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Research Article
Ground Covering Characteristics of Sugarcanes Using High-angle Images and Their Relationship with Growth Destructive Sampling

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Abstract
Background and Objective: One strategy to improve sugarcane yield is understanding the optimization of the photosynthetic organs, such as leaf area, to accelerate ground and canopy cover. Non-destructive sampling through image analysis, using a high-angle image as a simple method, could represent the percentage of ground cover. Therefore, the purpose of this study was to evaluate ground cover using high-angle images of diverse sugarcane cultivars and their relationship with destructively sampled growth. **Materials and Methods:** A Randomized Complete Block Design (RCBD) with four replications was used. Five commercial sugarcane cultivars with different canopy types were assigned as treatments. Destructive growth samples were collected at 30 days intervals from 30 to 120 days after transplanting (DAP). Image analysis for ground cover using Gimp 2.20.12 software and non-destructive growth samples were measured at 15 days intervals from 30 to 120 DAP. **Results:** The five cultivars in this study had different ground covering speeds. Cultivar 'KPS01-12' was identified as the fastest ground-covering cultivar and 'UT-13' had moderate ground coverage, whereas UT-12 and 'KK3' were slower ground-covering cultivars. The fastest ground-covering cultivar, 'KPS01-12', also revealed outstandingly high measurements during destructive growth sampling, namely leaf area index (LAI), leaf area (LA), leaf dry weight, stalk dry weight and biomass. Correlation between ground cover, LAI and leaf dry weight existed at 90 and 120 DAP in diverse sugarcane cultivars, but not 30 and 60 DAP. **Conclusion:** The ground coverage measurement using a high-angle image is a feasible indirect measurement of growth traits that would have to be measured destructively at some developmental stages. This information would help support further sugarcane research as a non-destructive criterion in several aspects.

Key words: Sugarcane, ground cover, LAI, image analysis, canopy development

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Sugarcane (*Saccharum officinarum* L.) is widely grown to produce sugar, bioenergy and biogas[1,2]. Sugarcane has been produced in more than 100 countries in the world[3]. Brazil has the highest sugarcane production value in the world at 32.9% of the total world sugarcane production, followed by India at 18.8%, China at 6.0% and Thailand at 4.4%[4]. Sugarcane production in the world has decreased by 85,500 tons from 2019 until 2020[4]. Great challenges, from pre- to post-harvest, affect sugarcane cultivation and reduce yields. Irrigation systems, soil quality, climatic change, harvest period, manpower shortage, plant density and weeds are factors that affect sugarcane yield[5-11].

Plant density effectiveness is dependent on increasing the LAI to optimum levels, accelerating ground cover and canopy cover and affecting the final yield[12]. Canopy development is influenced by genotype, environmental factors and managerial decisions, such as row spacing[13-15]. Moreover, the speed of canopy coverage is directly involved with a light interception and it consequently influences physiological processes, such as photosynthetic assimilation and plant community competition, among others, along with weed development. Castro-Nava *et al.*[16] reported that a large-canopy sugarcane cultivar was suitable for high-temperature rainfed conditions and varietal differences greatly affected canopy closure depending on conditions. In contrast, the small-canopy variety was able to continue canopy development processes for a longer period in the presence of increasing water stress[17].

Analysis of ground cover in sugarcane is possible with methods like genetic engineering, robotics and imaging analysis[18]. Remote sensing is one of the common methods for analyzing the ground cover in sugarcane because it is inexpensive and time-saving to obtain the data[19,20]. Another ground cover analysis method is image analysis, which is more appealing because it can predict the harvest yield with more than 90% accuracy[20,21]. Several studies have reported the relationship between ground cover and LAI in various types of plants, such as potatoes, cotton and legume. Boyd *et al.*[22] reported that the light interception efficiency of a potato canopy can be estimated by comparing the slopes of the LAI and ground cover correlation. The relationship between ground cover and LAI also showed high significance in barley[23]. The cotton canopy covers around 90% of the field when it reaches the maximum LAI, according to the link between ground cover data and LAI measured in the field[24].

However, there has been a lack of information on the relationship between ground cover and LAI in sugarcane. In addition, it is important to avoid destructive sampling. Hence, the image analysis using a simple high-angle image method could be used first to evaluate the required percentage of ground cover. Therefore, the objective of this study was to evaluate ground coverage using high-angle images of diverse sugarcane cultivars and their relationship with growth measurements collected through destructive sampling. This information would be an advantage to support further sugarcane research as a non-destructive criterion in several aspects.

MATERIALS AND METHODS

Site description and experimental details: This research was carried out at the Agronomy Field Crop Station, Khon Kaen University, Thailand (Latitude 16.4591873, Longitude 102.8119933 and 179 m above sea level), during the early rainy season, of a sugarcane production system from February, 2020 to June, 2020. A Randomized Complete Block Design (RCBD) with four replications was used. Each plot consisted of seven rows with a length of 15 m per plot and plant spacing was 1×1.5 m. The treatments were five commercial cultivars, namely 'KK3', 'KKU99-02', 'KPS01-12', 'UT12' and 'UT13'. Each cultivar has a different leaf orientation. The 'KK3', 'KKU99-02', 'KPS01-12' and 'UT12' cultivars have large leaf sizes, high leaf surface areas and horizontal leaves. Meanwhile, the 'UT13' cultivar has slender long and vertical leaves.

Cane sets were planted in plastic bags to prepare uniform seedlings before transplanting them to the field experiment. The seedlings were transplanted into the field with 1×1.5 m plant spacing 1 month after planting (MAP). The soil in this experiment was classified as being siliceous, is hypothermic, Oxic Paleustults, Yasothon series, WRB: Arenosols, a sandy soil with a sand content of 84.93%, a clay concentration of 5.07% and a silt component of 10.0%. The soil's pH value was 5.5, 3.10 c mol kg^{-1} the cation exchange capacity (CEC) and there was 0.847% organic matter (OM), 0.028% total nitrogen, 32 mg kg^{-1} of available phosphorus, 30 mg kg^{-1} of exchangeable potassium and 214 mg kg^{-1} of exchangeable calcium. On the transplanting date, a basal application of herbicide using carbofuran (2,3-dihydro-2,2-dimethyl benzofuran-7-yl methylcarbamate 3% granular) was applied. Fertilizer was applied at the transplanting date and a top dressing was applied at 90 DAP. The K was applied at a rate of 25 kg per ha, while 50 kg per ha of N and P were applied. Drip irrigation was used for supplementary irrigation from transplanting to 30 DAP for setting the uniformity of the plant stand. Depending on the number of weeds in the experimental area, weed management was carried out manually once per month throughout the experimental period.

Data collection: Growth characteristics such as plant height, the number of tillers and the number of leaves per tiller were measured for three plants per plot. The plant height was measured from the soil surface level to the last exposed dewlap of the main stem at 15 days intervals from 30 to 120 days after planting (DAP). The number of tillers and the number of leaves per tiller were counted. These traits were collected at 15 days intervals from 30 to 120 DAP. Stem and leaf dry weights were collected for three plants per plot at 30, 60, 90 and 120 DAP. The sugarcanes of each plot were cut at the ground level and then separated from stems and leaves. Leaf samples in each plot had LA measured using an LI-3100 area meter (Li-COR, Lincoln, Inc., Nebraska, USA). The LAI was then calculated from LA divided by ground area. Both stem and leaf samples were then dried in an oven at 80°C for 72 hrs or until constant weight and leaf and stem dry weights were determined.

Ground coverage data was taken with a high-angle picture from the ground to the camera with a height of 2 m using a steel frame (same distance between camera and plant sample). The steel frame was put in a base with a size of 1×1 m^2 covering sugarcane ground spacing. The pictures were taken with a Fuji Camera (XA-5) at 15 days intervals from 30 to 120 DAP and the photographed plant was marked to use the same plant on every measured date. The ratio was one-sided leaf area per unit ground area. The high-angle picture was analyzed for ground cover using the Adobe Photoshop application and the ground cover in the sugarcane image was converted to negative film and cropped to meet the dimensions 1×1 m. The negative film pictures were analyzed using Gimp 2.20.12 software to determine the area of the leaf from the picture. The percentage of ground cover was then calculated as follows:

$$\text{Ground cover (\%)} = \frac{\text{Area of leaf from the picture}}{\text{Total ground area}} \times 100$$

Statistical analysis: The analysis of variance for all parameters was carried out by following an RCBD model. The data were subjected to statistical analysis using Statistix 8 software program version 8.0 (Analytical Software, Tallahassee, Florida, USA). Least significant differences (LSD) were conducted to compare means for significant variables at the 0.05 level of probability. The correlations among these parameters were determined using simple correlations.

RESULTS

Percentage of ground cover area of sugarcane cultivars: The percentage of ground cover among all cultivars showed significant differences at 45, 60, 75 and 90 DAP. Meanwhile, there was no significant difference among cultivars at 30, 105 and 120 DAP. During 30 to 90 DAP, cultivar 'KPS01-12' had a high percentage value compared to other cultivars, meaning that 'KPS01-12' had a faster canopy closure rate (Fig. 1). However, for 60 DAP, cultivar 'KKU99-02' had a high percentage of canopy closure with a value of 85.30% and there was no significant difference from 'KPS01-12' (Fig. 1). 'UT12', 'UT13' and 'KK3' had low canopy percentage values at 45-90 DAP and these cultivars were classified as having slower canopy coverage. The 'KK3', 'KKU99-02' and 'KPS01-12' cultivars had decreased percentages of ground cover at 120 DAP, decreasing by 2.71, 1.25 and 6.34% from 105 to 120 DAP, respectively (Fig. 1).

Leaf area (LA), leaf area index (LAI) and leaf number of diverse sugarcane cultivars: At 30 DAP, in general, the LAs of all studied genotypes did not have significant differences. However, 'KK3' had a higher leaf area value compared to other cultivars at 60 DAP, except for 'KPS01-12'. The 'KPS01-12' had the highest LA at 90 and 120 DAP, meanwhile, 'UT12' revealed a low value of LA for almost all collected dates (Fig. 2a). The LAI of all these studied cultivars showed the same performance among cultivars and an increasing pattern with LA (Fig. 2b). From 30 to 120 DAP, in general, 'KK3', 'KPS01-12' and 'KKU99-02' showed a good performance in leaf number and had consistently high average numbers of leaves, meanwhile, 'UT12' and 'UT13' had smaller numbers of leaves on most collection dates (Fig. 2c).

Growth pattern of the sugarcane cultivars: The tiller and leaf numbers of the five cultivars showed similar patterns but at different magnitudes. The tiller number gradually increased from 30 to 90 DAT and it then decreased from 105 to 120 DAT. At the early growth stage, 'UT13' and 'UT12' had fewer tillers than other cultivars and 'UT12' revealed fewer tillers during 90-105 DAT. The five

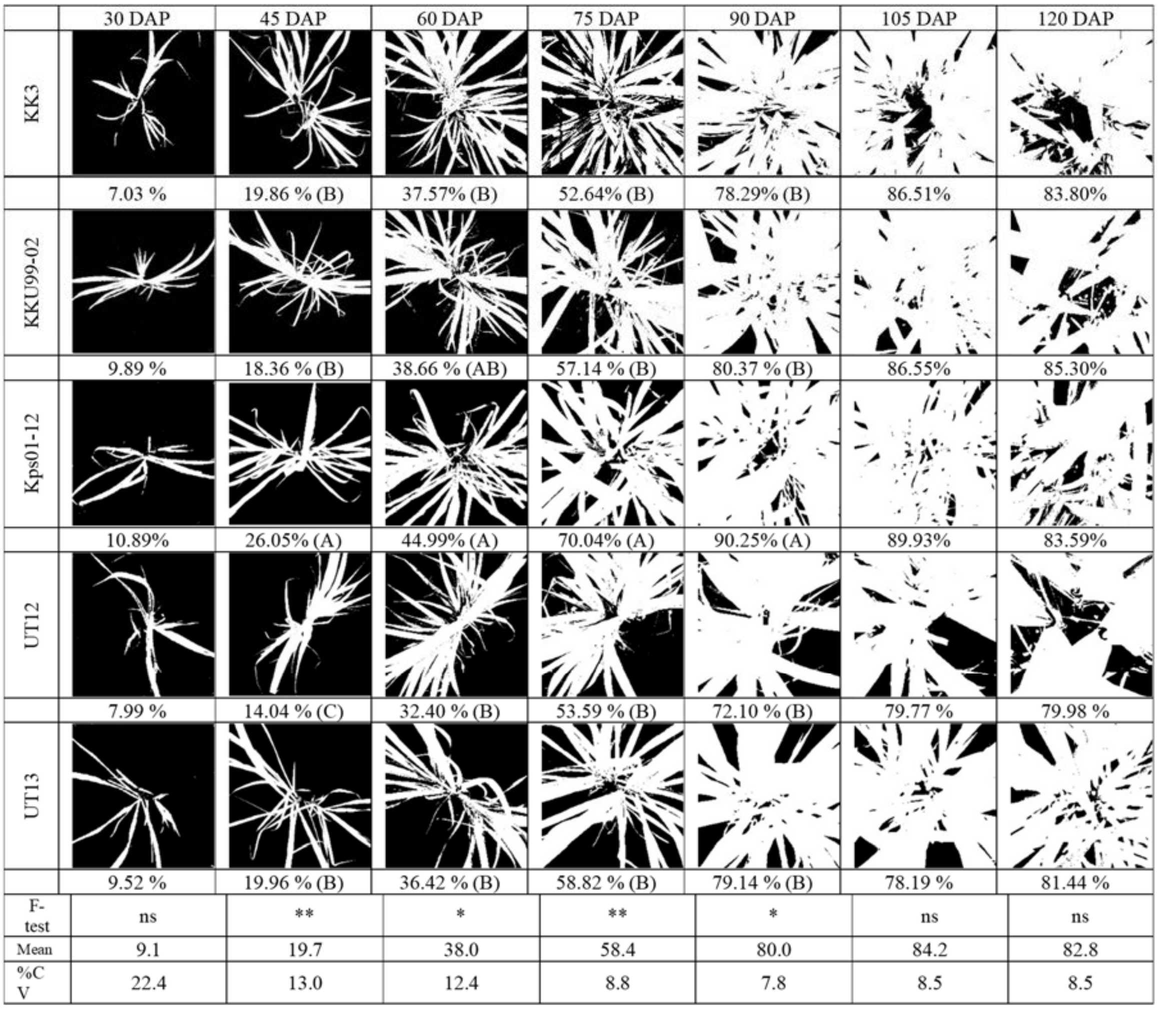


Fig. 1: Ground cover images of five cultivars (‘KK3’, ‘KKU99-02’, ‘KPS01-12’, ‘UT12’ and ‘UT13’) at 15 days intervals from 30 to 120 days after planting (DAP)

Mean values with different letters within a column show significance at p<0.05 by LSD

studied cultivars did not differ in tiller numbers at 120 DAT (Fig. 3a). In general, ‘KK3’ and ‘KPS01-12’ had high performance in tiller numbers during the tillering phase, but ‘UT12’ and ‘UT13’ had rather low tiller numbers during this phase. For height at 30-75 DAT, there was no significant difference. Cultivars ‘KPS01-12’ and ‘UT13’ had high height values at 105-120 DAT and ‘KKU99-02’, ‘UT12’ and ‘KK3’ were the shorter group (Fig. 3b).

Dry matter accumulation of diverse sugarcane cultivars: In terms of leaf dry weight, ‘KPS01-12’ had consistently high leaf dry weight, especially at 90 and 120 DAP. At 90 DAP, ‘UT13’ and ‘KK3’ showed higher leaf dry weights than ‘KKU99-02’ and ‘UT12’ (Fig. 4a). Cultivars ‘KPS01-12’ and ‘UT13’ showed high stalk dry weights and biomass at 90 DAP and ‘KPS01-12’ had the highest stalk and whole dry matter weights at 120 DAP (Fig. 4b and c).

Correlation analysis between ground cover percentage and growth parameters: During the initial germination phase at 30 and 60 DAP, the ground cover did not show any results correlated to LAI. However, at 60 DAP, the ground cover and the number of leaves and tillers had a significant positive correlation (Table 1). In the tillering and canopy development phase at 90 DAP, ground cover had a significant positive correlation with LAI, LA, leaf number, leaf dry weight and height. At 120 DAP, the correlation between ground cover and LAI, LA and leaf dry weight showed a highly positive relationship and a positive correlation was found between ground cover and biomass.

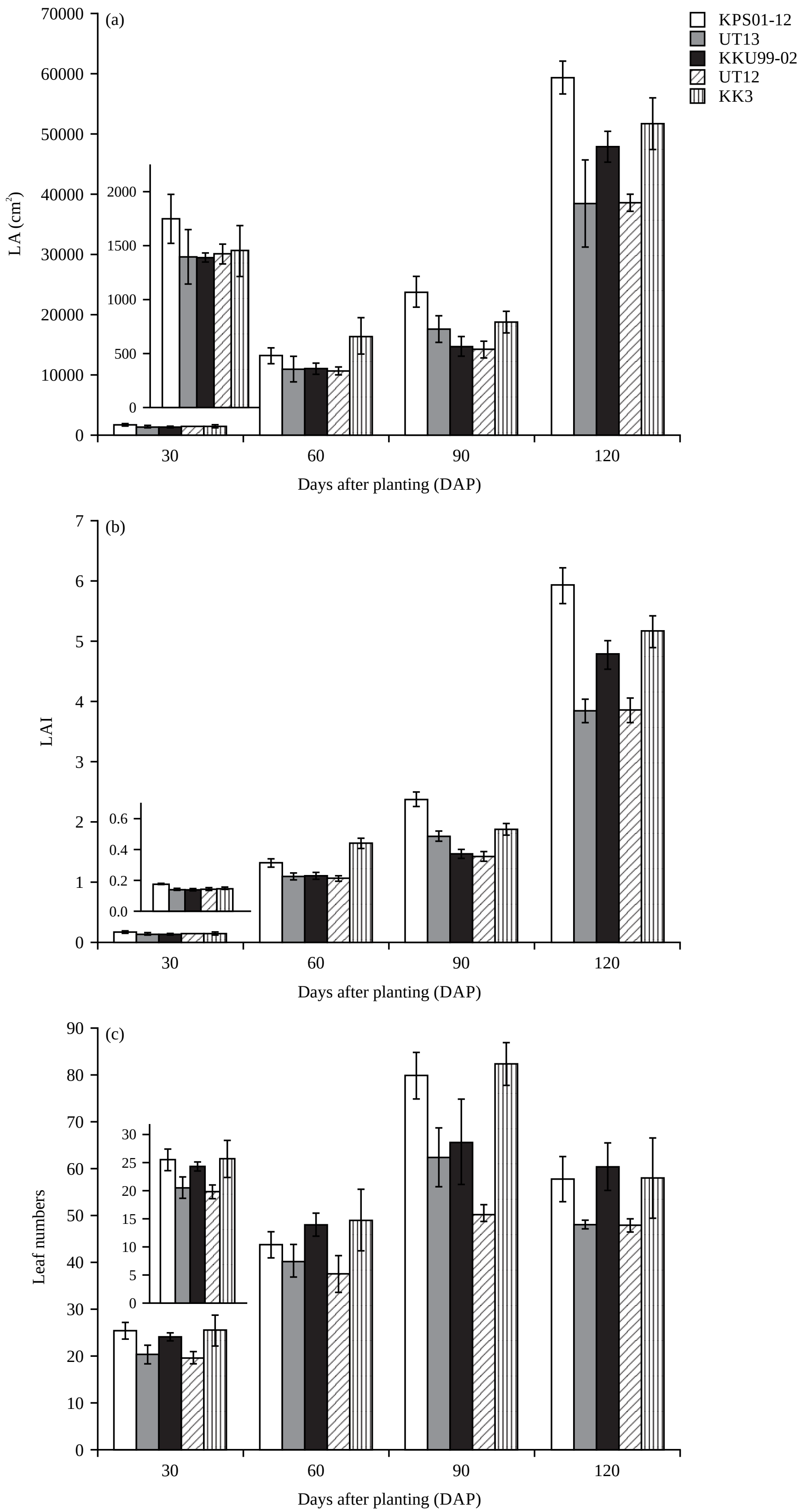
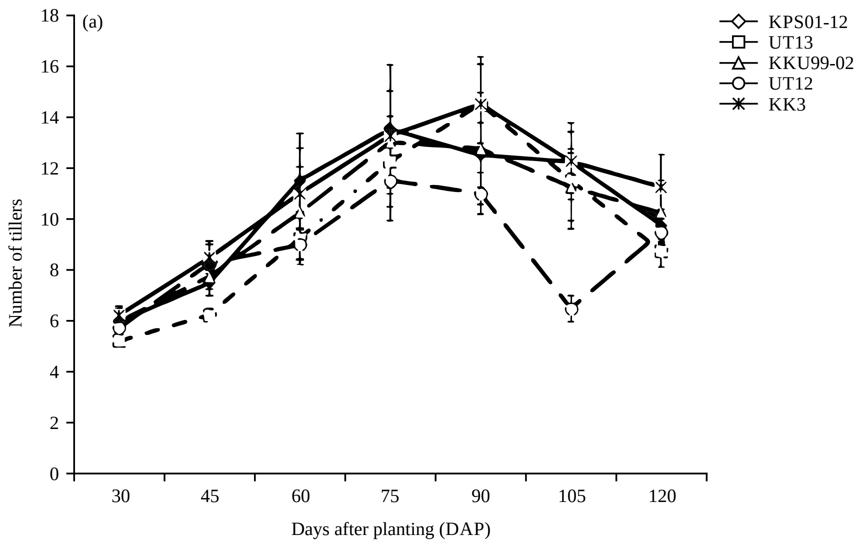


Fig. 2(a-b): Study of five cultivars for (a) Leaf area (LA), (b) Leaf area index (LAI) and (c) Leaf number at 30, 60, 90 and 120 days after planting (DAP)



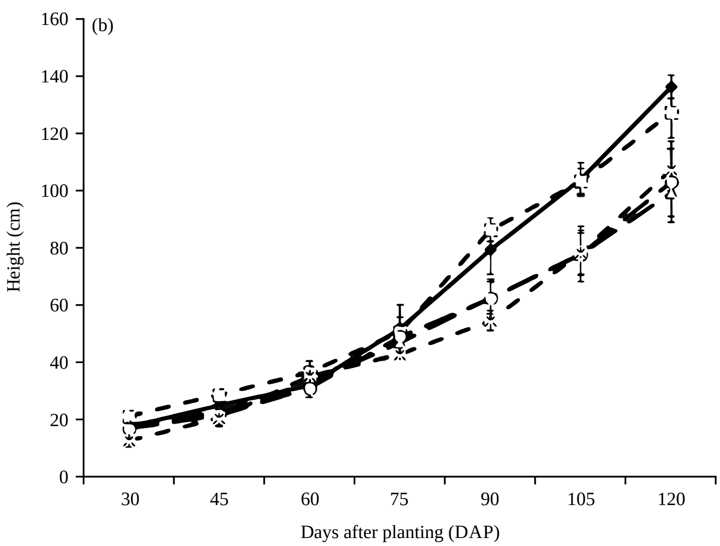


Fig. 3(a-c): Study of five cultivars for (a) Number of tillers and (b) Plant height at 30, 45, 60, 75, 90, 105 and 120 DAP

Table 1: Accumulation correlation of ground cover at 30, 60, 90 and 120 days after planting (DAP)

	Ground cover (%)			
	30 DAP	60 DAP	90 DAP	120 DAP
LAI	0.08	0.16	0.74*	0.72**
LA	0.08	0.16	0.74*	0.72**
Leaf number	0.15	0.71**	0.67*	0.29
LDW	0.38	0.33	0.53**	0.52**
SDW	0.33	0.38	0.17	0.34
Biomass	0.39	0.37	0.29	0.43*
Tillers number	0.28	0.48*	0.13	0.06
Height	0.34	0.08	0.45*	0.10

*,**Significant at 0.05 and 0.01 probability levels, respectively, DAP: Days after transplanting, LAI: Leaf area Index, LA: Leaf area, SDW: Stalk dry weight and LDW: Leaf dry weight

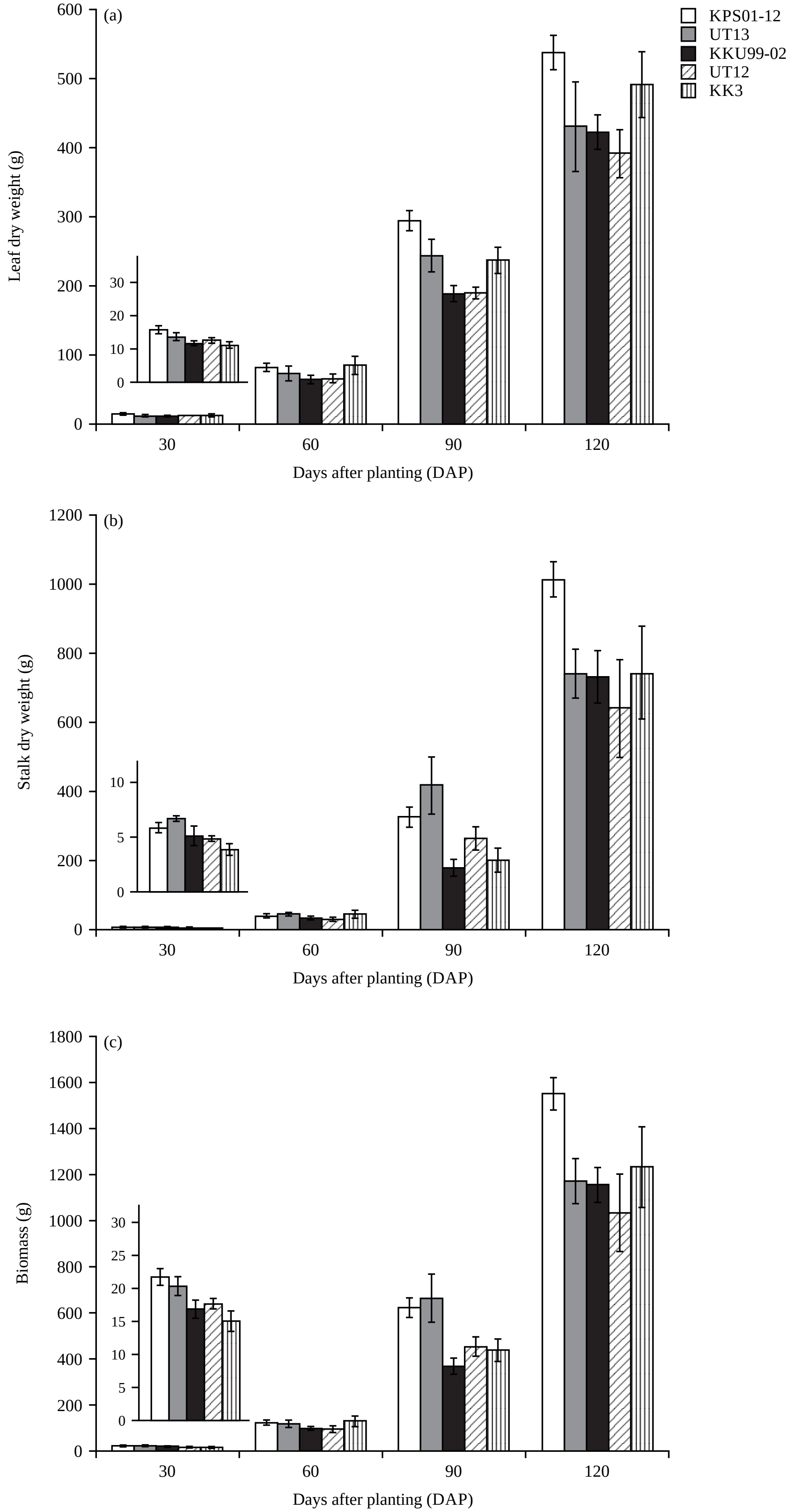


Fig. 4(a-c): Study of five cultivars for (a) Leaf dry weight, (b) Stalk dry weight and (c) Biomass at 30, 60, 90 and 120 DAP

DISCUSSION

Sugarcane cultivars in this study had different ground coverage in terms of percentage of ground cover and speed (Fig. 1). Sugarcane cultivar that had a large canopy in formative stage can support radiation use and canopy photosynthesis. The physiological feature of ground cover has a significant impact on crop radiation absorption, water usage effectiveness and yield[25]. The area of soil surface that is covered by plants as viewed from a nadir position is referred to as ground cover and can be measured in percentages or fractional units[26]. Although the ground cover is a crucial determinant of rangeland health, traditional methods for assessing ground cover require a lot of labor. There is a tendency for total leaf area to decrease after reaching maximum canopy development and the percentage of ground cover is also reduced due to the change of overlapping between leaves[16]. Moreover, the mortality of the leaves and tillers during the formative stage might affect the ground cover percentage (Fig. 1). The photosynthetic output of plants is ultimately dependent on the green leaf surface[27]. The cultivars that had high LAI produced high biomass as well. From this current result, LAI increased followed the crop duration, revealing the same performance with leaf number, but not LA. The lower leaves, which were older, were induced to senescence sooner by the reduction of light interception[28]. However, even though there was mortality in the leaves, the LAI still increased until the plant reached the mature stage[29].

Tillers changed following the developmental growth stages and depended on genetics and environmental factors[30]. The number of tillers constantly increased during the germination phase (Fig. 3). However, at 105 DAP, the tillers decreased because of tiller mortality from the effect of competition. In sugarcane, the mortality rate of tillers is up to 50–60% due to the variety, tiller class, time of planting and cultural conditions[31,32]. In contrast, the plant height of these studied cultivars constantly increased with increasing time since planting. This agreed with previous reports that revealed the height at this stage[33-35].

In this study, stalk dry weight contributed more to partitioning biomass accumulation than leaf dry weight. This result was corroborated by Kamble and Kharate[36], who showed that stem dry weight was approximately three times higher than green leaf dry weight. Dry weight can be affected by several factors, such as drought, variety, water and nutrient uptake[37,38]. Increasing biomass means increasing sugarcane yields because they are interrelated[39,40].

There was no correlation between ground cover and LAI at the early stage of sugarcane. This meant that the use of image analysis on the ground cover did not correspond with LAI at 30 and 60 DAP, but the ground cover and LAI were related at 90 and 120 DAP. The crop canopy efficiency, representing light interception, can be estimated by comparing the slopes of the LAI and ground cover correlation[22]. Moreover, the leaf growth rate, involved with LAI and light interception, was related to ground coverage[41]. These current results agreed with Haverkort *et al.*[42], who reported that there was a good correlation between LAI and ground cover during the early growing season before the ground cover reached 100%. The application of image analysis in ground cover has been proven as an alternative non-destructive way to study the ground coverage in many crops, such as the use of image analysis to determine the value of ground cover for various types of wheat, grass, potato and cotton[41,43,44]. However, the tiller number revealed no correlation with ground cover. This might be due to the mortality decreasing the number of tillers. Increasing LAI causes more shading and tiller mortality, as well as a slower tillering rate[45-47]. Therefore, there was no relationship between ground cover and tiller number at 90 and 120 DAP.

In this study, the methodology to evaluate the ground coverage using the high-angle image of diverse sugarcane cultivars was feasible to identify the variation between the cultivars in both magnitude and speed. Moreover, the output from the high-angle image is also related to growth traits that are usually measured during destructive sampling in some developmental periods, indicating that it was an alternative measurement to study the different growth metrics of sugarcane in the formative stage.

CONCLUSION

The five cultivars in this study had different ground coverage. The ‘KPS01-12’ was identified as the fastest ground-covering cultivar and UT-13 had a moderate ground coverage rate, whereas UT-12 and ‘KK3’ were slower cultivars. The fastest ground cover cultivar, ‘KPS01-12’, also revealed outstanding high values during destructive growth sampling, namely LAI, LA, leaf dry weight, stalk dry weight and biomass. Correlation between ground cover and LAI and leaf dry weight existed at 90 and 120 DAP in multiple sugarcane cultivars, but there was no correlation at 30 and 60 DAP. As a result, the measurement of ground coverage using a high-angle image is feasible to use as an indirect measurement of destructive growth traits in some formative developmental periods.

SIGNIFICANCE STATEMENT

This study discovers the measurement of ground coverage using a high-angle image is feasible to use as an indirect measurement of destructive growth traits in some formative developmental periods that can be beneficial for supporting further sugarcane research. This study will help the researcher to uncover the research as a non-destructive criterion in the evaluation of diverse sugarcane cultivars that many researchers were not able to explore. Thus, the alternative high throughput measurement for further sugarcane research may be arrived at.

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