and Figure 12 presents the predicted leaf inclination angle frequencies by AngleCam.

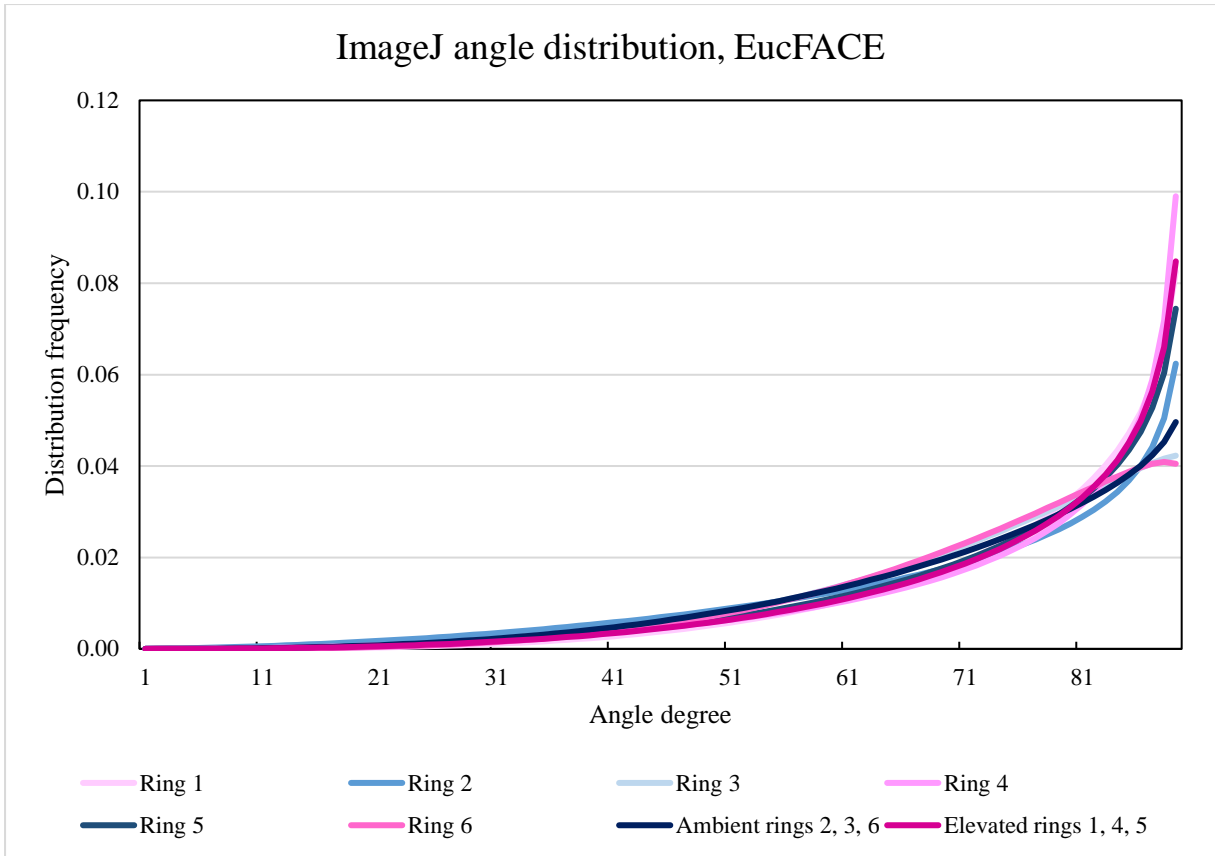


Figure 11. ImageJ leaf inclination angle distribution for EucFACE site.

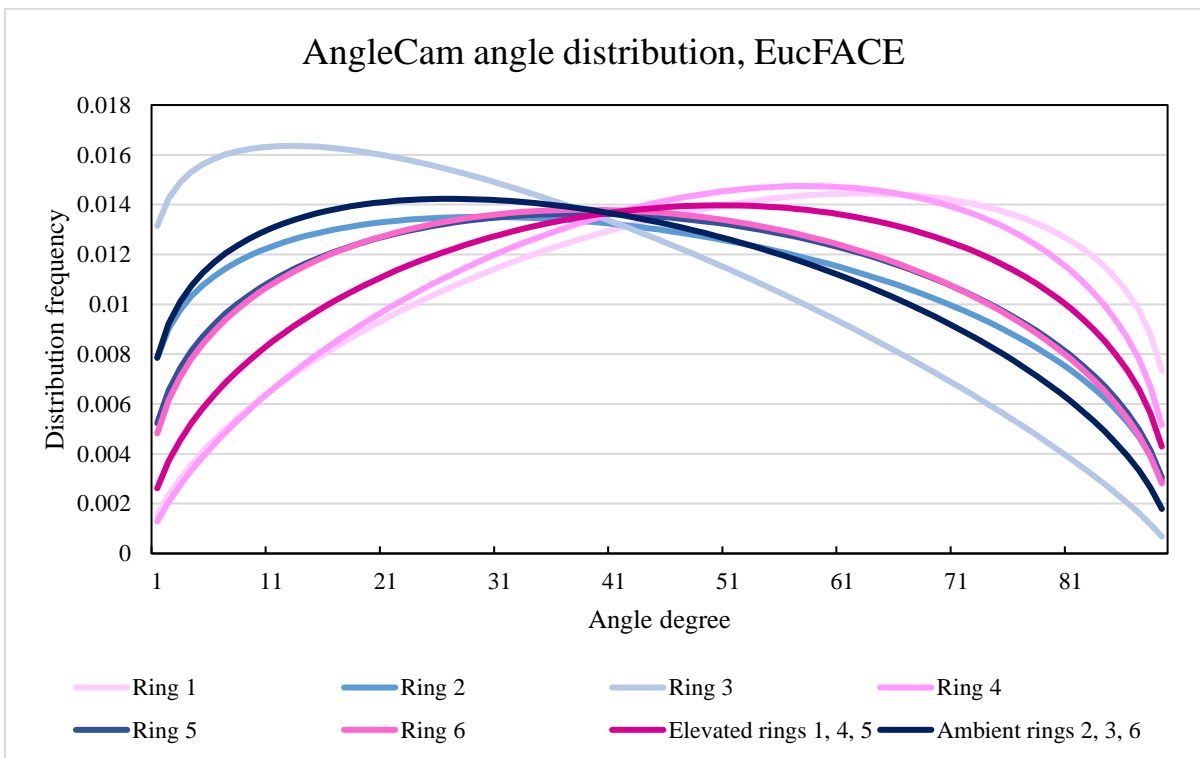


Figure 12. AngleCam leaf inclination angle distribution for EucFACE site.

8. DISCUSSION

The main aim of this thesis was to compare two methodologies of digital leveled photography (Pisek *et al.*, 2011; Ryu *et al.*, 2010) and a new machine learning-based program called AngleCam (Kattenborn *et al.*, 2022) to see the possibilities for future automation of leaf angle measurements. The input datasets were obtained from two distinct sites - BIFoR (U.K.) and EucFACE (Australia). At both sites, the same two methodologies were applied. While the study achieved its overall goal, the results and validation of AngleCam deviated more than anticipated from the established methodologies (ImageJ measurements).

First, it is important to acknowledge that the tested methodologies employed are still in the testing phases and are not yet flawless techniques. In addition, a few researchers have looked at finding an alternative and better method for doing *in situ* measurements for just under two decades. The dataset provided from the U.K. and Australia gave valuable input for the validation of AngleCam, a more targeted data collection may enhance future analyses. Moreover, using ImageJ for hand measurement, it is evident that the human eye plays a substantial role in the differentiation between the twigs, branches, and clouds in the pictures, where AngleCam might still struggle.

The first of our questions for this study was to find out if the two methods - leveled digital photography (Ryu *et al.*, 2010; Pisek *et al.*, 2011) and machine-learning-based AngleCam (Kattenborn *et al.*, 2022) - provide comparable results. After the data analyses, it is possible to see that while the data collected with each method seem consistent within the methodologies, comparing the two has quite a significant discrepancy. The measurements, specifically mean inclination angles of leaves, were at times up to 20 degrees different and cannot be suggested as slight errors or discrepancies. On the positive side, the two methodologies tend to exhibit the same trend in mean angles. For the BIFoR site, the trends matched better than those of the EucFACE. This could be because the AngleCam program had previously been trained mainly with broadleaf trees in the Leipzig test grounds in Germany, which tend to be more similar to that of the BIFoR site and less to the *Eucalyptus* tree leaves. The results here indicate the originally used training dataset by Kattenborn *et al.* (2022) might not be representative enough to get the reliable mean angles from the machine learning program AngleCam. In addition, it is quite possible that the AngleCam program suspected the artificial structures and clouds in the photographs to be part of the canopy (Fig. 13). AngleCam algorithm considers the scene as a whole, and these non-leaf elements might confuse/bias the predicted leaf angle result.

AngleCam was developed with a very limited training dataset for erectophile distribution class examples. Results presented here indicate the method might not be reliable in conditions with prevalently erectophile distribution type vegetation.



Figure 13. Photo at the BIFoR site at the 25-meter height level. The image shows that the structure of the site, as well as clouds and establishments further away, are within the frame.

The second question asked was: Is spherical leaf angle distribution appropriate for present species at the two sites? Based on the data gathered, the spherical distribution of leaves is currently the correct assumption for neither site. This could be because of the different conditions at the sites and the fact that perhaps the spherical distribution is incorrect for trees in a wooded area. However, ImageJ results indicate very different distribution types between BIFoR (planophile) and EucFACE (erectophile), which is to be expected. In contrast, AngleCam indicated the identical LIAD type for both sites, which is slightly unrealistic as it is very clear (Fig. 14) that the angles are not the same. Either way, none of the approaches classified any of the data as a spherical LIAD type.



Figure 14. *On the left side are English oaks from array 3 at 15m height, and on the right side are visible the leaves of the Eucalyptus tree in ring 3. In comparison, it is visible that the leaf inclination angles are dissimilar.*

Lastly, the third research question was: Are there any changes in leaf inclination angle distribution under different CO₂ concentrations? There seem to be slight changes when an extra 150 ppm CO₂ is introduced to the rings/arrays. However, no significant change can be witnessed when looking solely at the elevated vs ambient rings/arrays. Canopies slightly changed with elevated carbon dioxide, while the absolute values differed, both methods picked up the same trends. For the BIFoR site, the inclination angles tended to become more horizontal and smaller in degree values. This was the opposite though for the EucFACE site, where the eucalyptus tree leaves tended to become more vertical and higher in the degree values. Despite the differences between methods, results indicate no pronounced change in leaf inclination is to be expected due to elevated CO₂ conditions in the future as the changes are slight. At all height levels through the vertical canopy at BIFoR experiment, AngleCam indicates higher values than those measured with ImageJ.

Leaf inclination could be an interesting factor for the future of the environment in the changing climate scenarios. In warmer climates, forests would provide less coverage in the case of elevated CO₂, as the leaves are at steeper angles and are not trying to intercept but potentially even avoid the incoming radiation from the sun's rays in hotter climates, such as Australia. For

temperate forests, this could potentially be the opposite, the foliage will try to capture more of the incoming radiation the higher the amount of CO₂ that is in the atmosphere.

While somewhat similar trends are present at BIFoR site, this is completely overturned when looking at the EucFACE site, as the angles and distributions of these contradict each other heavily (Fig. 11 vs 12). This could be brought back to the fact that while AngleCam has been trained with over 60 different shrub and tree species, it has had little training with leaf angles identified as erectophile distribution type. The slightly better correlation between BIFoR site measurements for AngleCam and ImageJ could be because some of the species (English Oak, Sycamore Maple) present at the site were also used for training AngleCam. However, with more training with specific species and distribution types, AngleCam shows a lot of promise in automating leaf angle measuring. While the leveled digital photography approach seems more reliable, the amount of labour that goes into getting a decent-sized pool of samples is immense. Further work could be done using the different height data found at BIFoR to understand better the changes within a height level and per species rather than an array as a whole.

9. CONCLUSION

The aim for this thesis was to explore the two different methodologies, leveled digital photography approach and AngleCam, and their comparison on the results. In addition, it was assumed that the distribution is spherical for canopies. Last but not least, the aim was to see if the elevated carbon dioxide affected the leaf inclination angles.

While AngleCam might be a more convenient method for retrieving the leaf inclination information, this study showed its possibly limited performance, especially in conditions with a relatively limited training sample. The program performs better when the input is similar to the training materials used in the development of AngleCam, however, it lacks the training when dealing with more vertical inclinations such as the eucalyptus trees in Australia. This should be kept in mind when operating it in a different climate. While digital leveled photography method seems to be more reliable, then the main obstacle of time and resource is still apparent. However, while the actual values between the two methods vary greatly, both can provide information about the overall trend/change direction with treatment.

From the current dataset of values and the de Wit classification and Beta parameter calculations from this thesis, the overall assumption that a canopy tends to be spherical can be overturned. During no occasion was the canopy overall classification a spherical one with the current data. So, it is plausible to consider that the spherical classification is false for such species as present in the study.

In regard to the increased CO₂ levels at the test sites, the canopies reacted slightly. However, no significant change is noticeable when comparing the results for the ambient and the elevated arrays/rings. While a change is there in degree values, at most times it does not affect the distribution types amongst the arrays/rings. For the BIFoR site, steeper angles are noticed from the data for the ambient arrays rather than the elevated concentration arrays. The opposite is true for EucFACE, as the elevated rings tended to consist of steeper inclination angles while introduced to extra CO₂. While the ambient rings remained at a flatter inclination.

In conclusion, the research questions proposed at the start of thesis have been met with answers and the methodologies chosen have been sufficient to help resolve the questions. Both methodologies used have worked and provided results to work with, however the values themselves were a bit of a surprise. Leading to an understating that more work needs to be done to assess the readiness of AngleCam for fieldwork in different locations than it was trained for.

Both methodologies have a future in remotely measuring leaf inclinations, minimizing the need for expensive gear and/or time to receive trustworthy results. As climate change continues and the carbon dioxide in the atmosphere rises, this work gives an inkling of how that might effect trees in the future.

Lehe kaldenurga jaotus lehtpuu- ja igihaljas lehtpuumetsas kõrgendatud atmosfäärse CO₂ tingimustel

Anni Maria Pearson

KOKKUVÕTE

Lõputöö eesmärk oli valideerida AngleCami tarkvara, kui uut ja lihtsat lehe kaldenurga mõõtmise meetodit. Samal ajal oli ka eesmärk võrrelda kaht erinevat meetodit, mida antud töös kasutati – tasapinnaline digitaal kaamera lähenemine ja AngleCami tarkvara. Eraldi eesmärkideks olid veel sfäärilise jaotuse paikapidavuse uurimine ning kõrgendatud CO₂ keskkonnas lehe kaldenurkade muutuste jälgimine.

Uurimuse aluseks on *FACE* ehk '*Free Air Carbon Dioxide Enrichment*' projektid. Antud töös on kasutatud andmeid BIFoR *FACE* projektist, mis asub Birminghamis, Suurbritannias ja EucFACE, mis asub Sydney lähedal Austraalias. Katsealustes metsades on kuus „rõngast” (ingl k vastavalt „*array*” või „*ring*”), millest kolmele lisatakse neid ringe ümbritsevatest struktuursetest „rõngastest” 150 ppm CO₂ juurde, kolmele ülejäänule ei ole midagi lisatud. Nii UK kui ka Austraalia katsealast on tehtud pildid. BIFoR proovialalt on tehtud pildid iga viie meetri tagant, kuni 25 meetrini. EucFACE puhul on pilte tehtud kompaktsemalt ja samuti terves moodsus kuni 25 meetrini.

BIFoR piltidelt mõõdeti käsitsi ImageJ tarkvaraga lehtede kaldenurki, jälgides, et lehe serv oleks vaataja poole – tasapinnalise digitaalfotograafia meetod. Samu fotosid kasutati ka AngleCam sisendiks, mis põhineb CNN ehk *convolutional neural networki* ehk masinõppe peal. Antud programmi on varasemalt väljatöötatud ligi 60 erineva taime- ja puuliigi peal testides. EucFACE piltidelt oli juba varasemalt kogutud andmed tasapinnalise digitaal fotograafia meetodil. AngleCamile anti sisendiks samad pildid.

Tulemusena oli lõpuks üle mitmetuhande käsitsi mõõdetud lehtede kaldenurgad. Kaldenurkade järgi arvutasid programmid de Wit klassikalise jaotuse hulkadeks. Tulemused olid veidi üllatavad. AngleCam valideerimiseks kasutati võrdlusena ImageJ-st saadud tulemusi, kuid nende omavaheline vahe oli küllaltki märgiline. Mõlemad meetodid küll suutsid leida lehestikus esinevad trendid, kuid tulemuste absoluutväärtused olid kohati kuni 20 kraadi juurde. Tulemused klappisid veidi paremini BIFoRis kui EucFACEis. Seda võib käsitleda, kui olukorda, kus AngleCam oli varasemalt treenitud liikidega, mille lehed olid väiksema nurga all, kui EucFACEis olevatel eukalüptipuudel. Seega algselt loodetud validatsioon osutus

veidike ebapiisavaks antud tingimustes. Jaotuste puhul saadi teada, et liikidel, mis olid vaatluse all antud töös, ei esine sfääriline jaotus üldse, seega eeldus, et lehestik on sfäärilise jaotusega ei pea paika antud tingimustel. Viimase punktina sooviti teada, kas kõrgendatud CO₂ tasemed avaldavad lehe kaldenurkadele mõju. Leiti, et mõju on olemas, BIFoRis kõrgendatud süsinikdioksiidiga ringides olid lehtede kaldenurgas väiksemad, seega lehed püüdsid rohkem päikese kiirgust püüda kõrgendatud keskkonnas. Kuid EucFACEis on vastupidised tulemused – lehed kõrgendatud süsinikdioksiidiga keskkonnas olid tervamate nurkade all, kui muidu. Muutused kõrgendatud CO₂ keskkondades on märgatavad, kuid mitte tähelepanuväärselt suured.

ACKNOWLEDGEMENTS

I would like to express gratitude to my supervisor, Jan Pisek, for the guidance along the way and their patience with me while figuring out academic writing. Their prompt and straightforward feedback as well as encouragement have been instrumental in making sure the quality of work and direction are right.

I am also immensely thankful for my secondary supervisor, Margit Aun, who took time to talk me through the process of data retrieval and code mishaps without which this thesis would have been short-lived. Their help during the writing process was also greatly appreciated.

A huge thank you to my program coordinator, Kairi Põldsaar, for guidance during the moments of hesitation. Their support has been invaluable.

Furthermore, I would like to extend my appreciation to my classmates with whom the process of thesis writing seems to be a bit lighter.

This thesis would not have been possible without the support and encouragement of all those mentioned above, and for that, I am truly grateful.

REFERENCES

- Bala, G. (2014). Can planting new trees help to reduce global warming. *Current Science*, 1623–1624.
- BIFoR FACE*. (n.d.). University of Birmingham. <https://www.birmingham.ac.uk/research/bifor/face>, last accessed 23.03.2024
- Chianucci, F., Pisek, J., Raabe, K., Marchino, L., Ferrara, C., & Corona, P. (2018). A dataset of leaf inclination angles for temperate and boreal broadleaf woody species. *Annals of Forest Science*, 75, 1–7.
- de Wit, C. T. (1965). *Photosynthesis of leaf canopies*. Pudoc.
- Duursma, R. A., Gimeno, T. E., Boer, M. M., Crous, K. Y., Tjoelker, M. G., & Ellsworth, D. S. (2016). Canopy leaf area of a mature evergreen Eucalyptus woodland does not respond to elevated atmospheric [CO₂] but tracks water availability. *Global change biology*, 22(4), 1666–1676.
- Gallery – EUcFACE Experiment*. (n.d.). <https://eucface.hieresearch.org/index.php/portfolio/>, last accessed 23.03.2024
- Gimeno, T. E., Crous, K. Y., Cooke, J., O’Grady, A. P., Ósvaldsson, A., Medlyn, B. E., & Ellsworth, D. S. (2016). Conserved stomatal behaviour under elevated CO₂ and varying water availability in a mature woodland. *Functional Ecology*, 30(5), 700–709.
- Goel, N. S., & Strebel, D. E. (1984). Simple beta distribution representation of leaf orientation in vegetation canopies 1. *Agronomy Journal*, 76(5), 800–802.
- Hart, K. M., Curioni, G., Blaen, P., Harper, N. J., Miles, P., Lewin, K. F., Nagy, J., Bannister, E. J., Cai, X. M., & Thomas, R. M. (2020). Characteristics of free air carbon dioxide

- enrichment of a northern temperate mature forest. *Global change biology*, 26(2), 1023–1037.
- Kattenborn, T., Richter, R., Guimarães-Steinicke, C., Feilhauer, H., & Wirth, C. (2022). AngleCam: Predicting the temporal variation of leaf angle distributions from image series with deep learning. *Methods in Ecology and Evolution*, 13(11), 2531–2545.
- Oguchi, R., Onoda, Y., Terashima, I., & Tholen, D. (2018). Leaf anatomy and function. *The leaf: a platform for performing photosynthesis*, 97–139.
- Pisek, J., Řezníčková, L., Adamson, K., Ellsworth, D. S., & Williams, D. (2021). Leaf inclination angle and foliage clumping in an evergreen broadleaf Eucalyptus forest under elevated atmospheric CO₂. *Australian Journal of Botany*, 69(8), 622–629.
- Wang, W.-M., Li, Z.-L., & Su, H.-B. (2007). Comparison of leaf angle distribution functions: Effects on extinction coefficient and fraction of sunlit foliage. *Agricultural and Forest Meteorology*, 143(1–2), 106–122.

Lihtlitsents lõputöö reprodutseerimiseks ja üldsusele kättesaadavaks tegemiseks

Mina, Anni Maria Pearson,

1. annan Tartu Ülikoolile tasuta loa (lihtlitsentsi) minu loodud teose
„Leaf inclination angle distribution in deciduous broadleaf and evergreen broadleaf Eucalyptus forests under elevated atmospheric CO2“,
mille juhendajad on PhD Jan Pisek ja PhD Margit Aun,
reprodutseerimiseks eesmärgiga seda säilitada, sealhulgas lisada digitaalarhiivi Dspace kuni autoriõiguse kehtivuse lõppemiseni.
2. Annan Tartu Ülikoolile loa teha punktis 1 nimetatud teos üldsusele kättesaadavaks Tartu Ülikooli veebikeskkonna, sealhulgas digitaalarhiivi DSpace kaudu Creative Commons'i litsentsiga CC BY NC ND 4.0, mis lubab autorile viidates teost reprodutseerida, levitada ja üldsusele suunata ning keelab luua tuletatud teost ja kasutada teost ärieesmärgil, kuni autoriõiguse kehtivuse lõppemiseni.
3. Olen teadlik, et punktides 1 ja 2 nimetatud õigused jäävad alles ka autorile.
4. Kinnitan, et lihtlitsentsi andmisega ei riku ma teiste isikute intellektuaalomandit ega isikuandmete kaitse õigusaktidest tulevaid õigusi.

Anni Maria Pearson

15.05.2024