agronomy

MDPI

Article

Nitrogen Application Alleviates Impairments for *Jatropha curcas* L. Seedling Growth under Salinity Stress by Regulating Photosynthesis and Antioxidant Enzyme Activity

Zhao Yang [1,2,3], Shuai Tan [1,2,3,*], Qiliang Yang [1,2,3,*], Shaomin Chen [1,2,3], Changmin Qi [4], Xiaogang Liu [1,2,3], Jiaping Liang [1,2,3] and Haidong Wang [1,2,3]

1 Faculty of Modern Agricultural Engineering, Kunming University of Science and Technology, Kunming 650500, China; chenshaomin1989@163.com (S.C.)
2 Yunnan Provincial Field Scientific Observation and Research Station on Water-Soil-Crop System in Seasonal Arid Region, Kunming University of Science and Technology, Kunming 650500, China
3 Yunan Provincial Key Laboratory of High-Efficiency Water Use and Green Production of Characteristic Crops in Universities, Kunming University of Science and Technology, Kunming 650500, China
4 China Gezhouba Group Co., Ltd., Wuhan 430033, China
* Correspondence: tans90@163.com (S.T.); yangqilianglovena@163.com (Q.Y.)

Abstract: *Jatropha curcas* L. is a promising bioenergy source, and its seedling stage is sensitive to salinity. Nitrogen application presents an effective strategy for alleviating the adverse consequences of salinity stress. However, the responses of plant growth and physiology of *Jatropha curcas* L. seedlings to nitrogen application under salinity stress remain unclear. As a result, a one-year greenhouse plot experiment was conducted to investigate the effects of nitrogen application on the plant growth, antioxidant enzyme activity, and photosynthesis of *Jatropha curcas* L. seedlings under saline conditions. Experiment treatments consisted of three salinity stresses (mild salinity stress, S1: 2 g/kg; moderate salinity stress, S2: 4 g/kg; and severe salinity stress, S3: 6 g/kg), four nitrogen application rates (N0: 0 gN/plant; N1: 20 gN/plant; N2: 60 gN/plant; and N3: 100 gN/plant), and a control treatment (CK) which was without salinity stress and nitrogen application. The results showed that salinity stress substantially reduced plant growth of *Jatropha curcas* L. seedlings. As the salinity stress increased, the reduction in plant growth also increased. The S3 treatment had the lowest leaf area, leaf biomass, and total biomass, which decreased by an average of 70.4%, 66.3%, and 69.9%, respectively, compared to CK. Nitrogen application could compensate for these impairments of plant growth from salinity stress by promoting antioxidant enzyme activity and photosynthesis. As for mild and moderate salinity stresses, the maximum plant growth was found in the N3 treatment, with the maximum antioxidant enzyme activity, photosynthetic pigment, photosynthetic characteristic, and chlorophyll fluorescence. As for severe salinity stress, higher plant growth was found in N2 and N3 treatments, and there were no significant differences between N2 and N3 treatments. It also should be noted that the maximum photosynthetic characteristic and chlorophyll fluorescence were found in N2 treatment under severe salinity stress. In conclusion, nitrogen application could be an alternative strategy to improve the salinity tolerance of *Jatropha curcas* L. growth. The nitrogen application rate of 100 gN/plant could be recommended for low and moderate salinity stresses, while 60 gN/plant could be recommended for severe salinity stress. However, higher nitrogen application rate (>100 gN/plant) under mild and moderate salinity stress and the effects of reactive oxygen species under salinity stress should be further evaluated.

Keywords: salinity stress; nitrogen application; *Jatropha curcas* L.; antioxidant enzyme activity; photosynthesis

Citation: Yang, Z.; Tan, S.; Yang, Q.; Chen, S.; Qi, C.; Liu, X.; Liang, J.; Wang, H. Nitrogen Application Alleviates Impairments for *Jatropha curcas* L. Seedling Growth under Salinity Stress by Regulating Photosynthesis and Antioxidant Enzyme Activity. *Agronomy* **2023**, *13*, 1749. https://doi.org/10.3390/agronomy13071749

Academic Editor: Junfei Gu

Received: 17 May 2023
Revised: 21 June 2023
Accepted: 25 June 2023
Published: 28 June 2023

1. Introduction

Jatropha curcas L., a promising renewable bioenergy source, has been extensively cultivated in tropical and subtropical regions [1], due to its adaptability in stressful en-

vironments [2,3]. This species can survive in infertile and degraded soils [4], such as arid, saline-alkali, and erosion lands, consequently helping to restore vegetation, improve soil salinization, and prevent soil erosion [1,5,6]. As the climate characteristic of *Jatropha curcas* L. cultivation, it is essential to address soil salinity and plant salinity tolerance [7]. As *Jatropha curcas* L. seedlings are sensitive to salinity stress [8–10], soil salinity causes a reduction in water content and water potential of tissues, resulting in a significant reduction in stem and root elongation, leaf expansion, and biomass accumulation in seedlings [11]. When soil salinity reaches 0.6 g/kg, *Jatropha curcas* L. seedling occurs damage [9], while soil salinity of 5–10.9 g/kg significantly reduced growth and photosynthesis by over 50%, resulting in a significant decrease in biomass [12]. In addition, soil salinity (0.6–2.4 g/kg) had a negative impact on the total biomass production and photosynthetic activity and pigments of *Jatropha curcas* L. [9,13,14], whereas it elevated the activity of antioxidant enzymes [8]. Salinity can also impair the effectiveness of nutrients in plants, as well as their absorption, photosynthetic performance, leaf chlorophyll fluorescence parameters, and distribution of nutrients, causing nutrient imbalance [15–17]. Patel et al. [11] noted that salinity stress inhibited the uptake of nutrients such as nitrogen, calcium, and potassium. Therefore, understanding the responses of plant growth and physiological ecology in *Jatropha curcas* L. seedlings to salinity stress is crucial for mitigating the risk caused by salinity stress and developing effective strategies to maintain productivity under these conditions.

Salinity affects carbon and nitrogen metabolisms in plants, which are fundamental physiological processes for plant growth and development [15]. The interaction between salinity and nitrogen is crucial in determining the photosynthetic potential of plants and their salinity tolerance [15,18]. Nitrogen application appears to be an effective strategy to reduce the impairment of salinity stress on plant growth by regulating photosynthesis and enzyme activity [15,19]. Notably, nitrogen is the nutrient element required in the greatest quantity by plants. Most of the nitrogen in leaves is utilized for synthesizing photosynthetic organs [20], and nitrogen application can simulate the synthesis of photosynthetic pigments [21]. Generally, a higher photosynthetic rate is associated with a higher leaf nitrogen content [22], and there is a significant positive correlation between the photosynthesis of plant leaves and nitrogen content [23]. According to Iqbal et al. [15], nitrogen could protect photosynthesis against salinity stress. Moreover, an elevated nitrogen level (20 mM N) has beneficial effects on salinity-stressed plants by modulating nitrogen availability, which regulates ethylene levels and proline production to influence the response of photosynthesis and plant growth. In addition, nitrogen is an essential component of leaf enzymes. Nitrogen fertilizer application can increase the activity of antioxidant enzymes in plants [24], thereby helping in the resistance to oxidative damage caused by the accumulation of a large number of reactive oxygen species (ROS) under salinity stress [25]. Singh et al. [19] found that exogenous nitrogen supplementation could mitigate NaCl-induced toxicity in *Solanum lycopersicum* seedlings by regulating ROS, enhancing the ratio of K^+/Na^+, with a nitrogen application rate of 270 mg/kg, producing maximum growth. Conversely, excessive nitrogen application can reduce soil porosity, hinder plant growth, and even cause agricultural non-point source pollution. Therefore, the appropriate nitrogen application rate should be considered comprehensively based on soil conditions and plant growth status. However, the response of plant growth, photosynthesis, and antioxidant enzymes of *Jatropha curcas* L. seedlings to nitrogen application under salinity stress, as well as the contribution of nitrogen application to alleviating salinity stress, remain uncertain.

Based on this, a glasshouse experiment was conducted with one-year-old *Jatropha curcas* L. seedlings. Analytical pure NaCl mixed with air-dry soil was used to simulate a soil salinity stress environment. The purposes of this study were to: (1) determine the effects of salinity stress and nitrogen application rate on the growth of *Jatropha curcas* L. seedlings; (2) explore whether nitrogen application can alleviate the impairment caused by salinity stress; and (3) recommend the nitrogen application rate under salinity stress for *Jatropha curcas* L. seedlings.

2. Materials and Methods

2.1. Experimental Site Descriptions

A one-year glasshouse experiment was conducted from 1 May to 30 August 2021 at the Faculty of Modern Agricultural Engineering of Kunming University of Science and Technology (24°9′ N, 102°79′ E, 1979 m a.s.l.). During the experimental period, the maximum and minimum ambient temperatures in the glasshouse were 28.0 °C and 17.5 °C, respectively, with an average relative humidity of 68%. The red soil with an average bulk density of 1.25 g/cm^3 and a field capacity of 0.350 cm^3/cm^3 was used for the experimental soil. The contents of organic matter (13.08 g/kg), total nitrogen (0.87 g/kg), total phosphorus (0.66 g/kg), total potassium (13.9 g/kg), and soil pH (5.0–5.5) were measured. One-year-old *Jatropha curcas* L. seedlings were used in the experiment.

2.2. Experimental Design

The experiment was conducted at the beginning of leaf extension in the *Jatropha curcas* L. seedlings (approximately 30 days after transplanting) from 1 May to 30 August 2021, for a total of 122 days. Four nitrogen application rates (N0: 0 gN/plant, N1: 20 gN/plant, N2: 60 gN/plant, and N3: 100 gN/plant) and three salinity stress levels (mild salinity stress, S1: 2 g/kg; moderate salinity stress, S2: 4 g/kg; and severe salinity stress, S3: 6 g/kg) were arranged with the treatment without salinity stress and nitrogen application serving as a control (CK). Consequently, there were 13 treatments in total, and each treatment had six replicates, as detailed in Table 1. The soil was air-dried and passed through a 2 mm sieve before filling the plots with a volume of 6 m × 0.55 m × 0.65 m. The air-dry soil was mixed with different rates of purified NaCl to simulate salinity stress according to the mass ratio specified in the experiment design (Table 1). The *Jatropha curcas* L. seedlings were planted at a spacing of 1 m intervals in each plot with a total of six plants per plot. Urea containing 46.3% N was used as nitrogen fertilizer, derived from Sichuan Meifeng Chemical Industry Co., Ltd., (Deyang, Sichuan, China). During the experimental period, the urea was applied twice in equal quantities, i.e., at the beginning of the experiment (1 DAE) and 60 days after the experiment (60 DAE) using a ring method with a 15 cm radius and a 20 cm depth. The irrigation water was derived from tap water with a salinity of 260 mg/L. Irrigation was triggered when the soil water content fell below 75% of field capacity (0.263 cm^3/cm^3) and was terminated when the soil water content approached field capacity (0.350 cm^3/cm^3). Other field management practices, such as weed control and pesticide application, were the same for each treatment.

Table 1. The salinity stress and nitrogen application treatments arrangements during the experimental period.

Treatments	Soil Salinity (g/kg)	Nitrogen Application Rate (gN/Plant)		
		1 DAE	60 DAE	Total
CK	0	0	0	0
S1N0	2	0	0	0
S1N1		10	10	20
S1N2		30	30	60
S1N3		50	50	100
S2N0	4	0	0	0
S2N1		10	10	20
S2N2		30	30	60
S2N3		50	50	100
S3N0	6	0	0	0
S3N1		10	10	20
S3N2		30	30	60
S3N3		50	50	100

Note: DAE indicates the day after the experiment.

2.3. Data Collection

2.3.1. Plant Biomass and Leaf Area Measurements

At the end of the experiment (122 DAE), three plants for each plot were randomly collected, cut, and separated into root, stem, and leaf sections. The separate parts were oven-dried at 105 °C for 30 min to halt the metabolic processes and then dried at 75 °C until a constant weight was achieved. Next, leaf and total biomass were determined using an electronic balance with a precision of 0.01 g. The leaf area was estimated by using the gravimetric method. The total leaf area was calculated by adding the areas of all fully expanded green leaves.

2.3.2. Antioxidant Enzyme Activity Measurements

At 15 days after the second nitrogen application (75 DAE), the antioxidant enzyme activity in the leaves for each treatment was determined. Fresh leaves were added to a chilled phosphate-buffered solution with a pH of 7.0, ground into a homogenate, and centrifuged at 10,000 r/min for 10 min at a temperature of 4 °C. The supernatant was used to measure antioxidant enzyme activity, including peroxidase (POD), superoxide dismutase (SOD), and catalase (CAT). The POD activity was measured using the guaiacol method, SOD activity was measured using the nitrogen blue tetrazole method, and CAT activity was measured using the colorimetric method [26,27].

2.3.3. Photosynthetic Pigment Contents Measurements

Similar to antioxidant enzyme activity measurement, the leaf photosynthetic pigment contents and photosynthetic characteristics of *Jatropha curcas* L. seedlings were determined at 75 DAE. For each plot, five leaves were collected between 9:00 and 10:00 a.m. to measure photosynthetic pigment contents. Chlorophyll a (Chl a), chlorophyll b (Chl b), and carotenoid (Car) were determined by using the ultraviolet spectrophotometer method.

2.3.4. Photosynthetic Characteristics and Chlorophyll Fluorescence Measurement

The net photosynthetic rate (P_n), stomatal conductance (G_s), intercellular CO_2 concentration (C_i), and transpiration rate (T_r) were measured using a portable photosynthesis measurement system (LI-6400, Li-Cor, Inc., Lincoln, NE, USA). For each plot, five fully extended leaves were selected and measured between 9:00 to 11:00 a.m. on a clear sunny day with three replicates under photosynthetically active radiation of 1200 μmol/m^2/s. The CO_2 concentration in the leaf chamber was fixed at 380 μmol/mol.

The chlorophyll fluorescence of the same leaves was measured by using a MINI-PAM portable chlorophyll fluorometer (Heinz WAZL GmbH, Effeltrich, Germany). The maximum photochemical quantum yield of PSII (F_v/F_m), the effective photochemical quantum yield of PSII (Φ_{PSII}), coefficient of photochemical fluorescence quenching (qP), Stern–Volmer-type non-photochemical fluorescence quenching (NPQ), and apparent electron transport rate (ETR) were obtained.

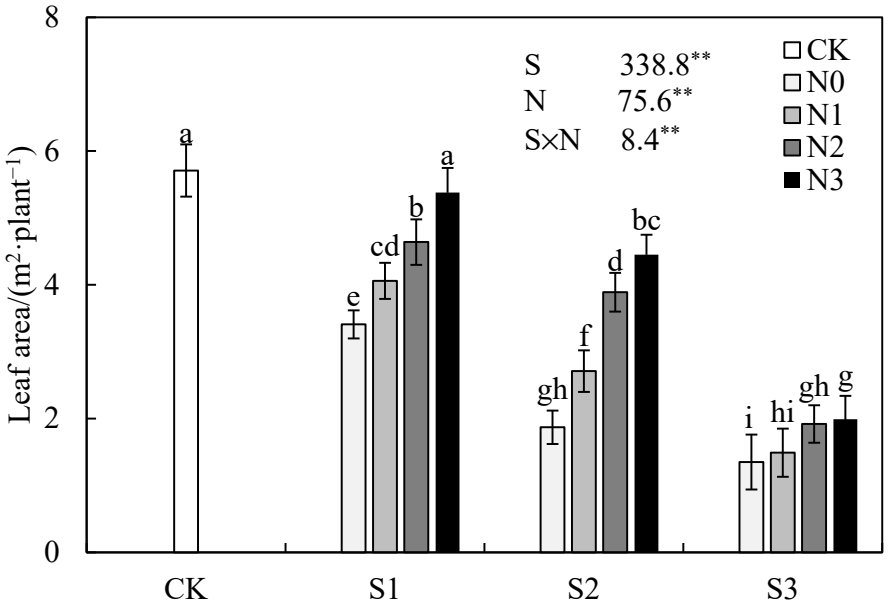
2.4. Statistical Analysis

Statistical analysis was carried out using the SPSS (Statistical Product and Service Solutions) software (version 21.0, IBM Corporation, Endicott, NY, USA). A two-way analysis of variance (ANOVA) at $\alpha = 0.05$ and $\alpha = 0.01$ was used to determine the significance of the effects of salinity stress, nitrogen application rate, and their interaction. When ANOVA results were significant ($p < 0.05$), Duncan's test was conducted at $\alpha = 0.05$ to compare the differences in the plant biomass, leaf area, antioxidant enzyme activity, photosynthetic pigment content, photosynthetic characteristics, and chlorophyll fluorescence parameters among all treatments. All data were shown as the mean ± standard deviation.

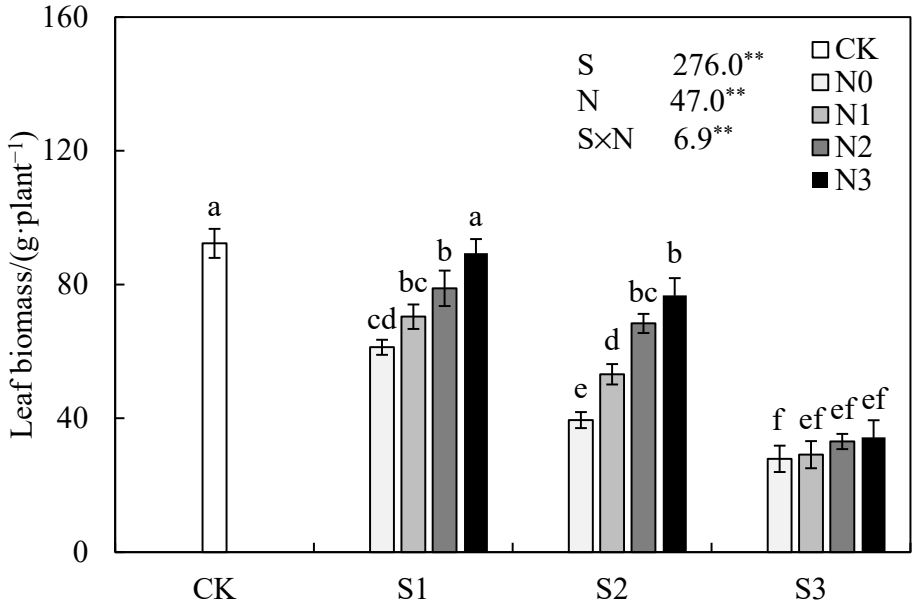
3. Results

3.1. Effect of Nitrogen Application Rate on Jatropha curcas L. Growth under Salinity Stress

Salinity stress, nitrogen application, and their interaction had a significant impact on the growth of *Jatropha curcas* L. seedlings ($p < 0.01$, Figure 1). Increasing salinity stress caused substantial reductions in plant growth. Compared to CK, S1–S3 treatments decreased leaf area by an average of 23.5–70.4%, leaf biomass by 18.8–66.3%, and total biomass by 24.1–69.9%, respectively. Furthermore, leaf area, leaf biomass, and total biomass exhibited a negative linear correlation with increases in salinity stress. The S3 treatment had the minimum leaf area, leaf biomass, and plant biomass.

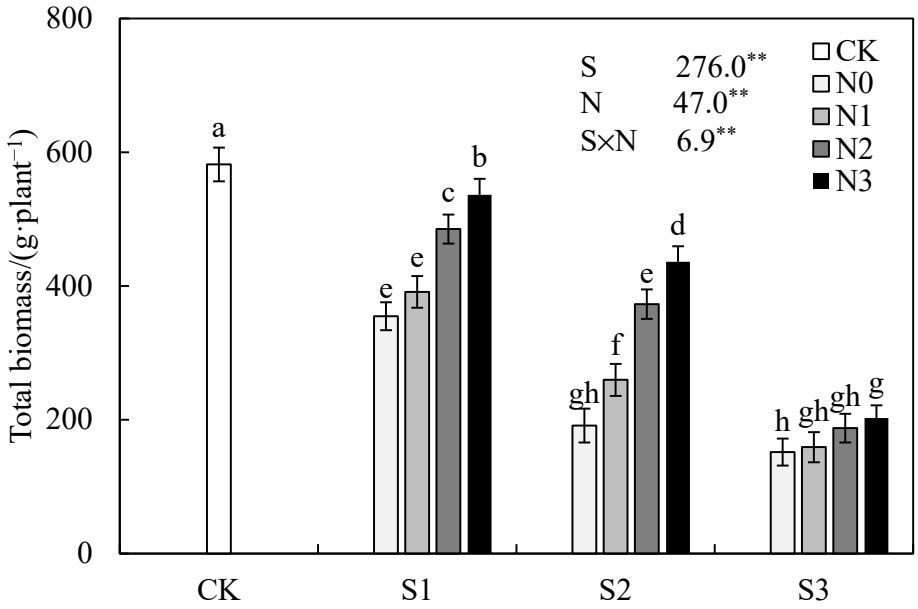


(**a**)



(**b**)

Figure 1. *Cont.*



(**c**)

Figure 1. Effects of nitrogen application rate on leaf area (**a**), leaf biomass (**b**), and total biomass (**c**) of *Jatropha curcas* L. seedlings under various salinity stresses. The error bars represent standard deviations. Different lowercase letters above the bars indicate significant differences among treatments at $p < 0.05$. The number in the figure represents the F value. ** represents $p < 0.01$. S1, S2, and S3 represent mild, moderate, and severe salinity stress, respectively. N0, N1, N2, and N3 represent the nitrogen application rate of 0, 20, 60, and 100 gN/plant, respectively.

At the same salinity stress, leaf area, leaf biomass, and total biomass increased with the increasing nitrogen application rate. On average, in comparison to the N0 treatment, N1, N2, and N3 treatments increased leaf area by 24.4%, 57.5%, and 78.3%, leaf biomass by 18.8%, 40.2%, and 55.8%, total biomass by 16.0%, 49.8%, and 68.3%, respectively. As for mild and moderate salinity stress, N3 treatment showed significantly higher plant growth. As for the severe salinity stress, there were no significant differences between N2, N1, and N3 treatments ($p > 0.05$). These results indicate that nitrogen application could reduce the adverse influence of salinity stress on the growth of *Jatropha curcas* L. seedlings, although it may be difficult to achieve equivalent compensation, especially under severe salinity stress.

3.2. Effect of Nitrogen Application Rate on Antioxidant Enzyme Activity of Jatropha curcas L. Seedlings under Salinity Stress

The POD, SOD, and CAT activities in S1–S3 treatments were significantly greater than those in CK ($p < 0.01$, Table 2), with an average increase of 54.2–104.1%, 80.6–151.6%, and 31.3–66.7%, respectively. There were no significant differences in SOD and CAT between S3N0 and CK ($p > 0.05$), and SOD in S3N0 was slightly lower than that in CK. In addition, the POD, SOD, and CAT activities represented a single peak curve trend with salinity stress and reached peak value at S2. This demonstrates that salinity stress could enhance the antioxidant enzyme activity of *Jatropha curcas* L. seedlings, but severe salinity stress could inhibit this promotion.

Additionally, nitrogen application also had a positive influence on POD, SOD, and CAT activities under salinity stress ($p < 0.01$, Table 2). Compared to the N0 treatment, N1, N2, and N3 treatments increased POD activity by 35.0%, 71.3%, and 105.6%, SOD activity by 35.2%, 102.9%, and 160.6%, and CAT activity by 19.5%, 46%, and 67.4%, respectively. It can be seen that the antioxidant enzyme activities increased gradually as the nitrogen application rate increased.

Table 2. Effects of nitrogen application rate on POD, SOD, and CAT activities of *Jatropha curcas* L. seedlings under various salinity stresses.

Treatments	POD/(U·g^{-1}·min^{-1})	SOD/(U·g^{-1}·min^{-1})	CAT/(U·g^{-1}·min^{-1})
CK	83.9 ± 1.0 ^{k}	20.4 ± 0.8 ^{k}	60.3 ± 1.2 ^{l}
S1N0	85.4 ± 1.0 ^{k}	23.2 ± 0.3 ^{j}	68.0 ± 0.6 ^{k}
S1N1	115.5 ± 0.5 ^{h}	29.8 ± 0.3 ^{i}	78.0 ± 0.8 ^{h}
S1N2	144.9 ± 0.6 ^{f}	48.2 ± 0.2 ^{e}	92.1 ± 0.6 ^{e}
S1N3	176.4 ± 0.6 ^{c}	67.3 ± 0.3 ^{b}	108.9 ± 0.4 ^{c}
S2N0	107.3 ± 0.6 ^{i}	31.2 ± 0.2 ^{h}	71.1 ± 0.7 ^{j}
S2N1	148.7 ± 0.5 ^{e}	40.1 ± 0.3 ^{g}	87.1 ± 0.9 ^{f}
S2N2	198.1 ± 0.8 ^{b}	59.4 ± 0.3 ^{c}	116.1 ± 0.7 ^{b}
S2N3	231.1 ± 0.7 ^{a}	74.2 ± 0.6 ^{a}	128.1 ± 0.6 ^{a}
S3N0	89.3 ± 0.4 ^{j}	20.1 ± 0.3 ^{k}	61.0 ± 1.1 ^{l}
S3N1	116.4 ± 0.6 ^{h}	30.8 ± 0.3 ^{h}	74.0 ± 0.6 ^{i}
S3N2	139.9 ± 0.6 ^{g}	43.5 ± 0.2 ^{f}	84.0 ± 0.5 ^{g}
S3N3	172.3 ± 0.7 ^{d}	52.6 ± 0.7 ^{d}	97.9 ± 0.6 ^{d}
Significant of factors (F-value)			
Salinity stress (S)	5452 **	2252 **	943 **
Nitrogen application (N)	13,188 **	9947 **	2349 **
S × N	270 **	138 **	76 **

Note: Means followed by different lowercase letters within the same column indicate significant differences among treatments at $p < 0.05$ using Duncan's test. ** represents $p < 0.01$. S1, S2, and S3 represent mild, moderate, and severe salinity stress, respectively. N0, N1, N2, and N3 represent the nitrogen application rate of 0, 20, 60, and 100 gN/plant, respectively.

3.3. Effect of Nitrogen Application Rate on the Photosynthetic Pigment of Jatropha curcas L. Seedlings under Salinity Stress

Significant differences in Chl a, Chl b, Chl a+b, Chl a/b ratio, and Car were observed among all treatments ($p < 0.01$, Table 3). Generally, salinity stress significantly reduced these five leaf photosynthetic pigments. On average, S1, S2, and S3 treatments decreased Chl a by 8.5%, 25.2%, and 37.3%, Chl b by 7.7%, 29.1%, and 44.6%, Chl a+b by 7.9%, 26.7%, and 39.6%, Chl a/b ratio by 0.9%, 6.1%, and 14.2%, and Car by 0.2%, 20.8%, and 35.0%, respectively. Similarly to plant growth, a negative linear relationship was found between salinity stress and Chl a, Chl b, Chl a+b, and Car, while there was a positive linear relationship with the Chl a/b ratio.

In contrast, nitrogen application significantly increased the photosynthetic pigment under salinity stress. The trend for Chl a, Chl b, Chl a+b, and Car was N3 > N2 > N1 > N0. In comparison to the N0 treatment, N1, N2, and N3 treatments increased Chl a by 11.4%, 23.6%, and 34.3%, Chl b by 19.3%, 45.6%, and 63.2%, Chl a+b content by 14.3%, 30.6%, and 43.4%, and Car by 17.5%, 40.0%, and 57.5%, respectively. It should be noted that greater leaf photosynthetic pigment was observed with higher nitrogen application rates under mild salinity stress. S1N3 had maximum Chl a, Chl b, Chl a+b, and Car, even exceeding those in CK by 5.5%, 16.1%, 9.1%, and 24.4%, respectively. As for moderate and severe salinity stresses, N3 treatment also had maximum Chl a, Chl b, Chl a+b, and Car. Additionally, the Chl a, Chl b, Chl a+b, and Car demonstrated a positive linear correlation with the nitrogen application rate under various salinity stresses, whereas the Chl a/b ratio exhibited the opposite relationship.

Table 3. Effect of nitrogen application on leaf photosynthetic pigments of *Jatropha curcas* L. seedlings under various salinity stresses.

Treatments	Chl a/(mg/g FW)	Chl b/(mg/g FW)	Chl a+b/(mg/g FW)	Chl a/b ratio	Car/(mg/g FW)
CK	2.15 ± 0.21 ^{b}	1.03 ± 0.02 ^{b}	3.18 ± 0.11 ^{a}	2.09 ± 0.13 ^{j}	0.63 ± 0.01 ^{c}
S1N0	1.74 ± 0.00 ^{e}	0.72 ± 0.01 ^{f}	2.46 ± 0.01 ^{f}	2.43 ± 0.01 ^{c}	0.51 ± 0.00 ^{e}

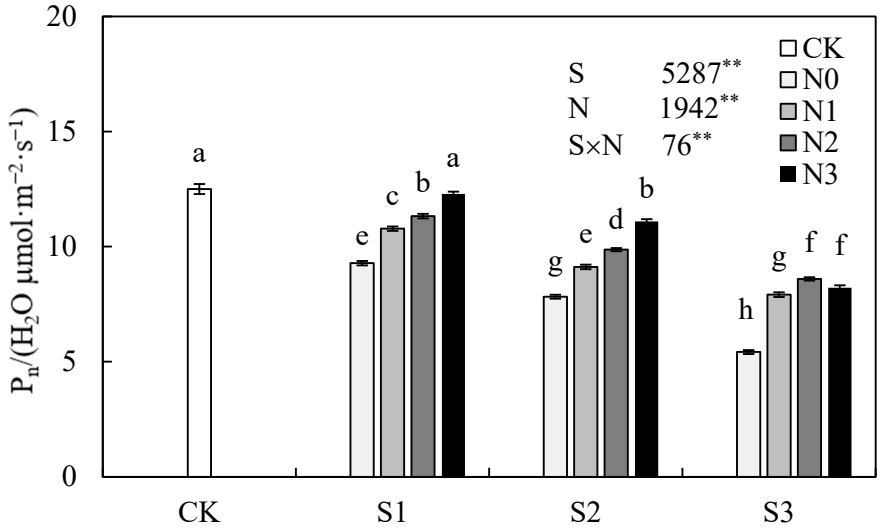
Table 3. *Cont.*

Treatments	Chl a/(mg/g FW)	Chl b/(mg/g FW)	Chl a+b/(mg/g FW)	Chl a/b ratio	Car/(mg/g FW)
S1N1	1.81 ± 0.02 ^{c}	0.83 ± 0.00 ^{d}	2.65 ± 0.02 ^{e}	2.18 ± 0.01 ^{i}	0.56 ± 0.01 ^{e}
S1N2	2.06 ± 0.01 ^{c}	1.07 ± 0.01 ^{b}	3.14 ± 0.02 ^{c}	1.93 ± 0.01 ^{l}	0.67 ± 0.01 ^{b}
S1N3	2.27 ± 0.00 ^{a}	1.20 ± 0.01 ^{a}	3.47 ± 0.01 ^{b}	1.90 ± 0.01 ^{l}	0.79 ± 0.01 ^{a}
S2N0	1.41 ± 0.01 ^{j}	0.59 ± 0.01 ^{j}	2.00 ± 0.01 ^{k}	2.38 ± 0.02 ^{d}	0.41 ± 0.00 ^{j}
S2N1	1.49 ± 0.01 ^{h}	0.66 ± 0.01 ^{h}	2.15 ± 0.01 ^{i}	2.26 ± 0.02 ^{f}	0.44 ± 0.01 ^{i}
S2N2	1.69 ± 0.01 ^{f}	0.77 ± 0.01 ^{e}	2.46 ± 0.01 ^{g}	2.20 ± 0.01 ^{h}	0.53 ± 0.00 ^{f}
S2N3	1.83 ± 0.01 ^{d}	0.89 ± 0.00 ^{c}	2.72 ± 0.02 ^{d}	2.05 ± 0.01 ^{k}	0.61 ± 0.01 ^{d}
S3N0	1.03 ± 0.01 ^{l}	0.40 ± 0.00 ^{l}	1.43 ± 0.02 ^{m}	2.61 ± 0.01 ^{a}	0.27 ± 0.00 ^{k}
S3N1	1.36 ± 0.01 ^{k}	0.55 ± 0.00 ^{k}	1.91 ± 0.01 ^{l}	2.48 ± 0.02 ^{b}	0.40 ± 0.00 ^{j}
S3N2	1.45 ± 0.01 ^{i}	0.64 ± 0.01 ^{i}	2.09 ± 0.01 ^{j}	2.26 ± 0.02 ^{e}	0.47 ± 0.01 ^{h}
S3N3	1.54 ± 0.01 ^{g}	0.69 ± 0.01 ^{g}	2.23 ± 0.01 ^{h}	2.23 ± 0.02 ^{g}	0.50 ± 0.01 ^{g}
Significant of factors (F-value)					
Salinity stress (S)	4219 **	3306 **	1734 **	815 **	1262 **
Nitrogen application (N)	1419 **	1640 **	359 **	426 **	773 **
N × S	54 **	76 **	22 **	37 **	24 **

Note: Means followed by different lowercase letters within the same column indicate significant differences among treatments at $p < 0.05$ using Duncan's test. ** represents $p < 0.01$. S1, S2, and S3 represent mild, moderate, and severe salinity stress, respectively. N0, N1, N2, and N3 represent the nitrogen application rate of 0, 20, 60, and 100 gN/plant, respectively.

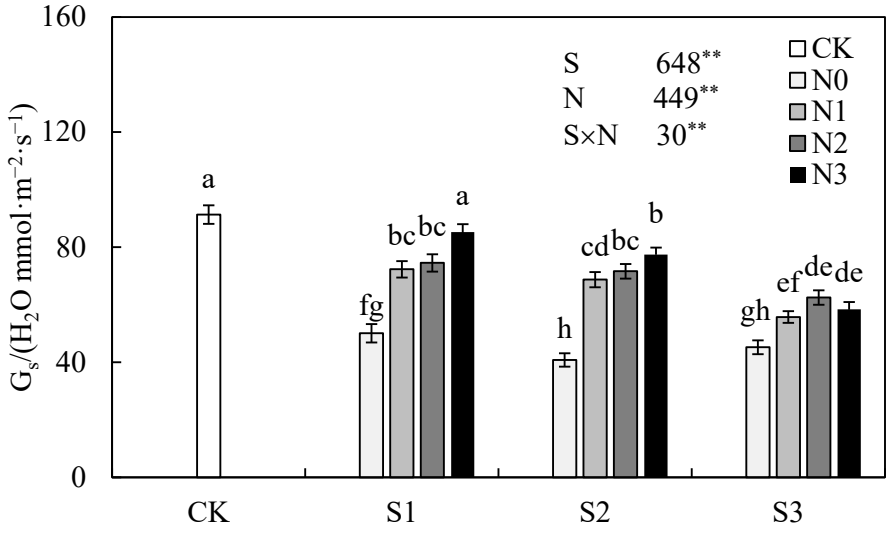
3.4. Effect of Nitrogen Application Rate on Leaf Photosynthetic Characteristic of Jatropha curcas L. Seedlings under Salinity Stress

As expected, salinity stress, nitrogen application, and their interaction exhibited significant effects on P_n, G_s, C_i, and T_r ($p < 0.01$, Figure 2). Salinity stress dramatically reduced leaf photosynthesis, showing that P_n, G_s, C_i, and T_r in S1–S3 treatments were decreased by an average of 12.7–39.7%, 22.8–39.3%, 6.7–20.6%, and 20.1–31.5%, respectively, relative to CK. As salinity stress increased, P_n, G_s, and T_r gradually decreased, while C_i declined initially and then rose. The minimum P_n, G_s, and T_r were observed in the S3 treatment, whereas the minimum C_i was found in the S2 treatment.

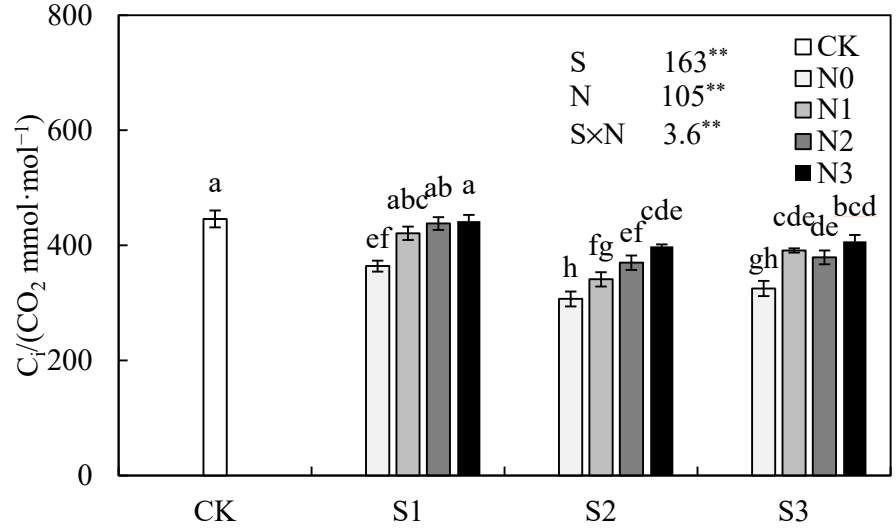


(**a**)

Figure 2. *Cont.*

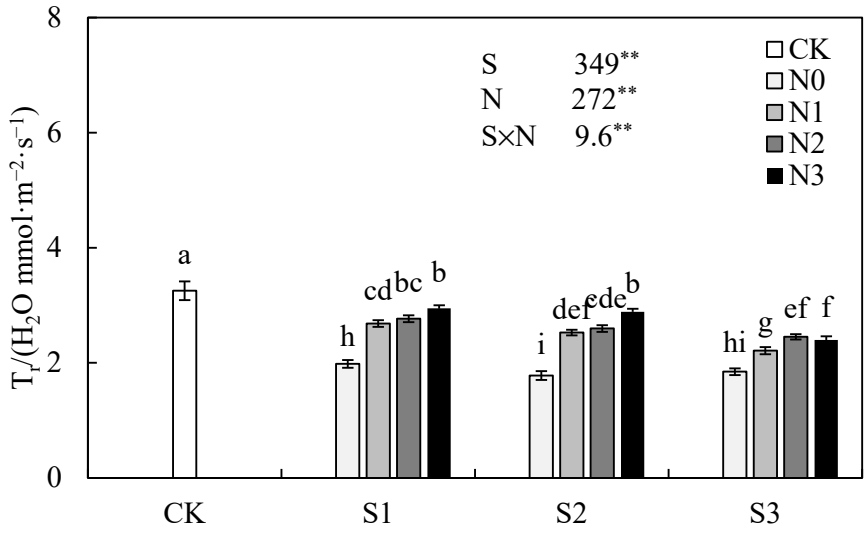


(**b**)



(**c**)

Figure 2. *Cont*.



(**d**)

Figure 2. Effects of nitrogen application rate on net photosynthetic rate (**a**), stomatal conductance (**b**), intercellular CO_2 concentration (**c**), and transpiration rate (**d**) under various salinity stresses. The error bars represent standard deviations. Different lowercase letters above the bars indicate significant differences among treatments at $p < 0.05$. The number in the figure represents the F value. ** represents $p < 0.01$. S1, S2, and S3 represent mild, moderate, and severe salinity stress, respectively. N0, N1, N2, and N3 represent the nitrogen application rate of 0, 20, 60, and 100 gN/plant, respectively.

Nitrogen application could substantially mitigate the adverse impacts of salinity stress. Generally, N3 treatment had the greatest promotion of leaf photosynthesis under mild and moderate salinity stresses. As for mild salinity stress, N3 treatment increased P_n, G_s, C_i, and T_r by an average of 32.4%, 70.2%, 21.4%, and 25.2%, respectively. As for moderate salinity stress, N3 treatment enhanced P_n, G_s, C_i, and T_r by an average of 41.8%, 89.8%, 29.7%, and 62.2%, respectively. As for severe salinity stress, the maximum P_n, G_s, and T_r were observed in the N2 treatment, and the maximum C_i was found in the N3 treatment. It should be noted that there were no significant differences in P_n, G_s, C_i, and T_r between N2 and N3 under severe salinity stress. These results demonstrate that the promotion of photosynthesis may not be significant with a higher nitrogen application under severe salinity stress.

3.5. Effect of Nitrogen Application Rate on Chlorophyll Fluorescence of Jatropha curcas L. Seedlings under Salinity Stress

Both salinity stress and nitrogen application significantly influenced leaf chlorophyll fluorescence parameters, including F_v/F_m, Φ_{PSII}, qP, NPQ, and ETR (Table 4, $p < 0.05$). Their interaction did not significantly affect F_v/F_m and Φ_{PSII}. Generally, the differences in F_v/F_m among all treatments and CK were insignificant ($p > 0.05$), ranging from 0.749 to 0.818, with the exception of S3N2, which was significantly lower than CK ($p < 0.05$). In addition, all treatments had lower Φ_{PSII}, qP, and ETR than CK, by an average of 2.9–16.1%, 2.3–17.3%, and 1.4–41.2%, respectively. Furthermore, most of treatments displayed a significant negative effect on NPQ ($p < 0.05$). Notably, NPQ for S1N0, S2N0, S3N0, and S3N1 was higher than CK by 3.3%, 8.8%, 16.7%, and 1.3%, respectively, and there were no significant differences between S1N0, S3N1, and CK ($p > 0.05$). Overall, F_v/F_m, Φ_{PSII}, qP, and ETR declined linearly with increasing salinity stress, while NPQ increased.

Table 4. Effect of nitrogen application on chlorophyll fluorescence of *Jatropha curcas* L. seedlings under various salinity stresses.

Treatments	Fv/Fm	Φ_{PSII}	qP	NPQ	ETR
CK	0.781 ± 0.011 ^{ab}	0.521 ± 0.014 ^{a}	0.751 ± 0.012 ^{a}	1.473 ± 0.016 ^{c}	199.3 ± 5.4 ^{a}
S1N0	0.777 ± 0.009 ^{abc}	0.449 ± 0.008 ^{ef}	0.651 ± 0.004 ^{fg}	1.522 ± 0.011 ^{c}	142.3 ± 4.4 ^{g}
S1N1	0.802 ± 0.005 ^{a}	0.485 ± 0.007 ^{abc}	0.714 ± 0.005 ^{ab}	1.034 ± 0.013 ^{i}	177.8 ± 3.3 ^{bc}
S1N2	0.809 ± 0.007 ^{a}	0.489 ± 0.007 ^{abc}	0.720 ± 0.005 ^{ab}	0.913 ± 0.006 ^{j}	185.0 ± 3.5 ^{ab}
S1N3	0.818 ± 0.008 ^{a}	0.506 ± 0.010 ^{a}	0.734 ± 0.009 ^{a}	0.807 ± 0.012 ^{k}	196.5 ± 1.8 ^{a}
S2N0	0.774 ± 0.008 ^{abc}	0.441 ± 0.010 ^{f}	0.638 ± 0.006 ^{gh}	1.603 ± 0.013 ^{b}	131.4 ± 3.78 ^{h}
S2N1	0.793 ± 0.005 ^{abc}	0.475 ± 0.006 ^{bcd}	0.691 ± 0.006 ^{cd}	1.314 ± 0.011 ^{f}	164.4 ± 2.9 ^{de}
S2N2	0.799 ± 0.004 ^{ab}	0.481 ± 0.007 ^{abcd}	0.703 ± 0.007 ^{bc}	1.281 ± 0.010 ^{g}	170.2 ± 5.0 ^{cd}
S2N3	0.813 ± 0.005 ^{a}	0.495 ± 0.011 ^{ab}	0.727 ± 0.009 ^{a}	1.197 ± 0.006 ^{h}	191.9 ± 5.4 ^{a}
S3N0	0.752 ± 0.005 ^{bc}	0.437 ± 0.004 ^{f}	0.621 ± 0.004 ^{h}	1.719 ± 0.011 ^{a}	117.2 ± 3.7 ^{i}
S3N1	0.782 ± 0.004 ^{abc}	0.455 ± 0.006 ^{def}	0.663 ± 0.007 ^{ef}	1.492 ± 0.009 ^{c}	149.6 ± 3.7 ^{fg}
S3N2	0.749 ± 0.046 ^{c}	0.467 ± 0.009 ^{cde}	0.683 ± 0.008 ^{d}	1.397 ± 0.009 ^{d}	159.2 ± 2.6 ^{def}
S3N3	0.784 ± 0.005 ^{abc}	0.463 ± 0.011 ^{cdef}	0.672 ± 0.006 ^{de}	1.352 ± 0.008 ^{e}	153.4 ± 3.4 ^{efg}
Significant of factors (F-value)					
Salinity stress (S)	6.5 **	10.9 **	52.0 **	1722 **	68.4 **
Nitrogen application (N)	3.6 *	17.8 **	80.4 **	1342 **	103.24 **
S × N	0.48 ^{NS}	0.80 ^{NS}	2.31 **	69.4 *	2.99 **

Note: Means followed by different lowercase letters within the same column indicate significant differences among treatments at $p < 0.05$ using Duncan's test. * represents $p < 0.05$, ** represents $p < 0.01$, and ^{NS} represents $p > 0.05$. S1, S2, and S3 represent mild, moderate, and severe salinity stress, respectively. N0, N1, N2, and N3 represent the nitrogen application rate of 0, 20, 60, and 100 gN/plant, respectively.

By contrast, in comparison to the N0 treatment, N1–N3 treatments increased F_v/F_m, Φ_{PSII}, qP, and ETR by an average of 2.6–5.2%, 6.8–11.4%, 7.8–10.9%, and 25.8–38.6%, respectively, while NPQ reduced by 20.5–30.4%. The maximum F_v/F_m, Φ_{PSII}, qP, and ETR, and minimum NPQ were found in S1N3. As the nitrogen application rate increased, F_v/F_m, Φ_{PSII}, qP, and ETR also increased linearly, whereas NPQ decreased. It is worth noting that when *Jatropha curcas* L. seedlings were subjected to severe salinity stress, F_v/F_m, Φ_{PSII}, qP, and ETR in N1–N3 were considerably higher than those in the N0 ($p < 0.05$), but there were no significant differences among N1–N3. This demonstrates that nitrogen application could inhibit the negative effects of salinity stress on photosynthesis and contribute to compensation under severe salinity stress, but higher nitrogen application did not have a clearly positive influence.

4. Discussion

Soil salinity is an essential environmental factor affecting plant growth. Numerous studies have demonstrated that salinity stress can inhibit plant development and photosynthesis. Typically, a decline in biomass is the most visible symptom of salinity stress in plants. In the present study, similar results were found. Salinity stress reduced the leaf area, leaf biomass, and total biomass of *Jatropha curcas* L. seedlings (Table 2). This may be attributed to plant transpiration and photosynthesis impairment from leaf growth inhibition and stomata function deceleration. Moreover, nitrogen application appeared to promote leaf growth and biomass accumulation of *Jatropha curcas* L. seedlings, which is in agreement with the effect of nitrogen application on the growth of wheat and tomato seedlings under salinity stress conditions [28,29]. Indeed, appropriate nitrogen application could induce salinity tolerance as well as stimulate plant growth and biomass accumulation.

The reduction in photosynthesis is a primary cause of low plant biomass under salinity stress [9,11,12]. Photosynthetic pigments offer a direct description of plant growth and development status, as well as the intensity of photosynthesis [30]. In this study, photosynthetic pigments of *Jatropha curcas* L. seedlings decreased with the increase in salinity stress (Table 3). This observation could be caused by salinity-induced increased soil

pH promoting chlorophyll degrading enzyme activity (such as chlorophyllase), ultimately leading to a breakdown in chlorophyll [31]. A higher chlorophyll a/b ratio signifies greater peroxidation activity within leaf membranes [32]. Consequently, under salinity stress, this ratio increases, but nitrogen application alleviates this stress and enhances the chlorophyll a/b ratio. The research findings of Liu et al. on the impact of salt stress on the physiological characteristics of pomegranates align with the results of this experiment [33]. Furthermore, the presence of ROS could trigger oxidative damage to plants, resulting in protease inactivation and membrane lipid peroxidation [15,19,34]. Antioxidant enzymes (e.g., SOD, POD, and CAT) could help mitigate ROS in plants [8,10,35]. In this present study, POD, SOD, and CAT activities increased under low and moderate salinity stress (S1 and S2) and declined under severe salinity stress (S3). These results are in accordance with the findings reported in the hydroponic experiments on seedlings of *Jatropha curcas* L. [8] and cucumber [36] under various saline concentrations. This demonstrates that greater antioxidant enzymes under lower salinity stress could facilitate the mitigation of oxidative damage, whereas the damage was difficult to alleviate from lower POD, SOD, and CAT contents under higher salinity stress. The lower antioxidant enzyme activity could elevate ROS accumulation in plants and further cause a reduction in photosynthetic membrane permeability and decomposition in photosynthetic pigments [37]. The lower photosynthetic pigments found in severe salinity stress treatments in this study (Table 3) could consolidate the above speculation. Additionally, nitrogen application could increase the photosynthetic pigments of *Jatropha curcas* L. seedlings, reducing the vulnerability of photosynthesis to salinity stress. The possible reason is that the nitrogen application could enhance the nitrogen utilization in the leaf for synthetic chlorophyll under salinity stress [29], as nitrogen is an important component in antioxidant substances. The lower photosynthetic pigment contents under severe salinity stress may result from exceeding the threshold of salinity tolerance in *Jatropha curcas* L. seedlings, consequently reducing the benefits of nitrogen application.

Photosynthesis is highly affected by the CO_2 concentration gradient and diffusion resistance between inside and outside plant leaves [38], resulting in stomatal conductance as a critical factor in determining plant photosynthesis. The reduction in photosynthesis caused by salinity stress can be attributed to two main reasons for stomatal closure. One reason is a stomatal limitation which occurs when the decline in stomatal conductance inhibits the diffusion of CO_2 from the outside of leaves to the carboxylated sites in chloroplasts, thereby leading to a decrease in photosynthetic rate [39]. The other reason is a non-stomatal limitation where the photosynthetic rate decreases as a result of salinity stress affecting the assimilation ability of mesophyll cells, and this decrease is unrelated to the intercellular CO_2 concentration [40]. In the present study, P_n, G_s, and T_r gradually decreased, while C_i decreased initially and eventually increased with the increase in salinity stress, resulting from stomatal contraction hindering the transport of CO_2 to the chloroplast under low and moderate salinity stresses, and the reduction in photosystem II and enzyme activities lowering CO_2 availability under severe salinity stress causing CO_2 accumulation in cells [41]. In addition, the nitrogen application was found to improve the photosynthesis in *Jatropha curcas* L. seedlings under salinity stress. According to a stomatal limitation in low and moderate salinity stresses and a non-stomatal in severe salinity stress, nitrogen application could alleviate water loss in plant cells and compensate for stomatal closure under low and moderate salinity stresses, whereas nitrogen application could increase antioxidant substances and photosynthetic pigment content under severe salinity stress, ultimately improving photosynthesis.

Chlorophyll fluorescence parameters can effectively characterize the intrinsic relationship between salinity stress and photosynthesis. Φ_{PSII} represents the ratio of the actual light energy consumed in the photochemical reaction to the total light energy absorbed by the plant leaves. The greater the value of Φ_{PSII}, the more the part of the light energy is effectively utilized. Light energy absorbed by photosynthetic pigments in leaves is used for photoelectron transport, chlorophyll fluorescence, and heat dissipation. The photo-

chemical quenching coefficient (qP) and non-photochemical quenching coefficient (NPQ) represent the photochemical consumption and heat dissipation, respectively. qP is used to characterize the openness of PSII reaction centers. In the present study, Φ_{PSII}, qP, and ETR decreased while NPQ increased with the increase in salinity stress. However, nitrogen application could enhance Φ_{PSII}, qP, and ETR, as well as reduce NPQ. One reason for the effect of nitrogen application on chlorophyll fluorescence parameters could be that the presence of nitrogen could increase the light absorption intensity and light capture ability of leaves of *Jatropha curcas* L. seedlings from the increase in leaf area (Figure 1), promoting assimilation capacity (ATP and NADPH) of leaf cells to facilitate the fixation and assimilation of carbon [42], ultimately improving photochemical reaction efficiency. Nitrogen application could also regulate PSII to consume excessive light energy and enhance Calvin cycle activity, eventually avoiding damage to the reaction center from salinity stress [31]. However, the normal physiological function of plants was damaged, and the heat dissipation ability was gradually lost under severe salinity stress, indicating that the contribution of nitrogen application in alleviating chlorophyll fluorescence parameters may not be significant.

5. Conclusions

In the present experiment, *Jatropha curcas* L. seedlings were sensitive to salinity stress, and salinity stress significantly reduced plant growth and physiology. Nitrogen application could alleviate the negative effect of salinity stress on plant growth by promoting the antioxidant enzyme, photosynthetic pigment, photosynthesis, and chlorophyll fluorescence. In general, the N3 treatment in mild and moderate salinity stresses could achieve maximum plant growth, antioxidant enzyme activity, leaf photosynthetic pigment, and photosynthesis. As for severe salinity stress, higher nitrogen application rate (N2 and N3) had no significant effects on plant growth, photosynthetic characteristics, and leaf chlorophyll fluorescence parameters under severe salinity stress. Therefore, N3 treatment (100 gN/plant) was recommended under mild and moderate salinity stresses, while N2 (60 gN/plant) was recommended under severe salinity stress. This study could provide a theoretical basis for soil salinity management and nitrogen fertilization application for *Jatropha curcas* L. seedlings. However, the nitrogen application rate higher than 100 gN/plant in mild and moderate salinity stresses should be noted in further study, which may achieve a better performance of *Jatropha curcas* L. seedlings. In addition, the reactive oxygen species seems to be an essential indicator to reflect plant growth and physiological characteristics, which could be considered in future research.

Author Contributions: Conceptualization, Z.Y., Q.Y. and S.C.; methodology, Z.Y. and S.T.; software, Z.Y. and S.T.; validation, Q.Y. and X.L.; formal analysis, Z.Y., S.T. and S.C.; investigation, Z.Y. and C.Q.; data curation, Z.Y. and C.Q.; writing—original draft preparation, Z.Y. and S.T.; writing—review and editing, S.T., S.C., J.L. and H.W.; visualization, Z.Y. and S.T.; supervision, Q.Y., X.L., J.L. and H.W.; funding acquisition, Q.Y. and S.C. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by the National Natural Science Foundation of China grant number [51379004 and 51009073], the Yunnan Science and Technology Talent and Platform Program [202305AM070006], the Key Laboratory Project of Efficient Water Use and Green Production of Specialty Crops in Yunnan Universities [KKPS201923009], and the Scientific Research Fund Project of Yunnan Provincial Department of Education [2022J0061].

Data Availability Statement: The data used to support the findings of this study are included within the article.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Lozano-Isla, F.; Campos, M.L.; Endres, L.; Bezerra-Neto, E.; Pompelli, M.F. Effects of seed storage time and salt stress on the germination of *Jatropha curcas* L. *Ind. Crops Prod.* **2018**, *118*, 214–224. [CrossRef]
2. Corte-Real, N.; Endres, L.; Santos, K.; Figueirêdo, R.; Arruda, E.; Ulisses, C.; Pompelli, M.; Figueirêdo, R.; Pompelli, M. Morphoanatomy and ontogeny of the fruit and seeds of *Jatropha curcas* L.: A promising biofuel plant. *Promis. Future Jatropha Curcas Propr. Potential Appl. Hauppauge NY Nova Sci. Publ. Inc* **2016**, *8*, 141–158.
3. Silitonga, A.S.; Hassan, M.H.; Ong, H.C.; Kusumo, F. Analysis of the performance, emission and combustion characteristics of a turbocharged diesel engine fuelled with *Jatropha curcas* biodiesel-diesel blends using kernel-based extreme learning machine. *Environ. Sci. Pollut. Res.* **2017**, *24*, 25383–25405. [CrossRef]
4. Abrar, M.M.; Saqib, M.; Abbas, G.; Atiq-ur-Rahman, M.; Mustafa, A.; Shah, S.A.A.; Mehmood, K.; Maitlo, A.A.; Sun, N.; Xu, M. Evaluating the contribution of growth, physiological, and ionic components towards salinity and drought stress tolerance in Jatropha curcas. *Plants* **2020**, *9*, 1574. [CrossRef] [PubMed]
5. Reubens, B.; Achten, W.M.; Maes, W.; Danjon, F.; Aerts, R.; Poesen, J.; Muys, B. More than biofuel? *Jatropha curcas* root system symmetry and potential for soil erosion control. *J. Arid Environ.* **2011**, *75*, 201–205. [CrossRef]
6. Pompelli, M.F.; Barata-Luís, R.; Vitorino, H.S.; Gonçalves, E.R.; Rolim, E.V.; Santos, M.G.; Almeida-Cortez, J.S.; Ferreira, V.M.; Lemos, E.E.; Endres, L. Photosynthesis, photoprotection and antioxidant activity of purging nut under drought deficit and recovery. *Biomass Bioenergy* **2010**, *34*, 1207–1215. [CrossRef]
7. de Lima Cabral, G.A.; Binneck, E.; de Souza, M.C.P.; da Silva, M.D.; Costa Ferreira Neto, J.R.; Pompelli, M.F.; Endres, L.; Kido, É.A. First expressed TFome of physic nut (*Jatropha curcas* L.) after salt stimulus. *Plant Mol. Biol. Report.* **2020**, *38*, 189–208. [CrossRef]
8. Gao, S.; Ouyang, C.; Wang, S.; Xu, Y.; Tang, L.; Chen, F. Effects of salt stress on growth, antioxidant enzyme and phenylalanine ammonia-lyase activities in *Jatropha curcas* L. seedlings. *Plant Soil Env.* **2008**, *54*, 374–381. [CrossRef]
9. de Oliveira, D.F.B.; Endres, L.; Silva, J.V.; Clemente, P.R.A. Pre-colonized seedlings with arbuscular mycorrhizal fungi: An alternative for the cultivation of *Jatropha curcas* L. in salinized soils. *Theor. Exp. Plant Physiol.* **2017**, *29*, 129–142. [CrossRef]
10. Pompelli, M.F.; Jarma-Orozco, A.; Rodríguez-Páez, L.A. Salinity in Jatropha curcas: A review of physiological, biochemical, and molecular factors involved. *Agriculture* **2022**, *12*, 594. [CrossRef]
11. Patel, A.D.; Panchal, N.S.; Pandey, I.B.; Pandey, A.N. Growth, water status and nutrient accumulation of seedlings of *Jatropha curcas* L. (Euphorbiaceae) in response to soil salinity. In *Anales de Biología*; Facultad de Biología, Universidad de Murcia: Murcia, Spain, 2010; pp. 59–71.
12. Liu, B.; Yang, Q.; Xu, M. Effects of water-nitrogen addition on *Jatropha curcas* L. physiological characteristics under salt stress. *J. Drain. Irrig. Mach. Eng.* **2018**, *36*, 679–684.
13. da Silva Cavalcante, P.G.; dos Santos, C.M.; de Lins Wanderley Filho, H.C.; Avelino, J.R.L.; Endres, L. Morpho-physiological adaptation of *Jatropha curcas* L. to salinity stress. *Aust. J. Crop Sci.* **2018**, *12*, 563–571. [CrossRef]
14. Silva, E.d.; Ribeiro, R.; Ferreira-Silva, S.; Viégas, R.; Silveira, J. Comparative effects of salinity and water stress on photosynthesis, water relations and growth of *Jatropha curcas* plants. *J. Arid Environ.* **2010**, *74*, 1130–1137. [CrossRef]
15. Iqbal, N.; Umar, S.; Khan, N.A. Nitrogen availability regulates proline and ethylene production and alleviates salinity stress in mustard (*Brassica juncea*). *J. Plant Physiol.* **2015**, *178*, 84–91. [CrossRef]
16. Reda, M.; Migocka, M.; Kłobus, G. Effect of short-term salinity on the nitrate reductase activity in cucumber roots. *Plant Sci.* **2011**, *180*, 783–788. [CrossRef]
17. Syeed, S.; Anjum, N.A.; Nazar, R.; Iqbal, N.; Masood, A.; Khan, N.A. Salicylic acid-mediated changes in photosynthesis, nutrients content and antioxidant metabolism in two mustard (*Brassica juncea* L.) cultivars differing in salt tolerance. *Acta Physiol. Plant* **2011**, *33*, 877–886. [CrossRef]
18. Hessini, K.; Issaoui, K.; Ferchichi, S.; Saif, T.; Abdelly, C.; Siddique, K.H.; Cruz, C. Interactive effects of salinity and nitrogen forms on plant growth, photosynthesis and osmotic adjustment in maize. *Plant Physiol. Biochem.* **2019**, *139*, 171–178. [CrossRef]
19. Singh, M.; Singh, V.P.; Prasad, S.M. Nitrogen alleviates salinity toxicity in *Solanum lycopersicum* seedlings by regulating ROS homeostasis. *Plant Physiol. Biochem.* **2019**, *141*, 466–476. [CrossRef]
20. Novoa, R.; Loomis, R. Nitrogen and plant production. *Plant Soil* **1981**, *58*, 177–204. [CrossRef]
21. Peng, J.; Feng, Y.; Wang, X.; Li, J.; Xu, G.; Phonenasay, S.; Luo, Q.; Han, Z.; Lu, W. Effects of nitrogen application rate on the photosynthetic pigment, leaf fluorescence characteristics, and yield of indica hybrid rice and their interrelations. *Sci. Rep.* **2021**, *11*, 7485. [CrossRef]
22. Huang, G.; Shu, Y.; Peng, S.; Li, Y. Leaf photosynthesis is positively correlated with xylem and phloem areas in leaf veins in rice (*Oryza sativa*) plants. *Ann. Bot.* **2022**, *129*, 619–631. [CrossRef]
23. Ru, C.; Hu, X.; Chen, D.; Song, T.; Wang, W.; Lv, M.; Hansen, N.C. Nitrogen modulates the effects of short-term heat, drought and combined stresses after anthesis on photosynthesis, nitrogen metabolism, yield, and water and nitrogen use efficiency of wheat. *Water* **2022**, *14*, 1407. [CrossRef]
24. Muhammad, I.; Yang, L.; Ahmad, S.; Zeeshan, M.; Farooq, S.; Ali, I.; Khan, A.; Zhou, X.B. Irrigation and nitrogen fertilization alter soil bacterial communities, soil enzyme activities, and nutrient availability in maize crop. *Front. Microbiol.* **2022**, *13*, 105. [CrossRef]

25. Muhammad, I.; Yang, L.; Ahmad, S.; Farooq, S.; Al-Ghamdi, A.A.; Khan, A.; Zeeshan, M.; Elshikh, M.S.; Abbasi, A.M.; Zhou, X.-B. Nitrogen fertilizer modulates plant growth, chlorophyll pigments and enzymatic activities under different irrigation regimes. *Agronomy* **2022**, *12*, 845. [CrossRef]
26. Zhang, Y.; Luo, J.; Peng, F.; Xiao, Y.; Du, A. Application of bag-controlled release fertilizer facilitated new root formation, delayed leaf, and root senescence in peach trees and improved nitrogen utilization efficiency. *Front. Plant Sci.* **2021**, *12*, 627313. [CrossRef]
27. Shen, Z.; Pu, X.; Wang, S.; Dong, X.; Cheng, X.; Cheng, M. Silicon improves ion homeostasis and growth of liquorice under salt stress by reducing plant Na^+ uptake. *Sci. Rep.* **2022**, *12*, 5089. [CrossRef]
28. Ahanger, M.A.; Qin, C.; Begum, N.; Maodong, Q.; Dong, X.X.; El-Esawi, M.; El-Sheikh, M.A.; Alatar, A.A.; Zhang, L. Nitrogen availability prevents oxidative effects of salinity on wheat growth and photosynthesis by up-regulating the antioxidants and osmolytes metabolism, and secondary metabolite accumulation. *BMC Plant Biol.* **2019**, *19*, 479. [CrossRef] [PubMed]
29. Singh, M.; Singh, V.P.; Prasad, S.M. Responses of photosynthesis, nitrogen and proline metabolism to salinity stress in *Solanum lycopersicum* under different levels of nitrogen supplementation. *Plant Physiol. Biochem.* **2016**, *109*, 72–83. [CrossRef] [PubMed]
30. Zhang, J.; Zhang, D.; Cai, Z.; Wang, L.; Wang, J.; Sun, L.; Fan, X.; Shen, S.; Zhao, J. Spectral technology and multispectral imaging for estimating the photosynthetic pigments and SPAD of the Chinese cabbage based on machine learning. *Comput. Electron. Agric.* **2022**, *195*, 106814. [CrossRef]
31. Gong, B.; Wen, D.; VandenLangenberg, K.; Wei, M.; Yang, F.; Shi, Q.; Wang, X. Comparative effects of NaCl and $NaHCO_3$ stress on photosynthetic parameters, nutrient metabolism, and the antioxidant system in tomato leaves. *Sci. Hortic.* **2013**, *157*, 1–12. [CrossRef]
32. Zhang, M.S.; Tan, F. Relationship between ratio of Chlorophyll a and b under water stress and drought resistance of different sweet potato varieties. *Seed* **2001**, *4*, 23–25.
33. Liu, C.Y.; Yan, M.; Huang, X.B. Effects of salt stress on growth and physiological characteristics of pomegranate (*Punica granatum* L.) cuttings. *Pak. J. Bot.* **2018**, *50*, 457–464.
34. Suzuki, N.; Koussevitzky, S.; Mittler, R.; Miller, G. ROS and redox signalling in the response of plants to abiotic stress. *Plant Cell Environ.* **2012**, *35*, 259–270. [CrossRef]
35. Feghhenabi, F.; Hadi, H.; Khodaverdiloo, H.; Van Genuchten, M.T.; Pessarakli, M. Improving wheat (*Triticum aestivum* L.) antioxidative defense mechanisms against salinity stress by exogenous application of potassium silicate. *J. Plant Nutr.* **2022**, *45*, 2887–2905. [CrossRef]
36. Turan, M.; Ekinci, M.; Kul, R.; Boynueyri, F.G.; Yildirim, E. Mitigation of salinity stress in cucumber seedlings by exogenous hydrogen sulfide. *J. Plant Res.* **2022**, *135*, 517–529. [CrossRef]
37. Liang, Y.; Ding, R. Influence of silicon on microdistribution of mineral ions in roots of salt-stressed barley as associated with salt tolerance in plants. *Sci. China Ser. C Life Sci.* **2002**, *45*, 298–308. [CrossRef] [PubMed]
38. Harrison, E.L.; Arce Cubas, L.; Gray, J.E.; Hepworth, C. The influence of stomatal morphology and distribution on photosynthetic gas exchange. *Plant J.* **2020**, *101*, 768–779. [CrossRef] [PubMed]
39. Liao, Q.; Gu, S.; Kang, S.; Du, T.; Tong, L.; Wood, J.D.; Ding, R. Mild water and salt stress improve water use efficiency by decreasing stomatal conductance via osmotic adjustment in field maize. *Sci. Total Environ.* **2022**, *805*, 150364. [CrossRef]
40. Pan, T.; Liu, M.; Kreslavski, V.D.; Zharmukhamedov, S.K.; Nie, C.; Yu, M.; Kuznetsov, V.V.; Allakhverdiev, S.I.; Shabala, S. Non-stomatal limitation of photosynthesis by soil salinity. *Crit. Rev. Environ. Sci. Technol.* **2021**, *51*, 791–825. [CrossRef]
41. Jamil, M.; Lee, K.J.; Kim, J.M.; Kim, H.-S.; Rha, E.S. Salinity reduced growth PS2 photochemistry and chlorophyll content in radish. *Sci. Agric.* **2007**, *64*, 111–118. [CrossRef]
42. Peng, W.; Wang, C.-D.; Wang, X.-L.; Wu, Y.-L.; Zhang, Y.; Sun, Y.-G.; Yi, S.; Mi, G.-H. Increasing nitrogen absorption and assimilation ability under mixed $NO3^-$ and $NH4^+$ supply is a driver to promote growth of maize seedlings. *J. Integr. Agric.* **2023**, *22*, 1896–1908.