

Potential of Sorghum Stalk as Climate Resilient Resource for Paper Production

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Sorghum bicolor is a promising multi-purpose crop for the bioeconomy. To unfold its potential, genotypes must be matched to the respective industrial processes. Five sorghum genotypes were evaluated under contrasting field conditions in Southwest Germany, focusing on the stalk as a source of lignocellulosic biomass for paper production. Dual-purpose hybrids, particularly Ganymed and Tarzan, demonstrated high resilience, maintaining stable biomass and sugar yield under drought stress. Compositional analysis of the stalk showed substantial genotypic differences in extractive, carbohydrate, and lignin contents. Ganymed and Tarzan possessed higher glucose and xylose levels and lower extractives than KIT1 and Hannibal, making them more favorable candidates for biobased processing. Soda pulping results aligned with chemical profiles: Ganymed achieved the highest pulp yield (50.2%) and moderate kappa number (30.5), while KIT1 showed the lowest yield (38.0%) and the highest kappa number (40.3). Tarzan and Zerberus produced pulps with low kappa values (23.5 and 19.8 respectively) but moderately reduced yields due to hemicellulose loss (43.9% and 39.9%, respectively). Paper produced from Tarzan (6.6 mN.m²/g) had high tear indices compared to the other sorghum genotypes. Overall, Ganymed, Tarzan, and Zerberus showed strong suitability as lignocellulosic feedstocks, offering promising options for biorefinery and paper production.

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INTRODUCTION

The global transition towards a sustainable bioeconomy necessitates the development of efficient and resilient biomass feedstocks that can be integrated into multi-product biorefining systems (Bridgwater 2012; John *et al.* 2025). In the context of climate change, sorghum (*Sorghum bicolor* L. Moench) stands out among promising bioenergy crops because of its C4 photosynthetic pathway, high water-use efficiency, and adaptability to marginal lands (Tari *et al.* 2013; Vadez *et al.* 2014; Abuslima *et al.* 2022; Abuslima *et al.* 2025). Sweet sorghums are specialized to accumulate high amounts of soluble sugar in the stem, grain sorghums are bred for seed yield, and dual-purpose hybrids are developed to achieve both high biomass and satisfactory grain production (Almodares and Hadi 2009; Kanbar *et al.* 2020, 2021a,b,c, 2022).

One of the critical challenges in optimizing sorghum for the bioeconomy is the significant genotypic diversity in response to environmental stresses, especially drought. It is expected that summer droughts in Europe, particularly in Germany, will become more frequent and severe, posing a risk to agricultural productivity (Toreti *et al.* 2022). The drought resilience of sorghum is influenced by multiple mechanisms, such as osmotic adjustment (Blum 2009; Blum 2017), deep-root systems (Kanbar and Al Gehani 2013), and the stay-green trait (Borrell *et al.* 2014), which enables ongoing photosynthesis even in conditions of water deficit. The manifestation of these traits is significantly influenced by genotype-by-environment (G×E) interactions. Because sorghum performance is strongly mediated by G×E interactions (Souza *et al.* 2021), robust selection for climate resilience requires multi-environment evaluation under contrasting soil and moisture regimes to capture genotype-specific adaptation patterns and identify material suitable for variable growing conditions anticipated under climate change.

In addition to agronomic performance, the biochemical and structural composition of biomass play a crucial role in determining its suitability for industrial use. In first-generation biorefining, the concentration of fermentable sugars in the stem juice, indicated by °Brix, serves as an essential quality parameter (Almodares and Hadi 2009). In second-generation biorefining, which focuses on the structural carbohydrates in the cell wall, the composition of cellulose (glucan), hemicellulose (xylan), and lignin is crucial. This composition significantly affects the effectiveness of pretreatment and enzymatic hydrolysis, ultimately impacting the production of fermentable sugars (Zhao *et al.* 2020).

Lignin is a significant factor in biomass recalcitrance, obstructing enzymatic saccharification while enhancing energy yield in thermochemical conversion processes (Bridgwater 2012; Hubbe *et al.* 2025). Moreover, the non-structural, solvent-soluble components (extractives) might affect processing; water-soluble extractives promote fermentation, while lipophilic and phenolic chemicals may suggest alternate valorization pathways (Awika and Rooney 2004; Durand *et al.* 2024). A thorough evaluation of a novel feedstock must include its potential for material production. The pulping characteristics and subsequent papermaking properties of sorghum biomass serve as a direct indicator of its applicability in a lignocellulosic biorefinery. The soda pulping technique, using sodium hydroxide for biomass delignification, may elucidate genotypic variations in pulp yield and residual lignin (kappa number) (Alves *et al.* 2019). The pulping parameters dictate the physical and mechanical properties of the resultant paper, including tear strength, burst strength, and porosity, which define its end-use applications, ranging from high-quality printing paper to durable packaging materials (Hubbe *et al.* 2008; Hubbe 2014; Bajpai 2018). While earlier studies have typically concentrated on individual characteristics of sorghum, such as performance under agricultural stress or biochemical composition, integrated analyses linking field performance to end-product quality remain scarce. This study aims to bridge this gap by conducting a multi-tiered evaluation of five distinct sorghum genotypes-including sweet and dual-purpose types-under the contrasting soil and moisture conditions of Southwest Germany.

It is hypothesized that there is considerable genetic diversity not only in drought resilience but also in biochemical composition and technological properties, which will result in unique, genotype-specific valorization pathways in an integrated biorefinery. To investigate this, (1) the agronomic and sugar-related traits under both irrigated and rainfed environments at two locations were assessed, (2) the composition of extracts and cell walls was analyzed, (3) the pulping efficiency and paper quality were evaluated, and (4) these findings were integrated to recommend optimal genotype-application pairings for the bioeconomy. The biochemical and pulping analyses were

conducted on pooled samples from the irrigated treatment at the Forchheim site only, using fixed conditions. Consequently, the reported material properties reflect genotype-specific traits under well-watered conditions, and the potential effects of drought on these parameters were not evaluated in this study.

EXPERIMENTAL

Plant Materials

For this study five sorghum genotypes of *S. bicolor* L. Moench were chosen and grouped according to their use in agricultural practice (Table 1). These included sweet varieties (KIT1), and dual-purpose hybrids (Ganymed, Hannibal, Tarzan, and Zerberus). KIT1 is a sweet sorghum genotype developed by us using pedigree selection. All hybrids used in this study were bred by KWS company (KWS SAAT SE and Co. KGaA, Germany) with the targets of high yield and high disease tolerance for production of grain and biomass.

Table 1. List of Genotypes Used in this Study, their Agronomic Group, Type and Origin

Cultivars	Type	Pedigree	Source
KIT1	Sweet, stalk biomass	Variety	KIT, Germany
Hannibal	Grain, stalk biomass	Hybrid (<i>S. bicolor</i> × <i>S. bicolor</i>)	KWS, Germany
Tarzan	Grain, stalk biomass	Hybrid (<i>S. bicolor</i> × <i>S. bicolor</i>)	KWS, Germany
Ganymed	Grain, stalk biomass	Hybrid (<i>S. bicolor</i> × <i>S. bicolor</i>)	KWS, Germany
Zerberus	Grain, stalk biomass	Hybrid (<i>S. bicolor</i> × <i>S. bicolor</i>)	KWS, Germany

Field Experiment

The field experiment was carried out in the 2022 growing season at two locations in the Rhine Valley, Baden-Württemberg, Southwest Germany. The Ettlingen location, situated in the fertile Upper Rhine Plain (Ecological area 3a), had sandy loam to silty clay soil with a soil quality rating of 133. The Forchheim location near Karlsruhe, located in the less fertile Lower Rhine Plain (Ecological area 3b), featured clayey loam soil and a quality rating of 117 (Fig. 1). Despite similar climatic conditions (~742 mm annual precipitation; ~10.2 °C mean annual temperature), the soil variation offered a valuable gradient for evaluating genotype-by-environment interactions (Fig. 1).

In the first week of May 2022, seeds were sown at a depth of 2 to 4 cm. The experimental design was a randomized complete block design with three replications per genotype at each location. Each plot included six adjacent rows of 5.0 m in length, with a 75 cm gap between rows and 25 cm between plants. The two outer rows served as buffers, and data were collected from the two central rows. Fertilization (both organic and chemical) was applied as needed based on soil analysis. Weeding and crop protection were applied as required.

At the Forchheim location, two different moisture regimes were implemented: an irrigated treatment where soil moisture was maintained above 70% of field capacity, and a non-irrigated (rainfed) treatment. Soil moisture was monitored only in the

irrigated treatment using a portable moisture probe (Trime®-Pico 32, IMKO GmbH, Ettlingen, Germany) at 10 cm depth with five replications per plot, with measurements taken every five days. Irrigation was applied when soil moisture fell below the 70% threshold. The Ettlingen site work was conducted under natural rainfed conditions only, with no irrigation or active soil moisture monitoring. Drought stress was naturally imposed during the 2022 growing season due to high temperatures and low precipitation, particularly in July and August where monthly rainfall remained below 20 mm (Supplementary Fig. S1).



Location	Ettlingen	Forchheim (Rheinstetten)
Elevation (sea level, m)	133	117
Precipitation (mm) (long-term mean)	742	742
Annual temperature (°C) (long-term mean)	10.1	10.2
N _t (%)	0.096	0.064
pH	5.8	6.5
Bulk density (g.cm ⁻³)	1.33	1.2 to 1.5
Soil texture	35% sand, 30% clay, 35% silt	60.2% sand, 13.7% clay, 26.1% silt
Soil type	Clayey loam	Loamy sand

Fig. 1. Key environmental conditions and soil types at the Ettlingen and Forchheim field trial locations

The experimental design presented an unbalanced structure for genotype-by-environment (G×E) interaction analysis, as the irrigation treatment was implemented only at the Forchheim location. This means the factors of location (Ettlingen vs. Forchheim) and water regime (irrigated vs. rainfed) were not fully crossed. While this design was implemented to reflect realistic regional growing conditions and logistical constraints, it precludes a fully factorial analysis of location × water interactions. Therefore, for the purpose of G×E assessment, the three field conditions, Forchheim-irrigated, Forchheim-rainfed, and Ettlingen-rainfed, were treated as distinct environments representing a gradient of soil quality and water availability in Southwest Germany.

Plants were harvested at the dough seed stage, recognized for producing optimal stem sugar concentrations (Undersander *et al.* 1990; Kanbar *et al.* 2020, 2021c). From each replicate, five randomly chosen plants from the center of the plot were harvested, and the following agronomic parameters were recorded: Time to flower (day), plant height (cm), cane weight (g), juice yield (mL/plant), and sugar content (°Brix). A cane crusher (VEVOR Juicer 110LBS/H, India) was used to extract juice and measure sugar concentration in °Brix (1 °Brix indicates 1 g soluble solids per 100 mL stalk juice) using a manual refractometer (Model PAL, Atago Co. Ltd., Tokyo, Japan), as well as juice volume (see Kanbar *et al.* 2021c; Kanbar *et al.* 2022).

Sample Collection and Preparation

Sorghum stalk samples from each genotype were collected from the irrigated plots at the Forchheim site. For each plot, five plants were randomly selected from the central area and harvested. The stalks from the three biological replicates were pooled to create a composite sample representative of each genotype. The stalks were then cut into smaller pieces for pulping experiments, and a subsample was milled using a disc mill (SM 200, Retsch, Haan, Germany) for compositional analysis. All subsequent

chemical analyses were performed on these pooled composite samples, with technical replicates as specified below.

Extractive Content

The extractive content was determined using an accelerated solvent extraction system (Dionex ASE 350, Thermo Scientific, USA) at elevated temperature. Sequential extractions were performed using petroleum ether, acetone/water (9:1, v/v), and water. Each sample was analyzed in duplicate and reported on an oven-dry basis.

Carbohydrate Composition

The monomeric carbohydrate and lignin contents of extractive-free samples were determined following the two-step acid hydrolysis procedure described by Lorenz *et al.* (2016). Briefly, 200 mg (± 10 mg) of oven-dried, milled sample was hydrolyzed with 2 mL of 72% H₂SO₄ at 30 °C for 1 h. The mixture was then diluted with 56 mL of ultrapure water and subjected to post-hydrolysis in an autoclave at 120 °C and 1.2 bar for 40 min. The acid-insoluble residue (equivalent to Klason lignin) was collected by filtration through a G4 sintered glass frit, washed, dried at 105 °C, and weighed. The filtrate was analyzed for monomeric sugars (glucose, xylose, mannose, arabinose, galactose, rhamnose, and cellobiose) using borate anion-exchange chromatography (borate-HPAEC) according to Lorenz *et al.* (2016). Each sample was analyzed in triplicate.

Soda Pulping

Soda pulping was performed using a 15 L electrical rotary digester. For each experiment, 400 g (oven-dry weight) of chopped sorghum stalk was pulped with 16% (w/w) NaOH (Table 2), based on the oven-dry raw material, at a solid-to-liquid ratio of 1:4. The temperature was raised to 160 °C within 60 min and maintained for an additional 60 min under a pressure of approximately 6 bar (Table 2). After cooking, the pulp was thoroughly washed with water to remove residual chemicals and black liquor. The kappa number was determined in duplicate according to TAPPI standard T236 cm-85.

Table 2. Standard Soda Pulping Conditions Used for All Sorghum Stalk Genotypes

Cooking Conditions	
Maximum temperature [°C]	160
Preheating time [min]	60
Pulping time [min]	60
NaOH-Concentration [w/w %]	16
Pressure [bar]	6
Solid-to-liquid ratio	1:4

Hand Sheet Preparation and Evaluation

The never-dried pulp was beaten in a Jokro mill (FRANK-PTI, Germany) according to the German standard Zellcheming V/5/60 (1960) at 150 rpm for 5 min. The beaten pulp was disintegrated according to ISO 5263-2:2004, and the beating degree (°SR) was determined using a Schopper-Riegler freeness tester (Karl Schröder KG, Germany). Laboratory handsheets with a basis weight of approximately 80 g/m² were prepared using a Rapid-Köthen sheet-forming machine (FRANK-PTI, Germany). The sheets were conditioned overnight at 23 $\pm$ 1 °C and 55 $\pm$ 2% relative humidity prior to testing. Four handsheets were tested for each pulp, and the reported values represent the average of the measurements. The compression, burst, and tear indices were

measured according to DIN 54518:2004, ISO 2759:2014, and ISO 1974:2012-09, respectively, using instruments from FRANK-PTI GmbH (Germany). All results were normalized to the measured sheet grammage.

Statistical Analysis

Phenotypic data from the field experiment were analyzed using linear mixed-effects models in R (version 4.3.1; R Core Team 2023) to appropriately account for the unbalanced design structure, where irrigation was implemented only at the Forchheim location. The model treated location as a random effect and included water treatment as a factor nested within location, reflecting that water availability differed structurally between sites. The full model specification was,

$$Y_{ijkl} = \mu + G_i + L_j + W_k(L_j) + (G \times L)_{ij} + (G \times W)_{ik}(L_j) + \epsilon_{ijkl} \quad (1)$$

where Y_{ijkl} is the observed trait value, μ is the overall mean, G_i is the fixed effect of genotype i ($i = 1, \dots, 5$), L_j is the random effect of location j ($j = 1, 2$; $L_j \sim N(0, \sigma_L^2)$), $W_k(L_j)$ is the fixed effect of water treatment k nested within location j , $(G \times L)_{ij}$ is the genotype $\times$ location interaction, and $(G \times W)_{ik}(L_j)$ is the genotype $\times$ water treatment interaction nested within location j . Mean comparisons were performed using the least significant difference (LSD) test at a 5% significance level. The LSD values were calculated from the mixed model output.

For chemical analyses and pulping trials, technical replications were performed; no biological replication was included. As statistical analyses were not performed for compositional, pulping, and handsheet data, observed differences among genotypes should be interpreted as indicative trends rather than statistically validated differences.

RESULTS AND DISCUSSION

Drought Stress Impact on Agronomic Traits Reveals Genotypic Variation in Resilience Strategies

The linear mixed-effects model analysis confirmed significant effects of genotype, environment, and their interaction ($G \times E$; $p < 0.05$, Supplementary Table S1) on all measured agronomic traits, underscoring that drought response was strongly genotype- and environment-dependent. Drought stress substantially reduced plant growth, although the magnitude of reduction varied considerably among genotypes and locations (Table 3).

At Forchheim, where both irrigated and rainfed treatments were implemented, drought led to significant decreases in plant height and cane weight across most genotypes. This reduction aligns with the well-documented adaptive strategy of sorghum to limit transpirational water loss under moisture deficit (Blum 2009; Alhajturki *et al.* 2012; Almodares *et al.* 2013). KIT1 exhibited the greatest sensitivity, with plant height declining by 52 % and cane weight by 62 % under drought (Table 3). Such pronounced sensitivity may indicate a limited capacity for osmotic adjustment or other drought-tolerance mechanisms often associated with drought-susceptible genotypes (Vadez *et al.* 2014). In contrast, Tarzan demonstrated notable stability, maintaining cane weight under drought (358.3 g/plant) nearly equal to its irrigated performance (356.7 g/plant). This suggests the presence of effective tolerance mechanisms, which could include deeper rooting or sustained turgor maintenance (Vadez *et al.* 2014; Amelework *et al.* 2015).

Performance varied between the two rainfed sites, showing that drought resilience depends on environmental conditions. Most genotypes grew better at Ettlingen than at Forchheim under drought. For example, KIT1 reached 199.6 cm at

Ettlingen but only 132.3 cm at Forchheim (Table 3). This difference likely resulted from higher soil moisture at Ettlingen, where silty clay soil retains water better than the sandy loam at Forchheim (Fig. 1; Brady and Weil 2008). Ganymed showed clear adaptation to Ettlingen: its cane weight under drought there (492.7 g/plant) was even higher than under irrigation at Forchheim (435.9 g/plant, Table 3). This indicates that genotype performance can change with soil and climate, highlighting the importance of multi-environment testing (George *et al.* 2014).

Table 3. Agronomical and Product Parameters for a Field Experiment, where Five Genotypes of Sorghum were Cultivated Side by Side in Two Sites, Forchheim and Ettlingen (Baden-Württemberg, Southwest Germany) either under Drought Stress (Rainfed Fields in the Extreme Summer of 2022) or under Irrigation in the Forchheim Site

Trait	Genotype	Forchheim-irrigated	Forchheim-drought	Ettlingen-drought
Plant height (cm)	KIT1	277.0 ± 7.0 a	132.3 ± 17.6 b	199.6 ± 17.0 c
	Hannibal	310.1 ± 9.1 d	184.8 ± 5.1 e	276.7 ± 10.8 a
	Tarzan	313.6 ± 19.7 d	193.8 ± 15.2 e	312.8 ± 4.1 d
	Ganymed	332.8 ± 18.2 d	193.9 ± 4.3 e	302.6 ± 12.0 d
	Zerberus	383.1 ± 13.7 f	267.7 ± 12.1 g	389.3 ± 4.2 f
Cane weight (g.plant ⁻¹)	KIT1	358.4 ± 23.7 a	135.0 ± 27.8 b	296.1 ± 19.9 a
	Hannibal	479.8 ± 40.4 c	340.1 ± 13.6 d	434.9 ± 40.2 c
	Tarzan	356.7 ± 37.7 a	358.3 ± 47.8 a	349.4 ± 23.8 a
	Ganymed	435.9 ± 37.0 c	350.4 ± 12.5 a	492.7 ± 17.7 e
	Zerberus	545.8 ± 49.8 f	358.7 ± 2.4 a	496.4 ± 31.7 e
Juice yield (ml.plant ⁻¹)	KIT1	127.7 ± 8.2 a	44.0 ± 10.9 b	122.1 ± 9.3 a
	Hannibal	197.2 ± 20.6 c	137.2 ± 6.0 d	193.9 ± 16.0 c
	Tarzan	126.1 ± 21.1 a	153.7 ± 20.8 d	154.2 ± 21.8 d
	Ganymed	169.3 ± 20.3 ac	163.0 ± 9.8 d	250.7 ± 7.7 e
	Zerberus	239.9 ± 23.3 f	164.3 ± 6.6 d	195.4 ± 10.8 c
Sugar content (°Brix)	KIT1	15.6 ± 0.5 a	17.1 ± 1.4 a	15.3 ± 0.7 a
	Hannibal	12.1 ± 0.6 b	10.5 ± 1.8 b	12.9 ± 0.3 b
	Tarzan	12.4 ± 0.3 b	10.6 ± 1.5 b	11.1 ± 0.4 b
	Ganymed	11.6 ± 0.6 b	7.9 ± 1.1 c	12.5 ± 0.8 b
	Zerberus	11.0 ± 0.4 b	9.0 ± 0.6 bc	12.9 ± 0.8 b
Time to flower (day)	KIT1	69.7 ± 0.7 a*	76.3 ± 0.7 b	70.3 ± 0.3 a
	Hannibal	88.3 ± 1.5 c	118.0 ± 2.3 h	89.3 ± 2.0 c
	Tarzan	87.3 ± 2.6 c	103.7 ± 4.5 g	88.7 ± 1.8 c
	Ganymed	88.7 ± 2.0 c	116.7 ± 2.6 h	90.3 ± 1.8 c
	Zerberus	116.0 ± 2.1 e	129.3 ± 0.9 i	112.0 ± 1.5 d

* Data presented as mean ± standard error of three biological replications per genotype (n = 3). Different letters within each trait indicate significant differences at $p < 0.05$, according to LSD test comparing all 15 genotype × treatment combinations.

Sugar-related traits responded to drought in a manner consistent with the classic trade-off between biomass production and osmotic adjustment (Chaves *et al.* 2003; Turki *et al.* 2011). At Forchheim, sugar concentration (°Brix) increased under drought, most markedly in KIT1 (from 15.6 to 17.1 °Brix; Table 3), a response associated with solute accumulation to maintain cellular function at low water potential (Blum 2009). However, this adjustment came at a substantial cost in juice yield (KIT1: 127.7 to 44.0 mL/plant), highlighting the inherent conflict between survival (high solute concentration) and productivity (high yield) under water-limited conditions (Alhajturki *et al.* 2012; Tari *et al.* 2013; Babst *et al.* 2021).

At Ettlingen, Ganymed presented a favorable combination for bioenergy production, retaining both appreciable sugar content (12.5 °Brix) and high juice yield (250.7 mL/plant) under drought (Table 3). This ability to sustain yield and sugar concentration simultaneously under stress suggests improved water-use efficiency, which is a key target for breeding in water-limited regions (Xie and Su 2012; Mengistu *et al.* 2016). In contrast, under the more severe Forchheim-drought, Ganymed's °Brix dropped to 7.9, indicating that its capacity to maintain metabolic homeostasis was compromised under intense stress, resulting in diluted solutes or impaired sugar transport. Flowering time responses also varied by genotype and stress intensity (Table 3). Severe drought at Forchheim delayed flowering across all genotypes, whereas the milder stress at Ettlingen caused minimal delay, reinforcing that stress severity critically modulates phenotypic expression and that genetic differences become most apparent under high-stress conditions (Almodares *et al.* 2013).

The observed variation in drought performance has direct implications for breeding and cultivar deployment. Tarzan's stable biomass production across contrasting moisture regimes supports its potential for dual-purpose systems in drought-prone areas, consistent with earlier findings (Kanbar *et al.* 2022). Ganymed's ability to maintain favorable sugar-related traits under moderate drought indicates suitability for sugar-based bioenergy applications, particularly on soils with good water-retention capacity (Kanbar *et al.* 2022). These results align with previous work showing that the same dual-purpose hybrids (Ganymed, Tarzan, Hannibal) can yield heavier, sugar-rich internodes and higher ethanol output compared to other sorghum types (Kanbar *et al.* 2021a). Collectively, the data support Ganymed and Tarzan as promising, climate-resilient genotypes for biomass and sugar production in Central European conditions, while underscoring that G×E interactions necessitate multi-environment evaluation for robust genotype recommendation.

Chemical Composition of Sorghum Genotypes

Compositional analyses of sorghum stalk genotypes are essential for evaluating their potential as lignocellulosic feedstocks. The extractives profile of irrigated sorghum stalks grown at the Forchheim site showed clear variation in both the quantity and composition of solvent-soluble compounds.

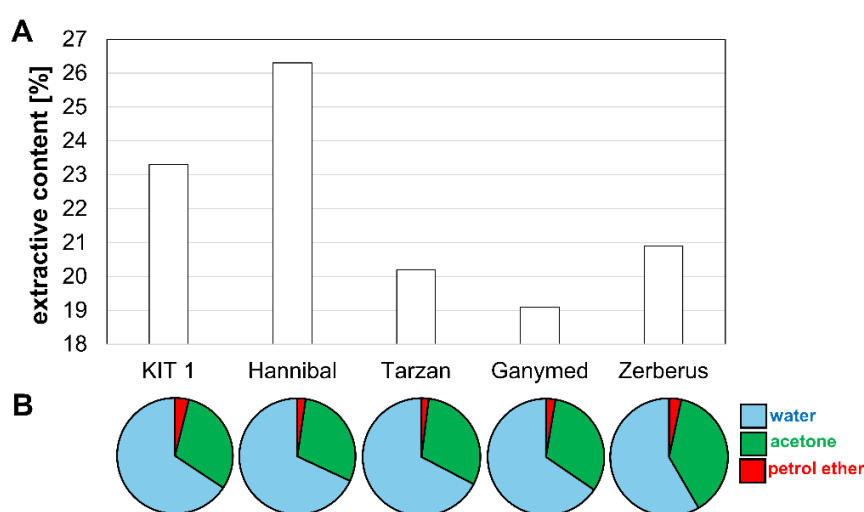


Fig. 2. Extractive content of five sorghum stalk genotypes. Total extractive content (**A**); Distribution of water-soluble, acetone/water-soluble, and petrol ether soluble fractions (**B**)

The total extractive content varied from 19.0% in Ganymed to 26.3% in Hannibal (Fig. 2A). Across all genotypes, water-soluble extractive constituted the predominant fraction (Fig. 2B). These results are in line with previous reports showing high extractive contents in sorghum stalk (29.1%) and are comparable to those reported for other agricultural residues such as corn stover (21.6%) and wheat straw (16.5%) (Theerarattananon *et al.* 2012). The predominance of water soluble constituents is a typical feature of agricultural residue biomass.

The genotypes exhibited apparent differences in metabolic profiles. KIT1, which had high sugar content ($^{\circ}$ Brix) and juice yield, also showed high total extractives (23.3%), which were dominated by water-soluble compounds (15.3%). This indicates substantial accumulation of soluble sugars (sucrose, glucose, fructose) and amino acids (Fig. 2B), characteristic of genotypes with high metabolic activity favorable for fermentation processes. Ganymed had the lowest total extractives content (19.0%), with 12.5% water-soluble compounds. The acetone-water extractive fraction ranged from 6.1% in Ganymed to 8% in Zerberus and was relatively similar across genotypes. This fraction typically contains secondary metabolites such as phenolic compounds and flavonoids, which have been extensively studied in sorghum tissues (Awika and Rooney 2004), indicating a considerable allocation towards defense-related compounds. The extractive profiles therefore indicate different carbon allocation strategies: Hannibal and KIT1 allocate more carbon to easily fermentable substrates, whereas Zerberus places more on secondary metabolism.

As illustrated in Fig. 3, slight variation in glucose, xylose, and Klason lignin contents was observed among sorghum genotypes. The extractive free Ganymed, Tarzan, and Zerberus genotypes exhibited relatively higher glucose contents 46.5%, 46.3%, and 46.5%, respectively (Fig. 3A) along with corresponding xylose contents of 24.4%, 24.7%, and 23.8% (Fig. 3B).

Lignin content across all genotypes was lower than wood biomass and comparable to other agricultural residues. KIT1 genotype displayed slightly lower lignin content than the other genotypes (Fig. 3C), suggesting that it may require less severe pretreatment conditions during processing.

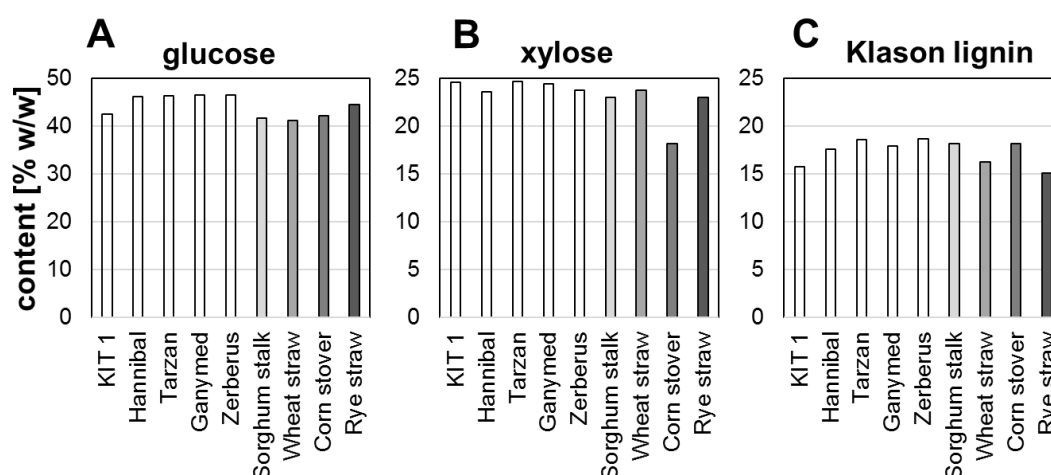


Fig. 3. Chemical composition: Glucose (A), Xylose (B) and Klason lignin (C) of extractive-free sorghum genotypes and other agricultural resources (Theerarattananon *et al.* 2012)

When expressed on an extractive-free basis, the carbohydrate contents of the sorghum genotypes examined here were comparable to values reported in earlier studies for sorghum and other agricultural residues (Theerarattananon *et al.* 2012). The high xylose content in all sorghum genotypes is particularly significant, as hemicellulose

sugars are co-products in a biorefinery context (Sipos *et al.* 2009; Li *et al.* 2010; Menegol *et al.* 2014).

Overall, the observed difference in chemical composition among the sorghum genotypes have important implications for biomass utilization. Variations in extractives, carbohydrates, and lignin may influence processing behavior, pretreatment requirements, and suitability for specific application.

Pulping Performance and its Paper Strength Properties of Sorghum Genotypes

Soda pulping of sorghum genotypes was performed under standard pulping conditions as previously described. This work was carried out with the stalks from the irrigated plots at Forchheim. The resulting pulp yields and kappa numbers are presented in Figs. 4A and 4B. Pulp yields ranged from 38.0% in KIT1 to 50.2% in Ganymed, demonstrating clear genotypic differences in pulping efficiency. Ganymed produced the highest yield (50.2%) together with a moderate kappa number (30.5), while KIT1 showed the lowest yield (38.0%) and the highest kappa number (40.3). Under the fixed soda pulping conditions applied in this study, KIT1 exhibited lower pulp yield and higher kappa number compared to other genotypes, indicating less favorable delignification behavior. In comparative terms, this suggests that KIT1 is less suitable for papermaking than the other genotypes evaluated under the condition tested.

This behavior aligns with the previously observed chemical composition of Ganymed, where it showed relatively lower extractive content (Fig. 2A) and higher glucose content (Fig. 3A). The strong correlation between high glucan content and high pulp yield (Fig. 4A) is well-established, since cellulose is the core component of pulp fiber (Bajpai 2018).

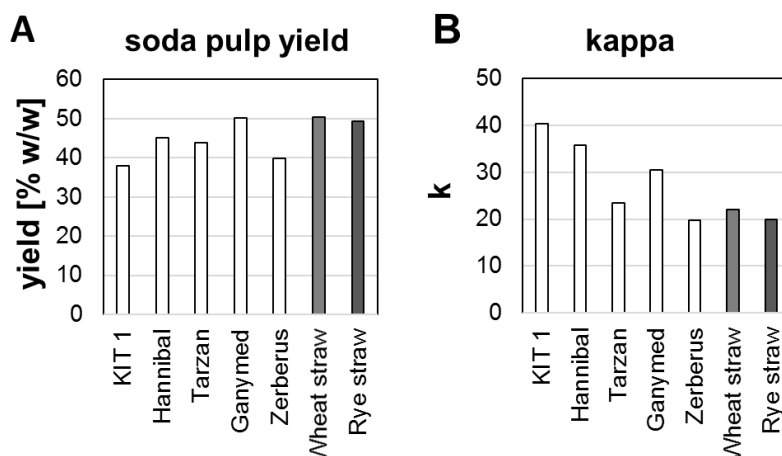


Fig. 4. Pulp yield (A) and kappa number (B) of sorghum genotype in comparison with wheat and rye straw (Salehi *et al.* 2014).

Tarzan and Zerberus produced pulps with the lowest kappa numbers (23.5 and 19.8, respectively), values comparable to wheat and rye straw (Fig. 4B). However, their pulp yields (43.9% and 39.9%, respectively) were slightly lower than those of reference materials. Low kappa number usually associated with lignin that is rich in less-condensed bonds, such as β -O-4 linkages, which are more easily cleaved during chemical pulping (Yao *et al.* 2022). The lower yields of Tarzan and Zerberus suggest substantial dissolution of carbohydrates, particularly hemicellulose during pulping. Hemicellulose is particularly vulnerable to degradation in alkaline environments, and its loss directly impacts the final pulp yield (Sixta 2006).

Despite its relatively low lignin content, KIT1 exhibited the highest kappa number and the lowest pulp yield, indicating that its lignin was more recalcitrant to delignification while its carbohydrates were highly susceptible to alkaline degradation (Figs. 4A, B) (Gierer 1980; Sjostrom 1993).

In summary, the pulping results indicate a significant trade-off between the degree of delignification and the preservation of carbohydrate. Compared with other agricultural residues, most sorghum genotypes show competitive potential as raw materials for pulp and paper production. Genotypes such as Ganymed represent promising candidates for papermaking, with additional potential for integration into biorefinery pathways. It should be noted that chemical composition and pulping evaluations were conducted on pooled stalk samples from the irrigated Forchheim site only, without biological replication across environments. Therefore, observed genotypic differences in chemistry and pulping are indicative but cannot be statistically generalized or linked to environmental variability. The findings should be interpreted as specific to the conditions tested.

To further evaluate the biomass suitability, the physical and mechanical properties of the handsheets made from the sorghum pulps were examined (Table 4). The burst index of the sorghum papers ranged from 2.0 to 2.8 kPa·m²/g, most of the genotypes showed values higher than those reported for bagasse, wheat straw pulps, and recycled paper pulp (Salehi *et al.* 2017a,b). Tarzan exhibited the lowest burst index (2.0 kPa·m²/g) and in contrast Zerberus showed the highest burst index (2.8 kPa·m²/g). However, both genotypes showed high tear indices (Tarzan: 6.6 mN·m²/g; Zerberus: 6.1 mN·m²/g). This suggests strong inter-fiber bonding, which is characteristic of flexible, well-delignified fibers that form a dense, uniform sheet network (Bajpai 2018).

Pulps from Tarzan and Zerberus resulted in papers with significantly different structural properties. They demonstrated very high porosity (814.6 mL/m and 630.0 mL/m) and roughness (1861.0 mL/m and 1402.6 mL/m), nearly twice that of other sorghum varieties and the reference straw pulps. The compression index, an indicator of sheet stiffness and resistance to crushing, was relatively consistent across all genotypes, though slightly higher for KIT1 and Hannibal (Table 4).

Table 4. Paper Strength and Related Properties from Sorghum Genotypes

	KIT1	Hannibal	Tarzan	Ganymed	Zerberus
Burst Index (kPa·m ² /g)	2.4	2.6	2.0	2.7	2.8
Tear Index (mN·m ² /g)	5.0	4.5	6.6	6.0	6.1
Porosity (mL/m)	213.0	335.4	814.6	334.0	630.0
Roughness (mL/m)	1282.0	920.3	1861.0	1238.0	1402.6
Compression Index (Nm/g)	30.7	30.0	27.1	28.2	26.7
Bulk (cm ³ /g)	2.1	1.8	2.0	2.0	2.0
SR ^o	23.0	21.0	18.0	21.0	18.0

CONCLUSIONS

1. The five sorghum genotypes demonstrated different drought response strategies. Tarzan maintained a high level of biomass stability, whereas Ganymed fared better in terms of sugar yield under stress. Their performance was greatly affected by specific soil conditions at each site, emphasizing the importance of genotype-by-environment interactions.

2. Significant variations in carbon allocation were observed, with Hannibal and Ganymed accumulating considerable amounts of water-soluble sugars suitable for fermentation. In contrast, Zerberus invested significantly on acetone-soluble secondary metabolites, highlighting their distinct biochemical aptitudes for various biorefinery pathways.
3. Chemical composition analysis showed that Ganymed, Tarzan, and Zerberus had lower extractive content and higher glucose contents compared with KIT1 and Hannibal. These characteristics make them candidates for paper production. Overall, the chemical composition of all genotypes were comparable to common non-wood agricultural residues such as wheat straw, corn stover, and rye straw.
4. Soda pulping performance of sorghum stalk was consistent with the chemical analysis: Ganymed, Tarzan, and Zerberus genotypes produced higher pulp yields and lower kappa numbers than KIT1 and Hannibal genotypes. Their tear indices were also higher.
5. Based on chemical composition and soda pulping results obtained from pooled samples collected at a single site (irrigated Forchheim site), the sorghum genotype Ganymed, Tarzan, and Zerberus demonstrate strong potential as lignocellulosic feedstock for paper production.
6. It is highly recommended that further studies be conducted on the separate characterization of sorghum pith and bark. Their potential valorization route may differ based on their chemical composition. The bark could be used as a fiber source for paper pulp production, while the pith could serve as a feedstock for sugar fermentation. Additionally, research should be conducted to assess how this separation could be implemented in an industrial preprocessing chain and to determine whether pith/bark ratio differ among genotypes or growing environments, as these factors may influence process efficiency and product quality.

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Author Contributions

Conceptualization: A.K., B.S., P.N.; Funding acquisition: B.S., K.S., P.N.; Methodology: A.K., H.L., B.S., P.N.; Investigation: A.K., H.L., K.S.; Formal analysis: A.K., H.L.; Data curation: A.K., H.L.; Visualization: A.K., H.L., P.N.; Resources: P.N., A.K., K.S., B.S.; Supervision: P.N., B.S.; Writing – original draft: A.K., H.L.; Writing – review & editing: P.N., B.S. All authors have read and agreed to the published version of the manuscript.

Conflict of Interest

There are no conflicts to declare.

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APPENDIX
Supplementary Materials

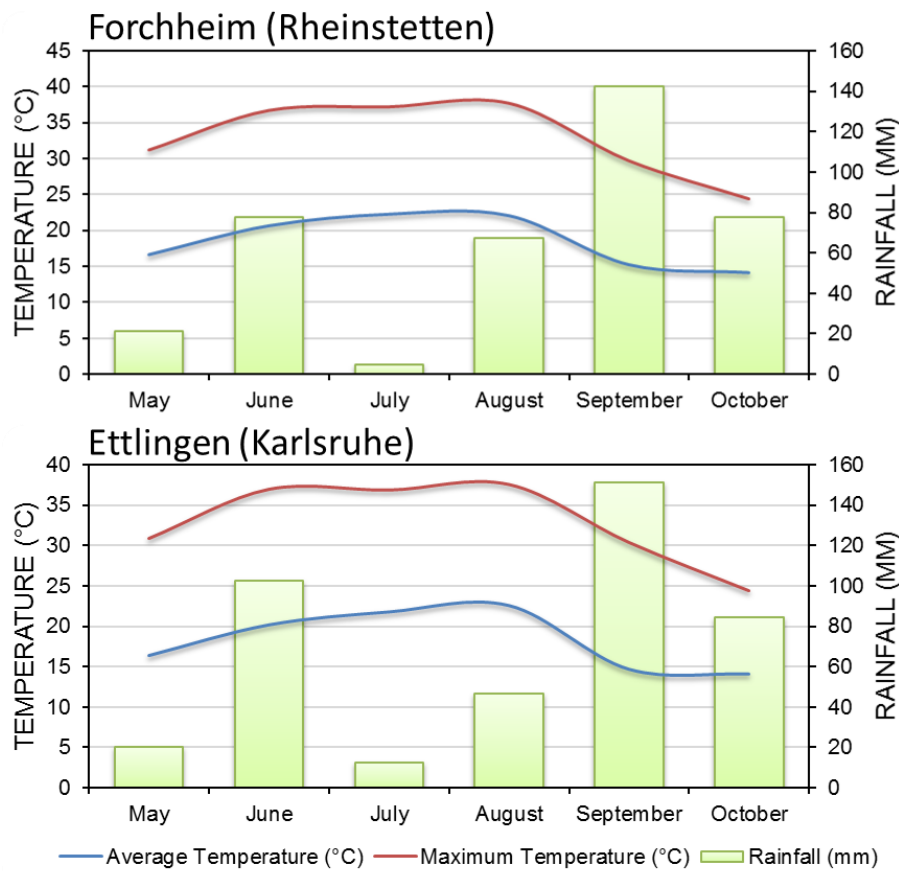


Fig. S1. Monthly weather conditions during the 2022 growing season at the Forchheim and Ettlingen experimental sites in Southwest Germany. Data source: www.klimadiagramme.de.

Table S1. Detailed Analysis of Variance for Sorghum Traits under Three Environmental Conditions Showing Mean Squares, and Significance Levels Based on the Nested Model Accounting for Missing Irrigation Treatment at Ettlingen Location. Significance Levels: ** $p < 0.01$, * $p < 0.05$, ns = Not Significant. Error df = 30 for all Traits.

Source	df	Time to flower	Plant height	Cane weight	Juice yield	°Brix
Genotype (G)	4	24.85**	18.67**	12.34**	9.87**	15.43**
Location (L)	1	0.12ns	15.43**	8.76*	5.43*	2.34ns
Water (L)	1	45.32**	62.89**	28.91**	34.21**	8.76**
G × L	4	2.34*	3.21*	1.89ns	2.12*	1.23ns
G × Water(L)	4	8.76**	12.45**	6.54**	7.89**	3.45*
Error	30					
Total	44					