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Research paper

Biochemical Response of Ten Varieties of Kenaf (Hibiscus Cannabinus) to Water Stress

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Abstract

Kenaf is a common wild crop of tropical and subtropical Africa and Asia with numerous applications. However, it required optimal water for growth and yields which may be a constraint in tropical areas. This study aims to investigate the biochemical response of ten kenaf varieties to water stress to develop drought-tolerant kenaf. Ten kenaf varieties (Ifeken 400, Ifeken 100, Cuba 108, Au-72, Tianung, Vi-100, A60-282, AC-313, Au-754, and Au-2452) were planted n=5 in a pot of soil in a completely randomised block design. They were arranged and divided into five groups according to the days of watering (WR10, WR8, WR6, WR4). WR10 (watered on every tenth day), WR8 (watered every eight days), WR6 (watered every six days), WR4 (watered every four days), while the control received water as required. The plants were later harvested at maturity (110 DAP); Compatible solutes (soluble protein, soluble carbohydrate and proline), lipid peroxidation, and ascorbic acid, photosynthetic pigments (chlorophyll a, b and carotenoid), and antioxidant enzymes (SOD, POD and catalase) were examined. Data were analysed using anova at ($P \leq 0.05$). The results showed that among the ten varieties of kenaf examined, Ifeken 400, Ifeken 100, Tianung and Cuba 108 showed the highest values in all the biochemical parameters, which indicates that these varieties could be tolerant to water stress and could be useful in an area with little or no rainfall.

Keywords: *Kenaf; Water Stress; Watered; Biochemical; Varieties.*

1. Introduction

Kenaf is an annual crop cultivated mainly for its fibre, although it's useful for other purposes like oil (which could be consumed as edible vegetable oil or used for cosmetics, industrial lubricants, and biofuel production (Bhardwaj et al.,1995; Swingle et al.,1978; Toole et al.,1960; and Ching et al.,1993). However, kenaf is a highly productive crop that grows well under favourable temperature and soil moisture and therefore takes up considerable quantities of nutrients from the soil (Tamargo et al.,1954, William,1966, White et al.,1971, Webber,1992). Meanwhile, good quality kenaf seed, under favourable temperature and moisture, will produce vigorous seedlings that emerge in 3 to 5 days and attain heights of 30 to 40 cm in three weeks (Xu et al.,2020; Chee and Kar, 2021; Suriyajantratong,1973, Webber,1994; Dengjie et al.,2007). The importance of water for kenaf growth has been studied. In fact, it has been shown that kenaf grown in the Wet tropics is likely to thrive well, but where there is irregular rainfall, irrigation will be required (Raghad et al.,2026; Molly et al.,2024; Wilson et al.,1965; Ali et al.,2025; Burnside and Williams, 1968; Higgins and White,1969; Whiteley et al.,1967; Jones et al.,1955; Webber and Bledsoe,1993). With respect to these important factors in kenaf growth, there is a need to determine some biochemical indicators of suboptimal water availability to kenaf growth to give an insight into how to develop kenaf varieties that could tolerate water deficit. However, increasing antioxidant enzyme activities have been linked to some genes, such as HcERF2, which is a positive regulator of drought tolerance. Additionally, HcWRKY44 is also linked to ABA signaling, and HcLEA113, a late embryogenesis abundant protein, is another gene implicated. (Luo et al., 2023, Niu et al., 2022) Also, kenaf modifies proteins, including V-type proton ATPase subunit G1-like, which is essential for energy management, to modify metabolism under drought. Also, studies have shown that six separated Histone Deacetylase genes (HcHDA2, HcHDA6, HcHDA8, HcHDA9, HcHDA19, and HcSRT2. Transcriptome analysis, the discovery of important transcription factors, and the use of molecular markers to find differentially expressed genes (DEGs) that support drought resistance are examples of recent developments (Kashif et al., 2020). Therefore, this study aims to determine biochemical attributes of kenaf exposed to water stress in order to identify and develop kenaf varieties with high yield and desirable levels in stress conditions, most especially in some temperate regions marked by irregular or no rainfall.

2. Materials and Methods

The experiment was carried out in a screen house at I.A.R&T, Ibadan. The seeds of kenaf (number of seeds=5) were sown in each pot containing 10kg of soil in a completely randomized blocked design (CRBD) with 3 replicates for each group in a rain-free screen house. The pots were arranged and divided into five groups according to the days of watering [Water Restriction: WR, (WR_{10}, WR_8, WR_6, WR_4). WR_4 group was (watered on every 4^{th} day), WR_6 (watered on every 6^{th} day), WR_8 (watered every 8^{th} day), WR_{10} (watered every 10^{th} day), while the control received water as required. A portable moisture meter was used to check the water content, temperature, and relative humidity every week. The plants were maintained at 80% relative humidity, soil pH 5.6 and temperature 35ºC At maturity both leaves and seeds were harvested at maturity (110 Day After Planting) and analysed for viz: Photosynthetic pigments (chlorophyll a, b and carotenoid), antioxidant enzymes [(Superoxide dismutase (SOD), Peroxidase: (POD) and Catalase], Compatible solutes (soluble protein, soluble carbohydrate and proline), lipid peroxidation and ascorbic acid were determined.

2.1. Determination of photosynthetic pigments

Chlorophyll a, b, and carotenoids were determined according to the methods of Masood et al. (2019) with slight modifications. 1g of leaf was ground in 85% acetone, and centrifugation was done for 20 minutes at 15000rpm and the filtrate was kept in a coolant. The filtrate was later diluted with 85% chilled acetone solution to a suitable volume for reading on a spectrophotometer. The solution obtained above was estimated against a blank at three wavelengths: 452nm, 663nm, and 644nm. The values of chlorophyll a, b, and total carotenoid were estimated using Arnon's equation (1949).

Chl a = $10.31\ E_{663.1}$-$0.918E_{644.1}$ = mg/ml

Chl b = $19.71E_{644.2}$-$3.870E_{663.2}$ = mg/ml

Car = $4.21\ E_{452.1}$-(0.0264 chl a + 0.426 chl b) = mg/ml

2.2. Determination of antioxidant enzymes

Superoxide dismutase, Peroxidase, and Catalase were determined according to the methods of Dengjie et al. (2007).
Superoxide dismutase
1ml was dissolved in 9.0 mL of distilled water to make a 1/10 dilution. 0.20 ml of the dissolved sample was transferred into 2.5 ml of 0.05M sodium carbonate buffer, pH 10.2, using a spectrophotometer, and 0.3 ml of newly prepared 0.3 mM adrenaline was added to the mixture, which was immediately mixed by agitation. Then, the mixture was observed at 480 nm absorbance every 30 seconds for 2 minutes and 30 seconds.
Calculation

$$\text{Increase in absorbance per minute} = \frac{A_3 - A_0}{2.5}$$

Where A_0 = absorbance after 0 seconds
A_3 = absorbance after 150 seconds

$$\%\ \text{Inhibition} = \frac{\text{Increase in absorbance of substrate X 100}}{\text{Increase in absorbance of blank}}$$

A unit of SOD activity is the amount of SOD necessary to produce 50% inhibition of the oxidation of adrenaline.
Determination of Peroxidase
About 4 mL phosphate buffer was dispensed into a cuvette, 100µl of the sample was added, followed by the addition of 2ml of 0.05M pyrogallol. It was mixed and kept in a spectrophotometer (reading was adjusted to zero at 420nm). 1ml of 1 mL of 1% H_2O_2 was added to the cuvette kept in the spectrophotometer. The absorbance was read at 30 sec interval up to 180 seconds. Peroxidase (POD) activity was expressed as units/mg protein.
Determination of Catalase
About 19Mm of 2.95ml Hydrogen peroxide solution was dissolved in a cuvette containing 50.0µl of the sample. The mixture was quickly upturned for mixing, and changes in absorbance were read at 240 nm every 5 minutes
Calculation

$$\text{Catalase activity} = \frac{A_{240}/\text{min x ml of reaction x dilution factor}}{0.0435\text{x ml of sample x mg protein/ml}}$$

2.3. Determination of compatible solutes

Soluble protein, soluble carbohydrate, and proline were determined according to the methods of Chen et al. (2000, Dengjie et al., 2007 and Mai et al. (2005).
Determination of Soluble Protein
Soluble proteins were analysed based on the method of Chen et al. (2002) using the Folin-Ciocalteu reagent. 250mg of the dried leaf and seed samples were weighed and digested by hot ethanol 40% twice, and the extract was then diluted to 25 ml with double-distilled water. Absorbance was read at 660 nm on a spectrophotometer. The amount of soluble protein was calculated from bovine serum albumen.
Soluble Carbohydrate
Soluble carbohydrate was analysed spectrophotometrically based on Dengjie et al. (2007) methods with slight modification. 5 mg of fresh samples were extracted in 1.5ml of 96 % ethanol for 15 min. Thereafter, samples were centrifuged at 21,000g for 15 min, and 40 µl of supernatant was poured into 10 ml test tubes that contained 400 µl deionised water, 400 µl of 5 % phenol, and 2 ml of concentrated

Hydrogen teteraoxosulphate VI was added. The resulting solution from above was incubated for 20 min. It was later read at 490 nm. The result was expressed in µg g-

Determination of Proline

The concentration of proline was estimated based on Mai et al.(2005). 0.5 g of the plant was ground with 10 ml of 3 W/V sulphosalicylic acid, and the mixture was filtered. Thereafter, supernatant, which was made up of 2 ml extract of plant, 2 ml of acid ninhydrin reagent, and 2 ml glacial acetic acid, was heated in a boiling water bath for 1 hour. It was later stopped using an ice bath, followed by adding 4 ml toluene. The mixture was shaken vigorously and later left to separate into phases. The chromophase, which contains the upper toluene phase, was transferred into another tube, and the absorbance was read at 520nm in a UV-Visible spectrophotometer. Proline concentration was then determined from a standard curve prepared with L-proline. Proline content was expressed as µg-1FW

2.4 Statistical analysis

Data were analysed using descriptive statistics and ANOVA with Statistical Analysis System (SAS) Version 9. Sample means were compared using Duncan Multiple Range Test (DMRT), and data were considered significantly different at P≤0.05. All the results were presented as mean ± standard error

3. Results

Table 1: Effect of Water Stress on the Soluble Protein of Kenaf Varieties

Kenaf Varieties	SPmg/g/ FW Control	SP mg/g/ FW (WR_4)	SP mg/g/ FW (WR_6)	SP mg/g/ FW (WR_8)	SP mg/g/ FW (WR_{10})
Ifeken100	25.09±0.35^{b}	25.11±0.18^{b}	21.12±0.12^{b}	34.11±0.02^{b}	46.14±0.03^{b}
Ifeken400	37.12±0.29^{a}	40.18±0.24^{a}	25.11±0.13^{a}	45.81±0.08^{a}	51.40±1.06^{a}
Cuba108	17.11±0.22^{d}	30.11±0.07^{d}	17.13±0.18^{d}	26.50±0.10^{d}	29.09±0.09^{d}
Tianung	20.10±0.27^{c}	35.10±0.22^{c}	19.12±0.28^{c}	29.12±0.09^{c}	37.12±0.11^{c}
Vi-100	10.10±0.17i	11.04±0.03^{i}	9.81±0.01^{i}	8.41±0.09^{i}	16.11±0.09^{i}
Au-72	13.00±0.89^{h}	13.51±0.07^{h}	11.11±0.05^{h}	13.31±0.11^{h}	13.21±0.04^{h}
Au2452	7.51±0.07^{j}	9.91±0.05^{j}	7.71±0.03^{j}	10.41±0.01^{j}	10.81±0.01^{j}
A60-282	15.11±0.11^{g}	15.53±0.02^{g}	12.51±0.07^{g}	16.11±0.02^{g}	17.02±0.03^{g}
AC-313	17.61±0.28^{f}	16.10±0.02^{f}	13.91±0.22^{f}	19.11±0.19^{f}	18.11±0.17^{f}
Au-754	19.15±0.12^{e}	18.10±0.14^{e}	15.12±0.18^{e}	23.09±0.08^{e}	23.51±0.14^{e}

SP: Soluble Protein, FW: Fresh Weight, WR: Water Restriction

Means with the different letters are significantly different (P≤0.05). Values are Mean±Standard error n=3.

Table 2: Effect of Water Stress on the Soluble Carbohydrate of Kenaf Varieties

Kenaf Varieties	SC (mg/g/ FW) Control	SC(mg/g/ FW) (WR_4)	SC(mg/g/ FW) (WR_6)	SC(mg/g/ FW) (WR_8)	SC(mg/g/ FW) (WR_{10})
Ifeken100	24.00±0.11^{b}	35.02±0.14^{b}	44.11±0.09^{b}	49.12±0.06^{b}	51.09±0.23^{b}
Ifeken400	26.71±0.12^{a}	41.05±0.61^{a}	46.04±0.34^{a}	52.91±0.18^{a}	55.09±0.75^{a}
Cuba108	19.24±0.05^{d}	23.11±0.23^{d}	29.13±0.43^{d}	41.50±0.14^{d}	39.90±0.18^{d}
Tianung	21.90±0.11^{c}	28.51±0.45^{c}	35.92±0.38^{c}	43.11±0.13^{c}	46.81±0.47^{c}
Vi-100	11.11±0.87^{i}	13.92±0.10^{i}	14.52±0.04^{i}	21.53±0.05^{i}	19.09±0.14^{i}
Au-72	12.12±0.08^{h}	14.71±0.11^{h}	17.52±0.12^{h}	25.51±0.10^{h}	22.91±0.17^{h}
Au2452	9.63±0.05^{j}	11.11±0.08^{j}	13.21±0.11^{j}	18.31±0.09^{j}	17.11±0.12^{j}
A60-282	14.10±0.02^{g}	16.11±0.07^{g}	19.91±0.22^{g}	27.91±0.11^{g}	24.12±0.15^{g}
AC-313	16.91±0.07^{f}	18.51±0.06^{f}	25.11±0.26^{f}	33.61±0.55^{f}	26.91±0.34^{f}
Au-754	17.11±0.11^{e}	20.51±0.15^{e}	21.11±0.17^{e}	37.61±0.11^{e}	31.10±0.11^{e}

SC: Soluble Carbohydrate, FW: Fresh Weight, WR: Water Restriction,

Means with the different letters are significantly different (P≤0.05). Values are Mean±Standard error n=3.

Table 3: Effect of Water Stress on the Lipid Peroxidation of Kenaf Varieties

Kenaf Varieties	LPO (nmol/g/FW) Control	LPO (nmol/g/FW) (WR_4)	LPO (nmol/g/FW) (WR_6)	LPO (nmol/g/FW) (WR_8)	LPO (nmol/g/FW) (WR_{10})
Ifeken100	0.08±0.001^{i}	3.02±0.01^{b}	8.12±0.02^{b}	12.51±0.03^{b}	15.11±0.19^{b}
Ifeken400	0.05±0.001^{j}	1.02±0.01^{a}	6.51±0.01^{a}	10.72±0.01^{a}	13.93±0.00^{a}
Cuba108	3.13±0.01^{h}	5.21±0.02^{d}	13.81±0.02^{d}	15.01±0.01^{d}	20.12±0.04^{d}
Tianung	0.14±0.01^{g}	4.03±0.01^{c}	11.11±0.03^{c}	14.51±0.06^{c}	17.10±0.16^{c}
Vi-100	1.21±0.05^{b}	17.11±0.06^{i}	41.11±0.17^{i}	32.91±0.08^{i}	34.11±0.56^{i}
Au-72	0.41±0.01^{d}	14.51±0.01^{h}	39.11±0.24^{h}	29.11±0.09^{h}	30.05±0.01^{h}
Au2452	4.03±0.02^{a}	19.91±1.23j	48.71±0.11^{j}	36.91±0.13^{j}	40.14±0.45^{j}
A60-282	0.32±0.15^{e}	12.32±0.02^{g}	33.95±0.02^{g}	21.32±0.01^{g}	27.25±0.01^{g}
AC-313	0.25±0.02^{f}	10.51±0.02^{f}	30.22±0.04^{f}	20.61±0.02^{f}	24.85±0.02^{f}
Au-754	0.52±0.01^{c}	8.61±0.01^{e}	27.45±0.04^{e}	18.09±0.06^{e}	22.23±0.01^{e}

SP: LPO: Lipid Peroxidation, DM: Dry Matter, WR: Water Restriction

Means with the different letters are significantly different (P≤0.05). Values are Mean±Standard error n=3.

Table 4: Effect of Water Stress on the Ascorbic Acid of Kenaf Varieties

Kenaf Varieties	Ascorbic acid mg/g/FW Control	Ascorbic acid mg/g/FW (WR_4)	Ascorbic acid mg/g/FW (WR_6)	Ascorbic acid mg/ g/ FW (WR_8)	Ascorbic acid mg/g/FW (WR_{10})
Ifeken100	97.53±0.28^{b}	80.00±0.58^{b}	65.50±0.29^{b}	51.97±0.55^{b}	45.61±0.17^{b}
Ifeken400	105.30±0.35^{a}	89.16±0.44^{a}	74.10±0.67^{a}	63.00±0.58^{a}	51.11±0.23^{a}

Cuba108	77.11±0.58[d]	60.00±0.58[d]	53.00±0.58[d]	35.27±0.82[d]	29.05±0.13[d]
Tianung	86.02±0.21[c]	71.27±0.63[c]	58.83±0.44[c]	43.90±0.21[c]	37.12±0.61[c]
Vi-100	35.97±0.55[i]	21.51±0.20[i]	18.00±0.58[i]	11.50±0.29[i]	7.30±0.65[i]
Au-72	47.83±0.44[h]	26.56±0.44[h]	21.87±0.14[h]	13.53±0.15[h]	9.77±0.17[h]
Au2452	20.45±0.32[j]	19.57±0.30[j]	15.73±0.19[j]	10.53±0.15[j]	5.70±0.06[j]
A60-282	59.10±0.57[g]	32.02±0.58[g]	26.17±0.44[g]	15.83±0.44[g]	12.83±0.44[g]
AC-313	62.00±0.58[f]	40.33±0.60[f]	35.00±0.58[f]	19.17±0.60[f]	17.00±0.58[f]
Au-754	66.00±0.42[e]	51.50±0.29[e]	41.97±0.55[e]	26.00±0.58[e]	20.00±0.22[e]

SP: Soluble protein, FW: Fresh Weight, WR: Water Restriction,
Means with the different letters are significantly different (P≤0.05). Values are Mean±Standard error n=3.

Table 5: Effect of Water Stress on the Proline Content of Kenaf Varieties

Kenaf Varie-ties	Proline (µMol/g/FW) Control	Proline (µMol/g/FW) (WR_4)	Proline (µMol/g/FW) (WR_6)	Proline (µMol/g/FW) (WR_8)	Proline (µMol/g/FW) (WR_{10})
Ifeken100	8.27±0.02[b]	21.17±0.04[b]	25.97±0.07[b]	38.30±0.09[b]	48.60±0.17[b]
Ifeken400	9.43±0.03[a]	22.60±0.07[a]	31.90±0.05[a]	43.60±0.08[a]	54.02±0.19[a]
Cuba108	6.43±0.05[d]	18.00±0.09[d]	20.63±0.02[d]	26.33±0.18[d]	32.50±0.22[d]
Tianung	7.42±0.06[c]	20.65±0.03[c]	21.83±0.02[c]	30.23±0.02[c]	37.13±0.23[c]
Vi-100	4.02±0.02[i]	11.63±0.04[i]	14.50±0.01[i]	14.00±0.03[i]	19.30±0.09[i]
Au-72	4.57±0.01[h]	12.76±0.02[h]	15.40±0.11[h]	16.33±0.06[h]	21.43±0.17[h]
Au2452	3.63±0.01[j]	10.77±0.02[j]	12.50±0.11j	12.33±0.09[j]	14.11±0.19[j]
A60-282	5.02±0.04[g]	14.00±0.02[g]	16.43±0.04[g]	18.03±0.35[g]	23.93±0.23[g]
AC-313	5.60±0.02[f]	15.46±0.07[f]	17.33±0.02[f]	20.80±0.06[f]	26.00±0.33[f]
Au-754	6.23±0.03e	16.41±0.03[e]	19.60±0.05[e]	22.00±0.02[e]	29.00±0.25[e]

Means with the different letters are significantly different (P≤0.05). Values are Mean±Standard error n=3.

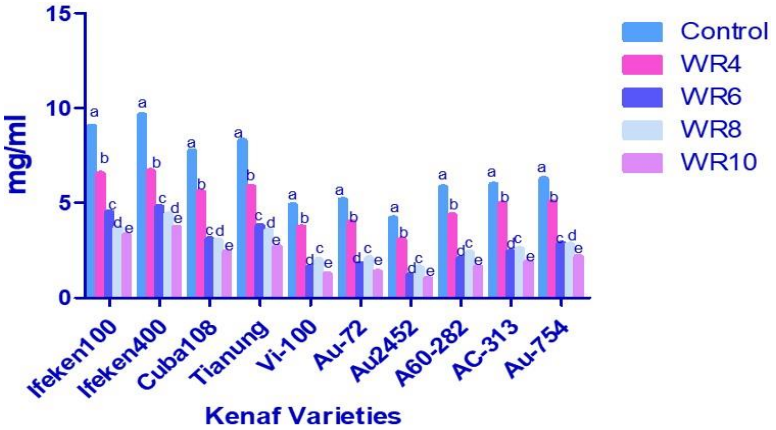

Fig. 1: Effect of Water Stress on Chlorophyll A Content of Kenaf Varieties.

Means with the different letters are significantly different (P≤0.05). Values are Mean±Standard error n=3.

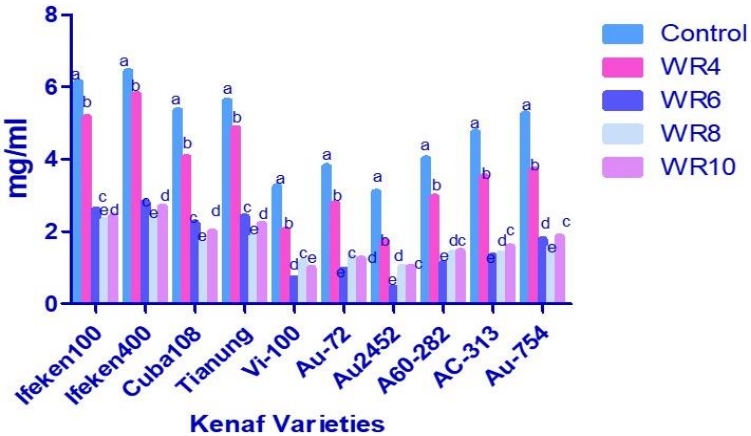

Fig. 2: Effect of Water Stress on Chlorophyll B Content of Kenaf Varieties.

Means with the different letters are significantly different (P≤0.05). Values are Mean±Standard error n=3.

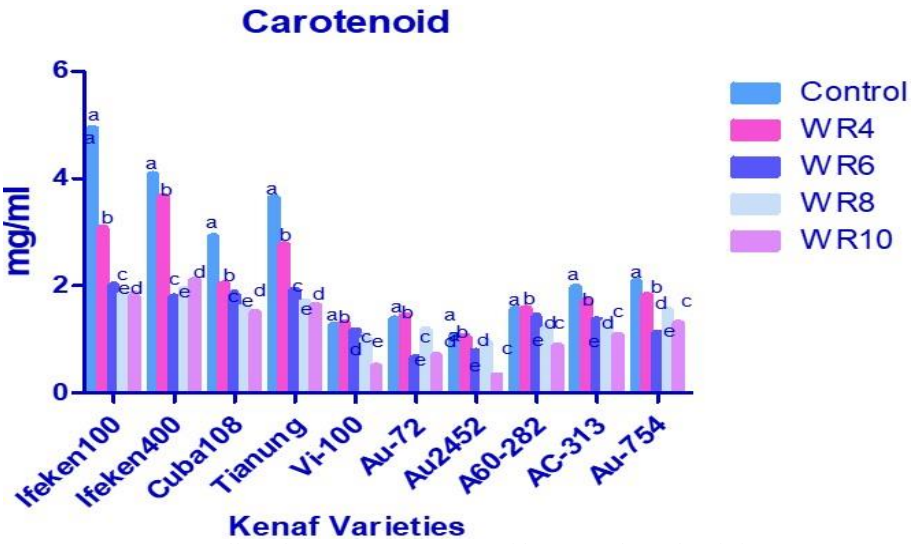

Fig. 3: Effect of Water Stress on Carotenoid Content of Kenaf Varieties.

Control: Unstressed plants, Stressed plants WR4(Watered every four days), WR6 (Watered every six days), WR8 (Watered every eight days), WR10 (Watered every ten days).

Table 6: Effect of Water Stress on the SOD of Kenaf Varieties

Kenaf Varieties	SOD(U/mgP) Control	SOD(U/mgP) (WR_4)	SOD(U/mgP) (WR_6)	SOD(U/mgP) (WR_8)	SOD(U/mgP) (WR_{10})
Ifeken100	0.188 ± 0.002^{b}	0.162 ± 0.003^{b}	0.147 ± 0.02^{b}	0.140 ± 0.01^{b}	0.157 ± 0.003^{b}
Ifeken400	0.192 ± 0.004^{a}	0.197 ± 0.03^{a}	0.155 ± 0.02^{a}	0.162 ± 0.01^{a}	0.178 ± 0.004^{a}
Cuba108	0.115 ± 0.002^{d}	0.131 ± 0.001^{d}	0.118 ± 0.003^{d}	0.103 ± 0.01^{d}	0.125 ± 0.002^{d}
Tianung	0.144 ± 0.001^{c}	0.142 ± 0.01^{c}	0.131 ± 0.003^{c}	0.129 ± 0.01^{c}	0.146 ± 0.002^{c}
Vi-100	0.053 ± 0.001^{i}	0.068 ± 0.001^{i}	0.079 ± 0.001^{i}	0.043 ± 0.001^{i}	0.062 ± 0.001^{i}
Au-72	0.064 ± 0.002^{h}	0.082 ± 0.001^{h}	0.087 ± 0.001^{h}	0.059 ± 0.001^{h}	0.078 ± 0.001^{h}
Au2452	0.035 ± 0.002^{j}	0.057 ± 0.001^{j}	0.062 ± 0.001^{j}	0.032 ± 0.003^{j}	0.053 ± 0.02^{j}
A60-282	0.069 ± 0.001^{g}	0.096 ± 0.002^{g}	0.089 ± 0.001^{g}	0.062 ± 0.001^{g}	0.095 ± 0.001^{g}
AC-313	0.081 ± 0.002^{f}	0.111 ± 0.001^{f}	0.095 ± 0.001^{f}	0.078 ± 0.02^{f}	0.107 ± 0.01^{f}
Au-754	0.089 ± 0.001^{e}	0.123 ± 0.02^{e}	0.103 ± 0.01^{e}	0.098 ± 0.002^{e}	0.118 ± 0.01^{e}

SOD: Superoxide dismutase, WR: Water Restriction, U/mgP; Unit/mg protein
Means with the different letters are significantly different ($P\leq0.05$) , Values are Mean±Standard error n=3,

Table 7: Effect of Water Stress on the POD of Kenaf Varieties

Kenaf Varieties	POD (U/mgP) Control	POD (U/mgP) (WR_4)	POD (U/mgP) (WR_6)	POD (U/mgP) (WR_8)	POD (U/mgP) (WR_{10})
Ifeken100	0.131 ± 0.002^{b}	0.167 ± 0.04^{b}	0.121 ± 0.032^{b}	0.122 ± 0.01^{b}	0.158 ± 0.009^{b}
Ifeken400	0.165 ± 0.007^{a}	0.211 ± 0.03^{a}	0.135 ± 0.01^{a}	0.141 ± 0.003^{a}	0.165 ± 0.05^{a}
Cuba108	0.099 ± 0.002^{d}	0.021 ± 0.001^{d}	0.098 ± 0.006^{d}	0.103 ± 0.02^{d}	0.137 ± 0.004^{d}
Tianung	0.104 ± 0.005^{c}	0.169 ± 0.03^{c}	0.112 ± 0.04^{c}	0.112 ± 0.01^{c}	0.145 ± 0.02^{c}
Vi-100	0.048 ± 0.002^{i}	0.018 ± 0.001^{i}	0.042 ± 0.001^{i}	0.013 ± 0.001^{i}	0.072 ± 0.001^{i}
Au-72	0.054 ± 0.004^{h}	0.032 ± 0.002^{h}	0.057 ± 0.001^{h}	0.057 ± 0.002^{h}	0.083 ± 0.001^{h}
Au2452	0.036 ± 0.001^{j}	0.016 ± 0.001^{j}	0.035 ± 0.001^{j}	0.048 ± 0.002^{j}	0.063 ± 0.005^{j}
A60-282	0.064 ± 0.005^{g}	0.145 ± 0.03^{g}	0.066 ± 0.02^{g}	0.062 ± 0.01^{g}	0.098 ± 0.01^{g}
AC-313	0.075 ± 0.01^{f}	0.134 ± 0.02^{f}	0.075 ± 0.01^{f}	0.078 ± 0.02^{f}	0.115 ± 0.01^{f}
Au-754	0.086 ± 0.007^{e}	0.189 ± 0.03^{e}	0.087 ± 0.02^{e}	0.097 ± 0.01^{e}	0.128 ± 0.02^{e}

POD: Peroxidase, WR: Water Restriction
Means with the different letters are significantly different ($P\leq0.05$). Values are Mean±Standard error n=3.

Table 8: Effect of Water Stress on Catalase in Kenaf Varieties

Kenaf Varieties	Catalase(U/mgP) Control	Catalase (U/mgP) (WR_4)	Catalase (U/mgP) (WR_6)	Catalase (U/mgP) (WR_8)	Catalase (U/mgP) (WR_{10})
Ifeken100	0.151 ± 0.01^{b}	0.183 ± 0.02^{b}	0.205 ± 0.01^{b}	0.292 ± 0.008^{b}	0.360 ± 0.007^{b}
Ifeken400	0.192 ± 0.003^{a}	0.207 ± 0.02^{a}	0.225 ± 0.03^{a}	0.313 ± 0.02^{a}	0.378 ± 0.01^{a}
Cuba108	0.154 ± 0.008^{d}	0.181 ± 0.03^{d}	0.185 ± 0.01^{d}	0.252 ± 0.02^{d}	0.307 ± 0.01^{d}
Tianung	0.165 ± 0.003^{c}	0.192 ± 0.02^{c}	0.191 ± 0.01^{c}	0.279 ± 0.01^{c}	0.346 ± 0.02^{c}
Vi-100	0.091 ± 0.005^{i}	0.108 ± 0.003^{i}	0.129 ± 0.003^{i}	0.173 ± 0.002^{i}	0.202 ± 0.006^{i}
Au-72	0.104 ± 0.005^{h}	0.122 ± 0.003^{h}	0.137 ± 0.002^{h}	0.189 ± 0.001^{h}	0.221 ± 0.001^{h}
Au2452	0.078 ± 0.02^{j}	0.097 ± 0.004^{j}	0.102 ± 0.003^{j}	0.162 ± 0.005^{j}	0.193 ± 0.03^{j}
A60-282	0.127 ± 0.02^{g}	0.136 ± 0.004^{g}	0.159 ± 0.003^{g}	0.202 ± 0.002^{g}	0.255 ± 0.001^{g}
AC-313	0.135 ± 0.02^{f}	0.165 ± 0.004^{f}	0.165 ± 0.006^{f}	0.218 ± 0.03^{f}	0.268 ± 0.02^{f}
Au-754	0.146 ± 0.005^{e}	0.173 ± 0.03^{e}	0.173 ± 0.02^{e}	0.239 ± 0.01^{e}	0.288 ± 0.02^{e}

Means with the different letters are significantly different ($P\leq0.05$). Values are Mean±Standard error n=3.

4. Discussion

Table 1-8 and Figures 1-3 showed the Biochemical response [compatible solutes (soluble protein, soluble carbohydrate, and proline), lipid peroxidation and ascorbic acid, photosynthetic pigments; chlorophyll a, b, and carotenoid], antioxidant enzymes (SOD, POD, and catalase)] of kenaf varieties exposed to water stress. There is a significant decrease in the chlorophyll a, b, and carotenoid content, SOD, POD, catalase, and ascorbic acid in the water-stressed kenaf varieties as compared with the control. This is a result of their imbalance in the uptake of essential minerals needed for photosynthesis, as well as lowered antioxidant activities due to the accumulation of oxidant molecules induced by water deficit. In addition, lipid peroxidation, proline, soluble carbohydrate, and soluble protein were significantly increased in water-stressed kenaf varieties as compared with the control due to oxidation of lipid molecules, plant response, and adaptation to induced stress and accumulation of free radicals induced by inadequate water. However, Ifeken 400, Ifeken 100, Tianung, and Cuba 108 showed higher photosynthetic pigments, enzyme activities, and compatible solutes, as well as the lowest values in lipid peroxidation in all the water regimes demonstrated. This is because Kenaf, which is resistant to water stress, frequently has better osmotic adjustment and improved antioxidant systems, such as increased proline, soluble carbohydrate, soluble protein, SOD, POD, and catalase activities. In addition, the modulation of the hormonal signal and the metabolism of carbohydrates play key roles in the potential expressed gene patterns. The result is in accordance with the work of Dengjie et al. (2007), who reported that soluble sugar and proline accumulation were significantly higher, while malonaldehyde (MDA) content was lower in the kenaf varieties exposed to drought stress. (Mai et al.,2005) Also reported that kenaf varieties showed improved photosynthesis under severe drought stress. Moreover, Masood et al.(2019) reported a significant decrease in stomatal conductance, water potential, and photosynthetic efficiency of Sorghum bicolor exposed to drought stress. Li et al. (2024) reported that pre-treatment of kenaf with 5-azacytidine significantly reduced genomic DNA methylation, alleviated salt stress in kenaf and significantly increased the seedlings biomass, antioxidant enzyme activities, and contents of chlorophyll and carbohydrate while reducing ROS production via expression of differentially methylated genes (HcMDH, pyruvate kinase, triosephosphate isomerase, G6PDH, NADPH, and Hsps) which are involved in carbon metabolism, amino acid biosynthesis, and fatty acid metabolism. Luo et al. (2022) had reported that Methyl-Sensitive Amplification Polymorphism (MSAP) Analysis Provides Insights into the DNA Methylation Underlying Heterosis in Kenaf (Hibiscus Cannabinus L.) Drought Tolerance, and that DnaJ, ERF5, ZIP2, and PATL3 are responsible for drought tolerance observed in kenaf, and that the virus-induced gene silencing (VIGS) mediated knockdown of DnaJ significantly increased the sensitivity of kenaf seedlings to drought. All these suggested the importance of biochemical mechanisms and pathways of water stress tolerance in kenaf, which is an indicator and important guide in breeding programmes for crop improvement.

5. Conclusion

Kenaf is an important and highly productive crop beneficial for its nutrients and industrial purposes, but requires an appreciable amount of moisture and nutrients from the soil. In this study, Ifeken 400, Ifeken 100, Tianung, and Cuba 108 were found to show high tolerance to water stress up to ten days through their biochemical response. This serves as useful information for farmers, breeders, researchers, and other relevant stakeholders. In addition, it also reveals and gives insights into their possible mechanisms of tolerance to water stress, which may serve as the main function and useful tools in predicting their gene expression patterns. Therefore, these varieties could be adopted by farmers who cultivate kenaf at subsistence and commercial levels, most especially where irrigation is the only option for growing kenaf. Finally, it could be recommended that these varieties will be suitable in an arid and semi-arid region characterised by dryness and low rainfall.

5.1. Contributions, recommendations, and future perspectives

Based on the results obtained in this study, the following recommendations could be deduced:

1) Ifeken 100, Ifeken 400, Tianung, and Cuba 108 could tolerate 10 days of water stress, which could complement irrigation and be found useful applications in arid regions.
2) Plant tolerance to water stress emanates from their intrinsic biochemical reaction occurring in plants, which then proceeds to their physiological characteristics exhibited by each plant.
3) Ifeken 100, Ifeken 400, Tianung, and Cuba 108 could be genetically engineered and used for breeding purposes for crop improvements.

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