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Effects of Harvest Time on Medicinal Qualities of Hemp

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Abstract

Harvest time impacts the physicochemical properties of hemp. This study investigated the relationship between harvest time on growth parameters, physiological parameter, and color, in addition to Cannabidiol (CBD), Tetrahydrocannabinol (THC) and Cannabigerol (CBG) concentrations. Siskiyou cannabis was harvested at 5 different stages after flowering 50% at week five to week nine. The rational explanation for stages on growth parameters, physiological parameters, color, and total physiochemical properties was found, while total color changes ranged from brightness (L*) decreased gradually from 66.06 at 5 weeks to a minimum of 16.14 at 9 weeks of flowering. On the contrary, the redness (a*) and yellowness (b*) increased from –0.65 and 8.95 at 5 weeks to their peak values of 19.99 and 34.24 at 9 weeks, respectively. The hue also decreased from 110.03 at 5 weeks to a minimum of 80.14 at 9 weeks, with samples being significantly ($p < 0.05$) different. Tetrahydrocannabinol (THC) was higher than 1 % at week 5, which was lower than their Cannabidiol (CBD) concentrations, reaching 14.35% at week 8. Cannabigerol (CBG) in dried samples reached 2.01% at week 7. The average dry weight of inflorescence per plant peaked at 36.75 g in week 8 and week 9 respectively. Significant differences in Crop Growth Rate (CGR) were noted across the harvesting periods, notably 17.13 and 16.70 g cm^{-2} day^{-1} at weeks 8 and 9, respectively, representing the highest dry weight accumulation per unit area. This increase in dry weight accumulation indicates higher efficiency. Finally, the Harvest Index (HI) showed notable discrepancies among the post-flowering harvesting times, with the greatest total dry weight observed at 0.357 and 0.345 at weeks 8 and 9, respectively. These findings could be of industrial relevance for improving post-harvest processes while maintaining the quality of this regulated crop.

Keywords**:** *Hemp; cannabidiol content; Cannabis sativa L.; harvested time*

1. Introduction

Hemp, *Cannabis sativa* L. is a short-day plant. The flowering is induced by short days and is genetically controlled. However, the actual time of inflorescence initiation is modified by weather, growth conditions, light, humidity and management practices (Kozlowski, & Pallardy, 1996; Lisson, & Mendham, 2000; Pallardy, 2010). For consistent phytochemical profiles in hemp, which is crucial in herbal medicine, standardization of hemp is

imperative. This ensures uniform cannabidiol (CBD), tetrahydrocannabinol (THC) and cannabigerol (CBG) compositions, leading to stable medical efficacy. Optimizing and standardizing growth conditions and agronomic practices become pivotal for yield maximization (Janatová et al., 2018).

The cultivation of active pharmaceutical ingredients (APIs) for medical applications necessitates the standardization of product quality, such as cannabidiol (CBD), and cultivation processes (Danziger, & Bernstein, 2021). As a result, medicinal hemp is often grown in indoor and greenhouse systems to gain better control over environmental conditions, facilitating a higher level of standardization in cultivation processes. This is particularly crucial for managing photoperiodism and temperature, especially in tropical and temperate regions, to enable year-round cultivation.

The significance of optimizing indoor cultivation systems has grown due to the increasing demand for maximizing yield and improving the efficiency of the growing process (ElSohly et al., 2017). The final yield quantity is highly variable and depends on several factors, including genotype and agronomic practices. The therapeutic potential of the harvested material is greatly influenced by the initial quality of the hemp plant, determined by factors such as harvesting time, harvesting technique, and post-harvesting technologies. These elements collectively shape the chemical composition and quality of the end products. Safety remains a paramount concern in this process (Burgel et al., 2009; Burgel et al., 2020; Reichel et al., 2021).

The concentration of cannabidiol (CBD) exhibits a notable contrast between unripe and ripe buds. Optimal potency is achieved when the hemp reaches maturity, making the ripe stage the ideal time for harvesting in hemp cultivation. To ensure the highest quality harvest for processing, a meticulous approach involves daily inspections of the influence and dedicating additional time to conduct multiple harvesting sessions. The subsequent section will elaborate on the methods for determining the optimal harvest time and selecting the best cannabidiol (CBD) concentrations for preserving medicinal hemp qualities using effective harvesting technology (Crispim Massuela et al., 2022).

2. Objectives

The study aimed to investigate the effect of harvest times on growth parameters and the optimum total cannabidiol (CBD) content in hemp.

3. Materials and methods

3.1 Plant materials

Cannabis plant seedlings (*Cannabis sativa* L. cv. Siskiyou) (Siskiyou-Sanfansico, USA) were cloned from standardized stock plants. This cannabis strain was chemotype III genotype, which was purchased from ACC CANNABIS CO., LTD. Pattaya city, Bang Lamung district, Chonburi, Thailand, in August 2022.

3.2 Planting location

The experiment was conducted from October 2022 to March 2023. An indoor growing experiment was conducted at ACC cannabis plant factory (Chonburi, Thailand). Cannabis was grown legally under license No. 36/2565 (G) which was approved by Food and Drug Administration (FDA), Ministry of Public Health, Royal Thai Government in 2022s.

3.3 Growing conditions

The experiment was arranged in a randomized complete block design with four replications. 16 plants were grown for each replicated. Harvesting times was an experimental treatment which was varied in five harvesting stages. Harvesting stages were 5 (Wk.5), 6 (Wk.6), 7 (Wk.7), 8 (Wk.8), and 9 (Wk.9) weeks after 50% of flowering stage, respectively. The experimental treatments were placed in a row-column pattern with 16 rows and 5 columns that followed in Figure 1.

Experimental plants were produced from standardized plant seedling. 80% of relative humidity levels was controlled by humidifier machine with air circulation system. seedlings were grown in biodegradable plastic nursery bags (3.5 cm ×7 cm × 3.5 cm) filled with a mixture of 20% perlite, 20% vermiculite, 40% soil, and 20% peat moss under nurseries condition (Zheng, 2019). The first day after planting (DAP) is regarded as the start of the experiment after the plants had been transferred into plastic plant pots for 14 days. The plants were moved to round pots using the same substrate composition after 7 DAP. A total of 250 g of media was added to the soil mixture for the first and second reports. With a density of 20 plants per square meter, the pots were arranged in four rows, each with 16 colonial horticultural tables (1 × 4 m). Weeks to flowering stages from 50% emergence.

At the vegetative growth, two pruning techniques were applied. First, 30 days after planting (DAP), topping involved an apical cut at the main stem. Second, lollipop pruning involved removing

branches and apical growth for branch control which was conducted at 45 DAP. At the flowering stage, the lowest branches were removed, controlling 8 branches per plant.

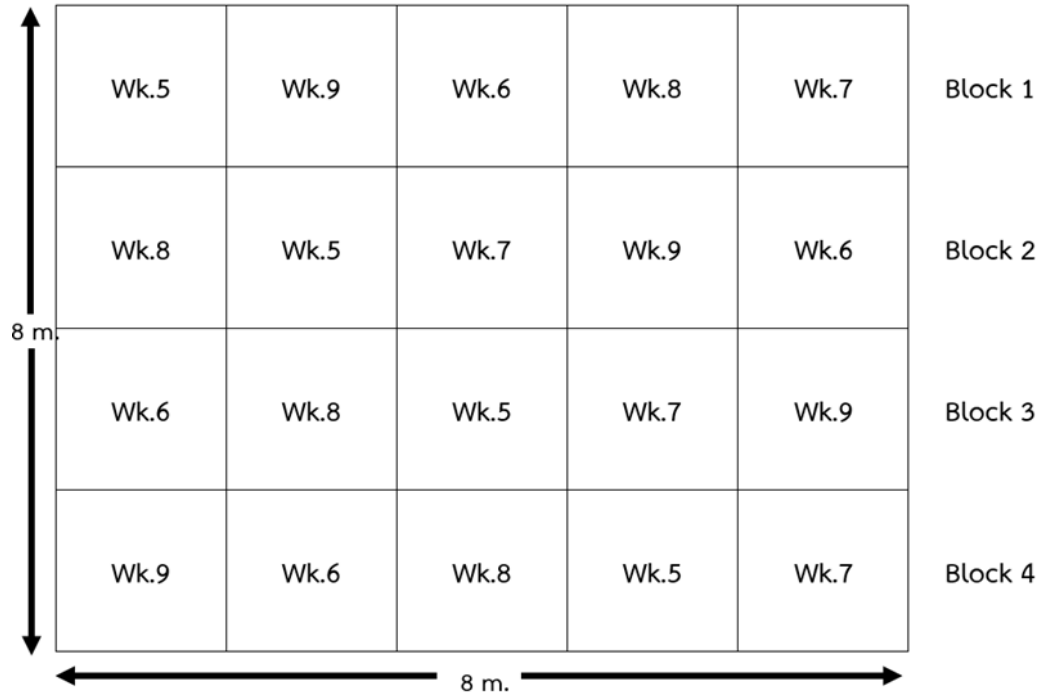

Figure 1 Diagram of the experiment arranged in randomized complete block design

Notes: Wk. 5, Wk. 6, Wk. 7, Wk. 8, and Wk. 9 were experimental treatments that varied in harvesting time for 5, 6, 7, 8, and 9 weeks after flowering 50%, respectively.

3.3.1 Light

Red, green, blue, and infrared (IR) LED fixtures with adjustable spectra (1000 w 4-bar LEDs) were used for vegetative growth with 18 hours of continuous light, while a 12-hour light and 12-hour dark schedule were utilized to initiate flowering.

3.3.2 Temperature

Cultivation room temperature was controlled, seedlings were grown at a constant temperature of 23°C, while the temperature ranged from 23°C to 25°C during the vegetative and flowering stage.

3.3.3 CO_2 concentration

CO_2 concentration ranged from 800 to 1500 μmol/m^2 /PPFD and was supplied from vegetative stages until the end of the flowering stage (Gaffney, 1996; Hendry, & Grime, 1993).

3.3.4 Irrigation system and humidity

A drip irrigation system with a controller provided a constant water supply of 100–500 ml per day. Relative humidity was controlled at 75% for seedling stages and between 55% to 60% for vegetative and flowering stage was controlled by humidifier machine with air circulation system.

3.3.5 Fertilization

Different concentrations of N, P, and K were applied three times a week. Dilution concentrations were followed as per the producer's recommendation, with a specific N-P-K ratio of 4:1.3:1.7. The electrical conductivity (EC) ranged from 0.9 to 3.9 mS/cm, and the pH in irrigation water was maintained between 5.8 and 6.0 (Caplan et al., 2017; Zheng, 2022).

3.3.6 Harvest techniques

The harvesting times followed experimental treatments which focused on the later stages of flower maturation, from 5 to 9 weeks after 50% flowering stage. Harvested plants were divided into four parts: the stem, leaves, roots, and flowering tops. Samples were collected separately from the top, middle, and bottom (lower) parts of one in four plants to examine

intra-plant variation in the concentration and overall yield of cannabidiol (CBD), referred to as “flowering top position” (Crispim Massuela et al., 2022).

3.3.7 Drying techniques

The cool drying technique was applied to dry plant samples. Plant samples were placed in a well-ventilated room equipped with an environmental temperature control system. The drying temperature was set between 18°C and 21°C. Relative humidity was controlled between 50% and 55%. Air circulation was provided by circulating fan. Plant samples were dried until moisture content reached 10% - dry basis (Coffman, & Gentner, 1974; Ross, & ElSohly, 1996; Challa et al., 2021).

3.4 Data collections

3.4.1 Physiological parameter

Number of leaves, plant height, and stem diameter were recorded weekly. Plant photosynthesis was measured by LCpro T Advanced Portable Photosynthesis System (ADC Bio scientific Ltd.) (Tobiasz-Salach et al., 2021; Priya et al., 2022). Leaf greenness or chlorophyll content (SPAD) was measured by SPAD-502Plus model by Centasia®. Plant stress value was measured by Pocket PEA Chlorophyll Fluorosensor model by Hansatech®.

3.4.2 Growth parameters

1) Leaf Area Index (LAI):

The leaf area index (LAI) was calculated following Williams (1946) using equation (1):

$$LAI = LA/Ga \quad (1)$$

where LAI is the leaf area index, LA is the leaf area, and Ga is the ground area covered by the plant.

2) Leaf Area Ratio (LAR)

Leaf Area Ratio (LAR) analysis of leaf dispersion in the canopy is the ratio of leaf area to light assimilation distribution. This ratio indicates the amount of foliage in each square centimeter per gram. It was calculated according to Sampet (1999) using equation (2):

$$LAR = LA / Wl \quad (2)$$

where LAR is the leaf area ratio, LA is the leaf area, and Wl is the dry weight of the leaves.

3) Net Assimilation Rate (NAR)

The Net Assimilation Rate (NAR) is calculated by dividing the total dry weight by the leaf area and the time interval. NAR indicates the efficiency of light absorption or photosynthesis per leaf. It was calculated according to Watson (1958) using equation (3):

$$NAR = Wl / LA \quad (3)$$

where NAR is the net assimilation rate, Wl is the dry weight of the leaves, and LA is the leaf area.

4) Crop Growth Rate (CGR)

The Crop Growth Rate (CGR) is calculated by dividing the total change in dry weight by the planting area and the time interval. CGR serves as an index indicating the rate of dry weight accumulation of plants per unit area per unit time. It was calculated according to Sampet (1999) using equation (4):

$$CGR = NAR \times LAI \quad (4)$$

where CGR is the crop growth rate, NAR is the net assimilation rate, and LAI is the leaf area index.

5) Harvest Index (HI)

The Harvest Index (HI) was calculated using the formula provided by Sampet (1999);

$$HI = \text{Economic yield} / \text{Biological yield} \quad (5)$$

where HI represents the harvest index, the economic yield refers to the dry weight of the cannabis inflorescence, and the biological yield is the total above-ground plant biomass weight excluding the dry weight of the inflorescence.

3.4.3 Yield

Four plant samples were harvested at the base of the stem then each plant was separated into four parts: stems, leaves, roots, and inflorescences, respectively. Additionally, inflorescence samples were taken from the top, middle, and low parts of the plant. Then, inflorescence samples were dried by the cool drying technique until moisture content reached 10%. Finally, all plant samples were kept in plastic sealed bag and then stored in cool conditions (5°C to 10°C) before sampling for medicinal quality analysis (Crispim Massuela et al., 2022).

3.4.4 Cannabidiol (CBD), Tetrahydrocannabinol (THC) and Cannabigerol (CBG) Analysis

The dried inflorescence sample was submitted for analysis of Cannabidiol (CBD), Tetrahydrocannabinol (THC), and Cannabigerol (CBG) using the Near Infrared Spectroscopy (NIRS) technique, which conducted by GemmaCert Cannabis Analyzer Professional, GemmaCert®.

3.4.5 Color analysis

Cannabis inflorescence color was detected by ColorFlex EZ and then color analyzed by EasyMatch® QC Electronic Record (ER) software. Color data, color plots, spectral data, and spectral plots of samples were analyzed, respectively.

3.4.6 Statistical analysis

Analysis of variance (ANOVA) and least significant different (LSD) tests with $\alpha = 0.05$ were analyzed by R version 4.4.1 (R-tools Technology Inc., Málaga, Spain).

4. Results and discussion

4.1 Growth parameters

LAI, LWR, and NAR were significantly decreased from Wk. 5 to Wk. 9. Conversely, RGR, CGR, and HI were increased (Table 1).

The study showed significant differences in LAI during five harvesting times, with the highest LAI observed at 5 and 6 weeks after 50% of flowering (2.21 and 1.95 cm^2, respectively). It underscored the importance of considering both size and quality of the source. Larger LAI did not necessarily translate to an optimum LAI. The optimum LAI promoted optimal light interception for photosynthesis, while excessive LAI might result in shading effects. Moreover, LWR discussed how photosynthate was translocated for leaf area formation, relative to generating plant dry matter accumulation (De Oliveira, 2019). The significantly decrease of LWR among the five harvesting times indicated that the cannabis plants were undergoing senescence. Plant senescence decreased in leaf function that correlated to a decrease in photosynthesis and dry matter accumulation (Pallas et al., 1967). This result referred to a significant decrease in NAR. This highlighted the decreased efficiency of cannabis plants in assimilating light energy, which is vital for growth and development, with implications for sink-source dynamics. Plant photosynthesis and dried matter accumulation decreased, affecting the decrease of overall plant yield productivity in both quantity and quality (Aslani et al., 2020; Norman et al., 2011). This result showed that optimum harvesting time before plant senescence was most emphasized that could maintain in both plant yield quantity and quality.

RGR, CGR, and HI significantly increased during the observed harvesting time. These results showed that leaf function promoted light interception and photosynthetic efficiency, leading to increased dry matter accumulation efficiency. The maintenance of dry matter accumulation could promote plant yield production that related to increase of HI (De Oliveira, 2019). It was predicted that cannabis yield would exhibit a saturating response to optimum of light interception, while decreased of optimal light interception would affect cannabis yield productivity (Rodriguez-Morrison et al., 2021). These results emphasized the importance of harvesting time in maintaining optimal harvested yield.

Table 1 The effect of harvest times on Leaf area index (LAI), Leaf weight ratio (LWR), Net assimilation rate (NAR), Relative Growth Rate (RGR), Crop Growth Rate (CGR), and Harvest Index (HI)

Treatments	LAI	LWR ($cm^2\ g^{-1}$)	NAR ($g\ cm^{-2}\ day^{-1}$)	RGR ($g\ cm^{-2}\ day^{-1}$)	CGR ($g\ cm^{-2}\ day^{-1}$)	HI
Wk. 5	2.21±0.334^{a}	13.28±0.048^{a}	0.34±0.042^{a}	2.23±0.265^{b}	13.00±1.537^{b}	0.126±0.021^{c}
Wk. 6	1.95±0.381^{ab}	11.35±0.043^{a}	0.15±0.618^{b}	2.32±0.207^{b}	13.52±1.201^{ab}	0.134±0.020^{c}
Wk. 7	1.64±0.027^{b}	9.54±0.028^{bc}	0.11±0.789^{bc}	2.35±0.186^{b}	13.68±1.085^{b}	0.308±-.020^{b}
Wk. 8	1.47±0.086^{b}	6.53±0.510^{c}	0.09±0.027^{c}	2.94±0.148^{a}	17.13±0.863^{a}	0.357±0.014^{b}
Wk. 9	0.86±0.052^{c}	3.24±0.510^{d}	0.07±0.727^{c}	2.86±0.126^{a}	16.70±0.736^{a}	0.345±0.024^{b}
Mean	1.63	8.79	0.15	2.54	14.8	0.254
LSD 0.05	0.55*	3.92*	0.05*	0.30*	1.75*	0.27*
CV (%)	22.22	24.34	25.59	7.82	2.14	7.065

Notes: Wk.5, Wk. 6, Wk. 7, Wk. 8, and Wk. 9 represent harvesting time at 5, 6, 7, 8, and 9 weeks after 50% flowering, respectively. Difference letters in same column indicate statistically significant different at 95% level of confidence ($p<0.05$), ns indicates not statistically significant different at the 95% level of confidence

4.2 Plant photosynthesis and plant stress

Simultaneously, the impact of harvest times on leaf temperature, leaf greenness (SPAD), transpiration rate, sub-stomatal CO_2, and net photosynthesis rate showed a significantly decrease throughout the harvesting time. Conversely, plant stress did not show significant differences (Table 2). This result indicated that plant senescence decreased leaf area and leaf function, which affected photosynthesis reduction (Thomas, 2013). Understanding these relationships holds paramount importance for optimizing conditions conducive to plant growth, particularly in controlled environments or agricultural settings (Wall et al., 2023). Diligent monitoring and precise adjustment of these parameters can significantly enhance plant productivity, ensuring a more efficient and fruitful cultivation process. This result was confirmed by Rodriguez-Morrison et al., (2021), who reported that photosynthesis of cannabis plant decreased linearly from flowering to the harvesting time of cannabis inflorescence. Moreover, it is important to appreciate that light quality in range of PPFD represents an instantaneous light interception level that affected cannabis yield in both quality and quantity.

4.3 Yield

This result indicated that harvesting time significantly affected leaf function and photosynthesis efficiency, which most directly impact dry matter accumulation and yield productivity. The biomass of inflorescence, leaves, stems, roots, and the total weight per plant significantly decreased (Figure 1). The dry weight of leaves significantly decreased. On the other hand, the dry weight of stems and roots, and the size of inflorescences in both width and length, significantly increased (Table 3). However, this result showed that plant senescence during harvesting time decreased plant dry matter accumulation. These results correlated with the discussion above that the optimum harvesting time of cannabis plants could maintain plant dry matter accumulation. Increased dry matter accumulation promoted plant yield productivity (Crispim Massuela et al., 2022). This corresponds to the increased biomass of the inflorescence (Spitzer-Rimon et al., 2019).

Table 2 The effect of harvest times on plant stress rate, leaf temperature, leaf greenness (SPAD), transpiration rate, sub-stomata CO_2, and net photosynthesis rate

Treatment	Plant Stress	Leaf temperature (ºC)	SPAD (Unit SPAD)	Transpiration rate (µmol m^{-2} s^{-1})	Sub-Stomata CO_2 (vpm)	Net photosynthesis rate (µmol m^{-2} s^{-1})
Wk. 5	0.71	33.70±1.240^{a}	70.52±4.291^{a}	2.80±0.970^{a}	396.83±72.013^{a}	7.60±0.461^{a}
Wk. 6	0.71	31.95±0.140^{b}	63.27±4.129^{a}	1.35±2.266^{a}	365.50±3.012^{ab}	5.58±0.487^{b}
Wk. 7	0.71	31.15±0.139^{bc}	50.84±4.126^{b}	0.71±2.264^{b}	314.41±3.072^{b}	3.81±0.483^{c}
Wk. 8	0.71	30.50±0.242^{c}	35.54±6.217^{d}	0.44±0.226^{c}	314.08±24.031^{b}	2.46±0.651^{d}
Wk. 9	0.66	30.52±0.360^{c}	18.15±4.093^{d}	0.31±0.782^{c}	304.41±77.518^{b}	1.45±0.782^{e}
Mean	0.7	31.58	47.74	1.12	339.05	4.81
LSD 0.05	ns	0.95*	7.94*	0.58*	80.95*	0.52*
CV (%)	6.91	1.95	6.77	33.48	15.49	8.09

Notes: Wk.5, Wk. 6, Wk. 7, Wk. 8, and Wk. 9 represent harvesting time at 5, 6, 7, 8, and 9 weeks after 50% flowering, respectively. Different letters in same column indicate statistically significant different at 95% level of confidence ($p<0.05$), ns inditate no statistically significant different at the 95% level of confidence.

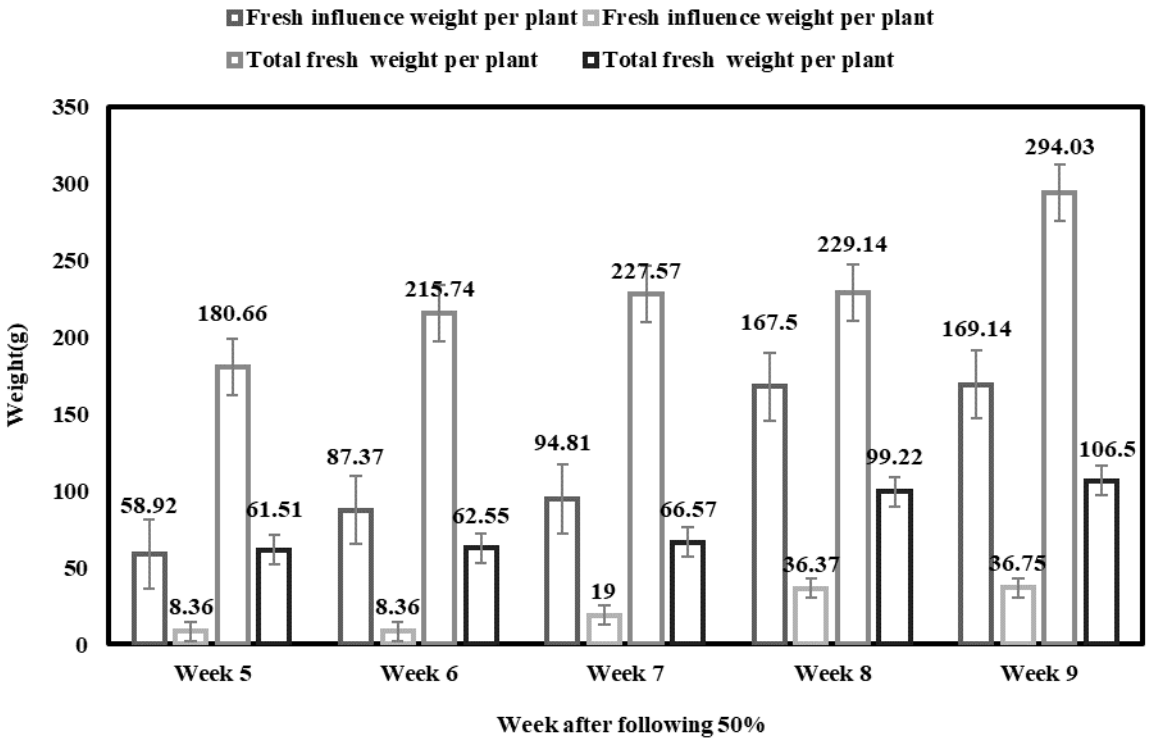

Figure 1 The effect of harvest times on total weight per plant and inflorescence weight per plant

Table 3 The effect of harvest times on leave, stem, root dry weight, inflorescence width and length

Treatments	Leave dried weight per plant (g)	Stem dried weight per plant (g)	Root dried weight per plant (g)	inflorescence width (cm)	inflorescence length (cm)
Wk. 5	15.07±6.976^{a}	26.20±8.210^{c}	5.63±0.647^{b}	1.92±0.340^{c}	1.42±0.853^{d}
Wk. 6	14.20±6.976^{ab}	37.43±8.207^{b}	5.88±0.530^{bc}	2.54±0.043^{c}	2.54±0.427^{c}
Wk. 7	11.12±2.707^{bc}	37.43±3.856^{b}	6.36±0.278^{c}	3.06±0.502^{b}	5.11±0.526^{a}
Wk. 8	9.97±2.187^{c}	38.39±5.300^{b}	11.25±0.841^{b}	3.07±0.483^{b}	5.11±0.510^{a}
Wk. 9	8.25±2.217^{c}	49.00±4.617^{a}	12.50±0.913^{a}	5.74±0.213^{a}	5.81±0.726^{a}
Mean	11.72	0.88	8.32	2.78	4.28
LSD 0.05	3.66*	0.20*	4.24*	0.43*	1.24*
CV (%)	20.32	29.57	20.32	10.21	6.45

Notes: Wk.5, Wk. 6, Wk. 7, Wk. 8, and Wk. 9 represent harvesting time at 5, 6, 7, 8, and 9 weeks after 50% flowering, respectively. Difference letters in same column indicate statistically significant different at 95% level of confidence ($p<0.05$), ns indicates no statistically significant different at the 95% level of confidence

4.4 Color

The color of cannabis inflorescence changed significantly during the harvesting period. The brightness (L*) decreased gradually from 66.06 at 5 weeks to a minimum of 16.14 at 9 weeks of flowering. On the contrary, the redness (a*) and yellowness (b*) increased from –0.65 and 8.95 at 5 weeks to their peak values of 19.99 and 34.24 at 9 weeks, respectively. The hue value also decreased from 110.03 at 5 weeks to a minimum of 80.14 at 9 weeks (see Table 4). Overall, as the flowering period progressed, the flowers became darker, redder, and more yellow (Figure 2). Chroma, however, remained relatively stable throughout the experiment.

Table 4 The effect of harvest times on inflorescence colors

Harvest time	L*	a*	b*	c*	Hue
Wk. 5	60.06±14.213^{ab}	-0.65±0.697^{b}	8.95±1.746^{b}	19.29	110.03±4.804^{a}
Wk. 6	61.63±2.298^{a}	-4.33±0.601^{c}	15.26±3.637^{b}	15.88	113.75±9.640^{a}
Wk. 7	54.42±2.442^{a}	1.87±0.552^{b}	18.08±0.566^{b}	9.77	88.15±2.526^{b}
Wk. 8	44.66±5.268^{c}	01.43±0.228^{b}	12.69±2.823^{b}	9.77	86.10±6.453^{b}
Wk. 9	16.14±6.984^{d}	19.99±6.314^{a}	34.24±14.194^{a}	9.77	80.14±13.762^{b}
Mean	47.38	7.85	1.59	11.3	100.19
LSD 0.05	6.31*	4.31*	0.42*	ns	10.19*
CV (%)	8.64	124.24	17.44	33.25	7.06

Notes: Wk.5, Wk. 6, Wk. 7, Wk. 8, and Wk. 9 represent harvesting time at 5, 6, 7, 8, and 9 weeks after 50% flowering, respectively. Difference letters in same column indicate statistically significant different at 95% level of confidence ($p<0.05$), ns indicate no statistically significant different at the 95% level of confidence

The overarching trends suggested that inflorescence color of the tested cannabis strain evolved towards red and yellow over time. This aligned with the maturation of trichomes during this period. At this stage, trichomes transitioned from transparent (Stage I) to white (Stage II) and finally to yellow amber/brown (Stage III). The component ratio underwent rapid changes during Stage I, which were too swift for optimal harvesting, potentially compromising the overall yield. The final stage (Stage III) was labeled as "over-ripening" due to trichomes turning brown trichomes associated with the late-stage aging of the plant or over ripening stage. This might result in a decline in inflorescence quality by converting THCA and CBDA into cannabinolic acid (CBNA) (Phummisutthigoon, & Kummalue, 2022). In literature, it is suggested that the change in coloration of pistils and trichomes is due to flower maturity and plant senescence, indicating that the plant is ready to harvest (Livingston et al., 2020; Tobiasz-Salach et al., 2021).

4.5 Cannabidiol (CBD), Tetrahydrocannabinol (THC) and Cannabigerol (CBG) concentration

The measurement of Cannabidiol (CBD) concentration, encompassing both CBD and CBDA, highlighted distinctions between unripe and ripe inflorescences. The total CBD concentration exhibited a steady increase from week 5, peaking at 14.35% during the 8th week of harvesting, and subsequently decreased to 12.37% by the 9th week. The trend in Cannabidiol (CBD) concentration displayed fluctuations throughout the harvesting period. Previous studies have proposed a decline in Cannabidiol (CBD) concentration as plants matured, potentially linked to reduced synthesis capabilities (Aubin et al., 2015). The observed decrease in concentration during harvesting may result from dilution due to the associated biomass of the inflorescences. The notable rise in CBD concentration during the harvest period indicated an increased exposure to Cannabidiol (CBD), potentially stimulating oxidation and decarboxylation processes in trichomes (Ryu et al., 2021). Conversely, sample preparation techniques that preserved Cannabidiol (CBD) in its acidic form may contribute to higher Cannabidiol (CBD) values (Crispim Massuela et al., 2022). Another study also studied with chemotype III plants reported that genotypes presented maximum concentration of total CBD by six weeks of flowering, generally reaching a plateau with consequent reduction of concentrations after ten weeks of flowering (Yang et al., 2020). However, some genotypes already presented a significant reduction in total CBD concentrations after seven weeks of flowering.

In a study by Yang et al., (2020), similar patterns were observed for total CBD concentration in different hemp genotypes containing Cannabidiol (CBD). All genotypes exhibited a consistent increase in CBD concentration, peaking during the seventh to eighth week of flowering, followed by a decline as plants aged. Concentrations varied from 2% to 12% in cultivated genotypes. However, the measured tetrahydrocannabinol (THC) concentration decreased from 1% at 5 weeks to a minimum of 0.5% in the last week. In contrast, the measurement of Cannabigerol (CBG) concentration increased from 1.4% at 5 weeks to a peak of 2.01% at 7 weeks, after which it decreased to a minimum in the last week (Table 5). Aizpurua-Olaizola et al., (2016) reported that CBGA reached a maximum concentration around five weeks of flowering and decreased afterward.

Table 5 Cannabidiol (CBD), Tetrahydrocannabinol (THC) and Cannabigerol (CBG) in cannabis inflorescence

Treatments	CBD	THC	CBG
	%	%	%
Wk. 5	10.60±1.332^{c}	1.00±0.171^{a}	1.40±0.000^{b}
Wk. 6	12.15±0.673^{c}	0.97±0.665^{b}	1.40±0.000^{b}
Wk. 7	13.02±0.704^{b}	0.42±0.504^{b}	2.01±0.491^{a}
Wk. 8	14.35±0.597^{a}	0.52±0.057^{b}	1.73±0.385^{ab}
Wk. 9	12.37±0.971^{b}	0.50±0.221^{b}	1.40±0.000^{b}
Mean	12.51	0.68	1.59
LSD 0.05	1.24*	1.75*	0.42*
CV (%)	6.45	2.14	17.44

Notes: Wk.5, Wk. 6, Wk. 7, Wk. 8, and Wk. 9 represent harvesting time at 5, 6, 7, 8, and 9 weeks after 50% flowering, respectively. Difference letters in same column indicate statistically significant different at 95% level of confidence ($p<0.05$), ns indicates no statistically significant different at the 95% level of confidence.

5. Conclusion

This study investigated the effect of harvest times on growth parameters, physiological parameters, and concentrations of Cannabidiol (CBD), Tetrahydrocannabinol (THC) and Cannabigerol (CBG). The study examined the impact of different harvesting times on various parameters of cannabis plants, showing significant differences in leaf area, Leaf Area Ratio (LAR), Net Assimilation Rate (NAR), Crop Growth Rate (CGR), and Harvest Index (HI) among different harvesting periods. Additionally, physiological parameters such as sub-stomatal CO_2 exchange rate, transpiration rate, Photosystem II efficiency, SPAD values, and leaf temperature were affected by harvesting time. The study also highlighted changes in plant biomass, leaf size, and flower color over the flowering period. Moreover, variations in Cannabidiol (CBD) concentration were observed, with concentrations peaking during the 8th week of harvesting. These findings emphasize the importance of timing in optimizing cannabis cultivation for desired outcomes. These findings provide new and important insights for the cannabis industry, indicating that optimal harvesting time significantly affects the quantity and quality of harvested cannabis inflorescence. Optimizing post-harvest time not only impacts cannabis yield but also affects the direct and indirect process costs in the cannabis industry. More research on the effect of harvest time on secondary metabolites should be conducted to further explore other Cannabidiol (CBD) derivatives as well as terpene compounds that are most required by markets and influence cannabis product pricing.

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