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Performance of industrial hemp cultivars across U.S. Midwestern environments: evidence from multi-location trials in Missouri

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Introduction: Industrial hemp (*Cannabis sativa* L.) is a crop of considerable industrial, environmental, and economic importance due to its multipurpose applications and sustainability potential. However, following an initial surge in interest after legalization in the United States, hemp production has declined in recent years, highlighting the continued need for region-specific adaptability and cultivar performance data, particularly in the U.S. Midwestern production region.
Methods: Thirty-two industrial hemp cultivars, comprising fiber-type and dual-purpose genotypes, were evaluated across three agro-ecological regions in Missouri over three consecutive growing seasons (2021–2023). Key agronomic traits, including emergence rate, plant height, stem diameter, biomass, flowering time, and fiber yield, were assessed to determine cultivar suitability under southern U.S Midwestern growing conditions. An optimized high-performance liquid chromatography (HPLC) method was employed to rapidly and precisely quantify eleven cannabinoids, including Δ9-tetrahydrocannabinol (Δ9-THC) and cannabidiol (CBD).
Results: Substantial variability in cultivar performance was observed across locations and years. The fiber cultivar Jinma consistently exhibited robust establishment, high biomass production, and superior fiber yields ranging from 6 to 12 Mg ha^{-1}, although it displayed relatively late flowering. Among dual-purpose cultivars, Futura 83 demonstrated a favorable balance of relatively early flowering, vigorous growth, and competitive fiber and seed yields, indicating suitability for diversified production systems. Cannabinoid analysis confirmed that all cultivars maintained total THC concentrations well below the legal threshold of 0.3%, with minimal variation across environments.
Discussion: The findings provide critical insights into genotype × environment interactions and confirm the genetic stability of cannabinoid profiles across diverse agro-ecological conditions. Jinma, Futura 83, and Fibror 79 were identified as top-performing cultivars with strong potential for commercial

production under Missouri's climatic conditions. These results support strategic cultivar selection and advance efforts toward the reintroduction and expansion of industrial hemp production in the U.S. Midwest, while also providing a valuable foundation for broader multi-state comparisons.

KEYWORDS

cannabinoids, genotype × environment (G×E), industrial hemp, multi-location trials, U.S. Midwest

Introduction

Industrial hemp (*Cannabis sativa* L.) gained renewed attention following its relegalization in the U.S., but domestic production and coverage area has since contracted. In contrast, the crop continues to experience a global resurgence due to its adaptability, environmental benefits, and wide-ranging industrial applications, highlighting the need for regionally adapted, data-driven cultivar evaluations to support sustainable adoption. As low-THC chemotype of *Cannabis sativa* L. bred and cultivated for industrial purposes rather than psychoactive use, industrial hemp holds a deep-rooted historical significance in American agriculture and was once a dominant fiber crop throughout the United States (Darby et al., 2018). During the 19th century, Missouri, a key Midwestern state, emerged as a national leader in hemp cultivation, second only to Kentucky. By 1860, Missouri had produced more than 19,000 metric tons of hemp, accounting for approximately 26% of the total national production, with major production concentrated in counties such as Saline, Lafayette, and Pike. However, regulatory restrictions in the early 20th century led to a decline in its cultivation, resulting in a loss of agronomic knowledge and genetic resources (Rupasinghe et al., 2020). The 2014 U.S. Farm Bill marked a pivotal moment in the U.S history by allowing research institutions to initiate pilot programs for studying industrial hemp, laying the groundwork for broader legalization. Building on this foundation, the 2018 U.S. Farm Bill reintroduced industrial hemp as a legal agricultural commodity, fueling renewed interest in its potential for fiber, grain, and biomass production (Malone and Gomez, 2019).

Hemp's value lies in its versatility. It is a fast-growing, low-input crop that thrives in various agroecological regions, producing both bast and hurd fibers with distinct industrial applications (Cherney and Small, 2016; Darby et al., 2018). Its rapid growth and high biomass yield make industrial hemp an ideal candidate for atmospheric carbon fixation and sequestration, with the potential to capture up to four times more carbon dioxide than conventional crops such as corn (*Zea mays*), wheat (*Triticum aestivum*), and soybean (*Glycine max*) (Ahmed et al., 2022; Asiamah et al., 2025). Additionally, hemp's deep root system architecture enhances soil health, prevents erosion, and facilitates phytoremediation by absorbing heavy metals such as lead, cadmium, and nickel from contaminated soils (Asiamah et al., 2021; Placido and Lee, 2022). These traits make industrial hemp a valuable crop for sustainable farming systems, especially those focused on climate resilience, efficient resource use, and long-term environmental and economic sustainability.

Economic and environmental considerations further drive the demand for industrial hemp. With increasing concerns about climate change and dependency on petrochemicals, hemp offers a sustainable alternative for bioplastics, biofuels, and biodegradable materials (Modi et al., 2018). The economic potential of hemp farming is significant, as fiber hemp outperforms cotton in yield per hectare while requiring fewer pesticides and less water (Berg et al., 2014). Dual-purpose hemp cultivars are selectively developed to yield both industrial fiber and grain, maximizing economic value and versatility when adapted to appropriate agroecological regions (Tang et al., 2016). However, challenges remain in cultivar selection, as genetic variation among hemp cultivars affects fiber quality, cannabinoid content, and plant performance (Zhang et al., 2021). Despite its numerous advantages, including adaptability, sustainability, and diverse end-use applications, industrial hemp cultivation in the United States continues to face significant hurdles. These include limited agronomic guidance, complex regulatory frameworks, and the urgent need for robust breeding programs to enhance fiber and grain yields. Following decades of prohibition and regulatory decline, the recent revitalization of hemp has reestablished Missouri as a key player in the crop's resurgence. In this context, optimizing cultivation practices in historically significant regions like Missouri is not only timely but essential for realizing the full agronomic and economic potential of industrial hemp in the U.S. Extensive research across various regions of the world has highlighted the agronomic potential and sustainability of several industrial hemp cultivars, many of which show promise for adaptation in new growing environments such as Missouri. For instance, Futura, a dual-purpose cultivar from France, has demonstrated early flowering, high fiber content, and good grain yield (Amaducci et al., 2008; Lisson et al., 2000). Similarly, Santhica 27, also from France, is a monoecious fiber cultivar noted for its low cannabinoid content (CBD and THC) and excellent stem yield (Shah et al., 2024). In Eastern Europe, the Polish cultivar Bialobrzeskie has gained recognition for its cold tolerance, adaptability, and high seed oil production (Bócsa and Karus, 1998). Meanwhile, Jinma, a fiber-type cultivar from China, has shown exceptional performance in terms of plant height, stem thickness, and total biomass, particularly under field conditions in semi-arid west Texas conditions, USA (Bajwa et al., 2023). These findings underscore the need to evaluate these promising cultivars under local agroecological conditions to identify those best suited for sustainable production in this region (Adu et al., 2025).

This study aims to assess the performance of different use-type industrial hemp cultivars under the southern midwestern conditions, focusing on fiber yield, phenotypic variability, and biochemical composition. This study seeks to provide Missouri farmers with data-driven insights to enhance hemp production by evaluating genotype-environment interactions and refining biochemical assay protocols for cannabinoid profiling. Understanding the agronomic, genetic, and economic aspects of hemp will aid in optimizing crop selection and also contribute to the broader adoption of hemp as a sustainable resource for feed, fiber and industrial applications.

Materials and methods

Experimental site and planting material

The industrial hemp study was established at three locations in Missouri: George Washington Carver (GWC) farm in Jefferson City (Longitude: 38.5322° N; Latitude: - 92.1333° W), Sikeston Agri-Park in Sikeston (Longitude: 36.8831° N; Latitude:-89.5878° W) and Washington, Missouri (Longitude: 38.5581° N; Latitude: -91.0121° W) (Supplementary 1, Table 1). The experimental sites represented distinct agro-ecological conditions (Table 2, Figure 1). At Carver Farm, soils were classified as Menfro silt loam, well-drained, with a pH range of 6.0-6.5, and characterized by deep, loess-derived profiles with moderate organic matter. Sikeston soil was Alfisols (silty loam), well-drained, with pH 6.1-6.5, typical of the Missouri Bootheel row-crop region. Washington soils were loess-derived silty loam, moderately well-drained, slightly more acidic (pH 6.0-6.2), and exhibited variable moisture retention (Table 1). Thirty-two industrial hemp cultivars, consisting of both fiber and dual-purpose types, sourced from domestic and international seed suppliers were studied (Table 2). In the 2021 summer growing season, fifteen cultivars were selected and evaluated at two sites: GWC farm (Jefferson City) and Sikeston. In 2022, the study expanded to include twenty-two cultivars grown across all three locations. Based on performance data from the previous two years, the top eight cultivars were selected for further evaluation during the 2023 summer growing season.

Field experimental design

Industrial hemp cultivars were evaluated in a randomized complete block design (RCBD) with two replications per location (Tamang et al., 2025; Babaei and Ajdanian, 2020; Papastylianou et al., 2018). Prior to sowing, all field sites underwent standard land preparation, consisting of two passes of conventional tillage

TABLE 2 List of industrial hemp cultivars evaluated in the study.

Cultivar	Sexual type	Origin	Use	Evaluation year
Jinma	Diecious	China	Fiber	2021, 2022, 2023
Yuma	Diecious	China	Fiber	2022
Puma	Diecious	China	Fiber	2022
MS 77	Diecious	Australia	Fiber	2022
Fibror 79	Monoecious	France	Dual	2021, 2022, 2023
Futura 83	Monoecious	France	Dual	2022, 2023
Fibranova	Diecious	Italy	Fiber	2022
Santhica 70	Monoecious	France	Fiber	2022, 2023
Rajan	Monoecious	Poland	Fiber	2022
Uso 31	Monoecious	Ukraine	Dual	2022, 2023
Ferimon	Monoecious	France	Dual	2021, 2022, 2023
Felina 32	Monoecious	France	Dual	2022, 2023
Orion 33	Monoecious	France	Dual	2022
BVL1	Diecious	United States	Dual	2022
BVL2	Diecious	United States	Dual	2022
BVL4	Diecious	United States	Dual	2022
BVL5	Diecious	United States	Dual	2022
Gravity	Diecious	United States	Dual	2022
BVL-3	Diecious	United States	Dual	2022
Bialobrzeskie	Monoecious	Poland	Dual	2022, 2023
Tygra	Monoecious	Poland	Dual	2021, 2022
Altair	Monoecious	Canada	Dual	2021, 2022
Henola	Monoecious	Poland	Dual	2022
CFX-2	Monoecious	Canada	Dual	2021
CRS-1	Monoecious	Canada	Dual	2021
CFX-1	Monoecious	Canada	Dual	2021
Grandi	Monoecious	Canada	Dual	2021
Picolo	Monoecious	Canada	Dual	2021
Katani	Monoecious	Canada	Dual	2021
Joey	Monoecious	Canada	Dual	2021
Anka	Monoecious	Canada	Dual	2021
Hliana	Monoecious	Ukraine	Dual	2021

TABLE 1 Soil characteristics of agroecological sites in Missouri, U.S. selected for the study.

Location	Site	Soil series/ type	Drainage class	Soil pH range	Description
George Washington Carver farm (GWC)	38.5322° N; 92.1333° W	Menfro silt loam	Well-drained	6.0-6.5	Deep, loess-derived soil with moderate organic matter; classified as prime farmland.
Sikeston, MO	36.8831° N; 89.5878° W	Alfisols/silty loam	Well-drained	6.1-6.5	Typical of the Missouri Bootheel, it supports intensive row crop systems.
Washington, MO	38.5581° N; 91.0121° W	Loess-derived silty loam	Moderately well-drained	6.0-6.2	Slightly more acidic; more variable moisture retention across profile.

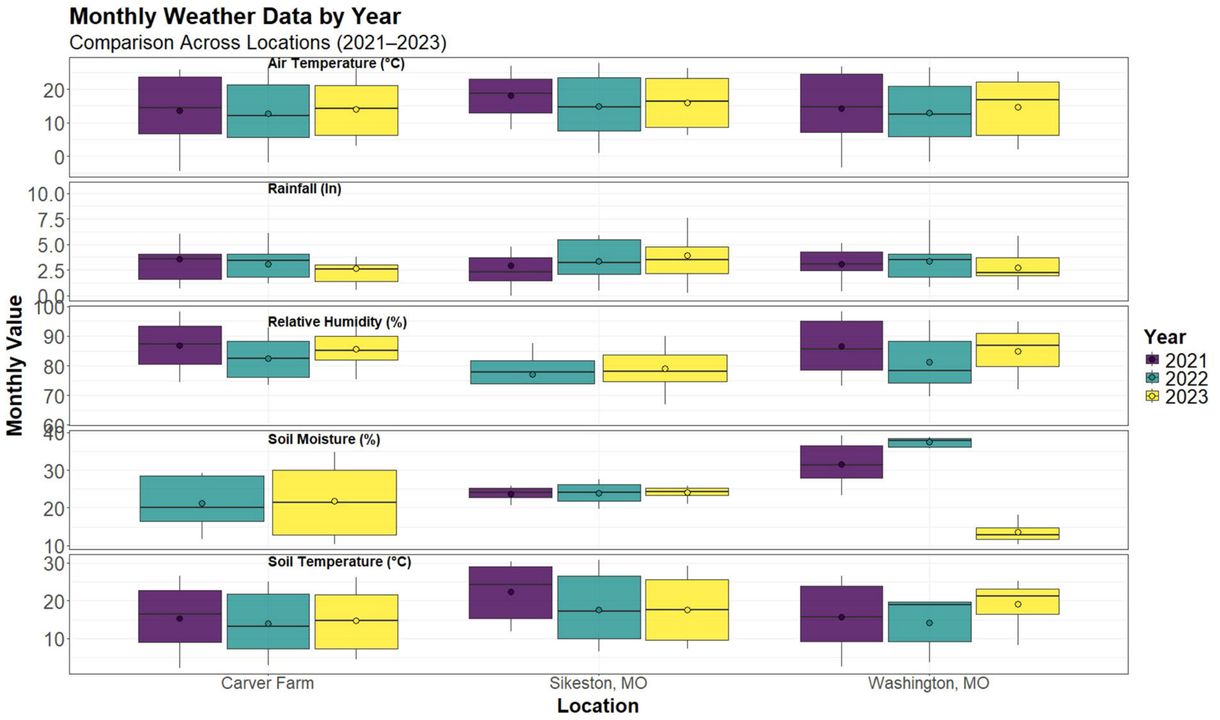


FIGURE 1
Monthly weather data (2021–2023) across three Missouri locations (GWC farm, Sikeston, and Washington). Panels show monthly distributions of air temperature (°C), rainfall (inches), relative humidity (%), soil moisture (%), and soil temperature (°C). Each box plot represents the distribution of monthly values, with median, interquartile range, and outliers displayed for each year and site.

followed by culti-packing to establish a uniform and firm seedbed. Basal fertilization was applied at a rate of 70-60–40 kg ha^{-1} of nitrogen (N), phosphorus (P), and potassium (K), respectively, in accordance with regional agronomic recommendations for hemp cultivation (Kaur et al., 2023; University of Missouri Extension,). Irrigation management was designed to simulate farmer field conditions, where cultivation is predominantly rainfed. Supplemental irrigation was only applied at critical growth stages (early vegetative, flowering, and seed set) when rainfall was insufficient or drought stress indicators were observed (leaf wilting, soil moisture depletion). Irrigation decisions were based on visual field assessments, with water requirement estimates informed by FAO modeling approaches (Food and Agriculture Organization (FAO), 1992) and supported by region-specific studies, such as Thevs and Aliev (2022), which reported ~350 mm seasonal water consumption for hemp under temperate conditions. Sowing was performed using a John Deere 1590 No-Till Drill, calibrated at a seed drop setting of 22 to accommodate the relatively larger seed size typical of industrial hemp. The seeding rate was approximately 56 kg ha^{-1} of viable seed, consistent with industry recommendations for fiber- and dual-purpose hemp production (45–65 kg ha^{-1}) (Cherney and Small, 2016; Giannoulis et al., 2024). However, following seed viability assessments (germination tests) conducted prior to sowing each year, cultivar-specific seeding rates were adjusted to compensate for differences in germination percentage (GP) (Table 3). Seeding rate was then calculated using the formula (International Seed Testing Association (ISTA), 2022):

$$\text{Adjusted Seeding Rate (kg ha}^{-1}\text{)} = \left(\frac{\text{Standard rate (kg ha}^{-1}\text{)}}{\left(\frac{GP}{100}\right)} \right)$$

Where:

Standard Rate = Target seeding rate for viable seeds (56 kg ha^{-1}).

Germination Percentage (GP) = percent viability from germination test.

Planting dates, plot dimensions, row spacing, and planting depth for each site and year are summarized in Table 4 to give an overview of the field setup used for industrial hemp trials conducted across three Missouri locations between 2021 and 2023.

In 2023, severe flooding events at the Sikeston and Washington locations affected planting activities, resulting in incomplete datasets from these sites. Accordingly, data from affected plots were excluded from subsequent statistical analyses.

Phenotypic data collection

Phenotypic traits were recorded throughout the growth cycle and included emergence rate, flowering time, plant density, plant height, stem diameter, biomass, and yield. Plant density was determined by counting all plants within the two 1-m^2 area within the plot (totaling 2 m^2 per plot). For plant height, twelve plants were randomly selected from each plot. Stem diameter was measured on the same twelve plants, with two measurements per plant: one taken 12.7 cm above the soil surface (basal diameter), and

TABLE 3 **Germination percentage of thirty-two industrial hemp (Cannabis sativa L.) cultivars evaluated during the study.**

Cultivar	Germination percentage (%)			Seeding rate (kg ha^{-1})		
	2021	2022	2023	2021	2022	2023
Altair	53.9	87.6	–	104	64	–
Anka	6.5	–	–	862.16*	–	–
Bialobrzeskie	56.7	92.2	81.1	98.86	60.8	69.16
BVL1	–	73.8	–	–	76	–
BVL2	–	56.4	–	–	99.4	–
BVL3	–	55.5	–	–	101	–
BVL4	–	76.7	–	–	73.1	–
BVL5	–	0	–	–	0.0	–
CFX-1	37.6	–	–	149.1	–	–
CFX-2	66.9	–	–	83.7	–	–
CRS-1	8.6	–	–	651.7*	–	–
Felina 32	–	80	80.7	–	70.1	69.5
Ferimon	78.7	80	80.5	71.2	70.1	69.6
Fibranova	–	24	–	–	–	–
Fibror 79	68.1	89.9	79.9	82.3	62.3	70.2
Futura 83	–	78.3	88	–	71.6	56.8
Grandi	1.00	–	–	–	–	–
Gravity	–	70.9	–	–	79	–
Henola	–	54.8	–	–	102.2	–
Hliana	57.5	–	–	97.5	–	–
Jinma	64.5	90.9	85.6	86.9	617	58.4
Joey	60.5	–	–	92.6	–	–
Katani	7.5	–	–	747.27*	–	–
MS 77	–	49.5	–	–	113.2	–
Orion 33	–	76.6	–	–	73.2	–
Picolo	28	–	–	200.2	–	–
Puma	–	71.5	–	–	78.4	–
Rajan	–	47.6	–	–	117.7	–
Santhica 70	–	48	85.8	–	116.8	58.3
Tygra	27.9	63.7	–	200.9	88	–
Uso 31	–	71.7	80.4	–	78	62.2
Yuma	–	44.5	–	–	126	–
LSD	1.858	2.18	2.188			
Grand Mean	41.55	64.468	73.311			

'*'- Adjusted to 200kg/ha during planting due to seed availability.
'–' indicates that seed was not available for planting during the respective experimental year.
NB: The conclusions regarding the potential influence of seed quality and handling were therefore based on the observed variation in germination rates between cultivars and across years, as well as background information from seed suppliers on storage and transportation practices.

the other 12.7 cm below the apical tip (upper diameter). Germination performance (GP) was evaluated under controlled greenhouse and laboratory conditions, without any pre-treatment of seeds (e.g., scarification or chemical priming). For each cultivar, 100 seeds were sown in three replications under uniform conditions. Germination percentage (GP) was then calculated using the standard formula (Gadissa et al., 2022; Shah et al., 2021):

$$\mathrm{GP} = \frac{Number\ \ of\ \ seeds\ \ germinated}{Total\ \ number\ \ of\ \ seeds\ \ tested} \times 100$$

Emergence rates were visually scored on a 1–5 scale (1 = 0%, 5 = 100%) between 5 and 14 days after planting. Flowering time was recorded when 50% of the plants within a plot exhibited visible flowers.

Biomass was assessed at full maturity by uprooting entire plants and recording their fresh weight. The samples were then dried for 2–3 weeks at 49 °C to obtain constant dry weight. Once a constant moisture content was achieved, the dry weight was recorded. Biomass percentage was then calculated using the formula:

$$\%\ \ Biomass = \frac{Dry\ \ Weight}{Fresh\ \ Weight} \times 100$$

Climatic data, including temperature, precipitation, and humidity, were collected throughout the growing seasons using on-site weather stations (Campbell Scientific Inc., Logan, UT, USA) (Figure 1).

Multilocation analyses and statistical approach

To evaluate cultivar performance across locations, we selected a core subset of eight industrial hemp cultivars from the total set evaluated. These cultivars were chosen because they consistently produced uniform stands and progressed through phenological stages at all three experimental sites, even under challenging environmental conditions (e.g., flood). Their stable performance across diverse environments made them suitable for valid and reliable multi-location comparative analysis. The analysis for this subset focused on key agronomic parameters, including stem diameter, plant height, plant density, whole-plant biomass, and fiber yield. To account for environmental variation across sites, data were normalized using Z-score standardization, following the method described by Cheadle et al. (2003), using the formula:

$$Z = (X_{\_Value} - \mu\_\mathrm{site})\ \ /\sigma\_site$$

Where:

- x value = observed value.
- μ_site = mean of the trait at a specific site.
- σ_site = standard deviation of the trait at specific site.

Agronomic and yield data were log-transformed prior to statistical analysis to normalize distributions and stabilize variances. However, original (non-transformed) values were retained for graphical presentations and comparison of cultivar trends. Statistical analyses were conducted using RStudio (version 2022.07.1 Build 554) and Minitab$^{®}$ version 22.1. The general linear model (GLM) procedure for analysis of variance (ANOVA) was used to test the effects of cultivar, location, year, and interactions. Means were separated using Tukey's Least Significant Difference (LSD) at a 95% confidence level (α = 0.05). Correlation coefficients

TABLE 4 **Planting dates, plot sizes, and seeding details for industrial hemp trials at three Missouri locations, 2021-2023.**

Study locations	Planting dates			Plot size (m^2)	Row spacing (m)	Planting depth (cm)
	2021	2022	2023			
GWC farm (Jefferson City), MO	25-May	11-May	5-Jun	92.9	0.2	1.3
Sikeston Agri-Park, MO	5-Jun	18-May	15-May	743.2 (2021 & 2022), 371.6 (2023)	0.2	1.3
Washington, MO	–	5-May	8-May	92.9	0.2	1.3

were computed according to the method described by Pearson (1895) (Figure 2).

Moreover, a *post-hoc* power analysis was performed to evaluate the ability of the experimental design to detect meaningful differences among cultivars. The analysis was conducted separately by site and year using residual mean square error (MSE) values obtained from one-way ANOVA models (trait- cultivar).

The minimum detectable difference (MDD) was calculated for plant density and biomass (kg m^{-2}) at 80% statistical power and a significance level of $\alpha = 0.05$ using the following equation:

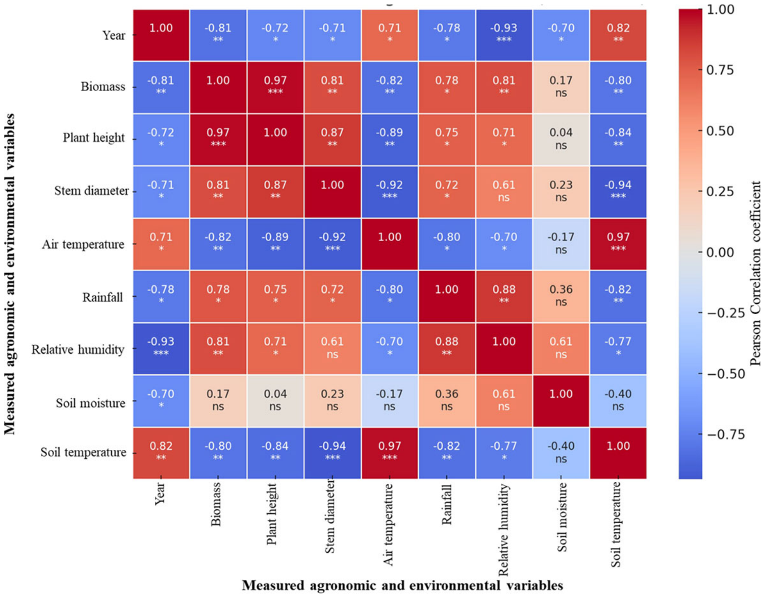
$$\text{MDD} = (t_{\alpha/2,df} + t_{\beta,df}) \times \sqrt{\frac{2 \times \text{MSE}}{n}}$$

where t values correspond to the student's t distribution, *df* represents residual degrees of freedom, and n is the number of replications per cultivar.

Cannabinoid testing

Fresh leaf samples from GWC farm-Jefferson City and Sikeston were collected and immediately flash-frozen in liquid nitrogen and stored at -80 °C until further analysis. The flash frozen samples were freeze-dried and ground into a fine powder. A sample size of 200 mg was used for extraction, with three solvent mixtures tested: 100% methanol, methanol: water (9:1), and methanol: acetonitrile (9:1) (Tzimas et al., 2024). The powdered samples were mixed with 5 mL of solvent, vortexed for 10 minutes, and sonicated for 60 minutes. After centrifugation at 4500 rpm for 10 minutes, the supernatant was filtered through a 0.22 µm PTFE syringe filter before High-Performance Liquid Chromatography (HPLC) analysis.

HPLC analysis was conducted using a Shimadzu Hemp Analyzer, injecting 10 µL of the extract under a column temperature of 35 °C for 10 minutes. Cannabinoid concentrations, including THC and CBD, were determined using calibration standards and peak area ratios (Table 5). A mobile phase system consisting of phosphoric acid in water, acetonitrile, and methanol-based solvents were used for system stabilization. Methanol: acetonitrile (9:1 v/v) mixture was employed based on its proven efficiency in solubilizing phenolic compounds in hemp leaf tissues. This combination of methanol and acetonitrile offers high polarity and low viscosity, which enhances the extractability of both structural and biochemical constituents, including cannabinoids and secondary metabolites for downstream analysis. While this ratio was optimized within the context of the current study, the decision was guided by previous research demonstrating its effectiveness in extracting a broad range of phytochemicals from *Cannabis sativa* (Tzimas et al., 2024). Statistical analysis was performed using RStudio (version 2022.07.1 Build 554).



	Year	Biomass	Plant height	Stem diameter	Air temperature	Rainfall	Relative humidity	Soil moisture	Soil temperature
Year	1.00	-0.81 **	-0.72 *	-0.71 *	0.71 *	-0.78 *	-0.93 ***	-0.70 *	0.82 **
Biomass	-0.81 **	1.00	0.97 ***	0.81 **	-0.82 **	0.78 *	0.81 **	0.17 ns	-0.80 **
Plant height	-0.72 *	0.97 ***	1.00	0.87 **	-0.89 **	0.75 *	0.71 *	0.04 ns	-0.84 **
Stem diameter	-0.71 *	0.81 **	0.87 **	1.00	-0.92 ***	0.72 *	0.61 ns	0.23 ns	-0.94 ***
Air temperature	0.71 *	-0.82 **	-0.89 **	-0.92 ***	1.00	-0.80 *	-0.70 *	-0.17 ns	0.97 ***
Rainfall	-0.78 *	0.78 *	0.75 *	0.72 *	-0.80 *	1.00	0.88 **	0.36 ns	-0.82 **
Relative humidity	-0.93 ***	0.81 **	0.71 *	0.61 ns	-0.70 *	0.88 **	1.00	0.61 ns	-0.77 *
Soil moisture	-0.70 *	0.17 ns	0.04 ns	0.23 ns	-0.17 ns	0.36 ns	0.61 ns	1.00	-0.40 ns
Soil temperature	0.82 **	-0.80 **	-0.84 **	-0.94 ***	0.97 ***	-0.82 **	-0.77 *	-0.40 ns	1.00

FIGURE 2
Relationships between growth performance and environmental conditions in industrial hemp grown under variable field climates (2021–2023). Significant codes: '***'<0.00; '**'< 0.001; '*'<0.05; ns, non-significant.

TABLE 5 Calculations for the cannabinoid percentage conversions.

% Cannabinoid	(Concentration of cannabinoid mg/L) * (1000)/(vol/mg) * 100% Potency:(% THCA * 0.877) + %Δ9 - THC [THCA]: Concentration of THCA, DIL=Dilution Factor, VOL=External Volume, MG=dry sample weight (mg), 0.877¼molecular weight ratio of cannabinoids to cannabinoid acids.
Total THC %	Δ9-THC % + (THCA %*0.877)

A one-way ANOVA was conducted to evaluate differences in cannabinoid concentrations among cultivars.

Results

Climatic conditions

Monthly weather conditions varied notably across the years and locations. Air temperatures ranged from 10 °C to 25 °C, with higher means observed in 2023, particularly at GWC farm and Washington. In 2022, both the Sikeston and Washington sites recorded the highest total rainfall observed during the study period. Relative humidity remained above 70% overall, though Washington experienced a decline in 2022. Soil moisture was highest at GWC farm in 2022–2023 but notably reduced in Washington in 2023. Soil temperatures mirrored air temperature trends, reaching their highest in 2023 across all sites, indicating warmer growing conditions during that season.

Germination performance of hemp cultivars

Germination percentages among the thirty-two industrial hemp cultivars varied across the three experimental years (Table 3), reflecting both genetic diversity and potential differences in seed quality and viability. In 2021, germination ranged from as low as 1% in Grandi to 78.7% in Ferimon, with a grand mean of 41.55%. Cultivars such as Bialobrzeskie (56.7%), Fibror 79 (68.1%), and Jinma (64.5%) performed well above the grand mean, whereas Anka (6.5%), Katani (7.5%), and CRS-1 (8.6%) showed extremely poor viability (<10%).

In 2022, overall germination improved substantially (grand mean 64.47%), with top-performing cultivars including Bialobrzeskie (92.2%), Jinma (90.9%), and Fibror 79 (89.9%). Poor-performing lines included BVL5 (0%) and Fibranova (24%). The 2023 results, though covering fewer cultivars, showed the highest overall viability (grand mean 73.31%), with Futura 83 (88%), Bialobrzeskie (81.1%), and Jinma (85.6%) maintaining strong performance. Statistical analysis confirmed significant differences among cultivars in each year ($p< 0.05$), with least significant differences (LSD) of 1.86, 2.18, and 2.19 for 2021, 2022, and 2023, respectively. European cultivars generally exhibited high germination rates, consistent with previous studies (Amaducci et al., 2015; Tang et al., 2022), whereas poor performance in certain cultivars likely resulted from genetic limitations, seed viability or dormancy. The associated seeding rate calculations, adjusted for germination percentage, revealed the practical implications of seed viability on field establishment. For instance, cultivars with high viability such as Ferimon and Felina 32 required ~70 kg ha^{-1}o achieve the target viable seed rate, while low-performing cultivars such as Anka and Katani required >747 kg ha^{-1} rendering them impractical for commercial planting. These findings underscore the importance of pre-sowing germination testing and cultivar selection to ensure optimal stand establishment and resource efficiency in industrial hemp production.

Phenotypic variations among cultivars in the 2021 growing season

In 2021, fifteen industrial hemp cultivars were evaluated at two agroecological sites, GWC farm (Jefferson City) and Sikeston, Missouri, revealing significant variation ($P< 0.001$) in all measured traits, including emergence rate, plant height, stem diameter, plant density, and biomass (Tables 6, 7).

Emergence rate

The emergence rate ranged from 21% (Grandi) to 91.5% (Jinma and Fibror 79) across sites. High emergence was also observed for Bialobrzeskie (81-82%) and Tygra (81-84%), while cultivars like Cfx-2 (30-31.5%) and Grandi (20.5-21%) had the lowest establishment.

Plant height

Jinma exhibited the tallest plant height, reaching 335.1 cm at GWC and 302.5 cm at Sikeston. Fibror 79 and Tygra also displayed exceptional height (259.1 cm and 209.2 cm at Sikeston, respectively). In contrast, Grandi and Katani recorded the shortest plants (<60 cm), with grand mean height consistent between sites (~131 cm).

Stem diameter

Significant differences in stem diameter were recorded ($P< 0.001$), with Jinma again leading (13.4 mm at GWC, 16.7 mm at Sikeston), followed by Fibror 79 (12.3 mm at Sikeston). Thinner stems were observed in Grandi, Cfx-2, and CRS-1, with values between 2.7 mm and 4 mm.

Plant density

Plant density was highest in Jinma (41–415 plants/m^2), followed closely by Fibror 79 and Altair (36–41 plants/m^2). Grandi recorded the lowest density (9 plants/m^2 at both sites). Grand means were ~27 plants/m^2 for both locations.

TABLE 6 Phenotypic variation among fifteen cultivars grown at GWC farm, Jefferson City during the 2021 planting season.

Cultivars	Emergence rate (%)	Plant height (cm)	Stem diameter (mm)	Plant density (No. of plants/m^{2})	Biomass (kg/plant)	Biomass (kg/m^{2})
Altair	81.5^{a}	128.3^{bcde}	5.3^{c}	37^{a}	0.08^{bc}	2.96^{bc}
Anka	63.5^{b}	188.0^{b}	6.2b^{c}	29^{b}	0.02^{c}	0.58^{e}
Bialobrzeskie	81.0^{a}	121.9^{bcde}	4.8^{c}	36^{a}	0.01^{c}	0.36^{e}
Cfx-1	42.5^{de}	94.5^{cde}	4.9^{c}	19de	0.03^{bc}	0.57^{e}
Cfx-2	31.5^{fg}	48.3^{de}	7.0^{bc}	14^{fg}	0.03^{bc}	0.42^{e}
CRS-1	42.0^{def}	98.3^{bcde}	5.9^{bc}	18^{def}	0.01^{c}	0.18^{c}
Fibror 79	91.5^{a}	286.6^{a}	9.8^{ab}	41^{a}	0.13^{ab}	5.33 ^{ab}
Grandi	21.0^{g}	36.8^{e}	5.1^{c}	9^{g}	0.02^{c}	0.18^{f}
Ferimon	70.0^{b}	77.5^{cde}	5.5^{c}	32^{b}	0.07^{c}	2.24^{c}
Hliana	61.0^{bc}	103.4^{bcde}	6.4^{bc}	28^{bc}	0.05^{bc}	1.40^{cd}
Jinma	90.5^{a}	335.1^{a}	13.4^{a}	41^{a}	0.20^{a}	8.20^{a}
Joey	40.5^{ef}	96.5^{bcde}	6.7^{bc}	18^{ef}	0.04^{bc}	0.72^{cde}
Katani	41.5^{def}	54.3^{de}	6.8^{bc}	19^{def}	0.01^{c}	0.19^{f}
Picolo	52.0^{cd}	134.5^{bcd}	4.3^{c}	23^{cd}	0.07^{bc}	1.61^{cd}
Tygra	84.0^{a}	163.8^{bc}	5.3^{c}	38^{a}	0.23^{a}	8.74^{a}
Grand mean	59.600	131.177	6.511	26.821	0.067	1.80
P value	0.001	0.001	0.001	0.001	0.001	0.001

Different letters within a column indicate significant differences at p< 0.05 based on Tukey's Least Significant Difference (LSD) test following ANOVA.

Biomass

Biomass production differed significantly among cultivars at both locations ($P < 0.001$; Tables 6, 7). Jinma and Tygra consistently produced the highest biomass on a per-plant basis, with values ranging from 0.20 to 0.23 kg plant^{-1}, whereas Bialobrzeskie, Katani, CRS-1, and Grandi exhibited consistently low biomass production (≈0.01–0.02 kg plant^{-1}). Intermediate biomass levels were observed

TABLE 7 Phenotypic variation among fifteen cultivars grown at Sikeston, MO during the 2021 planting season.

Cultivars	Emergence rate (%)	Plant height (cm)	Stem diameter (mm)	Plant density (No. of plants/m^{2})	Biomass (kg/plant)	Biomass (kg/m^{2})
Altair	79.5^{ab}	132.7^{bc}	7.2^{de}	36^{ab}	0.06^{bc}	2.16^{b}
Anka	73.0^{abcd}	72.0^{c}	8.0^{cd}	33^{abcd}	0.01^{de}	0.33^{e}
Bialobrzeskie	82.0^{ab}	152.7^{bc}	5.9^{def}	37^{ab}	0.01^{e}	0.37^{e}
Cfx-1	41.5^{de}	92.5^{c}	4.0^{ef}	19de	0.03^{de}	0.57^{de}
Cfx-2	30.5^{e}	64^{c}	3.5^{f}	14^{e}	0.03^{de}	0.42^{de}
CRS-1	46.0^{cde}	95.4^{c}	3.2^{f}	21^{cde}	0.01^{de}	0.21^{ef}
Fibror 79	91.0^{a}	259.1^{a}	12.3^{b}	4^{a}	0.01^{b}	0.40^{de}
Grandi	20.5^{e}	55.4^{c}	2.7^{f}	9^{e}	0.01^{de}	0.09^{f}
Ferimon	74.5^{abc}	139.8^{bc}	4.5^{def}	34^{abc}	0.04^{cde}	1.36^{c}
Hliana	72.0^{abcd}	118.8^{bc}	5.6^{def}	32^{abcd}	0.04^{cd}	1.28^{cd}
Jinma	91.5^{a}	302.5^{a}	16.7^{a}	415^{a}	0.20^{a}	8.20^{a}
Joey	50.5^{bcde}	90.5^{c}	4.5^{def}	23^{bcde}	0.03^{de}	0.69^{d}
Katani	40.5^{de}	95.2^{c}	3.8^{ef}	18de	0.01^{e}	0.18^{e}
Picolo	51.5^{bcde}	87.0^{c}	3.4^{f}	23^{bcde}	0.01^{e}	0.23^{e}
Tygra	81.5^{ab}	209.2^{ab}	11.1^{bc}	37^{ab}	0.20^{a}	7.40^{a}
Grand mean	61.733	131.120	6.440	27.780	0.052	1.44
P value	0.001	0.001	0.001	0.001	0.001	0.033

Different letters within a column indicate significant differences at p< 0.05 based on Tukey's Least Significant Difference (LSD) test following ANOVA.
Phenotypic variations among cultivars in the 2022 growing season.

for cultivars such as Fibror 79, Ferimon, Hliana, and Picolo, reflecting moderate growth potential across environments.

When expressed on an area basis, Jinma and Tygra also achieved the highest biomass yields, reaching 8.20–8.74 kg m^{-2} at GWC and 7.40–8.20 kg m^{-2} at Sikeston, driven by a combination of high plant density and vigorous vegetative growth. In contrast, cultivars with lower plant density and reduced biomass per plant produced substantially lower biomass per unit area (<1.5 kg m^{-2}). Grand mean biomass was higher at GWC (1.80 kg m^{-2}; 0.067 kg $plant^{-1}$) than at Sikeston (1.44 kg m^{-2}; 0.052 kg $plant^{-1}$), indicating a favorable site effect on overall biomass accumulation (Tables 6, 7).

Emergence rate

Twenty-two industrial hemp cultivars were evaluated for emergence rate across three locations: GWC farm (Jefferson City), Sikeston, and Washington, during the 2022 growing season (Tables 8, 9; Supplementary 1). The highest emergence was recorded for Jinma (100%) at GWC and Sikeston, followed by Felina 32 and Ferimon with emergence rates exceeding 90% at GWC. Conversely, lower emergence rates were observed in Uso 31 and Santhica 70 at Washington, with Santhica 70 registering below 40%. Significant differences in emergence rates were observed among cultivars and locations ($P< 0.05$), with the trend generally indicating superior emergence at GWC, moderate at Sikeston, and lower at Washington.

Days to 50% flowering

Late flowering was observed in China-origin cultivars such as Yuma and Puma (107 days), and Jinma (105 days), while Canadian and European cultivars like Ferimon (54 days), Henola, Altair, and Uso 31 (58 days) exhibited early flowering. Most cultivars, including Fibror 79, Felina 32, Futura 83 and Tygra, were classified as mid-flowering, reaching 50% flowering between 60 and 87 days (Table 8).

Stem diameter

Stem diameter varied significantly among cultivars and locations ($P< 0.001$). Jinma exhibited the thickest stems across all sites, with an exceptional value of ~36 mm at Washington and over 15 mm at GWC and Sikeston (Tables 8, 9; Supplementary 1). Other cultivars with thick stems included Yuma, Puma, and MS 77. In contrast, Uso 31, Henola, and BVL5 consistently showed narrower stems (4–7 mm). Stem diameter was positively correlated with plant height (r = 0.897) and biomass (r = 0.871) (Figure 2).

Plant density

Plant density showed significant variation across cultivars and locations ($P< 0.001$). The highest plant densities were observed for Futura 83 and Ferimon at Sikeston (42 and 41 plants/m^2, respectively), and for BVL-3, Jinma, and Felina 32 at GWC (26–34 plants/m^2) (Tables 9, 10; Supplementary 1). The lowest densities were recorded for Bialobrzeskie, Rajan, and Tygra at Washington (<10 plants/m^2). Generally, plant densities were highest at Sikeston and lowest at Washington, reflecting environmental and emergence challenges at the specific site.

TABLE 8 Days to 50% flowering of twenty-two industrial hemp cultivars planted at the GWC farm, Jefferson City, MO in the 2022 growing season.

Cultivar	Day to 50% flowering	Flowering status
Yuma	107	Late Flowering
Puma	107	Late Flowering
Jinma	105	Late Flowering
MS 77	104	Late Flowering
BVL-3	87	Mid Flowering
BVL5	85	Mid Flowering
BVL1	84	Mid Flowering
Gravity	82	Mid Flowering
BVL2	80	Mid Flowering
Rajan	77	Mid Flowering
Fibranova	72	Mid Flowering
Fibror 79	71	Mid Flowering
Bialobrezeskie	70	Mid Flowering
Orion 33	67	Mid Flowering
Felina 32	63	Mid Flowering
Tygra	62	Mid Flowering
Futura 83	61	Mid Flowering
Santhica 70	60	Mid Flowering
Uso 31	58	Early Flowering
Altair	58	Early Flowering
Henola	58	Early Flowering
Ferimon	54	Early Flowering

NB: Early flowering: ≤ 60 day; Mid flowering: 61–95 day; Late flowering: > 95 days.

Plant biomass

Biomass accumulation per plant varied significantly across cultivars and locations ($P< 0.001$). Jinma produced the highest biomass at all three sites, peaking at approximately 0.99 kg/plant at GWC farm (Tables 8, 9; Supplementary 1). Other high-performing cultivars included Puma, MS 77, and Yuma, with values between 0.33 and 0.70 kg/plant. In contrast, Felina 32, Ferimon, Bialobrzeskie, and Uso 31 consistently showed low biomass values (<0.1 kg/plant). When expressed on a field-area basis (kg m^{-2}), biomass production showed consistent cultivar-specific patterns across locations. Jinma exhibited the highest and most stable performance, ranging from approximately 1.45 kg m^{-2} at Sikeston to 27.72 kg m^{-2} at GWC, followed by Puma (4.81–19.60 kg m^{-2}), MS 77 (5.55–8.55 kg m^{-2}), and Yuma (2.48–8.91 kg m^{-2}). In contrast, low-performing cultivars such as Felina 32, Ferimon, Bialobrzeskie, Henola, Rajan, Tygra, and Uso 31 consistently produced<0.1–0.8 kg m^{-2} across sites. Overall, biomass was greater at GWC than at Sikeston, and across environments biomass accumulation was strongly correlated with plant height ($r = 0.818$) and stem diameter ($r = 0.871$) (Figure 2).

TABLE 9 **Phenotypic variation among the twenty-two hemp cultivars grown in GWC farm, Jefferson City, MO in the year 2022 growing season.**

Cultivar	Emergence rate (%)	Plant height (cm)	Stem diameter (mm)	Plant density/m^2	Biomass (kg/plant)	Biomass (kg/m^2)	Fiber yield (bast + hurd) (kg/m^2)
Altair	45^{bc}	115.1^{ef}	6.2^{cd}	10^{c-g}	0.10^{b}	1.00^{de}	0.3^{f}
Bialobrzeskie	70^{abc}	109.4^{ef}	5.4^{a-g}	20^{a-g}	0.02^{b}	0.40^{fg}	0.20^{f}
Felina 32	95^{abc}	129.8^{def}	5.5^{cd}	26^{abc}	0.01^{b}	0.26^{g}	0.44^{ef}
Ferimon	90^{ab}	123.8^{def}	6.9^{cd}	24^{a-e}	0.02^{b}	0.48^{fg}	0.44^{ef}
Fibranova	60^{abc}	211.2^{bcd}	12.9^{a-d}	8^{efg}	0.10^{b}	0.80^{ef}	0.98^{cdef}
Fibror 79	65^{abc}	168.5^{cde}	6.4^{cd}	17^{b-g}	0.04^{b}	0.68^{ef}	0.49^{def}
Futura 83	85^{abc}	177.4^{cde}	8.5^{bcd}	25^{a-d}	0.08^{b}	2.00^{cd}	0.53^{def}
Gravity	60^{abc}	153.3^{c-f}	11.4^{a-d}	$6f^{g}$	0.20^{ab}	1.20^{de}	0.40^{f}
Henola	50^{abc}	67.3^{f}	4.7^{d}	9^{d-g}	0.02^{b}	0.18^{g}	0.06^{f}
BVL3	100^{a}	239.5^{bc}	9.7^{a-d}	34^{a}	0.42^{ab}	14.28^{ab}	2.28^{bcd}
Jinma	100^{a}	333.3^{a}	16.5^{ab}	28^{ab}	0.99^{a}	27.72^{a}	5.80^{a}
MS 77	90^{ab}	284.9^{ab}	14.6^{abc}	15^{b-g}	0.57^{ab}	8.55^{bc}	2.63^{bc}
BVL2	70^{abc}	147.3^{c-f}	9.1^{a-d}	18^{a-g}	0.13^{b}	2.34^{cd}	0.79^{def}
BVL1	80^{abc}	123.6^{def}	6.0^{c-d}	24^{a-e}	0.03^{b}	0.72^{ef}	0.69^{def}
BVL5	70^{abc}	102.9^{ef}	4.5^{d}	13^{b-g}	0.05^{b}	0.65^{ef}	0.23^{f}
Orion 33	80^{abc}	134.6^{def}	5.3^{cd}	22^{a-f}	0.08^{b}	1.76^{d}	0.27^{f}
Puma	100^{a}	357.3^{a}	16.8^{ab}	28^{ab}	0.70^{ab}	19.60^{ab}	2.23^{bcde}
Rajan	35^{c}	157.5^{c-f}	7.9^{bcd}	5^{g}	0.03^{b}	0.15^{g}	0.20^{f}
Santhica 70	80^{abc}	138.0^{def}	7.6^{bcd}	19^{a-g}	0.05^{b}	0.95^{de}	0.20^{f}
Tygra	35^{c}	124.4^{def}	5.5^{cd}	5^{a-g}	0.02^{b}	0.10^{g}	0.11^{f}
Uso 31	80^{abc}	111.8^{ef}	4.7^{d}	20^{a-g}	0.04^{b}	0.80^{ef}	0.46^{f}
Yuma	100^{a}	336.1^{a}	18.20^{a}	27^{abc}	0.33^{ab}	8.91^{bc}	3.23^{b}
Grand mean	3.73	174.86	6.21	19.33	0.13	2.51	1.05
P value	<0.001	<0.001	<0.001	<0.001	<0.001	0.034	0.014

Different letters within a column indicate significant differences at $p < 0.05$ based on Tukey's Least Significant Difference (LSD, $\alpha = 0.05$) test following ANOVA.

Fiber yield

Fiber yield differed significantly among cultivars at all locations (GWC: P = 0.014; Sikeston: P = 0.02; Washington: P = 0.04) and closely reflected biomass production on a per-area basis, indicating clear genotype × environment (G×E) effects (Figure 3; Supplementary 1). At GWC, Jinma consistently produced the highest fiber yield across years (5.83 kg m^{-2}), followed by Futura 83 and Fibror 79, whereas several cultivars, including Uso 31 and Felina 32, produced less than 1.0 kg m^{-2}. In Sikeston, overall fiber yields were lower, and cultivar differences were less pronounced, with MS 77 and Puma exhibiting comparatively higher yields (approximately 2.0–2.3 kg m^{-2}). At Washington, Jinma again ranked highest (approximately 3.18 kg m^{-2}), while most other cultivars produced less than 1.0 kg m^{-2}. Least significant difference (LSD) analysis confirmed strong cultivar separation at GWC and Washington, whereas discrimination among cultivars at Sikeston was more limited, reflecting site-specific environmental constraints on fiber productivity, but weaker discrimination at Sikeston, reflecting site-specific environmental constraints on fiber productivity.

Correlation analysis was carried out to quantify the relationships between agronomic performance (biomass, plant height, and stem diameter) and major environmental variables (air temperature, rainfall, relative humidity, soil moisture, and soil temperature) recorded across the 2021–2023 growing seasons (Figure 2). Correlation analysis revealed that, biomass, plant height, and stem diameter were positively correlated with rainfall (r = 0.72 to 0.78, $p < 0.05$) and plant height (r = 0.71, $p < 0.05$) and biomass (r = 0.81, $p < 0.01$) were significantly positively corelated with relative humidity. But biomass, and other growth parameters were significantly negatively correlated with air and soil temperatures (r ≈ −0.82 to −0.90, $p < 0.01$). These results indicate that hemp growth was enhanced under cooler and more humid field conditions, confirming the strong influence of environmental variability on cultivar performance.

Multilocation analysis

Biomass, plant height, and stem diameter exhibited inter-annual and inter-cultivar variability across all three study sites (GWC, Sikeston, and Washington) over the 2021–2023 growing seasons (Figures 4–6).

TABLE 10 **Phenotypic variation among the nineteen hemp cultivars grown in Sikeston Agri-Park, Sikeston, MO in the year 2022.**

Cultivars	Emergence rate (%)	Plant height (cm)	Stem diameter (mm)	Plant density (No. of plants/m^2)	Biomass (kg/plant)	Biomass kg/m^2	Fiber yield (bast + hurd) (kg/m^2)
Altair	60^{abc}	119.5^{fg}	8.1^{b}	15^{bc}	0.03^{a}	0.45^{ef}	0.19^{ef}
Bialobrzeskie	40^{c}	124.5^{fg}	9.5^{b}	7^{c}	0.01^{a}	0.07^{h}	0.03^{g}
Felina 32	80^{a}	104.7^{gh}	7.5^{b}	37^{ab}	0.02^{a}	0.74^{d}	0.31^{d}
Ferimon	80^{a}	106.5^{gh}	5.7^{b}	41^{a}	0.02^{a}	0.82^{d}	0.34^{d}
Fibror 79	65^{abc}	205.9^{d}	8.7^{b}	22^{abc}	0.03^{a}	0.66^{d}	0.28^{d}
Futura 83	85^{a}	119^{fg}	8.2^{b}	34^{ab}	0.02^{a}	0.68^{d}	0.29^{d}
Henola	60^{abc}	77.083^{h}	7.35^{ab}	20^{abc}	0.01^{a}	0.20^{fg}	0.08^{g}
Jinma	80^{a}	336^{a}	11.6^{b}	29^{abc}	0.05^{a}	1.45^{ef}	0.61^{c}
MS 77	85^{a}	326^{ab}	12.5^{ab}	37^{ab}	0.15^{a}	5.55^{a}	2.33^{a}
BVL1	70^{ab}	150.5^{ef}	8.2^{b}	23^{abc}	0.03^{a}	0.69^{d}	0.29^{d}
BVL3	70^{ab}	159.5^{e}	8.3^{b}	20^{abc}	0.03^{a}	0.60^{de}	0.25^{de}
BVL4	70^{ab}	100.5^{gh}	8.0^{b}	17^{abc}	0.01^{a}	0.17^{g}	0.07^{g}
BVL5	60^{abc}	104.5^{gh}	7.7^{b}	16^{bc}	0.03^{a}	0.48^{cde}	0.20^{ef}
Orion 33	80^{a}	119.5^{fg}	8.1^{b}	29^{abc}	0.02^{a}	0.58^{de}	0.24^{de}
Puma	85^{a}	292.1^{bc}	21.6^{a}	37^{ab}	0.13^{a}	4.81^{a}	2.02^{a}
Rajan	50^{bc}	261^{c}	12.03^{ab}	7^{c}	0.18^{a}	1.26^{c}	0.53^{c}
Santhica 70	60^{abc}	124.75^{fg}	9.5^{b}	17^{abc}	0.03^{a}	0.51^{de}	0.21^{ef}
US0 31	50^{bc}	80.4^{h}	6.5^{b}	20^{abc}	0.02^{a}	0.40^{ef}	0.17^{f}
Yuma	80^{a}	293^{bc}	14.2^{ab}	31^{abc}	0.08^{a}	2.48^{b}	1.04^{b}
Grand mean	68.94	168.68	9.69	24.43	0.04	0.98	0.50
P value	<0.001	<0.001	0.008	<0.001	0.158	0.01	0.02

Different letters within a column indicate significant differences at $p < 0.05$ based on Tukey's Least Significant Difference (LSD) test following ANOVA.

Biomass

Biomass production varied substantially by year, cultivar, and site (Figure 4). In 2021, Jinma produced the highest biomass across both locations, with mean values of approximately 8–9 kg m^{-2} at GWC and >15 kg m^{-2} at Sikeston, well above the yearly mean (~5–6 kg m^{-2}). Fibror 79 and Ferimon showed intermediate biomass, averaging ~4–6 kg m^{-2} and ~1–3 kg m^{-2}, respectively, whereas

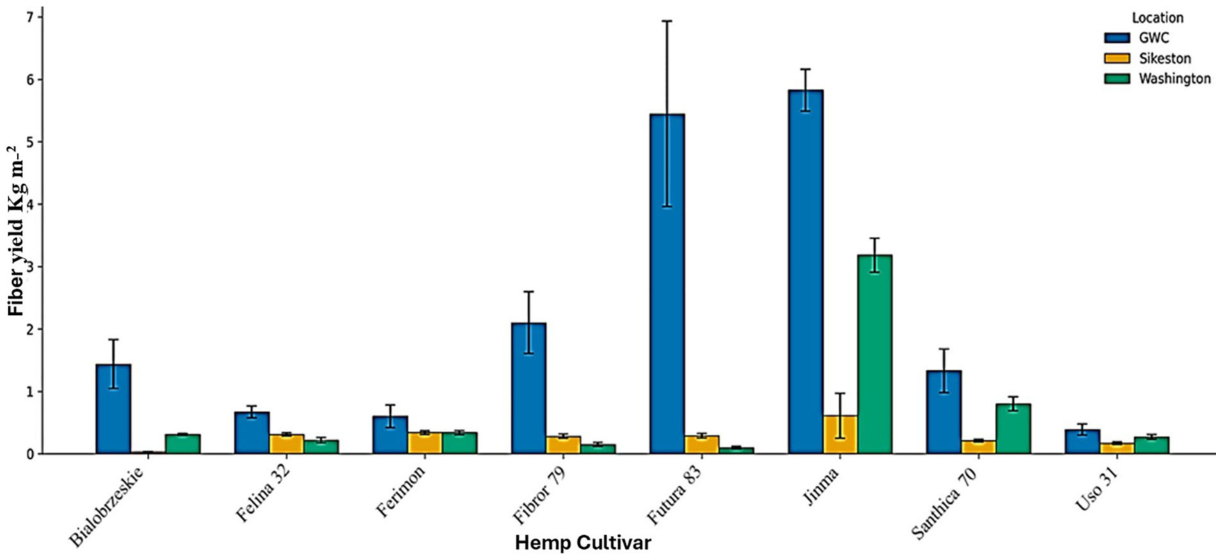


FIGURE 3
Genotype × environment effects on mean fiber yield (kg m^{-2}, ± SE) of core industrial hemp cultivars across Missouri locations (2022–2023).

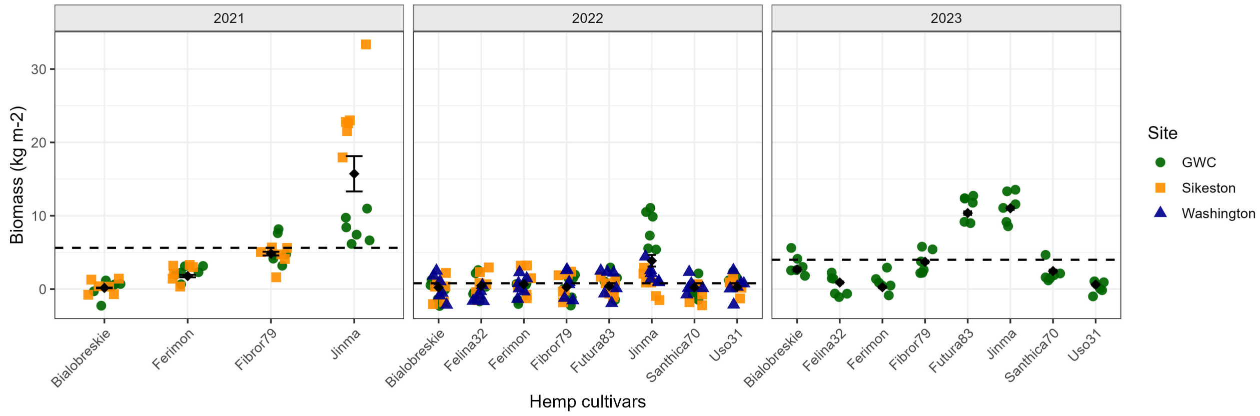


FIGURE 4
Mean yearly biomass per meters square of industrial hemp cultivars represented across all three study years. Error bars represent the 95% confidence interval calculated using the average and standard error for each cultivar, and the gray dashed line depicts an average biomass for all cultivars and sites.

Bialobreskie remained consistently low (<0.5 kg m^{-2}). In 2022, overall biomass was reduced across cultivars and sites, with most cultivars producing<1 kg m^{-2}. Jinma nevertheless remained the top performer, reaching approximately 4–6 kg m^{-2} at GWC and ~2–4 kg m^{-2} at Washington, while values at Sikeston were markedly lower. Other cultivars, including Fibror 79, Ferimon, and Felina 32, generally remained below 1 kg m^{-2} across sites.

In 2023, biomass was assessed at GWC only, where cultivar differences remained pronounced. Futura 83 and Jinma produced the highest biomass, averaging ~9–11 kg m^{-2}, followed by Fibror 79 (~3–4 kg m^{-2}). In contrast, Felina 32, Ferimon, Santhica 70, and Uso 31 consistently produced ≤3 kg m^{-2} (Figure 4).

Plant height

Trends in plant height mirrored those of biomass (Figures 4, 5). Jinma was consistently the tallest cultivar, surpassing 330 cm in 2021 and remaining above 300 cm in subsequent years. Fibror 79 followed closely in height across most environments, while Ferimon maintained intermediate height. Bialobrzeskie and Uso 31 consistently demonstrated shorter stature, with Uso 31 being the shortest across all years and locations (Figure 5).

Stem diameter

Stem girth varied significantly by cultivar, site, and year. In 2021, Jinma recorded the largest stem diameters (~17 mm), followed by Fibror 79. Cultivars such as Bialobrzeskie and Ferimon had narrower stems (~5–7 mm). In 2022, although diameter values were more evenly distributed, Jinma remained dominant, especially at Sikeston and Washington. By 2023, stem diameters declined across all cultivars, yet Jinma and Fibror 79 continued to lead in performance (Figure 6).

Cannabinoid analysis

Tetrahydrocannabinol concentrations

Total THC concentrations remained below the federal legal threshold of 0.3% across all cultivars, locations, and years (Figure 7),

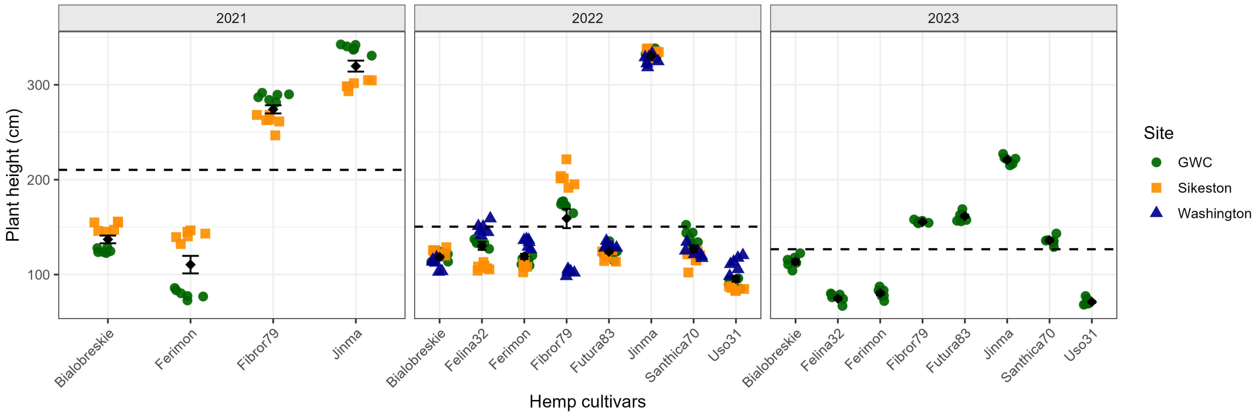


FIGURE 5
Yearly mean plant height of industrial hemp cultivars across all three study years. Error bars represent the 95% confidence interval calculated using the average and standard error for each cultivar, and the gray dashed line depicts an average plant height for all cultivars and agroecological sites.

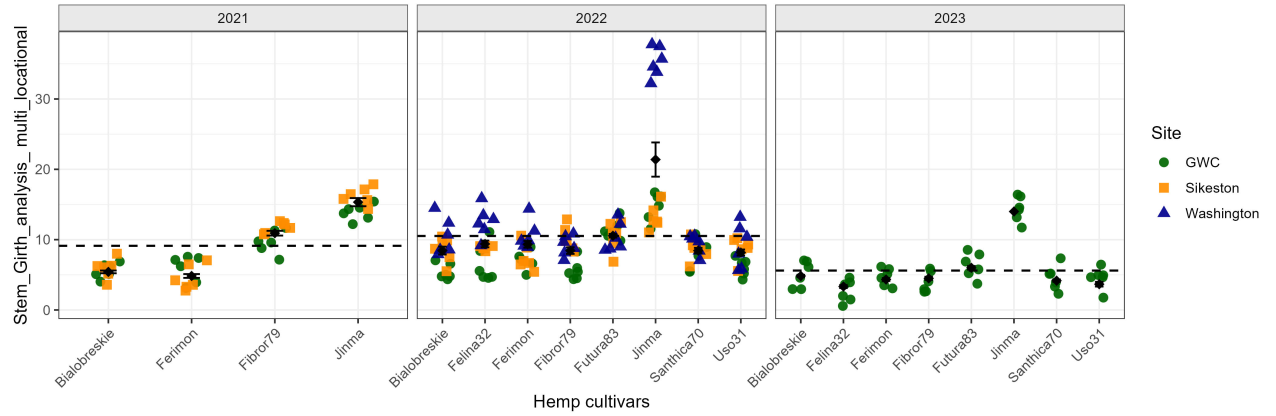


FIGURE 6
Yearly means stem diameter of core industrial hemp cultivars across all three study years. Error bars represent the 95% confidence interval calculated using the average (black circle) and standard error for each cultivar, and the gray dashed line depicts an average stem diameter for all cultivars and agroecological sites.

although statistically significant differences were observed among cultivars. Slightly elevated THC levels were recorded in Felina 32, Ferimon, and Futura 83 during the 2022 season, particularly in the GWC farm-Jefferson City trial. THC concentrations were relatively stable across years within GWC trials, although some cultivars, such as Bialobrzeskie, Ferimon, and USO 31 in 2023, approached 0.10-0.15%. cultivars such as Fibror 79, Futura 83, and Santhica 70 in 2023 and 2024 consistently maintained THC concentrations below 0.3%. These findings affirm the suitability of these genotypes for industrial hemp classification and regulatory compliance.

Total cannabidiol

In 2022, cannabidiol (CBD) concentrations varied significantly between study sites, with cultivars generally accumulating higher levels at the Sikeston location compared to the GWC-Jefferson City site. Cultivars such as Bialobrzeskie, Felina 32, Ferimon, and Fibror 79 exhibited moderate to high CBD levels at Sikeston, whereas GWC plots recorded lower levels for the same cultivar, except for Ferimon, which maintained elevated CBD concentrations across both sites. Across the 2022, 2023, and 2024 growing seasons, substantial inter-annual variation in CBD accumulation was observed at GWC (Figure 8). In 2024, both Bialobrzeskie and Fibror 79 showed notable increases in CBD content compared to the previous years, with Bialobrzeskie reaching approximately 0.5% and Fibror 79 around 0.3%. In the 2022 GWC farm trials, Felina 32 and Ferimon emerged as top performers, achieving mean CBD concentrations near 0.6%. In 2023, Bialobrzeskie and USO 31 recorded moderate levels ranging from 0.2% to 0.3%. Conversely, Futura 83, Jinma, and Santhica 70 consistently exhibited low CBD levels across all years and locations.

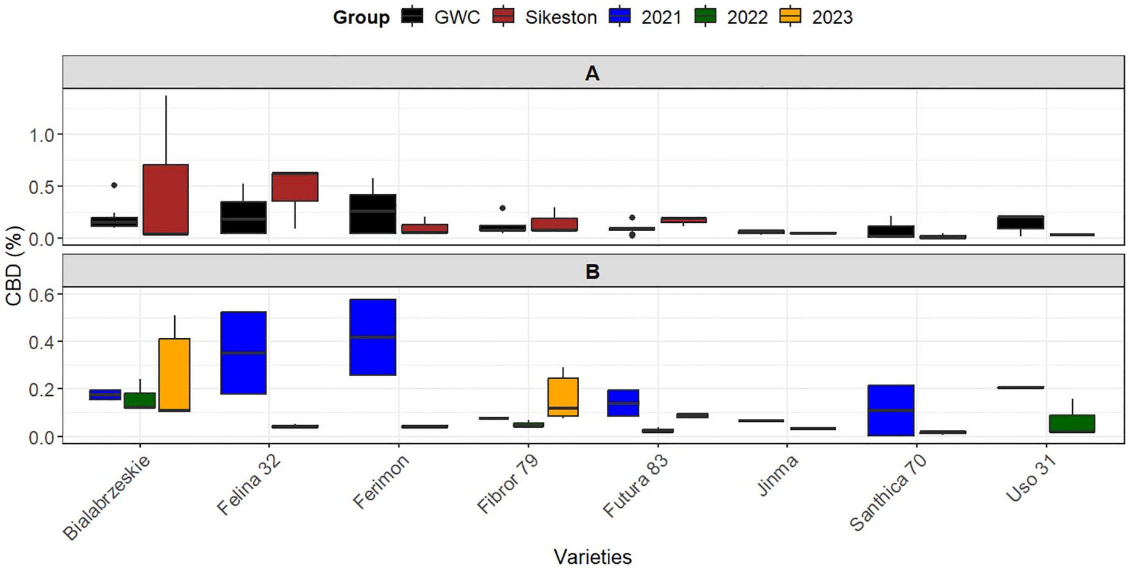


FIGURE 7
Boxplots showing the concentration of total THC (%) across eight industrial hemp cultivars. **(A)** Variation by study locations (GWC-Jefferson City and Sikeston). **(B)** Variation by year (2021, 2022 and 2023).

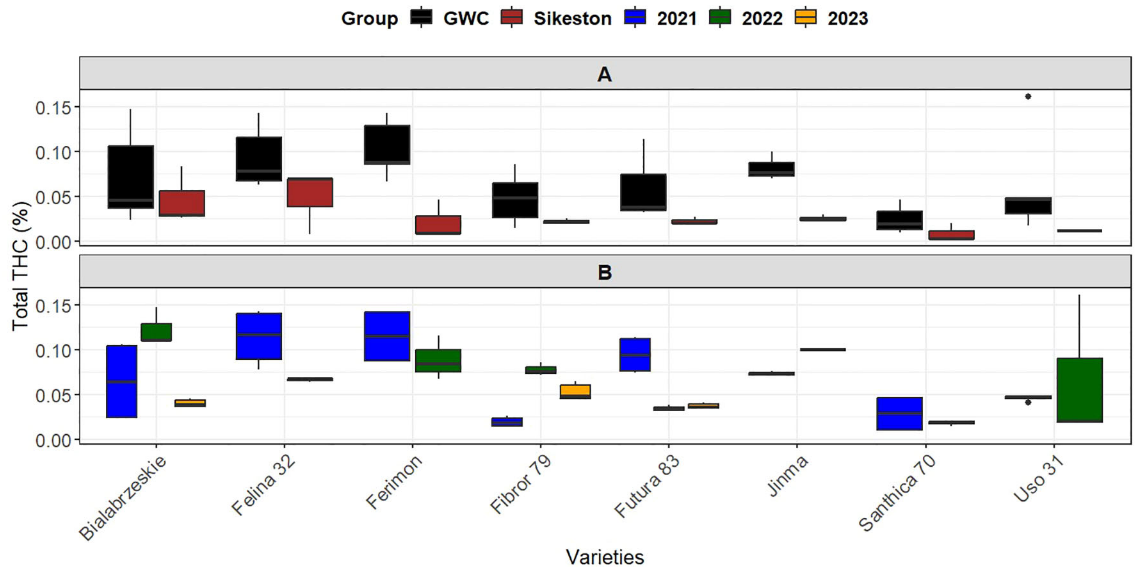


FIGURE 8
Boxplots showing the concentration of total CBD (%) in eight industrial hemp cultivars. **(A)** Variation by experimental location (GWC-Jefferson City and Sikeston); **(B)** Variation by year (2021, 2022, and 2023).

Discussion

Environmental influences on agronomic performance

Environmental conditions strongly influence hemp growth, particularly during early establishment and flowering (Struik et al., 2000; Amaducci et al., 2015). Across the three-year study, variation in temperature, rainfall, humidity, and soil moisture significantly affected cultivar performance. Reduced biomass at Washington in 2023 was likely driven by high evapotranspiration and limited soil moisture, consistent with stress-related constraints on canopy development and carbon assimilation reported by Lisson et al. (2000) and Ortmeier-Clarke et al. (2023) under water-limited conditions. Conversely, favorable conditions at Sikeston in 2022, characterized by higher rainfall, moderate temperatures, and greater humidity, likely enhanced biomass accumulation, particularly for high-yielding cultivars such as Jinma. This agrees with findings by Campiglia et al. (2017) and Adesina et al. (2020), who showed that hemp growth is optimized under moderate temperatures (20–25 °C) and adequate soil moisture. Similar location-by-year effects across U.S. agroecological zones were reported by Williams et al. (2025) and Stack et al. (2021), emphasizing the importance of genotype–environment matching. Accordingly, climate-adapted cultivar selection combined with locally responsive agronomic practices is essential for sustaining hemp productivity under increasing climatic variability (Panday et al., 2025).

Hemp emergence is influenced by genetic and environmental factors such as seed vigor, soil moisture, and temperature (Poudel et al., 2020; Amaducci et al., 2015; Campiglia et al., 2017). Higher emergence at the GWC farm in 2022 was likely due to moderate temperatures and adequate moisture that favored uniform seedling establishment. In contrast, lower emergence at Washington may reflect elevated soil temperatures and moisture deficits that constrain early growth. Cultivars including Jinma, Felina 32, and Ferimon showed stable emergence across environments, indicating greater seedling vigor and tolerance to early-season stress. The strong correlation between emergence and plant density (r = 1.00) aligns with Panday et al. (2025), reinforcing the role of uniform emergence in maximizing biomass production. These results highlight the importance of environment-specific cultivar selection and planting strategies in stress-prone regions.

Flowering time variation among hemp cultivars is governed by photoperiod sensitivity and genotype-environment interactions, both of which significantly impact regional adaptability and harvest scheduling (Zhang et al., 2021; Petit et al., 2020). Late-flowering cultivars such as Jinma, developed at lower latitudes (e.g., China), typically require shorter photoperiods to trigger reproductive development. In contrast, northern genotypes like Ferimon and USO 31 are either photoperiod-neutral or have reduced sensitivity, leading to earlier flowering under Missouri's extended summer daylight conditions (Van der Werf et al., 1994; Lisson et al., 2000). Our field data confirm this divergence, with Jinma flowering at approximately 105 DAPS: well within the region's frost-free window. No frost damage was observed at any site during the three-year study period, as air temperatures remained above freezing in October, and soil temperatures consistently exceeded 15 °C during the flowering-to-harvest phase (Figure 1). This climatic stability confirms that late-flowering cultivars like Jinma are viable in central and southern Missouri. However, in more northern regions or in years with early frost onset, such cultivars could be at greater risk of biomass and quality loss, particularly in fiber-production systems. Genetic studies by Toth et al. (2022) and Dowling et al. (2024) underscore the influence of FLOWERING LOCUS T (FT) homologs and other photoperiod-regulatory genes in controlling flowering time across latitudes, supporting the phenological variation we observed. These findings align with previous work by Amaducci et al. (2008), who emphasized the importance of synchronizing cultivar phenology with regional frost-free periods to ensure reliable production and fiber quality.

Significant variation in plant density was observed across cultivars and locations, reflecting the combined influence of emergence rate, seedling vigor, and environmental conditions. Higher plant densities at Sikeston in 2022 were likely supported by favorable rainfall and soil moisture conditions, promoting uniform germination. In contrast, lower densities recorded at Washington were consistent with the site's drier and warmer conditions. Cultivars such as Futura 83, Felina 32, and Jinma demonstrated consistently high stand densities, indicative of strong early vigor and adaptability. These attributes are particularly beneficial for fiber-production systems, where dense stands promote vertical growth and reduce lateral branching (Small and Marcus, 2002). Conversely, reduced densities in cultivars like USO 31 and Santhica 70 at Washington suggest the need for improved seed quality, adjusted seeding rates, or site-specific agronomic practices. These findings support previous recommendations by Tang et al. (2017), suggesting higher seeding densities (90–150 plants m^{-2}) for fiber production and lower densities (30–75 plants m^{-2}) for seed-production systems (Tamang et al., 2025), to balance stem quality and minimize intra-specific competition. Overall, the correlation analysis between agronomic and climatic variables demonstrated that hemp growth and yield performance were closely linked to environmental conditions (Figure 2). This reinforces the role of genotype-environment interactions in shaping hemp productivity across diverse field sites and growing seasons.

Biomass production and fiber yields of core industrial hemp cultivars (2021-2023)

Jinma, a tall Chinese fiber-type cultivar, consistently exhibited superior aboveground biomass and fiber yield across locations and years, demonstrating strong genotypic stability and broad agroecological adaptability within the southern U.S. Midwest. Its performance was particularly robust at the GWC farm, where fiber yields reached approximately 5.8 kg m^{-2} (~ 3–5 t $acre^{-1}$), and remained comparatively high at Sikeston and Washington despite contrasting environmental conditions. These results are especially notable given the limited availability of region-specific management guidelines for fiber hemp following its recent reintroduction in the United States.

When contextualized against other bast fiber crops, Jinma's productivity is comparable to reported kenaf yields (5–10 t $acre^{-1}$) and substantially exceeds typical flax bast fiber yields (~0.8 t $acre^{-1}$) under U.S. field conditions (Austin et al., 2024; Arefin et al., 2021). Other cultivars, including Fibror 79 and Ferimon, exhibited moderate biomass and fiber potential but showed greater sensitivity to environmental variability, particularly under suboptimal conditions. The reduced biomass and fiber yields observed in 2023, especially at the Washington site, were likely driven by early-season flooding followed by elevated summer temperatures, which constrained stand establishment and vegetative growth. These trends are consistent with previous studies indicating that tall, fiber-oriented cultivars maximize biomass and fiber accumulation under favorable temperature, moisture, and plant density conditions (Amaducci et al., 2015; Tang et al., 2017). Jinma's high productivity can be attributed to complementary morphological traits, such as tall stature, thick stems, and strong early vigor, that enhance resource-use efficiency and fiber yield. Its stable performance across diverse environments supports its suitability for resilient fiber-based production systems, while reduced performance under stress underscores the importance of site-specific cultivar selection (Finnan and Burke, 2013).

Plant height of core industrial hemp cultivars across three years (2021-2023)

Plant height is a critical determinant of stem biomass, bast fiber and hurd yield in fiber hemp. Among the cultivars evaluated, Jinma and Fibror 79 consistently achieved the tallest height across years and locations, reflecting both their genetic selection for fiber production and adaptability to variable environments. Favorable conditions at GWC and Sikeston in 2022, characterized by moderate temperatures and sufficient soil moisture, further promoted elongation, consistent with findings by Williams et al. (2025), who reported strong genotype × environment effects on hemp height in the Midwestern region, US. A strong positive correlation between plant height and biomass reinforces the role of vertical growth in fiber yield potential, corroborated by previous work (Small and Marcus, 2002; Salentijn et al., 2015; Musio et al., 2018; Rehman et al., 2021; Ortmeier-Clarke et al., 2023). In contrast, Washington's hotter and drier conditions in 2023 suppressed stem elongation across all genotypes, an outcome attributed to heat and drought stress that limits internodal development (Campiglia et al., 2017; Panday et al., 2025).

Photoperiod-temperature interactions also modulated plant height variation, particularly in photoperiod-sensitive cultivars, which showed strong responses to day length and growing degree accumulation (Petit et al., 2020; Zhang et al., 2021). These dynamics underscore the importance of matching cultivar phenology to local climatic regimes. Although Bialobrzeskie maintained reasonable emergence and stand density (Figure 6), its moderate height reduced its suitability for fiber-production systems. Shorter cultivars tend to exhibit limited internodal elongation and reduced bast fiber proportion, making them more appropriate for dual-purpose or seed-oriented production where earlier flowering and easier mechanical processing are prioritized (Rahemi et al., 2021; Ortmeier-Clarke et al., 2023). Overall, these findings highlight the need for cultivar-environment matching in hemp cropping systems. Tall, late-flowering genotypes like Jinma are best suited to regions with extended growing seasons and sufficient late-season moisture, while compact cultivars may perform better in stress-prone or short-season environments. As noted by Panday et al. (2025), optimizing spacing, moisture availability, and temperature during vegetative stages is essential for maximizing height and fiber biomass.

Stem diameter of hemp cultivars across three growing seasons (2021-2023)

Stem diameter is a critical morphological trait in fiber hemp, closely linked to structural biomass, bast fiber yield, and mechanical processing efficiency. Across all environments and years, Jinma consistently exhibited the thickest stems, even under suboptimal moisture conditions at the Washington site, suggesting robust genetic vigor and potential water-use efficiency. These results support earlier findings by Tang et al. (2017), Petit et al. (2020), and Musio et al. (2018), which highlight thicker stems as indicators of enhanced bast fiber content and total stem biomass in fiber-bred cultivars. A strong positive correlation was observed between stem diameter and both plant height and biomass production, reinforcing the interconnected nature of architectural traits and their cumulative impact on fiber productivity. This aligns with the understanding that stem thickness contributes significantly to both yield and harvest quality in fiber hemp. Inter-annual variation also influenced diameter patterns. In 2022, reduced variability across cultivars pointed to favorable and uniform moisture and temperature conditions (Panday et al., 2025), which may have buffered environmental stress. Conversely, the 2023 decline in stem diameter across all genotypes likely reflects climate-induced stress, including heat and late-season drought, factors known to suppress cambial activity and secondary thickening (Campiglia et al., 2017; Ortmeier-Clarke et al., 2023). Cultivars such as Bialobrzeskie and USO 31, which consistently produced narrower stems, may be better suited for dual-purpose or seed-oriented systems, where lower lignification can facilitate mechanical harvesting and decortication (Rahemi et al., 2021; Petit et al., 2020). These findings underscore the importance of matching cultivar architecture to production goals and environmental constraints, particularly in variable Midwestern agroecosystems.

Although the multi-location, multi-year experimental design across Missouri strengthened the environmental representativeness of our findings, we acknowledge that the use of only two replications per site may have reduced the overall statistical power of the study and potentially mask minor differences among cultivars. Nonetheless, the consistent trends observed across years and location, particularly for high-performing cultivars such as Jinma and Fibror 79, further support the robustness of our findings. Moreover, power analysis indicates that the experimental design was adequate to detect biologically meaningful differences among cultivars for key traits. At the Washington site, the minimum detectable difference was estimated at 5.34 plants m^{-2} for plant density and 0.62 kg m^{-2} for biomass, thresholds that are well within the observed range of cultivar variation.

These results suggest that large and agronomically relevant differences, such as those observed between high-performing cultivars (e.g., Jinma, Puma) and low-performing cultivars, were detected reliably. However, smaller differences among intermediate cultivars may have gone undetected, particularly under environmentally stressful conditions such as flooding in 2023. It is important to acknowledge that the 2023 flooding events at the Sikeston and Washington sites resulted in the exclusion of complete datasets from those environments. This data loss reduced the temporal balance of the three-year dataset and may have limited our ability to fully capture inter-annual variability across all locations. However, consistency in cultivar performance observed between 2021 and 2022, as well as the unaffected 2023 GWC site, indicates that the exclusion did not materially alter overall conclusion. However, future experiments should incorporate higher replication numbers (≥3 per site) or spatially augmented designs to enhance precision and reliability of genotype × environment interaction analyses.

Cannabinoid analysis

The observed variation in THC concentration among cultivars across environments highlights the complex genotype × environment (G×E) interactions that govern cannabinoid expression. Warmer temperatures, delayed harvest timing, and abiotic stressors such as drought have been shown to elevate THC accumulation, likely by extending the flowering phase or activating stress-induced biosynthetic pathways (Chandra et al., 2020; Berthold et al, 2020). Despite these fluctuations, cultivars such as Fibror 79, Futura 83, and Santhica 70 consistently remained below the legal THC threshold, affirming their suitability for compliant commercial production. The stable performance of these cultivars aligns with recent profiling studies, which confirm their low-THC genotypic backgrounds (Lindekamp et al., 2024; Nahler et al., 2019: Süzerer et al., 2023). At the same time, elevated CBD concentrations in cultivars like Bialobrzeskie, Fibror 79, Felina 32, and Ferimon suggest strong dual-purpose potential, offering value for both fiber and phytochemical applications. This confirms the importance of multi-trait selection strategies that integrate biomass performance with stable cannabinoid profiles (Small, 2015; Adesina et al., 2020).

Year-to-year and site-specific variation in CBD concentrations (Figure 8) highlights the strong influence of genotype × environment interactions. Higher CBD accumulation at Sikeston likely reflects favorable soil or microclimatic conditions relative to GWC farm, while elevated CBD levels in cultivars such as Bialobrzeskie and Fibror 79 in 2024 suggest responsiveness to improved agronomic or climatic conditions, consistent with Andre et al. (2016). In contrast, fiber-dominant cultivars including Jinma, Futura 83, and Santhica 70 maintained consistently low CBD levels, confirming their limited phytochemical potential and suitability for compliant fiber production. These inter-cultivar and inter-site patterns have direct implications for value chain specialization: cultivars with low cannabinoid accumulation are best suited for fiber systems, whereas genotypes expressing moderate to high CBD levels (e.g., Bialobrzeskie, Ferimon, Felina 32, and Fibror 79) offer potential for dual-purpose or phytochemical-oriented production. Overall, integrating agronomic performance with cannabinoid stability provides a practical framework for cultivar selection across industrial and pharmaceutical hemp value chains. Collectively, these findings emphasize the need for multi-year, multi-location evaluations of hemp cultivars, especially under variable climatic conditions, to ensure cannabinoid consistency and regulatory compliance. Advances in rapid cannabinoid quantification technologies, such as NIR spectroscopy (Jarén et al., 2022), and molecular characterization of biosynthetic enzymes like THCA and CBDA synthase (Van Bakel et al., 2011), offer promising avenues for precision phenotyping and cultivar certification. Moreover, the concept of sustainability in hemp production can be further interpreted through the input–output balance observed in this study. Industrial hemp demonstrated high biomass productivity under moderate input conditions, specifically, basal fertilization of 70-60–40 kg ha^{-1} (NPK) and largely rainfed irrigation supplemented only at critical growth stages. Across sites, cultivars such as Jinma and Fibror 79 produced between 6 and 12 t ha^{-1} of dry fiber yield, translating to an estimated nitrogen-use efficiency of 85–120 kg biomass per kg N applied, which is comparable or superior to conventional fiber crops like cotton and kenaf (Campiglia et al., 2017; Kaur et al., 2023). These results highlight hemp's capacity to achieve substantial biomass gains with limited nutrient and water inputs, thereby reducing its environmental footprint. Moreover, its short growth cycle and high biomass production contribute to atmospheric carbon sequestration, reinforcing its role as a climate-smart rotation as well as cover crops. Although this study did not include a full life-cycle assessment, the observed yield-to-input ratio provides a practical indication of hemp's potential as a sustainable, resource-efficient crop for Midwestern production systems.

Conclusion

This multi-year, multi-location study confirms the agronomic suitability and regulatory stability of selected industrial hemp (*Cannabis sativa L.)* cultivars for fiber production in southern Midwestern U.S. environments. Cultivars such as Jinma consistently outperformed others in key traits, including emergence, stem diameter, height, and biomass, demonstrating strong adaptation and yield potential. Futura 83 also showed promise under favorable conditions, supporting its use in targeted systems. Across all sites–year combinations, total THC levels remained below the 0.3% legal threshold, with low interannual variation, indicating cannabinoid stability and regulatory compliance. High-performance liquid chromatography (HPLC) enabled precise profiling of cannabinoid content, supporting reliable cultivar differentiation and market classification. These results highlight the central role of genotype-by-environment interactions in shaping both morphological and phytochemical traits and emphasize the need for site-specific cultivar deployment. The combination of agronomic consistency and cannabinoid reliability observed in this study strengthens the case for integrating industrial hemp into diversified, climate-adaptive cropping systems. Future efforts should prioritize genetic diversification, precision agronomy, and breeding strategies aimed at improving dual-use performance and cannabinoid uniformity under increasingly variable climatic conditions. Further studies incorporating fiber-quality assessments, such as fiber length, tensile strength and fineness, are warranted to complement the yield data presented here and to guide cultivar selection for specific industrial end uses.

Data availability statement

The datasets presented in this study can be found in online repositories. The names of the repository/repositories and accession number(s) can be found in the article/Supplementary Material.

Author contributions

SM: Conceptualization, Data curation, Investigation, Methodology, Resources, Validation, Visualization, Writing – original draft, Writing – review & editing. JA: Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Software, Validation, Visualization, Writing – original draft, Writing – review & editing. KT: Data curation, Formal analysis, Investigation, Methodology, Validation, Visualization, Writing – original draft, Writing – review & editing. PK: Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Visualization, Writing – original draft, Writing – review & editing. CA-M: Conceptualization, Data curation, Formal analysis, Funding acquisition, Investigation, Methodology, Project administration, Resources, Supervision, Validation, Visualization, Writing – original draft, Writing – review & editing. CC: Data curation, Investigation, Methodology, Project administration, Supervision, Validation, Visualization, Writing – review & editing. ER: Formal analysis, Investigation, Methodology, Resources, Validation, Visualization, Writing – review & editing. KN: Data curation, Formal analysis, Investigation, Methodology, Validation, Visualization, Writing – original draft, Writing – review & editing. SS: Data curation, Formal analysis, Investigation, Methodology, Validation, Visualization, Writing – original draft, Writing – review & editing. SP: Conceptualization, Formal analysis, Investigation, Methodology, Software, Validation, Visualization, Writing – original draft, Writing – review & editing. JP: Conceptualization, Data curation, Funding acquisition, Investigation, Methodology, Project administration, Resources, Supervision, Validation, Visualization, Writing – original draft, Writing – review & editing. BV: Conceptualization, Data curation, Formal analysis, Funding acquisition, Investigation, Methodology, Project administration, Resources, Supervision, Validation, Visualization, Writing – original draft, Writing – review & editing.

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Conflict of interest

The authors declared that this work was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Correction note

A correction has been made to this article. Details can be found at: 10.3389/fagro.2026.1822224.

Generative AI statement

The author(s) declared that generative AI was not used in the creation of this manuscript.

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