

### Using Rice Husk Biochar For Ming Aralia (*Polyscias fruticosa* (L.) Harm) Production Under Saline Conditions

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#### ABSTRACT

Polyscias fruticosa (L.) Harms is a traditional medicinal plant widely used in the treatment of ischemia and inflammation in the Southeast Asia region. Salinity stress has a negative impact on plant growth and development. Therefore, the application of biochar could help alleviate the problem. This research aims to find the optimal growing medium components for Ming Aralia to produce active compounds under saline conditions. A pot experiment was conducted from June to December 2019 with 2-month-old plants based on complete randomized design (CRD). The treatments include 1) NBNS (non-biochar, non-saline), 2) NBMD (nonbiochar, moderate salinity), 3) NBSI (non-biochar, saline severity I), 4) NBSII (non-biochar, saline severity II), 5) 25NS (25% biochar, non-saline), 6) 25MD (25% biochar, moderate salinity), 7) 25SI (25% biochar, saline severity I), 8) 25SII (25% biochar, saline severity II), 9) 50NS (50% biochar, non-saline), 10) 50MD (50% biochar, moderate salinity), 11) 50 SI (50% biochar, saline severity I), 12) 50SII (50% biochar, saline severity II) with 8 replications. Results showed that increases in salinity from non-saline to saline severity II significantly reduced plant height, leaf area, leaf and root fresh weight, and dry weight. Antioxidant activity, as well as total phenolic and flavonoid compound content were found to be higher when rice husk biochar was used at 25% and 50% under moderate and severe saline conditions.

Keywords: Active compound; Biochar; Polyscias fruticosa; Salinity

#### 1. Introduction

Ming aralia (*Polyscias fruticosa*) is a perennial shrub belonging to the Araliaceae family [1]. It's used as both an ornamental and medicinal plant in tropical and subtropical regions such as China, Malaysia, and Vietnam. *P. fruticosa* leaf and root extracts contain a high content of active compounds: saponin, flavonoids, alkaloids, glycosides, and polyacetylenes [2-5].

The main production area is in Hai Hau district, Vietnam, a coastal region located in the Red River Delta. Ming aralia is supplied at around 450 tons/year to companies to produce dietary supplement products for neurology in Vietnam [6]. In this area, around 21-28% of the total area is considered saline soil, with the electrical conductivity (EC) ranging from 0.46 - 8.91 dS·m<sup>-1</sup> [7]. This value is higher than the classification threshold set for saline soil by the USDA (2008) (EC = 4 dS·m<sup>-1</sup>, equivalent to 40 mM of NaCl). As such, farmers have to cope with this soil salinity, which is also increasing every year due to climate change.

Soil salinity becomes a significant factor that limits P. fruticosa production in Hai Hau district. Plants growing under saline conditions may experience growth reduction during their growth and development stages. The initial phase of growth reduction comes from a high concentration of salt in the soil, leading to a reduction in the plant's capacity to uptake water, resulting in a water deficit. Inside the plant tissue, salt stress causes cellular dehydration, osmotic stress, and water removal from the cytoplasm, which results in a decrease in cytosolic and vacuolar volumes [8]. Later, the high amount of toxic salts inside the plant leaves may cause severe ion toxicity, resulting in an imbalance of nutrient uptake [9].

Accumulation of secondary metabolites often occurs when plants are subjected to stress, this phenomenon can be referred to as the plant antioxidant defense system. Salinity stress increases the production of reactive oxygen species (ROS) such as  $H_2O_2$  (hydrogen peroxide),  $O_2^-$  (superoxide),  ${}^1O_2$  (singlet oxygen), and •OH (hydroxyl radical) in plant tissue, which can damage cell membranes to the point that programmed cell death (PCD) is activated [10]. Salinity stress enhances the availability of secondary compounds; however, excess salinity may reduce their levels significantly. It is, therefore, quite worthwhile to find a cultivation method that can overcome this problem.

Biochar is the thermochemical combustion of organic materials produced under a low concentration of oxygen, at temperatures in the range of 300 - 1000° C. Recently, it has become a global trend in soil application to enhance soil fertility [11] and improve plant growth and development [12]. These strategies play an essential role in increasing soil nutrient capacity, soil cation exchange capacity, and plant water retention. Biochar is considered an important tool for the reclamation of salt-affected soil [13-16]. In fact, successful applications of biochar have been shown to reduce salinity stress on many plants including maize [17], legume crops [18], potato [19], and wheat [20]. A wide range of biomass feedstocks is available for biochar production such as wood, energy crops, agriculture waste residues, sewage sludge, anaerobic digested materials, municipal waste, etc. It can be produced through various types of processes, including pyrolysis, pyrolysis, slow fast and gasification. These processes differ from each other in their temperature, heating rate, biomass, and vapor residence times. Different processing types are expected to yield biochar with different physicochemical properties.

Rice husk is rice residue from rice production. A large quantity of rice husk is produced every year in the field. Nowadays, rice husk is considered to be a good candidate for biochar production in Asia, and as a new soil amending material, because it is available in large amounts, low cost, and is easy to collect [21]. Rice husk biochar has high nutrient content, including compounds such as nitrogen, potassium, phosphorus, silicon, and black carbon. Notably, black carbon could be a valuable source of organic matter which has great potential for amending soil [22]. Studies on medicinal plants indicate that the application of rice husk biochar not only promotes the growth of these plants but also significantly affects plant chemical compositions via an enhancement in plant nutritional status [23]. Therefore, this research aimed to evaluate the effect of using varying amounts of rice husk biochar as a growing component for Ming Aralia, under varying salinity conditions.

#### 2. Materials and Methods 2.1 Plant materials

This experiment was performed from June to December 2019. The stem cuttings of Ming aralia 'Elegan' dwarf type (dwarf P. fruticose 'Elegans') at one-month-old were purchased from the wholesale plant market, Chatuchak, Bangkok. Each pot contained 3 cuttings (15 cm height). The plants were maintained in a greenhouse at Kasetsart university, Bangkhen campus, Bangkok (13°51'13.5"N 100°34'09.0"E) for one month, and were then transplanted into a 10inch pot (5,661 cm<sup>3</sup>). Substrates were composed of different ratios of coco coir (100%, 75%, and 50%) and rice husk biochar (RHB) (0%, 25%, and 50%), mixed with 300 g of compost fertilizer, and then treated with 1 of 4 levels of saline solution (non-saline: NS, moderate: MD: 1.5 dS·m<sup>-1</sup>, severe I: SI: 4 dS·m<sup>-1</sup>, and severe II: SII: 8 dS·m<sup>-1</sup>). The experimental design was Complete Randomized Design (CRD) with 12 treatments, 8 replications, and one-pot per replication. Saline solutions were prepared by dissolving NaCl (purity 99.9%) into tap water and then adjusting EC values to 1.5  $dS \cdot m^{-1}$ , 4  $dS \cdot m^{-1}$  and 8  $dS \cdot m^{-1}$ . The EC value of tap water was 0.33 dS·m<sup>-1</sup>. The saline solutions were applied to the plants via watering two times a week until being harvested (total 52 times). EC and pH of substrate solutions were measured by the pour-through method, with quick and practical method on-site. Drainage solutions were collected and measured for EC and pH by TDS & EC meter hold and pH meter (pH-006 Pen-Type Automatic Correction pH-meter) every week. Foliage fertilizer (Pokon® 21-21-21) was applied weekly to the plants. The temperature and humidity inside the greenhouse were recorded by the data logger (WatchDog-No.9). The mean temperature was  $27.5\pm1.99^{\circ}$ C and the relative humidity was  $58.11\pm8.40\%$  during the experiment period.

### 2.2 Phytochemical analysis

### 2.2.1 Preparation of plant extraction and chemical reagents

Leaf and root samples were cleaned and oven-dried at 60°C to stabilize weights. Fresh (0.5 g) and dry (0.3 g) samples were ground by liquid nitrogen, blended and extracted with ethanol (1 ml and 1.2 ml, respectively) and then sonicated using HP Series 53 kHz Ultrasonic Cleaners (for 30 min at room temperature). The supernatant was collected after being centrifuged for 5 min at 8000 rpm and maintained at -20°C until further analysis.

All chemicals used for phytochemical analysis were of analytical grade and manufactured by Sigma-Aldrich, purchased from Singapore, including Standard Quercetin (Q4951-10G. > 95% (HPLC), CAS Number: 117-39-5); Gallic acid (G7384-100G, CAS Number: 149-91-7), DPPH (CAS number: 1898-66-4), and all other solvents used were of HPLC grade.

# 2.2.2 DPPH radical scavenging activity

The DPPH radical scavenging activity was done following the procedure with modifications, carried out in a 96-well microplate using a SPECTRO star Nano Microplate Reader. Then, 22  $\mu$ l of the extract was added to 200  $\mu$ l of 0.1 mM DPPH (Diphenyl-2-pierlyhydrazyl) radical solution

in ethanol and incubated for 30 min in dark conditions at room temperature [24]. The mixed solutions were measured at 517 nm and 95% aqueous ethanol was used as a control. The scavenging activity of DPPH radicals (%) was calculated using the equation:  $[(A_{blank}-A_{sample}) \div A_{blank}] \times 100\%$ .

### 2.2.3 Total phenolic content determination

The total phenolic content determination was done with the Folin-Ciocalteu assay with modifications [25]. The ethanol extract, 100 µl, was mixed with 2.0 ml of distilled water and 1.0 ml of 20% sodium carbonate. After 3 min, 1.0 ml of Folin-Ciocalteu reagent was added and incubated for 60 min in dark conditions at The solution room temperature. was measured at 725 nm by a T70 UV/VIS spectrophotometer. Gallic acid (0.25, 0.125, 0.0625, and 0.03125 g/l) was used as a reference standard calibration. The total phenolic content is expressed as mg<sub>GAE</sub>/g which was determined from the linear equation: y = 3.5155x + 0.1276 (R<sup>2</sup> = 0.9985).

# 2.2.4 Isolation of total Flavonoid by HPLC

Flavonoid content in the leaves was detected by HPLC-DAD Agilent series 1100 following the method of [26] with modifications. Chromatographic separations were achieved using a Zorbax Eclipse XDB C18 column ( $4.6 \times 250$  mm, 5 µm). A reverse phase HPLC assay was carried out using isocratic elution with a flow rate of 1 ml/min, a column temperature of 35°C, a mobile phase of acetonitrile, 2% v/v acetic acid (40%: 60% v/v) and a detection wavelength of 262 nm. The injection volume was 20 µL with a total run time of 10 minutes for each injection and a retention time of 4 minutes. The calibration curve was calculated with five concentrations (10, 3.33, 1.25, 0.625, and 0.3125 mg/l) of quercetin. The equation was calculated in the form of y = 58663x +

3.4291, where y and x were peak area and compound concentration, respectively, and  $R^2=0.9993$ .

#### 2.3 Data analysis

The data were subjected to one-way analysis of variance (ANOVA). A comparison between means was performed using Duncan's multiple range test.

#### 3. Results and Discussion

### **3.1** Change in EC and pH of substrate solution

EC values in Table 1 did not show significant values (4.09 - 6.55 dS·m<sup>-1</sup>) at the onset of the experiment whereas EC values at the end were significantly different (p < 0.01) and increased to the concentrations of salinesolution as shown in Table 1. The solution with the highest EC was SII, followed by SI, MD, and finally NS; this pattern was true for all substrate components (Fig. 1). EC values at the beginning showed values higher than the plant requirement, which resulted from the measurement method. Other studies have revealed that the pour-thru method provides EC values higher than the 1:2 method and displays a linear correlation [27]. According to the equation, EC values from the experiment ranged from 0.78 - 1.19 dS·m<sup>-1</sup> according to the 1:2 method, which is in the plant requirement range. Moreover, other reports have also found that the EC values considered as equivalent to other laboratorybased methods of EC measurement range from 1.4 - 1.8 dS·m<sup>-1</sup> using the 1:2 dilution method, and 3.6 - 5.0 dS·m<sup>-1</sup> using the saturated media extract (SME) methods [28]. It could be suggested that EC values from this experiment were in the range acceptable for plant growth and development. However, the high EC values at the beginning may have come from RHB and compost fertilizer, which had values at  $4.69 \pm 0.12$  dS·m<sup>-1</sup> and  $2.08 \pm 0.02$  dS·m<sup>-1</sup>, respectively (data not shown).

The substrate's pH values ranged from neutral (7.07) to slightly alkaline (7.94) at the

beginning. They increased gradually after that, in the range of very slightly alkaline (7.26) to slightly alkaline (8.04) (Table 1).

The increase in pH could have possibly resulted from the accumulation of

pH in tap water (pH~7.34). RHB treatments' pH values at the onset were higher than those of non-RHB (discussed in Table 1).

**Table 1.** EC and pH of substrate solutions measured by the pour-through method of Ming Aralia grown in different RHB components under various saline conditions (NS = non-saline, MD =  $1.5 \text{ dS} \cdot \text{m}^{-1}$ , SI =  $4 \text{ dS} \cdot \text{m}^{-1}$  and SII =  $8 \text{ dS} \cdot \text{m}^{-1}$ ).

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Codo	Treatment -		$EC (dS \cdot m^{-1})$			pH		
Code			onset	end	Δ	onset	end	$\Delta$
NBNS	Non- RHB	NS	5.59±1.08	1.02±0.19e	-4.57	$7.07{\pm}0.17^{g}$	$7.87{\pm}0.42^{ab}$	0.8
NBMD		MD	5.08±1.26	$4.24{\pm}0.96^{d}$	-0.84	$7.29{\pm}0.21^{defg}$	$7.58{\pm}0.21^{bcd}$	0.29
NBSI		SI	4.09±2.15	$11.83 \pm 3.58^{\circ}$	7.74	$7.09{\pm}0.25^{\mathrm{fg}}$	$7.26{\pm}0.36^{d}$	0.17
NBSII		SII	4.29±1.16	$17.10\pm0.89^{b}$	12.81	$7.30{\pm}0.07^{defg}$	$7.50{\pm}0.29^{cd}$	0.20
25NS	25% RHB	NS	5.38±1.35	$1.44{\pm}0.42^{e}$	-3.93	$7.22{\pm}0.26^{efg}$	$7.49{\pm}0.23^{cd}$	0.27
25MD		MD	5.72±1.18	$5.31{\pm}1.15^{d}$	-0.41	$7.38{\pm}0.46^{cdefg}$	$7.37{\pm}0.18^{d}$	-0.01
25SI		SI	5.43±1.95	12.14±1.93°	6.71	$7.49{\pm}0.21^{bcde}$	$7.41 \pm 0.13^{cd}$	-0.08
25SII		SII	6.55±1.27	18.96±3.28 <sup>ab</sup>	12.41	$7.45{\pm}0.15^{bcdef}$	$7.34{\pm}0.13^{d}$	-0.11
50NS	50% RHB	NS	4.59±1.33	1.13±0.33e	-3.46	$7.77 \pm 0.24^{ab}$	8.04±0.13ª	0.27
50MD		MD	4.97±1.09	$6.02{\pm}1.40^{d}$	1.05	$7.94{\pm}0.19^{a}$	$7.76 \pm 0.15^{abc}$	-0.18
50SI		SI	5.35±1.47	11.92±2.72°	6.57	$7.68{\pm}0.31^{abc}$	$7.85{\pm}0.33^{ab}$	0.17
50SII		SII	5.50±1.13	$20.05{\pm}1.92^{a}$	14.55	$7.60\pm0.34^{abcd}$	$7.88{\pm}0.19^{ab}$	0.28
C.V%			26.99	20.59		3.46	3.25	
F-test			ns	**		**	**	

RHB = rice husk biochar. ns = not significant \*\* Means within a column followed by the same letter are not statistically different by DMRT at 0.01 level.



**Fig. 1.** EC of substrate solutions of Ming Aralia grown in different RHB components under various saline conditions (NS = non-saline, MD =  $1.5 \text{ dS} \cdot \text{m}^{-1}$ , SI =  $4 \text{ dS} \cdot \text{m}^{-1}$  and SII =  $8 \text{ dS} \cdot \text{m}^{-1}$ ).

The high pH values were related to the percentage of RHB which resulted from the alkalinity (OH<sup>-</sup>) of RHB. Various researches have reported that soil pH increased due to biochar application; therefore, this must be considered as a soil amendment to improve acidic soils [29]. However, at the end of the experiment, the differences in pH found in the 25% RHB and 50% RHB treatments were less than those seen in the non-RHB component (Table 1). This may have resulted from biochar that had particles with absorbed ions, thus retaining positively charged ions on its surface; therefore, it could capture cations like H<sup>+</sup> in the substrate. Nevertheless, it could have been related to the substrate porosity. It is important to note that Thai coco coir has various particle-size diameters, a considerable amount of fine particles, 95.2-96.2% total pore space, and 786-533 mL·L<sup>-1</sup> of total water holding capacity, similar physical properties to peat [30]. With these physical properties, non-RHB (coco coir only) can hold tap water and render a pH value higher than RHB treatments. Whereas for RHB treatments, rice husk biochar may reduce the substrate's porosity, resulting in more stable pH values. The results showed the potential of RHB to maintain pH values for long durations, when plants are subjected to saline conditions.

#### **3.2** Plant growth and development of Ming Aralia cultivated under different RHB components and saline conditions.

Ming Aralia plants were subjected to saline conditions for six months and showed significant results (p<0.01) in all parameters. These results revealed that plant growth and development, including plant height, leaf area (LA), root length, and leaf/root fresh and dry weights, all decreased with increasing salinity conditions, going from non-saline to saline severity II (Fig. 2). These results indicate that Ming Aralia could be considered to have salt tolerance, due to the fact that all plants in this trial were still living by the end of the study period. These results are supported by other research [31], which reported that *Polyscias* spp. had a slight salt tolerance potential. Similar negative effects of salinity stress on plant growth and development (such as plant height and LA) were reported in studies on wheat [32], sweet corn [33], tomato [34], potato [19], artichoke [35], *Schizonepeta tenuifolia* [36], bean seedling [37], and *Suaeda Salsa* [38].

According to LA and plant weight (Figs. 2A,D,E), Ming Aralia plants cultivated with 25% RHB components had higher values than those cultivated with 50% RHB and non-RHB, especially when the plants were grown in the MD and SI saline conditions. The results show that biochar application at a moderate rate could ameliorate salinity stress, compared with a higher rate of biochar application [20, 32]. Appropriate biochar application could reduce sodium ion uptake and release mineral nutrients such as K, Ca, and Mg into the soil, leading to water availability in the soil. A high rate of biochar application has been shown to cause excessive sodium-ion build-up which in turn increases soil EC [39]. Further, it also induces soluble salts from biochar to leech into the soil solution [40, 41], physiological disrupting various and biochemical processes in cells, ultimately leading to reduced plant growth and development [42]. The results of this study indicate that applying the appropriate amount of biochar is essential in growing the Ming Aralia plant under saline conditions. To grow Aralia under saline conditions Ming (salinity:  $1.5 - 4.00 \text{ dS} \cdot \text{m}^{-1}$ ), rice husk biochar at 25% can be used for promoting plant growth and development. Apart from ornamental benefit, Ming Aralia is also used as an herbal plant; therefore, for the cultivation of this plant for medical purposes, the yield of its active compounds has to be considered along with plant yield.



**Fig. 2.** Plant height (a), leaf area (b), root length (c), leaves/root fresh weight (d) and leaves/root dry weight (e) of Ming Aralia grown in different RHB components under various saline conditions (NS= non-saline, MD =  $1.5 \text{ dS} \cdot \text{m}^{-1}$ , SI =  $4 \text{ dS} \cdot \text{m}^{-1}$  and SII =  $8 \text{ dS} \cdot \text{m}^{-1}$ ).

# **3.3** Active compounds in Ming Aralia cultivated under different RHB components and saline conditions.

#### 3.3.1 Antioxidant Activity (AOA)

The antiradical properties of the extract samples were measured by DPPH scavenging activity assay under wavelength 517 nm. AOA of dry and fresh leaf extracts are shown as  $IC_{50}$  values, while the AOA of dry and fresh root extracts are shown as % DPPH scavenging activity (Fig. 3). The results suggest that the leaves' antioxidant activity was higher than that of the roots, and

dry plant parts showed higher AOA than fresh. Concerning the AOA in fresh leaves, 50NS exhibited AOA higher than the other treatments, while control and 50SI showed the lowest AOA (Fig. 3A). Biochar addition increased AOA when the plants were subjected to non-saline conditions. This resulted from the biochar promoting plant growth (Fig. 2), by providing primary metabolites and altering the level of antioxidants in the plants. But the effectiveness of biochar increasing AOA in plants subjected to saline conditions is not

clear yet. AOA in dry leaves was higher than it was in fresh leaves, and varied according to saline conditions. The pattern of plant response to saline conditions was very clear (Fig. 3B) in dry leaves. AOA of SI plants was higher than those of other saline conditions. This trend appeared in all components (nonbiochar, 25% biochar, and 50% biochar). The results indicate that stress from salinity induced AOA in plants. Plant cells formed antioxidants in order to regulate osmotic solutes to enable water absorption and also to prevent cell damage from ROS. These results are similar to other research findings [43-45] that report that under salinity stress, oxidative stress was induced. The increase of AOA in plant tissues is considered to be a result of irregularities in the electron transport chain and accumulation of photoreducing power.

The high AOA of leaves in high salinity conditions indicates plants are responding to and coping with stress conditions; biochar application under saline conditions also results in higher AOA compared to non-biochar application plants. This can be explained by a higher rate of sodium ion absorption by the biochar, which leads to a significantly greater increase in soil EC compared to non-biochar application soil. Various studies have reported that active compounds were found in the roots of Polyscias fruticosa but data from % DPPH scavenging activity suggests that plant roots under 6-months old showed AOA lower than that of the leaf extract. Similar results have also reported on the low AOA of root extract when compared to leaf extract of Mentha pulegium under saline conditions from 0 mM to 75 mM NaCl [46]. These results may be related to plant age, which is linked to the accumulation of active compounds in this species [47].

#### 3.3.2 Total Phenolic Content (TPC)

TPC describes the whole of antioxidants used in defense systems to balance ROS production while under

environmental stress, especially salinity stress. The TPC showed significant differences among treatments in fresh leaves, dry leaves, and dry roots (Fig. 4). TPC in leaves was higher than in roots and was concentrated in the dry form. According to the TPC in dry leaves (Fig. 4B), SI tended to increase TPC, especially in 25% RHB which had the highest values  $(397.12 \text{ mg}_{GAE}/100 \text{ mg}_{GAE})$ g<sub>Dw</sub>). However, the TPC values measured from the other treatments demonstrated the plant's ability to produce high TPC values under various conditions. While some treatments may result in high TPC, it may negatively affect plant yields. Therefore, in terms of production capacity of TPC per pot (Fig. 5), whole plant production should also be considered. As seen in Fig. 5, TPC decreased as saline concentrations increased. and RHB promoted higher TPC in the plants. The results indicate that salinity SII caused too much stress for the plants. Biochar application at 25% was able to ameliorate the salt stress by increasing TPC to levels higher that those seen in the non-saline and 50% biochar applications. Similar results were also seen in the TPC of artichoke, which was reduced when the salinity rose above 6.9 dS·m<sup>-1</sup> [35]. A similar pattern was also reported in Salvia officinalis, when salinity rose higher than 75 mM NaCl [48]. Therefore. concerning Ming Aralia production. shows notable RHB а proficiency in increasing plant TPC while under saline conditions within the range of  $1.5 - 4 \text{ dS} \cdot \text{m}^{-1}$ , but higher than this, TPC will decrease. As with dry leaves, 25% RHB showed a tendency to increase TPC under saline conditions.

### 3.3.3 Total Flavonoid Content (TFC)

Due to the sample measurements all using same amount of 100 g of fresh leaves, the highest TFCs were found in plants subjected to non-biochar under MD and SI conditions, including 50% RHB under nonsaline and MD conditions. Whereas control (NBNS) NBSII and 50SII showed the lowest

TFC values (Fig. 6). The results indicate that 25% RHB produced TFC values lower than non-RHB and 50% RHB plants, but when plants were subjected to SII, this treatment could produce TFC at a higher level than non-RHB and 50%RHB. However, the results showed that the highest TFC according to plant yield was found in 50% RHB under non-saline conditions, followed by MD (Fig. 6). The results obtained resemble the accumulation of TFC in Schizonepeta tenuifolia, which increases with salinity in the range of 25-50 mM NaCl, but decreases in the range of severe salinity conditions (75-100 mM NaCl) [36]. For biochar application, a similar trend was also

seen in research on the ability of biochar to alleviate fluoride toxicity and oxidative stress in safflower (*Carthamus tinctorius* L.) seedlings. At higher fluoride concentrations (400 gNaF/kg soil and 800 gNaF/kg soil), TFC was reduced when compared to 0, 100, and 200 gNaF/kg soil. The application of biochar at 25 g/kg soil and 50 g/kg soil enhanced TFC as compared to the nonbiochar application plants [49]. Therefore, the results indicate that for the cultivation of Ming Aralia fresh leaves, 50% RHB should be used to promote optimal plant TFC, and that these plants can be grown under moderate saline conditions.



**Fig. 3.** Antioxidant activity of fresh leaves (a), dry leaves (b), fresh root (c), and dry root (d) of Ming Aralia grown in different RHB components under various saline conditions (NS= non-saline, MD = 1.5 dS·m<sup>-1</sup>, SI = 4 dS·m<sup>-1</sup> and SII = 8 dS·m<sup>-1</sup>).



**Fig. 4.** Total phenolic content of fresh leaves (a), dry leaves (b), fresh root (c), and dry root (d) of Ming Aralia grown in different RHB components under various saline conditions (NS= non-saline, MD = 1.5 dS·m<sup>-1</sup>, SI = 4 dS·m<sup>-1</sup> and SII = 8 dS·m<sup>-1</sup>).



**Fig. 5.** Total phenolic content per plant pot of Ming Aralia grown in different RHB components under various saline conditions (NS= non-saline, MD =  $1.5 \text{ dS} \cdot \text{m}^{-1}$ , SI =  $4 \text{ dS} \cdot \text{m}^{-1}$  and SII =  $8 \text{ dS} \cdot \text{m}^{-1}$ ).



**Fig. 6.** Flavonoid content of Ming Aralia grown in different RHB components under various saline conditions (NS= none saline, MD =  $1.5 \text{ dS} \cdot \text{m}^{-1}$ , SI =  $4 \text{ dS} \cdot \text{m}^{-1}$  and SII =  $8 \text{ dS} \cdot \text{m}^{-1}$ ).

#### 4. Conclusion

This study shows that the growth and development of Ming Aralia reliably decreases in response to increases in saline concentration. For antioxidant activity, total phenolic content and total flavonoid content were both found to be higher in the leaf than in the root. Saline concentrations at  $1.5 \text{ dS} \cdot \text{m}^{-1}$  (moderate saline) and 4 dS·m<sup>-1</sup> (severe saline) tended to promote the production of active compounds that are of pharmaceutical and industrial interest. Rice husk biochar at 25% and 50% has been shown to ameliorate the negative impacts of moderate to severe saline stress on plants.

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