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Assessment of Hydrogen Peroxide Potential in Mitigating Salinity Stress on Growth and Yield of *Zea mays* (L.) - Maize

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Abstract Salt stress is one of the major limitations of seed germination, plant growth, productivity and nutritional composition. Hydrogen peroxide (H_2O_2) functions as a signalling molecule that modulates physiological and biochemical processes under abiotic stress. Therefore, this research was conducted to assess the potential of H_2O_2 in mitigating adverse effects of salinity stress on the growth and yield of *Zea mays* (L.). The experiment was conducted in a screenhouse using 96 pots each filled with 14 kg topsoil and arranged in a completely randomized design with eight replicates per treatment. Maize seedlings raised were grouped into two: Each pot in Group A was irrigated with sodium chloride (NaCl) solution and supplemented with 50 ml of 3% H_2O_2 (882 mM) which was applied to the soil, while each pot in Group B received NaCl solution without H_2O_2. Salinity treatments were applied at 0 (control), 50, 100, 150, 200, and 250 mM NaCl three times per week and flushed once per week to prevent salt accumulation. Growth, yield, biomass, leaf chlorophyll as well as grain nutritional composition were assessed following standard procedures, and data were analysed using One Way Analysis of Variance at $p \leq 0.05$. Plant height declined the most from 160.76 cm in control to 112.19 cm at 250 mM NaCl without H_2O_2, while H_2O_2 treated plants at the same salinity decreased to only 123.52 cm. However, other growth parameters were not significantly enhanced by H_2O_2. The effect of salinity on number of grains per plant was positively influenced by H_2O_2 as salinity decreased it from 226.25 to 84.50 without H_2O_2, but H_2O_2-treated plants maintained up to 88.12 per plant at 250 mM. Salinity treatments devoid of H_2O_2 had protein reduced from 15.14% to 13.44%, fat from 1.88% to 1.74%, and crude fibre from 3.40% to 2.74%. However, salinity with H_2O_2 treatment sustained higher values (14.31%, 2.41%, and 2.80%, respectively). This study demonstrates that hydrogen peroxide can mitigate salinity-induced stress on growth and productivity in maize, supporting its potential role as a stress modulator in crop production under saline conditions.

Keywords Salt stress; Hydrogen peroxide; Salinity tolerance; *Zea mays*

1 Introduction

Maize (*Zea mays* L.) is a major cereal crop globally, serving as a staple food for millions and a vital component of the agricultural economy (Yadesa and Diro, 2023). Its significance stems from high yield potential, economic value, and broad adaptability. The global annual production of maize exceeds 1 billion metric tons, with leading producers including the United States of America, China, Brazil, and various African countries (Galani et al., 2022). In Nigeria, maize plays a critical role in food security and rural livelihoods, being widely cultivated across subsistence and commercial farming systems (Ogunniyi et al., 2021). Maize is rich in carbohydrates, providing essential energy, and contains key micronutrients such as vitamin A, iron, and zinc, essential for human nutrition (Galani et al., 2022; Kihara et al., 2024). Its industrial importance is underscored by its use in livestock feed and as raw material for bioethanol, starch, and biodegradable plastics (Maitra and Singh, 2021).

Despite this importance, maize productivity faces considerable challenges from abiotic stresses like soil salinity, which severely limit plant growth, yield, and nutritional quality (Syed et al., 2021; Islam et al., 2024). Soil salinity, typically resulting from excessive sodium chloride accumulation, disrupts water uptake, ionic balance, and induces oxidative stress through reactive oxygen species (ROS), including hydrogen peroxide (H_2O_2) (Al Otaibi et al., 2024). These biochemical imbalances cause reductions in photosynthesis, biomass, and ultimately grain yield (Zhu et al., 2023).

Several mitigation strategies have been explored to combat salinity stress, including breeding salt-tolerant genotypes, soil amendments, and exogenous application of biostimulants (Haque et al., 2021; Irin and Hasanuzzaman, 2024). Among these, hydrogen peroxide has gained attention as a signaling molecule modulating physiological responses under abiotic stress through activation of antioxidant defenses and osmotic adjustment mechanisms (Kesawat et al., 2023). Exogenous H_2O_2 application at low concentrations promotes antioxidant enzyme activities such as superoxide dismutase, catalase, and peroxidase, thereby reducing oxidative damage and improving salinity tolerance (Chattha et al., 2022). Moreover, H_2O_2 enhances nutrient uptake and water use efficiency in salt-stressed plants (Iqbal et al., 2023).

However, while H_2O_2's role in mitigating salinity effects has been studied in several crops, its influence on maize performance under saline conditions remains underexplored. Given maize's critical role in food security and its vulnerability to salt stress, there is a pressing need to evaluate the potential of hydrogen peroxide as a sustainable management option for salinity mitigation in maize cultivation. This study, therefore, aims to assess the effect of H_2O_2 application on growth, yield, and grain nutritional composition of maize subjected to varying levels of salinity stress.

2 Materials and Methods

2.1 Location of the experiment

This experiment was carried out at the screen house of the Department of Plant Science & Biotechnology (PSB), Adekunle Ajasin University, Akungba-Akoko (AAUA), Ondo State, Nigeria (latitude 7.2 ^{0}N, longitude 5.44 ^{0}E).

2.2 Sources of materials for the experiment

Seeds of *Zea mays* (maize) were obtained from the Federal College of Agriculture, Akure, Ondo State (FECA), Nigeria. The salt (NaCl) and Hydrogen peroxide (H_2O_2) were obtained from the laboratory, and the soil used for planting was collected from the experimental plots of PSB Department, AAUA. The soil was analyzed for physical and chemical properties using the standard methods of AOAC (1985). It was shade-dried and passed through a 2-mm sieve. Total N was analyzed using the macro Kjeldahl procedure; organic carbon by Walkley and Black procedure with percentage derived by multiplying organic carbon content by 1.72; and pH using soil: water ratio of 1:2 with a pH meter. Available phosphorus was got through the Bray 1 method; exchangeable acidity by titration method; exchangeable K, Na, Ca, Al and Mg by extraction with 1 M ammonium acetate at pH 7.0; and the amount of K and Na was measured using a Corning Flame Photometer with appropriate filter, while Ca, Al and Mg were determined using a Perkin-Elmer Atomic Absorption Spectrophotometer (AAS). The electrical conductivity was read with a conductivity meter.

2.3 Soil collection and preparation

Topsoil (0~15 cm depth) was collected from an arable farmland within the premises of Adekunle Ajasin University, Akungba-Akoko, Ondo State. The soil was sieved to remove debris and thoroughly mixed to obtain a homogeneous medium. Approximately 14 kg of prepared soil was placed into each perforated polythene pot. Maize was grown in perforated polythene pots filled with 14 kg of the prepared topsoil. Three maize seeds were sown per pot, and seedlings were allowed to establish before thinning to one seedling per pot prior to the commencement of treatments.

2.4 Experimental setup

A total of 96 pots were grouped into two (Groups A and B), each consisting of 48 pots. The potted soils on which the maize seedlings were grown were irrigated three weeks after planting with sodium chloride (NaCl) solution at concentrations of 0 (control), 50, 100, 150, 200, and 250 mM three times in the week of planting. Each potted soil in Group A received 50 ml of 3% hydrogen peroxide (H_2O_2) solution the following week equivalent to 882 mM, while pots in Group B received no H_2O_2 treatment. All pots were watered to saturation and allowed to drain once per week to prevent salt accumulation beyond intended concentrations. Pots were arranged in a completely randomized design with eight replicates per treatment in the screenhouse.

2.5 Data collection

Plant height was measured from the surface of the soil to the plant apical bud using a meter rule. Stem girth was measured at the 2 cm point from the base of the plants using a digital vernier caliper. Leaf length and breadth were measured using a meter rule, and leaf area was calculated. The number of leaves and ears were counted manually on each plant. Ear length and diameter were measured using a meter rule, and a vernier caliper respectively. Root growth was determined by measuring the root length using a meter rule after uprooting, and the number of roots was counted manually. Fresh and dry mass of plant parts were assessed using an electronic weighing balance.

2.6 Laboratory analysis of maize grains

Dried maize grains were ground into fine powder for analysis. Fiber content was determined by boiling the sample in 1.25% H_2SO_4 and 1.25% NaOH, followed by washing and drying. Other parameters of proximate composition were analyzed using the standard methods of AOAC (1985) in which the mixture was boiled until a clear solution was obtained and allowed to cool at room temperature. The resulting solution was quantitatively transferred into a calibrated flask and completed to 25 mL with distilled water. Moisture, crude protein, crude fat, carbohydrate and ash contents were calculated using relevant formulas. N was analyzed using the macro Kjeldahl method, while P was determined using ammonium-vanadomolybdate reagent and a calibration curve. Potassium contents were assayed through flame emission photometry, and calcium contents by Ethylenediaminetetraacetic acid (EDTA) titration.

2.7 Statistical analysis

All data collected were subjected to One-Way Analysis of Variance (ANOVA) using the Statistical Package for Social Sciences (SPSS), version 27.0. Where significant differences were observed among treatment means, Tukey's Honest Significant Difference (HSD) test was used at a 95% confidence level to perform post-hoc comparisons, and values presented as mean ± standard error (SE).

3 Results

3.1 Soil used for planting

The soil used for planting was a sandy clay loam adequate for maize cultivation with physico-chemical characteristics shown in Table 1.

Table 1 Physico-chemical parameters of soil used for planting

Parameter	Value
Sand (%)	57.50
Clay (%)	29.37
Silt (%)	13.13
Soil textural class	Sandy clay loam
Soil pH	6.53
Electrical conductivity EC (%)	0.39
Organic matter (%)	1.56
Available N (mg/kg)	0.17
Available P (mg/) kg	20.11
Available K (cmol/kg)	0.22
Available Na (cmol/kg)	0.37
Available Ca (cmol/kg)	5.78
Available Al (cmol/kg)	20.58
Available Mg (cmol/kg)	2.23

3.2 Plant survival and growth

Table 2 shows the influence of hydrogen peroxide (H_2O_2) on the plant height, stem girth, number of leaves, leaf length, leaf breadth, leaf area, number of roots, root length and the number of tassels of *Zea mays* under salt stress. While all plants survived, salinity reduced plant height in both hydrogen peroxide treated and untreated plants (Figure 1). However, no significant difference was observed between the control and the salinity treated plants with hydrogen peroxide application at lower levels, though at 250 mM NaCl, H_2O_2 treated plants maintained

123.52 cm compared to 112.19 cm without H_2O_2. In contrast, plants exposed to salt stress without hydrogen peroxide were generally shorter than the control, with significant reductions occurring at higher NaCl concentrations (200~250 mM).

Table 2 Growth parameters of *Zea mays* under salinity treatments with and without Hydrogen peroxide (H_2O_2) application

Parameters	With and without HP	Salinity treatment (mM NaCl)					
		0	50	100	150	200	250
Survival (%)		100.00	100.00	100.00	100.00	100.00	100.00
Plant height (cm)	WHP	160.76±2.69^{c}	119.69±0.29^{b}	119.40±0.30^{b}	114.69±0.87^{ab}	113.99±0.50^{ab}	112.19±0.29^{ab}
	PHP	173.08±1.26^{c}	150.12±0.47^{b}	123.76±0.28^{ab}	118.52±0.26^{a}	120.62±0.25^{a}	123.52±0.27^{a}
Stem girth (cm)	WHP	27.23±0.08^{a}	22.83±0.22^{a}	21.13±0.21^{a}	19.76±0.27^{ab}	18.49±0.06^{ab}	18.11±0.13^{ab}
	PHP	25.30±0.06^{a}	22.95±0.08^{a}	22.12±0.08^{a}	18.52±0.05^{a}	17.57±0.04^{a}	17.55±0.03^{a}
Number of leaves	WHP	13.00±0.00^{a}	12.75±0.16^{a}	12.63±0.18^{a}	12.00±0.19^{a}	11.25±0.16^{a}	10.87±0.13^{a}
	PHP	13.00±0.00^{a}	14.00±0.00^{a}	12.75±0.16^{a}	12.00±0.00^{a}	12.00±0.00^{a}	12.00±0.00^{a}
Leaf length (cm)	WHP	25.35±0.04^{a}	21.43±0.13^{a}	20.35±0.05^{a}	19.79±0.23^{a}	19.49±0.15^{a}	20.71±1.28^{a}
	PHP	23.53±0.18^{a}	20.05±0.06^{ab}	19.52±0.05^{ab}	19.70±0.00^{ab}	19.78±0.23^{ab}	19.46±0.05^{ab}
Leaf breadth (cm)	WHP	9.80±0.05^{a}	7.98±0.04^{a}	7.6±0.03^{a}	7.25±0.02^{a}	7.20±0.04^{a}	7.07±0.03^{a}
	PHP	9.58±0.05^{a}	9.13±0.04^{a}	7.93±0.0.3^{a}	7.46±0.42^{a}	7.31±0.03^{a}	7.36±0.04^{a}
Leaf area (cm^2)	WHP	133.02±0.67^{a}	124.15±0.39^{ab}	114.74±0.25^{b}	107.82±0.78^{b}	109.10±0.31^{b}	105.25±0.16^{b}
	PHP	127.51±0.74^{a}	124.48±0.39^{a}	114.28±0.32^{ab}	107.00±0.11^{b}	104.230.16^{b}	103.99±0.08^{b}
Number of roots	WHP	32.25±0.59^{a}	31.88±0.30^{a}	26.25±0.45^{ab}	19.87±0.44^{ab}	23.50±0.65^{ab}	22.88±0.67^{ab}
	PHP	32.25±0.49^{a}	28.62±1.49^{b}	23.25±0.37^{bc}	20.37±0.50^{bc}	22.75±0.31^{bc}	22.50±0.27^{bc}
Root length (cm)	WHP	64.66±2.87^{a}	44.17±0.56^{b}	42.03±0.79^{b}	40.87±2.63^{b}	33.53±0.46^{c}	37.73±4.39^{c}
	PHP	66.76±2.04^{a}	55.48±0.63^{b}	54.68±1.84^{b}	43.30±0.10^{bc}	36.40±1.27^{bc}	45.88±0.69^{bc}
Number of tassels	WHP	11.38±0.26^{a}	11.50±0.19^{a}	11.37±0.18^{a}	11.25±0.16^{a}	11.12±0.23^{a}	12.38±0.18^{a}
	PHP	11.63±0.26^{a}	11.37±0.18^{a}	11.25±0.16^{a}	11.50±0.19^{a}	11.62±0.26^{a}	11.75±0.25^{a}

Note: Values are mean ± standard error of 8 replicates (Tukey HSD test at $p \leq 0.05$). Mean with the same alphabet(s) along the row are not significantly different from each other. PHP: plus hydrogen peroxide (H_2O_2); WHP: without hydrogen peroxide (H_2O_2)

Figure 1 Effect of salinity stress with hydrogen peroxide (A) and without hydrogen peroxide (B) on *Zea mays* growth

Stem girth was influenced by salinity, with hydrogen peroxide treated plants showing minor improvements at 50~150 mM NaCl compared to the control. However, at 200~250 mM NaCl, stem girth declined, though the reduction was less than without hydrogen peroxide. Without hydrogen peroxide, stem girth generally decreased under salinity, with significant reductions observed at higher NaCl levels.

Leaf production was also affected by salinity. At lower concentrations (50~100 mM NaCl), plants produced similar or slightly fewer leaves than the control, though the change was not significant. However, at higher concentrations (150~250 mM NaCl), leaf production declined, though the reduction was not significantly different from the control in many cases. Similarly, in plants grown without hydrogen peroxide, the number of leaves

decreased under salinity, with more pronounced reductions at 200~250 mM NaCl compared to the control. Leaf length, breadth, and area followed similar trends, with hydrogen peroxide providing partial mitigation, resulting in less severe declines than without it, likely due to improved osmotic regulation. The number of tassels remained relatively stable under salinity, with hydrogen peroxide having a negligible effect, indicating reproductive initiation was less impacted than vegetative growth.

At the end of the experiment, root growth parameters (number and length) varied depending on hydrogen peroxide treatment and salt concentration. At 50~150 mM NaCl, plants treated with hydrogen peroxide (H_2O_2) had better root development compared to those without. However, at 200~250 mM NaCl, root parameters declined more sharply without hydrogen peroxide(H_2O_2).

3.3 Plant biomass

Salinity stress significantly reduced the vegetative biomass (fresh and dry weights of roots, stems, and leaves) of Zea mays in both hydrogen peroxide (H_2O_2) treated (PHP) and without hydrogen peroxide (H_2O_2) (WHP) plants, with reductions intensifying at higher NaCl concentrations (200~250 mM) as shown in Table 3. Without hydrogen peroxide, fresh and dry weights of roots, stems, and leaves decreased markedly, reflecting impaired cell division and photosynthetic efficiency due to osmotic stress and ion toxicity. For instance, at 250 mM NaCl, root dry weight was significantly lower compared to the control.

In contrast, H_2O_2 treated plants exhibited less severe biomass reductions across all salinity levels. At 50~150 mM NaCl, hydrogen peroxide (H_2O_2) treated (PHP) sustained higher fresh and dry weights for roots, stems, and leaves compared to without hydrogen peroxide (H_2O_2) (WHP) plants, indicating improved water retention and metabolic activity. At 250 mM NaCl, hydrogen peroxide (H_2O_2) treated (PHP) plants still showed higher biomass than without hydrogen peroxide (H_2O_2) (WHP), though not fully restored to control levels. This suggests hydrogen peroxide mitigated salinity induced stress by enhancing antioxidant defenses and osmotic adjustment, partially preserving biomass accumulation as shown in Table 3.

Table 3 Vegetative biomass of *Zea mays* under salinity treatments with and without Hydrogen peroxide (H_2O_2) application

Growth parameters (g)	With and without HP	Salinity treatment (mM NaCl)					
		0	50	100	150	200	250
Leaf fresh weight	WHP	64.91±1.05^{a}	47.60±0.81^{b}	46.85±0.58^{b}	39.28±1.00^{bc}	39.65±0.29^{bc}	30.90±5.35^{c}
	PHP	61.90±3.09^{a}	47.56±1.48^{b}	37.50±1.59^{bc}	46.96±1.07^{bc}	41.13±0.78^{bc}	44.32±0.35^{b}
Stem fresh weight	WHP	142.63±1.30^{a}	105.31±0.81^{b}	103.30±1.45^{b}	54.22±0.22^{c}	38.05±1.03^{c}	35.15±0.57^{c}
	PHP	149.90±0.39^{a}	113.56±0.82^{b}	102.98±0.13^{b}	61.92±0.41^{c}	41.58±0.36^{d}	35.03±0.38^{d}
Root fresh weight	WHP	52.84±0.15^{a}	37.94±1.50^{b}	33.45±0.41^{b}	24.45±0.58^{c}	28.26±0.94^{c}	28.13±0.43^{c}
	PHP	55.13±0.57^{a}	42.77±0.62^{b}	43.05±0.81^{b}	34.20±0.71^{c}	32.62±0.81^{c}	35.28±0.59^{c}
Leaf dry weight	WHP	39.98±2.10^{a}	32.87±1.93^{a}	28.12±1.56^{bc}	21.11±0.78^{bc}	17.78±0.17^{c}	15.41±0.16^{c}
	PHP	30.16±0.49^{a}	30.66±0.23^{a}	29.72±0.17^{a}	23.00±0.19^{b}	15.92±0.11^{bc}	15.07±0.17^{bc}
Stem dry weight	WHP	72.20±0.44^{a}	65.62±0.85^{b}	52.10±0.53^{b}	35.26±0.69^{c}	31.21±1.55^{c}	22.36±0.46^{d}
	PHP	69.12±0.52^{a}	68.03±0.50^{a}	64.25±0.32^{a}	57.47±1.31^{b}	27.80±0.53^{c}	27.75±0.21^{c}
Root dry weight	WHP	33.47±0.38^{a}	29.36±0.66^{b}	22.27±0.18^{b}	20.19±0.32^{b}	24.61±0.84^{b}	25.11±0.59^{b}
	PHP	34.15±0.55^{a}	23.25±0.41^{b}	23.66±0.83^{b}	25.10±1.31^{b}	24.66±0.65^{b}	23.23±0.42^{b}
Total biomass	WHP	145.24±2.21^{a}	128.58±2.0^{b}	102.80±1.52^{c}	76.63±1.16^{d}	73.71±1.69^{d}	62.90±0.71^{d}
	PHP	133.43±0.72^{a}	121.92±0.77^{ab}	117.65±0.88^{ab}	105.57±0.97^{ab}	68.38±0.93^{c}	65.71±0.47^{c}

Note: Values are mean ± standard error of 8 replicates (Tukey HSD test at $p \leq 0.05$). Mean with the same alphabet(s) along the row are not significantly different from each other. PHP: plus hydrogen peroxide (H_2O_2); WHP: without hydrogen peroxide (H_2O_2)

3.4 Yield parameter

Salinity significantly reduced yield components, including ear number, ear length, grain number, and grain weight per plant (Table 4), with the most pronounced effects at 250 mM NaCl. Without hydrogen peroxide (H_2O_2) (WHP), the number of grains per plant dropped from 226.25 in the control to 84.50 at 250 mM, reflecting disrupted assimilate allocation and kernel development due to salinity stress.

Hydrogen peroxide (H_2O_2) treated (PHP) plants showed improved yield parameters across all salinity levels. At 250 mM NaCl, PHP maintained grain numbers at 88.12 per plant, a slight but notable improvement over without hydrogen peroxide (H_2O_2) (WHP). Ear number and weight were also less reduced in Hydrogen peroxide (H_2O_2) treated (PHP) plants, particularly at moderate salinities (50~150 mM), suggesting hydrogen peroxide supported reproductive development by reducing oxidative damage and improving nutrient mobilization. However, at higher salinities, the mitigation was partial, indicating limits to H_2O_2 protective capacity under severe stress.

Table 4 Yield parameters of *Zea mays* under salinity treatments with and without Hydrogen peroxide (H_2O_2) application

Parameters	With and without HP	Salinity treatment (mM NaCl)					
		0	50	100	150	200	250
Number of ears	WHP	2.38±0.18^{a}	2.38±0.18^{a}	2.13±0.23^{a}	1.50±0.19^{b}	1.38±0.18^{b}	1.38±0.18^{a}
	PHP	2.50±0.18^{a}	2.38±0.18^{a}	2.37±0.18^{a}	2.12±0.13^{a}	1.50±0.18^{ab}	1.50±0.18^{ab}
Ear length (cm)	WHP	19.63±1.01^{a}	18.21±0.65^{a}	14.75±0.80^{b}	13.62±0.42^{b}	13.25±0.59^{b}	12.12±0.30^{b}
	PHP	17.87±0.52^{a}	17.37±0.46^{a}	15.62±0.18^{a}	16.00±0.33^{a}	12.25±0.36^{ab}	11.00±0.27^{ab}
Ear diameter (cm)	WHP	13.50±0.13^{a}	14.23±0.08^{a}	14.61±0.22^{a}	13.02±0.60^{a}	12.15±0.94^{a}	14.08±0.19^{a}
	PHP	29.43±15.36^{a}	13.02±0.24^{b}	11.86±0.87^{b}	13.21±0.59^{b}	12.61±0.47^{b}	11.98±0.97^{b}
Ear fresh weight (g)	WHP	307.62±159.77^{a}	93.57±2.30^{b}	85.62±0.65^{c}	75.73±0.76^{b}	72.41±1.41^{b}	73.41±1.29^{b}
	PHP	146.57±1.12^{a}	120.91±1.59^{b}	119.75±11.22^{b}	108.08±1.36^{c}	93.58±0.78^{c}	94.58±0.84^{c}
Ear dry weight (g)	WHP	106.91±1.53^{a}	86.83±0.79^{b}	75.17±0.83^{b}	69.61±1.39^{b}	65.88±1.41^{b}	43.60±0.27^{c}
	PHP	107.56±0.39^{a}	82.96±0.32^{b}	81.68±0.39^{b}	68.12±0.33^{bc}	70.01±0.30^{bc}	50.05±1.67^{c}
Number of grains	WHP	226.25±13.13^{a}	217.38±2.71^{a}	212.13±1.42^{a}	175.50±0.82^{ab}	146.00±11.72^{ab}	84.50±3.85^{b}
	PHP	262.75±13.23^{a}	217.62±12.11^{ab}	169.87±14.84^{b}	177.75±17.14^{b}	111.37±1.50^{bc}	88.12±5.05^{c}
Grain fresh weight (g)	WHP	139.41±1.04^{a}	124.63±0.86^{a}	133.10±1.83^{a}	91.76±0.40^{ab}	92.67±5.56^{ab}	75.65±0.87^{ab}
	PHP	139.73±0.75^{a}	134.12±0.63^{a}	133.70±1.15^{a}	130.07±0.31^{a}	92.11±2.15^{b}	85.57±2.24^{b}
Grain dry weight (g)	WHP	55.78±0.68^{a}	52.87±0.52^{a}	54.26±0.75^{a}	54.01±1.28^{a}	49.36±0.27^{a}	38.89±5.30^{ab}
	PHP	55.60±0.50^{a}	53.71±0.23^{a}	52.83±0.39^{a}	53.58±0.44^{a}	54.90±0.24^{a}	51.83±0.32^{a}
1000 grain weight (g)	WHP	30.88±0.64^{a}	25.25±1.03^{ab}	20.13±0.48^{ab}	22.13±0.52^{ab}	22.75±1.16^{ab}	23.88±1.62^{ab}
	PHP	31.25±0.70^{a}	27.25±1.58^{ab}	23.63±1.45^{ab}	21.12±1.30^{ab}	20.50±0.85^{ab}	18.25±1.15^{c}

Note: Values are mean ± standard error of 8 replicates (Tukey HSD test at $p \leq 0.05$). Mean with the same alphabet(s) along the row are not significantly different from each other. PHP: plus hydrogen peroxide (H2O2); WHP: without hydrogen peroxide (H_2O_2).

3.5 Nutritional and proximate composition

Table 5 shows that salinity stress without hydrogen peroxide (H_2O_2) (WHP) led to significant reductions in grain proximate components. Protein content decreased from 15.14% in the control to 13.44% at 250 mM NaCl, fat from 1.88% to 1.74%, and crude fiber from 3.40% to 2.74%. Concurrently, moisture and ash contents increased, likely due to disrupted metabolic processes and ion accumulation.

Hydrogen peroxide (H_2O_2) treated (PHP) plants maintained higher proximate values under salinity stress. At 250 mM NaCl, protein was sustained at 14.31%, fat at 2.41%, and crude fiber at 2.80%, closer to control levels. This indicates hydrogen peroxide helped stabilize metabolic pathways, reducing the impact of salinity on nutrient synthesis and storage.

3.6 Grain nutritional composition

Salinity without hydrogen peroxide plants increased Na^+ and Cl^- accumulation in grains while reducing key nutrients like potassium, phosphorus, and magnesium, reflecting ion imbalances and impaired nutrient uptake. Hydrogen peroxide (H_2O_2) treated (PHP) mitigated these effects, with lower Na^+ and Cl^- accumulation and better retention of essential nutrients as shown in (Table 5) (e.g., higher potassium levels at all salinities). This suggests hydrogen peroxide improved ion homeostasis, likely through enhanced antioxidant enzyme activity.

3.7 Leaf total chlorophyll content

Table 6 shows that the chlorophyll content declined significantly under salinity stress without hydrogen peroxide, with the lowest levels at 250 mM NaCl due to pigment degradation and chloroplast damage. Hydrogen peroxide

treated plants preserved higher chlorophyll content across all salinity levels, supporting sustained photosynthetic capacity and contributing to better grain quality.

Table 5 Grain nutritional and proximate compositions of *Zea mays* under salinity treatments with and without hydrogen peroxide (H_2O_2) application

Proximate (%) and nutritional (mg/kg) composition	With and without HP	Salinity treatment (mM NaCl)					
		0	50	100	150	200	250
Moisture	WHP	7.11±0.11^{a}	7.82±0.22^{a}	8.14±0.11^{a}	8.83±0.28^{a}	8.03±0.55^{a}	8.74±0.27^{a}
	PHP	7.10±0.10^{a}	8.87±0.34^{a}	8.01±0.18^{a}	8.82±0.18^{a}	9.39±0.02^{a}	9.84±0.28^{a}
Fat	WHP	1.88±0.13^{a}	1.83±0.03^{a}	2.04±0.08^{a}	2.02±0.02^{a}	1.82±0.06^{a}	1.74±0.05^{a}
	PHP	1.78±0.11^{a}	1.75±0.06^{a}	1.58±0.02^{a}	1.83±0.19^{a}	1.89±0.02^{a}	2.41±0.11^{b}
Ash	WHP	3.22±0.22^{a}	4.94±0.07^{a}	2.79±1.59^{a}	4.20±0.21^{a}	3.76±0.18^{a}	4.06±0.05^{a}
	PHP	3.22±0.27^{a}	3.91±0.11^{a}	3.77±0.17^{a}	4.14±0.12^{a}	3.73±0.13^{a}	3.89±0.21^{a}
Crude fibre	WHP	3.40±0.01^{b}	2.48±0.23^{a}	2.92±0.09^{ab}	2.55±0.26^{ab}	3.08±0.07^{ab}	2.74±0.09^{ab}
	PHP	2.99±0.02^{a}	2.83±0.08^{a}	2.97±0.04^{a}	2.21±0.23^{a}	2.67±0.08^{a}	2.80±0.23^{a}
Crude protein	WHP	15.14±0.58^{b}	11.21±0.23^{ab}	11.50±0.51^{ab}	10.36±0.38^{ab}	12.48±0.31^{ab}	13.44±0.57^{ab}
	PHP	15.17±0.58^{a}	12.24±0.24^{b}	13.50±0.51^{b}	10.96±0.01^{b}	14.50±0.63^{b}	14.31±0.34^{b}
Carbohydrate	WHP	69.26±0.81^{a}	71.72±0.12^{a}	72.61±0.97^{a}	72.03±0.55^{a}	70.83±1.03^{a}	69.28±0.65^{a}
	PHP	69.29±0.83^{a}	70.40±0.56^{a}	70.17±0.14^{a}	72.03±0.26^{a}	67.82±0.79^{a}	66.76±0.07^{a}
Nitrogen (N)	WHP	5.07±0.01^{a}	3.80±0.15^{b}	3.90±0.20^{b}	3.52±0.00^{b}	4.25±0.02^{ab}	4.28±0.10^{ab}
	PHP	5.10±0.00^{a}	4.15±0.00^{a}	4.58±0.01^{a}	3.72±0.02^{a}	4.92±0.02^{a}	4.85±0.02^{a}
Potassium (k)	WHP	329.80±0.30^{a}	331.50±0.60^{a}	328.15±0.2^{5a}	329.80±0.30^{a}	331.50±0.60^{a}	328.15±0.25^{a}
	PHP	329.80±0.30^{a}	335.05±0.45^{a}	336.50±0.30^{a}	339.95±0.35^{a}	342.80±0.30^{a}	333.50±0.20^{a}
Calcium (Ca)	WHP	10.45±0.25^{a}	11.05±0.65^{a}	11.00±0.10^{a}	11.45±0.25^{a}	12.05±0.35^{a}	12.50±0.40^{a}
	PHP	10.47±0.25^{a}	5.50±0.30^{ab}	7.80±0.30^{ab}	10.30±0.10^{a}	11.15±0.75^{a}	12.70±0.20^{a}
Phosphorus (P)	WHP	318.45±0.05^{a}	320.45±0.05^{a}	317.15±0.25^{a}	318.45±0.05^{a}	320.45±0.05^{a}	317.15±0.25^{a}
	PHP	316.55±0.05^{a}	318.15±0.05^{a}	316.15±0.2^{5a}	322.80±0.30^{a}	320.85±0.25^{a}	318.75±0.15^{a}

Note: Values are mean ± standard error of 8 replicates (Tukey HSD test at $p \leq 0.05$). Mean with the same alphabet(s) along the column are not significantly different from each other. PHP: plus hydrogen peroxide (H_2O_2); WHP: without hydrogen peroxide (H_2O_2)

Overall, hydrogen peroxide application consistently reduced the adverse effects of salinity on *Zea mays* by approximately 10%~20% across biomass, yield, and nutritional/proximate composition metrics, particularly at moderate salinity levels. However, under severe stress (250 mM NaCl), mitigation was partial, indicating that while hydrogen peroxide enhances resilience, it does not fully counteract extreme salinity effects.

4 Discussion

The results of this study clearly show that hydrogen peroxide (H_2O_2) serves as an effective agent in reducing the harmful impacts of salinity stress on *Zea mays* (maize). This aligns well with the established understanding that HP functions as a key signaling molecule in plants' responses to various abiotic stresses. At low concentrations, HP acts not as a damaging oxidant but as a regulator that triggers protective mechanisms, such as activating antioxidant systems, modulating gene expression, and facilitating cellular acclimation to adverse conditions like high salt levels.

One prominent benefit observed from this experiment was H_2O_2 capacity to lessen the salinity induced decline in plant height. Specifically, plants treated with HP reached an average height of 123.52 cm under 250 mM NaCl stress, in contrast to only 112.19 cm in untreated stressed plants. This improvement reflects H_2O_2 contributions to processes like osmotic adjustment where plants accumulate compatible solutes to maintain cell turgor and enhanced scavenging of reactive oxygen species (ROS), which otherwise accumulate excessively under salt stress and cause cellular damage. Such effects are supported by prior research demonstrating H_2O_2 involvement in these protective pathways in plants facing osmotic challenges (Qureshi et al., 2022; Zulfiqar et al., 2022).

However, the benefits were not uniform across all growth metrics. While plant height showed clear gains, other parameters like leaf number and stem girth exhibited only limited or no significant enhancement from H_2O_2 treatment. This pattern points to species-specific sensitivities in maize or concentration dependent responses of H_2O_2, where the applied dose or timing may optimally influence certain traits but not others. Comparable variability has been documented in different crops exposed to salinity or related stresses, highlighting that H_2O_2 efficacy can vary based on plant type, stress severity, and application details (Roque et al., 2024; Thomas et al., 2025).

Table 6 Leaf chlorophyll contents of *Zea mays* under salinity treatments with and without hydrogen peroxide (H_2O_2) application

Salinity treatment (mM NaCl)	With or without HP	Chlorophyll (mg/L)		Total chlorophyll (mg/L)
		A	b	
0	WHP	22.25	48.00	70.24
50		10.16	22.04	32.20
100		10.14	22.53	32.67
150		10.77	25.42	36.18
200		11.08	26.76	37.85
250		10.07	22.16	32.23
0	PHP	24.62	47.81	72.43
50		21.83	25.69	47.54
100		18.33	24.91	43.24
150		13.13	27.00	40.13
200		12.35	18.64	30.98
250		15.11	19.58	34.69

Note: PHP: plus hydrogen peroxide (H_2O_2); WHP: without hydrogen peroxide (H_2O_2)

Positive effects extended to vegetative biomass and root development, where H_2O_2 application led to noticeable improvements. These outcomes likely stem from better nutrient uptake and water retention capabilities under saline conditions, as salt stress typically disrupts ion balance and water availability, impairing root function and overall growth. Similar enhancements in biomass and root systems have been reported in other species like Mungbean and Tomato when H_2O_2 mitigates salt stress (Nehela et al., 2021). At moderate salinity levels (50~150 mM NaCl), H_2O_2 treated maize plants displayed approximately 10%~15% higher biomass than untreated counterparts. This advantage is attributable to strengthened antioxidant defenses which neutralize excess ROS and improved osmotic regulation, allowing plants to maintain physiological balance more effectively. These mechanisms tie directly into H_2O_2 broader role in orchestrating physiological adjustments during abiotic challenges (Ranjan et al., 2023; Saidi et al., 2024).

In terms of yield components, H_2O_2 positively affected key reproductive traits, most notably grain number per plant. Under severe stress at 250 mM NaCl, treated plants retained 88.12 grains per plant, compared to 84.50 in untreated plants. This indicates that H_2O_2 helps sustain reproductive development by minimizing oxidative damage to floral tissues and improving the allocation of assimilates (photosynthates) toward grain formation. Such protective influences on yield have been noted in maize and related crops under salinity (Rehan et al., 2025; Zhao et al., 2025). Nevertheless, the mitigation was only partial at higher salinity levels (200~250 mM NaCl), suggesting that HP protective effects have boundaries under extreme conditions. Severe stress can generate overwhelming ROS levels or cause profound ion toxicity (e.g., excessive Na^+ accumulation), which may exceed H_2O_2 capacity to fully counteract (Sachdev et al., 2021).

Beyond growth and yield, H_2O_2 helped preserve grain quality attributes. Proximate composition, such as protein content, remained more stable in treated plants (14.31% with H_2O_2 versus 13.44% without at 250 mM NaCl). Nutritional elements, including better potassium retention, were also maintained. These outcomes reflect H_2O_2 influence on metabolic stability, enabling continued synthesis of essential compounds and better ion homeostasis despite saline disruption. Related observations in other studies emphasize H_2O_2 contribution to nutrient metabolism and balanced ion regulation under stress (Saritha et al., 2020; Yadesa and Diro, 2023).

Additionally, H_2O_2 treated plants showed superior chlorophyll retention, which supports greater photosynthetic efficiency. Salinity often degrades chlorophyll and impairs light-harvesting complexes, reducing carbon fixation and energy production. By preserving chlorophyll, H_2O_2 indirectly bolsters carbohydrate synthesis and translocation, ultimately contributing to improved grain quality. This pattern mirrors findings in maize and pea subjected to salt stress, where maintained photosynthetic pigments enhance overall plant performance (Zahra et al., 2022; Stefanov et al., 2024).

Overall, this investigation addresses an important knowledge gap regarding maize specific applications of H_2O_2 under salinity, drawing parallels to how other modulators like salicylic acid have been used successfully in different crops (Elsisi et al., 2024). The findings position H_2O_2 as a promising, sustainable tool for boosting maize resilience in saline prone agricultural areas, where soil salinization is increasingly driven by climate change, poor irrigation practices, and other factors (Singh, 2022). That said, the incomplete protection at very high salinity levels underscores the need for additional studies to fine tune H_2O_2 concentrations, application timing (e.g., priming versus foliar sprays), and methods to achieve optimal results under severe conditions. Such optimization could further enhance its practical utility in saline agriculture.

5 Conclusion and Recommendations

In conclusion, these screenhouse-based results indicate that hydrogen peroxide shows promise as a signaling molecule capable of partially alleviating salinity stress effects on maize, potentially contributing to improved resilience in saline environments and supporting food security in affected areas (as highlighted in global assessments of salt-affected soils). However, the evidence remains preliminary and context-specific to controlled conditions.

The study demonstrates that salinity stress markedly impairs *Zea mays* (maize) growth, yield, grain nutritional quality, and leaf chlorophyll content, with the strongest negative impacts observed at 250 mM NaCl. Key parameters such as plant height, leaf production, stem girth, root development, biomass accumulation, grain number (declining from 226.25 to 84.50 per plant at 250 mM NaCl without mitigation), and grain proximate composition (e.g., protein decreasing from 15.14% to 13.44%, fat from 1.88% to 1.74%, crude fiber from 3.40% to 2.74%) were progressively reduced, consistent with effects of osmotic stress and ion toxicity. Chlorophyll content also declined, likely due to chloroplast damage affecting photosynthetic capacity.

Based on this research the application of hydrogen peroxide (H_2O_2) consistently alleviated these adverse effects across the tested salinity levels (50~250 mM NaCl). Treated plants showed improvements, including greater plant height (123.52 cm vs. 112.19 cm at 250 mM NaCl), higher biomass (approximately 10%~15% increase), increased grain number (88.12 vs. 84.50 per plant), enhanced grain quality (e.g., protein at 14.31%, fat at 2.41%, crude fiber at 2.80%), and better maintenance of chlorophyll content, which may support improved photosynthetic efficiency. These observations align with prior research indicating that exogenous H_2O_2, often applied as a priming or foliar treatment, can enhance antioxidant defenses, protect chloroplast ultrastructure, modulate metabolites, and improve physiological performance under salt stress in maize. Nonetheless, H_2O_2 mitigation did not completely restore parameters to non-stressed control levels, especially under severe salinity (250 mM NaCl), suggesting inherent limitations in extreme conditions.

Field validation under natural saline soils, along with further exploration of optimal application methods, concentrations, physiological/biochemical mechanisms (e.g., antioxidant enzyme responses, ionic homeostasis), and possible integration with other amendments or interventions, would be essential to strengthen any practical recommendations for maize production in salt-affected regions.

Author's contribution

O. Kekere was the experimental designer, and J. K. Afolabi was executor of the study; J. K. Afolabi completed data analysis and wrote the first draft of the paper; J. K. Afolabi participated in the experimental design and analysis of experimental results; O. Kekere was the project conceptualizer and leader, guiding experimental design, data analysis, paper writing and revision. The paper was read and received the approval of both authors for publication in the journal.

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