

Morphological and physiological responses of *Coccoloba uvifera* (L.) L. seedlings of different origin to salt stress

Respuestas morfológicas y fisiológicas de plántulas de *Coccoloba uvifera* (L.) L. de diferente origen al estrés salino

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Section Editor: Dr. Antonio Juárez Maldonado

SUMMARY

The salinity of soils and water for irrigation is a limitation that influences the productivity of crops that has increased worldwide and affects the morphology and physiology of plants since it causes osmotic stress and ionic toxicity and impairs the absorption of water and nutrients. The effect of salt stress on some morphological and physiological traits in *Coccoloba uvifera* (L.) L. (sea grape) seedlings from different origins was evaluated and the traits whose behavior under salt stress could be used as an effective criterion of tolerance to salinity were determined. A completely randomized design experiment with ten repetitions was performed. The factors under study were salinity (0, 5, 15, and 25 dS m⁻¹), which was maintained for a period of sixteen weeks, and the second factor was the origin of the plant material. The salinity level and the seedlings' origin significantly influenced the morphological and physiological variables controlled. However, it is necessary to carry out *in situ* experiments to obtain more information related to the mechanisms used by sea grape to tolerate salinity conditions, because in the consulted sources no research focused on evaluating the effect of salinity and the origin on the morphology and physiology of *C. uvifera* seedlings in Cuba has been found. The results of the research provide preliminary information to understand the negative effect of salt stress on this type of plant. The behavior of some controlled traits under salt stress can be used as an effective criterion of tolerance to salinity.

Index words: *chlorophyll fluorescence, morpho physiology, salinity, sea grape, water status.*

RESUMEN

La salinidad de los suelos y del agua para riego es una limitante que influye en la productividad de los cultivos que se ha incrementado a nivel mundial y afecta la morfología y fisiología de las plantas, ya que provoca estrés osmótico y toxicidad iónica, lo que perjudica la absorción de agua y nutrientes. Se evaluó el efecto del estrés salino en características morfológicas y fisiológicas de plántulas de *Coccoloba uvifera* (L.) L. (uva de mar) de diferente origen, y se determinaron las características cuyo comportamiento ante el estrés salino podrían utilizarse como indicador de tolerancia a la salinidad. El experimento se estableció en un diseño



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Recommended citation:

Bullaín-Galardis, M., Campos-Posada, R., Campos-Posada, G., Eichler-Löbermann, B., Pruneau, L., Bâ, A., and López-Sánchez, R. (2023). Morphological and physiological responses of *Coccoloba uvifera* (L.) L. seedlings of different origin to salt stress. *Terra Latinoamericana*, 41, 1-15. e1655. <https://doi.org/10.28940/terra.v41i0.1655>

Received: September 23, 2022.

Accepted: January 21, 2023.

Article, Volume 41.

April 2023.

completamente aleatorizado con diez repeticiones. Los factores en estudio fueron la salinidad (0, 5, 15 y 25 dS m⁻¹), la cual se mantuvo durante un período de doce semanas, y el segundo factor fue el origen del material vegetal. El nivel de salinidad y el origen de las plántulas influyeron significativamente en las variables morfológicas y fisiológicas controladas. Sin embargo, es necesario realizar experimentos *in situ* para obtener mayor información relacionada con los mecanismos utilizados por la uva de mar para tolerar las condiciones de salinidad, pues en las fuentes consultadas no se han encontrado investigaciones enfocadas en evaluar el efecto de la salinidad y el origen en la morfología y fisiología de plántulas de *C. uvifera* en Cuba. Los resultados de la investigación aportan información preliminar para comprender el efecto negativo del estrés salino en este tipo de plantas. El comportamiento de algunas características controladas bajo estrés salino puede ser utilizado como criterio efectivo de tolerancia a la salinidad.

Palabras clave: fluorescencia de clorofila, morfofisiología, salinidad, uva de mar, estado hídrico.

INTRODUCTION

Salinity is considered a significant factor that affects crop production in arid and semiarid regions of the world (Tester and Davenport, 2003). Depending on the cause, soil salinity is classified as primary (due to natural processes) and secondary or anthropic (due to human activity). In some regions, the main natural causes of salinity are the capillary rise of the phreatic layers with saline characteristics, where the source of salinity is the weathering of the original soil material of a saline nature and the constant contribution of salts received by the soils near the sea or the gradual withdrawal of the seas which leads these soils to salinization (Courel, 2019; Arulmathi and Porkodi, 2020). Among the main anthropic causes are irrigation with saline water, sudden changes in land use, and the misuse of irrigation and fertilizers (Courel, 2019).

Salinity can disrupt cell function, through the toxic effects of specific ions and by osmotic effects, or both (Munns, 2005). The detrimental effects of high salinity on plants can be seen at the whole plant level as plant death or decreased productivity. Most terrestrial plants, including agricultural crops, are glycophytes and cannot tolerate a high concentration of salt, such as the case of corn (*Zea mays*), wheat (*Triticum aestivum*), rice (*Oryza sativa*) and tomato (*Solanum lycopersicum*) (Majeed and Muhammad, 2019). Many plants develop mechanisms either to exclude salt from their cells or to tolerate its presence within the cells.

During the onset and development of salt stress within a plant, all the major processes such as photosynthesis, protein synthesis, and energy and lipid metabolism are affected. The earliest response is a reduction in plant leaf area and stomatal density, followed by growth interruption as stress intensifies, but growth resumes as salt stress decreases (Romero-Aranda, Soria and Cuartero, 2001).

Carbohydrates, among other substances, are necessary for cell growth and are supplied primarily through the process of photosynthesis. When plants are exposed to salt stress, especially NaCl, photosynthesis rates are usually lower and vary depending on the type of plant (glycophytes or halophytes) and the level of salinity to which they are exposed (Parida and Das, 2005; Hniličková, Kraus, Vachová and František 2021; Wang *et al.*, 2022).

Salt tolerance in plants is a complex phenomenon that includes morphological changes and physiological and biochemical processes such as the elimination of salts by regulating the fall of leaves and in extreme cases of fruits (Del Amor-Saavedra, 2001), the elimination of excess salts through glands or specialized structures such as vesicular hairs, alteration in membrane structure (González, González and Ramírez, 2002), change in photosynthetic pathways, the accumulation or synthesis of proteins and amino acids that intervene in the osmotic adjustment (Ashraf and

Harris, 2004), the selective accumulation or exclusion of ions at the root level and the retention of ions in the vacuoles of growing roots or in different organs (ElYacoubi *et al.*, 2022), the accumulation of soluble carbohydrates to increase the osmotic potential (Murakeözy, Nagy, Duhazé, Bouchereau and Tuba, 2003).

To cultivate plants on salt-affected soils, tolerant plants with valued economic traits are required. Various investigations have identified halophytic plants of agroecological importance that grow well in coastal habitats, for example, *Crithmum maritimum*, *Inula chritmoides*, and *Salicornia bigelovii* (Ravindran, Venkatesan, Balakrishnan, Chellappan and Balasubramanian, 2007; Qadir *et al.*, 2008; Rueda-Puente, López and Huez, 2012; González-Villaveces, 2022¹).

Coccoloba uvifera (L.) L. (Polygonaceae), also called sea grape, is a tree with branches at a low level that grows up to 15 m in height, it is widely distributed along the Atlantic, Caribbean, and Pacific coasts of the American tropics and subtropics (Parrotta, 2000). It is used as a fruit tree (edible fruit), an ornamental plant for exteriors, and as a windbreak along the beaches and roads of the Caribbean.

Sea grape is considered a woody plant, drought resistant and non-halophilous relatively salt tolerant, growing mainly in stands within well-drained sandy soils with a slightly alkaline pH (Parrotta, 2000; Larcher, 2003). Although several authors have reported that the sea grape is tolerant to salt stress, in the sources consulted, there is no information related to the physiological and biochemical mechanisms used by this plant to grow in environments with high salt content. However, Guzmán, Ramírez, Miller, Lodge and Baroni (2004), Bâ, McGuire and Diédhiou (2014) and Séné *et al.* (2018) reported that this tree is associated with ectomycorrhizal fungi of the genera *Amanita*, *Inocybe*, *Cantharellus*, *Melanogaster*, *Cenococcum*, *Lactarius*, *Russula*, *Thelephoraceae*, *Xerocomus* and *Scleroderma*, on the other hand, Bandou *et al.* (2006) and Bullaín *et al.* (2022) observed that the tolerance of sea grape to salt stress is considerably improved by *S. bermudense*.

The knowledge of the effect of salt stress on the morphological and physiological traits of plants is important for the selection of plant material tolerant to salinity and the management of crops in soils affected by high concentrations of salts and can be used as one of the most challenging strategies to help organisms under stress conditions (Vahdati and Leslie, 2013).

The objective of the study was to evaluate the effect of salt stress on morphological and physiological traits in *C. uvifera* seedlings from different edaphic conditions and to select traits whose behavior under salt stress can be used as an effective criterion of tolerance to salinity under greenhouse conditions.

MATERIALS AND METHODS

Collection of Sea Grape Fruits

Sea grape seeds from three representative origins were collected in the northern and southern coastal areas of Cuba based on the presence of 40-year-old sea grape populations. The geographic location, annual precipitation, mean annual temperature, pH, and electrical conductivity of the soil of the selected origins are given in Table 1. Samples of 20 sea grape trees were randomly taken from each origin. Ripe fruits without damage or malformations were collected from different positions in the crown and placed in labeled plastic bags.

The fresh fruits were transported to the laboratory and the pulp surrounding the seed was manually removed, washed with potable water, and dried at a room temperature of 32-35 °C for 72 hours on 50 × 30 cm cardboard trays.

¹ González-Villaveces, A. F. (2022). *Especies halófitas de importancia agrícola como alternativa de biorremediación de suelos con problemas de salinización en Colombia*. Tesis para obtener el título de Ingeniero agrónomo. Facultad de Ciencias Agropecuarias. Universidad de Ciencias Aplicadas y Ambientales. Disponible en <https://repository.udca.edu.co/handle/11158/4907>

Table 1. Geographic location and environmental conditions of the place of origin of the fruits and seedlings of *C. uvifera*.

Origin	Location and environmental conditions of the three origins					
	Latitude	Longitude	MAP	MAT	pH	EC
			mm	°C		dS m ⁻¹
Las Coloradas	19° 56' 00"	77° 41' 00"	942	27.0	8.91	7.62
Guardalavaca	21° 07' 30"	75° 49' 44"	974	26.4	8.84	7.31
Cayo Coco	22° 32' 10.06"	78° 21' 19.44"	855	25.6	8.75	7.84

† MAP = mean annual precipitation; MAT = mean annual temperature; EC = electric conductivity.

Experimental Design, Plant Material and Environmental Conditions

C. uvifera seedlings grew for sixteen weeks following a completely randomized design with a factorial arrangement, consisting of three origins (Cayo Coco, Guardalavaca, and Las Coloradas) and four salinity levels (0, 5, 15, and 25 dS m⁻¹). A total of 12 treatments were compared, each with ten repetitions, and one plant per pot was established as the experimental unit.

To break dormancy, seeds were scarified in 95% sulfuric acid for 2 hours, and every 20 min the container was shaken to achieve the uniform action of the acid on the surface of the seeds (Bandou *et al.*, 2006). Then, they were rinsed with abundant distilled water and then germinated in pots (20 × 12 cm) with 1 kg of river sand, collected from the banks of the Yao River (20°15' 32.3" N, 76°45' 19.3" W) in Granma province, previously sterilized for one hour at 121 °C and 1.2 kg/cm² pressure in a vertical autoclave (BK-75, ТЗМОИ, Ukraine). After germination, the seedlings were maintained under good irrigation conditions for eight weeks (50 ml of potable water every 48 hours).

The analysis of the substrate was carried out in the Soil Laboratory of the Ministry of Agriculture of the Granma Province (Cuba) and the following determinations were made following the methodology proposed by Paneque *et al.* (2010):

K and Na: by the flame photometry method.

Ca and Mg: by the volumetric method with EDTA.

Assimilable P in the soil: by the Olsen method.

pH (KCL): by the potentiometric method, in a ratio of substrate: solution 1:2.5.

% of organic matter (OM): by the wet combustion method.

Electrical Conductivity (EC): by the method of estimation of salinity by the electrical conductivity of the extract of saturation of the substrate, using a portable conductivity meter (HANNA. HI 9033. Romania).

The result of the analysis of the substrate showed the following composition (mg L⁻¹): 67 K, 23.2 Na, 82.14 Ca, 28.12 Mg, 3.7 Olsen-P, pH (KCL) 7.2, 1.36% OM, and electrical conductivity (EC) 0.81 deciSiemens per meter.

The seedlings grew in a climatic cabinet (Snijders Scientific. ECL02. Netherlands) (day/night temperatures of 35/25 °C, relative humidity of 80%, and photoperiod of 12 hours of day-length).

Experimental Conditions and Controlled Variables

The NaCl concentration range was based on a field assessment of soil salinity in sea grape stands (Bojórquez *et al.*, 2008; Herrera-Romero, Bojórquez, Can, Madueño and García, 2019). Over the course of four weeks, the seedlings were brought to the desired salinity level for each treatment by adding NaCl (PanReac, Reagent Grade) in the irrigation. To avoid osmotic shock, a volume of 50 ml of NaCl solution with an electrical conductivity of 2 dS m⁻¹ was added to the substrate every

3 days. Upon reaching the desired salinity level for each treatment, exposure to salinity through irrigation was extended for sixteen weeks. The Na⁺ ions present in the substrate were leached with potable water every seven days to reduce salt accumulation. After each leaching, a volume of 50 ml of fresh saline solution was added to keep the NaCl concentration. The salinity was controlled weekly by the method of estimation of salinity by the electrical conductivity of the extract of saturation of the substrate (Torres, Camberato, Lopez and Mickelbart, 2010) using a portable conductivity meter (HANNA. HI 9033. Romania).

The chlorophyll fluorescence parameters, basal fluorescence (F_o), maximum fluorescence (F_m), variable fluorescence (F_v) the maximum quantum yield of photosystem II (F_v/F_m), the variable fluorescence initial fluorescence ratio (F_v/F_o), and performance index on absorption basis (PI_{abs}) were measured with a portable chlorophyll fluorimeter (Hansatech Instruments, RS232, United Kingdom), only one measurement was made in the fourth fully expanded leaf of each plant (ten plants per treatment, n = 10) in the morning. Before taking the measurement, the leaves were dark-adapted for 30 min using the clips provided with the kit. A saturating pulse of radiation (3500 μmol m⁻² s⁻¹ with the help of three light-emitting diodes of 650 nm) was given for (from 10 μs to 1 s) allowing to determine these parameters related to the fluorescence of chlorophyll.

After harvest, root, and stem length were measured and leaf area was determined using a foliar area meter (ADC BioScientific, AM350, United Kingdom) (ten plants per treatment, n = 10). The excess of the substrate was removed from the roots gently with potable water, then the excess water was removed from the surface of the plant using absorbent paper and the fresh weight of the aerial parts (FWAP) and the roots were immediately determined (FWR) with an analytical laboratory balance (Denver Instrument, TP-214, USA). The dry weight of aerial parts (leaves and stem) (DWAP) and roots (DWR) was measured after drying the samples at 80 °C until reaching a constant weight in an oven with air recirculation (Memmert, UNB 200, Germany). Root shoot ratio (RSR) was calculated as the ratio of the dry weight of roots to the dry weight of aerial parts (DWR/DWAP = RSR) (Rogers, Zalesny, Hallett, Headlee and Wiese, 2019).

The specific leaf area (SLA) was calculated using the formula (SLA = Leaf area (cm²)/dry weight of leaves (g) (Nageswara, Talwar and Wright, 2001). The leaf water content (LWC) and root water content (RWC) were calculated following the methodology of Cuni, De Smedt, Haq and Samson (2011):

$$\text{LWC} = 100 * (\text{fresh weight of leaves} - \text{dry weight of leaves}) / \text{fresh weight of leaves.}$$

$$\text{RWC} = 100 * (\text{fresh weight of roots} - \text{dry weight of roots}) / \text{fresh weight of roots.}$$

Statistical Analysis

Prior to ANOVA analysis, the normality of the data and the homogeneity of the variances were verified using the Shapiro Wilk and Levene tests, respectively ($P \geq 0.05$), with the statistical software MINITAB Release 13.20 (Minitab, 2000). All data were subjected to a two-way (origin and salinity) analysis of variance, the mean values were compared using Tukey's test ($P \leq 0.05$). A simple linear regression analysis was performed and the Pearson's correlation coefficient was determined with the InfoStat software version 2020 (Di Rienzo et al., 2020).

RESULTS AND DISCUSSION

Effects of NaCl on Morphological Traits

Salt stress had a negative effect on the morphological variables evaluated in *C. uvifera* seedlings. Significant differences ($P \leq 0.05$) were observed in all traits as the salinity level increased. The highest level of salinity (25 dS m⁻¹) caused the death

of *C. uvifera* seedlings. The origin, salinity, and the interaction between origin and salinity had a significant influence on each of the traits analyzed (Table 2).

In the seedlings from the three locations, the growth parameters evaluated, stem length and root length, FWAP, FWR, DWAP, RSR, and the leaf area were negatively and significantly affected as the salinity level increased from 0 to 25 dS m⁻¹. On the other hand, the DWR of the seedlings of the three localities did not show severe affections due to exposure to increased levels of salinity from 0 to 5 dS m⁻¹ and only seedlings from Cayo Coco showed significant differences between 5 and 15 dS m⁻¹. The RSR showed that at 15 dS m⁻¹ the seedlings presented a significantly higher amount of biomass in the root system (Table 2).

The values of the morphological traits evaluated were significantly higher in the seedlings from Cayo Coco, while the lower values were observed in the seedlings from Las Coloradas (Table 2).

Salt stress affected the vegetative growth and leaf development of *C. uvifera* seedlings from the three origins. Stem length, root length, and leaf area of seedlings decreased as the salinity level increased from 0 to 25 dS m⁻¹, although growth continued at 15 dS m⁻¹ (Table 2).

The reduction in the values of the controlled variables as the salinity level increased coincides with the results obtained by Bullaín *et al.* (2022) in *C. uvifera* seedlings from Las Coloradas beach, but they differ in that in this study the seedlings did not survive a salinity level of 25 dS m⁻¹, which may be the result of a longer exposure time to saline stress.

Table 2. Effect of salinity and origin in the morphological traits of *C. uvifera* seedlings.

Origin	Salt level	Morphological traits							
		Stem length	Root length	FWAP	FWR	DWAP	DWR	RSR	LA
	dS m ⁻¹	cm	cm	g					cm ²
Cayo Coco	0	15.62 a	10.20 a	0.78 a	0.14 a	0.53 a	0.12 a	0.22 cd	127.15 a
	5	10.42 d	7.66 d	0.61 b	0.13 b	0.32 d	0.12 a	0.37 b	60.18 d
	15	5.92 g	3.70 g	0.44 d	0.12 c	0.16 g	0.08 b	0.49 a	27.20 g
	25	0.00 j	0.00 j	0.00 g	0.00 f	0.00 j	0.00 e	0.00 e	0.00 j
Guardalavaca	0	14.92 b	9.80 b	0.76 a	0.12 c	0.51 b	0.08 b	0.15 cd	125.25 b
	5	9.66 e	7.36 e	0.58 c	0.07 e	0.31 e	0.07 bc	0.23 cd	58.27 e
	15	5.24 h	3.18 h	0.22 f	0.07 e	0.14 h	0.06 bc	0.42 ab	25.09 h
	25	0.00 j	0.00 j	0.00 g	0.00 f	0.00 j	0.00 e	0.00 e	0.00 j
Las Coloradas	0	13.38 c	8.86 c	0.62 b	0.13 b	0.49 c	0.07 bc	0.14 d	122.22 c
	5	8.26 f	6.40 f	0.45 d	0.09 d	0.28 f	0.05 cd	0.19 cd	55.22 f
	15	3.74 i	2.34 i	0.27 e	0.06 e	0.12 i	0.03 d	0.25 c	22.04 i
	25	0.00 j	0.00 j	0.00 g	0.00 f	0.00 j	0.00 e	0.00 e	0.00 j
Origin		S	S	S	S	S	S	S	S
Salinity		S	S	S	S	S	S	S	S
Origin × Salinity		S	S	S	S	S	S	S	S
C.V. (%)		0.25	0.46	5.46	2.77	0.79	19.83	26.42	0.06

† Distinct letters in the same column indicate significant differences (Tukey, $P \leq 0.05$). Values are the means ($n = 10$). C.V. = Coefficient of variation. S = significant ($P \leq 0.05$); FWAP = fresh weight of the aerial parts; FWR = fresh weight of roots; DWAP = dry weight of aerial parts; DWR = dry weight of roots; RSR = root shoot ratio; LA = leaf area.

El-Juhany, Ali, Basalah and Shehatah (2014) mentioned that the reduction of the leaf area in plants under the effect of salt stress is due to the decrease in the number of leaves. Also, Chamekh *et al.* (2014) observed a significant reduction in leaf area as salinity level increased in 17 of 25 genotypes of *Triticum turgidum* ssp. *durum*. On the other hand, Rahnesan, Nasibi and Moghadam (2018) reported that exposure to moderate and high salinities (0, 4.5, 9.1, and 13.7 dS m⁻¹) negatively affected stem and root length in two cultivars of *Pistacia vera* L.

Ahanger, Aziz, Alsahli, Alyemeni and Ahmad (2019) and Zhao, Zhang, Song, Zhu and Shabala (2020) observed that under salt stress conditions, the reduction of the values of the traits evaluated is due to the high osmotic pressure caused by the excessive accumulation of NaCl that interferes with the absorption of water and nutrients by the roots, this reduces growth and affects physiological processes such as photosynthetic rate, stomatal conductance, and transpiration rate.

Similar damage showed the FWAP, FWR, and DWAP. The values of FWAP and DWAP were higher in comparison to the FWR and DWR (Table 2), this coincides with the results obtained by Demir-Kaya, Ipek and Öztürk (2003) in which root biomass rarely overcomes stem biomass.

Plant growth is an important character in determining the salt tolerance capacity of plants. Although plant length is genetically controlled, environmental factors also have a strong influence on gene expression (Deinlein *et al.*, 2014). The main cause of decreased plant growth based on salinity stress is reduced photosynthesis (Chartzoulakis, Loupassaki, Bertaki and Androulakis, 2002; Sivritepe, Sivritepe, Çelik and Katkat, 2010). Also, under saline stress, competition is generated between the aerial organs and the roots for the absorption of photosynthetic materials and this affects these organs (Chookhampaeng, 2011; Hsiao & Xu, 2000).

Effects of NaCl on Physiological Traits

Salt stress also had a significant negative effect on controlled physiological traits in *C. uvifera* seedlings. Under salt stress, the reduction in LWC was 15.5% at 5 dS m⁻¹ and 36.34-36.35% at 15 dS m⁻¹. While the RWC showed the same behavior, the reduction due to salt stress was 17.02-17.2% at 5 dS m⁻¹ and 25.7-32.52% at 15 dS m⁻¹ (Table 3).

The origin, salinity, and interaction between these two experimental factors had a significant influence on the water content and the SLA of the seedlings of the three origins.

Salinity generates both water stress and osmotic stress in plants by decreasing water potential within the cells, and ionic stress due to the inhibition of specific metabolic processes such as the transport and elimination of ions, (mainly Na⁺, Cl⁻, and SO₄²⁻), (Heidari, 2012; Hniličková, Hnilička, Martinková and Kraus, 2017). The reduction of water uptake inhibits stomatal conductance, protein synthesis, and photosynthetic activity (Munns and Tester, 2008). Osmotic regulation is evidence of response to osmotic stress and in the conditions of water scarcity due to any stress, the osmotic potential decreases, and thus the relative water content of the leaves decreases (Karimi and Yusef-Zadeh, 2013). Osmotic regulation capacity is important for plants and the reduction of water potential is one of the mechanisms of resistance to salinity to maintain the higher relative water content of the leaf. The RWC is mostly correlated with the leaf area, dry weight of the leaf, amount of chlorophyll, and other growth rates indicators such as root and stem length (Kafi and Mahdavi, 2003).

The SLA is reduced under saline stress conditions and constitutes a way to improve Water-use efficiency (Bayuelo-Jiménez, Debouck and Lynch, 2003) This trait has a relation with leaf area and photosynthesis due to the sensitivity of photosynthesis and the reduction of leaf area under salt stress (El-juhany, Aref and Ahmed, 2008). Similar results were obtained by Ziaf *et al.* (2009) in plants of *Capsicum annuum* L.

Table 3. Effect of salinity and origin in the physiological traits of *C. uvifera* seedlings.

Origin	Salt level	Physiological traits		
		LWC	RWC	SLA
	dS m ⁻¹	----- % -----		cm ² g ⁻¹
Cayo Coco	0	81.12 a	79.22 a	253.22 a
	5	65.62 d	62.20 d	202.18 d
	15	44.77 g	46.52 h	188.28 g
	25	0.00 j	0.00 j	0.00 j
Guardalavaca	0	79.32 b	75.62 b	252.27 b
	5	63.82 e	58.42 e	200.24 e
	15	42.97 h	49.92 g	187.14 h
	25	0.00 j	0.00 j	0.00 j
Las Coloradas	0	77.72 c	72.72 c	249.28 c
	5	62.22 f	55.52 f	197.24 f
	15	41.38 i	40.20 i	184.14 i
	25	0.00 j	0.00 j	0.00 j
Origin		S	S	S
Salinity		S	S	S
Origin × Salinity		S	S	S
C.V. (%)		0.04	0.06	0.06

[†] Distinct letters in the same column indicate significant differences (Tukey, $P \leq 0.05$). Values are the means ($n = 10$). C.V.= Coefficient of variation. S = significant ($P \leq 0.05$). LWC = leaf water content; RWC = root water content; SLA = specific leaf area.

exposed to 2, 4, 6, and 8 dS m⁻¹ and Elfeel and Bakhshwain (2012) in plants of *Acacia saligna* (Labill.) H. Wendl subjected to 1.5, 7, 9 and 12 deciSiemens per meter.

The origin, the salinity, and the interaction between these two factors significantly influenced the parameters of chlorophyll fluorescence evaluated (Table 4).

As the salinity level increased, a negative effect on photochemical processes was observed. The values of F_o increased and the values of F_m , F_v/F_m , F_v/F_o , and PI_{abs} decreased in the seedlings of the three origins. The most critical values of the four parameters evaluated were observed in the seedlings from Las Coloradas.

A behavior similar to F_o was observed by De Lucena, Lopez, Prieto and Cecon (2012) and (Hniličková *et al.* (2017). This could be related to a decrease in the capacity to channel solar energy through photochemical pathways due to damage to the PSII reaction center or a reduction in the capacity to transfer excitation energy from the antenna to the reaction center as a result of exposure to salt stress (Baker, 2008; Jiménez-Suanca, Alvarado and Balaguera, 2015).

The reduction of F_m values with increasing salinity was observed by Erdal & Çakırlar (2014) and (Hniličková *et al.* (2017). This reduction may be the result of decreased chlorophyll levels, reduction in the number or severe damage to the antenna complex, accumulation of inactive reaction centers of photosystem II, and low efficiency in the photoreduction of quinone A and in the flow of electrons between the photosystems (Dan-Tatagiba, Kling, Telles and de Figueiredo 2014; de Melo, de Souza and Cunha, 2017; Tsai *et al.*, 2019).

The reduction of F_v/F_m in plants subjected to increasing levels of salinity has been reported by several investigations (Kalaji, Govindjee, Karolina, Kościelniak and Żuk-Gofaszewska, 2011; Casierra-Posada, Peña and Vaughan, 2013; Tsai *et al.*, 2019).

Table 4. Effect of salinity and origin in chlorophyll fluorescence parameters.

Origin	Salt level	Chlorophyll fluorescence parameters				
		Fo	Fm	Fv/Fm	Fv/Fo	PI _{abs}
	dS m ⁻¹	----- bits -----				
Cayo Coco	0	262.00 c	1806.80 a	0.86 a	5.91 a	2.04 a
	5	281.80 bc	1644.20 b	0.83 ab	4.85 b	1.29 b
	15	286.60 bc	1501.80 c	0.81 bc	4.25 bc	1.00 c
	25	0.00 d	0.00 e	0.00 e	0.00 e	0.00 g
Guardalavaca	0	270.20 c	1809.60 a	0.85 a	5.71 a	2.04 a
	5	284.80 bc	1649.00 b	0.83 ab	4.81 b	1.29 b
	15	288.40 bc	1509.20 c	0.81 bc	4.24 bc	1.00 c
	25	0.00 d	0.00 e	0.00 e	0.00 e	0.00 g
Las Coloradas	0	310.40 b	1496.00 c	0.79 c	3.84 c	0.28 d
	5	374.20 a	1442.80 cd	0.74 d	2.88 d	0.20 e
	15	378.60 a	1341.60 d	0.72 d	2.57 d	0.14 f
	25	0.00 d	0.00 e	0.00 e	0.00 e	0.00 g
Origin		S	S	S	S	S
Salinity		S	S	S	S	S
Origin × Salinity		S	S	S	S	S
C.V. (%)		7.46	4.74	2.63	12.03	6.88

[†] Distinct letters in the same column indicate significant differences (Tukey, $P \leq 0.05$). Values are the means ($n = 10$). C.V. = Coefficient of variation. S = significant ($P \leq 0.05$). Fo = basal fluorescence; Fm = maximum fluorescence; Fv/Fm = maximum quantum yield of photosystem II; PI_{abs} = performance index on absorption basis.

The presence of (Fv/Fm) values below 0.85 suggest exposure to some type of biotic or abiotic stress (Hansatech Instruments, 2006²). However, a value of 0.83 is considered acceptable for most plants (Maxwell and Johnson, 2000).

A sensitive indicator of photosynthetic activity in healthy and stressed plants is Fv/Fo (Li *et al.*, 2010). The significant differences in the values of Fv/Fo between 0 and 5 dS m⁻¹ in the seedlings of the three locations suggest that photosynthetic activity was affected by a slight increase in salinity, considering the high levels of salinity of the medium in which this species normally develops. The reduction of the value of Fv/Fo with the increase in the salinity level coincides with that observed by Kalaji *et al.* (2011), Khalid *et al.* (2015), Killi and Haworth (2017) and Pereira, de Siqueira, Martínez and Puiatti (2000) indicated that the reduction of Fv/Fo is a manifestation of structural damage in the photosynthetic apparatus, this causes damage to the efficiency of the photochemical processes and in the electron transport chain.

Plant vitality could be characterized by PI_{abs} it reflects the functionality of both photosystems I and II and provides quantitative information related to the current state of plant performance under stress conditions (Strasser, Srivastava and Tsimilli-Michael, 2000; Strasser, Tsimilli-Michael and Srivastava, 2004) The decrease in PI_{abs} values and the presence of significant differences between treatments as the salinity level increased in the seedlings of the three origins coincides with a similar behavior previously observed by Kalaji *et al.* (2011), Sayyad-Amin, Jahansooz, Borzouei and Ajili (2016), Yasmeen and Siddiqui (2017) and Estaji, Kalaji, Karimi, Roosta and Moosavi, (2019)

² Hansatech Instruments. (2006). *Handy PEA, Pocket PEA and PEA Plus Manual (versión 1.0)*. <https://studylib.net/doc/25578469/handy-pea-manual>

The correlation analysis between the traits evaluated and the saline stress levels (Table 5) showed that the correlation coefficient was highly significant and negative in all cases, therefore, the increase in the level of salinity will decrease the values of the variables evaluated. Except for RSR where the correlation coefficient between the evaluated traits and the salinity level was between -0.68 and -0.99.

The variation of stem length, root length, FWAP, DWAP, LWC, and RWC were determined by more than 90% by the linear relationship between the salinity level and these traits. This suggests that in *C. uvifera* seedlings these were the variables most sensitive to salt stress. The variation of the FWR, DWR, Leaf area, SLA, Fo, Fm, Fv/Fm, and Fv/Fo was determined by more than 50% by the relationship between these with the level of salinity. The RSR and the PI_{abs} were the traits whose variation was less determined by the salinity level, with a coefficient of determination of 0.10 and 0.47, respectively, it is then inferred that these were the variables least affected by exposure to this type of stress.

These results coincide with those obtained by Bandou *et al.* (2006) who reported the reduction of stem length, leaf area, DWAP, DWR, and leaf water potential in *C. uvifera* seedlings subjected to increasing levels of salinity (0, 18, 30, and 45 dS m⁻¹). This may be related to the fact that under salinity conditions macroscopic changes are observed, such as the reduction of leaf area and RSR (Leidi and Pardo, 2002). Hasan, Kawasaki, Taniguchi and Miyake (2018) indicated that the osmotic stress caused by salt accumulation within cells and tissues contributes to cell distortion and acceleration of cell death. On the other hand, it is widely reported that salt stress inhibits plant growth (Sam, 2007; Martínez-Villavicencio, López, Pérez and Basurto, 2011; Zhao *et al.*, 2020).

Table 5. Regression equations and correlation and determination coefficients between the salinity level and the controlled variables in *C. uvifera* seedlings.

Controlled variables	Equation	r	R ²
Stem length (cm)	$y = -0.5561x + 13.519$	-0.98**	0.95
Root length (cm)	$y = -0.3833x + 9.2703$	-0.99**	0.97
FWAP (g)	$y = -0.028x + 0.7076$	-0.96**	0.93
FWR (g)	$y = -0.0048x + 0.1317$	-0.88**	0.77
DWAP (g)	$y = -0.0192x + 0.4532$	-0.97**	0.94
DWR (g)	$y = -0.0034x + 0.0939$	-0.78**	0.61
RSR	$y = -0.0057x + 0.2684$	-0.32**	0.10
Leaf area (cm ²)	$y = -4.5389x + 102.95$	-0.93**	0.86
LWC (%)	$y = -3.0672x + 81.085$	-0.99**	0.97
RWC (%)	$y = -2.8461x + 77.046$	-0.97**	0.93
SLA (cm ² g ⁻¹)	$y = -9.1667x + 262.62$	-0.92**	0.85
Fo (bits)	$y = -10.652x + 347.91$	-0.75**	0.56
Fm (bits)	$y = -63.993x + 1903.3$	-0.88**	0.78
Fv/Fm (bits)	$y = -0.031x + 0.9511$	-0.85**	0.72
Fv/Fo (bits)	$y = -0.1907x + 5.4005$	-0.86**	0.74
Plabs	$y = -0.0528x + 1.3673$	-0.68**	0.47

[†] R² = determination coefficient; r = correlation coefficient; ** = highly significant ($P \leq 0.01$). FWAP = fresh weight of the aerial parts; FWR = fresh weight of roots; DWAP = dry weight of aerial parts; DWR = dry weight of roots; RSR = root shoot ratio; LWC = leaf water content; RWC = root water content; SLA = specific leaf area; Fo = basal fluorescence; Fm = maximum fluorescence; Fv/Fm = maximum quantum yield of photosystem II; PI_{abs} = performance index on absorption basis.

Under saline stress, seedlings from Cayo Coco showed higher morphological and physiological values than those from other origins, the tolerance of salt stress was more evident indicating a better ability to grow with salt stress. The differences in the response to salt stress between sea grape seedlings from different origins are related to the ecological conditions where these tree populations are established (Gilman *et al.*, 2014). The seedlings from Las Coloradas showed greater susceptibility to saline stress, apparently, the presence of mean annual precipitation of 942 mm, a mean annual temperature of 27.0 °C and a pH of 8.91 and an electrical conductivity of 7.62 dS m⁻¹ in the soil solution, or the synergy of these variables, makes the environmental conditions on the southeastern coast of Cuba more favorable for the growth and development of *C. uvifera* seedlings compared to the eastern and central north coast.

CONCLUSIONS

Exposure to increasing levels of salinity, the origin of the seedlings, and the interaction between these two factors had a significant effect on all the morphological and physiological traits controlled. RSR and PI_{abs} are not good markers of salt tolerance in *C. uvifera* seedlings, since their variation was the least influenced by the salinity level. The fact that more than 90% of the variation of stem and root length, FWAP, DWAP, LWC, and RWC is determined by the salinity level would allow using the behavior of these traits under salt stress as an effective criterion for salinity tolerance.

ETHICS STATEMENT

Not applicable.

CONSENT FOR PUBLICATION

Not applicable.

AVAILABILITY OF SUPPORTING DATA

The sets of data used or analyzed during this study are available through the corresponding author upon reasonable request.

COMPETING INTERESTS

The authors declare that they have no competing interests.

AUTHORS' CONTRIBUTIONS

Conceptualization: R.L.S., A.B., and M.B.G. Methodology: R.L.S., A.B., and M.B.G. Validation: M.B.G., and L.P. Formal analysis: M.B.G., B.E.L., R.C.P., G.C.P. and L.P. Investigation: M.B.G. and R.L.S. Data curation: M.B.G., R.L.S. and B.E.L. Writing-original draft preparation: M.B.G. and R.L.S. Writing-review and editing: M.B.G., R.L.S., A.B. and B.E.L. Statistical analysis: M.B.G. Supervision: R.L.S., A.B. and M.B.G. Funding acquisition: R.L.S.

ACKNOWLEDGMENTS

The authors acknowledge the support provided by the Abiotic Stress Laboratory of the University of Granma, the Laboratory of Plant Biology and Physiology of the

University of the Antilles and the Department of Plant Sciences of the University of Rostock. The funding provided by the University of Granma to carry out this research is also appreciated.

REFERENCES

- Ahanger, M. A., Aziz, U., Alsahli, A. A., Alyemeni, M. N., & Ahmad, P. (2019). Influence of exogenous salicylic acid and nitric oxide on growth, photosynthesis, and ascorbate- glutathione cycle in salt stressed vigna angularis. *Biomolecules*, 10(1), 1-16. <https://doi.org/10.3390/biom10010042>
- Arulmathi, C., & Porkodi, G. (2020). Characteristics of Coastal Saline Soil and their Management: A Review. *International Journal of Current Microbiology and Applied Sciences*, 9(10), 1726-1734. <https://doi.org/10.20546/ijcmas.2020.910.209>
- Ashraf, M., & Harris, P. J. C. (2004). Potential biochemical indicators of salinity tolerance in plants. *Plant Science*, 166(1), 3-16. <https://doi.org/10.1016/j.plantsci.2003.10.024>
- Bâ, A. M., McGuire, K. L., & Diédhiou, A. G. (2014). *Ectomycorrhizal Symbioses in Tropical and Neotropical Forests*. Boca Ratón, FL, USA: CRC Press. <https://doi.org/10.1201/b16536>
- Baker, N. R. (2008). Chlorophyll Fluorescence: A Probe of Photosynthesis In Vivo. *Annual Review of Plant Biology*, 59(1), 89-113. <https://doi.org/10.1146/annurev.arplant.59.032607.092759>
- Bandou, E., Lebailly, F., Muller, F., Dulormne, M., Toribio, A., Chabrol, J., ... Bâ, A. M. (2006). The ectomycorrhizal fungus *Scleroderma bermudense* alleviates salt stress in seagrape (*Coccoloba uvifera* L.) seedlings. *Mycorrhiza*, 16(8), 559-565. <https://doi.org/10.1007/s00572-006-0073-6>
- Bayuelo-Jiménez, J. S., Debouck, D. G., & Lynch, J. P. (2003). Growth, gas exchange, water relations, and ion composition of Phaseolus species grown under saline conditions. *Field Crops Research*, 80(3), 207-222. [https://doi.org/10.1016/S0378-4290\(02\)00179-X](https://doi.org/10.1016/S0378-4290(02)00179-X)
- Bojórquez, I., Hernández, A., García, D., Nájera, O., Flores, F., Madueño, A., & Bugarín, R. (2008). Características de los suelos de las barras paralelas, playas y dunas de la llanura costera norte del estado de Nayarit, México. *Cultivos Tropicales*, 29(1), 37-42.
- Bullaín-Galardis, M. M., López Sánchez, R. C., Fall, F., Eichler-Löbermann, B., Pruneau, L., & Bâ, A. M. (2022). Growth and physiological responses of ectomycorrhizal *Coccoloba uvifera* (L.) L. seedlings to salt stress. *Journal of Arid Environments*, 196, 104650. <https://doi.org/10.1016/j.jaridenv.2021.104650>
- Casierra-Posada, F., Peña-Olmos, J. E., & Vaughan, G. (2013). Photochemical efficiency of photosystem II and growth in banana passionfruit plants (*Passiflora tripartita* (Juss.) var. *mollissima* (Kunt) L.H. Bailey under salt stress. *Acta Agronómica*, 62(1), 21-26.
- Chamekh, Z., Ayed, S., Sahli, A., Ayadi, S., Hammemi, Z., Jallouli, S., ... Amara, H. (2014). Effect of salt stress on the flag leaf area and yield components in twenty five durum wheat genotypes (*Triticum turgidum* ssp. durum). *Journal of New Sciences*, 6(3), 15-23.
- Chartzoulakis, K., Loupassaki, M., Bertaki, M., & Androulakis, I. (2002). Effects of NaCl salinity on growth, ion content and CO₂ assimilation rate of six olive cultivars. *Scientia Horticulturae*, 96(1-4), 235-247. [https://doi.org/10.1016/S0304-4238\(02\)00067-5](https://doi.org/10.1016/S0304-4238(02)00067-5)
- Chookhampaeng, S. (2011). The effect of salt stress on growth, chlorophyll content proline content and antioxidative enzymes of pepper (*Capsicum annum* L.) seedling. *European Journal of Scientific Research*, 49, 103-109.
- Courel, G. F. (2019). *Suelos salinos y sódicos*. Tucuman, Argentina: Universidad Nacional de Tucumán.
- Cuni, A., De Smedt, S., Haq, N., & Samson, R. (2011). Variation in baobab seedling morphology and its implications for selecting superior planting material. *Scientia Horticulturae*, 130(1), 109-117. <https://doi.org/10.1016/j.scienta.2011.06.021>
- Dan-Tatagiba, S., Kling-Moraes, G. A. B., Telles-Nascimento, K. J., & de Figueiredo-Peloso, A. (2014). Limitações fotossintéticas em folhas de plantas de tomateiro submetidas a crescentes concentrações salinas. *Revista Engenharia na Agricultura*, 22(2), 138-149.
- De Lucena, C. C., Lopez-de Siqueira, D., Prieto-Martínez, H. E., & Cecon, P. R. (2012). Salt stress change chlorophyll fluorescence in mango. *Revista Brasileira de Fruticultura*, 34(4), 1245-1255. <https://doi.org/10.1590/S0100-29452012000400034>
- de Melo, H. F., de Souza, E. R., & Cunha, J. C. (2017). Fluorescence of chlorophyll a and photosynthetic pigments in *Atriplex nummularia* under abiotic stresses. *Revista Brasileira de Engenharia Agrícola e Ambiental*, 21(4), 232-237. <https://doi.org/10.1590/1807-1929/agriambi.v21n4p232-237>
- Deinlein, U., Stephan, A. B., Horie, T., Luo, W., Xu, G., & Schroeder, J. I. (2014). Plant salt-tolerance mechanisms. *Trends in Plant Science*, 19(6), 371-379. <https://doi.org/10.1016/j.tplants.2014.02.001>
- Del Amor-Saavedra (2001). Optimización del manejo de aguas salinas en el cultivo del tomate en invernadero. *Agrícola Vergel: Fruticultura, Horticultura, Floricultura*, 239, 588-592
- Demir-Kaya, M., Ipek, A., & Öztürk, A. (2003). Effects of different soil salinity levels on germination and seedling growth of safflower (*Carthamus tinctorius* L.). *Turkish Journal of Agriculture and Forestry*, 27(4), 221-227.
- Di Rienzo, J. A., Casanoves, F., Balzarini, M. G., González, L. A., Tablada, E. M., & Robledo, C. W. (2020). *InfoStat versión 2020*. software estadístico. Córdoba, Argentina: Grupo InfoStat.
- El-Juhany, L. I., Ali, H. M., Basalah, M. O., & Shehatah, A. M. S. (2014). Effects of water stress and salinity on the growth of Hibiscus tiliaceus Trees. *Journal of Pure and Applied Microbiology*, 8(2), 399-408.

- El-juhany, L. I., Aref, I. M., & Ahmed, A. I. M. (2008). Response of *Eucalyptus camaldulensis*, *Eucalyptus microtheca* and *Eucalyptus intertexta* seedlings to irrigation with saline water. *World Journal of Agricultural Sciences*, 4, 825-834.
- Elfeel, A. A., & Bakhashwain, A. A. (2012). Salinity Effects on Growth Attributes Mineral Uptake, Forage Quality and Tannin Contents of *Acacia saligna* (Labill.) H. Wendl. *Research Journal of Environmental and Earth Sciences*, 4(11), 990-995.
- ElYacoubi, H., Mouhssine, F., Imtara, H., Ouallal, I., Ech-cheddadi, S., Koutoua, A., ... Rochdi, A. (2022). Insight into Membrane Stability and Physiological Responses of Selected Salt-Tolerant and Salt-Sensitive Cell Lines of Troyer Citrange (*Citrus sinensis* [L.] x *Citrus trifoliata* [L.] Raf.) under Salt Stress. *Sustainability*, 14(15), 1-17. <https://doi.org/10.3390/su14159583>
- Erdal, Ş., & Çakırlar, H. (2014). Impact of salt stress on photosystem II efficiency and antioxidant enzyme activities of safflower (*Carthamus tinctorius* L.) cultivars. *Turkish Journal of Biology*, 38, 549-560. <https://doi.org/10.3906/biy-1401-33>
- Estaji, A., Kalaji, H. M., Karimi, H. R., Roosta, H. R., & Moosavi-Nezhad, S. M. (2019). How glycine betaine induces tolerance of cucumber plants to salinity stress? *Photosynthetica*, 57(3), 753-761. <https://doi.org/10.32615/ps.2019.053>
- Gilman, E. F., Watson, D. G., Klein, R. W., Koeser, A. K., Hilbert, D. R., & McLean, D. C. (2014). *Coccoloba uvifera*: Sea grape. Orlando, FL, USA: University of Florida.
- González, L. M., González, M. C., & Ramírez, R. (2002). Reseña bibliográfica. Aspectos generales sobre la tolerancia a la salinidad en las plantas cultivadas. *Cultivos Tropicales*, 23(2), 27-37.
- Guzmán, G., Ramírez-Guillen, F., Miller, O. K., Lodge, D. J., & Baroni, T. J. (2004). *Scleroderma stellatum* versus *Scleroderma bermudense*: the status of *Scleroderma echinatum* and the first record of *Veligaster nitidum* from the Virgin Islands. *Mycologia*, 96(6), 1370-1379. <https://doi.org/10.2307/3762153>
- Hasan, R., Kawasaki, M., Taniguchi, M., & Miyake, H. (2018). The Damage of Root, Leaf and chloroplast ultrastructure on maize seedlings caused by salinity stress. *IOP Conferences Series: Earth and Environmental Science*, 197, 1-6. <https://doi.org/10.1088/1755-1315/197/1/012054>
- Heidari, M. (2012). Effects of salinity stress on growth, chlorophyll content and osmotic components of two basil (*Ocimum basilicum* L.) genotypes. *African Journal of Biotechnology*, 11(2), 379-384. <https://doi.org/10.5897/ajb11.2572>
- Herrera-Romero, J. A., Bojorquez-Serrano, J. I., Can-Chulim, Á., Madueño-Molina, A., & García-Paredes, J. D. (2019). Salinity and soil properties of beach ridge in national marshlands in Mexico. *Revista Bio Ciencias*, 6, 1-14. <https://doi.org/10.15741/revbio.06.e412>
- Hnilíčková, H., Hnilička, F., Martinková, J., & Kraus, K. (2017). Effects of salt stress on water status, photosynthesis and chlorophyll fluorescence of rocket. *Plant Soil Environment*, 63(8), 362-367. <https://doi.org/10.17221/398/2017-PSE>
- Hnilíčková, H., Kraus, K., Vachová, P., & František, H. (2021). Salinity stress affects photosynthesis, malondialdehyde formation, and proline content in *Portulaca oleracea* L. *Plants*, 10(5), 1-14. <https://doi.org/10.3390/plants10050845>
- Hsiao, T. C., & Xu, L. K. (2000). Sensitivity of with of roots versus leaves to water stress: Biophysical analysis and relation to water. *Journal of Experimental Botany*, 51(350), 1595-1616. <https://doi.org/10.1093/jexbot/51.350.1595>
- Jiménez-Suanca, S. C., Alvarado, O. H., & Balaguera-López, H. E. (2015). Fluorescencia como indicador de estrés en *Helianthus annuus* L. Una revisión. *Revista Colombiana de Ciencias Hortícolas*, 9(1), 149-160. <https://doi.org/10.17584/rcch.2015v9i1.3753>
- Kafi, M., & Mahdavi, A. M. (2003). Mechanisms of plant resistance to environmental stresses. Mashhad, Razavi, Iran: Ferdowsi University Publications of Mashhad.
- Kalaji, H. M., Govindjee., Karolina, B., Kościelniak, J., & Żuk-Gołaszewska, K. (2011). Effects of salt stress on photosystem II efficiency and CO₂ assimilation of two Syrian barley landraces. *Environmental and Experimental Botany*, 73(11), 64-72. <https://doi.org/10.1016/j.envexpbot.2010.10.009>
- Karimi, H., & Yusef-Zadeh, H. (2013). The effect of salinity level on the morphological and physiological traits of two grape (*Vitis vinifera* L.) cultivars. *International Journal of Agronomy and Plant Production* 4(5), 1108-1117.
- Khalid, A., Athar, H., Zafar, Z. U., Akram, A., Hussain, K., Manzoor, H., ... Ashraf, M. (2015). Photosynthetic capacity of canola (*Brassica napus* L.) plants as affected by glycinebetaine under salt stress. *Journal of Applied Botany and Food Quality*, 88, 78-86. <https://doi.org/10.5073/JABFQ.2015.088.011>
- Killi, D., & Haworth, M. (2017). Diffusive and metabolic constraints to photosynthesis in quinoa during drought and salt stress. *Plants*, 6(4), 1-15. <https://doi.org/10.3390/plants6040049>
- Larcher, W. (2003). *Physiological Plant Ecology. Ecophysiology and Stress Physiology of Functional Groups* (4th ed.). Berlin Heidelberg, Germany: Springer. ISBN: 3-540-43516-6
- Leidi, E. O., & Pardo, J. M. (2002). Tolerancia de los cultivos al estrés salino: qué hay de nuevo. *Revista de Investigaciones de la Facultad de Ciencias Agrarias de la Universidad Nacional de Rosario*, 2, 70-91.
- Li, G., Wan, S., Zhou, J., Yang, Z., & Qin, P. (2010). Leaf chlorophyll fluorescence, hyperspectral reflectance, pigments content, malondialdehyde and proline accumulation responses of castor bean (*Ricinus communis* L.) seedlings to salt stress levels. *Industrial Crops and Products*, 31(1), 13-19. <https://doi.org/10.1016/j.indcrop.2009.07.015>
- Majeed, A., & Muhammad, Z. (2019). Salinity: A Major Agricultural Problem-Causes, Impacts on Crop Productivity and Management Strategies. In M. Hasanuzzaman, K. R. Hakeem, K. Nahar, & H. F. Alharby (Eds.). *Plant Abiotic Stress Tolerance, Agronomic, Molecular and Biotechnological Approaches* (pp. 83-99). Berlin, Germany: Springer Nature. <https://doi.org/10.1007/978-3-030-06118-0>

- Martínez-Villavicencio, N., López-Alonzo, C. V., Pérez-Leal, R., & Basurto-Sotelo, M. (2011). Efectos por salinidad en el desarrollo vegetativo. *Tecnociencia Chihuahua*, 5(3), 156-161.
- Maxwell, K., & Johnson, G. N. (2000). Chlorophyll fluorescence - a practical guide. *Journal of Experimental Botany*, 51(345), 659-668.
- Minitab (2000). *Minitab Statistical Software User's Guide. Version 13.20*. State College, PA, USA: Minitab Inc.
- Munns, R. (2005). Genes and salt tolerance: Bringing them together. *New Phytologist*, 167(3), 645-663. <https://doi.org/10.1111/j.1469-8137.2005.01487.x>
- Munns, R., & Tester, M. (2008). Mechanisms of salinity tolerance. *Annual Review of Plant Biology*, 59, 651-681. <https://doi.org/10.1146/annurev.arplant.59.032607.092911>
- Murakeözy, É. P., Nagy, Z., Duhazé, C., Bouchereau, A., & Tuba, Z. (2003). Seasonal changes in the levels of compatible osmolytes in three halophytic species of inland saline vegetation in Hungary. *Journal of Plant Physiology*, 160(4), 395-401. <https://doi.org/10.1078/0176-1617-00790>
- Nageswara, R. C., Talwar, H. S., & Wright, G. C. (2001). Rapid assessment of specific leaf area and leaf nitrogen in peanut (*Arachis hypogaea* L.) using a chlorophyll meter. *Agronomy & Crop Science*, 186, 175-182. <https://doi.org/10.1046/j.1439-037X.2001.00472.x>
- Paneque, V. M., Calaña, J. M., Calderón, M., Borges, Y., Hernández, T. C., & Caruncho, M. (2010). *Manual de técnicas analíticas para análisis de suelo, foliar, abonos orgánicos y fertilizantes químicos*. La Habana, Cuba: Ediciones INCA. ISBN: 978-959-7023-51-7
- Parida, A. K., & Das, A. B. (2005). Salt tolerance and salinity effects on plants: a review. *Ecotoxicology and Environmental Safety*, 60, 324-349. <https://doi.org/10.1016/j.ecoenv.2004.06.010>
- Parrotta, J. A. (2000). *Coccoloba uvifera* (L.) L. Uva de playa, sea grape. *Polygonaceae*. Familia de los polígonos. En J. K. Francis, & C. A. Lowe (Eds.). *Bioecología de arboles nativos y exóticos de Puerto Rico y las Indias Occidentales* (pp. 148-151). Rio Piedras, Puerto Rico: USDA
- Pereira, W. E., de Siqueira, D. L., Martínez, C. A., & Puiatti, M. (2000). Gas exchange and chlorophyll fluorescence in four citrus rootstocks under aluminium stress. *Journal of Plant Physiology*, 157(5), 513-520. [https://doi.org/10.1016/S0176-1617\(00\)80106-6](https://doi.org/10.1016/S0176-1617(00)80106-6)
- Qadir, M., Tubeileh, A., Akhtar, J., Larbi, A., Minhas, P. S., & Khan, M. A. (2008). Productivity enhancement of salt-affected environments through crop diversification. *Land Degradation & Development*, 19(4), 429-453. <https://doi.org/10.1002/ldr.853>
- Rahneshan, Z., Nasibi, F., & Moghadam, A. A. (2018). Effects of salinity stress on some growth, physiological, biochemical parameters and nutrients in two pistachio (*Pistacia vera* L.) rootstocks. *Journal of Plant Interactions*, 13(1), 73-82. <https://doi.org/10.1080/17429145.2018.1424355>
- Ravindran, K. C., Venkatesan, K., Balakrishnan, V., Chellappan, K. P., & Balasubramanian, T. (2007). Restoration of saline land by halophytes for Indian soils. *Soil Biology and Biochemistry*, 39(10), 2661-2664. <https://doi.org/10.1016/j.soilbio.2007.02.005>
- Rogers, E. R., Zalesny, R. S., Hallett, R. A., Headlee, W. L., & Wiese, A. H. (2019). Relationships among root-shoot ratio, early growth, and health of hybrid poplar and willow clones grown in different landfill soils. *Forests*, 10(1), 1-18. <https://doi.org/10.3390/f10010049>
- Romero-Aranda, R., Soria, T., & Cuartero, J. (2001). Tomato plant-water uptake and plant-water relationships under saline growth conditions. *Plant Science*, 160(2), 265-272. [https://doi.org/10.1016/S0168-9452\(00\)00388-5](https://doi.org/10.1016/S0168-9452(00)00388-5)
- Rueda-Puente, E. O., López-Elias, J., & Huez-López, M. A. (2012). *Las halófitas en la agricultura de zonas áridas*. East Finchley, London, United Kingdom: Editorial Académica Española. ISBN: 978-3-659-03196-0
- Sam, O. (2007). Influencia de la salinidad en las radículas de plántulas de arroz (*Oryza sativa*). *Cultivos Tropicales*, 28(1), 25-28.
- Sayyad-Amin, P., Jahansooz, M. R., Borzouei, A., & Ajili, F. (2016). Changes in photosynthetic pigments and chlorophyll- a fluorescence attributes of sweet-forage and grain sorghum cultivars under salt stress. *Journal of Biological Physics*, 42, 601-620. <https://doi.org/10.1007/s10867-016-9428-1>
- Séne, S., Avril, R., Chaintreuil, C., Geoffroy, A., Ndiaye, C., Diédhiou, A. G., ... Bâ, A. (2015). Ectomycorrhizal fungal communities of *Coccoloba uvifera* (L.) L. mature trees and seedlings in the neotropical coastal forests of Guadeloupe (Lesser Antilles). *Mycorrhiza*, 25(7), 547-559. <https://doi.org/10.1007/s00572-015-0633-8>
- Séne, S., Selosse, M. A., Forget, M., Lambourdière, J., Cissé, K., Diédhiou, A. G., ... Bâ, A. (2018). A pantropically introduced tree is followed by specific ectomycorrhizal symbionts due to pseudo-vertical transmission. *The ISME Journal*, 12(7), 1806-1816. <https://doi.org/10.1038/s41396-018-0088-y>
- Sivritepe, N., Sivritepe, H. Ö., Çelik, H., & Katkat, A. V. (2010). Salinity responses of grafted grapevines: Effects of scion and rootstock genotypes. *Notulae Botanicae Horti Agrobotanici Cluj-Napoca*, 38(3), 193-201. <https://doi.org/10.15835/nbha3834677>
- Strasser, R. J., Srivastava, A., & Tsimilli-Michael, M. (2000). The fluorescence transient as a tool to characterize and screen photosynthetic samples. En M. Yunus, U. Pathre, & P. Mohanty (Eds.). *Probing photosynthesis: Mechanism, regulation & adaptation* (pp. 445-483). New York, NY, USA: Taylor & Francis. ISBN: 0-7484-0821-5
- Strasser, R. J., Tsimilli-Michael, M., & Srivastava, A. (2004). Analysis of the Chlorophyll a Fluorescence Transient. En G. C. Papageorgiou & Govindjee (Eds.). *Advances in Photosynthesis and Respiration*. (pp. 321-362). Berlin, Germany: Springer. https://doi.org/10.1007/978-1-4020-3218-9_12
- Tester, M., & Davenport, R. (2003). Na Tolerance and Na Transport in Higher Plants. *Annals of Botany*, 91(5), 503-527. <https://doi.org/10.1093/aob/mcg058>
- Torres, A. P., Camberato, D., Lopez, R. G., & Mickelbart, M. (2010). *Medición de pH y conductividad eléctrica en sustratos*. West Lafayette, IN, USA: Purdue Extension.

- Tsai, Y. C., Chen, K. C., Cheng, T. S., Lee, C., Lin, S. H., & Tung, C. W. (2019). Chlorophyll fluorescence analysis in diverse rice varieties reveals the positive correlation between the seedlings salt tolerance and photosynthetic efficiency. *BMC Plant Biology*, 19(1), 1-17. <https://doi.org/10.1186/s12870-019-1983-8>
- Vahdati, K., & Leslie, C. (2013). *Abiotic Stress - Plant Responses and Applications in Agriculture*. Gate Court, London, United Kingdom: InTech. <https://doi.org/10.5772/45842>
- Wang, C. W., Wong, S. L., Liao, T. S., Weng, J. H., Chen, M. N., Huang, M. Y., & Chen, C. I. (2022). Photosynthesis in response to salinity and submergence in two Rhizophoraceae mangroves adapted to different tidal elevations. *Tree Physiology*, 42(5), 1016-1028. <https://doi.org/10.1093/treephys/tpab167>
- Yasmeen, R., & Siddiqui, Z. S. (2017). Physiological responses of crop plants against *Trichoderma harzianum* in saline environment. *Acta Botánica Croata*, 76(2), 154-162. <https://doi.org/10.1515/botcro-2016-0054>
- Zhao, C., Zhang, H., Song, C., Zhu, J. K., & Shabala, S. (2020). Mechanisms of Plant Responses and Adaptation to Soil Salinity. *The Innovation*, 1(1), 1-41. <https://doi.org/10.1016/j.xinn.2020.100017>
- Ziaf, K., Amjad, M., Pervez, M. A., Iqbal, Q., Rajwana, I. A., & Ayyub, M. (2009). Evaluation of different growth and physiological traits as indices of salt tolerance in hot pepper (*Capsicum annuum* L.). *Pakistan Journal of Botany*, 41(4), 1797-1809.