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Growth analysis, leaf gas exchange and biochemical response of *Elaeis guineensis* to irrigation regimes and different sources of potassium fertilisers

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The role of potassium in mitigating drought stress is well-established. However, studies on different sources of fertiliser to alleviate this abiotic stress in oil palm seedlings are limited. Three types of potassium fertiliser (KCl: potassium chloride, K₂SO₄: potassium sulphate, and KNO₃: potassium nitrate) under three levels of water stress (100% ER; evapotranspiration replacement, 75% ER, and 25% ER) were exposed to 3-month-old *Elaeis guineensis* (oil palm) seedlings to investigate the influence of potassium source in minimising water stress effects in this crop. All treatments were arranged in Randomized Complete Block Design (RCBD) with three replications. It was found that as the level of water reduced, the Relative Growth Rate (RGR), Leaf Area Ratio (LAR), Leaf Weight Ratio (LWR), and Specific leaf area (SLA) of oil palm seedlings also reduced, but the value of Root: Shoot (R:S) improved. The result revealed that K₂SO₄ increased the net photosynthetic rate, transpiration rate, stomatal conductance, and Fv/Fm, as well as reduced the leaf temperature compared to KCl and KNO₃. There was an interaction between water stress and the source of K on respiration rate and electrolyte leakage where K₂SO₄ reduced both activities. K₂SO₄ too reduced the proline, total phenolics, and soluble sugar by 25%, 28% and 37% respectively compared to KCL. Thus, the study has shown that water stress deteriorated the growth of oil palm seedlings while the application of K₂SO₄ fertiliser could minimise the effects of water stress on the leaf gas exchange and the biochemical properties of this crop.

Keywords: Oil palm seedlings, drought, potassium fertiliser

INTRODUCTION

Elaeis guineensis is a C₃ plant that requires an abundance of rainfall, that is, at least 2000 mm year⁻¹ for optimum growth (Carr, 2011; Apichatmeta et al. 2017). However, water limitation is a major constraint for the growth and production

of this crop (Oettli et al. 2018). For the past 20 years, the amount of rainfall in Malaysia has experienced irregularities and this phenomenon is alarming to this high economic value industry which covers almost 6 million hectares of land (Tang, 2019; Malaysian Palm Oil Board [MPOB], 2020).

Climate and weather have a great influence on the oil palm seedlings under drought depending on its duration and intensities (Kamil and Omar, 2016). Lots of researchers have conducted plant growth analysis to evaluate the impact of water stress on the plant's physiology such as in roselle, capsicum, beans and goldenrod (Mehar-un-Nisa et al. 2016; Okunlola et al. 2017; Sofi et al. 2018; Du et al. 2019). The analysis of fundamental growth traits, such as relative growth rate (RGR), leaf area ratio (LAR), leaf weight ratio (LWR), specific leaf area (SLA), root to shoot ratio (R:S) and plant growth rate can help to understand the variation in growth and development of this crop. RGR is defined as rate of increase in dry matter per unit dry matter where it expresses growth in terms of a rate of increase in size per unit of size (Hunt, 2017) while LAR indicates the leafiness of plant (Price and Munns, 1999). LWR is ratio of leaf area to the plant dry weight, and it measures biomass allocation to the leaves (Price and Munns, 1999). SLA measures the ratio of leaf area to leaf weight. Pandey et al. (2017) stated that higher SLA signifies less thick and or less density of leaf while the determination of R:S shows index of the growth balance between root and shoot components of the plant (Hunt, 2017).

Apart from water, oil palm also needs great quantities of macronutrients, including potassium (K) fertiliser for its higher production (Sudradjat et al. 2018). It is reported that about 30-40% cost of production comes from fertiliser for this potassium devourer species (Goh and Teo, 2011; Lee, 2019). K fertiliser plays important roles in various mechanism, including improves photosynthesis, reduces respiration and oxidative stress, activates enzyme for plant growth, and most crucially role, under water scarcity, it can control the closing and opening of stomata (Kumar et al. 2016; Bahrami-Rad and Hajiboland, 2017; Tiemann et al. 2018).

There are a few available K fertilisers in Malaysia, such as potassium chloride (KCl), potassium sulphate (K_2SO_4), and potassium nitrate (KNO_3). These fertilisers are sold at different prices and the most commonly used source of K in the oil palm industry is KCl as it is the cheapest fertiliser available (Hussain et al. 2015). The price of K_2SO_4 is a little bit higher than KCl, yet the former is crucial when the soil lacks sulphate anion (Gunadi, 2009). There is little agreement between researchers on the effectiveness of different sources of potassium fertiliser. Some studies agree that KCl may give high salinity and chloride toxicity to the crop (Teixeira et al. 2011; Kumar and Kumar, 2008; Mancuso et al. 2014). Concurrently, Hussain et al.

(2015) do not recommend KCl fertiliser under limited water resources as it contains a high salt index. Meanwhile, K_2SO_4 has been shown to improve the quality of some crops, such as vegetable and pineapple colour (Samuels and Díaz, 1960; Hanaka et al. 2016), banana bunch size, and total soluble sugar (Kumar and Kumar, 2008). However, in some field experiments, no significant differences between KCl and K_2SO_4 were found (Ranjha et al. 2002). In another experiment, KCl was superior to K_2SO_4 (Surendran, 2005). On the other hand, Borowski and Michalek (2009) reported that KNO_3 had the highest content of potassium in spinach leaves compared to KCl and K_2SO_4 . Thus, in this study, K_2SO_4 and KNO_3 were suggested as an alternative to the chloride-free K source.

It is believed that different types of potassium could give different effects on the growth, physiology, and biochemical properties of oil palm seedlings. They might be affected not only by K, but also by their companion ions, such as Cl^- , SO_4^{2-} , and NO_3^- . The constant increase in the price of fertiliser demands oil palm grower to use it with greater efficiency to maintain the productivity under limited resources of water. The role of potassium fertiliser in mitigating water stress has been tested in many field experiments worldwide, but relatively few studies have investigated which type of K fertiliser is the best in alleviating the drought stress on *E. guineensis*. Thus, this experiment was intended to analyse the growth of oil palm seedlings under water scarcity and investigate which type of potassium fertiliser is the best for alleviating drought stress. Understanding the growth, leaf gas exchange, and biochemical changes of oil palm seedlings under different levels of water treatment and types of K fertiliser is important for efficient management of agronomical inputs (water and fertiliser), particularly in this changing environment. It was hypothesised that different types of potassium fertilisers could alleviate the negative impact of water stress. The level of water and fertiliser doses selected for the experiment were based on earlier studies where it described about effects of different levels of water stress on the growth, physiology and biochemical properties of oil palm seedlings (Najihah et al. 2019) and activity of the oil palm seedlings exposed to a different rate of potassium fertiliser under water stress condition (Najihah et al. 2020) respectively.

MATERIALS AND METHODS

Experimental design and plant materials

In the present study, a field trial was conducted at the Department of Biology, Faculty of Science in Universiti Putra Malaysia from August to December 2019 to evaluate the efficacy of potassium fertilisers under drought stress. Seedlings of 3-month-old Tenera (Dura X Pisifera) were acclimatised and grown in growth chambers for one month. The microclimatic condition under the glasshouse is measured by using CAREL® relative humidity/temperature sensor model DPDC11000 and the data is presented in Table 1. Prior to the acclimatisation, the seedlings were transplanted into polybags (10 x 12 cm with 16 cm diameter) containing Munchong series topsoil. A total of 81 polybags were arranged in randomised complete block design (RCBD) with three times replication. The treatments consisted of three levels of water treatment, which were 100% ER (evapotranspiration replacement; well-watered), 75% ER (moderate water stress), and 25% ER (severe water stress) with three sources of K treatments, i.e., potassium chloride (KCl), potassium sulphate (K_2SO_4), and potassium nitrate (KNO_3). Based on the MPOB (2013) guideline, the fertiliser should be applied once in two weeks with different ratio. The current study took around 3 months, thus five times applications were required. The amount of potassium was calculated based on the ratio and double of the amount suggested (Table 2). Plants were also fertilised with the source of N (ammonia) and P (CIRP) using standard rates. The ER treatment was applied following the method adopted by Klapwijk and Lint (1974). The water was calibrated once a week and plants were harvested in Week 3, Week 6, and Week 9 after the treatments.

Table 1: The microclimate condition under the glass house for oil palm seedlings during the experiment

Microclimate parameters	Quantification
Relative humidity	56.17–68.21%
Light intensity	210–1780 $\mu\text{mol m}^{-2} \text{s}^{-1}$
Day temperature	30–34 °C
Night temperature	17–24 °C
Ambient CO_2	388.70 $\mu\text{mol mol}^{-1}$

Table 2: The amount of K fertilisers used in the study.

Ratio (N:P:K:Mg) ^a	Amount (control) ^a	Kg/ha (KCl/ K_2SO_4 / KNO_3) ^b
15:15:6:4	15 g	565 (KCl), 678 (K_2SO_4) and 737 (KNO_3)
12:12:17:2	15 g	1600 (KCl), 1921 (K_2SO_4) and 2088 (KNO_3)
15:15:6:4	15 g	565 (KCl), 678 (K_2SO_4) and 737 (KNO_3)
12:12:17:2	30 g	3202 (KCl), 3842 (K_2SO_4) and 4176 (KNO_3)
12:12:17:2	30 g	3202 (KCl), 3842 (K_2SO_4) and 4176 (KNO_3)

^aBased on MPOB (2013) recommendation

^bDouble amount of different potassium source calculated based on the ratio and amount suggested

Vegetative measurements

Plant total dry weight, leaf area, plant height increment, root dry weight, stem dry weight, and shoot dry weight were measured according to MPOB (2017). The leaf area of oil palm seedlings was measured using a Leaf Area Meter (Li-3100 area meter, LICOR). Plant height was measured as the distance from soil surface to upper end of the longest leave. Dry weights were obtained by drying the samples using an oven at 75°C for 48 h until the weights were constant. Samples were then weighed using digital balances. Before harvesting, SPAD meter (502, Minolta Inc, USA) was used to measure the leaf chlorophyll content. The formula of RGR, LAR, LWR, SLA, and R:S could be deduced from the following equation based on Pandey et al. (2017).

$$RGR = (\ln W_2 - \ln W_1) / (t_2 - t_1) \quad (1)$$

Where, W_1 = Initial dry weight of plant at time t_1

W_2 = Final dry weight of plant at time t_2

$$LWR = \text{Leaf dry weight} / \text{Total dry weight} \quad (2)$$

$$SLA = \text{Leaf area} / \text{Leaf weight} \quad (3)$$

$$LAR = SLA \times LWR \quad (4)$$

$$R:S = \text{Root dry weight/shoot dry weight} \quad (5)$$

Electrolyte leakage

Electrolyte leakage was measured by following the method of Cha-um et al. (2013). The leaf samples were cut and kept in closed vials containing 10 mL deionised water. The samples were then maintained for 15 min at room temperature before initial conductivity (EC_0) was measured by using an electrical conductivity meter. After that, the same samples were incubated in the water bath (100°C) for 15 min and cooled down at 25°C. The following readings of electrical conductivity were measured (EC_1). The results were expressed as $REL = (EC_0 / EC_1) \times 100$.

Chlorophyll fluorescence

The Fv/Fm (quantum efficiency of PS II), Fv/Fo (maximum primary yield of photochemistry of PS II), Fo (initial fluorescence) and Pi (performance index) were estimated according to Ibrahim et al. (2012). The data were measured around 8.00 am to 10.00 am on fully expanded young leaves number two using a portable chlorophyll fluorescence metre (Handy PEA, Hansatech Instruments Ltd, Kings Lynn, UK). Leaves were darkened for 15 min by attaching dark-acclimation clips to the central region of the leaf surface before measurements, then the chlorophyll fluorescence was obtained from this procedure.

Leaf gas exchange

The measurement was obtained by using LI-6400XT Portable Photosynthesis System closed infrared gas analyser (Li-COR Inc; Nebraska; USA). The data of net photosynthesis (A), stomatal conductance (gs), transpiration rate (E), plant respiration (Rd), and temperature of oil palm seedlings were taken between 9.00 am to 11.00 am on fully expanded young leaves number two. The water use efficiency (WUE) was calculated by dividing the net photosynthesis (A) with corresponding transpiration rate (E). In this experiment on *E. guineensis*, the instrument was set at 400 $\mu\text{mol mol}^{-1} \text{CO}_2$, cuvette temperature of 30°C, air relative humidity of 60% with the airflow rate fixed at 0.000372 mol/s, and modified cuvette at 800 $\mu\text{mol m}^{-2} \text{s}^{-1}$ photosynthetically photon flux density (PPFD) (Zainuren et al. 2019).

Biochemical properties

The method used for proline determination was that of Bates et al. (1973). The oil palm leaves used for gas exchange and chlorophyll fluorescence measurements were removed and straightaway frozen in liquid nitrogen after collection to be grounded with mortar. The

homogenate powder was mixed with 1 mL aqueous sulfosalicylic acid (3% w/v) and filtered through filter paper (Whatman #1, England). Another 1 mL of glacial acetic acid and ninhydrin reagent (1.25 mg ninhydrin in 30 mL glacial acetic acid and 20 mL 6 M H_3PO_4) were added to this solution, then incubated for 1 h at 95°C before being placed in an ice bath for reaction termination. The reaction was mixed vigorously with 2 mL toluene, warmed to 25°C, and the chromophore was measured with a B UV-visible spectrophotometer (model DR/4000, HACH, Loveland, Colorado, USA) at 520 nm using L-proline as a standard.

For the determination of phenolics, 0.1 g ground dried tissue samples were extracted with 10 mL ethanol (80%) before being filtered with Whatman™ No.1. Then, 200 μL of sample extract was mixed with 1.5 mL of Folin-Ciocalteu reagent (diluted 10-fold), and stood at 22°C for 5 min. The mixture was then added with 1.5 mL, 60 g L^{-1} NaNO_3 solution. The absorbance was measured after two hours at 725 nm (Ibrahim et al. 2012).

The soluble sugar was identified according to Ibrahim et al. (2011). 0.5 g of samples were mixed with 10 mL of distilled water, then vortexed, and incubated in a 15 mL conical tube for ten min. For anthrone reagent preparation, 0.1 g of anthrone was dissolved in 50 mL sulphuric acid (95%). To prepare a standard curve for the quantification of sucrose in the sample, sucrose was used as a standard stock solution. Then, the dried sample was mixed with distilled water before being centrifuged at 3,400 rpm (10 min) and filtered. 8 mL of anthrone reagent was added with 4 mL aliquots and placed in a 100 °C water bath for five min. The sample was then measured with absorbance 620 nm using a UV160U spectrophotometer (Shimadzu, Japan) and results were expressed as mg sucrose/g dry.

Statistical analysis

All data were analysed using SPSS statistical software (ver. 25.0 SPSS, Chicago, USA) and were subjected to two-way analysis of variance (ANOVA). Duncan's multiple range test (DMRT) was used to compare the mean with the least significant level of 5%.

RESULTS

Plant growth analysis

Exposure to water stress subsequently affected the RGR of *E. guineensis* (Figure 1; $P \leq 0.05$). There was no interaction effect between water stress and potassium fertiliser on the plant growth analysis of oil palm seedlings. The RGR of

the seedlings under 100% ER (control) and 75% ER (mild water stress) seemed to be significantly higher than 25% ER (severe water stress) in all the weeks measured.

Under 25% ER, water stress reduced the RGR by 20%, 37%, and 76% in Week 3, 6, and 9 respectively compared to control treatment. However, there was no significant difference between the control and 75% ER.

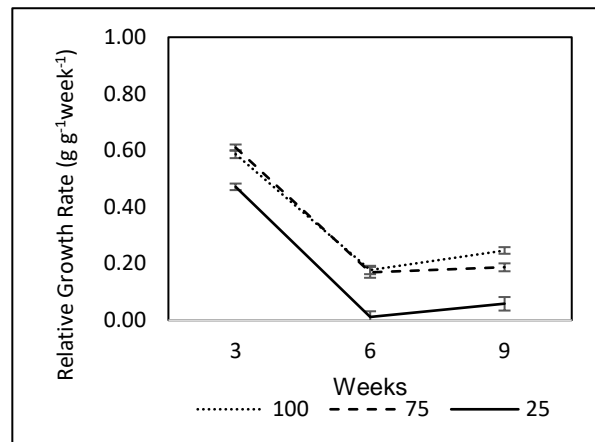


Figure 1. The effects of different water stress treatments on the RGR oil palm seedlings for 9 weeks. Bars indicate the standard error of mean (SEM) (n = 3).

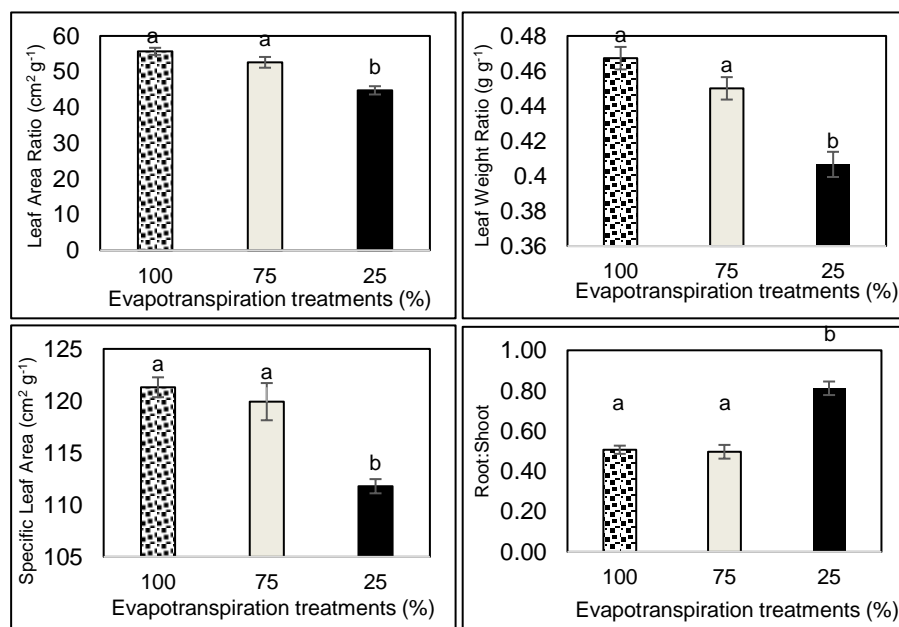


Figure 2. The effects of different water stress treatments on the LAR, LWR, R:S and SLA of oil palm seedlings on week 9. Bars indicate the standard error of mean (SEM) (n = 3). Different letters indicate significant difference at 5% level.

Figure 2 shows the effects of water treatment level on LAR, LWR, SLA, and R:S of oil palm seedlings. The growth analysis of *E. guineensis* seedlings was significantly influenced by the water applied ($P \leq 0.05$). As shown in Figure 2, the highest LAR was obtained by 100% ER ($55.67 \text{ cm}^2 \text{ g}^{-1}$), followed by 75% ER ($52.61 \text{ cm}^2 \text{ g}^{-1}$) and 25% ER ($44.77 \text{ cm}^2 \text{ g}^{-1}$). The LWR and SLA were also showing the same trend where, as the level of water decreased (100% ER > 75% ER > 25% ER), less LWR and SLA were produced. Severe water stresses dramatically reduced LWR and SLA about 0.06 g g^{-1} and $9.52 \text{ cm}^2 \text{ g}^{-1}$ respectively compared to control. However, oil palm seedlings receiving the least water had the highest R:S which was almost twice the control.

Water stress also affected the plant growth rate (plant height increment) as shown in Figure 3. Plant height is associated positively with week progressed. As the levels of water decreased from 100% > 75% > 25 ER, the plant height increment was also decreased, yet there were no significant differences between 75% ER and 100% ER.

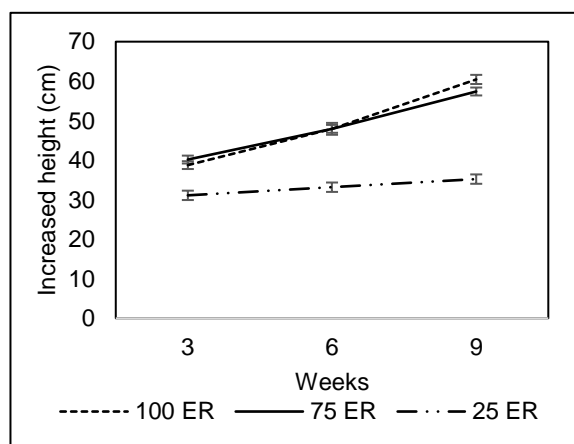


Figure 3. The effects of different water stress treatments on the plant growth rates (plant height increment) for 9 weeks. Bars indicate the standard error of mean (SEM) (n = 3).

Apparently, different sources of potassium fertiliser significantly influenced the leaf gas exchange of oil palm seedlings (Figure 4; $P \leq 0.05$). There was no interaction effect between water stress and potassium fertiliser on the leaf gas exchange of oil palm seedlings. Plants under K_2SO_4 had the highest ($7.79 \mu\text{mol m}^{-2}\text{s}^{-1}$) net photosynthesis (A) compared to KCl ($5.97 \mu\text{mol m}^{-2}\text{s}^{-1}$) and KNO_3 ($5.05 \mu\text{mol m}^{-2}\text{s}^{-1}$) while K_2SO_4 (0.51

$\text{mmol m}^{-2}\text{s}^{-1}$) also increased the stomata conductance of oil palm seedlings more than two-fold compared to KCl ($0.22 \text{ mmol m}^{-2}\text{s}^{-1}$) with no significant difference between KCl and KNO_3 ($0.22 \text{ mmol m}^{-2}\text{s}^{-1}$). The transpiration of oil palm seedlings also seemed to be higher in K_2SO_4 ($2.27 \text{ mol m}^{-2}\text{s}^{-1}$) followed by KCl ($1.61 \text{ mol m}^{-2}\text{s}^{-1}$) and KNO_3 ($1.42 \text{ mol m}^{-2}\text{s}^{-1}$). However, the leaf temperature dropped approximately about 2°C when plants were applied with K_2SO_4 (25.13°C) compared to KCl (27.27°C) and KNO_3 (27.82°C).

Figure 5 and 6 illustrate the Fv/Fm value under different levels of water stress treatments and different types of K correspondingly. It was found that the Fv/Fm value was statistically lower as the level of water reduced ($P \leq 0.05$). Only 100% ER had a ratio above 7, while as for 75% ER and 25% ER, the ratios were around 6. Instead, the Fv/Fm value was significantly higher with the application of K_2SO_4 compared to others. It increased the Fv/Fm to 0.77 while it was only almost 6 for KCl and KNO_3 .

The respiration of *E. guineensis* was affected by the level of water and potassium fertilisation (Figure 7; $P \leq 0.05$). Of all the combined treatments, it was found that plants which were fertilised with K_2SO_4 had the lowest respiration regardless of the water treatment (under $0.05 \mu\text{mol m}^{-2}\text{s}^{-1}$). The same trend was experienced with plants under KCl, but there was no significant difference with 75% ER (KNO_3). The highest respiration recorded under KNO_3 was the plants under severe water stress which is $0.27 \mu\text{mol m}^{-2}\text{s}^{-1}$.

Figure 8 shows the electrolyte leakage of oil palm seedlings as influenced by the level of water and the source of K fertiliser. Electrolyte leakage was significantly increased as the level of water decreased ($P \leq 0.05$). Water stress caused an increase of about four times higher under 25% ER (14.7%) compared to 100% ER applied with the same fertiliser (K_2SO_4) (46.73%). Overall, there was no significant difference between fertilisers applied under the same level of water except for under 75% ER.

Biochemical properties

Apart from leaf gas exchange, biochemical properties of oil palm seedlings were also affected by a different type of K applied (Figure 9; $P \leq 0.05$). There was no interaction effect between water stress and potassium fertiliser on the biochemical properties of oil palm seedlings. Proline was reduced by 25% under K_2SO_4 (1.33 mg g^{-1} fresh weight) compared to KCl (1.78 mg g^{-1} fresh weight).

Meanwhile there was a slight increase in the proline content under KNO_3 (1.83 mg g^{-1} fresh weight) from KCl. As for soluble sugar, the application of K_2SO_4 significantly reduced this chemical of about $8.38 \text{ mg sucrose g}^{-1}$ dry from KCl. The phenolics of oil palm seedlings were also decreased by about 28% compared to control while

KNO_3 produced the highest phenolics in all fertilisers. Overall, it seems that K_2SO_4 reduced the biochemical properties of oil palm seedlings while KNO_3 increased these parameters compared to KCl.

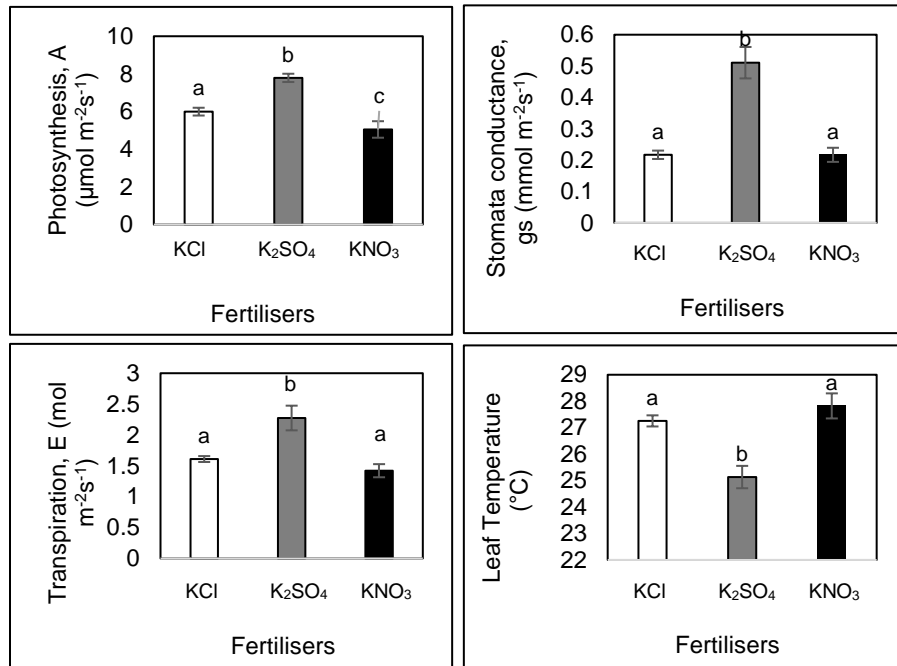


Figure 4. The effects of different types of K on leaf gas exchange of oil palm seedlings on week 9. Bars indicate the standard error of mean (SEM) ($n = 3$). Different letters indicate significant difference at 5% level.

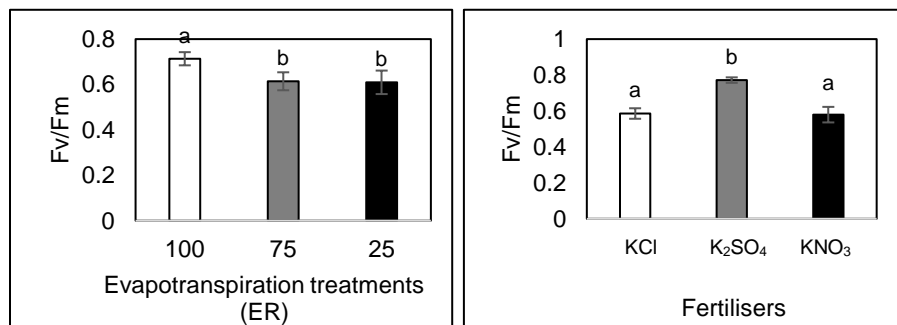


Figure 5. The effects of different water stress treatments on F_v/F_m of oil palm seedlings on week 9. Bars indicate the standard error of mean (SEM) ($n = 3$). Different letters indicate significant difference at 5% level.

Figure 6. The effects of different types of K on F_v/F_m of oil palm seedlings on week 9. Bars indicate the standard error of mean (SEM) ($n = 3$). Different letters indicate significant difference at 5% level.

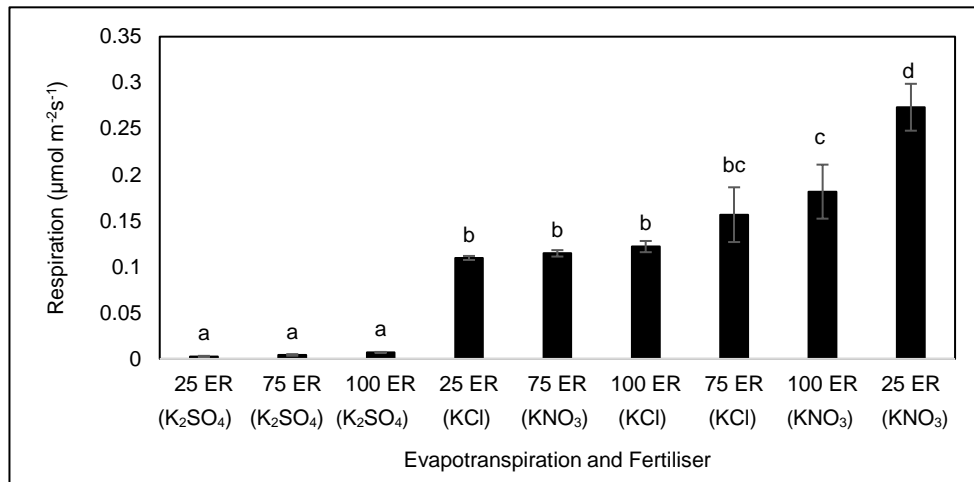


Figure 7. The interaction effects of water stress treatments and different types of K on respiration of oil palm seedlings on week 9. Bars indicate the standard error of mean (SEM) (n = 3). Different letters indicate significant difference at 5% level.

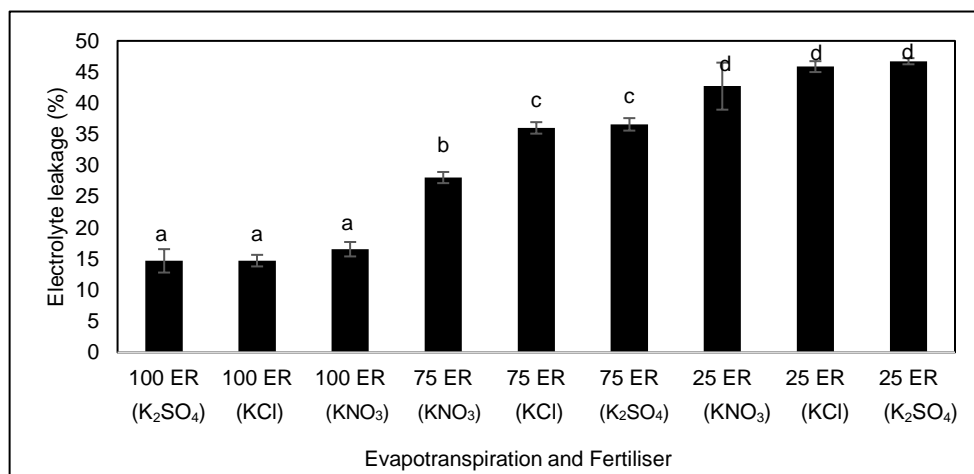


Figure 8. The interaction effects of water stress treatments and different types of K on electrolyte leakage of oil palm seedlings on week 9. Bars indicate the standard error of mean (SEM) (n = 3). Different letters indicate significant difference at 5% level.

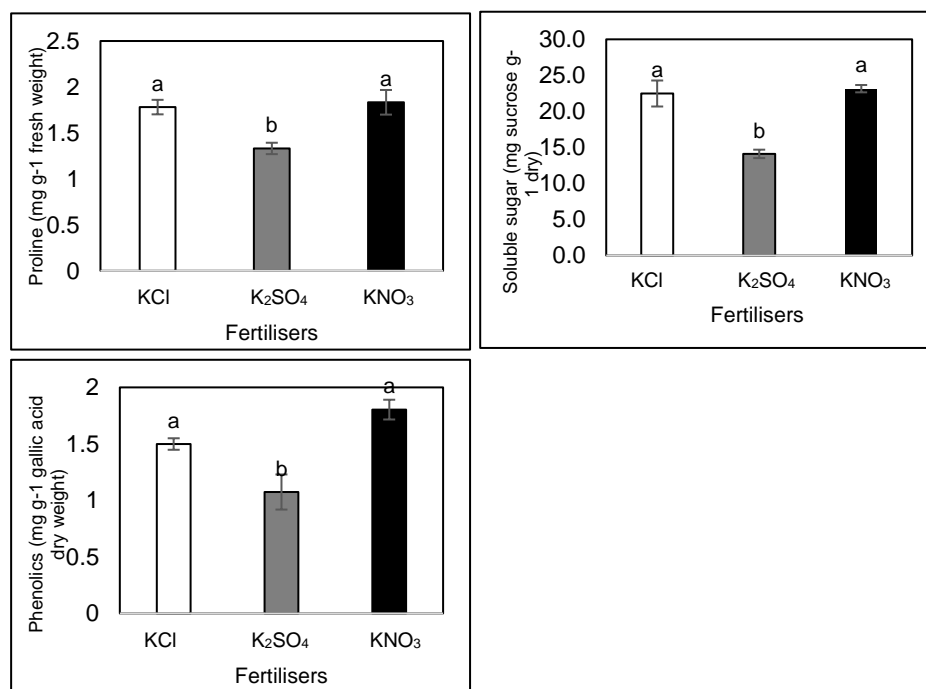


Figure 9. The effect of different types of K on biochemical properties of oil palm seedlings on week 9. Bars indicate the standard error of mean (SEM) (n = 3). Different letters indicate significant difference at 5% level.

Table 3: Pearson correlation between measured parameters in week 9 of the experiment.

Parameters	1	2	3	4	5	6	7	8	9	10	11
RGR	1										
LAR	0.681**	1									
LWR	0.615**	0.783**	1								
R_S	-0.743**	0.710**	-0.823**	1							
SLA	0.474*	0.793**	0.244	-0.313	1						
TDW	0.866**	0.717**	0.760**	-0.858**	0.385*	1					
A	-0.032	0.222	0.266	-0.255	0.088	0.016	1				
gs	0.176	0.306	0.448*	-0.409*	0.05	0.192	0.746**	1			
E	0.089	0.287	0.31	-0.327	0.154	0.167	0.682**	0.640**	1		
Respiration	-0.11	-0.267	-0.357	0.318	-0.073	-0.154	-0.593**	-0.679**	-0.530**	1	
Temperature	-0.072	-0.243	-0.257	0.341	-0.142	-0.062	-0.687**	-0.858**	-0.680**	0.613**	1

Note: RGR = relative growth rate; LAR= leaf area ratio; LWR= leaf weight ratio; R_S = root to shoot ratio; SLA= specific leaf area; TDW= total dry weight; A = net photosynthesis; gs = stomatal conductance; E= transpiration rate. * and ** significant at $P \leq 0.05$ and $P \leq 0.01$ respectively.

DISCUSSIONS

Plant growth analysis

The relationship between RGR and water stress has been studied in various field experiments, including in *Eucalyptus* seedlings (Maseda and Fernández, 2016), Mediterranean tree species (Forner et al. 2018), *Juglans* rootstocks (Liu et al. 2019), and *Capsicum* species (Okunlola et al. 2017). The reduction of RGR under water stress was particularly caused by the decreased in SLA (Galmés et al. 2005) where when the SLA reduced, the growth of oil palm seedlings was also affected as translated by the positive relationship between RGR and SLA ($R_2 = 0.474$; Table 3; $P \leq 0.05$).

The reduction of LAR under water deficit had also been reported by Bargali and Tewari (2004) in their study with two Chihuahuan desert grass species. In this study, they confirmed that under water deficit, the LAR of both species was significantly reduced due to the reduction in SLA and LWR. This was because, mathematically, LAR is a product of SLA and LWR. However, another theory was also proposed by Du et al. (2019) and Navarro Rocha et al. (2017) where the decreased of LAR under constant drought from Week 3 to Week 6 was most probably due to the protective mechanism of plant to reduce its transpiration rate as LAR is an indicator of plant relative leafiness. In this experiment, the correlation analysis in Table 3 showed that SLA ($R_2 = 0.793$; $P \leq 0.01$) and LWR ($R_2 = 0.783$; $P \leq 0.01$) were strongly correlated with LAR. However, the positive correlation between gs and LAR was not significant in this study.

On the other hand, SLA and LWR also decreased as the level of water decreased. SLA indicates the leaf thickness, while LWR is a partitioning of plant biomass to leaf component (Spitters and Kramer, 1986). According to Maseda and Fernández (2016) in their study on gum trees, drought tends to reduce the SLA of plants. Another study with roselle plants also reported the same result (Mehar-un-Nisa et al. 2016). Under water stress, plants reduce their SLA by increasing the thickness of leaves, thus resulting in the thicker cell wall that tighter the connection to reduce water loss by evaporation and increase water use efficiency (WUE) (Zhou et al. 2020). As SLA decreased, total dry weight showed a reduction that displayed a significant positive correlation trend ($R_2 = 0.385$; $P \leq 0.05$) which indicates that a smaller leaf area reduces the plant total dry weight.

The reduction of LWR is believed to be due to plants allocating proportionally more biomass to

roots and less to leaves under water scarcity (Widuri et al. 2018) as LWR is expressed as dry weight of leaves to whole plant dry weight. The same discovery was also obtained by a few plants under water stress (Erice et al. 2011; Fernández et al. 2002). This would be explained by higher R:S in 25% ER compared to 100% ER and 75% ER. R:S is usually used to measure the relative biomass distribution between roots and shoots and as an important parameter in water stress experiments (Xu et al. 2015). Plants tend to reduce its shoot growth and invest more biomass to root under severe drought to decrease the water lost by transpiration in leaves and increase the water absorption by root (Brunner et al. 2015; Erice et al. 2011). High R:S in plants under water stress was also being discussed in several field experiments such as in soybeans and common beans (Du et al. 2020; Sofi et al. 2018). The findings by Sun et al. (2011) are also in line with this result in their research of oil palm seedlings under water stressed conditions. From the correlations in Table 3, it is shown that R:S ($R_2 = -0.823$; $P \leq 0.01$) had a highly significant negative correlation with LWR, indicating that the reduction of LWR under water stress was triggered by high R:S.

Plant height can be used as an indicator of crop performance. In the current study, the effect of height seems to be contributed by the water stress, whereby the lower the level of water, the shorter the height of oil palm seedlings. In the paper written by Sun et al. (2011) showed that growth rates of oil palm were affected by water and nutrient availability, but it appeared more affected by water stress. The decrement of stem elongation under water stress might be due to the reduction in cell expansion, which is in agreement with Litvin et al. (2016) in their experiment with tomato plants.

Leaf gas exchange

In Figure 4, it is clear that feeding of K_2SO_4 had the greatest effect on gas exchange components (A, gs, and E) of *E. guineensis* and this fertiliser also dropped its leaf temperature. The mechanism for this effect is not known and could be from the efficacy of sulphur present in K_2SO_4 which produces high chlorophyll content (Ashraf et al. 2013) or accumulation of K^+ ions in the leaves (Borowski and Michalek, 2009). However, the same findings were reported by Kumar and Kumar (2008) in their research with Banana c.v. robusta. In the study, the plants treated with K_2SO_4 had the highest photosynthesis compared to KCl and this was believed to be due to the involvement of sulphate ions in the chlorophyll synthesis, thus

increasing the content of chlorophyll in the leaves. In the current experiment, it was proven by the analysis of variance where even though there was no significant difference of fertiliser sources on the chlorophyll content, yet K_2SO_4 (61.32) gave the highest number of chlorophylls compared to KCl (60.44) and KNO_3 (57.11).

Stomata conductance (gs) is the degree of stomatal opening in the leaves where it regulates carbon dioxide entering and plant transpiration rate (Hussain et al. 2015). In the present study, high gs was observed in plants treated with K_2SO_4 compared to KCl and KNO_3 . This might be explained by a higher concentration of K^+ ions in the guard cell, as the function of these ions is to help in opening and closing of stomata (Hasanuzzaman et al. 2018), as well as contributing to the rate of phloem transport (Pervez et al. 2004). An experiment conducted by Borowski and Michalek (2009) showed that the increase of gs in spinach leaves is the result of the high concentration of K^+ ions in its stomata, thus high transpiration (E) and photosynthesis rate (A) followed. There is a highly significant correlation between A, gs, and E as shown in Table 3.

On the other hand, as E is lower, the plant tends to rise its leaf temperature. Several studies have related the rising in leaf temperature with environmental stress. These include oil palm, maize, and peanut (Aucique-Perez et al. 2020; Liu et al. 2011; Isoda, 2010). According to Bahrami-Rad and Hajiboland (2017) and Liu et al. (2011), the increase of leaf temperature under stress is due to low transpiration caused by stomatal closure. Table 3 showed a negative correlation between leaf temperature ($R_2 = -0.680$; $P \leq 0.01$) with E signifying low E contributed to high leaf temperature. The rise of this temperature when plants were applied with KCl and KNO_3 could also be associated with a low value of chlorophyll fluorescence. High temperature in leaves comes from the dissipation of excess excitation energy to protect their photosynthetic apparatus from photodamaging effects, thus it explained that plants under K_2SO_4 had lower leaf temperature as they had the highest photosynthesis rate and high Fv/Fm value, indicating less heat had been dissipated (Aucique-Perez et al. 2020; Isoda, 2010).

Chlorophyll fluorescence serves as an indicator of the photosynthetic performance of plants and the value between 0.79 to 0.83 is considered as healthy (Khandakher et al, 2017; Khandakher et al 2018; Naidoo and Naidoo, 2018). However, in this experiment, none of the young

palms had their ratio above 0.8, which indicates that there was a photoinhibition in the photosystem II (Liu et al. 2019; Maxwell and Johnson, 2000) yet, the Fv/Fm of well-irrigated seedlings was still higher than under mild and severe drought. On the other hand, only plants treated with K_2SO_4 had Fv/Fm ratio close to 0.8. This result reflected the assertion by Hanaka et al. (2016) of their experiment with *Eruca sativa* MILL., an edible vegetable in Poland. In their study, vegetables treated under K_2SO_4 fertiliser combined with high dose of nitrate tended to have higher Fv/Fm compared to the combination of nitrate and KCl. It is believed that this was due to a more efficient photosynthetic apparatus in this treatment compared to other treatments.

Plants under stress have higher leaf respiration rate compared to normal plants. For instance, oil palm seedlings under water deficit have higher leaf respiration rate ($0.125 \mu\text{mol m}^{-2} \text{s}^{-1}$) compared to plants under field capacity ($0.067 \mu\text{mol m}^{-2} \text{s}^{-1}$) (Rivera-Mendes et al. 2016). One of the essential roles of potassium fertiliser is to reduce respiration (Hasanuzzaman et al. 2018; Rawat et al. 2016). Thus, the inadequate amount of K will increase plant respiration rate (Kafkafi et al. 2001). An experiment on chickpea showed that the respiration of this crop decreased with the increased potassium level (Kataria and Singh, 2013), which proved the function of K in reducing the respiration rate. Hamoda et al. (2016) agreed that a high respiration rate contributes to slower plant growth and development, and this process is increased when photosynthesis decreases. As shown in Table 3, there is a highly negative relationship between respiration and photosynthesis rate in *E. guineensis* ($R_2 = -0.593$; $P \leq 0.01$).

As mentioned in the paper written by Jungklang et al. (2017), plants under water stress experience electrolyte leakage which indicates the membrane injury. This phenomenon is associated with K^+ ions efflux from plant cells and usually happens when the plant is under stress (Demidchik et al. 2014). Even though there was no significant difference of fertiliser effects under severe water stress in the current study, Douglas José Marques et al. (2014) reported that electrical conductivity treated with KCl is significantly higher than K_2SO_4 due to soil salinization in eggplant. In this experiment, the effect of water stress is greater than fertiliser treatment on *E. guineensis*.

Biochemical properties

Proline, total soluble sugar (TSS), and

phenolics content usually emerge under environmental stress, such as drought, salinity, and extreme temperature, which they are often used as plant stress indicators (Ju et al. 2018; Cha-um et al. 2013; Rosa et al. 2009). High level of these osmoprotectants indicate that plant is under stress as it is increased to protect the cell from damages (Bahrami-Rad and Hajiboland, 2017; Hisyam et al, 2017). In the current study, under a high level of potassium, only plants treated with K_2SO_4 had the lowest proline, TSS and phenolics level compared to other salts. This result reflected the experiment conducted by Marques et al. (2011) where when they increased the concentration of potassium, the eggplant tended to increase its proline level. Concurrently, the level of proline was always lower in the form of K_2SO_4 compared to KCl. They believed that this situation was due to the adaptation of plants towards salinity stress.

CONCLUSIONS

Overall, in the present research work, water stress consistently decreased the growth traits (RGR, LAR, LWR, and SLA) of oil palm seedlings and improved R:S. The use of K_2SO_4 appeared to be more favourable than KCl and KNO_3 as it increased net photosynthesis, transpiration rate, stomatal conductance, and Fv/Fm, as well as reduced the leaf temperature. There was an interaction between water stress and the source of K on respiration rate and electrolyte leakage where K_2SO_4 reduced both activities. K_2SO_4 also reduced the biochemical properties of oil palm seedlings, such as proline, total phenolics, and soluble sugar. However, it depends on the cost of the production as oil palm growers should calculate the cost of loss due to drought with

expected output for fertiliser investment. Further studies on other factors of climate change, such as high temperature under water stress should be taken into account.

CONFLICT OF INTEREST

The authors declared that present study was performed in absence of any conflict of interest.

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AUTHOR CONTRIBUTIONS

TSN performed practical work, data analysis and wrote the manuscript. MHI participate in designed the experiment, data analysis and reviewed the final manuscript while RN and PEMW helped supervise the project. All authors read and approved the final version.

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