



Nano-NPK and Nano-Sulfur Boost Vegetative Growth and Chemical Constituents of African Mahogany (*Khaya senegalensis* L.) Seedlings under Saline Soil Conditions



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SOIL salinity is considered a serious and limiting factor reducing the crop productivity in particular under arid, and semi-arid conditions. Pot experiments were conducted at the Ornamental Department of the Horticulture Research Station in Sakha, Kafr El-Sheikh Governorate, Egypt during the season of 2021 and 2022. This study aimed to evaluate the effect of foliar application with nano-NPK and nano-sulfur using different doses on the growth and chemical compounds of *Khaya senegalensis* L. under salinity stress. The studied treatments were mineral NPK fertilizer at 5 g L⁻¹ (as a control), applied nano-NPK at 1, 2 and 3 ml L⁻¹, besides three combinations of nano NPK (2 ml L⁻¹) and applied nano-sulfur at 1, 2 and 3 ml L⁻¹, separately. The obtained results showed that the treatment of nano-NPK 2 ml L⁻¹ + nano-sulfur 2 and/or 3 ml L⁻¹ significantly increased plant height (cm), stem diameter (cm), fresh and dry weights of vegetative parts per seedling (g), membrane stability index, as well as root length (cm), number of roots and root fresh and dry weights per plant. The superiority of the nano-NPK at 2 ml L⁻¹ + nano-sulfur at 2 ml L⁻¹ treatment comparing with the control in both seasons. Most treatments recorded significantly higher chlorophyll a, b, carotenoids, catalase and peroxidase activity in the leaves than control. A superiority was found after applying nano-NPK 2 ml L⁻¹ + nano-sulfur 2 ml L⁻¹ in both seasons for nitrogen, phosphorus and potassium content in the leaves, which was significantly increased over the control.

Keywords: Nanofertilizers, Salinity stress, Catalase, Peroxidase, Photosynthetic pigments, Carotenoids.

1. Introduction

Khaya senegalensis Desr A. Juss., is a woody and ornamental plant commonly known as African mahogany, belongs to Meliaceae family. It is an evergreen Savanna hard wood tree species, with a crown of dark shining pinnate leaves that bear round capsules. This species is regenerated mainly naturally. Khaya provides a major source of revenue to many countries because of its hard, durable timber, which is widely sought for construction, furniture and carpentry (Sahu *et al.* 2023).

Nano-fertilizers play an important role in plant nutrition, through their applying to soil and foliar spraying on the vegetative system (Singh *et al.*

2024a, b). Many benefits have reported on the applied nano-fertilizers particularly under stressful conditions (Shalaby *et al.* 2022a), such as drought (Sári *et al.* 2024a), climate change (Sári *et al.* 2024b), salinity (Sári *et al.* 2023; Singh *et al.* 2023c; El-Ramady *et al.* 2024; Mahawar *et al.* 2024), nutrient deficiency (El-Bialy *et al.* 2023), and biotic stress (Tortella *et al.* 2023) for sustainable agriculture (Singh *et al.* 2023b). Nanofertilizers are promising candidates for sustainable agriculture, and nano-farming approach particularly the biological nanofertilizers (El-Ramady *et al.* 2023). These kinds of fertilizers may consider slow-release of nutrients, and high nutrient use efficiency, which supply cultivated plants with the right amounts of nutrients

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for a long time comparing with mineral fertilizers (Haydar *et al.* 2024).

Based on the nutrient kind, nanofertilizers can be classified into nanofertilizers of macro- (N, K, P, etc.) and micro-nutrients (Cu, Mo, Mn, Zn, etc.), along with nanocomposites-based fertilizers (Chakraborty *et al.* 2023). Nano-fertilizers enhance growth parameters such as plant height, leaf area and number of leaves per plant, dry matter production, chlorophyll synthesis and photosynthesis rate, which result in huge production compared to mineral fertilizers particularly during the seedling growing stage (Shalaby *et al.* 2022b; Sundararajan *et al.* 2023; Haydar *et al.* 2024; Zhao *et al.* 2024). Nano-composite fertilizer consists of N, P and K, micronutrients which increase the uptake and utilization of nutrients by crops owing to the properties such as large surface area and capability (Yang *et al.* 2023). Nano-formulations of sulfur have proved a sound potential in enhancing plant growth and reducing disease incidence against phytopathogens (Hammerschmidt *et al.* 2023; Steven *et al.* 2024).

Soil salinity is a limiting factor reducing the crop productivity and tree growth, leading to a serious threat to feed a growing population (Ul Hassan *et al.* 2023). Applied nanofertilizers can mediate plant tolerance and mitigation of salinity through many mechanisms including enhancing plant retaining of K^+ and eliminating Na^+ , maintaining reactive oxygen species (ROS) homeostasis, increasing nitric oxide production, and lowering lipoxygenase activities (Singh *et al.* 2023a). Many approaches have been applied to ameliorate salinity including nanomaterials such as nanofertilizers (Ibrahim and Hegab 2022; Singh *et al.* 2024a), nano-biochar (Rasheed *et al.* 2024), nano-amendments like sulfur (EL-Madah *et al.* 2024), organic fertilizers (Elshaboury *et al.* 2024), biostimulants (AbdEL-Azeiz and Faiyad 2024), and nano-priming of seeds (Zhao *et al.* 2024).

Therefore, this investigation aimed to evaluate the effect of foliar application of nano-NPK and nano sulfur using different doses on the growth, and chemical constituents of *Khaya senegalensis* Desr A. Juss during the seedling growing stage. The vegetative growth, antioxidant activities, photosynthetic pigments, and chemical composition of leaves under soil salinity were reported.

2. Materials and Methods

Pot experiments were conducted at the Ornamental Department of the Horticulture Research Station in

Sakha, Kafr El-Sheikh Governorate (the site is located at 31.07° N altitude and 30.57° E longitude with an elevation of about 6 meters above sea level) during two seasons 2021 and 2022. Data climatic conditions of this study were obtained from the Agriculture Research Center during the two experimental seasons and presented in Table (1). On woody plants few studies were done on the effect of nanofertilizer on the growth, so this study aimed to evaluate the effect of nano-NPK and nano sulfur with different levels as foliar application on the growth and chemical compounds of *Khaya senegalensis* L. seedling. The used soil was collected from the Station in Sakha, Kafr El-Sheikh Governorate.

The used soil in each season was physically and chemically analyzed according to Sparks *et al.* (2020) before cultivation as the soil texture was clayey loam and the chemical properties were pH 8.12, and SOM 1.02 %, available nutrients $mg\ kg^{-1}$ were nitrogen 37.06, phosphorus 7.03 and potassium (K_2O) 212.47 in the first season. In the second one soil texture was clay loam and the chemical properties were pH 8.03, EC 2.12 dsm^{-1} and OM 1.15%, available nutrients ($mg\ kg^{-1}$) were N 42.16, phosphorus 8, 34 and potassium 209.82.

2.1 Plant materials

Three-month-old produced seedlings were obtained from a private nursery on march 25th for the two seasons at a uniform length of 15 ± 1 cm then on April 5th the plants were planted in pots of 20 cm diameter with a clayey loamy soil (8 kg). The pots were manually watered every 3 days by using fresh water. Randomized completely block design was performed seven treatments were assessed each treatment consisted of three replicates as every replicate contained 10 plants. After one month later seedlings were foliar sprayed with mineral NPK (5g L^{-1}), as a control, nano-NPK at 1, 2 and 3 $ml\ L^{-1}$ and nano-NPK at 2 $ml\ L^{-1}$ + nano sulfur at 1, 2, 3 $ml\ L^{-1}$ for each (Figure 1). During the two experimental seasons at interval of 15 day the foliar spraying was done in early morning until run off. Nano-sulfur was foliar sprayed in the second day from adding NPK at 2 $ml\ L^{-1}$.

2.2 Source of chemicals

The mineral NPK (EGY FLEX, as commercial fertilizer 20:20:20) is composed of total nitrogen 20 % N, phosphorus (20% P_2O_5), potassium (20 % K_2O) produced by Egypt Chem International for Agrochemicals. Nano-NPK (19:6:20) produced by Biota EG Company with concentration of nitrogen of (3.8% N), phosphorus (1.2 % P_2O_5) and potassium

(4% K₂O). Nano sulfur solution concentrated at 4% produced by Biota EG Company. Transmission Electron Microscopy (TEM) of both nano-NPK and nano-sulfur can be observed in **Figure 2**. These nanoparticles were measured directly by TEM

(Model Talos L120CG2 – TEM – Thermo- Fisher, Europe). The mean diameters of applied nanomaterials were **308 and 350 nm** for nan-NPK and nano-sulfur, respectively.

Table 1. Monthly air temperature (max., min. and mean °C), relative humidity (RH %), at the experimental during the two growing seasons of 2021 and 2022.

Months	Air temperature (°C)			RH, %	Air temperature (°C)			RH, %
	Max	Min	Mean		Max	Min	Mean	
	First season (2021)				Second season (2022)			
March	22.3	17.6	19.9	62.6	21.7	16.8	19.2	63.5
April	26.9	20.4	23.6	59.6	27.9	19.7	23.8	61.0
May	32.4	24.7	28.5	58.3	31.0	21.8	26.4	59.5
June	30.9	25.5	28.2	65.1	33.0	25.7	29.3	64.7
July	33.9	27.9	30.9	69.6	33.4	26.6	30.0	71.7
Augustus	35.6	28.3	31.9	66.2	34.5	25.9	30.2	72.7
September	32.5	25.1	28.8	66.8	32.9	25.9	29.4	69.3
October	28.5	22.3	25.4	69.6	29.6	20.4	25.0	74.9
November	26.6	19.9	23.2	72.1	25.7	18.5	22.1	74.3

Table 2. Physical and chemical analysis of the used soil the 1st and 2nd two Experimental season 2021 and 2022.

Season	Soil Physical properties					pH*	EC** dS m ⁻¹	OM, %	Soil chemical properties		
	Coarse sand	Fine sand	Silt	Clay	Texture class				Available nutrients (ppm)		
									N	P	K
Frist season	2.53	22.05	37.79	37.63	Clay loam	8.12	4.56	1.02	37.06	7.03	212.4
Second season	2.71	21.59	35.28	40.42	Clayey	8.03	4.66	1.15	42.16	8.34	209.8

In both season pH* was determined in soil suspension (1: 5) and Soil Electrical Conductivity (EC) ** was determined in saturated soil paste extract.

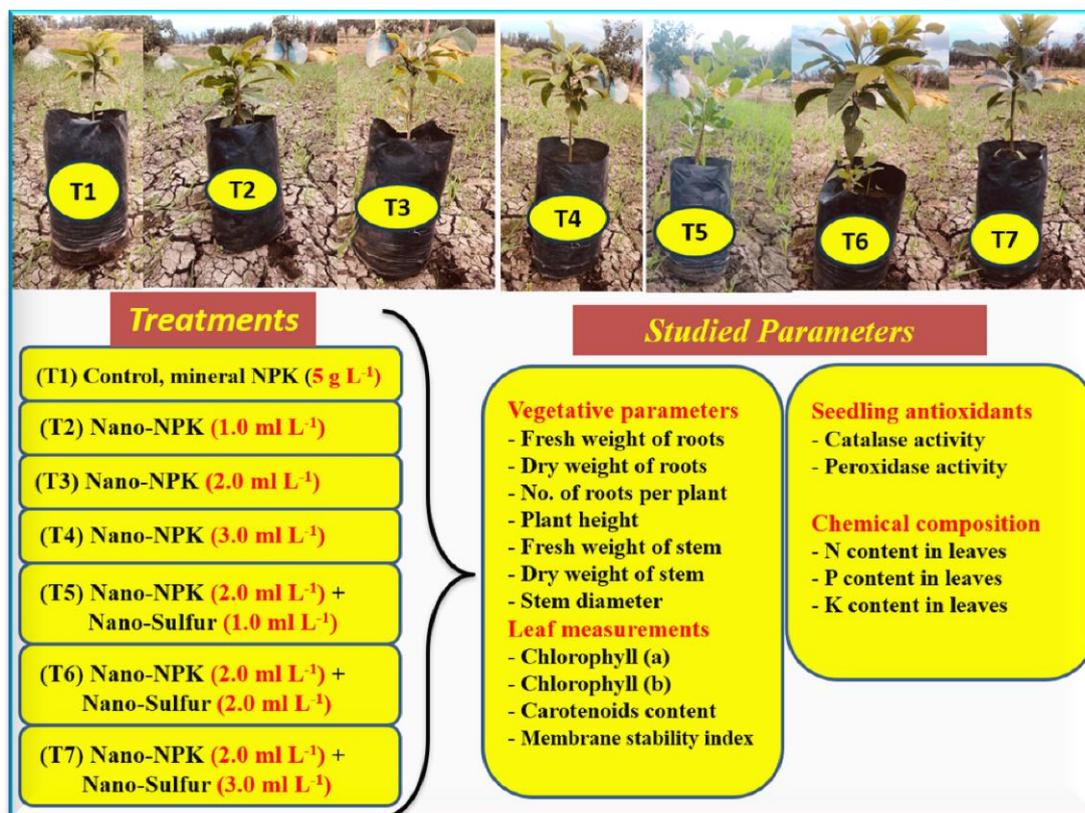


Fig. 1. An overview on the main treatments and measurements during the study.

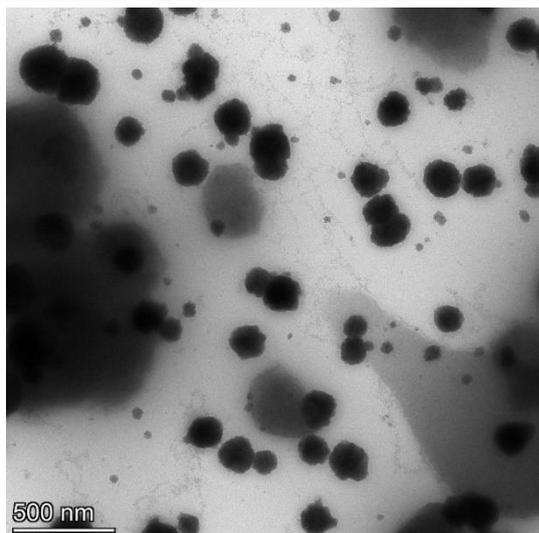


Fig. 2. Transmission Electron Microscope (TEM) imaging of nano-sulfur.

2.3 Vegetative growth parameters

At the end of experimental seasons on November 10th to 15th 2021 and 2022 respectively, the following data were recorded including plant height (cm), stem diameter (cm), fresh and dry weights of stem per plant (g), as well as root length (cm), number of roots and root fresh and dry weights (g) per plant.

2.4 Estimation of chlorophyll content

Chlorophyll content was measured using 20 ml of acetone 80% + 1 g leaf tissue and 0.5 g of (MgCO₃) powder was added and further grinded gently. The mixture was then incubated at 4°C for 3 hrs. The mixture was centrifuged at 2500 rpm for 5 min and the supernatant was transferred to a 100 ml volumetric flask and the volume was made up to 100 ml with the addition of acetone 80% and the solution was used for estimation chlorophyll a and b (Chl. a and Chl. b) were calculated using the following equation according to Rajalakshmi and Banu (2014):

$$\text{Chlorophyll a (mg/gm fw)} = [12.7(A_{663}) - 2.69(A_{645})] V/1000W$$

$$\text{Chlorophyll b (mg/gm fw)} = [22.9(A_{645}) - 4.68(A_{663})] V/1000W$$

Where A = Absorbance of specific wavelength, V = Final volume of chlorophyll extract in 80% acetone and W = Fresh weight of tissue extract.

2.5 Estimation of total carotenoids

The content of carotenoids was estimated according to Sumanta *et al.* (2014) using the same chlorophyll extract as measured at 470 nm in spectrophotometer (Jenway 6405, the UK) to estimate the total content of carotenoids (including both xanthophylls + carotene), as follows:

$$\text{Total carotenoids (mg/gm fw)} = (1000A_{470} - 1.82 \text{ Chl. a} - 85.02 \text{ Chl. b})/198$$

Where, A = Absorbance at respective wave length, Chl. a = chlorophyll a and Chl. b = chlorophyll-b.

2.6 Membrane stability index

Membrane stability index (MSI) or electrolyte leakage (EL) was determined according to Sairam *et al.* (1997). Leaf disks (200 mg) were taken in two sets of test tubes containing 10 ml of distilled water. One set was kept at 40°C in a water bath for 30 min and electrical conductivity (C1) was measured. The second set was incubated at 100°C for 15 min and electrical conductivity (C2) was measured. MSI was calculated according to the following formula:

$$\text{MSI (\%)} = (1 - C1/C2) \times 100$$

2.7 Leaf chemical composition

The content of nitrogen (N), phosphorus (P) and potassium (K) in the dried leaves were achieved using 0.2 gm were digested according to Peterburgski, (1968) as digested solution was used to determine N, P and K by Kjeldahl device, spectrophotometer (Jenway 6405) and flame photometer (Jenway PFP7, Staffordshire, UK) of the three elements, respectively (Mertens, 2005).

2.8 Determining of enzyme activities

The crude enzymatic extracts for determining the activities of catalase and peroxidase were obtained by soaking 0.3 g of leaves in liquid nitrogen and then adding 2 mL of homogenized medium; followed by centrifugation at 12,000×g for 15 min at 4 °C. The homogenization media were 0.1 M potassium phosphate buffer, pH 6.8, 0.1 Mm EDTA, 1 mM phenyl methyl sulfonyl fluoride (PMSF), and 1 % poly vinyl pyrrolidone (PVPP) (w/v) (Peixoto *et al.* 1999) for the enzymes CAT, POX 50 mM potassium phosphate buffer, pH 7.0, 1 mM ascorbate, and 1 mM EDTA (Nakano and Asada, 1981).

The catalase (EC 1.11.1.6) activity was determined by the addition of 0.1 mL of enzymatic extract to 2.9 mL of reaction solution that was composed of 50 mM potassium phosphate buffer at pH 7.0 and 12.5 mM H₂O₂ (Havir and McHale 1987). The decreasing absorbance during the first minute of reaction was measured at 240 nm at 25 °C. The enzymatic activity was calculated using a molar extinction coefficient of 36 mol L⁻¹ cm⁻¹ and expressed by $\mu\text{mol of H}_2\text{O}_2 \text{ min}^{-1} \text{ mg}^{-1} \text{ fw}$ (Anderson *et al.*, 1995). Catalase and peroxidase were determined by spectrophotometer (Jenway 6405).

The POX activity (EC 1.11.1.7) was assessed through the production rate of purpurogallin at 420 nm according to the proposed method of Nakano and Asada (1981) with a molar extinction

coefficient of $2.47 \text{ mmol}^{-1} \text{ L cm}^{-1}$. The enzymatic activity was expressed in $\mu\text{mol purpurogallin min}^{-1} \text{ g}^{-1} \text{ fw}$ (Chance and Maehley 1955).

2.9 Statistical analyses

Data were statically analyzed by analysis of variance (ANOVA) using Costat (Version 6.303, Co Hort, USA, 1998–2004)] program for data set of the two independent experimental and combined analysis was carried out after checking homogeneity of the variance Bartlett's test. Duncan's multiple range test (Duncan, 1955) was used in order to compare the mean at the $P \leq 0.05\%$ probability level according to Gomez and Gomez (1984). Resulted were presented as average mean of the two independent seasons \pm SE.

3. Results

3.1 Vegetative characteristics

In general, all studied values were recorded that highest values of vegetative parameters comparing with the control with superiority to nano-form than mineral form. Data of Khaya growth parameters included, plant height stem diameter, fresh and dry weight, beside the leaves number, are listed in **Table (3)**. The treated plants with N, P and K nano-fertilizer at different levels or plus nano-sulfur at all levels were significantly superior in all traits compared to control (mineral fertilizer) treatment. Nano-NPK (NPK-NPs) at 2 ml L^{-1} plus nano-sulfur

(S-NPs) at 2 ml L^{-1} gave the tallest plants (69.36 cm) compared to the control followed by the increase in seedling treated with nano-NPK at 2 ml L^{-1} plus nano-sulfur at 3 ml L^{-1} then nano-NPK at 2 ml L^{-1} plus nano-sulfur at 1 ml L^{-1} while the control treatment gave the lowest value (63.88 cm).

Nano-NPK fertilizer under different doses significantly increased in the length as compared to the control. Data listed in **Table (3)** showed that spraying plants with nano-NPK at different doses or nano-NPK at 2 ml L^{-1} plus nano-sulfur at 1, 2, 3 ml L^{-1} precipitate the mineral fertilizer (control). Nano-NPK plus nano-sulfur at 2 ml L^{-1} gave the highest value of stem diameter (13.0 cm) followed in terms of the increase treating with NPK-NPs at 2 ml L^{-1} plus nano-sulfur at 2 ml L^{-1} which gave average stem diameter compared to the control treatment which resulted the lowest stem diameter per plant (8.98 cm). Data resulted in **Table (3)** showed significantly superiority in fresh and dry weight of plants treated with NPK-NPs and nano-NPK plus sulfur at different doses compared to the control (mineral fertilizer). Adding sulfur-NPs at different doses along with the nano-NPK (2 ml L^{-1}) caused increase in fresh and dry weights of plant with increasing the applied dose of S-NPs.

Table 3. Effect of nano NPK and nano sulfur foliar application on stem growth of *Khaya senegalensis* L plants (average of two seasons, 2021 and 2022).

Treatments	Plant height (cm)	Stem diameter (cm)	Fresh weight of stem (g)	Dry weight of stem (g)
(T1) Control (mineral NPK 5 g L^{-1})	63.88d	8.98g	43.85e	11.05g
(T2) NPK-NPs 1 ml L^{-1}	64.15d	9.83f	46.20d	13.74f
(T3) NPK-NPs 2 ml L^{-1}	64.81cd	11.19d	51.00c	15.78d
(T4) NPK-NPs 3 ml L^{-1}	64.57cd	10.44e	46.59d	14.10e
(T5) NPK-NPs 2 ml L^{-1} + S-NPs 1 ml L^{-1}	65.47c	11.58c	55.76b	16.82c
(T6) NPK-NPs 2 ml L^{-1} + S-NPs 2 ml L^{-1}	69.36a	13.00a	65.20a	20.23a
(T7) NPK-NPs 2 ml L^{-1} + S-NPs 3 ml L^{-1}	66.83b	12.26b	64.17a	19.36b
F-test	**	**	**	**

Mean followed by the same letter in the same column do not differ significantly by Duncan's multiple range test at 5% level.

3.2 Root growth parameters

Table (4) shows a significant prominence of the treatment of NPK-NPs at different doses as well as supplying (S-NPs) to the NPK-NPs at 2 ml L^{-1} improved the root length, root number, root fresh and dry weights in comparison with the control treatment (mineral NPK). The highest root length (40.96 cm) was observed for using foliar spray NPK-NPs at 2 ml L^{-1} plus S-NPs at 2 or 3 ml L^{-1} whereas, the lowest value belongs the control treatment (35.64 cm). A similar trend was recorded for all parameter of roots as shown in **Table (4)** comparing with the control treatment.

3.3 Photosynthetic pigments

The values of NPK-NPs or NPK-NPs plus S-NPs at different applied doses increased by increasing the applied doses for chlorophyll a, b and carotenoids (**Figure 3A, B, and C**). The highest values of previous parameters (1.19 , 0.48 and $0.37 \text{ mg g}^{-1} \text{ fw}$) were recorded after applying T6 (nano-NPK at 2 ml L^{-1} plus nano-sulfur at 2 ml L^{-1}), whereas the lowest value of chlorophyll a, b and carotenoids were recorded by the control (1.02 , 0.4 and $0.29 \text{ mg g}^{-1} \text{ fw}$, respectively).

Table 4. Effect of nano-NPK and nano- sulfur foliar application on root growth parameter of *Khaya senegalensis* L. plants (average of two seasons, 2021 and 2022).

Treatments	Root length (cm)	No. of roots per seedling	Root fresh weight (g)	Root dry weight (g)
(T1) Control (mineral NPK 5 g L ⁻¹)	35.64 e	3.33 e	9.80 f	3.78d
(T2) NPK-NPs 1 ml L ⁻¹	36.16de	3.83 de	12.33 e	4.57 c
(T3) NPK-NPs 2 ml L ⁻¹	37.50 c	4.67 c	14.17d	4.75 c
(T4) NPK-NPs 3 ml L ⁻¹	36.85cd	4.33 cd	14.10 d	4.70 c
(T5) NPK-NPs 2 ml L ⁻¹ + S-NPs 1 ml L ⁻¹	39.92 b	5.67 b	15.36 c	5.24 b
(T6) NPK-NPs 2 ml L ⁻¹ + S-NPs 2 ml L ⁻¹	40.96 a	6.83 a	22.68 a	7.10 a
(T7) NPK-NPs 2 ml L ⁻¹ + S-NPs 3 ml L ⁻¹	40.68 a	6.67 a	22.13 b	7.02 a
F-test	**	**	**	**

Mean followed by the same letter in the same column do not differ significantly by Duncan's multiple range test at 5% level

3.4 Membrane stability index (MSI)

Data presented **Figure (3D)** showed that there were non-significant differences between all treatment and control in membrane stability index (MSI) in leaves but the treating seedlings with nano-NPK 2

ml L⁻¹ plus nano-sulfur at dose of 2 ml L⁻¹ gave the highest value of MSI (60.9 %) compared the control treating with mineral fertilizer, which recorded the lowest value (58.9%).

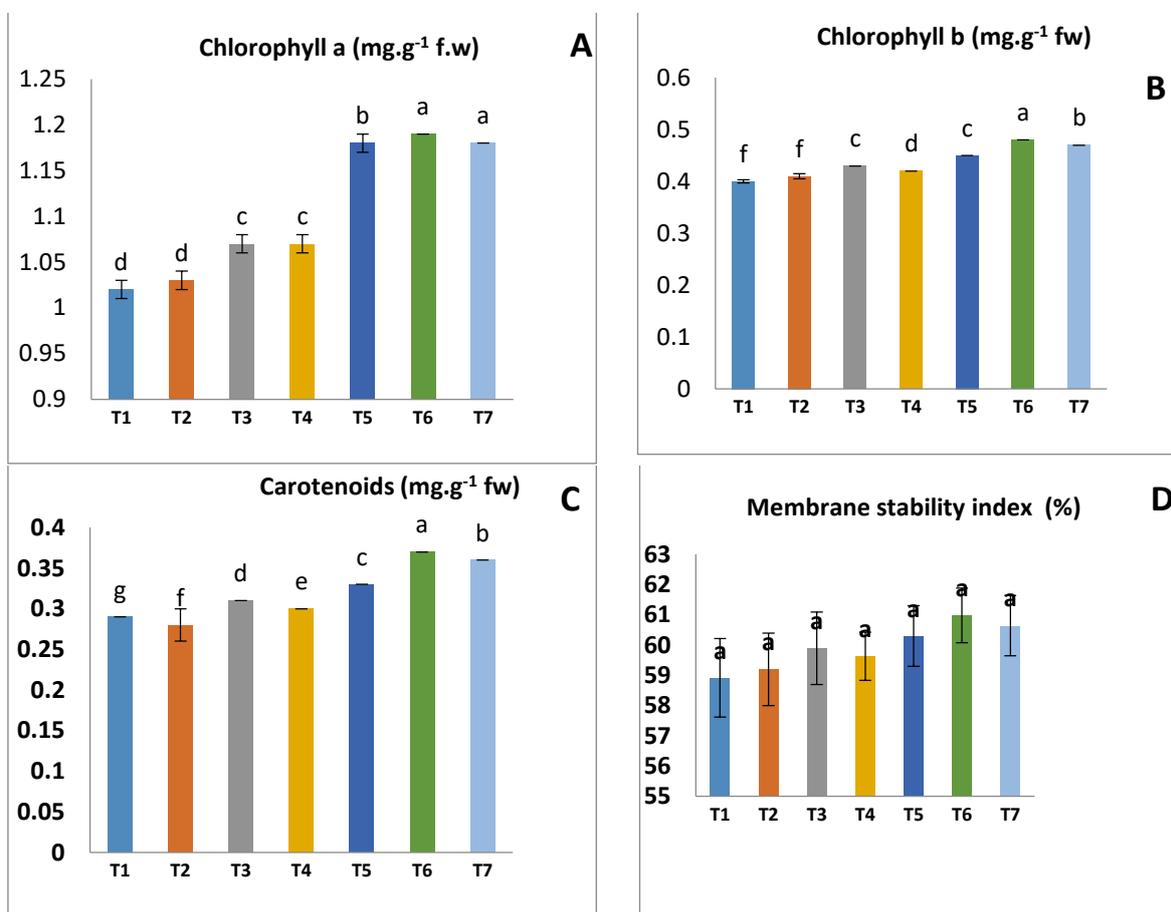


Fig 3. Effect of nano-NPK and nano-sulfur foliar application on photosynthetic pigments and Membrane stability index (MSI) of *Khaya senegalensis* L. as (A) chlorophyll a (mg g⁻¹ fw), (B) chlorophyll b (mg g⁻¹ fw), (C), carotenoids (mg g⁻¹ fw) and (D) membrane stability index (%). Data are mean value ± SE. Bars at the same letter are not significant at P≤0.05 level. (T1) Control (mineral NPK 5 g L⁻¹), (T2) NPK-NPs 1 ml L⁻¹, (T3) NPK-NPs 2 ml L⁻¹, (T4) NPK-NPs 3 ml L⁻¹, (T5) NPK-NPs 2 ml L⁻¹ + S-NPs 1 ml L⁻¹, (T6) NPK-NPs 2 ml L⁻¹ + S-NPs 2 ml L⁻¹, (T7) NPK-NPs 2 ml L⁻¹ + S-NPs 3 ml L⁻¹ (average of two seasons, 2021 and 2022).

3.5 Leaf chemical composition

An increase was observed in all treatments after applying different doses of both NPK-NPs and S-NPs regarding the content of N, P, and K in the seedling leaves (**Figure 4**). The highest value of N, P, and K content (2.62, 0.56 and 1.8 %) in plant leaves achieved after foliar application of nano-

NPK along with nano-sulfur at 2 ml L⁻¹ followed by treated plants with nano-NPK at 2 ml L⁻¹ plus nano-sulfur using 2 or 1 ml L⁻¹ whereas, the control treatment gave the lowest values (2.04, 0.48, and 1.22 %).

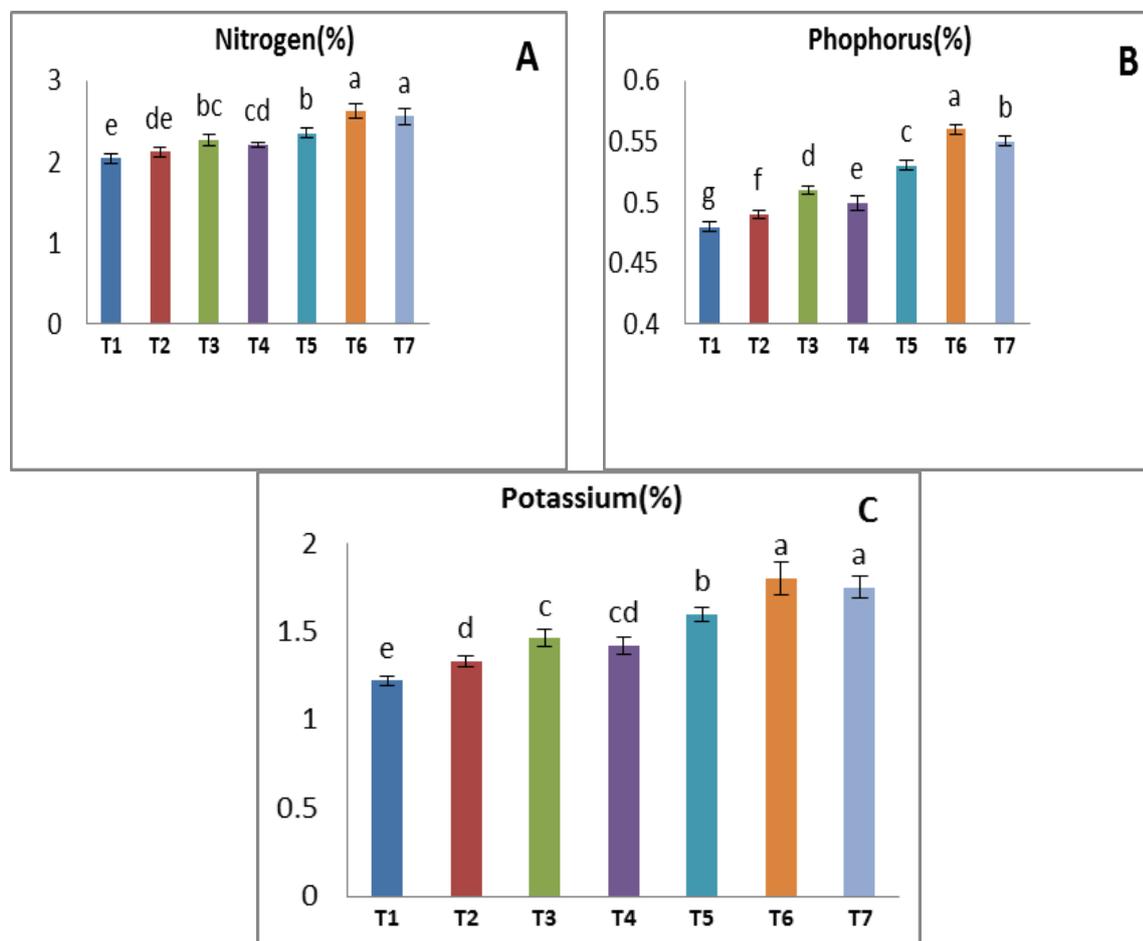


Fig. 4. Effect of nano-NPK and nano-sulfur foliar application on chemical composition nitrogen, phosphorus and potassium of *Khaya senegalensis* L. in leaves (%) for N, P, K (A), (B) and (C), respectively. Data are mean value \pm SE. Bars at the same letter are not significant at $P \leq 0.05$ level. (T1) Control (mineral NPK 5 g L⁻¹), (T2) NPK-NPs 1 ml L⁻¹, (T3) NPK-NPs 2 ml L⁻¹, (T4) NPK-NPs 3 ml L⁻¹, (T5) NPK-NPs 2 ml L⁻¹ + S-NPs 1 ml L⁻¹, (T6) NPK-NPs 2 ml L⁻¹ + S-NPs 2 ml L⁻¹, (T7) NPK-NPs 2 ml L⁻¹ + S-NPs 3 ml L⁻¹ (average of two seasons, 2021 and 2022).

3.6 Antioxidant activity

Data illustrated in **Figure (5A)** revealed that the treated plants with NPK-NP fertilizer at different doses or plus nano-sulfur at all levels were significantly superior in the catalase and peroxidase activities. Concerning catalase activity ($\mu\text{mol of H}_2\text{O}_2 \text{ min}^{-1} \text{ mg}^{-1} \text{ fw}$) data showed that the treating plants with nano-NPK at 2 ml L⁻¹ along with nano-sulfur at either 2 or 3 ml L⁻¹ resulted the highest significant values of catalase activity (30.52, and

30.02) which gave the equivalent values in significant, followed by nano-NPK at 2 ml L⁻¹ plus nano-sulfur at 2 ml L⁻¹, whereas the lowest value was recorded from the control (24.54). For peroxidase activity ($\mu\text{mol purpurogallin min}^{-1} \text{ g}^{-1} \text{ fw}$), the same previous trend that recorded by catalase (**Figure 5B**).

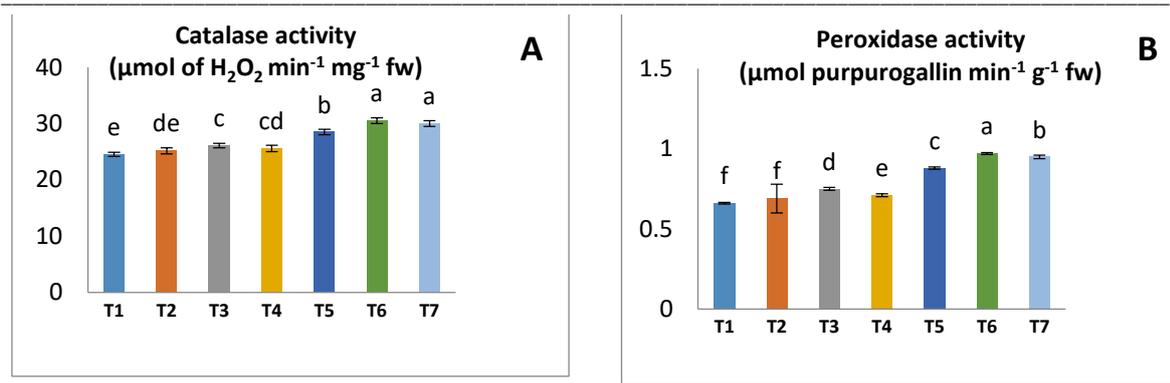


Fig. 5. Effect of nano-NPK and nano-sulfur foliar application on antioxidant activity of *Khaya senegalensis* L. as (A) catalase activity ($\mu\text{mol of H}_2\text{O}_2 \text{ min}^{-1} \text{ mg}^{-1} \text{ fw}$) and (B) peroxidase activity ($\mu\text{mol purpurogallin min}^{-1} \text{ g}^{-1} \text{ fw}$). Data are mean value \pm SE. Bars at the same letter are not significant at $P \leq 0.05$ level. (T1) Control (mineral NPK 5 g L⁻¹), (T2) NPK-NPs 1 ml L⁻¹, (T3) NPK-NPs 2 ml L⁻¹, (T4) NPK-NPs 3 ml L⁻¹, (T5) NPK-NPs 2 ml L⁻¹ + S-NPs 1 ml L⁻¹, (T6) NPK-NPs 2 ml L⁻¹ + S-NPs 2 ml L⁻¹, (T7) NPK-NPs 2 ml L⁻¹ + S-NPs 3 ml L⁻¹ (average of two seasons, 2021 and 2022).

4. Discussion

This section will answer the following main questions in the present study:

- 1- What are the NPK-NP and S-NP-fertilizers?
- 2- What is the suggested mechanism of such nano-fertilizers?
- 3- What is the expected combined application of the studied nanofertilizers?
- 4- To what extent can consider the studied candidates are promising nanofertilizers under salinity stress?
- 5- Are *Khaya* seedlings tolerant plants to salinity?

The reason beyond the selection of this tree for the current study may back to its importance in many therapeutic activities in both people and animals by treat a variety of diseases including dermatoses, malaria, jaundice, leprosy, syphilis, and mental illness (Amang *et al.* 2023). The growing of such tree depends on the growing media and environmental stresses like soil salinity under arid and semi-arid zones (**Table 1**). The mitigation of soil salinity was investigated by applying many materials and/or certain framing practices including nanomaterials (NMs). Under forestry cultivation, the potential application of nanotechnology or nanofertilizers still require research and evidence particularly in producing good quality transplants under nursery growing conditions (Singh *et al.* 2021).

The influence of nano-fertilizers through foliar spraying has been suggested for amplifying plant growth of different horticultural species under environmental stress particularly seedling stage such as banana (Shalaby *et al.* 2022b), and strawberry (El-Bialy *et al.* 2023). In the current investigation, the

combined foliar spraying of NPK-NPs fertilizer and/or with nano-sulfur at different applied rates had significantly positive effects on the morphological parameters including seedling height, stem diameter, fresh and dry weights and leaf number comparing with the control. This effect significantly increased with increasing the added dose of nano-NPK and/or nano-S up to 2 ml L⁻¹ comparing with the control. This may be attributed to the role of nano-fertilizers in ameliorating salinity stress by modulating the oxidative and salt stress, as well as promoting the antioxidative role under such stress (Singh *et al.* 2023). The increase in leaves number almost resulted from the physiological role of studied nutrients (N, P, K, and S), which are essential and responsible for improving the shoot growth and the accumulation of the carbohydrate substance and stimulate the vegetative growth as well (Bang *et al.* 2020). It could notice that applied NPK-NPs depends on applying soluble forms of NPK mineral fertilizers in a polymer, which let these NPK nutrients release slowly to plants as -slow-release fertilizer. The applied nano-fertilizers are a source of needed nutrients (N, P, K, and S), which are essential for all morphological (all vegetative parameters), physiological (photosynthetic pigments), and biochemical attributes (enzymatic antioxidants) of studied seedlings.

The role of nano-NPK and or nano-S for increasing total fresh and dry weight and elevated percent of chlorophyll a, b and carotenoids content in this study may be due to the beneficial effect of nano fertilizers in increasing the bioavailability of such necessary nutrients to the growing plants leading to increase

chlorophyll forming, dry matter production and improved overall growth of the plant under such studied stress (Saad-Allah and Ragab 2020; EL-Madah *et al.* 2024)

How can foliar nanofertilizers attribute to the previous impact on mahogany seedlings? Foliar sprayed nanoparticles to plant may enter stomata by increasing the osmotic pressure of the stomata cell, the frequently opening the stomatal cells and enabled the plant to receive more nutrients loaded on the surface of NPs as indicated by Qureshi *et al.* (2018). These results an agreement with those obtained by Haggag (2018), (Shalaby *et al.* 2022b), and (El-Bialy *et al.* 2023), who observed that nanofertilizers significantly enhanced the uptake of nitrogen, phosphorus and potassium in olive, banana and strawberry seedlings, respectively.

Data of the study showed that adding nano-sulfur at different levels can play an important role in the growth of *Khaya senegalensis* seedlings under salinity stress compared to the control as these treatments augmented the morphological traits including plant height, stem diameter, number of leaves fresh and dry weight as in **Table (3)** also increased root parameters as root length, number of roots, root fresh and dry weight as recorded in **Table (4)**. These results are confirmed with those obtained by Thirunavukkarasu *et al.* (2023). Chlorophyll a, b and carotenoids content as well as catalase and peroxidase activities were recorded the highest values in the seedling leaves after applying nano-NPK plus nano-sulfur using 2 ml L⁻¹ from both nanofertilizers as compared to untreated plants.

Therefore, application of such nanofertilizers can play a promising role in increasing studied seedlings to be tolerant against soil salinity. The role of S-NPs as a nanofertilizer is distinguished for promoting the plant chlorophyll content due to sulfur is a part of carbon compounds found in two amino acids and is a precursor for many coenzymes and vitamins essential for metabolism (Taiz and Zeiger 2002). Nano-S can also improve plant growth by increasing the efficiency of chemical energy production in the photosynthesis system and also improving plant growth and biomass production and many studies have showed that the application of nanofertilizer led to an increase in the content of photosynthetic pigments when using appropriate dose of sulfur-NPs which plays antioxidant role or modulate the antioxidant defense system (Najafi *et al.* 2020).

Nanoparticles of sulfur hold promise for enhancing plant nutrition when incorporated into saline/alkaline soil due to their significantly increased surface area, facilitating the transformation into soluble sulfur compounds. These compounds are readily absorbed by plant roots, thereby augmenting the plant's sulfur uptake. However, when applied via foliar spraying, sulfur nanoparticles may encounter limitations, which needs more studies for more details. The insoluble nature of sulfur granules can hinder their incorporation into plant metabolites, potentially resulting in reduced efficacy. Moreover, foliar application may lead to sulfur deposition on leaf surfaces, limiting stomatal exchange and impeding nutrient absorption. Therefore, while nano-sulfur shows potential for soil application to improve plant nutrition, its effectiveness via foliar application warrants further investigation to optimize its delivery and utilization in agricultural practices.

Based on the increasing possibility of global soil salinization due to climate change, more studies on soil salinity and its mitigation are needed in particular under nano-farming system and modern agriculture. Agrican mahogany proved from the current study its promising candidate to be successfully cultivated under saline conditions with more opening questions can be answered in the upcoming studies.

Conclusions

African mahogany is important tree that has many benefits for the human and animal as well. It can be concluded that the combined application of both nano-NPK fertilizer and nano-sulfur at 2 ml L⁻¹ every 15 days interval through the growth season positively gained the best morphological, biochemical, and physiological parameters. These parameters included mainly the content of nutrients in leave, the content of chlorophyll a and b, carotenoids, catalase, peroxidase activity. The suggested mechanism of cultivated seedlings to be tolerant to soil salinity may back to promoting the defense system. This system was supported by enzymatic antioxidants, and bioavailability of nutrients leading to be tolerant the oxidative and salt stress. More studies are needed to answer more open questions regarding this important ornamental crop.

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